Changes in Acute Phase Proteins after Anti-Tumor Necrosis Factor Antibody (Infliximab) Treatment in Patients with Crohn´s Disease

V. KUPČOVÁ¹, L. TURECKÝ², Z. DETKOVÁ¹, M. PRÍKAZSKÁ³, A. KELEOVÁ³

¹Third Department of Internal Medicine, Dérer´s Hospital, Medical Faculty of Comenius University, ²Institute of Chemistry, Biochemistry and Clinical Biochemistry, Medical Faculty of Comenius University, and ³Institute of Preventive and Clinical Medicine, Bratislava, Slovak Republic

Received February 26, 2002
Accepted July 2, 2002

Summary
Acute phase proteins and markers of proteosynthetic activity reflect the clinical activity in Crohn’s disease (CD). The impact of anti-tumor necrosis factor antibody (anti-TNF) therapy on serum levels of acute phase proteins and proteosynthetic markers was studied. Fourteen patients with active CD were treated with 5 mg per kg of anti-TNF in intravenous infusion. Clinical activity (assessed by Crohn’s disease activity index – CDAI), α-1-acid glycoprotein, haptoglobin, cholinesterase and prealbumin were assessed before and in months 1 and 5 after treatment. A sustained decrease in CDAI was observed. This was accompanied by a significant decrease in α-1-acid glycoprotein and haptoglobin in month 1 (p=0.005 and p=0.01, respectively) while in month 5 the levels of both acute phase proteins rose significantly (p=0.003 for α-1-acid glycoprotein and p=0.02 for haptoglobin). Cholinesterase and prealbumin significantly increased in month 1 after the treatment (p=0.02 and p=0.0006, respectively), the increase was sustained in cholinesterase while prealbumin levels diminished in month 5. We conclude that the clinical improvement after anti-TNF therapy for CD is accompanied by changes of acute phase proteins and proteosynthetic markers. The assessment of these laboratory markers may be useful in the management of CD patients treated with anti-TNF.

Key words
Crohn’s disease • Anti-tumor necrosis factor antibody • Acute phase proteins • Cholinesterase • Prealbumin

Introduction
Crohn’s disease (CD) is an inflammation of the intestinal mucosa which is mediated by T helper 1 (Th 1) cytokines (Mullin et al. 1992, Pullman et al. 1992) that stimulate hepatic synthesis of acute phase proteins, namely α-1-acid glycoprotein (Pous et al. 1990) and haptoglobin (Raynes et al. 1991).

Clinical studies showed that acute phase proteins rise in clinically active CD and correlate with the clinical activity (Andre et al. 1981, Cellier et al. 1994). On the other hand, the synthesis of proteosynthetic markers
(cholinesterase, albumin, prealbumin) is diminished in active CD (Richter et al. 1982, Tromm et al. 1992).

A novel therapeutic procedure for CD, chimeric anti-tumor necrosis factor antibody (anti-TNF, Infliximab) down-regulates the Th 1 response (Plevy et al. 1997, Baert et al. 1999) and seems to have a considerable effect on clinical, histological and endoscopic activity (Targan et al. 1997, D'Haens et al. 1999). Therefore, the administration of this agent in CD may be followed by changes of acute phase proteins and proteosynthetic markers.

In the study presented here, acute phase proteins and markers of proteosynthetic activity were assessed in CD patients treated with anti-TNF together with the follow-up of the clinical therapeutic effect.

Methods

Fourteen patients (9 women and 5 men, mean age 36 ± 9 years) with active moderate to severe CD were treated with 5 mg per kg of anti-TNF in intravenous infusion. Concomitant medication, 5-aminosalicylic acid-containing drugs (13 patients) and corticosteroids (3 patients), remained unchanged. Before the patients had been included in the study, basic hematological (blood count, parameters of coagulation) and biochemical (urea, creatinine, aspartate aminotransferase, alanine aminotransferase, γ-glutamyl transpeptidase, alkaline phosphatase, sodium, potassium) parameters were checked together with the chest X-ray in order to exclude any contraindication against the treatment with anti-TNF. Clinical activity, α-1-acid glycoprotein, haptoglobin, cholinesterase and prealbumin were assessed in all patients before treatment (Sample 1), in month 1 after treatment (Sample 2) and in 12 patients in month 5 after the administration of anti-TNF (Sample 3). At the same time, basic hematological (blood count, parameters of coagulation) and biochemical (urea, creatinine, aspartate aminotransferase, alanine aminotransferase, γ-glutamyl transpeptidase, alkaline phosphatase, sodium, potassium) parameters were followed in all patients.

Clinical activity was assessed by Crohn's disease activity index (CDAI) (Best et al. 1979) that represents a composite measure of frequency of loose bowel movements, severity of abdominal pain, general well-being, extraintestinal manifestations, presence of abdominal mass, use of anti diarrheal drugs, hematocrit and body weight.

Serum α-1-acid glycoprotein, haptoglobin, cholinesterase and prealbumin levels were obtained by immunodiffusion method (Laurell 1966) using a monospecific antiserum (USOL Prague).

The study was conducted with the agreement of the Institutional Ethics Board and an informed consent was obtained from each patient.

For statistical analysis non-parametric methods were used. Data are expressed as means and standard deviations and were evaluated by Wilcoxon’s test with a significance limit of p<0.05.

Results

Treatment was well tolerated, no side-effects necessitating interruption of the infusion were observed. After the treatment, no important changes in basic hematological and biochemical parameters were noted. In clinical activity, there was a significant drop in CDAI from 236 ± 54 before treatment (Sample 1) to 132 ± 76 in month 1 after treatment (Sample 2, p=0.0003). During the further follow-up a slight non-significant increase in CDAI to 145 ± 93 was observed in month 5 (Sample 3, Fig. 1).

![Fig. 1. Changes in Crohn’s disease activity index (CDAI, means ± SD), *p<0.05.](image)

In both biochemical markers of inflammatory activity, α-1-acid glycoprotein and haptoglobin, a significant decrease in month 1 (Sample 2) was noticed, followed by an increase in month 5 (Sample 3, Figs 2a,b). α-1-acid glycoprotein levels dropped from 1138 ± 411 mg/l before treatment (Sample 1) to 880 ± 230 mg/l in month 1 (Sample 2, p=0.005); this decrease was followed by a significant increase in month 5 (Sample 3) to 1207 ± 376 mg/l (p=0.003). Haptoglobin decreased from 1721 ± 894 mg/l (Sample 1) to 1179 ± 613 mg/l (Sample 2),
A sustained, significant increase was observed in cholinesterase and prealbumin levels (Figs 2c,d). Cholinesterase levels rose from $55.9 \pm 14.6 \, \mu$kat/l (Sample 1) to $62.6 \pm 16.5 \, \mu$kat/l (Sample 2, p=0.02), and $63 \pm 18.2 \, \mu$kat/l (Sample 3). Prealbumin increased from $215 \pm 47 \, \text{mg/l}$ (Sample 1) to $283 \pm 53 \, \text{mg/l}$ (Sample 2, p=0.0006); this was followed by a slight decrease to values of $245 \pm 54 \, \text{mg/l}$ (Sample 3) that were still significantly higher than the values obtained before treatment (p=0.01).

**Fig. 2.** Changes in $\alpha$-1-acid glycoprotein (2a), haptoglobin (2b), cholinesterase (2c), prealbumin (2d), before treatment (Sample 1), in month 1 (Sample 2) and in month 5 (Sample 3), data are presented as means ± SD, * p<0.05.

**Discussion**

In the present study we observed that both acute phase proteins studied ($\alpha$-1-acid glycoprotein and haptoglobin) diminished significantly after anti-TNF therapy for active CD. Together with a decrease in biochemical markers of inflammation, clinical improvement was observed. $\alpha$-1-acid glycoprotein is considered to be a good laboratory marker of inflammatory activity in CD (Lubega and Davies 1990) whilst haptoglobin, probably due to its polymorphism (Turecký et al. 2001), seems to be less reliable (Lubega and Davies 1990). $\alpha$-1-acid glycoprotein synthesis is enhanced in response to proinflammatory cytokines (interleukin 1, tumor necrosis factor-$\alpha$ – TNF-$\alpha$) (Pous et al. 1990) and these cytokines are thought to mediate the inflammation in CD. Therefore, anti-TNF therapy that is targeting the key proinflammatory cytokine in CD pathogenesis, TNF-$\alpha$, is expected to be related to changes in acute phase protein levels as observed in the present study.

However, in the further follow-up only a slight, non-significant increase in clinical activity was observed whilst both acute phase proteins raised significantly and, in the case of $\alpha$-1-acid glycoprotein, exceeded levels observed before treatment. The possibility of a flare of an opportunistic infection was excluded on the basis of the absence of medical history, normal clinical examination, and a physiological leukocyte count. $\alpha$-1-acid glycoprotein seems to predict a clinical relapse in CD (Brignola et al. 1986, Wright et al. 1987, Louis et al. 1997) that may account for the reported difference.
between clinical and laboratory observations. A close further follow-up of these patients is therefore needed with the aim of early therapeutic adjustment.

The two other biochemical parameters studied, cholinesterase and prealbumin, showed a significant increase in response to anti-TNF treatment in active CD. The increase was sustained in cholinesterase whilst prealbumin levels diminished after the initial rise. Prealbumin is a rapid turnover marker of proteosynthetic status (Malejev et al. 1979) and the ratio of α-1-acid glycoprotein and prealbumin was found to correlate with clinical (Richter et al. 1982) and endoscopic (Simonis et al. 1998) activity in CD. The observation of the decreasing level of prealbumin would therefore be in accordance with the above-mentioned increase in α-1-acid glycoprotein.

Cholinesterase is a marker of proteosynthetic activity (Malejev et al. 1979). Its serum levels were found to be decreased in active CD and it has been suggested that this decrease might be explained by a suppression of cholinesterase synthesis in severe CD mediated by endotoxins and cytokines rather than by an increased intestinal loss (Tromm et al. 1992). Administration of anti-TNF was found to reduce the number of lamina propria mononuclear cells producing TNF and interferon-gamma (Plevy et al. 1997) and the observed increase in cholinesterase after anti-TNF therapy might be related to diminished production of CHE synthesis suppressing cytokines.

Based on the above presented results, we can conclude that the clinical improvement after anti-TNF therapy in active CD is accompanied by changes in acute phase proteins, namely α-1-acid glycoprotein and haptoglobin, as well as in proteosynthetic markers, cholinesterase and prealbumin. The assessment of these laboratory markers may be useful in the further management of CD patients treated with anti-TNF.

Acknowledgements
The authors are grateful to Mrs H. Jahodníková for her help in laboratory and secretarial work.

References


---

**Reprint requests**
Doc. Dr. Kupčová Viera, CSc., Department of Internal Medicine, Dérer’s Hospital, Medical Faculty of Comenius University, Limbova 5, 833 05 Bratislava, Slovak Republic, e-mail: kupcova@cent.sk