

Anti-interleukin-5 (mepolizumab) therapy for hypereosinophilic syndromes

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Background: IL-5 is a cytokine critically involved in regulating several aspects of eosinophils including their production, activation, and tissue recruitment. As such, IL-5 may be involved in the pathogenesis of hypereosinophilic syndromes, a group of poorly treated diverse disorders characterized by sustained peripheral blood and/or tissue eosinophilia.

Objective: We aimed to assess the safety and efficacy of a humanized blocking monoclonal antibody against IL-5 (mepolizumab) in patients with several forms of hypereosinophilic syndromes.

Methods: We performed an open-label trial of anti-IL-5 in which 3 intravenous doses (10 mg/kg, maximum 750 mg) were administered at 4-week intervals to 4 patients with hypereosinophilic syndromes (defined by peripheral blood and/or tissue eosinophilia). The effects of treatment on safety, eosinophil levels (in peripheral blood and/or diseased tissue), pulmonary function, and quality of life were measured over a 28-week period.

Results: Anti-IL-5 was well tolerated in all patients and lowered peripheral blood eosinophil counts despite ongoing systemic glucocorticoid therapy. The decline in circulating eosinophil counts was sustained for at least 12 weeks after the last dose of anti-IL-5. In addition, anti-IL-5 improved clinical and quality of life measurements. In one patient with striking tissue eosinophilia (eosinophilic esophagitis), anti-IL-5 resulted in a 10-fold reduction in tissue eosinophil levels.

Conclusions: These results suggest that anti-IL-5 is safe, effective in lowering eosinophil levels, and has potential glucocorticoid-sparing effects in patients with a variety of hyper-

eosinophilic syndromes. As such, anti-IL-5 may have significant therapeutic potential for hypereosinophilic syndromes. (*J Allergy Clin Immunol* 2004;113:115-9.)

Key words: Eosinophils, esophagitis, IL-5, cytokine, hypereosinophilic, humanized antibody

Hypereosinophilic syndromes (HES) encompass diverse disorders defined by the accumulation of a large number of eosinophils in the blood and/or tissues. These disorders include the idiopathic hypereosinophilic syndrome (IHES), which is characterized by elevated levels of blood eosinophils (>1500 eosinophils/ μ L) and the involvement of multiple organs as well as tissue-specific disorders such as eosinophilic esophagitis (EE).¹⁻⁴ The IHES typically involves the heart, lung, and skin, whereas EE is part of a series of gastrointestinal-specific eosinophilic disorders that appear to be occurring with increasing incidence.⁴ Patients with EE usually have symptoms that mimic gastroesophageal reflux disease (GERD), but the disease is primarily resistant to typical anti-GERD therapy.⁵ Accordingly, the pathogenesis appears to be markedly different from GERD in that the esophagus has much higher levels of eosinophils compared with GERD, pH probes are typically normal in EE, and there is a higher prevalence of the male sex in EE. Additionally, EE is strongly associated with atopic disease; most patients have IgE sensitization to a variety of food and inhaled allergens and coexisting asthma.⁴

Interleukin (IL)-5 is a cytokine that has been demonstrated to regulate a variety of processes associated with eosinophils.⁶ These include antigen-induced eosinophilia, bone marrow release of eosinophils, eosinophil tissue survival, and eosinophil activation. Based on these properties, primarily demonstrated in rodents, neutralizing antibodies against murine IL-5 were generated and shown to be safe and effective in lowering eosinophil blood and tissue levels in models of asthma and parasitic infection.⁷ Subsequently, humanized neutralizing antibodies against human IL-5 have been developed, and early clinical trials in patients with asthma have shown that anti-IL-5 is safe and effective at lowering blood and sputum eosinophil levels but does not produce the desirable effect on asthma outcome measures.⁸⁻¹⁰ Anti-IL-5

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Abbreviations used

CTC:	Clinical toxicity criteria
EE:	Eosinophilic esophagitis
FIP1L1:	Fip1-like 1
GERD:	Gastroesophageal reflux disease
HES:	Hypereosinophilic syndrome
HPF:	High-power field
IHES:	Idiopathic HES
PDGFRA:	Platelet-derived growth factor receptor- α
QOL:	Quality of life

may be particularly useful for the treatment of diseases such as HES, which generally have much higher levels of eosinophils in the blood and/or tissue compared with asthma.¹ Notably, IL-5 has been shown to be overproduced in some patients with IHES and EE^{11,12} and involved in the pathogenesis of experimental EE in mice.¹³ At present, patients with IHES are often treated with a variety of potentially toxic and often only partially effective medications such as glucocorticoids, hydroxyurea, and interferon- α . Patients with EE are often treated with allergen avoidance, topical or systemic glucocorticoids, and a variety of anti-GERD medications, but these approaches are often not successful. More recently, the tyrosine kinase inhibitor imatinib mesylate has been shown to be useful in certain subsets of HES, particularly the myeloproliferative variants resulting from the fusion of the platelet-derived growth factor receptor- α (PDGFRA) and Fip1-like 1 (FIP1L1) genes.^{14,15} Based on the need to develop more effective and safe treatments for HES, we aimed to test the safety and efficacy of anti-IL-5 in these diseases. Because the effect of anti-IL-5 in patients with HES had not been previously studied and it remained possible that anti-IL-5 could be toxic in patients with markedly elevated levels of eosinophils, we designed an open-label phase I/II trial, designed to primarily assess the safety of this new therapeutic approach.

METHODS

After local institutional review board and FDA approval and informed consent was obtained, patients with HES involving peripheral blood and/or tissue eosinophilia (18 to 65 years old) were monitored (by complete blood counts and physical examination) at 2- to 4-week intervals for a period of 28 weeks. Patients were also evaluated by pulmonary function testing, electrocardiograms, and echocardiograms at weeks 0, 8, and 20. It is important to note that the entry criteria for this study permitted enrollment of patients with severe eosinophilic tissue disease (in the absence of peripheral blood eosinophilia). As such, for the purposes of this study, patients with HES included patients with IHES as well as EE. Patients with severe end-organ damage defined as grade III/IV toxicity on the National Cancer Institute Clinical Toxicity Criteria (CTC) were excluded except for those with stable congestive heart failure. During the run-in period (weeks 0 to 8), patients with a history of IHES had their antieosinophil therapy (eg, glucocorticoids) reduced until eosinophil levels increased 2-fold over baseline and/or until the absolute eosinophil levels were >750 cells/ μ L. The antieosinophil therapy was gingerly reduced at 2-week intervals, on a case-by-case basis, to avoid exacerbation of clinical symptoms. This provided reassurance

that the disease was not in remission and allowed the development of a moderate eosinophilia for assessment of the efficacy of anti-IL-5. At 8 weeks, patients were intravenously treated with anti-IL-5 (mepolizumab [SB-240563], provided by GlaxoSmithKline, Research Triangle Park, NC) at a dose of 10 mg/kg (maximum, 750 mg), and this was repeated twice at 4-week intervals. After week 8, patients did not have their conventional (preexisting) antieosinophil therapy modified. At week 8, before the first infusion, plasma IL-5 levels and mononuclear cell production of IL-5 (48-hour supernatant after PHA stimulation) was determined (by OptEIA ELISA kit, according to the manufacturer [BD Biosciences Pharmingen, San Diego, Calif]). The safety of anti-IL-5 was determined by recording all adverse events and scoring them according to the CTC. The National Cancer Institute CTC was developed for use in adverse drug experience reporting to the FDA and for publications (see <http://ctep.info.nih.gov/reporting/ctc.html> for complete information on the CTC). Quality of life (QOL) parameters were measured at 2- to 4-week intervals, using the validated Short Form Health Questionnaire SF-36, and reported as general health scores.¹⁶

RESULTS

Patient 1 was a 48-year-old woman with a 10-year history of IHES with biopsy-proven involvement of the lungs, gastrointestinal tract, and skin (eosinophilic cellulitis). Her treatment had been primarily oral methylprednisolone and methotrexate with previous trials of hydroxyurea, interferon- α , and phototherapy. At the time of the study, her gastrointestinal symptoms were inactive and her main target organs were the skin and lungs. During the initial 8 weeks of the protocol, she had an exacerbation of cellulitis requiring escalation of therapy (methylprednisolone and methotrexate). Because of the medical need to increase therapy, the blood eosinophil count at week 8 was low.

Patient 2 was a 55-year-old man with a 2-year history of IHES primarily involving biopsy-proven eosinophilic pneumonia, cellulitis (inactive at the time of study), and sinusitis/nasal polyposis. He had been treated primarily with oral prednisone and hydroxyurea. His past medical history is notable for atopic asthma and rhinitis and myocardial infarction at the age of 40 years. During weeks 0 to 8, his antieosinophil therapy was not adjusted because his blood eosinophil counts were already >750 cells/ μ L.

Patient 3 was a 40-year-old woman with a 3-year history of IHES who initially presented with biopsy-proven acute eosinophilic cardiomyopathy (14 days after commencing L-tryptophan); after stabilization, she had low-grade congestive heart failure (New York Heart Association class I; her ejection fraction was 50% at baseline) and asthma. She has been maintained on oral prednisone and cardiac medications. During weeks 0 to 8, her antieosinophil therapy (prednisone) was reduced by a total of 2.5 mg every other day, which allowed her blood eosinophil counts to increase >750 cells/ μ L.

Patient 4 was an 18-year-old man who presented with a diagnosis of EE at 17 years of age. He had a lifelong history of dysphagia, progressive inability to swallow solid foods, and was primarily maintained on a liquid diet. His initial upper endoscopy revealed severe esophageal narrowing (stricture), marked esophageal

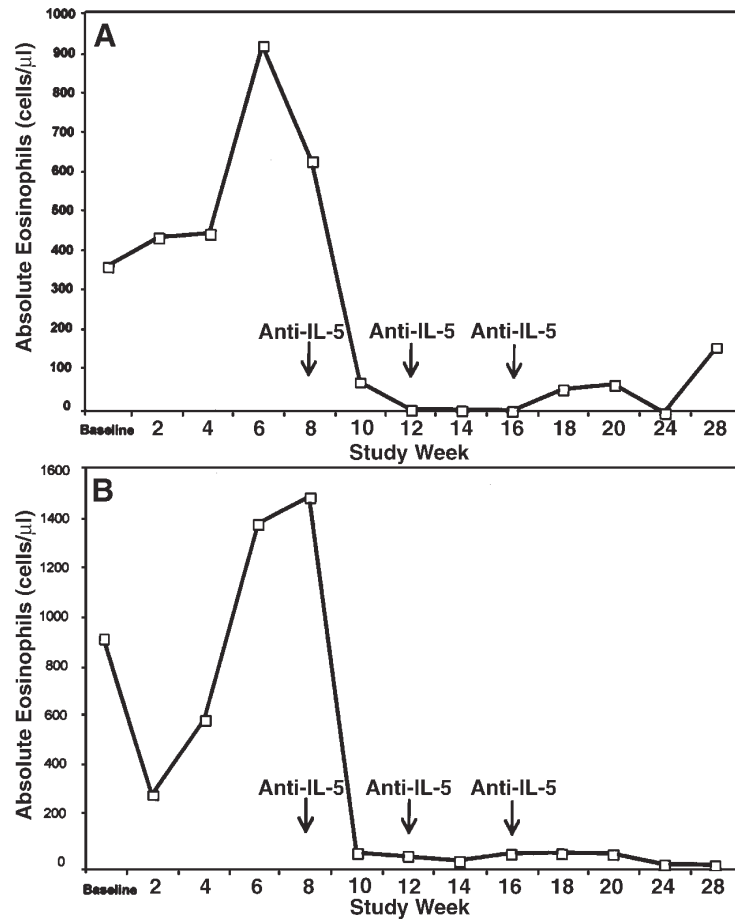


FIG 1. Effect of anti-IL-5 on eosinophil blood counts. Peripheral blood eosinophil counts in patient 2 (**A**) and patient 3 (**B**) are shown. Arrows indicate points of intervention with anti-IL-5 therapy.

TABLE I. Summary of patient data

Patient	Medication at wk 8 (daily mg dose)	Eos count* (wk 0–8)	Eos count* (wk 10–20)	Eos count* (wk 28)	QOL pre/post†	FEV ₁ pre/post‡	Plasma IL-5§	Mononuclear cell IL-5
1	Methylprednisolone (8) MTX (17.5)	300	113	105	20/32	65/70	3	147
2	Prednisone (10) HXU (1000)	1500	58	20	27/40	79/90	4	258
3	Prednisone (5)	924	69	170	57/67	79/90	6	112
4	None	600	171	50¶	42/37	101/102	39	402

*Eosinophil values represent maximum absolute eosinophil count (cells/μL) during the stated time interval.

†QOL parameters from the validated Short Form Health Questionnaire SF-36 are recorded as general health scores at weeks 8 (pre) and 20 (post). Normal mean range is 72 to 76.

‡FEV₁ measurements (prebronchodilator) are at weeks 8 (pre) and 20 (post).

§Plasma levels of IL-5 are presented as pg/mL; normal values are <7 pg/mL.

||Levels of IL-5 secreted in mononuclear cell cultures stimulated with PHA are expressed as pg/mL; normal values in our laboratory are 106 ± 52 pg/mL (mean ± SD, n = 11).

¶At week 32.

MTX, Methotrexate weekly; HXU, hydroxyurea.

eosinophilia, and epithelial hyperplasia. Skin testing was positive to a variety of foods, but dietary eliminations, topical fluticasone therapy (MDI 880 μg swallowed BID), and oral prednisone all failed to improve his symptoms and endoscopic findings. He did not have a history of peripheral blood eosinophilia.

Anti-IL-5 effectively reduced peripheral blood eosinophilia in all patients. Table I summarizes the blood eosinophil counts in all patients by showing the maximum level of eosinophils before and after anti-IL-5. Fig 1, A and B, depicts the peripheral blood eosinophil counts in patients 2 and 3, respectively, during the entire

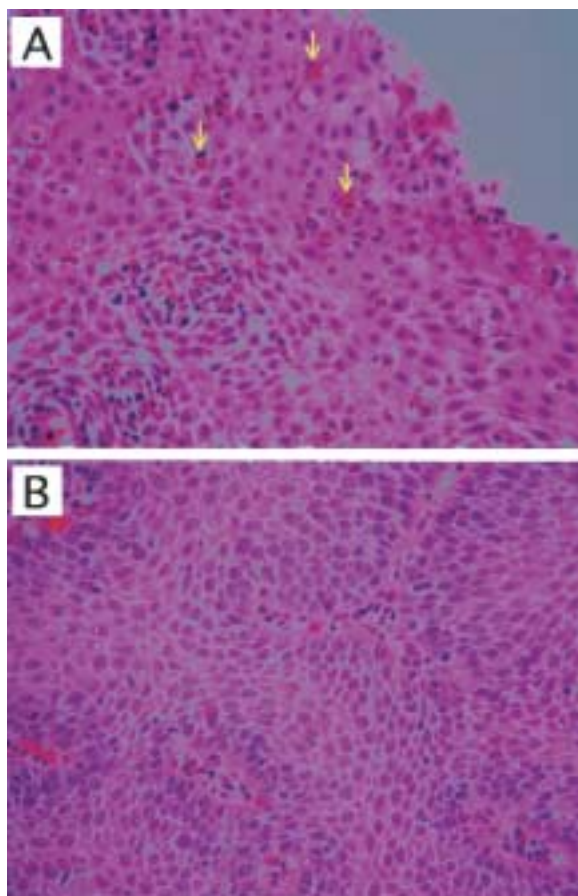


FIG 2. Histologic analysis of esophageal biopsy specimens before and after anti-IL-5. Representative sections from esophageal biopsies taken before (**A**) and after (**B**) anti-IL-5 were stained with hematoxylin and eosin. **A**, Biopsy specimen from the proximal esophagus shows numerous eosinophils throughout the epithelium, including at the luminal surface. Representative eosinophils are indicated with arrows. **B**, Only scattered intraepithelial eosinophils are present. Original magnification $\times 200$.

study. After anti-IL-5, eosinophil levels plummeted for the duration of the study. Patients reported progressive improvements in their specific symptoms during the duration of the study. For example, patient 1 had improvement in skin pruritis and induration; patient 2 reported remarkable improvement in nasal congestion, polyposis on physical examination, constitutional symptoms, and exercise tolerance. There were also improvements noted in FEV₁ measurements and objective QOL measurements (Table I). Notably, patient 4 reported a remarkable advancement in his diet; for the first time in many years he was able to advance his diet and was now able to swallow solid foods. In addition, before the study, he was vomiting 3 to 4 times per week because of dysphagia, but after 3 doses of anti-IL-5, this was no longer occurring. Subsequently, a repeat endoscopic evaluation at week 20 revealed that the esophagus was grossly less narrow and inflamed; this was confirmed by an improved

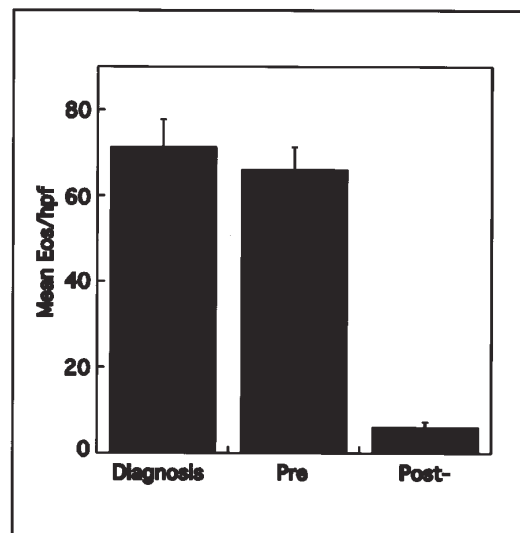


FIG 3. Effect of anti-IL-5 on esophageal eosinophil counts. Values are mean $\pm$ SD of the number of eosinophils in all HPF per specimen at time of diagnosis, before (week 8) and after (week 20) anti-IL-5. There was a statistically significant difference in eosinophil levels between post-anti-IL-5 and time of diagnosis ($P < .01$) and week 8 ($P < .01$) values as determined by Student *t* test.

esophagram (data not shown). Importantly, his esophagus contained markedly reduced levels of tissue eosinophils compared with his pre-anti-IL-5 biopsy specimen. Representative photomicrographs of his esophageal biopsies are shown in Fig 2. Notably, his pre-anti-IL-5 biopsy samples, consistent with biopsy samples taken on 3 prior occasions (data not shown), had abundant eosinophils and free eosinophil granules detected throughout the hyperplastic mucosa (Fig 2, A). However, after anti-IL-5 therapy, only scattered eosinophils were present in the mucosa (Fig 2, B). Quantitative analysis of his esophageal biopsy specimens revealed a statistically significant, >10 -fold decrease in the mean number of tissue eosinophils (Fig 3). There was also a decrease in the maximum number of eosinophils detected in any high-power field (HPF); before therapy (week 8), the maximum eosinophil count exceeded 200 cells/HPF, whereas after IL-5, the maximum number was 38 (and this was only noted in a surface exudate). Responsiveness to anti-IL-5 occurred in the patients with both normal and elevated production of IL-5 (Table I). There were no drug-associated adverse events except for fatigue associated with the first 2 infusions in patient 2 and recurrent infusion-associated headaches in patient 4, who had a history of headaches.

DISCUSSION

In summary, we have shown that anti-IL-5 appears to be safe in 4 patients with diverse manifestations of HES. In addition, several disease parameters appear to be improved by anti-IL-5 therapy, including peripheral blood

eosinophil counts in all patients, FEV₁ measurements, a variety of clinical symptoms, QOL measurements, and tissue eosinophilia in 1 patient. However, since this study is an open-label, noncontrolled trial, definitive proof that anti-IL-5 is responsible for these improvements cannot be ensured. Nevertheless, it is notable that 3 of these patients (1 through 3) normally maintain an abnormal level of blood eosinophils, especially at the relatively low doses of antieosinophil therapy that they were taking between weeks 8 and 28. Furthermore, the decline in blood eosinophils was noted at week 10 (right after anti-IL-5) and was maintained for the duration of the study, strongly supporting a causal association between anti-IL-5 and the decrease in blood eosinophilia.

The ability of anti-IL-5 to lower peripheral blood eosinophilia in the 3 patients already taking concurrent glucocorticoids (Table I) suggests that anti-IL-5 may have a steroid-sparing effect in this disease. The profound decline in tissue eosinophilia in patient 4 compared with the only modest decrease of lung eosinophils in asthmatic patients after anti-IL-5⁹ suggests that this agent may provide relatively more tissue effectiveness in patients with HES than with asthma. At present, patients with EE and other eosinophilic gastrointestinal disorders have limited medical options (except for glucocorticoids).⁴ This highlights the importance of further examining the efficacy of anti-IL-5 for this series of poorly treated gastrointestinal diseases. Although only one of the studied patients had detectable increases in IL-5 production (Table I), the ability of anti-IL-5 to lower eosinophils in all patients supports a contributory role for IL-5 in regulating eosinophilia despite the primary pathogenesis of HES. It will be interesting to determine the utility of anti-IL-5 in patients with HES who harbor the FIP1L1-PDGFR α fusion gene; notably, patients 1 through 3 did not have a detectable FIP1L1-PDGFR α gene fusion as assessed by nested reverse-transcriptase PCR from blood RNA (Y. Yamada and M. E. Rothenberg, unpublished results). Collectively, these preliminary results suggest that anti-IL-5 may have clinical utility for HES, raising the need to further evaluate this compound for these disorders.

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