

A SHORT-TERM STUDY OF CHIMERIC MONOCLONAL ANTIBODY cA2 TO TUMOR NECROSIS FACTOR α FOR CROHN'S DISEASE

STEPHAN R. TARGAN, M.D., STEPHEN B. HANAUER, M.D., SANDER J.H. VAN DEVENTER, M.D., PH.D., LLOYD MAYER, M.D., DANIEL H. PRESENT, M.D., TANJA BRAAKMAN, M.D., KIMBERLY L. DEWOODY, M.S., THOMAS F. SCHAIBLE, PH.D., AND PAUL J. RUTGEERTS, M.D., PH.D., FOR THE CROHN'S DISEASE cA2 STUDY GROUP

ABSTRACT

Background Studies in animals and an open-label trial have suggested a role for antibodies to tumor necrosis factor α , specifically chimeric monoclonal antibody cA2, in the treatment of Crohn's disease.

Methods We conducted a 12-week multicenter, double-blind, placebo-controlled trial of cA2 in 108 patients with moderate-to-severe Crohn's disease that was resistant to treatment. All had scores on the Crohn's Disease Activity Index between 220 and 400 (scores can range from 0 to about 600, with higher scores indicating more severe illness). Patients were randomly assigned to receive a single two-hour intravenous infusion of either placebo or cA2 in a dose of 5 mg per kilogram of body weight, 10 mg per kilogram, or 20 mg per kilogram. Clinical response, the primary end point, was defined as a reduction of 70 or more points in the score on the Crohn's Disease Activity Index at four weeks that was not accompanied by a change in any concomitant medications.

Results At four weeks, 81 percent of the patients given 5 mg of cA2 per kilogram (22 of 27 patients), 50 percent of those given 10 mg of cA2 per kilogram (14 of 28), and 64 percent of those given 20 mg of cA2 per kilogram (18 of 28) had had a clinical response, as compared with 17 percent of patients in the placebo group (4 of 24) ($P < 0.001$ for the comparison of the cA2 group as a whole with placebo). Thirty-three percent of the patients given cA2 went into remission (defined as a score below 150 on the Crohn's Disease Activity Index), as compared with 4 percent of the patients given placebo ($P = 0.005$). At 12 weeks, 41 percent of the cA2-treated patients (34 of 83) had had a clinical response, as compared with 12 percent of the patients in the placebo group (3 of 25) ($P = 0.008$). The rates of adverse effects were similar in the groups.

Conclusions A single infusion of cA2 was an effective short-term treatment in many patients with moderate-to-severe, treatment-resistant Crohn's disease. (N Engl J Med 1997;337:1029-35.)

©1997, Massachusetts Medical Society.

CROHN'S disease is a chronic inflammatory disorder characterized by patchy granulomatous inflammation of any part of the gastrointestinal tract.¹ Patients have a spectrum of clinical features, with great variation in the course of the disease. Mesalamine is considered first-line therapy. The majority of patients have relapses

requiring glucocorticoid treatment.² Immunomodulatory agents, including azathioprine or mercaptopurine,³ methotrexate,^{4,5} and cyclosporine,⁶⁻⁹ may be used to treat severe, persistent disease that is refractory to treatment with corticosteroids, or symptoms that recur on tapering of the dose of corticosteroids.

In animal models, antibodies to tumor necrosis factor α (anti-TNF- α) prevent or reduce inflammation,¹⁰⁻¹⁴ suggesting that therapy with such antibodies may be useful for disorders in which chronic inflammation may be due to an increase in cytokines produced by the T helper 1 subclass of T cells. In vitro studies have shown that the production of TNF- α is increased in the mucosa of patients with Crohn's disease^{15,16} and that the mucosal inflammatory process reflects a shift in the balance of cytokine production by T cells toward the T helper 1 subclass.^{17,18} Similar findings were reported in the synovia of patients with rheumatoid arthritis,¹⁹ and anti-TNF- α reduces clinical signs and symptoms of this disease.^{20,21} The role of TNF- α in the pathogenesis of Crohn's disease and the successful use of anti-TNF- α in the treatment of rheumatoid arthritis stimulated an open-label trial of chimeric monoclonal antibody cA2 (infliximab, Centocor, Malvern, Pa.) for Crohn's disease. In that preliminary trial, clinical remission occurred after one infusion of cA2 in eight of nine patients with Crohn's disease.²² We report the results of a multicenter randomized, placebo-controlled, double-blind trial of cA2 for the treatment of active Crohn's disease.

METHODS**Patients**

To be eligible for the study, patients had to have had Crohn's disease for six months,¹ with scores on the Crohn's Disease Activity Index²³ between 220 and 400. The Crohn's Disease Activity Index incorporates eight variables related to the disease: the number of liquid or very soft stools, the severity of abdominal pain or cramping, general well-being, the presence of extraintestinal man-

From the Cedars-Sinai Medical Center, Los Angeles (S.R.T.); the University of Chicago, Chicago (S.B.H.); Academisch Medisch Centrum, Amsterdam (S.J.H.D.); Mt. Sinai Medical Center, New York (L.M., D.H.P.); Centocor, Inc., Malvern, Pa. (T.B., K.L.D., T.F.S.); and Academisch Ziekenhuis Gasthuisberg, Leuven, Belgium (P.J.R.). Address reprint requests to Dr. Targan at Cedars-Sinai Medical Center, Division of Gastroenterology and Inflammatory Bowel Disease Center D4063, 8700 Beverly Blvd., Los Angeles, CA 90048.

ifestations, abdominal mass, use of anti-diarrheal drugs, hemato-crit, and body weight. These items yield a composite score ranging from 0 to approximately 600. Higher scores indicate greater disease activity. Scores below 150 indicate remission, whereas scores above 450 indicate severe illness. Patients were eligible for the study if they had been receiving any of the following: mesalamine for eight or more weeks, with the dose remaining stable during the four weeks before screening; a maximum of 40 mg of corticosteroids per day for eight or more weeks, with the dose remaining stable during the two weeks before screening; and mercaptopurine or azathioprine for six or more months, with the dose remaining stable during the eight weeks before screening. Patients were excluded from the study if they had received treatment with cyclosporine, methotrexate, or experimental agents within three months before screening. Patients were also excluded if they met any of the following criteria: symptomatic stenosis or ileal strictures; proctocolectomy or total colectomy; stoma; a history of allergy to murine proteins; prior treatment with murine, chimeric, or humanized monoclonal antibodies; or treatment with parenteral corticosteroids or corticotropin within four weeks before screening.

Patients were enrolled at 18 centers in North America and Europe. The protocol was approved by the institutional review boards and ethics committees at all sites, and all patients gave written informed consent before enrolling in the trial. The study began on June 21, 1995, and concluded on March 12, 1996. A total of 203 patients were screened for the study, 95 of whom were excluded. The most common reasons for exclusion were a requirement for contraindicated medications, refusal to give informed consent, or disease activity that did not meet the study criteria.

Protocol

Subjects were screened one week before the administration of cA2 to establish base-line scores on the Crohn's Disease Activity Index and the Inflammatory Bowel Disease Questionnaire,²⁴ and base-line C-reactive protein concentrations. The Inflammatory Bowel Disease Questionnaire, a 32-item questionnaire, evaluates quality of life with respect to bowel function (e.g., loose stools and abdominal pain), systemic symptoms (fatigue and altered sleep pattern), social function (work attendance and the need to cancel social events), and emotional status (angry, depressed, or irritable). The score ranges from 32 to 224, with higher scores indicating a better quality of life. Patients in remission usually score between 170 and 190.²⁴

Patients were randomly assigned to receive a single dose of either placebo or 5 mg of cA2 per kilogram of body weight, 10 mg of cA2 per kilogram, or 20 mg of cA2 per kilogram in an intravenous infusion, administered over a two-hour period. The cA2 monoclonal antibody is a chimeric mouse-human IgG1 that binds to both soluble²⁵ and transmembrane²⁶ human TNF- α with high affinity and specificity. It neutralizes the functional activity of TNF in a variety of bioassays by blocking the binding of the factor to the p55 and p75 receptors.²⁷ The placebo preparation contained 0.1 percent human serum albumin instead of cA2 and was identical in appearance to the cA2 solution. Patients were enrolled from June 21, 1995, to October 31, 1995. Randomization was performed centrally by an independent organization (PPD Pharmaco, Austin, Tex.). The cA2 and placebo solutions were prepared by a pharmacist at each site who was aware of the treatment assignments. The investigators, all other study personnel, and the patients were blinded to the treatment assignments.

The primary end point was defined before the initiation of the trial as a reduction of 70 points or more in the score on the Crohn's Disease Activity Index at the four-week evaluation that was not accompanied by a change in any concomitant medications. Patients who did not have a clinical response at that time were enrolled in a parallel, open-label study and received a single infusion of 10 mg of cA2 per kilogram and were followed for 12 additional weeks. Patients who were receiving mesalamine, corti-

steroids, azathioprine, or mercaptopurine before the study continued to receive a stable dose during the trial period. The dose of corticosteroids could be tapered beginning eight weeks after the initiation of the study. Treatment with these drugs or with methotrexate or cyclosporine could not be initiated during the trial. After all patients had completed 12 weeks of the trial and the data were finalized, the treatment assignments were revealed.

Immunologic Investigations

Serum samples were obtained at base line and at 12 weeks for the evaluation of antinuclear antibodies and human anti-cA2. Antinuclear antibodies were detected by immunofluorescence on Hep-2 cells. Serum samples positive by immunofluorescence for antinuclear antibodies were tested for antibodies to double-stranded DNA by an enzyme-linked immunosorbent assay (North American centers) or by Crithidia immunofluorescence (European centers). Human anti-cA2 was measured by a double-antigen enzyme-linked immunosorbent assay.

Statistical Analysis

An adaptive stratified design was used to assign patients to a treatment group, with investigational site and corticosteroid use as the strata. We calculated that approximately 25 patients were needed in each treatment group to detect a difference in the number of patients who responded with 80 percent power ($\alpha = 0.05$), assuming a response rate of 30 percent in the placebo group, 80 percent in the cA2 group with the greatest response, and 55 percent in the remaining cA2 groups. The original study protocol did not specify the use of intention-to-treat analysis. Two patients were assigned to a treatment but did not receive it: one declined to participate and one did not meet eligibility criteria. No further data were collected on these two patients, and they are not included in the analysis. Otherwise, all patients were analyzed according to the treatment to which they were assigned. When we assessed the response or remission rates in all evaluation periods after the initial blinded infusion, patients who received an open-label infusion or those with a change in concomitantly administered medications were considered to have had no response.

Categorical variables (clinical response and remission) were compared with use of the Mantel-Haenszel chi-square test for general association stratified according to investigational site.²⁸ Analyses comparing each of the cA2 treatment groups with placebo were performed only when the treatment effect was considered significant ($P < 0.05$). The changes from base line in continuous variables (Crohn's Disease Activity Index score, Inflammatory Bowel Disease Questionnaire score, and C-reactive protein concentration) were compared with use of analysis of variance, with the van der Waerden normal scores blocked according to center.²⁹ If the treatment effect was significant, the cA2 treatment groups were compared with the placebo group with linear contrasts. All P values are two-sided.

RESULTS

Base-Line Characteristics of the Study Patients

A total of 108 patients were studied, with 25 to 28 patients in each group. There were no significant differences in age, weight, race (all patients were white), sex, duration of disease, scores on the Crohn's Disease Activity Index and Inflammatory Bowel Disease Questionnaire, or C-reactive protein concentrations at base line among the groups, although patients in the placebo group had a lower mean concentration of C-reactive protein (Table 1). Significantly more patients had ileal disease alone in the placebo group than in the other three groups ($P = 0.02$), but there were no significant differences

TABLE 1. BASE-LINE CHARACTERISTICS OF THE 108 PATIENTS.*

CHARACTERISTIC	PLACEBO (N=25)	5 mg OF cA2/kg (N=27)	10 mg OF cA2/kg (N=28)	20 mg OF cA2/kg (N=28)
Duration of disease — yr	10.4±7.7	12.5±10.3	11.5±9.6	13.5±8.8
Involved intestinal area — no. of patients (%)				
Ileum only	8 (32)†	3 (11)	4 (14)	2 (7)
Ileum and colon	10 (40)	15 (56)	14 (50)	19 (68)
Colon only	7 (28)	9 (33)	10 (36)	7 (25)
Previous segmental resection — no. of patients (%)	13 (52)	12 (44)	14 (50)	14 (50)
Age — yr	38.5±11.0	37.0±11.8	39.3±10.6	36.0±9.7
Male sex — no. (%)	15 (60)	14 (52)	13 (46)	13 (46)
Weight — kg	71.4±14.4	68.1±17.7	74.2±19.5	68.4±16.0
Height — cm	172±11	169±8	171±10	171±9
Medications — no. of patients				
Prednisone equivalent				
<20 mg/day orally	10 (40)	8 (30)	8 (29)	10 (36)
≥20 mg/day orally	6 (24)	7 (26)	8 (29)	7 (25)
Mercaptopurine	4 (16)	4 (15)	4 (14)	4 (14)
Azathioprine	7 (28)	5 (19)	4 (14)	8 (29)
Oral aminosalicylates	17 (68)	16 (59)	18 (64)	13 (46)
Score on Crohn's Disease Activity Index	288±54	312±56	318±59	307±50
Score on Inflammatory Bowel Disease Questionnaire	128±29	122±29	116±23	118±28
C-reactive protein — mg/liter	12.8±13.9	22.1±23.6	23.2±34.2	22.4±23.9

*Plus-minus values are means ±SD. Higher scores on the Crohn's Disease Activity Index indicate greater disease activity, and higher scores on the Inflammatory Bowel Disease Questionnaire indicate better quality of life.

†P=0.02 for the comparison with the other three groups.

in the number who had undergone previous segmental resections among the groups. Similar numbers of patients in each group had been treated with oral corticosteroids, mercaptopurine, azathioprine, and oral mesalamine at base line. All treatment groups had a mean score on the Crohn's Disease Activity Index of approximately 300, despite concurrent treatment with drugs other than cA2; thus, the patients had moderate-to-severe, treatment-resistant Crohn's disease.

Clinical Response and Remission

Week 2

Figure 1A demonstrates that clinical response was achieved early: 61 percent of cA2-treated patients had a clinical response by week 2, as compared with 17 percent of patients in the placebo group ($P<0.001$). At two weeks, 27 percent of cA2-treated patients were in clinical remission (defined as a score of less than 150 on the Crohn's Disease Activity Index), as compared to 4 percent of the patients in the placebo group ($P=0.06$) (Fig. 1B).

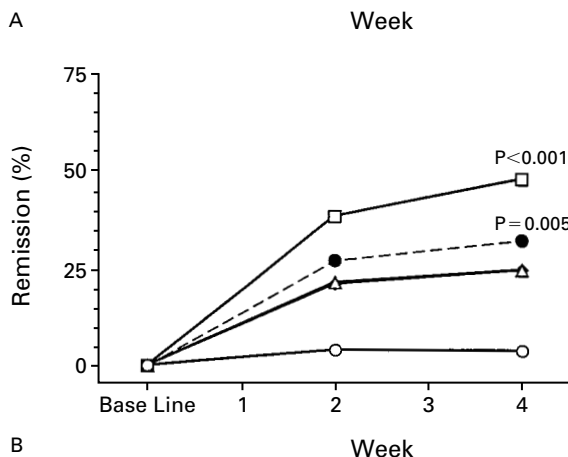
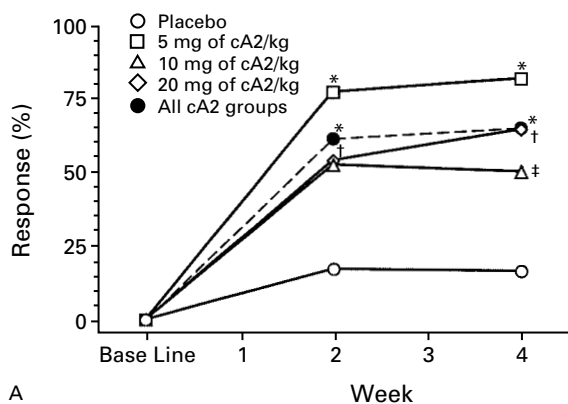
Week 4

Four weeks after the infusion, the primary end point of a reduction of 70 or more points in the score on the Crohn's Disease Activity Index was

reached in 81 percent of those given 5 mg of cA2 per kilogram (22 of 27 patients), 50 percent of those given 10 mg of cA2 per kilogram (14 of 28 patients), and 64 percent of those given 20 mg of cA2 per kilogram (18 of 28 patients), as compared with 17 percent of those given placebo (4 of 24 patients) (Fig. 1A). The overall response of the cA2 groups was 65 percent ($P<0.001$ for the comparison with placebo). No dose-response relation was seen during this period. At four weeks, 33 percent of the cA2-treated group were in remission, as compared with 4 percent of the placebo group ($P=0.005$). Thus approximately half of the patients who had a clinical response at either two or four weeks also entered remission. Consistent treatment effects were observed when the analyses for both response and remission at week 4 were stratified according to the location of disease or concurrent drug regimens (data not shown).

Changes in Clinical and Inflammatory Measures during the First Four Weeks

The mean change in the scores on the Crohn's Disease Activity Index in the cA2-treated group as a whole was significant at weeks 2 and 4 of the trial, as compared with the changes in scores in the placebo group ($P<0.001$) (Table 2). The mean decrease in the score on the Crohn's Disease Activity Index



NO. OF PATIENTS EVALUATED

Placebo	25	24	24
5 mg of cA2/kg	27	26	27
10 mg of cA2/kg	28	23	28
20 mg of cA2/kg	28	28	28
All cA2 groups	83	77	83

Figure 1. Rates of Clinical Response and Remission after a Single Infusion of cA2 or Placebo.

Clinical remission was defined as a score of less than 150 on the Crohn's Disease Activity Index and a score of 170 to 190 on the Inflammatory Bowel Disease Questionnaire. The asterisks ($P < 0.001$), daggers ($P < 0.01$), and double dagger ($P < 0.05$) indicate a significant difference from placebo.

in the cA2 group as a whole was 110 at four weeks, as compared with 13 in the placebo group. Most of this decrease had occurred by week 2, with a mean decrease of 103 in the cA2 group and 16 in the placebo group.

The Inflammatory Bowel Disease Questionnaire was given at base line and four weeks. There was a mean increase of 46, 30, and 32 in the groups treated with cA2 at 5, 10, and 20 mg of cA2 per kilogram, respectively, yielding a mean increase of 36 in the cA2 group as a whole, as compared with a mean increase of 5 in the placebo group ($P = 0.001$) (Table 2).

Concentrations of C-reactive protein were measured at base line and weeks 2 and 4. At four weeks,

the mean decrease in C-reactive protein was 16.3, 11.1, and 15.0 mg per liter in the groups treated with 5, 10, and 20 mg of cA2 per kilogram, respectively, yielding a mean decrease of 14.3 mg per liter in the group as a whole, as compared with a mean increase of 2.0 mg per liter in the placebo group ($P < 0.001$). The maximal reduction in C-reactive protein occurred within the first two weeks. At two weeks, the mean decrease in C-reactive protein in the cA2 groups as a whole was 16.0 mg per liter, as compared with a mean increase of 3.9 mg per liter in the placebo group ($P < 0.001$).

Week 12

The differences in the rates of clinical response between the cA2-treated groups and the placebo group remained significant through the 12 weeks of follow-up: it was 48 percent in the group given 5 mg of cA2 per kilogram (13 of 27 patients), 29 percent in the group given 10 mg of cA2 per kilogram (8 of 28 patients), and 46 percent in the group given 20 mg of cA2 per kilogram (13 of 28 patients), for an overall rate of response of 41 percent (34 of 83 patients), as compared with a rate of 12 percent in the placebo group (3 of 25 patients) ($P = 0.008$). The difference in the percentage of patients who were in remission was not significant at 12 weeks: 30 percent of the group given 5 mg of cA2 per kilogram (8 of 27 patients), 18 percent of the group given 10 mg of cA2 per kilogram (5 of 28 patients), and 25 percent of the group given 20 mg of cA2 per kilogram (7 of 28 patients), for an overall rate of remission achieved of 24 percent (20 of 83 patients), as compared with a rate of 8 percent in the placebo group (2 of 25 patients) ($P = 0.31$).

Characterization of the Response to Treatment

The magnitude and duration of response were characterized through the 12-week follow-up period in the 54 patients who responded to a single infusion of cA2. The improvement in the scores on the Crohn's Disease Activity Index (clinical remission was defined as a score below 150) and Inflammatory Bowel Disease Questionnaire (remission was defined as a score between 170 and 190) in patients with a response was maintained. The mean (\pm SD) score on the Crohn's Disease Activity Index was 318 ± 52 at base line, 144 ± 67 at week 4, 151 ± 86 at week 8, and 182 ± 91 at week 12, and the mean score on the Inflammatory Bowel Disease Questionnaire was 121 ± 26 at base line, 175 ± 26 at week 4, 165 ± 36 at week 8, and 162 ± 35 at week 12. Concentrations of C-reactive protein (normal, < 8 mg per liter) began to rise at 12 weeks, potentially indicating a relapse of disease (from 25.8 ± 2.7 mg per liter at base line to 7.5 ± 1.5 mg per liter at week 4, 11.0 ± 2.1 mg per liter at week 8, and 14.1 ± 2.2 mg per liter at week 12).

TABLE 2. MEASURES OF CLINICAL RESPONSE AND INFLAMMATION AT BASE LINE AND WEEKS 2 AND 4.*

VARIABLE	PLACEBO (N=25)	5 mg OF cA2/kg (N=27)	10 mg OF cA2/kg (N=28)	20 mg OF cA2/kg (N=28)	ALL cA2 GROUPS (N=83)
Score on Crohn's Disease Activity Index					
Base line	288±54	312±56	318±59	307±50	312±55
2 weeks	272±75	182±79†	238±92‡	217±92†	212±90†
4 weeks	271±82	166±76†	226±115§	211±107†	201±103†
Score on Inflammatory Bowel Disease Questionnaire					
Base line	128±29	122±29	116±23	118±28	118±27
4 weeks	133±28	168±36†	146±41¶	149±35	154±38‡
C-reactive protein (mg/liter)					
Base line	12.8±13.9	22.1±23.6	23.2±34.2	22.4±23.9	22.6±27.4
2 weeks	16.4±18.9	4.2±3.0†	6.7±7.3**	8.7±13.8†	6.5±9.3†
4 weeks	14.8±18.6	5.7±9.3††	12.1±18.6§	6.9±11.6†	8.3±13.9†

*Plus-minus values are means ±SD. Higher scores on the Crohn's Disease Activity Index indicate greater disease activity, and higher scores on the Inflammatory Bowel Disease Questionnaire indicate better quality of life. All P values are for the change from base line in cA2-treated groups as compared with the placebo group. See the Methods section for a description of the statistical analyses used.

†P<0.001. ‡P=0.001. §P=0.003. ¶P=0.02.
||P=0.03. **P=0.002. ††P=0.004.

Effects of Open-Label cA2 in Patients with No Response Four Weeks after the Initial Infusion of cA2

Patients who did not have a clinical response after the first infusion were given a second infusion of open-label cA2 in a dose of 10 mg per kilogram and followed for an additional 12 weeks. Among 19 patients who initially received placebo, the response rate was 58 percent and the remission rate was 47 percent four weeks after the second infusion — rates that were similar to those in the initial, blinded study (Fig. 1). By contrast, among the 29 patients who had no response to the initial cA2 infusion, the rates of response and remission after the second infusion were 34 percent (P=0.14 for the comparison with the patients who received placebo initially) and 17 percent (P=0.05), respectively, confirming that this group was less responsive to cA2.

Adverse Effects

Adverse effects were recorded at the time of infusion and 2, 4, 8, and 12 weeks after the infusion. In patients who received an open-label infusion 4 weeks after an infusion of placebo or cA2, adverse effects were monitored for an additional 12 weeks. As shown in Table 3, the percentages of patients with adverse effects were similar in the placebo and cA2 groups. Of the 29 patients who received two cA2 infusions, 2 had a reaction (chest pain, dyspnea, or nausea) that led to the discontinuation of the infusion. These reactions resolved spontaneously within minutes after the infusion was discontinued. In addition, complications requiring hospitalization developed in two patients (abdominal abscess in one

patient in the placebo group and salmonella colitis in one patient given an infusion of 20 mg of cA2 per kilogram). Both patients were treated successfully. A bowel obstruction occurred in one patient given cA2, and a flare of Crohn's disease occurred in another.

Immunologic Responses

Serum samples were screened for antibodies to double-stranded DNA at base line and at 12 weeks in 98 patients who received cA2 (in either a blinded

TABLE 3. ADVERSE EFFECTS.*

VARIABLE	PLACEBO	ONE DOSE OF cA2	TWO DOSES OF cA2
No. of patients evaluated	25	102	29
Average length of follow-up — wk	6.9	10.4	12.4
Adverse effect — no. of patients (%)			
Any adverse effect	15 (60)	76 (75)	23 (79)
Headache	5 (20)	19 (19)	3 (10)
Nausea	2 (8)	11 (11)	5 (17)
Upper respiratory tract infection	3 (12)	8 (8)	4 (14)
Fatigue	1 (4)	6 (6)	3 (10)
Myalgia	1 (4)	4 (4)	3 (10)
Rhinitis	1 (4)	3 (3)	3 (10)
Pain	0	4 (4)	3 (10)
Pruritus	1 (4)	1 (1)	4 (14)
Chest pain	1 (4)	2 (2)	3 (10)
Vomiting	0	2 (2)	3 (10)
Dyspnea	0	1 (1)	3 (10)

*Adverse effects that occurred in 10 percent or more of the patients in any of the groups are reported.

or an open-label infusion); all patients were negative for these antibodies at base line, and 3 were positive at 12 weeks. Serum samples from 101 patients treated with cA2 (in either a blinded or an open-label infusion) were also tested for human anti-cA2; 6 patients tested positive. In two thirds of the patients, however, cA2 was still detectable in serum samples taken at 12 weeks, and this may have interfered with the assay.

DISCUSSION

Many of our patients with moderate-to-severe Crohn's disease that was resistant to treatment had a rapid response to cA2. Other treatments for such patients have not had beneficial effects over the long term and may not be well tolerated.³⁰⁻³³ Our short-term study suggests that anti-TNF- α therapy with cA2 may represent a new treatment option for patients with moderate-to-severe Crohn's disease. Further studies will be necessary to determine the long-term efficacy of a single infusion of cA2 as well as the efficacy and safety of repeated treatments.

The mechanisms of the chronic mucosal inflammatory processes manifesting as Crohn's disease are not clear. However, our results add strength to the suggestion that TNF- α may have a central role in the inflammatory process in at least two thirds of the patients with Crohn's disease.²² Studies in rodent models have provided insight into the role of anti-TNF- α in the pathogenesis of Crohn's disease. Studies involving the transfer of CD45RB^{high} cells to mice with severe combined immunodeficiency have shown that these cells can induce a chronic, transmural inflammatory process in the colon.³⁴ Powrie et al. have demonstrated that this colitis can be reversed or ameliorated by the use of agents, including anti-TNF- α , capable of down-regulating the production of the T helper 1 subclass of T cells.¹⁰ These cells produce proinflammatory cytokines including interferon- γ , interleukin-2, and potentially, TNF. A mechanistic role for TNF- α in intestinal inflammation has been suggested on the basis of these studies.

Other studies involving animal models have shown that cytokines produced by the T helper 1 subclass are expressed in the mucosa of people with Crohn's disease.^{17,18} Anti-TNF- α may participate in the down-regulation of mucosal inflammation in this disease by inhibiting the T helper 1 population of active T cells. Extensive follow-up studies are needed to determine how the elimination of TNF affects mucosal inflammation after treatment with anti-TNF- α .

In this study, a clinical response or remission occurred in 65 percent of patients with severe Crohn's disease after a single infusion of cA2. Furthermore, results in the open-label retreatment phase corroborated the finding that patients with no response to the first infusion of cA2 were less likely to have a response to a second infusion. Thus, this group of pa-

tients may differ from those that responded. The similarities among the patients, including age, duration of disease, types of concomitant treatment, and disease activity at base line, suggest that any differences are more likely to be detected subclinically. There was no apparent dose-response relation between a dose of 5 mg of cA2 per kilogram and a dose of 20 mg per kilogram with respect to either the magnitude or the duration of the clinical response. In a previous open-label trial of cA2 doses of 1 mg, 5 mg, 10 mg, and 20 mg per kilogram, the group receiving 1 mg of cA2 per kilogram had a more transient response than the groups given the higher doses.³⁵ This transient response was similar to that in a small trial of a different anti-TNF- α antibody.³⁶ The results of these trials support the use of a dose of 5 mg of cA2 per kilogram in future trials.

In summary, we found that a single infusion of cA2 was an effective short-term treatment for patients with moderate-to-severe Crohn's disease that was resistant to treatment.

Supported by Centocor, Inc., and by a grant (FD-R-001276) from the Food and Drug Administration Orphan Products Development Division. Drs. Hanauer, van Deventer, Present, and Rutgeerts have received honorariums from Centocor for lectures.

APPENDIX

The Crohn's Disease cA2 Study Group consists of the following centers and investigators (the number of patients enrolled at each center is given in parentheses): *Massachusetts General Hospital, Boston* (5 patients): D. Podolsky, B.E. Sands, M.T. Marcucci; *Cedars-Sinai Medical Center, Los Angeles* (20 patients): S.R. Targan, E.A. Vasiliauskas, B. Voigt, J. Gaiennie; *University of Chicago Hospitals and Clinics, Chicago* (11 patients): S.B. Hanauer; *University of Alabama School of Medicine, Birmingham* (2 patients): C.O. Elson, R.P. McCabe, Jr.; *Mount Sinai Medical Center, New York* (8 patients): L. Mayer, D.H. Present, C. Stamaty; *Washington University School of Medicine, St. Louis* (2 patients): W.F. Stenson, J.J. O'Brien; *Virginia Mason Medical Center, Seattle* (5 patients): R. Kozarek, M. Gelfand; *Hospital of the University of Pennsylvania, Philadelphia* (4 patients): D. Bachwich, G. Lichtenstein, L. Hurd; *McMaster University Medical Center, Hamilton, Ont., Canada* (2 patients): E.J. Irvine, S. Collins; *Lahey Clinic, Burlington, Mass.* (3 patients): A.S. Warner, L.J. Costa; *University of North Carolina, Chapel Hill* (5 patients): K.L. Isaacs; *University of Maryland Medical System, Baltimore* (5 patients): S. James, B. Greenwald, M.L. Mullen; *University of Kentucky, Lexington* (5 patients): G.W. Varelek, B. Vivian; *Academisch Ziekenhuis Leiden, Leiden, the Netherlands* (4 patients): R.A. van Hogezaand, M.J. Wagtmans; *Institute for Clinical Immunology and Rheumatology, Erlangen, Germany* (1 patient): H. Schönekäs, J.R. Kalden, J.M.L. Bauer; *University of Amsterdam, Amsterdam* (9 patients): S.J.H. van Deventer, C.M.J. Kothe, O.J.B. de Smit; *Leeds General Infirmary, Leeds, United Kingdom* (4 patients): D.M. Chalmers, S. Chitturi, D. Todi; and *Academisch Ziekenhuis Gasthuisberg, Leuven, Belgium* (13 patients): P.J. Rutgeerts, G.R.A.M. D'Haens, A.F.M. Verstraeten.

REFERENCES

- Levine DS. Clinical features and complications of Crohn's disease. In: Targan S, Shanahan F, eds. *Inflammatory bowel disease: from bench to bedside*. Baltimore: Williams & Wilkins, 1993:296-316.
- Plevy SE, Targan SR. Specific management of Crohn's disease. In: Targan S, Shanahan F, eds. *Inflammatory bowel disease: from bench to bedside*. Baltimore: Williams & Wilkins, 1993:582-609.
- O'Brien HH, Bayless TM, Bayless JA. Use of azathioprine or 6-mercaptopurine in the treatment of Crohn's disease. *Gastroenterology* 1991;101:39-46.
- Feagan BG, Rochon J, Fedorak RN, et al. Methotrexate for the treatment of Crohn's disease. *N Engl J Med* 1995;332:292-7.
- Hanauer SB. Inflammatory bowel disease. *N Engl J Med* 1996;334:841-8. [Erratum, *N Engl J Med* 1996;335:143.]

6. Brynskov J, Freund L, Rasmussen SN, et al. A placebo controlled, double-blind, randomized trial of cyclosporine therapy in active chronic Crohn's disease. *N Engl J Med* 1989;321:845-50.
7. Feagan BG, McDonald JWD, Rochon J, et al. Low-dose cyclosporine for the treatment of Crohn's disease. *N Engl J Med* 1994;330:1846-51.
8. Jewell DP, Lennard-Jones JE, Cyclosporin Study Group of Great Britain and Ireland. Oral cyclosporin for chronic active Crohn's disease: a multicentre controlled trial. *Eur J Gastroenterol Hepatol* 1994;6:499-505.
9. Stange EF, Modigliani R, Peña AS, et al. European trial of cyclosporine in chronic active Crohn's disease: a 12 month study. *Gastroenterology* 1995;109:774-82.
10. Powrie F, Lesch MW, Mauze S, Menon S, Caddle LB, Coffman RL. Inhibition of Th1 responses prevents inflammatory bowel disease in scid mice reconstituted with CD45RBhi CD4+ T cells. *Immunity* 1994;1:553-62.
11. Neurath MF, Fuss I, Kelsall BL, Presky DH, Waegell W, Strober W. Experimental granulomatous colitis in mice is abrogated by induction of TGF- β -mediated oral tolerance. *J Exp Med* 1996;183:2605-16.
12. Davidson NJ, Leach MW, Fort MM, et al. T helper cell 1-type CD4+ T cells, but not B cells, mediate colitis in interleukin 10-deficient mice. *J Exp Med* 1996;184:241-51.
13. Mosmann TR, Coffman RL. TH1 and TH2 cells: different patterns of lymphokine secretion lead to different functional properties. *Annu Rev Immunol* 1989;7:145-73.
14. Pirmez C, Yamamura M, Uyemura K, Paes-Oliveira M, Conceicao-Silva F, Modlin RL. Cytokine patterns in the pathogenesis of human leishmaniasis. *J Clin Invest* 1993;91:1390-5.
15. MacDonald TT, Hutchings P, Choy M-Y, Murch S, Cooke A. Tumor necrosis factor-alpha and interferon-gamma production measured at the single cell level in normal and inflamed human intestine. *Clin Exp Immunol* 1990;81:301-5.
16. Murch SH, Braegger CP, Walker-Smith JA, MacDonald TT. Location of tumour necrosis factor α by immunohistochemistry in chronic inflammatory bowel disease. *Gut* 1993;34:1705-9.
17. Mullin GE, Lazenby AJ, Harris ML, Bayless TM, James SP. Increased interleukin-2 messenger RNA in the intestinal mucosal lesions of Crohn's disease but not ulcerative colitis. *Gastroenterology* 1992;102:1620-7.
18. Fuss IJ, Neurath M, Boirivant M, et al. Disparate CD4+ lamina propria (LP) lymphokine secretion profiles in inflammatory bowel disease: Crohn's disease LP cells manifest increased secretion of IFN-gamma, whereas ulcerative colitis LP cells manifest increased secretion of IL-5. *J Immunol* 1996;157:1261-70.
19. Maini RN, Elliott MJ, Brennan FM, Feldmann M. Beneficial effects of tumour necrosis factor-alpha (TNF- α) blockade in rheumatoid arthritis. *Clin Exp Immunol* 1995;101:207-12. [Erratum, *Clin Exp Immunol* 1995;102:443.]
20. Cope AP, Londei M, Chu NR, et al. Chronic exposure to tumor necrosis factor (TNF) in vitro impairs the activation of T cells through the T cell receptor/CD3 complex: reversal in vivo by anti-TNF antibodies in patients with rheumatoid arthritis. *J Clin Invest* 1994;94:749-60.
21. Elliott MJ, Maini RN, Feldmann M, 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.
22. van Dullemen HM, van Deventer SJH, Hommes DW, et al. Treatment of Crohn's disease with anti-tumor necrosis factor chimeric monoclonal antibody (cA2). *Gastroenterology* 1995;109:129-35.
23. Best WR, Becktel JM, Singleton JW. Rederived values of the eight coefficients of the Crohn's Disease Activity Index (CDAI). *Gastroenterology* 1979;77:843-6.
24. Irvine EJ, Feagan B, Rochon J, et al. Quality of life: a valid and reliable measure of therapeutic efficacy in the treatment of inflammatory bowel disease. *Gastroenterology* 1994;106:287-96.
25. Knight DM, Trinh H, Le J, et al. Construction and initial characterization of a mouse-human chimeric anti-TNF antibody. *Mol Immunol* 1993;30:1443-53.
26. Scallon BJ, Moore MA, Trinh H, Knight DM, Ghraieb J. Chimeric anti-TNF-alpha monoclonal antibody cA2 binds recombinant transmembrane TNF-alpha and activates immune effector functions. *Cytokine* 1995;7:251-9.
27. Siegel SA, Shealy DJ, Nakada MT, et al. The mouse/human chimeric monoclonal antibody cA2 neutralizes TNF in vitro and protects transgenic mice from cachexia and TNF lethality in vivo. *Cytokine* 1995;7:15-25.
28. Agresti A. Categorical data analysis. New York: John Wiley, 1990:230-5.
29. Conover WJ. Practical nonparametric statistics. 2nd ed. New York: John Wiley, 1980:318-26.
30. Stronkhorst A, Tytgat GN, van Deventer SJ. CD4 antibody treatment in Crohn's disease. *Scand J Gastroenterol Suppl* 1992;194:61-5.
31. Emmerich J, Seyfarth M, Fleig WE, Emmerich F. Treatment of inflammatory bowel disease with anti-CD4 monoclonal antibody. *Lancet* 1991;338:570-1.
32. Sandborn WJ. A critical review of cyclosporine therapy in inflammatory bowel disease. *Inflam Bowel Dis* 1995;1:48-63.
33. Sachar DB. Maintenance therapy in ulcerative colitis and Crohn's disease. *J Clin Gastroenterol* 1995;20:117-22.
34. Powrie F, Leach MW, Mauze S, Caddle LB, Coffman RL. Phenotypically distinct subsets of CD4+ T cells induce or protect from chronic intestinal inflammation in C. B-17 scid mice. *Int Immunol* 1993;5:1461-71.
35. McCabe RP, Woody J, van Deventer S, et al. A multicenter trial of cA2 anti-TNF chimeric monoclonal antibody in patients with active Crohn's disease. *Gastroenterology* 1996;110:Suppl:A962. abstract.
36. Stack WA, Mann SD, Roy AJ, et al. Randomised controlled trial of CDP571 antibody to tumour necrosis factor-alpha in Crohn's disease. *Lancet* 1997;349:521-4.