

# Effect of SCH55700, a Humanized Anti-Human Interleukin-5 Antibody, in Severe Persistent Asthma

## A Pilot Study

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Antagonizing the effect of interleukin (IL)-5 is a potential new treatment strategy in allergic disorders. We evaluated the safety, biological activity, and pharmacokinetics of SCH55700, a humanized anti-human IL-5 antibody, in subjects with severe persistent asthma treated with oral or high doses of inhaled steroids. In a double-blind, randomized, multicenter trial, a rising single dose of SCH55700 (0.03 mg/kg [n = 2], 0.1 mg/kg [n = 4], 0.3 mg/kg [n = 6], or 1.0 mg/kg [n = 12]) or placebo (n = 8) was administered intravenously. SCH55700 dose dependently reduced circulating eosinophil counts. At a dose of 1.0 mg/kg, the decrease remained significant up to Day 30 [(0.07 ± 0.01) × 10<sup>9</sup>/L versus (0.23 ± 0.04) × 10<sup>9</sup>/L at baseline] (mean ± SEM) (p = 0.05). After administration of SCH55700 at 0.3 and 1.0 mg/kg, a trend toward improvement in baseline FEV<sub>1</sub> was observed, which reached significance 24 hours after the 0.3-mg/kg dose (p = 0.019 versus placebo). No significant changes occurred in other clinical indices of disease activity. Adverse events were not different between active treatment and placebo. We conclude that SCH55700 is a biologically active anti-human IL-5 antibody that can be safely used in severe steroid-treated asthma. Its therapeutic potential needs to be addressed in specifically designed efficacy trials.

**Keywords:** anti-human interleukin-5 antibody; asthma; eosinophil; glucocorticosteroids; interleukin-5

The presence of activated eosinophils is a characteristic feature of the mucosal inflammation underlying allergic disorders such as asthma or rhinitis (1). Eosinophils are considered to play a key role in the pathophysiology of these disorders, by releasing a wide range of mediators, cytokines, and growth factors (2). Several cytokines can affect eosinophils. However, *in vivo* animal data indicate that interleukin-5 (IL-5) is the main cytokine involved in the production, differentiation, maturation, and activation of eosinophils (3).

The mucosa of asthmatic airways contains increased amounts of both IL-5 mRNA and protein (4). Expression of mRNA encoding IL-5, or the  $\alpha$  chain of its membrane-bound receptor, has been shown to correlate with clinical indices of disease severity (5). Inhalation of IL-5 appears to increase the percentage of eosinophils in induced sputum and to augment airway hyperresponsiveness in subjects with asthma (6). In addition, the effects of IL-5 in humans seem to be largely restricted to various aspects of eosinophil function, having little or no effect on other components of the immune system

(7). These various observations indicate that antagonizing IL-5 could represent an effective and selective anticosinophil approach that might be beneficial in asthma.

SCH55700 is a humanized anti-human IL-5 monoclonal antibody. It is based on 39D10, a rat IgG2a antibody against human IL-5, and incorporates antigen recognition sites for human IL-5 in consensus human IgG4( $\kappa$ ) constant regions using complementarity-determining region-grafting technology (8). *In vitro* studies have confirmed that SCH55700 displays high-affinity binding to human IL-5 (K<sub>d</sub> = 20 pmol/L), potentially inhibiting interaction of human IL-5 with Ba/F3 cells, which overexpress the recombinant human IL-5 receptor  $\alpha$  chain (IC<sub>50</sub> = 0.5 nmol/L).

The aim of this study was to evaluate the safety, biological activity, and pharmacokinetics of SCH55700 in patients with severe persistent asthma. Some of the results of this study have been previously reported in the form of an abstract (9).

## METHODS

### Patients

Subjects with an established diagnosis of asthma for at least 2 years were enrolled. They were at least 18 years of age and had severe persistent asthma, defined on the basis of being symptomatic despite being treated with high doses of inhaled and/or oral corticosteroids. "Symptomatic" was defined as the need for two or more puffs of short-acting inhaled  $\beta_2$  agonists per day and having a symptom score over a 7-day period that exceeded 10 out of a possible maximal score of 42. The score was built up of two questions, one on nighttime symptoms (0 = slept through the night; 3 = kept awake most of the night by asthma symptoms) and one on daytime symptoms (0 = no symptoms; 3 = cannot carry out daily activities due to asthma). On entry, the daily dose of corticosteroids had to be constant for at least 1 month and had to be  $\geq$  1,000  $\mu$ g of fluticasone, or  $\geq$  1,600  $\mu$ g of budesonide, or  $\geq$  2,000  $\mu$ g of beclomethasone dipropionate, or a maintenance dose of oral corticosteroids. Concomitant use of theophylline, short and long-acting inhaled  $\beta_2$  agonists, and antileukotrienes was allowed. Baseline forced expiratory volume in 1 second (FEV<sub>1</sub>) had to be between 40 and 80% of predicted. Reversibility of airway obstruction was confirmed either by demonstrating a 12% or greater increase from baseline after inhaling 200  $\mu$ g of salbutamol or by demonstrating an episode with an FEV<sub>1</sub>  $\geq$  15% below the personal best value over the past 2 years. None of the patients had experienced an acute asthma attack or a lower respiratory tract infection within 4 weeks of screening.

### Study Design

The study was conducted at four centers in Belgium, the United Kingdom, and the Netherlands. Approval from the local ethics committee was obtained at each center.

After obtaining written informed consent, patients entered a 7-day run-in phase. Treatment was kept unchanged and patients were asked to complete a diary card. Only patients with a total symptom score of 10 or more (maximum, 42) and needing short-acting inhaled  $\beta_2$  agonists as rescue on two or more occasions per day were included in the study.

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Eligible patients were randomized to receive either a single dose of SCH55700 or matching placebo. Both the patients and the evaluator were blinded to treatment. Randomization numbers were centrally allocated and the treatment was prepared by a third party. Four dose groups were sequentially enrolled: SCH55700 at 0.03 mg/kg ( $n = 2$ ) or placebo ( $n = 1$ ), SCH55700 at 0.1 mg/kg ( $n = 4$ ) or placebo ( $n = 1$ ), SCH55700 at 0.3 mg/kg ( $n = 6$ ) or placebo ( $n = 2$ ), and SCH55700 at 1.0 mg/kg ( $n = 12$ ) or placebo ( $n = 4$ ). Treatment was given intravenously as a bolus for the lowest two doses and as a 30-minute infusion for the two highest doses. Subjects were confined to the unit for a period of 48 hours after dosing.

### Outcome Measures

Safety analyses were performed in the clinic 1, 6, 24, and 48 hours after dosing, as well as on Days 8, 15, 30, 60, and 90. For the 0.3- and 1.0-mg/kg groups, the follow-up was extended to include data obtained on Days 120, 150, and 180. Safety assessment included symptom check, physical examination, ECG, blood and urine analysis, as well as pulmonary function testing. The decision to move to the next dosing level was based on the safety data obtained up to Day 15 after each dose.

Assessment of the biological activity of SCH55700 included measurement of eosinophil counts in peripheral blood and induced sputum, symptom severity, lung function parameters, and physician-evaluated overall condition. Blood eosinophil counts were automated on a 2-ml heparinized blood sample. Sputum was induced and processed according to a protocol that was strictly standardized for identical use in the participating centers. A previously validated protocol was used (10). Diary records included morning and evening premedication peak flow, symptom severity rated on a 0- to 3-point scale for dyspnea, cough and wheezing, as well as the use of short-acting inhaled  $\beta_2$  agonists as rescue medication. On the basis of the review of the diary card, pulmonary function tests, and physical examination, the physician assessed the overall condition of asthma, on a 0- to 3-point scale at each visit (0 = no asthma signs or symptoms; 3 = signs or symptoms of asthma are intolerable and interfere with normal daily activities and/or sleep).

Throughout the study, blood samples were taken for measuring plasma levels of SCH55700, using a validated enzyme-linked immunosorbent assay (limit of quantitation, 120 ng/ml), and for detection of neutralizing antibodies to SCH55700 (samples were first analyzed in a biosensor assay [Biacore 2000 system; Biacore, Uppsala, Sweden] to identify samples positive for antibodies and were then run in a bioassay to measure neutralizing activity).

### Statistical Analysis

Analyses of safety and efficacy were based on the data collected from all subjects randomized into the study (intention-to-treat principles). The eight patients treated with placebo from all dose groups were combined for the purpose of summaries and analyses. The baseline period was defined as the interval of time that began 7 days before the first day of treatment. Results are expressed as means  $\pm$  SEM. Baseline variables were analyzed by a Wilcoxon rank-sum test. Pairwise treatment comparisons were based on a one-way analysis of variance model with treatment effects. Changes in blood eosinophil counts over time were evaluated by repeated measure analysis of variance. Differences were regarded as statistically significant if  $p < 0.05$ .

## RESULTS

### Biological Activity and Clinical Outcome Measures

No significant biological activity on blood eosinophil numbers was noted in subjects treated with SCH55700 at 0.03 mg/kg ( $n = 2$ ) or 0.1 mg/kg ( $n = 4$ ).

Demographics of the subjects included in the placebo groups and in the groups treated with SCH55700 at 0.3 and 1.0 mg/kg are summarized in Table 1. Baseline characteristics of the three groups were not different.

After administration of SCH55700 at 0.3 mg/kg, a short-lived decrease in circulating blood eosinophil counts was observed, varying from a mean reduction of 52.5% at 48 hours to 18.9% at Day 30, compared with baseline. The decrease obtained with

TABLE 1. SUBJECT DEMOGRAPHICS

	SCH55700		
	Placebo ( $n = 8$ )	0.3 mg/kg ( $n = 6$ )	1.0 mg/kg ( $n = 12$ )
Age, yr	39.2 $\pm$ 4.2	37.3 $\pm$ 3.9	45.9 $\pm$ 4.4
Female/male	4/4	3/3	3/9
FEV <sub>1</sub> , % predicted	52.7 $\pm$ 2.0	61.7 $\pm$ 4.6	49.3 $\pm$ 4.9
Reversibility, % from baseline	32.1 $\pm$ 9.5	36.9 $\pm$ 8.6	17.2 $\pm$ 2.3
ICS/OCS	8/2	6/1	12/3
Blood eosinophils, $\times 10^9/L$	0.45 $\pm$ 0.16	0.28 $\pm$ 0.04	0.25 $\pm$ 0.04

Definition of abbreviations: ICS = inhaled glucocorticosteroids; OCS = oral glucocorticosteroids.

SCH55700 at 1.0 mg/kg was more pronounced and remained significant up to Day 30 ( $p = 0.05$  versus placebo) (Figure 1).

At baseline, induced sputum was obtained from five patients in the placebo group, from four in the 0.3-mg/kg group, and from seven in the 1.0-mg/kg group. There was substantial variability in sputum eosinophil counts (as a percentage of total cells, 22.9  $\pm$  12.5% [mean  $\pm$  SEM], 2.6  $\pm$  0.44%, and 5.5  $\pm$  3.92%, respectively). Paired samples collected at baseline and on Day 30 were obtained from four subjects in the placebo group and from six in the 1.0-mg/kg SCH55700 group (Figure 2). No consistent changes were seen, although in three of four subjects with elevated sputum eosinophil counts a fall was seen after SCH55700 treatment.

Mean baseline FEV<sub>1</sub> was 53.0% predicted in the placebo group, 61.7% predicted in the 0.3-mg/kg group, and 48.5% predicted in the 1.0-mg/kg group. These differences were not significantly different. Twenty-four hours after administration of SCH55700 at 0.3 mg/kg a significant increase in baseline FEV<sub>1</sub> was observed, when compared with placebo ( $p = 0.019$ ) (Figure 3). Although at subsequent time points a trend persisted, neither dose of SCH55700 induced significant changes in baseline FEV<sub>1</sub>. On Day 30, the increase in baseline FEV<sub>1</sub> was 11.2% predicted from baseline in the 0.3-mg/kg group and 8.6% in the 1.0-mg/kg group compared with 4.0% in the placebo group.

Similarly, no significant changes were observed in the FEV<sub>1</sub>/FVC ratio or peak flow recordings, nor in symptom score and physician-evaluated overall condition.

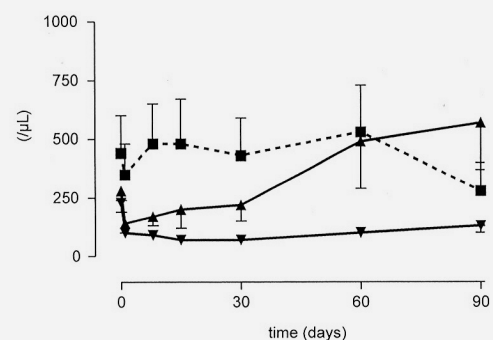
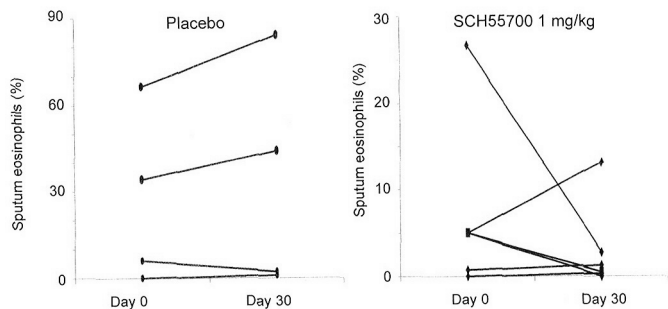


Figure 1. Blood eosinophil counts (means  $\pm$  SEM) after intravenous administration of placebo ( $n = 8$ ) (solid squares) or a single dose of SCH55700, an antihuman IL-5 antibody, at 0.3 mg/kg ( $n = 6$ ) (solid triangles) or 1 mg/kg ( $n = 12$ ) (solid inverted triangles). The decrease in the 1-mg/kg SCH55700 group remained significant up to Day 30 ( $p = 0.05$  vs. baseline).



**Figure 2.** Percentage of sputum eosinophils at baseline and 30 days after administration of placebo (n = 4) or a single dose of SCH55700 at 1 mg/kg (n = 6). No significant differences within or between groups were observed.

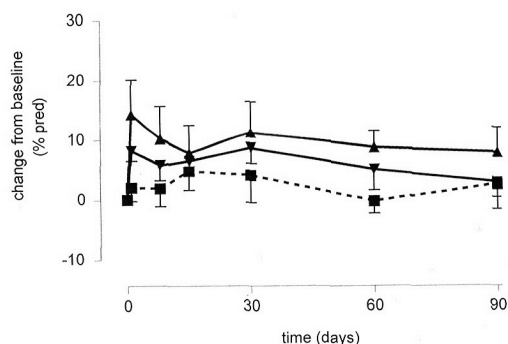
### Safety

Administration of a single dose of SCH55700 was well tolerated. No clinically meaningful changes in laboratory parameters, vital signs, or physical examinations were noted in any of the treatment groups. The most common treatment-related adverse events reported were headache (n = 6) and fatigue (n = 4). These occurred with the same frequency in the placebo group. Aggravation of asthma was noted in 3 of 12 subjects treated with 1.0 mg/kg and in 1 of 8 subjects treated with placebo (NS). None of these aggravations were classified as severe. In the 1.0-mg/kg group, two of the exacerbations were noted on Day 120 and one was noted on Day 180 of follow-up. In all cases, the patients also experienced symptoms of an infection and treatment with antibiotics was initiated. In view of the time frame and associated infectious nature, the relationship with anti-IL-5 seems unlikely.

In one subject treated with SCH55700 at 1.0 mg/kg, nonneutralizing antibodies to SCH55700 were detected in serum at Visit 10. A second patient treated with 0.03 mg/kg had a negative immunoassay, but a positive neutralization result. This indicates that the inhibition observed was probably due to serum (matrix) effects, and not to antibody formation.

### Pharmacokinetics

The plasma concentrations of SCH55700 were dose proportional. The mean SCH55700 systemic exposure (area under curve [I]) increased in a 1–2.2–5.9–23 proportion as the dose increased in a 1–3.3–10–33.3 proportion. The mean maximal concentration



**Figure 3.** Change in baseline FEV<sub>1</sub>, expressed as a percentage ( $\pm$  SEM) of the predicted value, after intravenous administration of placebo (n = 8) (solid squares), or a single dose of SCH55700 at 0.3 mg/kg (n = 6) (solid triangles) or 1 mg/kg (n = 12) (solid inverted triangles).  $p = 0.019$ , placebo versus SCH55700 (0.3 mg/kg) at 24 hours (analysis of variance).

obtained 6.9 hours after SCH55700 dosing at 1.0 mg/kg was 30.3  $\mu$ g/ml. Mean concentrations were 0.87  $\mu$ g/ml on Day 90 and 0.43  $\mu$ g/ml on Day 120. The elimination half-life ranged between 24.5 and 30.1 days.

### DISCUSSION

The present study illustrates that systemic administration of a single dose of SCH55700, a humanized anti-human IL-5 monoclonal antibody, reduces the number of circulating blood eosinophils in subjects with severe persistent asthma. In addition, a limited short-lived improvement in baseline FEV<sub>1</sub> is observed. The treatment was well tolerated.

The precise mechanisms that enhance eosinophilopoiesis in the bone marrow of individuals with asthma remain to be fully established. Inhalation of allergen is known to increase the number of IL-5-responsive eosinophil progenitor numbers in bone marrow (11). *In vivo* animal models indicate that production of IL-5 restricted to the airways is insufficient to explain this phenomenon and that either circulating IL-5 or local IL-5 production in the bone marrow is required to cause the antigen-induced increase in peripheral eosinophil counts (12, 13). In mild asthma, a moderate dose of inhaled steroids can reduce IL-5-responsive bone marrow eosinophil/basophil colony-forming units (14). The current study indicates that in severe persistent asthma even high doses of inhaled steroids do not entirely block IL-5-dependent blood eosinophilia, because SCH55700 was able to further reduce the number of peripheral blood eosinophils. This corresponds with the detection of IL-5 in the serum of steroid-treated patients with severe persistent asthma (15). The reduction of circulating eosinophil counts by SCH55700 in the current study further confirms the role of IL-5 in eosinophilopoiesis in humans and is in line with preclinical data that demonstrate an additive effect with steroids in reducing allergen-induced eosinophil recruitment (8).

Of particular interest is the prolonged effect of a single dose of anti-IL-5 on circulating eosinophil counts. A reduction to values in the normal range up to 30 days after dosing was found. A similar long-term reduction in blood eosinophil counts was obtained with another humanized anti-human IL-5 (SB240563) in non-steroid-treated patients with mild asthma (16). *In vivo* animal data indicate that these prolonged effects are primarily attributable to the pharmacokinetic properties of the antibody, and not to any untoward immunomodulatory effect (17, 18). The half-life of SCH55700 in this study was about 30 days, as normally expected for a native IgG4 immunoglobulin. Plasma levels up to Day 120 remained above 0.4  $\mu$ g/ml after the 1.0-mg/kg dose, suggesting that this could represent a threshold value of biological activity.

Administration of a single dose of SCH55700 in severe persistent asthma also proved safe. *In vitro* data indicate that, depending on their structure and concentration relative to the cytokine, some monoclonal anti-IL-5 antibodies can enhance instead of inhibit IL-5 activity (19). This effect is probably caused by binding of the monoclonal antibody to two IL-5 molecules. As IL-5 is a homodimer, the cytokine can still engage the IL-5 receptor via the remaining noninhibited receptor-binding site, the antibody thus facilitating simultaneous interaction of two IL-5 molecules with two IL-5 receptors. This concern was abated by the observation that in the present study SCH55700 neither increases circulating eosinophil counts nor causes any deterioration in asthma severity. It would therefore appear that the structure of SCH55700 is such that at the doses tested, it does not potentiate IL-5-mediated effects, confirming previous *in vitro* data. It needs to be recalled that the present data do not allow ascertainment of the safety of long-term suppression of eosino-

phils in humans. Eosinophils typically increase in response to parasite infections, but on the basis of *in vivo* animal data, it is unclear whether eosinophils actually protect against tissue-dwelling parasites (20). It is similarly unclear to what extent eosinophils are important in tumor surveillance. Some studies suggest a protective antitumor response whereas other studies link eosinophils to enhanced tumor spread (21–23).

Despite the profound long-lasting reduction in blood eosinophil counts, a single administration of SCH55700 in this group of steroid-treated patients with severe persistent asthma had little effect on clinical indices of disease severity. Only a short-lived transient increase in baseline FEV<sub>1</sub> was noted in the group treated with SCH55700 at 0.3 mg/kg. Several elements could contribute to this observation. First, the study was primarily a safety study and was not powered to detect clinical efficacy, especially as we conducted this study in the context of severe persistent asthma. The reason to do so was largely based on the argument that patients with severe persistent asthma are the prime candidates for this novel form of treatment. In view of the already mentioned potential dose-related enhancement of IL-5 activity by circulating anti-IL-5 antibodies, it is important to address the safety of biological compounds such as the current monoclonal antibodies in the patient population that is being considered for treatment. There is increasing evidence that the pathogenesis of severe persistent asthma shows considerable heterogeneity. It has been suggested that within the severe asthma phenotype, distinction can be made between an eosinophil-driven subgroup and a neutrophil-driven subgroup (24). The patients included in this study were not selected for eosinophils in sputum or blood. A second element that could explain the limited clinical effect relates to dose and the dosing frequency of the monoclonal antibody. Circulating eosinophils were reduced by about 85%, but were not totally abolished. Similar data emerge from a study with SB240563. A single administration of SB240563 at 10 mg/kg to patients with mild atopic asthma reduced circulating eosinophils to a comparable degree as SCH55700 at 1 mg/kg in the present study but did not influence the allergen-induced early or late asthmatic response or methacholine responsiveness (16). The remaining eosinophils might be sufficient to maintain airway inflammation. Although IL-5 has been shown to play an important role in the homing response of eosinophils to chemotactic stimuli, it cannot be excluded that even in the presence of anti-IL-5 other chemotactic stimuli in tissues are sufficiently strong to maintain large numbers of tissue-dwelling eosinophils despite the reduction in blood counts. There was a tendency toward reduction of eosinophils in induced sputum after administration of SCH55700 at 1 mg/kg. However, the variability was too large and the number of samples too limited to allow for any firm conclusion as to the effect of antibody on airway tissue eosinophil numbers. It was shown in a biopsy study that three monthly infusions of SB240563, although reducing BAL eosinophilia by more than 90%, decreased mucosal eosinophils in the airways only by 55%. No clinical effects were noted (25). An additional point that needs to be considered is that prolonged IL-5 antagonism might be required to inhibit the direct effects of IL-5 on airway smooth muscle (26, 27).

Finally, although circumstantial evidence links IL-5 and eosinophils to the pathophysiology of asthma, this remains to be proved. Human and *in vivo* animal studies clearly illustrate a dissociation between the eosinophil numbers and changes in airway responsiveness (28–32). Similarly, eosinophil numbers in biopsies or sputum do not consistently correlate with baseline FEV<sub>1</sub> or bronchial hyperresponsiveness in subjects with mild asthma (33, 34). To further address these important questions, well-powered clinical trials, aimed at assessing the efficacy of anti-IL-5 antibodies, need to be performed (35).

In conclusion, the present dose-rising study indicates that SCH55700 is a biologically active anti-human IL-5 antibody that can be safely used in asthma. Further studies will allow evaluation of the efficacy of this treatment modality in asthma and other eosinophilic disorders.

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