

Identifying Predictors of Response to Suplatast Tosilate among Patients with Moderate to Severe Bronchial Asthma Receiving Inhaled Steroid Therapy

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ABSTRACT

Background: The aim of this study was to identify the predictive factors, among laboratory test data and patient background variables, of an efficient response to the anti-allergic agent suplatast tosilate in patients with moderate to severe bronchial asthma.

Methods: The subjects were 44 patients with moderate to severe bronchial asthma on inhaled steroid therapy who were enrolled in a phase II clinical trial of suplatast (300 and 600 mg/day). Improvements in respiratory function parameters and symptom scores during the first 4 weeks of administration of suplatast were assessed to evaluate the response to the drug. Logistic regression analysis was used to relate the response to the independent variables. Secondly, to test whether these results were applicable to clinical practice, we examined the data from a phase III clinical trial of suplatast.

Results: Twenty-two patients were assessed as responders according to our criteria. The percentage of blood eosinophil (%EOS, $P = 0.015$) and basophil (%BASO, $P = 0.019$) counts were identified as significant variables to predict responders. When cut-off levels for %EOS and %BASO were set at 7.5 and 1.2, respectively, the sensitivity for prediction of responders with lower %EOS and %BASO was 81.8% (18/22). Furthermore, when we applied the same cut-off levels to subjects of a phase III clinical trial of suplatast, the sensitivity of prediction was found to be as high as 75.0% (6/8).

Conclusions: These results indicate that %EOS and %BASO are good candidates to predict the response to suplatast among patients with moderate to severe bronchial asthma on inhaled steroid therapy. These predictors may contribute, in combination with genomic information, to stratified medical treatment tailored to the individual needs of patients.

KEY WORDS

antiasthmatic agent, basophil, bronchial asthma, eosinophil, response prediction

INTRODUCTION

Suplatast tosilate (suplatast), a dimethylsulfonium compound, is used in Japan as a therapeutic drug for allergic diseases such as bronchial asthma, allergic rhinitis, and atopic dermatitis.^{1,2} Suplatast has been reported to attenuate the allergen-induced late asthmatic response,³ airway hyperresponsiveness and in-

filtration of eosinophils and CD 4⁺ lymphocytes in guinea pigs,^{3,4} human IgE synthesis,⁵ and antigen-elicited peritoneal eosinophilia.⁶ These effects are considered to be mediated through selective inhibition of interleukin-4 (IL-4) and interleukin-5 (IL-5) production by type-2 helper T cells (Th2-cells), without any effects on Interferon-gamma (IFN-gamma) production from type-1 helper T cells (Th1-cells).⁷

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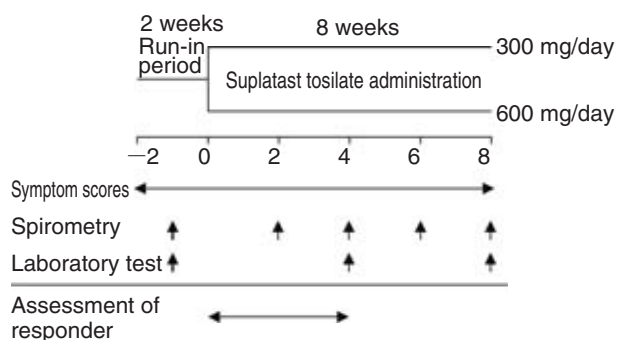


Fig. 1 Study protocol. After an initial 2-week run-in period, the treatment group was administered suplatast (300 mg/day or 600 mg/day) for 8 weeks. The responder was assessed first at 4 weeks after the start of treatment.

In clinical studies, suplatast has been shown to have beneficial effects in symptomatic asthmatic patients, and to reduce the levels of markers of inflammation and airway hyperresponsiveness. Tamaoki *et al.*⁸ evaluated the steroid-sparing effect of suplatast relative to placebo in patients with steroid-dependent asthma. When the steroid dose was halved, the patients in the suplatast group had less deterioration of pulmonary function and of asthma symptoms. In a recent study designed to further investigate the efficacy and mechanism of action of suplatast, Sano *et al.*⁹ sought to determine the suppressive effects of suplatast on the eosinophilic inflammation of the bronchial mucosa in patients with mild asthma. They reported a significant decrease in the number of infiltrating and activated eosinophils, besides improvement of the respiratory function parameters and symptom scores.

In 2002, a phase II clinical study of suplatast was conducted in patients with moderate to severe persistent bronchial asthma on inhaled steroid therapy to examine the reducing effect of a concomitantly administered 8-week course of suplatast (300 and 600 mg/day) on the dose level of inhaled steroids needed to control the asthma.¹⁰ In that study, the dose level of inhaled steroids could not be reduced in some patients, and these patients showed no improvement in the respiratory function parameters despite concomitant suplatast therapy. On the other hand, in other patients, improvement of the respiratory function parameters was observed by the 4th week; and the dose level of the inhaled steroids could be reduced by the 8th week after the start of the study. These patients were thus regarded as responders to suplatast. These results strongly suggest that there are responders and nonresponders to suplatast therapy.¹⁰ The data from this clinical study were analyzed in-depth to develop a method for predicting the response to suplatast. Focusing on the routine pre-treatment laboratory parameters and background variables of the

patients, we attempted to identify factors that might be predictive of the responses of these patients to this drug. The factors thus identified were retrospectively analyzed among the subjects of a previously performed phase III clinical study to verify their usefulness in predicting the response to suplatast therapy. In this way, we succeeded in developing a highly reliable method for predicting responders to suplatast therapy. In recent years, analysis of single-nucleotide polymorphisms (SNPs) and gene expression analysis in target tissues have often been used for predicting the responses and adverse reactions to drugs. Based on the results of the present study, we would like to emphasize that routinely measured laboratory parameters can also sometimes serve as valid predictors of responses to drug therapy.

METHODS

PATIENTS

Forty-four patients fulfilled the diagnostic criteria for moderate to severe bronchial asthma described in "Guidelines for Prevention and Management of Asthma", revised edition published in 1998 (JGL' 98), Japan.¹⁰ Fourteen centers participated in the trial and recruited asthma outpatients 20–64 years of age who were taking 400–1600 µg of inhaled beclomethasone dipropionate and less than 5 mg of oral prednisolone daily for at least 8 weeks prior to the commencement of the study. Other entry criteria included a forced expiratory volume in 1 s (FEV₁) of 50–80% of the predicted normal. Patients were excluded from the trial if they had used Chinese herbal medicines, leukotriene receptor antagonists, 5-lipoxygenase inhibitors, thromboxane synthase inhibitors, or thromboxane A₂ antagonists during the 4 weeks prior to their proposed entry into the study; or had received intramuscular or intravenous injections of corticosteroids or bronchodilator during the 2-week run-in period; or had been treated with allergen desensitization therapy or non-specific therapy during that period. A history of serious disease or other drug-induced disorders also precluded entry into the study. The protocol (Fig. 1) for this study was approved by the multi-center institutional review board, and written informed consent was obtained from each of the participants prior to their entry into the study.

STUDY DESIGN

The phase II clinical trial of suplatast (300 and 600 mg/day) in Japan¹¹ was a multi-center, open-label, randomized, parallel-group controlled study involving a 2-week run-in period (baseline) followed by an 8-week treatment period. During the 2-week run-in period, the patients continued to take their normal daily dose of beclomethasone dipropionate from pressurized metered-dose inhalers and received 3 capsules daily of matching placebo. During the 8-week treatment period, the patients were randomly assigned to

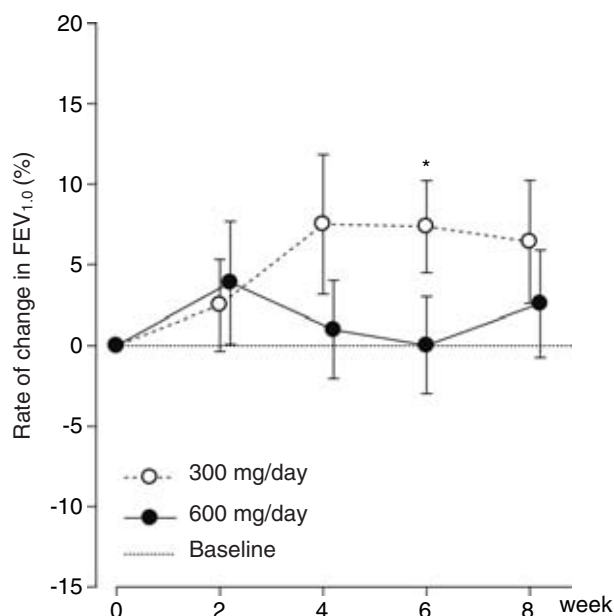


Fig. 2 Changes from baseline in FEV₁. FEV₁ in the 300 mg/day group increased significantly at week 6. (* $P < 0.05$ by t test)

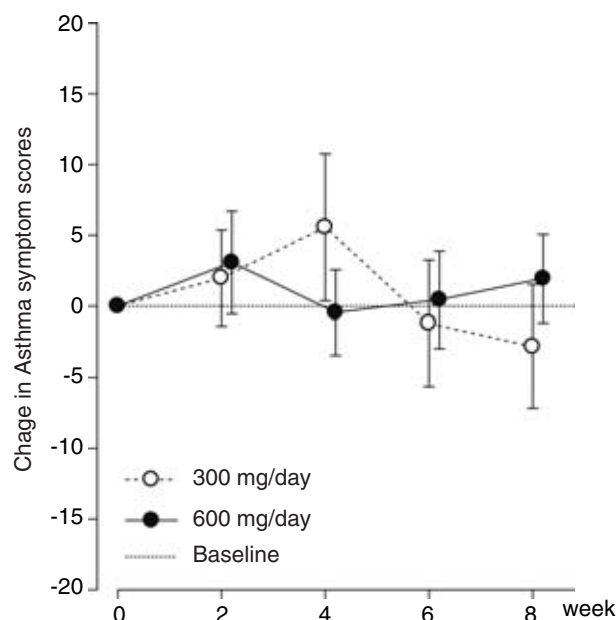


Fig. 3 Changes from baseline in asthma symptom scores. Asthma symptom scores did not change from the baseline in either the 300 mg/day group or the 600 mg/day group.

oral 300 mg suplatast (100 mg active capsule; 3 capsules daily) or oral 600 mg suplatast (100 mg active capsule; 6 capsules daily).

All the patients visited the outpatient clinic at least once every 2 weeks throughout the study period. At the baseline period, and at weeks 2, 4, 6, and 8 of treatment, spirometry was performed. Each patient was instructed to maintain a record in a booklet of all the medications taken during the study period. The asthma symptom scores were recorded along the guidelines¹⁰ commonly used in Japan, 4 times daily based on the severity of cough (severe, 1; mild, 0.5), presence of chest tightness (1), presence of wheeze (1), and the severity of asthma (mild, 3; moderate, 6; severe, 9); and the total symptom scores for 2 weeks were calculated. To assess safety, we did hematological, biochemical, and urine analyses at the baseline period, and at weeks 4 and 8 of treatment. Adverse events were recorded daily and were reviewed by the physician at every study-related visit to the study site.

An efficient response to suplatast was assessed first at 4 weeks after the start of treatment. A response to suplatast was judged as positive when an improvement of greater than 10% occurred in the respiratory function parameters (FEV₁), or when the asthma symptom scores improved by an average of 1 point/day or 14 points in a 2 week period. (The maximum possible symptom score of nonresponders and responders at baseline was 26.6 and 40.5, respectively).

STATISTICAL ANALYSIS

Primary endpoints were FEV₁ and asthma symptom

scores. FEV₁ was recorded every 2 weeks, and we calculated the rate of change from the values at the baseline. Asthma symptom scores recorded, 4 times daily, were expressed as 2-week totals, and we determined the variation from the values at the baseline. We used t tests to evaluate the significance of the differences within and between groups, and $P < 0.05$ was taken to indicate significant differences.

A logistic regression analysis was also used to relate the response to the independent variables. The independent variables considered in this study were the clinical parameters used as outcome parameters (respiratory function testing and asthma symptom scores at the baseline period), pre-treatment laboratory test data (hematological, biochemical and urine analyses), and the background variables (body weight, age of onset, severity, type of disease (atopic, infectious and mixed), presence of complications, medical history, family history and treatment history). A univariate analysis was first performed to select the independent variables to predict the responders, with a P value of 0.05. A multivariate regression model was then built using forward selection.

The software package SAS ver. 8.2 was used for the statistical analyses.

VERIFYING THE PREDICTORS

The usefulness of the identified predictors was verified using the data from an independently performed phase III clinical trial of suplatast (300 mg/day) in Ja-

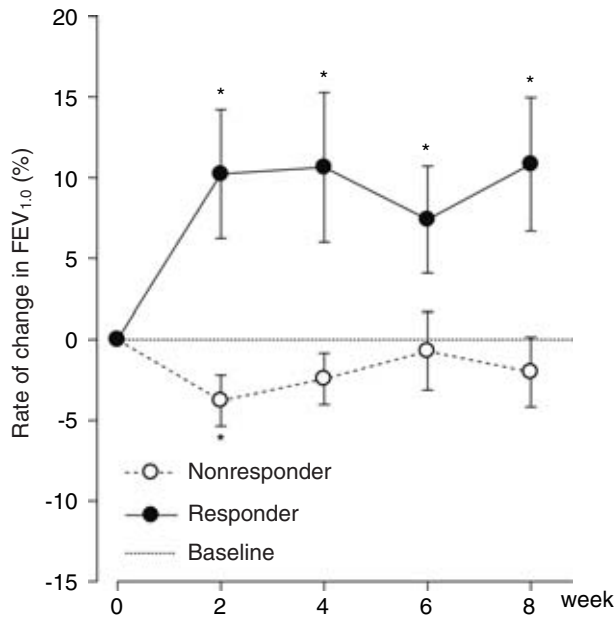


Fig. 4 Changes from baseline in FEV₁. FEV₁ in the responder group increased significantly; whereas in the nonresponder group there was no significant change from the baseline at weeks 4, 6, and 8, although the value decreased significantly at week 2. (* $P < 0.05$ by t test)

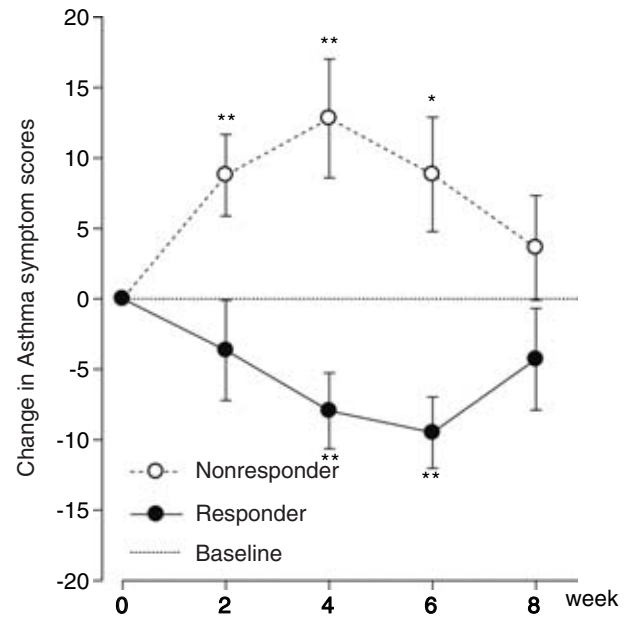


Fig. 5 Changes from baseline in asthma symptom scores. Asthma symptom scores in the responder group decreased significantly at weeks 4 and 6; whereas in the nonresponder group they increased significantly at weeks 2, 4, and 6. (* $P < 0.05$, ** $P < 0.01$ by t test)

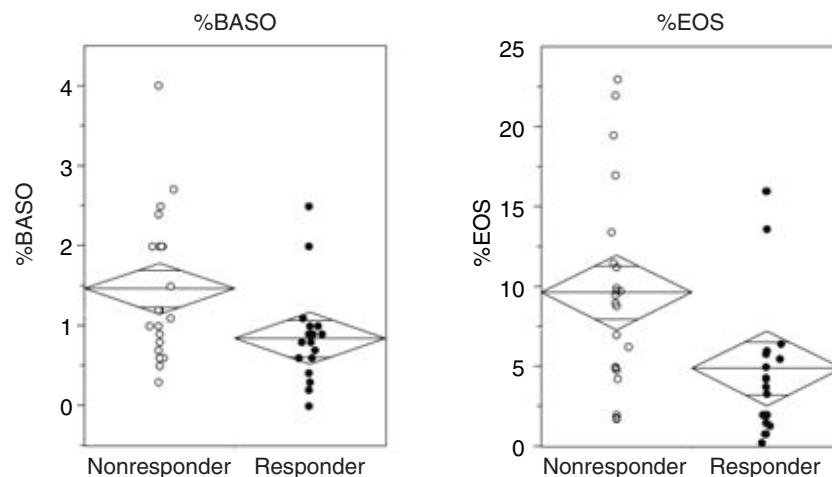


Fig. 6 Percentage of blood eosinophils (%EOS) and blood basophils (%BASO) before the suplatast treatment. The responder group had a statistically lower %EOS and %BASO than the nonresponder group ($P = 0.006$ for %EOS and $P = 0.009$ for %BASO according to the t test). The line across each diamond represents the group mean and the vertical span of each diamond represents the 95% confidence interval for each group.

pan.¹² Of the subjects of that study, 19 patients with moderate to severe bronchial asthma on steroid therapy who had undergone respiratory function testing during the study were selected for this analysis. The response of individual patients to suplatast was

judged using the same criteria as those employed in the phase II study, except that the symptom scores and respiratory function data in the 6th week were substituted for the data in the 4th week of dosing, because these data in the 4th week were unavailable in

Table 1 Measurements of run-in period

	Nonresponder (<i>n</i> = 22)	Responder (<i>n</i> = 22)	<i>p</i> value
Age (yr)			
Mean ± SD	49.6 ± 12.7	52.1 ± 13.3	0.535
Gender			
Female	8	10	0.540
Male	14	12	
Dose			
300 mg/day	13	8	0.131
600 mg/day	9	14	
Asthma severity			
Moderate	10	15	0.128
Severe	12	7	
Duration of asthma (yr)			
Mean ± SD	18.4 ± 14.9	14.1 ± 9.8	0.269
Serum IgE (IU/l)			
Mean + SD	345.3 ± 435.6	560.2 ± 759.3	0.256
Beclomethasone use (μg/day)			
≤ 400 – < 800	3	8	0.140
≤ 800 – < 1200	10	5	
< 1200 – ≤ 1600	9	9	
FEV ₁			
Mean ± SD	1774.7 ± 399.7	1739.5 ± 519.9	0.803
Symptom scores			
Mean ± SD	26.6 ± 21.9	40.5 ± 25.7	0.060

P values for categorical data are obtained by χ^2 test; *P* values for others are obtained by *t* test.

Baseline characteristics of patients for nonresponder and responder groups. Distribution of age, gender, doses, asthma severity, asthma duration, serum IgE, beclomethasone, FEV₁ and symptom scores did not differ significantly between the 2 groups. *P* values for categorical data are obtained by χ^2 test and for others are obtained by *t* test.

Table 2 Results of logistic regression

No.	Variable	No. of patients	Estimate	Odds Ratio	Lower CL	Upper CL	<i>P</i> Value
1	%blood eosinophils (%EOS)	44	– 0.174	0.840	0.730	0.966	0.015*
2	%blood basophils (%BASO)	44	– 1.297	0.273	0.092	0.811	0.019*
3	Alkaline phosphatase	43	0.011	1.011	1.000	1.022	0.042*
4	Potassium	44	1.755	5.783	0.875	38.217	0.069
5	Glutamic oxaloacetic transaminase	44	0.097	1.102	0.992	1.224	0.071
6	γ-glutamyl transpeptidase	43	0.029	1.030	0.993	1.068	0.110
7	Total protein	43	1.289	3.629	0.744	17.702	0.111
8	Severity	44	– 0.944	0.389	0.114	1.328	0.132
9	Dose	44	0.003	1.003	0.999	1.007	0.135
10	%blood lymphocytes	44	0.048	1.049	0.982	1.121	0.152

P values are obtained by χ^2 test. (**P* < 0.05)

Results of univariate logistic regression. The top 10 variables with smallest *P* values are listed. *P* values are obtained by χ^2 test. (**P* < 0.05)

the phase III study.

RESULTS

FEV₁ increased significantly only at week 6 in the 300 mg/day group (Fig. 2). Asthma symptom scores did not change from the baseline in either the 300 mg/

day group or the 600 mg/day group (Fig. 3). There were no significant differences between the 300 mg/day group and the 600 mg/day group with respect to FEV₁ or asthma symptom scores.

In accordance with our criteria, we found 22 responders and 22 nonresponders to suplatast. Pa-

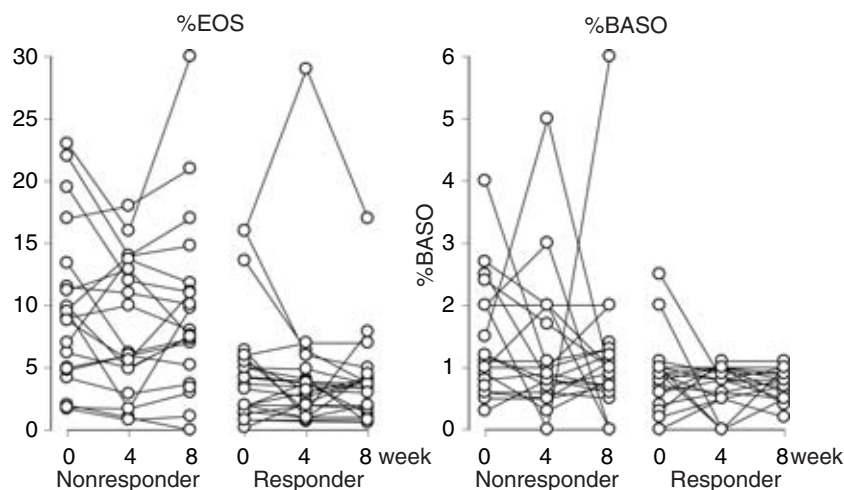


Fig. 7 Changes in percentage of blood eosinophils (%EOS) and blood basophils (%BASO).

tients' baseline characteristics are given in Table 1. Distribution of age, gender, doses, asthma severity, asthma duration, serum IgE, beclomethasone dipropionate, FEV₁ or symptom scores did not differ significantly between the 2 groups. In the nonresponder group the mean symptom score at baseline was 26.6 ± 21.9 and in the responder group it was 40.5 ± 25.7 . FEV₁ in the responder group increased significantly; whereas in the nonresponder group there was no significant change from the baseline at weeks 4, 6, and 8, although the value decreased significantly at week 2 (Fig. 4). There were significant differences between the 2 groups at all time points. Asthma symptom scores in the responder group decreased significantly at weeks 4 and 6, and in the nonresponder group increased significantly at weeks 2, 4 and 6 (Fig. 5). There were significant differences between the 2 groups at all time points except week 8.

Obtained by use of univariate logistic regression, the top 10 variables with smallest *P* values are listed in Table 2. In the univariate regression, the independent variables %EOS (*P* = 0.015), %BASO (*P* = 0.019) and alkaline phosphatase (*P* = 0.042) were found to be significantly related to the response to suplatast. Among these, %EOS and %BASO remained in the multivariate regression model. In fact, the absolute %EOS (*P* = 0.006) and %BASO (*P* = 0.009) before the suplatast treatment were significantly lower in the responder group than in the nonresponder group (Fig. 6). On the other hand, %EOS and %BASO did not reach statistically significant levels from the baseline in either the responder group or the nonresponder group (Fig. 7).

We then evaluated the accuracy of prediction of the response to suplatast on the basis of the %EOS and %BASO when the cut-off levels were set at 7.5 and 1.2, respectively (Table 3). When we used the combina-

tion of %EOS and %BASO, the positive predictive value was calculated to be 81.8% (18 out of 22 cases), whereas the percentage of responders in the entire population was 50.0% (Fig. 8). To verify the validity of this method of prediction, we applied the same cut-off levels to the data obtained during a phase III clinical trial of suplatast (Fig. 9). When the analysis was conducted using these cut-off levels in 19 cases of moderate to severe bronchial asthma on steroid therapy, the sensitivity of this method of prediction was found to be as high as 75.0%, whereas the percentage of responders in the total population was 52.6%.

DISCUSSION

Before a drug is prescribed to a given patient, attempts to predict the response and adverse reactions of the patients to the drug have, in recent times, often been made by means of SNP analysis, gene expression analysis, or proteome analysis of target tissues. In the present study, we sought to identify the factors potentially contributing to the response of individual patients to the drug therapy among routinely measured laboratory parameters and patient background variables.

In the Phase II study we could not show any dose-response effect of suplatast. However, it should be noted that the study group comprised both patients with and those without a positive response to suplatast. To assess the response to suplatast, we used FEV₁ and asthma symptom scores obtained first at 4 weeks after the start of treatment. Using our criteria, in the responder group, FEV₁ was also improved significantly at weeks 6 and 8, and asthma symptom scores were improved significantly at week 6. These results indicate that the criteria could discriminate responders and nonresponders successfully.

Table 3 Summary of response data

%EOS and %BASO levels	No. of patients	No. of Nonresponder	No. of responder	Ratio of responder (%)	<i>P</i> value
%EOS < 7.5	28	9	19	67.9	0.004
%EOS ≥ 7.5	16	13	3	18.8	
%BASO < 1.2	30	10	20	66.7	0.003
%BASO ≥ 1.2	14	12	2	14.3	
%EOS < 7.5 and %BASO < 1.2	22	4	18	81.8	< 0.001
%EOS ≥ 7.5 or %BASO ≥ 1.2	22	18	4	18.2	

P values are obtained by Fisher's exact test.

Summary of response data for suplatast with different levels of %EOS and %BASO. *P* values are obtained by Fisher's exact test.

We also used logistic regression to study the factors predicting an efficient response to suplatast. In this analysis %EOS and %BASO were selected as predictors. When the response to drug therapy was predicted on the basis of the predictors thus identified (%EOS and %BASO), the positive predictive value was as high as 81.8%. This method of prediction was found to be satisfactorily valid when it was verified by applying the same predictors to the data from an independently performed phase III clinical trial. These results suggest that the combination of %EOS and %BASO might be a predictive marker for responders to suplatast, although the mechanism has not yet been fully elucidated.

Our results suggest that asthma patients on steroid therapy who have a low %EOS and %BASO prior to the start of treatment are more likely to respond to suplatast over the relatively short term (4 weeks). Furthermore, in the responder group %EOS and %BASO were stable at a lower level during the treatment periods, whereas in the nonresponder group they fluctuated greatly (Fig. 7). The mechanism by which the pre-treatment %EOS and %BASO influence the response of asthma patients on inhaled steroid therapy to suplatast remains to be clarified. In the patients with low %EOS and %BASO not receiving inhaled steroids, the response rate to suplatast was not so high among asthmatic patients during the phase III clinical trial of suplatast (data not shown). This finding suggests that the effectiveness of suplatast may be related to the mechanism by which steroids exert their anti-allergy activity.

It was previously reported that treatment with suplatast reduced the elevated blood eosinophil count in patients¹³⁻¹⁶ or the number of eosinophils infiltrating the inflamed tissues.^{9,17} Also in the present phase II clinical trial, some of the patients with elevated %EOS who were on steroid therapy showed a decrease in their %EOS following 4 weeks of suplatast therapy. Therefore, we could speculate that suplatast provides better improvement in the patients with increased

eosinophils. However, our study indicated exactly the contrary results, suggesting that the responders of suplatast might be the patients who had markedly reduced eosinophils with steroid therapy. These findings suggest the necessity of following up and analyzing the course of asthma patients with elevated %EOS for long periods of time. It would seem essential to examine whether or not prolonged suplatast therapy may be useful over the long term in asthma patients on inhaled steroid therapy who showed a decrease in the %EOS during short-term suplatast therapy, even if the asthma symptoms showed no improvement during this period. It would be desirable for large scale prospective studies to be performed in the near future to resolve these issues.

Recently, analyses of SNPs,^{18,19} gene expression,^{20,21} proteomes,^{22,23} and metabolome²⁴ have been conducted in the hope of obtaining an important base for tailored medicine.²⁵ The results of the present study serve to emphasize that factors determining the sensitivity to a drug may even be identified by relatively simple analysis of the laboratory data and background variables of the patients. When combined with genomic information, these data may contribute to stratified medical treatment in the future that can be tailored to individual patients.

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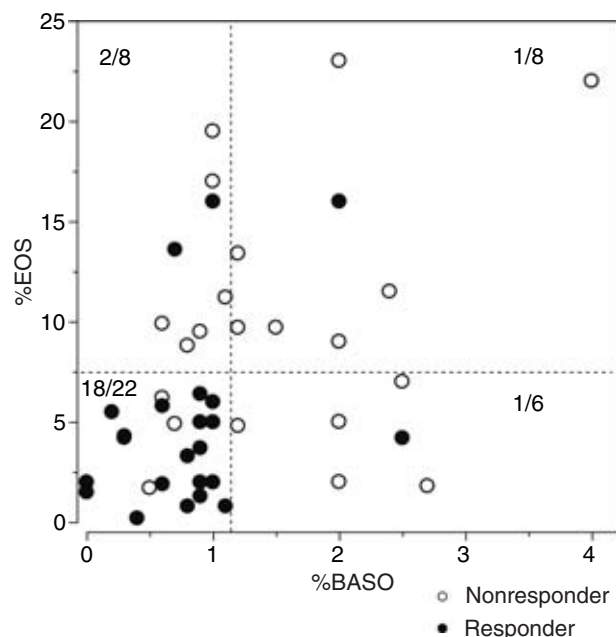


Fig. 8 Percentage of blood eosinophils (%EOS) plotted against blood basophils (%BASO) in the same set of 44 patients in a phase II clinical trial. Solid and empty circles indicate responders and nonresponders, respectively. The dotted lines indicate the response cut-off values (%EOS = 7.5 and %BASO = 1.2). The response rate was 81.8% (18 of 22) in patients with low %EOS and %BASO.

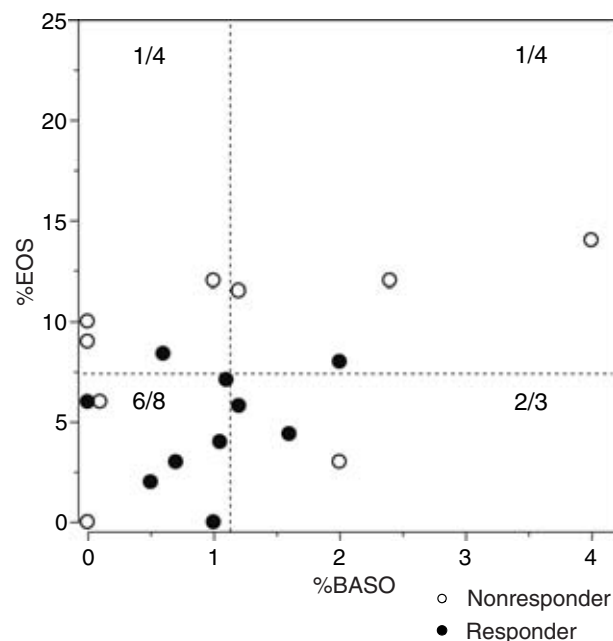


Fig. 9 The same cut-off levels were applied to 19 patients in a phase III clinical trial. Solid and empty circles indicate responders and nonresponders, respectively. The dotted lines indicate the response cut-off values (%EOS = 7.5 and %BASO = 1.2). The response rate was 75.0% (6 of 8) in patients with low %EOS and %BASO.

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