

Anti-TNF- α (infliximab) therapy for severe adult eosinophilic esophagitis

To the Editor:

Eosinophilic esophagitis (EE) is a chronic inflammatory disorder of the esophagus¹ with an increasing prevalence.²⁻⁴ EE is clinicopathologically characterized by proton pump inhibitor-refractory esophagus-related symptoms in combination with a dense esophageal eosinophilia.⁴ In addition to T_H2 cytokines,⁵ TNF- α is upregulated in EE⁶ and highly expressed by epithelial cells of the esophagus in patients with active EE.⁵ These results point to the possibility that TNF- α plays a critical role in the pathogenesis of EE.

Infliximab, a chimeric IgG1 mAb, is a potent inhibitor of TNF- α . Blocking TNF- α activity with infliximab demonstrated high efficacy in the treatment of several chronic inflammatory diseases, including Crohn's disease,⁷ rheumatoid arthritis,⁸ and asthma.⁹ We conducted a prospective T1 translational study with the purpose of evaluating the efficacy of infliximab monotherapy in 3 male adult patients with severe, corticosteroid-dependent EE. The primary end point was to determine the effect of infliximab on the esophageal eosinophilic infiltration. Secondary end points were to determine the effects of infliximab on symptoms, endoscopic alterations, and relevant biomarkers. Inclusion criteria were adult patients with isolated active EE and inadequate responses to conventional treatment. Active EE was defined clinically as having symptoms of dysphagia during almost every intake of solid food when not receiving anti-inflammatory therapy and histologically by a peak cell density of greater than 24 eosinophils/high-power field ($\times 400$) in the esophageal mucosa. The efficacy of the study medication was assessed before treatment, after a 4-week run-in period immediately before the first infusion of infliximab, and after treatment 4 weeks after the second infusion. The study was conducted with the approval of the local ethics committee, and all patients provided written informed consent.

Symptoms were assessed by using a slight modification of the previously published scoring system that focuses on esophageal symptoms.¹ Upper endoscopy was performed before treatment and again 4 weeks after the second infusion. The endoscopic findings were classified by using a simple, previously published overall score.¹ At least 4 biopsy specimens were sampled endoscopically from both the proximal half and the distal half of the esophagus. On each esophageal biopsy specimen, all levels were surveyed, and the eosinophils in the most densely infiltrated area were counted in 5 consecutive high-power fields. Data were expressed as maximum and mean eosinophils per high-power field (Zeiss Axiophot, Plan-Neofluar 40; Carl Zeiss, Peabody, Mass; ocular magnification $\times 10$; area of microscopic field, 0.3072 mm²). Infiltration of inflammatory cells and inflammatory mediators previously implicated in EE pathogenesis⁵ was assessed by means of immunofluorescence analysis. Indirect immunostaining was carried out on 4- μ m-thick, paraformaldehyde-fixed, paraffin-embedded esophageal sections by using the following primary antibodies: anti-eosinophil cationic protein (anti-ECP; EG1, Pharmacia & Upjohn Diagnostics AB, Uppsala, Sweden), anti-CD3 (DakoCytomation, Glostrup, Denmark), anti-tryptase (DakoCytomation), anti-TNF- α (Novus Biologicals, Inc, Littleton, Colo), and anti-eotaxin-3 (R&D Systems Europe Ltd,

Abingdon, United Kingdom). Serum TNF- α levels were determined by using a Cytometric Bead Array, Assay, and serum ECP levels were measured by using the ImmunoCap System for ECP. The clinical characteristics of the recruited patients with EE are provided in Table I. All patients had previously pursued several therapies, particularly high dosages of proton pump inhibitors, topical corticosteroids, dilations, or even long-term therapy with systemic corticosteroids. Patients 1 and 3 were almost free of symptoms during maintenance therapy with topical fluticasone, but immediately after cessation of the medication, symptoms reappeared. Patient 2 had been receiving systemic corticosteroid treatment for the past 8 years and needed at least 10 mg of prednisone per day for symptom control. Patients 2 and 3 had previously undergone repeated dilations for the treatment of strictures.

All ongoing treatments for EE were discontinued, and during a 4-week run-in period, patients were left without any specific antieosinophil therapy. The patients then received 2 infusions of infliximab, each containing 5 mg/kg body weight, at weeks 0 and 2. The infusions were given according to the manufacturer's instructions and precautions. Symptoms showed a decreasing trend in patients 1 and 3 and an increasing trend in patient 2. At baseline, 2 patients had severe and 1 patient had moderate abnormalities on endoscopy, including extensive white exudates, furrows, and corrugated rings. The stomach and duodenum were unremarkable in all subjects, and no signs of gastroesophageal reflux disease were found. Four weeks after the application of the 2 infliximab infusions, the endoscopic abnormalities appeared almost identical to the pretreatment examination.

Before treatment, the mean values of 40 analyzed high-power fields of eosinophils in the esophageal tissue were 102.5, 157.1, and 44.9 cells per high-power field. Four weeks after the 2 infliximab infusions, the corresponding values were 72.0, 237.5, and 54.2 cells per high-power field, respectively (Table I). In agreement with histologic data, eosinophil numbers, as assessed by means of an anti-ECP immunofluorescence analysis, decreased in patients 1 and 3 and increased markedly in patient 2. T-cell numbers (CD3⁺ cells) occurred at significantly lower numbers in patients 1 and 3 and showed a decreasing trend in patient 2. In contrast, mast cell numbers (tryptase-positive cells) did not markedly change in any patient under the influence of a TNF- α blockade. We also investigated the expression of TNF- α and eotaxin-3, which were both highly expressed at baseline. During the TNF- α blockade, the expression of TNF- α decreased markedly in patient 1, remained unchanged at a high level in patient 2, and decreased moderately in patient 3 (Fig 1 and Table I). Eotaxin-3 expression did not change in patients 1 and 3, whereas patient 2 experienced a slight increase. TNF- α levels in serum were not detectable either before or after treatment (data not shown).

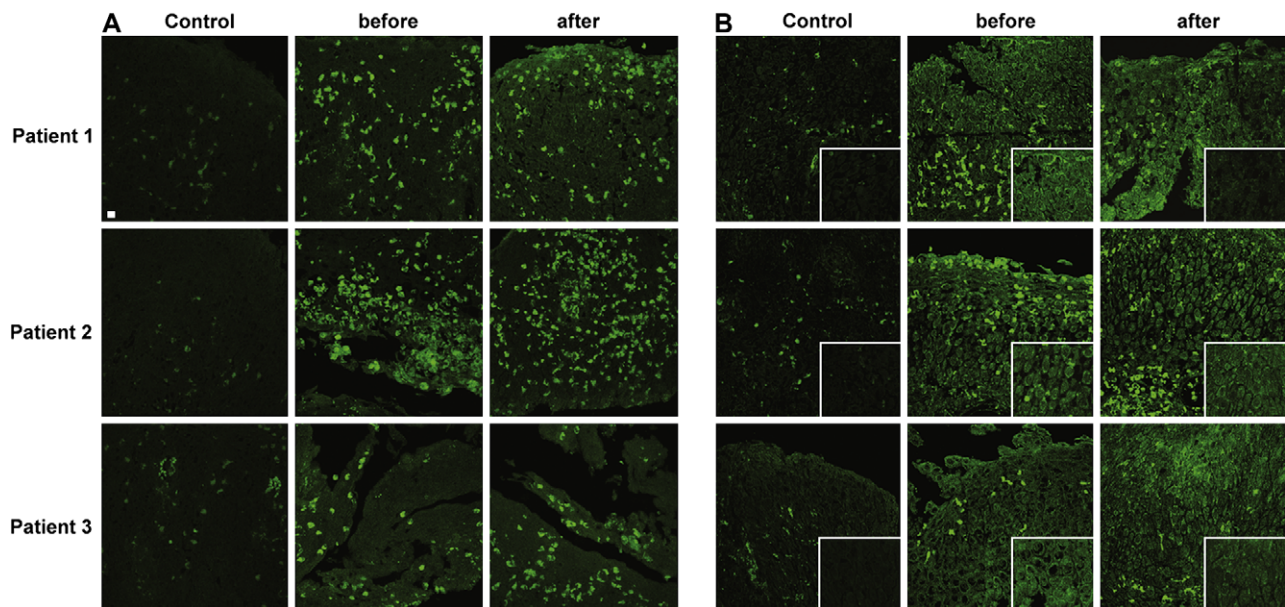
All 3 patients tolerated treatment with infliximab well, and no relevant adverse events occurred. Additionally, the follow-up after treatment was uneventful; in particular, no rebound flare-ups were observed, and the response to the reinstated corticosteroid therapy did not change after the infliximab administration.

In summary, we report the first translational study analyzing the efficacy of a TNF- α blockade in adult patients with severe, corticosteroid-dependent EE. These data demonstrate that infliximab, in a standard induction dosage schedule, is not able to

TABLE I. Clinical characteristics and both symptoms and inflammatory marker analysis in esophageal tissues in response to infliximab treatment

	Patient 1		Patient 2		Patient 3	
Patient characteristics						
Age (y)	34		39		41	
Age at EE onset (y)	28		26		29	
Duration of EE (y)	6		13		12	
Blood eosinophils (/mm ³)	96		165		361	
Atopic diseases	AR, AC, OAS		AR, AC		None	
Total IgE level (kU/L)	111		73		66	
Baseline esophageal peak (cells/hpf)	269		311		79	
	Before	After	Before	After	Before	After
Response to treatment						
Symptom score	10.25	9.25	6.50	7.25	9.00	7.00
Eosinophil load (histology)						
Mean (cells/hpf)	102.5	72.0	157.1	237.5	44.9	54.2
SD	77.1	48.6	61.9	62.8	18.5	28.9
ECP ⁺ cells						
Mean (cells/hpf)	117.4	95.8	103.8	179.9	51.6	41.8
SD	18.2	20.6	12.0	17.8	3.7	3.7
CD3 ⁺ cells						
Mean (cells/hpf)	57.8	32.0	59.4	53.9	67.9	35.6
SD	9.7	5.6	9.2	12.4	11.6	7.6
Tryptase-positive cells						
Mean (cells/hpf)	28.7	22.9	28.4	32.3	8.2	13.1
SD	12.9	13.9	8.8	17.1	4.7	5.6
TNF-α expression (score)*	+++	+	+++	+++	+++	++
Eotaxin-3 expression (score)*	++	++	++	+++	++	++

AR, Allergic rhinitis; AC, allergic conjunctivitis; OAS, oral allergic syndrome.

*Histologic grading system: TNF- α and eotaxin-3 stainings of epithelial cells were semiquantitatively evaluated by at least 2 investigators: -, no positive cells; +, weak staining; ++, clear positive staining; +++, strong positive staining).**FIG 1.** Identification of eosinophils (ECP⁺ cells) and TNF- α expression in esophageal epithelium of patients with EE before and after infliximab treatment, as determined by means of immunofluorescence analysis. **A**, ECP⁺ cells ($\times 400$ magnification). **B**, TNF- α ⁺ cells ($\times 400$ magnification). *Lower right corner*, Magnified view. Both panels show representative results from all patients. A quantitative analysis is shown in Table I.

induce a resolution of the eosinophilic tissue infiltration, nor does it markedly reduce the resulting symptoms. However, detailed analysis shows that infliximab evoked a heterogeneous reaction inasmuch as one of 3 patients achieved a partial response, one

experienced a mild flare-up, and the third was refractory to the TNF- α blockade.

The value of this study is limited by its design as an open-label, nonrandomized pilot trial with a small patient number. Further

studies working with alternative infliximab application modes and dosages are therefore needed to develop therapeutic options for patients with severe refractory EE.

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REFERENCES

1. Straumann A, Spichtin HP, Grize L, Bucher KA, Beglinger C, Simon HU. Natural history of primary eosinophilic esophagitis: a follow-up of 30 adult patients for up to 11.5 years. *Gastroenterology* 2003;125:1660-9.
2. Noel RJ, Putnam PE, Rothenberg ME. Eosinophilic esophagitis. *N Engl J Med* 2004;351:940-1.
3. Straumann A, Simon HU. Eosinophilic esophagitis: escalating epidemiology? *J Allergy Clin Immunol* 2005;115:418-9.
4. Furuta GT, Liacouras CA, Collins MH, Gupta SK, Justinich C, Putnam PE, et al. First International Gastrointestinal Eosinophil Research Symposium (FIGERS) Subcommittees. Eosinophilic esophagitis in children and adults: a systematic review and consensus recommendations for diagnosis and treatment. *Gastroenterology* 2007;133:1342-63.
5. Straumann A, Bauer M, Fischer B, Blaser K, Simon HU. Idiopathic eosinophilic esophagitis is associated with a Th2-type allergic inflammatory response. *J Allergy Clin Immunol* 2001;108:954-61.
6. Blanchard C, Wang N, Stringer KF, Mishra A, Fulkerson PC, Abonia JP, et al. Eotaxin-3 and a uniquely conserved gene-expression profile in eosinophilic esophagitis. *J Clin Invest* 2006;116:536-47.
7. Targan SR, Hanauer SB, van Deventer SJ, Mayer L, Present DH, Braakman T, et al. A short-term study of chimeric monoclonal antibody cA2 to tumor necrosis factor alpha for Crohn's disease. Crohn's disease cA2 Study Group. *N Engl J Med* 1997;337:1029-35.
8. Elliott MJ, Maini RN, Feldmann M, Kalden JR, Antoni C, Smolen JS, et al. Randomised double-blind comparison of chimeric monoclonal antibody to tumour necrosis factor alpha (cA2) versus placebo in rheumatoid arthritis. *Lancet* 1994;344:1105-10.
9. Erin EM, Leaker BR, Nicholson GC, Tan AJ, Green LM, Neighbour H, et al. The effects of a monoclonal antibody directed against tumor necrosis factor-alpha in asthma. *Am J Respir Crit Care Med* 2006;174:753-62.

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