Multiple Doses of Intravenous Interleukin 10 in Steroid-Refractory Crohn's Disease

SANDER J. H. VAN DEVENTER,* CHARLES O. ELSON,[†] and RICHARD N. FEDORAK[§] for the CROHN'S DISEASE STUDY GROUP

*Academic Medical Center, Amsterdam, The Netherlands; [†]Division of Gastroenterology, University of Gastroenterology, University of Alabama, Birmingham, Alabama; and [§]Division of Gastroenterology, University of Alberta, Edmonton, Alberta, Canada

Background & Aims: Interleukin 10 (IL-10) is a cytokine with immunosuppressive and anti-inflammatory activities. Gene-targeted IL-10-deficient mice develop a chronic intestinal inflammatory disease that is reminiscent of Crohn's disease. The present double-blind randomized multicenter trial was designed to evaluate the safety, tolerance, and pharmacokinetics of IL-10 in Crohn's disease. **Methods:** Forty-six patients with active steroid-resistant Crohn's disease were treated with one of five doses of recombinant human IL-10 (0.5, 1, 5, 10, or 25 µg/kg) or placebo administered once daily by intravenous bolus injection over 7 consecutive days. **Results:** Treatment was safe and well tolerated, and no evidence for IL-10 accumulation was observed at the end of the treatment period. At the end of the study, Crohn's disease activity scores were 179 in IL-10-treated patients and 226 in patients receiving placebo. The proportion of patients that experienced a complete remission at any time in the 3-week follow-up period was 50% in the IL-10 group and 23% in placebo-treated patients. **Conclusions:** These results indicate that IL-10 administered as a daily bolus injection over 1 week is safe and well tolerated and may be clinically efficacious.

Current medical therapy of Crohn's disease is unsatisfactory for many patients. In recent years, it has become apparent that initiation and perpetuation of mucosal inflammation is associated with a dysbalance of proinflammatory and anti-inflammatory cytokines produced within the intestinal mucosa. Of the proinflammatory cytokines, tumor necrosis factor α (TNF- α) and interleukin (IL)-1 β have so far been identified as therapeutic targets. However, evidence now also suggests that IL-10 and transforming growth factor β (TGF- β) may play anti-inflammatory roles in the intestine.¹⁻⁴

IL-10 is an 18-kilodalton cytokine produced by certain T cells, a subset of B cells, macrophages, and monocytes. IL-10 acts to suppress inflammation resulting from both antigen-specific and innate immune responses, by reducing monocyte HLA class-II expression,^{5,6} decreasing T-cell secretion of IL-2 and interferon gamma,⁷ and dimin-

ishing activated monocyte production of IL-1 α , IL-1 β , IL-6, IL-8, TNF- α , and granulocyte-macrophage colony-stimulating factor.^{8,9} In addition, in vitro synthesis of the proinflammatory cytokines and chemokines IL-1 β , TNF, and IL-8 by lipopolysaccharide-stimulated polymorphonuclear leukocytes is down-regulated by IL-10.¹⁰⁻¹² Also, IL-10 increases the production of the IL-1 receptor antagonist by monocytes.¹³

The importance of IL-10 for the regulation of mucosal inflammation has been well investigated in several animal models of inflammatory bowel disease. Gene-targeted IL-10-deficient mice develop a severe transmural and granulomatous inflammation of the small and large bowel, reminiscent of Crohn's disease,¹ and the inflammation can be prevented by the administration of IL-10. Administration of IL-10 also ameliorates inflammation in the CD4⁺/CD45RB^{high} T-cell adoptive transfer model in severe combined deficiency mice,¹⁴ and in immune complex colitis in rabbits.¹⁵ Furthermore, the recently described effects of IL-10 in stimulating intestinal electrolyte absorption and inhibiting chloride secretion and maintaining epithelial barrier integrity may have beneficial effects in inflammatory bowel disease.^{16,17} Finally, a preliminary clinical study has suggested that local administration of IL-10 in cases of human ulcerative colitis may suppress mucosal inflammation.¹⁸

These data from animal models and the potent anti-inflammatory properties of IL-10, resulting both from suppression of antigen-specific T-cell activation and from a direct effect on activated monocytes, suggest that this cytokine is a candidate for therapeutic suppression of mucosal inflammation in Crohn's disease.

The present study was designed to investigate the safety, tolerance, and pharmacokinetics of multiple intra-

Abbreviations used in this paper: CDEIS, Crohn's disease endoscopic index of severity; EKG, electrocardiogram; ELISA, enzyme-linked immunosorbent assay; HIV, human immunodeficiency virus; IL, interleukin; TGF- β , transforming growth factor β ; TNF- α , tumor necrosis factor α ; ULN, upper limit of normal.

© 1997 by the American Gastroenterological Association
0016-5085/97/\$3.00

venous doses of recombinant human IL-10 in patients with steroid-refractory Crohn's disease. The secondary objective was to look for preliminary evidence for efficacy of IL-10 treatment in this patient population.

Materials and Methods

Patients

To be eligible for the study, patients had to be between 18 and 65 years of age and have active steroid-resistant Crohn's disease involving the colon or both the ileum and colon, with or without external fistulae. Active steroid-resistant Crohn's disease was defined as having a Crohn's Disease Activity Index (CDAI) of ≥ 200 and < 350 despite treatment with prednisone at a dose ranging from 10 to 40 mg/day for more than 2 months. Prednisone could have been administered as a monotherapy or in combination with azathioprine or 6-mercaptopurine. Either of these latter therapies had to be stable for more than 6 months and were continued at a stable dose during the study. Patients treated with other immune-modulating drugs, including cyclosporine, interferons, and methotrexate, were excluded from the study. If patients had undergone such therapies previously, the shortest interval between the last dose and entrance into the study was specified as follows: methotrexate, 2 months; anti-TNF- α , 6 months; cyclosporine, 6 months; azathioprine/6-mercaptopurine, 6 months; and all other immune modulators, 6 months. Patients were allowed to take aminosalicylates and/or antibiotics, providing that the dose regimens were stable before and during the study. Other exclusion criteria were short bowel syndrome, colostomy or ileostomy, and isolated small bowel involvement. Patients had to have the following laboratory parameters: hemoglobin ≥ 10.0 g/dL, thrombocytes $\geq 100,000$ and $\leq 800,000/\mu\text{L}$, white blood cells $\geq 4000/\mu\text{L}$, granulocyte count $\geq 2000/\mu\text{L}$, total serum bilirubin ≤ 1.5 times the upper limit of normal (ULN), serum creatinine ≤ 2.0 times ULN, alanine aminotransferase/aspartate aminotransferase ≤ 2.5 times ULN, alkaline phosphatase ≤ 2.5 times ULN, and INR ≤ 1.5 . Women of child-bearing age might be allowed to begin infusion within 24 hours of a negative β human chorionic gonadotropin pregnancy test and were required to practice adequate birth control (either oral contraception or intrauterine devices with the use of a barrier method) for the duration of the study. Exclusion criteria included immediate need for surgery, known severe stenosis, prior treatment with anti-CD4 or anti-CD5 antibodies, a clinically abnormal chest radiograph or electrocardiogram (EKG), alcohol abuse, a positive test for anti-hepatitis C virus (HCV), hepatitis B surface antigen (HBsAg), or anti-human immunodeficiency virus (HIV), a history of thrombotic events or emboli, and presence or history of cancer.

Study Design

The study was randomized, double-blind, placebo-controlled, and increasing multiple dose in nature. One of five doses (0.5, 1.0, 5, 10, and 25 $\mu\text{g/kg}$ body wt) of recombinant human IL-10 (SCH 52000) was administered once daily by

intravenous bolus injection over 7 consecutive days. A central randomization group was used for this study. At the three lowest doses, patients were randomized in a 2:1 (IL-10:placebo) ratio; at the two highest doses, the ratio was 3:1. The study thus provided for evaluation of 6 patients at the 0.5-, and 1.0- $\mu\text{g/kg}$ level (4 active drug, 2 placebo), 9 patients in the 5.0- $\mu\text{g/kg}$ dose level (6 active drug, 3 placebo), and 12 patients (9 active drug, 3 placebo) in the two higher dose groups. Therefore, a total of 45 patients was included in the study.

Patients were screened for eligibility for a period of 14 days to 24 hours before the first infusion. During days -9 to -3, patients completed daily diary entries to determine their CDAI. Other assessments included a complete medical history and physical examination, chest radiography and EKG, laboratory screen and urinalysis, testing for anti-HIV, anti-HCV, and HBsAg, and a serum pregnancy test. On day -1, an ileocolonoscopy was performed. Dosing was initiated at day 1.

Recombinant human IL-10 (SCH 52000, Schering Plough, Kenilworth, NJ), provided as a sterile powder, was reconstituted with sterile water < 1 hour before bolus injection into an antecubital vein. Placebo was identical in appearance and volume at each dose level. The study medication, followed by a 10-mL saline flush, was administered at the same time of the day during 7 consecutive days through a heparin lock or butterfly. Patients were hospitalized on days 1, 2, and 7 of infusion. The 7-day infusion period was succeeded by a 3-week follow-up period. Investigators, members of the study staff, and patients were blinded throughout this 4-week period to study drug assignment, although the treatment level assignments were known to all. The treatment each patient was using for Crohn's disease at entry was continued unchanged during the 28-day study period. Safety evaluations included clinical assessments, vital signs, laboratory parameters, EKG, radiography, and assessment of anti-IL-10 antibody formation. IL-10 pharmacokinetics were assessed on days 1 and 7 of treatment.

All patients were to be evaluated for adverse events during the course of the study by nondirect questioning at each visit. An adverse event was defined as any adverse change in the patient's baseline condition, which occurred during the course of the study, regardless of whether the event was related to treatment. Each adverse event was graded to be mild, moderate, severe, or life-threatening. Clinical responses were defined as follows: (1) a complete clinical remission was a reduction of the CDAI to < 150 and a decrease of the CDAI ≥ 100 points, and (2) a clinical response involved a decrease of the CDAI by > 100 points. All other changes in CDAI were recorded as no change.

The Crohn's disease endoscopic index of severity (CDEIS) was calculated on the basis of ileocolonoscopy performed at days -1 and 28.¹⁹ In addition, a sigmoidoscopy was performed at day 7. To minimize interobserver variability, guidelines were provided to all investigators.

Detection of IL-10

IL-10 serum concentrations were evaluated using a validated enzyme-linked immunosorbent assay (ELISA) with a

detection limit of 0.1 ng/mL. Anti-IL-10 antibodies were measured by ELISA. Serial dilutions of serum samples were added to wells of a microtiter plate coated with human IL-10. After overnight incubation, the presence of IL-10 antibodies was detected by addition of biotin-labeled protein A followed by streptavidin conjugated to horseradish peroxidase. The ELISA results of each sample were compared with the results from pooled normal serum. Samples were considered positive if the ratio of optical density from the serum sample divided by the optical density of the normal human serum was ≥ 1.5 and if the ratio of optical density from the serum sample obtained after IL-10 treatment divided by the serum sample before dosing was ≥ 2.0 .

Statistical Analysis

Because this was primarily a safety and tolerance study, no inferential statistics were performed. Baseline characteristics and weekly CDAI scores were summarized using descriptive statistics. CDAI and CDEIS scores obtained between the end of week 1 and week 4 were tabulated according to the best response at any time. Because no major differences were observed between the different dose levels, data from the five placebo groups and the various dose groups were pooled in two groups, respectively, for presentation. Adverse events were listed and tabulated by dose group.

Results

Patients were enrolled at 14 sites (3 from the United States, 1 from Canada, and 10 from Europe) between December 1994 and June 1995. Of the 59 patients screened, 47 qualified for the study. One patient almost immediately developed vomiting, myalgia, and meteorism and was withdrawn at the discretion of the investigator before dosing. The remaining 46 patients were randomized (33 to IL-10 treatment and 13 to placebo treatment) and began the study. One patient who had been assigned to placebo treatment mistakenly received a single dose (10 $\mu\text{g/kg}$) of IL-10 and was withdrawn from the study. This patient's information is included in the safety and adverse events listings but excluded from efficacy analyses. With the exception of the patient who received the wrong medication, all patients received the full week of study medications. One patient on placebo discontinued the study during week 3 of the follow-up period as a result of worsening of abdominal pain considered by the investigator to be unrelated to treatment. Patient baseline characteristics were not different between the placebo group and the IL-10-treated patients. Those characteristics are presented in Table 1. The median duration of Crohn's disease was 9 years; 30 of 45 patients had combined ileocolonic disease, 10 of 45 had external fistulae, and 23 of 45 had a history of bowel resection. The therapies in use for Crohn's disease were also similar in the two treatment groups (Table 1).

Table 1. Baseline Characteristics

	All IL-10 dose levels (n = 32)	All placebo (n = 13)
Mean age (yr)	37 (20–60)	35 (23–47)
Male/female	8/24	6/7
Mean body weight (kg)	70 (45–98)	69 (53–97)
Duration of Crohn's disease (yr)	12 (1–31)	9 (1–16)
Disease location (%)		
Colon	10 (31)	5 (28)
Colon and ileum	21 (65)	8 (61)
Colon and ileum and above	1 (3)	0 (0)
History of resection (%)		
None	17 (53)	5 (38)
Small bowel	6 (18)	3 (23)
Large bowel	5 (15)	4 (31)
Large and small bowel	4 (12)	1 (7)
External fistula (%)		
Yes	6 (18)	4 (31)
No	26 (81)	9 (69)
Mean steroid dose (mg/day)	16 (10–30)	20 (10–40)
Duration of current steroid dose (days)	358 (14–4335)	74 (19–195)
Crohn's disease management (%)		
Steroids alone	7 (22)	7 (54)
Steroids and mesalamine	17 (53)	2 (15)
Steroids and azathioprine/6-MP	2 (6)	3 (23)
Steroids and mesalamine and azathioprine/6-MP	6 (18)	1 (7)
CDAI	280 (202–341)	292 (218–345)
CDEIS	15 (6–24)	16 (6–35)

6-MP, 6-mercaptopurine.

The mean steroid dose at entry into the study was 16 mg/day in the IL-10-treated group and 20 mg/day in the placebo group. Steroid dosage remained unchanged throughout the 28-day study period.

The baseline CDEIS in the IL-10 and placebo groups was 15 and 14, respectively, and the proportion of patients with deep ulceration was comparable between the treatment groups.

Adverse Events

Clinical adverse events were reported by almost all IL-10- and placebo-treated patients (Table 2). Other frequently reported events were increases of white blood cell counts and gastrointestinal events. The incidence of specific adverse events was comparable among the treatment groups, although the incidence of abdominal pain was lower in the IL-10-treated group (9% vs. 31%). Most adverse events occurring during the study period were rated as mild to moderate. Six of the 33 patients

Table 2. Adverse Events

Adverse events	All IL-10 dose levels (n = 33)			All placebo (n = 13)		
	Total (%)	Mild to moderate	Severe	Total (%)	Mild to Moderate	Severe
Neutrophilia	16 (48)	15	1	4 (31)	4	
Headache	9 (27)	8	1	2 (15)	2	
Injection site pain	7 (21)	7		3 (23)	3	
Anemia	6 (18)	5	1	5 (38)	5	
Nausea	6 (18)	6		1 (8)	1	
Leukocytosis	5 (15)	5		4 (31)	4	
Injection site inflammation	4 (12)	4		1 (8)	1	
Fatigue	4 (12)	4		0	0	
Back pain	4 (12)	3	1	3 (23)	2	1
Insomnia	4 (12)	4		0	0	
Abdominal pain	3 (9)	2	1	4 (31)	3	1
Hypokalemia	3 (9)	3		3 (23)	3	
Fever	1 (3)	1		3 (23)	3	
Monocytosis	0	0		2 (15)	2	

(18%) who received IL-10 and 4 of 13 patients (31%) in the placebo group reported severe adverse events. Severe adverse events experienced by 2 patients who received IL-10 were considered to be possibly related to treatment. One of those patients (10- μ g/kg dose) developed a perianal abscess and fistula at the end of 2 weeks of follow-up. Another patient (25 μ g/kg of IL-10) developed neutrophilia during the 7-day treatment period without symptoms of infection. In addition, 1 placebo-treated patient developed a severe adverse event (pulmonary embolism) at the end of 2 weeks of follow-up.

Significant shifts in laboratory results were observed in both IL-10-treated (23 of 33; 70%) and placebo-treated (11 of 13; 85%) patients. Most commonly, these shifts involved an increase in total or segmented neutro-

phils. No apparent pattern or dose relationship was identified, and no evidence for renal or hepatic toxicity was observed.

Clinical Responses

Stringent criteria were used for the definition of remission (decrease of baseline CDAI < 150 and a decrease of the CDAI > 100) and clinical response (decrease of baseline CDAI > 100). The mean CDAI decreased in both treatment groups, but the mean CDAI scores observed during the follow-up period were generally lower in the IL-10-treated patients (179 vs. 226 at the end of the study; Figure 1). No apparent dose-effect relationship was observed, but it should be noted that the small number of patients in each dose group precluded a meaningful analysis. The decrease in CDAI scores was sustained throughout the 3-week follow-up period for most of the patients. The number of patients who experienced

Figure 1. CDAI after IL-10 administration. Data represent a summary of mean CDAI scores in the intent-to-treat population. ■, IL-10; □, placebo.

Figure 2. Clinical responses (%) after administration of IL-10 in the 3-week follow-up period (best value extended). ■, IL-10; □, placebo.

Figure 3. Incidence of remission (%) by week of study in placebo- and IL-10-treated patients. ■, IL-10; □, placebo.

a remission in the IL-10 group was 50% (16 of 32) and 23% (3 of 13) in the placebo group. Clinical responses were observed in 31% of IL-10-treated patients and 23% of placebo-treated individuals (Figure 2). Of the patients treated with placebo, 54% had no change in disease activity. Only 19% of IL-10-treated patients experienced no change. In most patients for whom a complete clinical remission was obtained, the response was usually sustained throughout the study period (Figure 3). No potentially significant trends were observed for differences in CDEIS between the dose groups. The mean change was -3 (range, -13 to 5) in the IL-10-treated patients and -4 (range, -16 to 10) in the placebo group. A tabulation of CDEIS and CDAI failed to reveal a correlation (data not shown).

Pharmacokinetics and Anti-IL-10 Antibodies

Both the maximum serum IL-10 concentration and area under serum concentration time curve increased in a dose-related manner after the administration of IL-10 (Table 3). The drug was rapidly cleared from the body with a mean half-life of 2.6 hours on day 1 and 1.1 hours on day 7 (based on data for the 25- μ g/kg dose group). These results are similar to those observed for healthy volunteers. No evidence for IL-10 accumulation was found. No antibodies to human IL-10 were detected in serum obtained from patients at the end of the 3-week follow-up period.

Discussion

This is the first study to report on the effects of clinical application of IL-10 in human disease; the results indicate that treatment with short-term intravenous administration of IL-10 for patients with steroid-refractory Crohn's disease is safe and generally well tolerated. The

mild to moderate adverse events experienced by both placebo- and IL-10-treated patients are not surprising in view of the selection of patients with severe steroid-refractory Crohn's disease. Clinical adverse events were reported in 6 of 33 (18%) IL-10-treated patients and in 4 of 13 (31%) placebo-treated patients. Those events included headache, anemia, nausea, leukocytosis, fatigue, and insomnia, all consistent with the presence of severe Crohn's disease. The most important laboratory finding was the neutrophilia, which occurred in 16 of 33 (48%) patients in the IL-10 group and in 4 of 13 (31%) placebo-treated patients. An increase in neutrophil counts had previously been observed in IL-10-infused volunteers.²⁰ However, the neutrophil counts in this study may have been influenced by disease activity and steroid use. The pharmacokinetic profile for IL-10 was similar to that observed in healthy volunteers and indicates that IL-10 is rapidly removed from the body, without evidence for accumulation after repeated administration. Disease-associated parameters and/or concomitant drug therapy did not appear to have any effect on the pharmacokinetic profile of IL-10.

This study was not designed to assess the efficacy of IL-10 treatment in Crohn's disease or to show significant differences in clinical responses between the treatment groups. However, 81% of IL-10-treated patients experienced either a complete clinical remission or a clinical response, whereas only 46% of placebo-treated patients had such a response. The 23% rate of clinical remissions in placebo-treated patients with steroid-refractory Crohn's disease is similar to that reported in previous studies and may reflect a response to hospital admission and a general increase in intensity of medical care as a consequence of participation in the clinical study. Endoscopic findings for IL-10- and placebo-treated patients

Table 3. IL-10 Pharmacokinetics

Dose group	Serum IL-10 concentration				
	0.5 μ g/kg	1 μ g/kg	5 μ g/kg	10 μ g/kg	25 μ g/kg
C_{max}					
Day 1	7.0	20.1	102	191	451
Day 7	9.7	17.8	94	190	453
T_{max}					
Day 1	0.08	0.08	0.08	0.08	0.08
Day 7	0.08	0.13	0.08	0.10	0.08
AUC					
Day 1	3.2	12.7	84.1	155	393
Day 7	4.6	11.7	71.8	164	352
$t_{1/2}$					
Day 1	—	—	—	—	2.6
Day 7	—	—	—	—	1.1

C_{max} , maximum serum IL-10 concentration; T_{max} , time of maximum serum concentration; AUC, area under the serum concentration; $t_{1/2}$, terminal phase half-life (hours).

did not reveal striking differences, and, as has been reported by other investigators, no correlation appeared between CDEIS and disease activity.²¹ In addition, no evident relationship between IL-10 dose level and clinical response was observed. It should be noted that the dose-escalating design was chosen primarily to assess safety and tolerance and that the number of patients in each dose group was small. Therefore, the present study does not address the clinical efficacy of IL-10 in Crohn's disease, the effects on endoscopic findings, or the selection of active doses. These issues need to be addressed in future trials.

In view of the short half-life of IL-10, there is some question about whether IL-10 treatment of a chronic disease such as Crohn's disease is feasible. IL-10 has both anti-inflammatory and immunosuppressive activities, and we do not presently know which is the more important activity for suppression of Crohn's disease. Despite its short half-life, a single bolus infusion of IL-10 to healthy volunteers in an earlier study caused suppression of in vitro endotoxin-induced cytokine production during 24 hours.²² Local application of IL-10 in ulcerative colitis caused a reduction in disease activity most likely resulting from inhibition of the production of various proinflammatory cytokines (including TNF- α , IL-1 β , and IL-8) by lamina propria monocytes or macrophages.¹⁸ In addition to these anti-inflammatory effects, several recent observations indicate that exposure of CD4⁺ T cells to IL-10 during antigen-dependent activation may result in long-term antigen-specific anergy.²³ If IL-10 exerts this effect in vivo, short-term treatment with IL-10 may indeed have prolonged immunomodulatory effects. Although the mechanism of action of IL-10 remains unclear, a single week of intravenous IL-10 treatment does appear to have a generally sustained effect on the activity of Crohn's disease as assessed by the CDAI. Furthermore, studies on volunteers have indicated that subcutaneous IL-10 administration is feasible, and clinical studies in patients with inflammatory bowel disease using this route of administration are currently in progress.

In conclusion, the results of this study indicate that IL-10, administered as a daily bolus infusion over 1 week to patients with steroid-refractory Crohn's disease, is safe and well tolerated and may be clinically efficacious.

References

1. Kuhn R, Lohler J, Rennick D, Rajewsky K, Muller W. Interleukin-10-deficient mice develop chronic enterocolitis. *Cell* 1993;75:263-274.
2. Rennick D, Davidson N, Berg D. Interleukin-10 gene knockout mice: a model of chronic inflammation. *Clin Immunol Immunopathol* 1995;76:S174-S178.
3. Duchmann R, Schmitt E, Knolle P, Zum Büschenfelde KHM, Neurath M. Tolerance towards resident intestinal flora in mice is abrogated in experimental colitis and restored by treatment with interleukin-10 or antibodies. *Eur J Immunol* 1996;26:934-938.
4. Neurath ME, Fuss I, Kelsall BL, Presky DH, Waegell W, Strober W. Experimental granulomatous colitis in mice is abrogated by induction of TGF-beta-mediated oral tolerance. *J Exp Med* 1996;183:2605-2616.
5. Fiorentino DF, Zlotnik A, Viera P, Mosmann TR, Howard M, Moore KW, O'Garra A. IL-10 acts on the antigen-presenting cell to inhibit cytokine production by Th1 cells. *J Immunol* 1991;146:3444-3451.
6. De Waal Malefyt R, Haanen J, Spits H, Abrams J, Bennett B, Figdor CJ, de Vries JE. Interleukin 10 (IL-10) and viral IL-10 strongly reduce antigen-specific human T-cell proliferation by diminishing the antigen-presenting capacity of monocytes via down-regulation of class II major histocompatibility complex expression. *J Exp Med* 1991;174:915-924.
7. Taga K, Tosato G. IL-10 inhibits human T cell proliferation and IL-2. *J Immunol* 1992;148:1143-1148.
8. De Waal Malefyt R, Abrams J, Bennett B, Figdor CG, de Vries JE. Interleukin 10 (IL-10) inhibits cytokine synthesis by human monocytes: an autoregulatory role of IL-10 produced by monocytes. *J Exp Med* 1991;174:1209-1220.
9. Fiorentino DF, Zlotnik A, Mosmann TR, Howard M, O'Garra A. IL-10 inhibits cytokine production by activated macrophages. *J Immunol* 1991;147:3815-3822.
10. Cassatella MA, Meda L, Bonora S, Ceska M, Constantin G. Interleukin 10 (IL-10) inhibits the release of proinflammatory cytokines from human polymorphonuclear leukocytes. Evidence for an autocrine role of tumor necrosis factor and IL-1 β in mediating the production of IL-8 triggered by lipopolysaccharide. *J Exp Med* 1993;178:2207-2211.
11. Kasama T, Strieter M, Lukacs N, Burdick M, Kunkel S. Regulation of neutrophil-derived chemokine expression by IL-10. *J Immunol* 1994;152:3559-3569.
12. Wang P, Wu P, Anthes JC, Siegel MI, Egan KW, Billah MM. Interleukin-10 inhibits interleukin-8 production in human neutrophils. *Blood* 1994;83:2678-2683.
13. Cassatella MA, Meda L, Gasperini S, Calzetti F, Bonora S. Interleukin-10 (IL-10) upregulates IL-1 receptor antagonist production from lipopolysaccharide-stimulated human polymorphonuclear leukocytes by delaying mRNA degradation. *J Exp Med* 1994;179:1695-1699.
14. Powrie F, Leach MW, Mauze S, Menon S, Caddle LB, Coffman RL. Inhibition of TH1 responses prevents inflammatory bowel diseases in SCID mice reconstituted with CD45RB^{hi}CD4⁺T cells. *Immunity* 1994;1:553-562.
15. Grool TA, Meenan J, van Dulleman H, Koster F, ten Kate FJW, Lebeaut A, Tytgat GNJ, van Deventer SJH. The anti-inflammatory effect of interleukin-10 in a rabbit model of immune complex-induced colitis (abstr). *Gastroenterology* 1996;110:A918.
16. Madsen KL, Tavernini MM, Fedorak RN. Interleukin-10 inhibits modulates ion transport in rat small intestine. *Gastroenterology* 1996;111:936-944.
17. Madsen KL, Hibbard J, Tavernini MM, Fedorak RN. Interleukin-10 prevents disruption of T84 monolayer barrier integrity and limits chloride secretion (abstr). *Gastroenterology* 1996;110:A953.
18. Schreiber S, Heinig T, Thiele H, Raedler A. Immunoregulatory role of interleukin 10 in patients with inflammatory bowel disease. *Gastroenterology* 1995;108:1434-1444.
19. Mary JY, Modigliani R. Development and validation of an endoscopic index of the severity for Crohn's disease: a prospective multicentre study. *Gut* 1989;30:983-989.
20. Huhn RD, Radwanski E, O'Connell SM, Sturgill MG, Clarke L, Cody RP, Affrime MB, Cutler DL. Pharmacokinetics and immunomodulatory properties of intravenously administered recombinant human interleukin-10 in healthy volunteers. *Blood* 1996;87:699-705.

21. Cellier C, Shamoud T, Froguel E, Adenis A, Belaiche J, Bretagne JF, Florent C, Bouvry M, Mary JY, Modigliani R. Correlations between clinical activity, endoscopic severity, and biological parameters in colonic or ileocolonic Crohn's disease. A prospective multicentre study of 121 cases. *Gut* 1994;35:231-235.
22. Chernoff AE, Granowitz EV, Shapiro L, Vannier E, Lonneman G, Angel JB, Kennedy JS, Rabson AR, Wolff SM, Dinarello CA. A randomized controlled trial of rIL-10 in humans. Inhibition of inflammatory cytokine production and immune responses. *J Immunol* 1995;154:5492-5499.
23. Groux H, Bigler M, De Vries JE, Roncarolo MG. Interleukin-10 induces a long-term antigen-specific state in human CD4⁺ T cells. *J Exp Med* 1996;184:19-29.

Received December 5, 1996. Accepted April 22, 1997.

Address requests for reprints to: Sander J.H. van Deventer, M.D., Laboratory for Experimental Internal Medicine, Academic Medical

Center, Meibergdreef 9, 1105 AZ Amsterdam, The Netherlands. e-mail: s.j.vandeventer@amc.uva.nl; fax: (31) 20-6977192.

Supported by Schering Plough, Kenilworth, NJ.

The following investigators participated in the study: Austria: Dr. A. Gangl and Dr. C. Gasche, University Clinic for Internal Medicine IV, Vienna; Dr. H. Tilg, University Clinic for Internal Medicine, Innsbruck. Belgium: Dr. P. Rutgeerts and Dr. M. Peeters, UZ Gasthuisberg, University of Leuven, Leuven. Germany: Dr. K. Deusch and Dr. K. Reich, Abteilung für Allgemeinchirurgie, Klinikum der Eberhard-Karls-Universität, Tübingen; Dr. J. Schölmerich and Dr. V. Gross, Medizinische Klinik I, Universitätskliniken Regensburg. Netherlands: Dr. J. Koningsberger, Department of Gastroenterology, University Hospital Utrecht; Dr. M. van Blankenstein and Dr. J.H.P. Wilson, Department of Internal Medicine, University Hospital Rotterdam. United Kingdom: Dr. M. Jacyna, Gastroenterology Department, Northwick Park Hospital, Middlesex; Dr. D.P. Jewell, Radcliffe Infirmary NHS Trust, Oxford. United States: Dr. S. Hanauer, University of Chicago, Chicago, IL; Dr. K. Isaacs, University of North Carolina School of Medicine, Chapel Hill, NC; and Dr. W.J. Tremaine, Mayo Clinic, Rochester, NY.