MISSION STATEMENT

The mission of the journal is to serve the need of the internist to practice up-to-date medicine and to keep track with important issues in health care. With this purpose we publish editorials, original articles, reviews, controversies, consensus reports, papers on speciality training and medical education, book reviews and correspondence.

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Alphen aan den Rijn, the Netherlands
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Exposition ‘13 covers of the Netherlands Journal of Medicine’

We are pleased to invite you to the opening of gallery Unita’s latest exposition entitled ‘13 covers of the Netherlands Journal of Medicine’.

On Sunday 6 April 2003, Professor J.W.M. van der Meer, internist at St Radboud Hospital in Nijmegen and editor in chief of the Netherlands Journal of Medicine, will open the exposition.

The exposition includes graphic works by Klaas Gubbels, Annemari Petri, Bob Lejeune, Ruud Matthes, Els Maasson, Jops Jacobs, Miranda Penning, Karin Elfrink, Carole Witteveen, José Veugen, Gerdien Kroes, Bart Elfrink and Caroline Koenders.

The exposition is open every Sunday from 2 p.m. to 5 p.m. and upon request on Thursday, Friday or Saturday afternoons. The exposition runs until Sunday 11 May 2003.

Galery ‘Unita’
Rijksstraatweg 109
Beek/Ubbergen
tel.: +31 (0)24-684 40 49

These works of art will also be exhibited on the ‘Internistendagen’, 15 and 16 May 2003, MECC Maastricht, the Netherlands.

The winning abstract, presented at the ‘Internistendagen’, will be awarded one of these works (at the winners choice).
There are a number of reasons to evaluate the first year of the renewed Netherlands Journal of Medicine. As our readership knows, important changes to the journal took place in 2002. Firstly, the editorial board moved from Utrecht to Nijmegen, and at the same time it was decided to search for another, perhaps more flexible, publisher. Such changes may have had a negative influence on the citation index of the journal. For instance, we could not start with the first renewed issue until March 2002. Another disadvantage was that the publisher had to develop a complete electronic edition. The publisher has succeeded in this task and from now on the journal is available on the Internet with an attractive website (www.njmonline.nl).

There are some other editorial points to make. In the December 2002 issue of this journal, we mentioned that all submissions in all categories should include an abstract. Further, it was agreed that the editors would be selective in publishing abstracts and supplements on official journal pages, because this has a negative influence on the impact factor, as determined by the journal of citation reports. Table 1 shows the increase in the impact factor over the last four years as well as the rise of the journal on the list, from place 58 out of 107 to place 40 out of 112 journals, in the subject category medicine, general and internal. To be honest, we have to give our predecessors the credit for this spectacular advance. We hope that in this first, difficult year we have maintained this position and that the coming years will show a further increase.

The fact that the journal is again easily accessible on the net will hopefully contribute to a further improvement in the position of our journal.

Finally, we as editors would like to offer the following as food for thought. We are worried about the relatively low number of original articles submitted to us. We do, of course, appreciate that your most important and revolutionary work should be submitted to the top five to ten journals. But when your work is not suitable for the highest-ranked international journals, why go for an international journal in the subtop? Why not choose the Netherlands Journal of Medicine instead?

We are aware of the academic pressure to publish in journals with the highest possible impact. But should we also prefer the Scandinavian-based ‘Journal of Internal Medicine’ (formerly Acta Medica Scandinavia) or the Medical Journal of Australia purely for the higher impact factor and the higher chance of citation while your Dutch colleagues, and in particular the next generation of internists, are not aware of your contribution?

A last point of concern is the time between submission and the moment of acceptance: for 2002 this period amounted to an average of 98 days, which we consider much too long. Therefore, we need your support as potential referees to shorten the reviewing time. The next responsibility is for the authors; they should resubmit their revised versions more rapidly. Even for minor revisions, some authors take as long as three months.

After this mixture of good and less good news, the editors will work together with you to further improve your and ‘our’ journal.

Table 1

<table>
<thead>
<tr>
<th>YEAR</th>
<th>IMPACT FACTOR</th>
<th>JOURNALS IN CATEGORY MEDICINE GENERAL AND INTERNAL</th>
<th>PLACE NJM</th>
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<tbody>
<tr>
<td>1998</td>
<td>0.429</td>
<td>107</td>
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<td>1999</td>
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<tr>
<td>2001</td>
<td>0.925</td>
<td>112</td>
<td>40</td>
</tr>
</tbody>
</table>

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Reference

Infliximab therapy in Crohn’s disease: safety issues

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ABSTRACT

Infliximab has become a valuable addition to the therapeutic arsenal for Crohn’s disease. Although the rate of adverse events was relatively low in the premarketing trials, several investigators have recently reported experience in large groups of patients. This has shed more light on safety aspects of infliximab therapy, which should change the approach towards patients prior to infliximab infusion. This review discusses some immunological aspects that are relevant for infliximab therapy and provides guidelines for daily practice.

INTRODUCTION

It is only a decade ago that the first patient received anti-TNF antibodies for severe Crohn’s disease (CD), demonstrating a remarkable response after a single intravenous gift. Today, more than 300,000 patients, both with CD and with rheumatoid arthritis (RA), have been treated with infliximab. Infliximab can induce and maintain remission of Crohn’s disease that is refractory to standard therapy, while reducing or eliminating the need for steroid use. The success of this innovative therapeutic strategy is to a great extent the result of a better understanding of the immunological defects in the gut mucosa. Although the addition of infliximab to the therapeutic arsenal in CD has made a substantial impact on the daily life of large groups of patients, it is now known that this therapy is associated with immunosuppression that greatly increases the susceptibility to certain infections. In addition, infliximab is immunogenic, and is associated with the induction of antidouble-stranded DNA (anti-ds-DNA) antibody formation. Moreover, several important questions with regard to the safety of infliximab therapy remain to be answered. These questions include the long-term toxicity of infliximab, assessment of toxicity in patients with comorbid diseases, identifying patients at risk for infliximab toxicity, and evaluation of the effect of drug combinations on toxicity. This review will summarise the current knowledge on the safety profile of infliximab and attempt to bring these facts into a practical framework for physicians frequently treating CD patients.

GENERAL SAFETY ISSUES

Safety issues relating to biotechnologically manufactured drugs have been categorised into four groups.

Group 1: infectious agents
There is a risk of infectious agents originating from the cell lines used in the manufacturing process being transmitted to patients. This concern has been particularly focussed on viral contamination. For this purpose, the hybridoma cell line responsible for the production of the monoclonal antibody as well as the cell bank and end product are extensively screened, and no contaminating viruses have been detected.

Group 2: hypersensitivity
The likelihood that biotechnologically manufactured drugs will produce a host immune response is dependent on the quantity of ‘foreign’ material. A murine type of antibody will often induce hypersensitivity reactions, whereas with recombinant human antibodies this is not the case. With
infliximab, these hypersensitivity reactions occur in about 2% of patients, and associated symptoms include headache, nausea, shortness of breath, flushing, dizziness and pruritus (discussed below). Apparently, such reactions are mediated by mast cell release, but the precise mechanism has not been characterised. Infliximab is an engineered IgG1 murine human chimeric monoclonal antibody containing 75% human protein and 25% murine protein. The murine ‘business end’ of the molecule is immunogenic, and human antichimeric antibodies (HACAs) occur in 28% of patients. HACA formation is associated with an increase in the incidence of infusion reactions, and the formation of immune complexes can give rise to a syndrome that resembles serum sickness.

Three strategies can be employed to minimise these hypersensitivity reactions: first, co-administration of other immunosuppressive drugs has been shown to reduce the risk of sensitisation to therapeutic antibodies. Secondly, desensitisation is a strategy to diminish specific IgE-induced responses towards a specific therapeutic antibody. In general, small but increasing quantities of the antigen-containing antibody are administered over a period of hours or more gradually over weeks or months. As a result, specific IgE levels decrease and possibly specific T-cell tolerance is induced. The role of IgE during infliximab hypersensitivity reactions has not been established, however successful desensitisation has been reported in two Crohn’s disease patients. Finally, therapeutic biologicals can be ‘humanised’ for instance by grafting a complementarity determining region into the variable domain of a human antibody or by producing wholly humanised monoclonal antibodies.

**Group 3: opportunistic infections or neoplasm during biological treatment**

Integrity of the immune system is essential for defence against infectious organisms and the control of tumour antigens. Although our enthusiasm over the use of targeted immunotherapy is enormous, long-term immunosuppression holds the risk of interfering with the coherence of the immune system. A clear example has been the reactivation of tuberculosis (TBC) in patients treated with infliximab (this will be discussed further in this article). Although the postmarketing surveillance has not shown any increased risk of cancer yet, other biological therapy has been associated with for instance lymphomas.

**Group 4: monoclonal antibodies producing cell lysis of activated macrophages and T cells**

Antibodies directed against cell surface proteins are capable of inducing cell death either by activation of the complement system or by the induction of apoptosis. A sudden release of cytokines from the targeted immunocompetent cells can result in clinical symptoms of fever, chills and organ dysfunction. This problem can be overcome by only utilising the F(ab)2 fragments of the antibody, thereby retaining the capacity of antigen binding, but eliminating the Fc-fragment’s ability to activate complement. Although the mechanism of action of infliximab is thought to be related to induction of apoptosis of T lymphocytes, in contrast to other monoclonal antibodies (OKT3, anti-CD4 antibodies), no cell lysis or cytokine release syndromes are observed after infliximab infusion.

**REPORTED ADVERSE EVENTS**

Data from the first preclinical studies in both RA and CD patients did not reveal major safety concerns, partly because some of the adverse events occurred after a drug holiday. During recent years, numerous studies have been published, reporting on various events including infusion reactions, delayed-type hypersensitivity reactions, formation of HACAs and infections. We will review the most important adverse reactions and propose several management guidelines.

**Infusion reaction**

**Definition:** An infusion reaction is defined as any adverse event that occurs during or within two hours following an infusion of infliximab.

**Type of hypersensitivity:** Anaphylactoid (IgE independent).

**Signs and symptoms:** Most infusion reactions included headache, nausea, chest pain, dizziness, urticaria, dyspnoea, pruritus and flushing. These symptoms are usually self-limiting.

**Complication:** Severe anaphylaxis including shock, laryngeal oedema and bronchospasms.

**Management:** To stop infliximab infusion temporarily, and administer antihistamines and corticosteroids intravenously (i.e. 2 mg clemastine i.v. and 25 mg prednisolone i.v.). When symptoms have resolved, restart the infusion at a slower rate. Most patients respond adequately to treatment and can complete the infusion. Patients who have already experienced an infusion reaction should receive antihistamines and corticosteroids intravenously (i.e. 2 mg clemastine i.v. and 25 mg prednisolone i.v.) 30 minutes prior to infusion.

**Comment:** Infusion reactions develop in approximately 22% of infliximab-treated patients compared with 9% of placebo-treated patients. They are similar to infusion reactions observed during administration of intravenous immunoglobulins. Although severe anaphylactic reaction is rarely observed in infliximab-treated patients, if it does occur infliximab therapy should be discontinued. Successful desensitisation and therapeutic infusion using parenteral dose escalation in an intensive care unit setting has recently been reported. Infusion reactions are more common in patients who have developed HACAs. The
formation of HACAs occurs in approximately 28% of infliximab-treated patients, although higher rates have also been reported. Concomitant therapy with methotrexate, azathioprine or 6-mercaptopurine reduces the incidence of HACA formation, and probably reduces the rate of infusion reactions. The aforementioned premedication does not seem mandatory in patients who did not experience any infusion reaction during previous infusions. We are of the opinion that if the decision has been made to initiate infliximab treatment, be it for remission induction or maintenance therapy, then all patients should at the same time start an immunosuppressive drug (i.e. azathioprine, 6-MP or methotrexate).

Delayed-type hypersensitivity reaction (DTH)
Definition: An immune reaction in which T cell-dependent macrophage activation and cytokine-mediated inflammation cause tissue injury. This type of reaction usually occurs three to 12 days after infliximab infusion in a patient who had previously been treated with infliximab.

Type of hypersensitivity: Type IV hypersensitivity disorder.
Signs and symptoms: Days after the infliximab readministration, patients can develop myalgia, rash, fever, polyarthralgias, pruritus, facial, hand or lip oedema, urticaria, sore throat, dysphagia and headache. The incidence of DTH seems low, and was reported to be 2% in the ACCENT I study. Complication: A severe DTH reaction resulting in adult respiratory distress syndrome (ARDS) in a patient who received infliximab after a drug holiday of 15 months was recently reported.

Management: In our opinion, the occurrence of a DTH does not exclude patients from future infliximab re-infusion. However, our advice is to administer premedication, similar to that given during infusion reactions, on following infusions. Also, the use of concomitant immunosuppressive medication seems necessary. Naturally, infliximab therapy is discouraged in patients who have experienced a severe episode of DTH requiring hospitalisation.

Comment: Parallel to infusion reactions, the occurrence of DTH is probably more frequent in HACA-positive patients. Again, we would like to underscore the necessity to initiate immunosuppressive drugs alongside the infliximab treatment period. Since the majority of patients will neither experience infusion reactions or DTH, we do not recommend HACA assessment routinely in infliximab-treated patients.

Serum sickness
Definition: An immune reaction caused by a large dose of protein antigen (i.e. chimeric antibody) resulting in the deposition of antigen antibody complexes in blood vessel walls, especially in the kidneys and joints.

Type of hypersensitivity: Type III hypersensitivity disorder.
Signs and symptoms: Within days to weeks after the infliximab infusion, symptoms of skin rash, fever, myalgias, polyarthritis and even glomerulonephritis develop as a result of immune complex deposition, complement activation and neutrophil-driven inflammation.

Management: Management is similar to that of DTH; if a patient suffers from an episode of serum sickness, premedication is mandatory upon infliximab readministration as well as the use of an immunosuppressant. During such an episode, patients can be treated with a short course of prednisolone. The use of infliximab must be stopped, however, if symptoms have not resolved completely (i.e. renal function, arthralgias).

Comment: The incidence of serum sickness or a serum sickness-like reaction is probably lower than that of DTH. In our experience with 600 infliximab infusions, we observed similar symptoms in two patients. Neither of them required hospitalisation.

Tuberculosis and other infections
The rate of infection is higher in infliximab-treated patients (36%) than in those receiving a placebo (26%). Frequently reported infections are upper respiratory infections and urinary tract infections. Serious infections that have been associated with infliximab include TBC, Pneumocystis carinii pneumonia, aspergillosis, histoplasmosis, listeriosis and cytomegalovirus. The incidence seems to be very low (about 1:2000), but infliximab should not be administered to patients with a known active infection. Active screening for the above-mentioned micro-organisms has not been advised with the exception of TBC, which will be discussed below.

Clinical relevance: TBC has been reported in 101 of approximately 175,000 infliximab-treated patients, of whom 21% suffering from Crohn’s disease (data on file, Centocor Inc, Malvern, USA). In virtually all patients, disease was a result of reactivation of latent tuberculosis. Importantly, in most patients the clinical presentation was aspecific and many patients had extrapulmonary disease. Hence, diagnosis of active tuberculosis in infliximab-treated patients requires a high degree of suspicion and extensive diagnostic workup. Most patients were on concomitant therapy (corticosteroids, azathioprine, methotrexate). A higher incidence was seen in patients with rheumatoid arthritis, in female patients and in older patients. In a large Dutch cohort of infliximab-treated CD patients, TBC reactivation was not observed. However, the current recommendations are to screen all patients before infliximab therapy using the purified protein derivative skin test (PPD). Who is at risk: An increased risk of TBC exists in infliximab-treated patients with latent TBC or with a high risk of tuberculous reactivation. This later is defined as: i) a history of TBC treated before 1970 or not treated for at least six
months including at least two months on the rifampicin-pyrizinamide combination; 2) in response to an intradermal tuberculin test done more than ten years after the last BCG vaccination, a wheal larger than 10 mm in diameter or a blister, with no history of active TBC or of TBC treatment; 3) residual tuberculous lesions larger than 1 cm² in size with no certainty that eradicative treatment was received. TBC screening recommendations: General consensus has been reached over the requirement of a thorough medical history and physical examination together with the PPD test prior to infliximab infusion. French recommendations also include a routine chest X-ray for all patients. Naturally, all PPD-positive patients, or patients who have received TBC immunisation (bacillus Calmette-Guérin), should undergo a chest X-ray. If the chest X-ray is negative in a patient with a positive PPD he or she should be treated for a latent TBC before infliximab can be administered. If the chest X-ray is positive in PPD-positive patients, then patients should be treated for active TBC before infliximab therapy. TBC treatment schedules of either latent or active TBC are beyond the scope of this review.

Miscellaneous

Infliximab-induced lupus: Formation of the autoantibodies, antinuclear antibodies (ANA) and anti-ds-DNA antibodies has been reported in 20 to 40% of infliximab-treated patients. Usually, titres are low and only few patients with autoantibodies develop clinical evidence of lupus. Thus, evidence of autoantibodies does not exclude patients from infliximab therapy; concomitant use of immunosuppressants will reduce the frequency of autoantibody development.

CONCLUSIONS AND RECOMMENDATIONS

In the near future, several biological therapies for inflammatory bowel disease will be added to the therapeutic armamentarium. Infliximab was first registered in the Netherlands, and extensive worldwide infliximab use in large groups of patients has enabled a detailed risk analysis. Most of the safety issues discussed here are relatively predictable and directly related to the mechanism of action of these drugs. Being a chimeric antibody, hypersensitivity reactions and formation of antibodies were likely to appear. Recently published data on frequency and management of adverse events are important for guiding physicians. However, in our opinion, it will also be crucial for all physicians treating these groups of patients to be aware of and understand the fundamental immunological mechanisms that are related with this kind of therapy. Not only will he or she be better prepared for the management of these events, but also the proper use of concomitant immunosuppressive therapy will be promoted. Finally, adequate patient instruction has been shown critical for drug compliance, and thorough knowledge of basic concepts of biological therapy will probably facilitate the doctor-patient information process.

REFERENCES


Assessment of disease activity in inflammatory bowel disease; relevance for clinical trials

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ABSTRACT

In patients with inflammatory bowel disease, assessment of disease activity is important in order to monitor and adjust therapy. In individual patients, disease evaluation is largely based on subjective symptoms. However, for disease evaluation in clinical trials, an objective and reproducible disease activity index is needed. At present, a number of activity indices are available for Crohn’s disease and for ulcerative colitis. These indices may be distinguished in more subjective clinical indices, more objective endoscopic and histological indices, and in indices with combinations of both subjective and objective parameters. In the design of a new clinical trial, an appropriate disease activity index should be selected which is based on the patient selection criteria and the aims of the study.

INTRODUCTION

In clinical practice, the subjective assessment of disease activity is relevant for the treatment of individual patients with inflammatory bowel disease (IBD). For disease evaluation in clinical trials, this subjective assessment of disease activity should be translated into an objective disease activity index. Preferably, the index should consist of as few variables as possible, which are easily obtainable in any hospital and applicable in a majority of the patients. Furthermore, the index should be validated, objective, reproducible and accepted by physicians and investigators. Such an instrument is necessary to define the characteristics of patients taking part in therapeutic trials, and to compare the results of treatment. It may be expressed in discrete classes or as a continuous numerical scale. Because of the differences in objectives of the studies and the variability in patient characteristics, no single disease activity scoring system can be applied for both ulcerative colitis and Crohn’s disease. In the 1950s and 1960s the first disease activity indices were introduced.1-3 For disease activity in ulcerative colitis, Truelove and Witts introduced a three-grade scale of absent, mild, moderate and severe disease (table 1). For Crohn’s disease, Best et al. developed the Crohn’s disease activity index (CDAI), which has been used on a large scale in clinical trials. After multivariable regression analysis eight out of 18 variables were included in the CDAI (table 2).4 The recent introduction of (potential) new drugs in the treatment of inflammatory bowel diseases has revived interest in the assessment of disease activity. In the design of a new clinical trial, it is essential to select the appropriate index to

<table>
<thead>
<tr>
<th>Severity categories of ulcerative colitis (Truelove-Witts)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>MILD</strong></td>
</tr>
<tr>
<td>Diarrhoea &lt;4 times/day, non-bloody</td>
</tr>
<tr>
<td>No fever</td>
</tr>
<tr>
<td>Pulse rate &lt;90 beats/min</td>
</tr>
<tr>
<td>ESR &lt;30 mm/h</td>
</tr>
<tr>
<td><strong>MODERATE</strong></td>
</tr>
<tr>
<td>Intermediate between mild and severe</td>
</tr>
<tr>
<td><strong>SEVERE</strong></td>
</tr>
<tr>
<td>Diarrhoea &gt;4 times/day, bloody</td>
</tr>
<tr>
<td>Haemoglobin 75% or below</td>
</tr>
<tr>
<td>Evening temperature &gt;37.5°C</td>
</tr>
<tr>
<td>Pulse rate &gt;90 beats/min</td>
</tr>
<tr>
<td>ESR &gt;30 mm/h</td>
</tr>
</tbody>
</table>

ESR = erythrocyte sedimentation rate.
answer the aims of the study. In table 3, the main factors influencing the choice for a disease index are summarised. Recently, an international group of experts has published a consensus on endpoints for clinical trials in Crohn’s disease.

\textbf{INDICES IN CROHN’S DISEASE}

The most frequently used index for the assessment of disease activity in Crohn’s disease is the CDAI (table 2). The outcome of the CDAI varies between 0 and 600 points. A score below 150 is associated with clinical remission, 150 to 219 with mildly active disease, 220 to 450 with moderately active disease and above 450 with very severe disease. This index is used in trials to select groups of patients based on the severity of the disease activity and to evaluate the effect of treatments. In most studies complete remission is defined by a value of less than 150 points, and clinical response is characterised by a decrease in CDAI of more than 70 to 100 points. The CDAI is also used to monitor patients for worsening of the disease during a trial. Relapse or reactivation of disease is defined as an increase in CDAI by more than 150 points or a score above 150 together with an increase by more than 50 to 100 points (not well defined). An increase in the index by more than 60 to 100 points or a disease activity of more than 200 at the end of the treatment period indicates treatment failure. Although it is broadly applied in clinical trials, substantial criticism has been raised. The inter-observer variability can be substantial because of differences in interpretation in variables that are part of the CDAI. This is due to the subjective variables that are included in the index. For instance, the patient scores important variables like ‘Abdominal pain’ and ‘General well-being’. Also the number of liquid or soft stools is an important variable, more or less subjective, and influenced by the presence or absence of the ileum. Overflow of bile acids in the colon due to

\begin{table}
\centering
\caption{Crohn’s disease activity index (CDAI)\textsuperscript{3,4}}
\begin{tabular}{lccc}
\hline
\textbf{SUM OF 7 DAYS} & \textbf{FACTOR} & \textbf{SUBTOTAL} \\
\hline
Number of liquid or soft stools & – – – & 2 & – – – \\
Abdominal pain\textsuperscript{1} & – – – & 5 & – – – \\
General well-being\textsuperscript{1} & – – – & 7 & – – – \\
Number of complications (presence or absence):\textsuperscript{2}
\begin{itemize}
\item Arthritis or arthralgia
\item Iritis or uveitis
\item Anal fissure, fistula or abscess
\item Erythema nodosum, pyoderma gangrenosum, aphthous stomatitis
\item Other fistula
\item Fever over 37.8°C
\end{itemize}
\begin{itemize}
\item Loperamide or diphenoxylate for diarrhoea (none = 0, yes = 1)
\item Abdominal mass (none = 0, questionable = 2, definite = 5)
\item Haematocrit (males 47, Ht (%), females 42, Ht (%))
\item Body weight: (\text{-} Body weight/standard weight) x 100 =
\item Pain score per day: 0 = none, 1 = mild, 2 = moderate, 3 = severe.
\item General well-being score per day: 0 = generally well, 1 = slightly under par, 2 = poor, 3 = very poor, 4 = terrible.
\item Total number of complications from the list that are present.
\end{itemize}
\hline
\end{tabular}
\end{table}

\textsuperscript{1} Pain score per day: 0 = none, 1 = mild, 2 = moderate, 3 = severe. \textsuperscript{2} General well-being score per day: 0 = generally well, 1 = slightly under par, 2 = poor, 3 = very poor, 4 = terrible. \textsuperscript{3} Total number of complications from the list that are present.
resection of the ileum and the subsequent interruption of the entero-hepatic cycle may be the cause of diarrhoea without any inflammatory activity. Although it is a relevant score for improvement or worsening of the patient’s symptoms, it is not representative for disease activity in the intestinal wall. The CDAI is a complex index and is difficult to calculate in daily clinical practice. A more suitable index has now been developed, which is known as the Harvey-Bradshaw index or Simple index. It includes five of the main variables of the CDAI and is easier to calculate, but has the same disadvantages. Other indices have also been developed such as the Therapeutic Goals Score, the Cape Town index and the index of the Organisation Mondiale de Gastro-Entérologie (OMGE) of which the Therapeutic Goals Score is not validated. These indices correlate quite well and are appropriate for assessment of improvement in the patient’s symptoms in clinical trials.

For assessment of the quality of life in IBD patients, the Inflammatory Bowel Disease Questionnaire (IBDQ) has been introduced. At present, this questionnaire is not frequently used in clinical trials.

Because of the disadvantages of scoring systems based mainly on the complaints of the patient, an index mainly based on objective variables has been developed. This Van Hees index or Dutch index is partly based on the patient’s symptoms but also on laboratory data and physical examination (table 4). The Van Hees index does not correlate well with the indices mentioned above; it is probably more appropriate for trials in which disease activity should be assessed and correlated with the inflammatory parameters such as cytokines.

In fistulising Crohn’s disease, the CDAI is not the most suitable index because the presence or absence of fistulae is only one of a number of listed complications in the index. In 1980, the first attempt was made for an index evaluating fistula closure, but this index was not validated. Irvine proposed the Perianal Disease Activity Index with five categories and five grades for each category. However, not all the fistulae are perianal and no consensus has been reached about the criteria on activity of the fistulae, the period of inactivation of fistulae or the percentage of the number of fistulae that should be inactive. Recently, a simple and easily reproducible fistula drainage assessment score was described by Present et al. (table 5). This index is promising but should be validated in additional prospective trials.

Besides these clinical indices, there are other methods that can be used for assessment of disease activity. Radiological methods such as x-rays of the small bowel and colon are widely used. More recently, also sonography, computer tomography and magnetic resonance imaging techniques have been applied. For external fistulae photography may be useful. Disease activity may also be established by the nuclear scintigraphy such as the 111In-labelled leucocyte scan. Furthermore, scoring systems have been developed for the endoscopic disease activity assessment. Most frequently applied and also well validated is the Crohn’s Disease Endoscopic Index of Severity (CDEIS). Rutgeerts et al. have described an endoscopic score for activity of Crohn’s disease after surgical resection of the ileum. Korelitz and Sommers and D’Haens et al.

### Table 4

<table>
<thead>
<tr>
<th>Variable</th>
<th>Score</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum albumin g/l</td>
<td>_ x -5.48 _ _ _</td>
</tr>
<tr>
<td>ESR mm/h</td>
<td>_ x 0.29 _ _ _</td>
</tr>
<tr>
<td>Quetelet</td>
<td>_ x -0.22 _ _ _</td>
</tr>
<tr>
<td>Weight</td>
<td>_ x 7.83 _ _ _</td>
</tr>
<tr>
<td>Abdominal mass</td>
<td>_ x 12.3 _ _ _</td>
</tr>
<tr>
<td>Gender</td>
<td>_ x -12.3 _ _ _</td>
</tr>
<tr>
<td>Temperature °C</td>
<td>_ x 16.4 _ _ _</td>
</tr>
<tr>
<td>Consistency of the stools</td>
<td>_ x 8.46 _ _ _</td>
</tr>
<tr>
<td>Resection</td>
<td>_ x -9.17 _ _ _</td>
</tr>
<tr>
<td>Extraintestinal manifestations</td>
<td>_ x 10.7 _ _ _</td>
</tr>
</tbody>
</table>

Subtract -209

VHI

ESR = erythrocyte sedimentation rate.

### Table 5

#### Fistula drainage assessment

**IMPROVEMENT**

Closure of individual fistulas defined as no fistula drainage despite gentle finger compression. Improvement defined as a decrease from baseline in the number of open draining fistulas of ≥50% on at least two consecutive visits (i.e. at least four weeks).

**REMISSION**

Closure of individual fistulas defined as no fistula drainage despite gentle finger compression. Remission defined as closure of all fistulas that were draining at baseline on at least two consecutive visits (i.e. at least four weeks).
have proposed histological scores to evaluate the effect of medical treatment. The clinical relevance of these indices is a matter of debate because clinical symptoms will not only be influenced by the focal signs of the severity of the inflammatory process but also by the location and extent. However, these indices may be relevant in studies aiming at the effect of interventions on the inflammatory process in the intestinal mucosa.

INDICES FOR ULCERATIVE COLITIS

In ulcerative colitis, the intestinal inflammation is confined to the colon mucosa. In active disease, this results in specific symptomatology with frequent diarrhoea and blood loss. The first index to quantify disease activity in IBD patients was based on these symptoms and developed for ulcerative colitis specifically.1,2 This Truelove-Witts score was mainly used to characterise a severe relapse (table 1) and was frequently applied in clinical trials. The disadvantage of the index is the difficulty in classifying a number of patients to the appropriate disease category and changes in disease activity over time are difficult to quantify. Other activity indices have been proposed by various authors.24-30 The index by Talsted and Gjone24 consists of 15 variables and is not applicable due to its complexity. Also the Powell-Tuck index,25 consisting of ten items, has a low feasibility. Endoscopic assessment of disease activity is important, because the complaints of a patients do not always represent the severity and extent of the disease. Severe proctitis of 10 cm, or less, may provoke more complaints than a moderate colitis over more than half the colon. Another advantage of endoscopy is the opportunity to obtain biopsies. Histology obtained in a remission period may demonstrate features of acute inflammation, which increases the risk on a relapse.31 Furthermore, dysplasia and malignant tumours may be established by endoscopy.32 Based on endoscopic findings, Dick et al. introduced a five-scale endoscopic index33 and Rachmilewitz developed an endoscopic index, scoring for granularity, vascular pattern, vulnerability and mucosal damage. The Rachmilewitz score is numerical and has been used in clinical trials.39 The Baron scale, distinguishing three grades of activity, is the most commonly used score to evaluate the degree of activity endoscopically.34 To combine the advantages of the clinical Truelove score1,2 and the endoscopic Baron scale,34 the Mayo score was developed.30 Currently, the Mayo score is favourite in clinical studies. In table 7 indices for the activity of ulcerative colitis are summarised, with the contributing variables.

CONCLUSIONS

For the evaluation of IBD in clinical trials, a variety of disease activity indices are available. The patient selection criteria and aims of the study are major determinants to select

Table 6

<table>
<thead>
<tr>
<th>Disease activity indices in Crohn’s disease</th>
<th>CDAI</th>
<th>HARVEY-BRADSHAW</th>
<th>VHI</th>
<th>OXFORD INDEX</th>
<th>CAPE TOWN INDEX</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abdominal pain</td>
<td>X</td>
<td>X</td>
<td></td>
<td></td>
<td>X</td>
</tr>
<tr>
<td>Bowel habits</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Perianal complications</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td>X</td>
</tr>
<tr>
<td>Other complications</td>
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<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Palpable mass</td>
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<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Body weight</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td>X</td>
</tr>
<tr>
<td>Haemoglobin level</td>
<td>X</td>
<td>X</td>
<td></td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Well-being</td>
<td>X</td>
<td>X</td>
<td></td>
<td></td>
<td>X</td>
</tr>
<tr>
<td>Anti-diarrhoeal drugs</td>
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<td>X</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Quetelet index</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Temperature</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td></td>
</tr>
<tr>
<td>Serum albumen</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>ESR</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sex</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Bowel resections</td>
<td>X</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tenderness</td>
<td></td>
<td>X</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fistula</td>
<td></td>
<td></td>
<td></td>
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</tr>
</tbody>
</table>

CDAI = Crohn’s disease activity index, VHI = Van Hees index, ESR = erythrocyte sedimentation rate.
the activity index of choice. In Crohn’s disease patients, the Van Hees index as an overall index seems suitable to evaluate inflammatory disease activity. For local mucosal monitoring, endoscopic and histological indices are preferable. For the quality of life evaluation, the IBDQ is preferred. To evaluate the clinical effect of treatment in a group of patients the CDAI is suitable. Furthermore, CDAI was most widely used in previous trials which enables comparisons with previous results. For evaluation of fistulising disease, specific indices are suggested. For ulcerative colitis, classification of the extent of the inflammation is essential. This can be performed by endoscopy or X-ray examination of the bowel. For the assessment of disease activity, clinical scoring lists are available. The activity index of choice contains endoscopic as well as clinical variables.

**REFERENCES**


**Table 7**

*Most frequently used indices in ulcerative colitis*

<table>
<thead>
<tr>
<th>INDEX</th>
<th>POWELL-TUCK</th>
<th>SUTHERLAND</th>
<th>RACHMILEWITZ</th>
<th>MAYO</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bowel frequency</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Blood in stools</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Well-being</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Abdominal pain</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Stool consistency</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Nausea</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Weight loss</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Extraintestinal signs</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Fever</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>ESR</td>
<td>X</td>
<td>X</td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td>Physician’s global assessment</td>
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<td>X</td>
</tr>
<tr>
<td>Sigmoidoscopy</td>
<td>X</td>
<td>X</td>
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</tbody>
</table>

ESR = erythrocyte sedimentation rate.


ABSTRACT

According to several reports, the risk of active tuberculosis in patients who are latently infected with Mycobacterium tuberculosis is increased after treatment with tumour necrosis factor α (TNF)-blocking agents. These drugs have demonstrated effectiveness and are increasingly being used for treatment of several inflammatory diseases, including rheumatoid arthritis and Crohn’s disease. Specialists prescribing TNF-blocking agents should be aware of the risk of tuberculosis and other infections, the unusual and severe clinical presentations and the available preventive measures. In this review, we will weigh currently available data on the risk of infection with intracellular pathogens and in particular tuberculosis in patients treated with TNF-blocking agents, discuss the role of TNF in the pathogenesis of tuberculosis and describe the risk profile of this complication. Awaiting further consensus protocols, a provisional flow chart is presented that is based on clinical parameters to provide a logical framework to reduce and minimise the risk of tuberculosis during TNF blockade.

INTRODUCTION

An impairment of the cellular immune system due to treatment with immunosuppressive drugs increases the risk of infections with intracellular pathogens, which were either already latently present within the host and reactivated or are newly acquired and cannot be controlled. That novel and highly effective immunosuppressive or immunomodulating drugs may bring along new patterns of reactivation of latent infections is illustrated by recent reports of an increased risk of reactivation tuberculosis (TB) during treatment with the novel tumour necrosis factor (TNF) α-blocking agents. An unusually high proportion of these patients presented with extrapulmonary or disseminated TB resulting in delayed diagnosis and treatment. During the past decades, the development of novel immunosuppressive drugs and regimens has led to a considerable improvement in the management of rheumatic diseases, vasculitis, malignancies and solid organ or haematopoietic cell transplantations. An arsenal of immunosuppressive drugs are currently available, including prednisone and related glucocorticosteroids, methotrexate, cyclosporine, azathioprine, and antimicrobial agents such as mycophenolate mofetil, sirolimus, tacrolimus and antithymocyte globulin. The mechanism of action of these drugs has recently been summarised, their main collective effect being either to decrease T-cell numbers or their function, or interfere with the production or effect of interleukin (IL)-2. Thus, these drugs cause unequivocal suppression of the cellular immune system. The occurrence of opportunistic infections with micro-organisms that do not normally cause illness in immunocompetent persons, such as Pneumocystis carinii, is a well-known phenomenon during use of the above-mentioned agents. Other pathogens that can remain latently present for prolonged periods and may reactivate under failing cellular immune defences include Mycobacterium tuberculosis, Strongyloides stercoralis and Histoplasma capsulatum.

In this review, we will weigh currently available data as to whether, and if so by how much, the risk of infection with intracellular pathogens and in particular TB is increased in patients treated with TNF-blocking agents. Furthermore we will discuss the role of TNF in the pathogenesis of TB and describe the risk profile of this
complication. Awaiting further consensus protocols, a provisional flow chart is presented that is based on clinical parameters to provide a logical framework to reduce and minimise the risk of TB during TNF blockade.

**TNF-BLOCKING AGENTS: INFliximab and Etanercept**

Two TNF-blocking agents have thus far been approved for clinical use and others are currently being evaluated. The precise time line of the history of TNF-blocking agents has recently been reviewed in detail. Infliximab (Remicade®, Centocor Inc) is a chimeric anti-TNF antibody, consisting of two murine antigen-binding fragments (Fab) with a high affinity for soluble as well as membrane-bound TNF, coupled to three-quarters of the human constant part of immunoglobulin G (Fc-IgG1). Etanercept (Enbrel®, Wyeth/Immunex) is a hybrid molecule composed of two TNF-receptor 2 molecules linked to human Fc-IgG in a similar way as in infliximab. Infliximab has been approved for the treatment of patients with severe rheumatoid arthritis (RA) not responding to optimal treatment with at least two disease-modifying antirheumatic drugs (DMARDs) including methotrexate, and patients with Crohn’s disease that is refractory to optimal standard immunosuppressive treatment. Etanercept is approved for the treatment of similar RA patients as well as patients with juvenile idiopathic arthritis (formally known as juvenile chronic arthritis) and psoriatic arthritis.

TNF is a non-specific effector molecule that is excreted predominantly by macrophages in response to various stimuli and that can exert immunostimulatory or immunosuppressive effects depending on the precise setting in which it is produced. TNF was found to be abundantly present in active lesions of RA and Crohn’s disease and thus was thought to play a central role in the pathogenesis of these disorders, through the induction of the production of many other cytokines such as IL-1. This hypothesis was affirmed by the impressive favourable effect of TNF blockade on the course of RA and Crohn’s disease in the majority of treated patients in clinical studies.

**TNF Blockade and Tuberculosis**

The first case of active TB during treatment with infliximab was observed during a phase III study, in which 340 patients were treated with infliximab in one of four different regimens. In 2001, based on postmarketing surveillance data, a report of 70 cases of TB among approximately 147,000 patients treated with TNF-blocking agents worldwide was published. In the ensuing correspondence to this publication, additional cases were described and another report described a patient with presumed atypical Crohn’s disease who was treated with TNF blockade, which was followed by rapidly progressive tuberculous enteritis. By the end of 2001, the authors mentioned that the number of reported cases had increased to 117. From the data in the study by Maini et al., the following absolute risk or incidence of TB (by TNF-blocking agent, indication, origin) can be deducted: 24.4/100,000 (infliximab, RA, USA); 203.8/100,000 (infliximab, all indications, non-USA); 8.8/100,000 (etanercept, all indications, all countries). As these risks were calculated from figures obtained by voluntary reporting, they probably underestimate the true risks. The authors calculated the relative risk in the ‘infliximab, RA, USA’ subgroup by comparing with the risk in the whole population of RA patients, which differed by a factor of four. It may be argued, however, that TNF blockade had at least until then been restricted to the most seriously ill patients who already had a prolonged history of use of corticosteroids, methotrexate or similar agents. Patients thus treated may therefore not be comparable with the general RA population with regard to their immune status, who on average will have received less immunosuppressive treatment. The precise incidence of TB in a comparable group of similarly immunosuppressed RA patients, but without TNF blockade, is not known. In a previous study of patients with systemic lupus erythematosus, polymyositis or similar non-RA inflammatory disorders in a high-endemic area, the cumulative corticosteroid dose, the mean daily dose and pulse therapy were found to be significantly associated with the occurrence of TB. This indicates that previous immunosuppression cannot be ignored in this regard. In the last-mentioned study, the risk of TB was about five times greater than that found in the general population. It is therefore striking that patients who went through a prolonged period of treatment with corticosteroids or other immunosuppressive drugs without the development of active TB, first developed TB after TNF blockade was given. This raises the question whether this episode of TB resulted from reactivation of latent TB infection, primary infection or exogenous reinfection. The relatively short interval between start of TNF blockade and TB in most patients, with a median of 12 weeks, in combination with low background incidence rates in the reporting countries suggests reactivation rather than de novo infection. But it can also be asked why if these persons already harboured latent TB infection, progression to active TB did not occur during previous immunosuppression. Reactivation of TB after TNF blockade but not during earlier immunosuppressive treatment would indicate either a much stronger or a qualitatively different effect of TNF blockade compared with the previously used drugs on the risk of reactivation TB. By comparison, corticosteroids have a widespread effect.
on the immune system by regulation of the expression of many genes resulting in, among other things, decreased production of a number of cytokines including TNF, decreased production of prostaglandins and induction of apoptosis of lymphocytes, with a net anti-inflammatory effect. Cyclosporin A and tacrolimus inhibit T-cell activation by interference with intracellular signal transduction, resulting in decreased IL-2 production and T-cell proliferation. Serious opportunistic infections can occur when using these agents but the effect may be less specific than that of TNF-blocking agents. Indeed, the occurrence of TB first after TNF blockade but not during a previous period of immunosuppression strongly suggests a specific effect of TNF blockade and a crucial role of TNF in the maintenance of latency.

It cannot be excluded that at least some of the TB cases resulted from de novo infection, because despite low population average TB rates, the risk of infection is not homogenous within a population but instead concentrated in specific settings and population subgroups, such as in large cities or among recent immigrants or the homeless. More clarity about reactivation versus de novo infection could be provided by genotyping the causative M. tuberculosis isolates in a setting where all isolates are typed, such as has been done in the Netherlands since 1993: a unique fingerprint indicates reactivation TB whereas clustering of identical isolates points towards recent transmission. As long as such data are lacking, both reactivation TB and de novo infection must be considered as possible ways leading to active TB after TNF blockade. The considerably lower risk of TB after TNF blockade in the USA compared with other countries was not explained. This could result from differences in reporting or in background rates of latent TB infection among patients who are eligible for treatment with TNF blockade, or more effective screening procedures before starting immunosuppression. Thus far, TNF-blocking agents are expensive (more than € 2000 per dose) and have not yet been widely used in poorer areas where the rates of latent TB infection are much higher than in the USA and Western Europe. In high-incidence areas the risk of primary infection or reinfection with M. tuberculosis contributes significantly to the overall risk of TB.\(^5\)\(^6\)\(^7\) Except for animal studies, there are no data on the course of primary infection or exogenous reinfection with M. tuberculosis during TNF blockade, but an increased susceptibility is likely, given the key role of TNF in the innate immune response to M. tuberculosis.

It might be questioned whether TNF blockade alone, without previous immunosuppressive treatment, is actually sufficient to cause reactivation TB. The answer may be found in the results of a study of patients with ankylosing spondylitis, a disease for which standard immunosuppressive therapy is ineffective, who were treated with infliximab.\(^8\) One out of 34 patients treated with infliximab developed TB, suggesting that TNF blockade can in itself cause reactivation TB. Also, at least one case of TB was observed in several relatively small studies aimed at the evaluation of TNF blockade for various indications.\(^5\)\(^9\)\(^10\) This suggests an extraordinarily large increase in risk of reactivation TB during TNF blockade and this would point to an essential role for TNF in the maintenance of latency.

**Differences Between Infliximab and Etanercept**

From the available data, it appears that the risk of TB after etanercept is lower than after infliximab, and in fact not above the population background rate,\(^5\)\(^9\)-\(^7\) which could be related to differences in the mechanism of action of these drugs. Both agents probably work by binding and inactivating TNF at the site of inflammation. Infliximab not only neutralises soluble TNF, but also binds to membrane-bound and receptor-bound TNF,\(^8\)\(^9\)\(^10\) while etanercept has no such additional effects. Infliximab has been shown to induce apoptosis and cell-associated TNF infliximab complexes could initiate antibody-dependent cellular cytotoxicity resulting in cell lysis of the cells that contribute to the defence against mycobacteria.\(^8\)\(^9\)\(^10\)-\(^12\) However, it is unclear whether the proapoptotic effect of infliximab has clinical relevance for the risk of TB after TNF antibodies. An increased number of apoptotic cells were found in the lungs of mice who developed TB after treatment with TNF antibodies,\(^8\) which is discordant with the minimal apoptosis found in the lungs of similarly treated humans with TB.\(^5\) Downregulation of IFN-γ production of T cells by infliximab could add to the loss of resistance against infection with M. tuberculosis.\(^8\) Different kinetics, dosages and intervals could affect the level and continuity of TNF blockade. Etanercept binds lymphotoxin (TNF-β) in addition to TNF-α. Yet another factor that could contribute to the different effects could be that both infliximab and etanercept have a high affinity for TNF but only the binding of monoclonal TNF antibodies is irreversible. The naturally occurring soluble TNF receptor 2, which constitutes the TNF binding part of etanercept, is thought to be a ligand-passing receptor. TNF bound to this receptor may thus form a TNF reservoir from which TNF can be released in the presence of a low concentration of soluble TNF. Together, these differences between TNF antibodies and the soluble TNF receptor-based hybrid may translate into differences in efficacy and toxicity. In accordance with the above, an increased risk of TB was also observed during the clinical development of adalimumab (D2E7, Abbott Laboratories), a ‘human’ monoclonal antibody directed...
against TNF made by phage display from human components. A safety update on TNF antagonists by the Arthritis Drugs Advisory Committee of the USA Food and Drug Administration can be found at: http://www.fda.gov/ohrms/dockets/ac/01/briefing/3779b2.htm.

ROLE OF TNF DURING INFECTION WITH M. TUBERCULOSIS

During active TB disease, TNF appears to be involved with tissue necrosis and systemic symptoms but the study of its role during latent infection has been hampered because the immune responses responsible for maintaining the latent state of TB in humans are poorly understood. This is related to the fact that latent TB does not occur after infection with M. tuberculosis in animals and thus far no experimental model of latency is available that is truly comparable with latent TB infection in humans. Notwithstanding the limitations of animal models, both in TNF and TNF receptor knock-out mice and in mice treated with TNF antibodies, the course of TB was rapidly progressive. In an artificial murine latency model, using low-dose intravenous infection, TNF antibodies led to rapidly fatal ‘reactivation’ TB. In these mice, the infection was spread throughout the body, which reminds one of the frequent presentation with extrapulmonary or disseminated TB in patients after TNF blockade. These findings are in accordance with an important role of TNF in granuloma formation and local containment of infection. The factors that underlie reactivation TB in persons without recognised immune disorders are not known, although previous studies suggest that the risk is influenced by general health and deficiencies in specific nutrients such as vitamin D. It is generally thought that Th1 responses, especially IFN-γ production, confer protection to M. tuberculosis, but precise cell populations, the characteristics and kinetics of a protective immune response have not yet been defined, although they are of great interest in the light of the development of an improved vaccine against TB. If genetic determinants of the risk of active TB exist, no convincing factors have thus far been identified.

Other cytokines besides TNF play a role in the pathogenesis of inflammatory disorders and it is to be expected that novel drugs targeting different cytokines will become available. The question is then whether such new drugs will have a similar effect on the risk of TB. While TNF is central to the triggering of inflammation in RA, interleukin 1 appears to be particularly involved in tissue destruction. Treatment with recombinant IL-1 receptor antagonist (anakinra) was effective in a proportion of RA patients and appeared to increase the risk of serious infections, although so far it has not been associated with an increased risk of TB. Nevertheless, IL-1β and natural IL-1 receptor antagonists probably play a role in the defence against TB, and polymorphisms in the genes coding for these proteins were associated with clinical manifestations and the occurrence of false-negative tuberculin skin tests (TSTs) in TB patients. This both justifies an increased awareness of the possibility of reactivation TB during use of novel anticytokine drugs in general and suggests that interference with cytokines might decrease the reliability of the TST.

PREVENTION OF TB DURING TNF BLOCKADE

Identification of patients at risk
How can we identify patients eligible for TNF blockade who are at an increased risk of reactivation TB? Some of the patients who had developed active TB during TNF blockade were positive to the tuberculin skin test (TST) before the start of treatment or had a history of insufficiently treated TB. This highlights the importance of not missing the opportunity for prevention when it is still possible, i.e. before starting treatment with TNF blockade, or even better, before any immunosuppression is given. For an individual patient, the risk of TB during and after TNF blockade is the resultant of host factors, those determining the risk of latent TB infection and the status of the cellular immune system, and environmental factors determining the risk of de novo infection. The a priori risk of latent TB infection depends on factors such as age, country of origin, age at migration or immigration, travel and occupational history, recognised exposure to a patient with pulmonary TB or regular exposure to persons belonging to a risk group for TB (prison inmates, inhabitants of mental institutions, immigrants and asylum seekers from high-endemic countries, the homeless, drug abusers). The underlying disease and the type and intensity of the cellular immune defect induced by previous treatment can contribute to the risk of TB, although this effect may be overshadowed by the effect of TNF blockade, as was discussed above. The first and most important part of the evaluation should consist of an in-depth interview to establish the risk of prior exposure to M. tuberculosis. Such an interview could include questions regarding the place of birth of the patient, the present residence, a travel history including means of transport and the housing, whether TB has ever been diagnosed and how this was treated, if there has been any contact with a patient diagnosed with pulmonary TB or with persons belonging to a risk group for TB, and whether a TST has ever been performed at school, during military service or in a contact investigation. If a positive TST is reported, it is essential to find out if this was followed by drug treatment, which drug was prescribed, for how long and whether the patient adhered to the treatment.
Arend, et al. TNF-α blockade and tuberculosis: better look before you leap.

TNF blockade can be given but increased awareness of:
- Nonspecific presentations of TB
- Extrapulmonary TB
- Disseminated TB

Strongly consider treatment of latent TB infection
Delay TNF blockade

Currently active TB?
- Full diagnostic workup
- Withhold TNF blockade

Interpret according to:
- A priori risk of latent TB infection
- BCG vaccination state (false-positive?)
- Immunosuppressive drugs (false-negative?)

Full course multidrug treatment of TB
Postpone TNF blockade until completed

Figure 1
Provisional flow chart for the evaluation of tuberculosis infection in patients eligible for treatment with TNF-blocking agents

A Adequate treatment of active TB is defined as ≥6 months treatment, including ≥2 months rifampin combined with pyrazinamide. Adequate treatment of latent TB infection consists of either ≥6 months of isoniazid, ≥2 months rifampin plus pyrazinamide or ≥4 months rifampin.

B The risk of latent TB infection, as deducted from the presence of risk factors for prior exposure, depends on age, country of origin, travel and occupational history, recognised exposure to a patient with pulmonary TB or regular exposure to persons belonging to a risk group for TB (prison inmates, inhabitants of mental institutions, immigrants and asylum seekers from high-endemic countries, homeless persons, drug abusers).

C In persons who may have been infected long ago, the response to a first TST may be negative, but positive after a second test as a result of a booster phenomenon. Two-step testing could be useful in the cohort in which the prevalence of TB infection exceeds 5%, i.e. in persons born before 1945. Two-step testing may also be valuable in immunosuppressed persons, irrespective of age.

D There are no evidence-based data to determine a safe interval between the start of treatment of latent TB infection and TNF blockade (also, see text).
The history of the introduction of the anti-TB drugs, streptomycin in the mid-1940s, isoniazide in 1952 and rifampin in 1964, may act as a guideline to assess whether treatment has been effective according to current standards. A physical examination and chest radiograph are part of the workup and additional diagnostic tests should be performed if so indicated by the history or clinical findings.

Detection of latent TB infection: tuberculin skin testing and its limitations

Prevention of TB during TNF blockade requires the detection of latent TB infection, but this is problematic as the definition requires that *M. tuberculosis* can not be identified and that there are no signs or symptoms of active infection. A positive response to tuberculin (purified protein derivative, PPD) is currently the only and by its nature indirect method to detect latent infection with *M. tuberculosis*. In a statement by the American Thoracic Society and the Centres for Disease Control and Prevention (ATS/CDC), guidelines for targeted testing and treatment of latent TB infection are provided, including criteria to define a positive TST response. According to the ATS/CDC guidelines, the criterion to define a positive TST response depends on an individual’s clinical and epidemiological characteristics, resulting in three different cut-off levels.

In patients with a HIV infection and in those immunosuppressed by treatment or exposed to a patient with contagious pulmonary TB, an induration of ≥5 mm is defined to indicate a positive response; ≥10 mm is advocated in the presence of less severe risk factors and ≥15 mm in the absence of specific risk factors. In the Netherlands, a single cut-off value of ≥10 mm has thus far been applied to indicate a positive TST response, although the criterion of ≥5 mm has been adopted for HIV-infected persons. Along the same line, it may be argued that a cut-off level of ≥5 mm would be more appropriate for patients on immunosuppressive drugs or those with serious illness. However, following Dick Menzies’ dictum ‘once positive, no longer useful’, a TST should only be performed in the absence of a history of a positive TST or previously diagnosed TB.

The technical or biological reasons for false-positive and false-negative TST results have been reviewed. In the Netherlands, vaccination with *Mycobacterium bovis* Bacille Calmette-Guérin (BCG) has never been part of the routine vaccination policy but has been restricted to persons working in high-risk professions and long-term travellers to high-endemic countries. In contrast to the native Dutch population, most immigrants have been vaccinated with BCG and false-positive TST results due to cross-reactive immune responses can be expected in this population. Recent studies have shown that false-positive responses are avoided through the use of *M. tuberculosis*-specific antigens (named ESAT-6 and CFP-10) in an *in vitro* T-cell assay. Results from an enzyme-linked immunospot (ELISPOT) assay based on ESAT-6 in immunocompetent persons gave a stronger positive relation with exposure to *M. tuberculosis* than the TST. In a study of HIV-infected persons, an ESAT-6/CFP-10-based ELISPOT assay was found to be highly sensitive for detection of active TB as well as more specific and possibly more sensitive than the TST for detection of latent TB infection. Thus, such alternative diagnostic tests may provide important information in patients eligible for TNF-blocking agents and who had previously received immunosuppressive treatment. However, an assay based on such specific antigens is not yet available for daily practice.

In the context of TNF blockade, TST results can be false-negative in persons using immunosuppressive agents such as corticosteroids or methotrexate. The demonstration of skin test anergy could help to earmark a negative TST result as unreliable, but unfortunately there are no defined control antigens and results do not help to predict the risk of TB in either HIV-negative or HIV-positive patients. Anergy skin testing is therefore not advocated. It is highly preferable that a TST is performed before any immunosuppressive drug is given and not just before the first dose of a TNF-blocking agent, but a TST is nevertheless indicated in a person who is already being treated with immunosuppressive drugs, because a positive result still provides relevant information although a negative result does not rule out latent TB infection. As a result of waning immune responses, a first TST may be negative or false-negative in older persons who were infected with *M. tuberculosis* in the remote past, while a repeat TST may be positive due to the boosting effect of the first test.

This method of two-step testing could be advocated in all persons above a certain age, e.g., those born before 1945, the cohort in which the prevalence of TB infection exceeds 5%. Two-step testing may also be valuable in immunosuppressed persons, although this has not been studied.

Who to treat and when can TNF blockade be given safely?

In persons with a (history of) a positive TST response, strong epidemiological evidence for an increased risk of latent infection with *M. tuberculosis*, such as a family member being diagnosed with pulmonary TB in the past, or in case of untreated or insufficiently treated TB in the past it is mandatory to exclude active TB before a TNF-blocking agent is given. This implies that TNF blockade must be withheld until all results are available, including the results of cultures which may imply a period of six to eight weeks. When active TB is diagnosed or strongly suspected, a full course of antituberculosis treatment should be prescribed according to current guidelines and TNF-blocking agents withheld, preferably until treatment of TB is completed. When latent TB infection is recorded or strongly suspected and active TB is ruled out, the decision to
start early or pre-emptive treatment should always be made on an individual basis, weighing such factors as the risk of side effects, which depends on age, alcohol consumption, pre-existing liver function disturbances and co-medication.\[^{45}\] In general, the benefits of screening and preventive therapy were found to outweigh the risks for all risk groups, including immunocompromised persons.\[^{46}\] There are no data on which to base a safe interval between the start of treatment of latent TB infection and TNF blockade. In France, the ‘Groupe Tuberculose et infliximab’ and the French agency for healthcare product safety (AFSSAPS) have provisionally advocated an interval of two months.\[^{47}\]

### Recommendations during TNF blockade

In general, patients treated with TNF-blocking agents should be advised to avoid contact with persons known to have or who are at increased risk of pulmonary TB, including the risk of travel, and to seek medical advice immediately if symptoms or signs compatible with TB, such as weight loss, fever, sweats or persistent cough, occur during or after treatment. A high index of suspicion of TB is justified in all patients with unexplained symptoms, and those who have been or are being treated with a TNF-blocking agent. However, miliary TB can be notoriously difficult to diagnose despite a thorough diagnostic workup,\[^{48}\]\[^{49}\] arguing for empiric treatment of TB if the suspicion is high and no alternative diagnosis has been made.\[^{50}\] Apart from the risk of TB, the occurrence of other opportunistic infections such as *Pneumocystis carinii* pneumonia, histoplasmosis and listeriosis has been reported during TNF blockade and the clinician should keep an open mind regarding the differential diagnosis.

### COnclusion

TNF-blocking agents are not a panacea for all diseases in which TNF is thought to play a role, as they were found to be ineffective and possibly even deleterious in multiple sclerosis and congestive heart failure. However, the number of disorders for which TNF-blocking agents appear to be effective is rapidly increasing, starting with RA and Crohn’s disease, followed by ankylosing spondylitis, Still’s disease, psoriatic arthritis, progressive systemic sclerosis, Wegener’s granulomatosis, chronic uveitis and TRAPS (TNF receptor-associated periodic syndrome) and new indications might follow.\[^{45}\]\[^{51}\]\[^{52}\] Until the time of this writing, the TNF-blocking agents are only fully reimbursed according to the reimbursement system for medications in the Netherlands for use in RA, Crohn’s disease, juvenile idiopathic arthritis and psoriatic arthritis. If they prove to be highly effective for less severe cases of RA as well and if health insurance companies start reimbursing the costs the use of these drugs could increase considerably. It is therefore mandatory that protocols aimed at optimal prevention of reactivation TB and early detection of active TB are rapidly developed and evaluated prospectively.

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Diagnosing chronic fatigue syndrome: comparison of a protocol and computerised questionnaires

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** J.W.M. van der Meer was not involved in the handling and review of this paper.

ABSTRACT

Background: In the context of outpatient care and within the framework of scientific research, guidelines and measuring instruments have been developed to help improve CFS diagnostics. The purpose of this study was to measure the agreement between the evaluations of chronically fatigued patients by physicians using a CFS protocol and by researchers using computerised questionnaires.

Methods: The sample consisted of 516 patients referred to an internal medicine outpatient clinic with complaints of chronic fatigue. Retrospectively the medical records and the computerised questionnaires were checked separately and compared to see whether the criteria for diagnosis of CFS had been met. In addition, the reasons for not diagnosing CFS were evaluated.

Results: Agreement between the physicians’ and the researchers’ evaluations was 84%. Disagreement mostly concerned severity of fatigue and functional impairment, or premorbid exclusion criteria. A physical cause for the chronic fatigue was only found in 3% of the cases.

Conclusions: For physicians, questionnaire assessment may be complementary to the CFS protocol in optimising the process of diagnosing CFS.
METHODS

Patients and procedure
The sample consisted of all patients referred to the general internal medicine outpatient clinic of the University Medical Centre St Radboud in Nijmegen with complaints of chronic fatigue between October 1996 and January 1998. These patients were screened according to the CFS protocol. The protocol included an extensive anamnesis administered by the attending physician, followed by a medical examination and a restricted number of laboratory tests. This consultation lasted approximately one hour. Subsequently, after a trained nurse had instructed the patient on how to operate the computer, the patient was requested to fill in the questionnaires on a computer in a separate consulting room, which took about 30 minutes. The nurse remained available throughout the procedure for any questions. After four weeks the patient was called in for a second consultation during which the physician explained the findings of the clinical examination. The diagnosis of CFS was based solely on the physician’s judgement of these clinical findings. The outcome of the questionnaire assessment was used later to select CFS patients eligible for a randomised controlled trial and was not taken into account in the clinical judgement.

CFS protocol
To streamline and facilitate CFS diagnostics in patients referred with complaints of chronic fatigue, a CFS protocol for outpatients was developed containing guidelines for both the anamnesis and physical examinations as well as supplementary diagnostics.

CFS is defined as a self-reported fatigue that has lasted more than six months, is irrespective of physical exertion, leads to severe functional impairment, and where there is no medical explanation for the symptoms. For a diagnosis of CFS to be made, the physician needs to answer the following questions: Can a somatic explanation for the symptoms be excluded? Is this a case of severe fatigue associated with serious limitations in the patient’s professional, social and/or personal functioning? Have the symptoms and impairments been present for at least six months? Do any of the exclusion criteria as formulated by the CDC concerning depression, psychosis, eating disorders or alcohol abuse apply?

Anamnesis
The first step in the symptom-specific anamnesis is to try and gain insight into the patient’s expectations and objectives with respect to this consultation and this doctor. Frequently, CFS patients attribute their symptoms to a variety of factors, which cause them to have high expectations for the diagnostics. Also, there may be a hidden agenda involving insurance issues and invalidity benefit claims. It is essential to identify these issues and expectations at an early stage to make communication more transparent and to prevent both sides from digging in. When it has been established what the patient may or may not expect from his or her visit to the clinic, the severity and extent of the functional impairment is investigated. A suitable technique is to have the patient describe what a normal, average day looks like, for instance the day before.

Important details that should be discussed are: At what time does the patient get up? Does he or she take a shower, have breakfast, get dressed? Also issues such as who does the shopping, or the cooking, whether he/she goes to work, plays sports, etc. should be addressed. It is recommended to literally go through the patient’s day, hour by hour. Subsequently, the duration and the course of the symptoms are discussed. It is essential to try and establish whether the patient has been fatigued his/her entire life or whether the onset of the symptoms can be more or less clearly defined. This is assessed both from the physician’s and the patient’s perspective. Next, any concomitant complaints are investigated, which are often abundant. It is important to determine whether fatigue is indeed the principal complaint. In principle, the interview continues with a full internal anamnesis, use of medication (including alternative medication) and stimulants, and the patient’s case history, specifically with respect to psychiatric symptoms and eating disorders. Finally, any previous diagnoses and treatment(s) are discussed.

Physical examination
The patient is given a full physical examination during which specific attention is paid to the detection of so-called stigmata indicating possible endocrine causes for the fatigue symptoms, such as orthostatic hypotension, pigmentation, pattern of body hair, etc.

Supplementary diagnostics
Laboratory tests are restricted to erythrocyte sedimentation rate (ESR), haematological parameters, minerals, liver and renal functions, protein spectrum, thyroid stimulating hormone (TSH), ferritin and creatine phosphokinase (CPK). In rare cases this range of tests may be extended on the basis of the findings of the anamnesis and/or physical examination.

Computerised questionnaires
A total of four questionnaires were administered to verify whether patients fulfilled the international criteria for CFS as used in scientific research. The fundamental criterion, i.e. exclusion of physical causes, could not be included in this part of the study since a medical practitioner can only evaluate this aspect. The remaining criteria were all assessed by means of the various questionnaires, which
were administered on a personal computer. Patients completed the following five questionnaires: 1) a general questionnaire on the patient’s personal data, and the nature, duration and onset of the complaints, 2) the validated fatigue inventory checklist individual strength (CIS), 3) a functional impairment questionnaire consisting of eight subscales of the sickness impact profile (SIP-8): sleep/rest, housekeeping, mobility, social interaction, walking, alertness/intellectual functioning, work, recreational and leisure activities, 4) a questionnaire scoring premorbid functioning and finally 5) a questionnaire assessing additional CFS-related physical complaints. For the diagnosis of CFS as commonly applied in research, the following criteria needed to be met:
- Fatigue is the principal complaint
- The fatigue symptoms have been present for at least six months, excluding lifelong incidence
- A score of 35 or higher on the CIS subscale fatigue severity
- A score of 800 or higher on the eight subscales of the SIP
- Absence of premorbid eating disorders, alcohol-related problems in the two years prior to the assessment, premorbid depressive disorders or psychotic episodes.

The concomitant physical complaints were not included in the diagnosis since it has already been established that these are not contributing factors.

Analysis
A researcher from the department of general internal medicine (H. Koning) retrospectively evaluated the medical files of all the patients examined in the above-mentioned period. A researcher from the department of medical psychology (J.B. Prins) evaluated the computerised questionnaire data. Both evaluations were aimed at establishing whether a diagnosis of CFS had been made. In the absence of a CFS diagnosis, the rationale behind the judgement was determined. Next, the two datasets were linked and compared to determine statistically the agreement with respect to the CFS diagnosis for each patient. Concordance between the physician’s diagnosis and the researcher’s evaluation of the computerised questionnaires was evaluated by Cohen’s kappa, which is a measure of concordance between two dichotomous variables corrected for chance. A value of Cohen’s kappa of .40 or lower is considered moderate, between .40 and .70 satisfactory, and above .70 good.

RESULTS
Patient characteristics
In the period investigated, 567 patients were referred to our outpatient clinic because of complaints of chronic fatigue. Fifty patients were not included in the study. Their symptoms could be explained on the basis of existing data and a consultation was not expected to reveal any additional information. Of the remaining 517 patients, 212 were referred by their GPs, 46 by a medical specialist and 259 patients had contacted the clinic of their own accord. Nearly 75% of the patients attended the outpatient clinic in the expectation that they would be diagnosed with CFS, 16% mentioned participating in scientific research as their main reason for requesting the consultation and 10% reported both these motives. In one patient a full assessment proved to be impossible. Thus, the data of 516 patients could be analysed and compared. Of the patients included in the analyses 78% were female, 22% male, and their mean age was 36 years and 9 months (range 14-69 years).

CFS protocol
Figure 1 shows the results of the physical assessment of all 516 patients. Based on the protocol, the clinicians diagnosed 409 patients (79%) as suffering from CFS. In the remaining 107 patients CFS was not diagnosed on various grounds. In half of these patients (n=54) the fatigue-related symptoms or functional impairment were not judged sufficiently severe to justify a diagnosis of CFS. In 40 patients comorbidity, possibly explaining the fatigue, was diagnosed. The comorbidity included somatic illnesses (n=17), psychosocial problems (n=9), alcohol-related problems or eating disorders (n=4), and other principal complaints (n=10). Thirteen patients met the exclusion criteria for CFS relating to the premorbid condition, viz. eating disorders, depression or lifelong fatigue.

Computerised questionnaires
The results of the questionnaire-based assessment of all 516 patients referred are also listed in Figure 1. According to the outcome of the questionnaires, 369 patients (71%) met the CFS criteria investigated. The reasons why the remaining 147 patients were not diagnosed as suffering from CFS included insufficient scores on the CIS and/or SIP-8 (n=59), fatigue proved not to be the principal complaint (n=29) and the presence of premorbid eating disorders or alcohol-related problems, depression, psychoses or lifelong fatigue (n=59).

Comparison of the two assessments
Table 1 indicates that in 84% of the cases there was agreement between the clinicians’ assessments and the researchers’ evaluations of the questionnaires as regards the presence or absence of a CFS diagnosis. The degree of agreement was analysed using Cohen’s kappa and was .58 (SE .04), a correspondence that is common in scientific research in a clinical setting and is generally regarded as satisfactory.
Of all 516 patients examined, 21 (4%) were diagnosed as suffering from CFS on the basis of the computerised questionnaires whereas the internist excluded CFS. In these 21 patients, a different diagnosis was made in six of them: either somatic (n=3) or psychiatric (n=3). In the remaining 15 patients the physician found insufficient complaints and/or impairments for a diagnosis.

In 61 (12%) of the patients the inclusion criteria for CFS were not met according to the questionnaire-based assessment, whereas the specialist did diagnose CFS. The scores on the CIS or SIP were found to be too low in 40% of the patients concerned, while the physician judged the complaints and impairments as sufficiently severe. In the computer assessment 29% of the patients had not indicated fatigue as their main complaint and 31% had reported premorbid eating disorders or alcohol-related problems, depression or lifelong fatigue, aspects that had not come to light during the physician’s consultation.

**DISCUSSION**

It goes without saying that the diagnosis of CFS can and should never be solely based on an assessment using computerised questionnaires. First and foremost, any physical cause for the symptoms should be excluded, a criterion that always requires the judgement of a physician. In this study a physical cause for the fatigue symptoms could only be found in a few cases. Apparently, prior to their referral, the majority of patients had been screened in such a way that further diagnostics did not yield any additional information. We concluded that referral of CFS patients to our internal medicine outpatient clinic seldom
Table 2

Shortened fatigue questionnaire (SFQ) medical psychology. University Medical Centre St Radboud, Nijmegen, the Netherlands

Name: ............................................................................................................................... 
Gender: male/female ..........................................................................................
Date of birth: .................................................................................................................. 
Today’s date: .................................................................................................................

On this page you will find four statements indicating how you have been feeling during the past two weeks.
You can answer each question by placing a cross in one of the seven boxes. The position of the marking indicates to what extent you feel the statement applies to you.
For example: if you think the statement is completely true, you should place a cross in the left box, like this:

Yes, that is true

| 7 | 6 | 5 | 4 | 3 | 2 | 1 |

No, that is not true

If you think the answer is not ‘yes, that is true’ but also not ‘no, that is not true’, you should mark the box that best corresponds with your feeling, for example like this:

Yes, that is true

| 1 | 2 | 3 | 4 | 5 | 6 | 7 |

No, that is not true

Please answer all the statements and place only one cross for each statement.

1. I feel tired
   Yes, that is true

2. I tire very quickly
   Yes, that is true

3. I feel fit
   Yes, that is true

4. Physically I feel exhausted
   Yes, that is true

Score form SFQ
Chief complaint: ...................................................................................................... 
Date of origin: (month) (year)
Diagnosis: .............................................................................................................

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<tr>
<th>GROUPS</th>
<th>AVERAGE</th>
<th>&lt; LOW</th>
<th>AVERAGE</th>
<th>&gt; AVERAGE</th>
<th>&gt;&gt; HIGH</th>
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<td></td>
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<td>Healthy adults</td>
<td>37</td>
<td>4</td>
<td>4</td>
<td>5-8</td>
<td>9-14</td>
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<tr>
<td>Students, normal circumstances</td>
<td>22</td>
<td>4</td>
<td>5-7</td>
<td>8-14</td>
<td>15-21</td>
</tr>
<tr>
<td>Students, demanding circumstances</td>
<td>21</td>
<td>≤5</td>
<td>6-9</td>
<td>10-17</td>
<td>18-23</td>
</tr>
<tr>
<td>Servicemen at rest (normal)</td>
<td>21</td>
<td>4</td>
<td>5-6</td>
<td>7-14</td>
<td>15-22</td>
</tr>
<tr>
<td>Servicemen in field exercise</td>
<td>21</td>
<td>≤5</td>
<td>6-11</td>
<td>12-18</td>
<td>19-24</td>
</tr>
<tr>
<td>PATIENT GROUPS</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cancer</td>
<td>61</td>
<td>4</td>
<td>5-12</td>
<td>13-21</td>
<td>22-27</td>
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<tr>
<td>Functional bowel disease</td>
<td>41</td>
<td>≤6</td>
<td>7-12</td>
<td>13-21</td>
<td>22-27</td>
</tr>
<tr>
<td>Multiple sclerosis</td>
<td>36</td>
<td>≤12</td>
<td>13-19</td>
<td>20-26</td>
<td>27</td>
</tr>
<tr>
<td>Chronic fatigue syndrome</td>
<td>38</td>
<td>≤22</td>
<td>23-25</td>
<td>26-27</td>
<td>28</td>
</tr>
</tbody>
</table>

1. I feel tired
   Yes, that is true

2. I tire very quickly
   Yes, that is true

3. I feel fit
   Yes, that is true

4. Physically I feel exhausted
   Yes, that is true

Total score SFQ:
lead to new medical insights. Therefore, referrals could be limited to those patients for whom the expertise of a specialist is required to exclude any physical causes, for instance in cases of suspected adverse effects of medication, slightly deviating laboratory test results or somatic comorbidity. According to a recent unpublished survey among general practitioners, currently 78% of fatigued patients are still being referred to a medical specialist. It is our view that administration of the presented protocol for chronic fatigue complaints by GPs would not only lead to substantial reductions in public spending, but would also prevent undue expectations in patients about new or additional medical diagnoses.

When retrospectively comparing the diagnoses based on the CFS protocol with the diagnoses on the basis of the computerised questionnaires, agreement between both assessments was found in the majority of the cases. In 16% of the cases the clinicians’ and the researchers’ conclusions were contradictory. In quite a few instances, there was ambiguity about the severity of the fatigue and functional impairment. When a physician is having doubts about symptom severity, questionnaire assessment might be considered. Supplementary to the protocol, the shortened version of the fatigue questionnaire\textsuperscript{13,14} could be administered to assess fatigue severity or the physical functioning subscale of the SF-36 questionnaire (MOS-Short Form-36)\textsuperscript{15-17} to measure functional impairment (tables 2 and 3).

Physicians using the CFS protocol more often diagnosed CFS than researchers evaluating the computerised questionnaires (79 and 71% respectively). Premorbid exclusion criteria for the diagnosis CFS, such as alcohol dependency, eating disorders or depressive disorders, were found more often in the computerised questionnaires than in the physician’s consultation. Obviously, it is difficult to establish the patient’s case history or current situation with respect to psychological problems or psychiatric disorders. Questionnaire assessment might lead to additional information.

At our outpatient clinic, after consulting the internist patients with chronic fatigue routinely fill in computerised questionnaires to establish whether they meet the operational criteria for CFS. The physician is able to consider the questionnaire data concerning fatigue severity, functional impairment and actual and premorbid functioning before the second consultation. In this way the questionnaire assessment is complementary to the CFS protocol and the process of diagnosing CFS is optimised.

**REFERENCES**


**Table 3**

*SF 36 questionnaire, subscale physical functioning*\textsuperscript{15}

The following items are about activities you might do during a typical day. Does your health now limit you in these activities? If so, how much?

<table>
<thead>
<tr>
<th>YES, LIMITED A LOT</th>
<th>YES, LIMITED A LITTLE</th>
<th>NO, NOT LIMITED AT ALL</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vigorous activities, such as running, lifting heavy objects, participating in strenuous sports</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Moderate activities, such as moving a table, pushing a vacuum cleaner, bowling or playing golf</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Lifting or carrying groceries</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Climbing several flights of stairs</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Climbing one flight of stairs</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Bending, kneeling or stooping</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Walking more than a mile</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Walking several blocks</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Walking one block</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Bathing or dressing yourself</td>
<td>1</td>
<td>2</td>
</tr>
</tbody>
</table>

Score range 10-30. Score <25 indicative of severe impairment in physical functioning.


A 70-year-old Surinam woman visited the hospital because of a subcutaneous swelling on the instep of her left foot. She had had this problem for several years, but recently she was experiencing more pain when wearing certain shoes. Physical examination revealed a tender swelling on the foot without any other abnormalities. The clinical diagnosis of a ganglion was made.

At surgical excision, however, a more solid lesion was removed and sent in for histopathological examination. The histological findings showed a sharply demarcated nodule, consisting of partly concentrically arranged fibrous tissue (figure 1). The central area was composed of spherical structures (grains) surrounded by abundant neutrophils, necrotic debris and demarcated by a rim of palisaded epitheloid histiocytes and multinucleated giant cells (figure 2).

WHAT IS YOUR DIAGNOSIS?

See page 128 for the answer to this photo quiz.

Figure 1
Histology of the nodular lesion consisting of partly concentrically arranged fibrous tissue with a central core of neutrophils and spherical structures (H&E; 2x)

Figure 2
Detail of these structures demarcated with a rim of multinucleated giant cells (H&E; 200x)
The differential diagnostic considerations were mycetoma-like infections as (deep) dermatophytic granulomas (pseudomycetomas) and botryomycosis. The former is a fungal infection commonly known as tinea when superficial and limited to the keratinous parts of the skin, i.e. stratum corneum, hair and nails. Rarely the fungi invade the deeper nonkeratinised skin and subcutaneous tissue, where they evoke a granulomatous inflammatory reaction resulting in nodular lesions. Botryomycosis is a chronic, localised lesion of the skin and subcutaneous tissue caused by nonfilamentous bacteria.

Mycetomas are chronic, slowly progressive, but often indolent infections, mainly on the hands and feet, and they are not contagious. Patients are infected by percutaneous implantation of the agent, for instance by a small cut or splinter. Mycetomas are caused by two types of micro-organisms: true fungi (eumycotic mycetoma) or filamentous bacteria (actinomycotic mycetoma). Cultures are needed for the exact determination of those micro-organisms. Mycetomas are usually localised, but local invasion with destruction of soft tissues and bone can be a result. Haematogenous or lymphatic spread is rare. Most infections occur in (sub)tropical areas. Our patient was born in Surinam and has been living in the Netherlands for 15 years now. She had travelled abroad several times, Thailand being her last destination five years ago.

Surgical excision is the treatment of choice for mycetomas. Antifungal therapy does not seem to be necessary for an eumycotic mycetoma, in contrast to an actinomycotic mycetoma for which an early treatment with antibiotics is recommended.

Our patient remained free of pain and was, although not strictly indicated, treated with antifungals by her general physician.

**DIAGNOSIS**

The diagnosis is that of an eumycotic mycetoma. PAS-diastase staining shows that the spherical structures are composed of a dense network of hyphae (figure 3).
Improvement of glycaemic control in type 2 diabetes: favourable changes in blood pressure, total cholesterol and triglycerides, but not in HDL cholesterol, fibrinogen, von Willebrand factor and (pro)insulin

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ABSTRACT

Background: Diabetes mellitus causes a substantial increase in cardiovascular risk, which can only partly be reduced by antihyperglycaemic treatment. We were interested in whether improvement in glycaemic control is associated with improvement of other cardiovascular risk factors. Therefore, we studied among type 2 diabetic patients the association between on the one hand changes in glycaemic control and on the other hand within-subject changes of both classic cardiovascular risk factors and less conventional cardiovascular risk indicators that are typically associated with type 2 diabetes (proinsulin, insulin, fibrinogen, von Willebrand factor and the urinary albumin-creatinine ratio).

Methods: The 214 type 2 diabetic patients were randomly assigned to either a strict fasting capillary glucose target level (<6.5 mmol/l) or a less strict target (<8.5 mmol/l). Duration of follow-up was two years. Since the interventions did not yield statistically significant differences between the treatment arms, we reanalysed the data focusing on within-subject changes of cardiovascular risk factors and indicators across tertiles of average HbA₁c.

Results: Individuals in whom HbA₁c decreased had significant favourable concurrent changes in triglycerides, total cholesterol, blood pressure, and in the albumin-creatinine ratio in those who were normoalbuminuric at baseline. In contrast, these individuals had unfavourable, although not statistically significant, changes in HDL cholesterol, proinsulin, insulin, fibrinogen and von Willebrand factor. In the whole group, fibrinogen increased more than could be expected on the basis of the relationship between fibrinogen and age, namely from 3.5 ± 0.8 to 3.9 ± 0.9 g/l (p value <0.01).

Conclusions: Our results suggest that improvement in glycaemia in type 2 diabetes is associated with significant favourable changes in triglycerides, total cholesterol, blood pressure and, in normoalbuminuric individuals, albumin-creatinine ratio. In contrast, it is not consistently associated with favourable changes in some cardiovascular risk indicators typically associated with diabetes, which may in part explain why antihyperglycaemic treatment does not clearly lower atherothrombotic disease risk.
**INTRODUCTION**

Type 2 diabetes carries an increased risk of cardiovascular disease, which is not fully explained by several well-established risk factors (i.e., variables causally related to cardiovascular disease) associated with type 2 diabetes.\(^{1,2}\) Hyperglycaemia, an important factor in causing micro-angiopathy in type 1 and type 2 diabetes,\(^{3,5}\) is an additional possible explanation for the enhanced cardiovascular risk.\(^{7,9}\) However, the United Kingdom Prospective Diabetes Study (UKPDS) showed that over the first ten years after diagnosis, intensive glucose-control treatment, when compared with conventional treatment, reduced the frequency of microvascular complications but not diabetes-related mortality or myocardial infarction.\(^{10}\) Furthermore, the feasibility trial of the Veterans Affairs Cooperative Study Group (VA CSDM), which compared randomly allocated intensive and conventional glycaemic control, showed a trend towards an adverse effect of intensive glycaemic control on cardiovascular mortality.\(^{11}\)

Another explanation for the excess cardiovascular risk in type 2 diabetes may lie in less conventional cardiovascular risk indicators (i.e., variables whose association with cardiovascular disease may or may not be causal), such as proinsulin and treatment levels, and haemostatic and fibrinolytic abnormalities, which are typically associated with type 2 diabetes. For example, high levels of fibrinogen may partially explain the extra cardiovascular risk in type 2 diabetes.\(^{12,13}\) In addition, cardiovascular risk may be increased by high von Willebrand factor levels (vWF) and microalbuminuria,\(^{14-16}\) which may both reflect endothelial dysfunction.\(^{17,18}\)

We present data on a well-defined cohort of 214 type 2 diabetic patients followed for two years. After the baseline assessment, patients were randomly assigned to either a strict fasting capillary blood glucose target level (<6.5 mmol/l), or a less strict target (<8.5 mmol/l). This study design allowed us to study the effects of two different levels of treatment intensification and analyse the associations between within-subject changes of glycaemia, and changes in lipidemia, blood pressure, proinsulin, insulinemia, plasma fibrinogen, plasma vWF, and the urinary albumin-creatinine ratio (ACR). We were especially interested in whether improvement in glycaemic control was associated with improvement in these cardiovascular risk factors and indicators. Because the diabetic state is thought to be associated with an adverse cardiovascular risk profile, in part through the effects of hyperglycaemia (and insulin resistance), we reasoned that improvement of glycaemic control might be associated with favourable changes of cardiovascular risk factors and indicators. On the other hand, lack of any such associations might to some extent explain the inconsistent effects of improvement of glycaemic control on risk of atherothrombotic disease.\(^{19,20}\)

**MATERIALS AND METHODS**

**Design and measurements**

Participants were treated by their own general practitioner (GP). A single physician (FEEvdD) and a diabetes educator performed three-monthly surveillance of treatment-related parameters at the study centre. Results were sent to the GP. The GP (and the patient) then decided whether a subsequent treatment step according to a standard step-up therapy regimen should be taken. Enrolment to the trial was between June 1992 and December 1993, and the trial ended in December 1995. The Ethical Review Committee of the Free University Medical Centre approved the study protocol. The study was approved and performed before results of the UKPDS were reported. The regimen was a slightly modified version of the practice guidelines for type 2 diabetes of the Netherlands College of General Practitioners.\(^{21}\) An additional feature of the regimen was a stepwise protocol for initiation of insulin therapy by the GP. The regimen had the usual build-up: tablets in increasing doses up to their usual maximum before other blood-glucose-lowering agents were added. In patients with a body mass index ≥27 kg/m\(^2\), metformin was the first step. If the assigned target values for glycaemic control were not reached, a sulphonylurea (SU) – either glibenclamide, gliclazide or glipizide – was added. In patients with a body mass index <27 kg/m\(^2\) SU was the first step. If the assigned target values were not reached on tablets alone, bedtime intermediate-acting insulin was added (and metformin, if any, discontinued). If target values were not reached with this combination therapy, SU was discontinued and twice-daily injections of a mixture of short- and intermediate-acting insulin were started. If glycaemic control remained poor, multiple insulin injection therapy was considered.

Patients were randomly allocated to one of two groups, which differed only in target values for fasting capillary glucose levels. In ‘group 6’, fasting target values for capillary blood glucose were near-normal glycaemia (<6.5 mmol/l). In the other arm (‘group 8’), the fasting treatment target was <8.5 mmol/l, a value considered to be ‘acceptable’.\(^{22}\) Participating GPs were instructed to refrain from any further steps that might lower blood glucose as long as glucose levels were below the allocated target values. Further details of the study population have been described in detail elsewhere.\(^{23}\) Briefly, 372 Caucasian subjects between 40 and 75 years, were invited. After exclusion of subjects with comorbidity and those who were probably nondiabetic, 232 gave informed consent and participated in the study. Three-monthly assessments included levels of glucose, HbA\(_k\) and lipids, treatment modality, body mass index, blood pressure and early morning ACR. Six-monthly assessments included serum creatinine, proinsulin, insulin
and fibrinogen. VWf was measured at baseline, at one and at two years. Systolic and diastolic (Korotkoff V) blood pressure were measured on the right arm of the seated patient with a Hawksley random zero sphygmomanometer.

**Laboratory assessments**

All blood samples were taken in the fasting state. Serum and plasma were stored at -20°C for assessment of proinsulin, insulin and vWF, which took place after closure of the data collection. All other assessments were performed on the same day. Venous glucose was measured in sodium fluoride plasma by the glucose dehydrogenase method (Merck, Germany). HbA1c was determined in EDTA plasma by ion exchange HPLC (reference range: 4.3 to 6.1%; Modular Diabetes Monitoring System, BioRad, the Netherlands). Immunospecific insulin and proinsulin were measured in serum by double-antibody radioimmunoassays (lot SP21, Linco Research, St Louis, USA for insulin, and Lilly in serum by double-antibody radioimmunoassays (lot SP21, Linco Research, St Louis, USA for insulin, and Lilly Laboratory for Clinical Research, Indianapolis, USA for proinsulin).29 Serum total cholesterol, HDL cholesterol and triglycerides were determined by enzymatic colorimetric techniques (CHOD-PAP and CPO-PAP, Boehringer Mannheim, Germany). LDL cholesterol was calculated with the Friedewald formula (not for patients with TG >8.0 mmol/l).29 Fibrinogen was determined in citrated plasma by a spectrophotometric prothrombin time-derived method (ACL 1000, Instrumentation Laboratory, the Netherlands). vWF was determined in heparinised plasma by an ELISA and expressed as a percentage of normal pooled plasma (reference range: 50 to 150%).18,31,32 Urinary albumin was measured by an immunonephelometric method (sensitivity limit: 6.2 mg/l; Beckman, Ireland). For calculation of urinary ACR, we only used early morning samples negative to dipstick tests for nitrite and leucocytes (86% of all samples).

**Statistical analysis**

We studied the effects of the random assignment by comparison of groups 6 and 8 at the last available measurements, using t tests, χ² tests or Mann-Whitney U tests. Comparison of baseline and follow-up measurements were carried out using paired t tests, McNemar’s χ² tests or Wilcoxon’s signed-rank tests. Since the interventions did not yield statistically significant differences between the treatment arms we reanalysed the data focusing on within-subject changes in cardiovascular risk factors and indicators in the entire cohort. Cohort analysis of a randomised clinical trial (RCT) is legitimate, because an RCT is a cohort, with intervention as a determinant. Adjusting the analysis for the intervention (high versus low target) eliminates possible bias caused by this determinant.30 The cardiovascular risk factors and indicators considered were proinsulin, insulin, lipids, blood pressure, fibrinogen, vWF and ACR. Normoalbuminuria was distinguished from microalbuminuria and albuminurea using 3.5 mg/mmol as the cut-off.34 To study within-subject changes, change rates (slopes) were calculated for each patient separately by linear regression analysis based on all the available measurements. Preliminary analyses showed that patients with less than the full two years of follow-up were likely to confound analyses. Therefore, we chose to confine all analyses using these change rates to those patients who completed two years of follow-up (n=166). Relations between change rates of glycaemic parameters and change rates of outcome measures were studied by univariate scatter diagrams, and by partial correlations after adjustment for sex, diabetes duration, and group 6 or 8. We also addressed these associations by using analysis of variance (ANOVA) and trend analysis, comparing the change rates of the outcome measures across tertiles of HbA1c change rates. All multivariate analyses were performed with and without additional adjustment for change in body weight.

**RESULTS**

During the first year, ten patients found participation too much of a burden, six moved and one died (7%). One outlier (a woman with a BMI of 59) was excluded from the analyses. Thus, 106 patients in group 6 and 108 patients in group 8 were included in the analyses. During the second year, 12 of these remaining 214 patients found further participation too burdensome and dropped out, eight were lost to follow-up and two died. At closure of the data collection, 26 patients had not yet reached the two-year visit. Mean duration of follow-up at the time of the analysis was 22 months. Table 1 shows the characteristics of the study population. Fasting plasma glucose was significantly lower in group 6 as compared with group 8, but no clinically meaningful contrast in HbA1c had been achieved at a mean follow-up of 22 months. Furthermore, the difference in treatment intensity did not yield statistically significant differences between groups 6 and 8 at follow-up in any of the other outcome measures. In the total population, glycaemic control improved slightly from baseline to follow-up, as reflected by lowering of the fasting glucose levels and lower variability of mean HbA1c. Furthermore, total and LDL cholesterol, and blood pressure decreased. In contrast, body mass index, fibrinogen and ACR (increase), and HDL cholesterol (decrease) deteriorated during follow-up. Fasting serum insulin increased significantly due to 23 patients starting insulin injections during follow-up. At baseline 16.5% of the participants were on antihypertensive medication and 13.5% lipid-lowering medication, versus 24.1 and 16.7% at follow-up.
We subsequently analysed changes in the outcome measures in patients who had completed two years of follow-up (n=166) by tertiles of change rates of HbA1c (table 2). Results for tertiles of fasting glucose change rates were similar (data not shown). Triglycerides (figure 1), total cholesterol and blood pressure changed most favourably in the tertile with the HbA1c decrease, as compared with the other two tertiles. These associations could not be ascribed to the prescription of more lipid-lowering agents or antihypertensives in that tertile (data not shown). Only in patients with an ACR <3.5 mg/mmol at baseline did changes in ACR show a significant favourable association with HbA1c change rates. Changes in vWF were minor and not related to HbA1c change rates. Overall, fibrinogen increased, but mainly in the lowest and middle tertiles of HbA1c change (table 2 and figure 2). We checked whether adjustment for weight change altered any of the relations under study. This was not the case (data not shown).

As shown in tables 1 and 2, there were relatively large mean changes in fibrinogen, which could not be explained by changes in glycaemic control (table 2), or by changes in the prevalence of smokers (data not shown). Upon further

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**Table 1**

<table>
<thead>
<tr>
<th>Characteristics at baseline and at follow-up in group 6 (n=106), group 8 (n=108) and the total population (n=214)</th>
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</thead>
<tbody>
<tr>
<td><strong>AT BASELINE</strong></td>
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<tr>
<td><strong>GROUP 6</strong></td>
</tr>
<tr>
<td>Sex (% male)</td>
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<tr>
<td>Age (years)</td>
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<tr>
<td>Diabetes duration</td>
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<tr>
<td>Cardiovascular history</td>
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<tr>
<td>Hypertension (% yes)</td>
</tr>
<tr>
<td>Smoker (%)</td>
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<tr>
<td>Treatment modality (%)</td>
</tr>
<tr>
<td>Diet alone</td>
</tr>
<tr>
<td>Metformin (M)</td>
</tr>
<tr>
<td>Sulphonylurea (SU)</td>
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<tr>
<td>SU + M</td>
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<tr>
<td>Insulin (with or without SU)</td>
</tr>
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<td>Body mass index (kg/m²)</td>
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<td>Men</td>
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<tr>
<td>Women</td>
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<td>Fasting plasma glucose (mmol/l)</td>
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<tr>
<td>HbA1c (%)</td>
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<tr>
<td>HDL cholesterol (mmol/l)</td>
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<tr>
<td>Triglycerides</td>
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<td>Total cholesterol</td>
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<tr>
<td>LDL cholesterol</td>
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<tr>
<td>Systolic blood pressure (mmHg)</td>
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<tr>
<td>Diastolic blood pressure</td>
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<tr>
<td>Insulin (pmol/l) (n=143)</td>
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<tr>
<td>Pro-insulin (n=143)</td>
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<tr>
<td>Fibrinogen (g/l) (n=179)</td>
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<tr>
<td>Von Willebrand factor (%)</td>
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<tr>
<td>Albumin-creatinine ratio (mg/mmol)</td>
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<tr>
<td>Baseline</td>
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<tr>
<td>Baseline &gt;3.5 (n=40)</td>
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<tr>
<td>ACR at baseline &lt;3.5</td>
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<td>ACR at baseline ≥3.5</td>
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Data are percentages, means ± standard deviations, or medians (10th-90th centile). HbA1c = glycated haemoglobin, ACR = albumin creatinine ratio. Mean duration of follow-up: 22 months. Cardiovascular history is defined as at least one of the following: myocardial infarction, angina pectoris, stroke, transient ischaemic attack, and intermittent claudication. Hypertension is defined as a systolic blood pressure >165 mmHg, and/or a diastolic blood pressure >90 mmHg and/or blood pressure-lowering medication. Group 6 versus group 8, p<0.05; follow-up versus baseline, p<0.01; follow-up versus baseline, p<0.01.
There were significant baseline differences in fibrinogen levels among patients on diet treatment, those on tablets, and those using insulin, which remained when adjusted for age, sex and diabetes duration. These differences were still present at follow-up. Patients using insulin had significantly higher fibrinogen levels than those on diet, while those on tablets had intermediate fibrinogen levels. From baseline to follow-up, fibrinogen increased from 3.7 ± 0.9 to 4.3 ± 0.9 g/l (mean ± SD, p=0.03) in patients who started insulin treatment (n=23), and from 3.3 ± 0.6 to 3.8 ± 0.9 g/l (p<0.001) in those who changed from diet alone to tablet treatment (n=35). In the subgroup who retained baseline treatment throughout the study (n=93), fibrinogen increased from 3.5 ± 0.8 to 3.8 ± 0.9 g/l (p=0.003). In contrast, in the subgroup who stayed on diet treatment alone throughout the study (n=37), fibrinogen remained unchanged (3.4 ± 0.7 g/l), despite significant worsening of glycaemic control. Among individuals who started insulin treatment at some point during the observation period, the change in fibrinogen levels (measured at 0, 6, 12, 18 and 24 months) was about twice as large in the six-month interval in which the
insulin was started (0.28 g/l) as in the six-month intervals preceding and following the interval in which insulin was started (0.14 g/l). For comparison, plasma glucose continued to decrease after starting insulin, body mass index continued to increase, triglycerides decreased and ACR decreased. The vWF levels during the one-year interval with and without the initiation of insulin treatment showed a slight increase (7.2%) and a slight decrease (-5.2%), respectively. Changes in HDL cholesterol were not clearly associated with any specific treatment step. The duration of storage of serum and plasma samples (range: 18 to 48 months) did not significantly correlate with the measured levels of insulin, proinsulin and vWF (correlation coefficients: .01, .09, and .07, respectively). This suggests that the quality of the blood samples did not deteriorate significantly during storage at -20°C.

**DISCUSSION**

No differences between the two intervention arms were found, except for a slightly lower fasting plasma glucose level. The lack of a greater contrast in fasting glucose levels, and in HbA1c, may have been due to two obstacles. Firstly, the eligibility criteria employed may not have been sufficiently restrictive. Secondly, compliance with the study protocol may not have been sufficient to achieve the desired divergence between the treatment arms. Therefore, we reanalysed our data and focused on the question whether different levels of success in lowering the main parameter of antihyperglycaemic treatment, HbA1c, would be accompanied by different changes in various cardiovascular risk factors and indicators. As expected, we found that triglycerides, total cholesterol, blood pressure and ACR (only when <3.5 mg/mmol at baseline) exhibited significantly more favourable changes in the tertile with decreasing HbA1c compared with the other tertiles. In contrast, these favourable changes were not found for proinsulin, insulin, fibrinogen and vWF (*table 2*). For fibrinogen and vWF, this is in accordance with earlier studies. Moreover, in the whole group, fibrinogen even showed a significant increase. In conclusion, improvement of glycaemic control – with the strategy employed, i.e. a conventional step-up regime consisting of metformin or sulphonylurea in increasing dosages and insulin if the assigned target values were not reached with metformin or sulphonylurea – was associated with concurrent favourable changes in some, but not all, the cardiovascular risk factors and indicators.

Our results concerning the favourable association of HbA1c with ACR in those subjects without microalbuminuria and the absence of such an association in those with microalbuminuria are in accordance with findings in type 1 diabetes that intensified glycaemic control may not have favourable effects on the progression of the urinary albumin excretion rate once microalbuminuria exists. In addition, Levin et al. reported that creatinine clearance among subjects with microalbuminuria deteriorated more rapidly than among subjects without microalbuminuria, regardless of intensity of glycaemic treatment. Glycaemic changes were not significantly associated with changes in fibrinogen. However, fibrinogen increased after the initiation of insulin treatment (or after an increase of pharmacological treatment in general). Adjustment for diabetes duration, age and sex did not substantially affect differences between those who started insulin and the rest of the population (data not shown). Furthermore, fibrinogen showed a gradual overall increase during the study. However, the yearly change rate was far greater than the relationship of fibrinogen with age in nondiabetics would suggest (except in the subgroup who remained on diet treatment alone throughout the study). In a cross-sectional study, De Boever et al. found a 0.14 g/l increase per ten years in a nondiabetic population aged between 35 and 59 years. These findings may be of particular interest. Firstly, considerable variations in cardiovascular event rates could be attributable to relatively small variations in fibrinogen levels. Meade et al. have shown that a fibrinogen level elevation of 0.6 g/l was associated with an 84% increased risk of stroke within the next five years. In addition, Danesh et al. showed, in a meta-analysis of prospective studies, an 85% increased risk for coronary heart disease for each increment in fibrinogen of 1.0 g/l.42 Secondly, the VA CSDM feasibility trial reported an unexpectedly high incidence of cardiovascular events in a group of male type 2 diabetic patients who started insulin treatment. Fibrinogen level rose significantly in the first year and returned to baseline at two years.43 Insulin and proinsulin have been recognised as potentially atherogenic substances, although it is not clear whether insulin by injection has effects comparable with those seen in hyperinsulinaemia related to insulin resistance. A study in the offspring of hypertensive men demonstrated that fibrinogen is closely related to plasma insulin and most components of the insulin resistance syndrome. In contrast De Feo et al. demonstrated that induced insulin deficiency was associated with an increase in fibrinogen, suggesting an acute phase protein response. In vitro experiments lend support to possible adverse effects of proinsulin and insulin on fibrinolysis, acting via both liver and endothelium. The present findings could be in accordance with an effect of treatment intensification on the production of fibrinogen by hepatocytes in vivo. In vitro studies, however, have shown inconsistent results in this respect. Incubation of hepatocytes with insulin for less than 24 hours had no influence on fibrinogen synthesis, while studies with longer incubation showed no effect, decreased or increased fibrinogen synthesis. Moreover,
in our study fibrinogen change rate was not related to insulin and proinsulin change rates (data not shown). In sum, our data do not provide an explanation for the fibrinogen changes that were observed. In conclusion, our study suggests that further improvement of glycaemic control in moderately well-controlled type 2 diabetes is not associated with concurrent favourable changes in some cardiovascular risk indicators that are typically associated with type 2 diabetes. We stress that our findings on glycaemic control cannot be distinguished from the therapeutic strategy employed, i.e. a conventional step-up regime consisting of metformin or sulphonylurea in increasing dosages and insulin if the assigned target values were not reached. Therefore, we cannot exclude that improvement of glycaemic control through other strategies may yield better results in terms of cardiovascular risk factor amelioration. Nevertheless, our findings may provide a framework for understanding the disappointing effects of intensified glycaemic control on risk of macrovascular disease. Our finding that fibrinogen levels may increase after starting insulin treatment of particular concern and warrants further study.

REFERENCES


42. Danesh J, Collins R, Appleby P, Peto R. Association of fibrinogen, C-reactive protein, albumin, or leukocyte count with coronary heart disease:


We describe a patient with rheumatoid arthritis (RA) who developed a nephrotic syndrome during treatment with a fully human recombinant monoclonal antibody against TNFα (adalimumab, Humira, Abbott). The proteinuria disappeared spontaneously after cessation of anti-TNFα treatment and relapsed after rechallenge, pointing to anti-TNFα as the culprit. Although renal biopsy disclosed a membranous glomerulopathy, the clinical picture was more compatible with minimal lesion glomerulopathy. The pathogenesis of this side effect is not clear; several mechanisms could in theory lead to these abnormalities.

ABSTRACT

We describe a patient with rheumatoid arthritis (RA) who developed a nephrotic syndrome during treatment with a fully human recombinant monoclonal antibody against TNFα (adalimumab, Humira, Abbott). The proteinuria disappeared spontaneously after cessation of anti-TNFα treatment and relapsed after rechallenge, pointing to anti-TNFα as the culprit. Although renal biopsy disclosed a membranous glomerulopathy, the clinical picture was more compatible with minimal lesion glomerulopathy. The pathogenesis of this side effect is not clear; several mechanisms could in theory lead to these abnormalities.

INTRODUCTION

Blocking the action of tumour necrosis factor α (TNFα) with monoclonal antibodies directed against TNFα has been shown to yield fast and impressive responses in patients with rheumatoid arthritis (RA). Adverse effects are usually mild and except for enhanced susceptibility to certain infections, the most prominent being *M. tuberculosis*, and rare systemic lupus erythematosus (SLE)-like syndromes, no specific side effects have been reported. We describe a patient who developed a nephrotic syndrome as an adverse effect of treatment with the fully human anti-TNFα monoclonal antibody adalimumab.

CASE REPORT

Our 64-year-old patient was diagnosed with a rheumatoid factor positive, ANA negative erosive rheumatoid arthritis (RA) in 1982. His medical history was unremarkable, except for chronic obstructive pulmonary disease (COPD). In the following years he was treated with D-penicillamine followed by parenteral aurothioglucose. This was stopped in 1984 because of proteinuria up to 1.5 g/l. After cessation of intramuscular gold administration, the proteinuria completely disappeared. Therapy was continued with methotrexate, followed again by aurothioglucose, which was stopped in 1990 because of exacerbation of the RA. Proteinuria did not appear during this second episode of gold therapy nor did it later. Sulphasalazine treatment was initiated, later combined with methotrexate and prednisone. The latter combination therapy was stopped due to lack of efficacy. In light of the persistent disease activity the patient was recruited in May 1997 for a phase I trial with a fully human monoclonal antibody against TNFα (adalimumab, Humira®, Abbott). Treatment with anti-TNFα was started in a dose of 1.0 mg/kg body weight every two weeks intravenously, later increased to 3.0 mg/kg according to the study protocol. Concomitant medication included prednisone 10 mg a day, diclofenac 75 mg twice a day, alternating calcium and etidronic acid, omeprazol 20 mg twice a day, paracetamol 500 mg as needed, ipratropiumbromide 40 μg four times a day, salmeterol 100 μg twice a day and doxycycline 100 mg once daily. There was an excellent clinical response to anti-TNFα treatment: the disease activity score (DAS, a combined disease activity measure for RA) dropped from 5.89 to 2.41 and C-reactive protein levels went from 142 to 24 mg/l. Prednisone was gradually tapered to 2.5 mg/day.
In April 1998, the patient developed progressive pitting oedema of both lower extremities. He also noticed a weight gain from 73 to 84 kg in two weeks. On admission, blood pressure was 150/80 mmHg. Physical examination revealed bilateral leg oedema and evidence of pleural and peritoneal fluid. Laboratory investigations showed serum creatinine 82 μmol/l, serum albumin 15 g/l, cholesterol 7.0 mmol/l, haemoglobin 5.5 mmol/l. ANA negative, and normal complement C3 and C4. Proteinuria averaged 16.7 g/24 h, selectivity index was 0.07 (highly selective).

A renal biopsy was performed. By light microscopy segmental irregularities of the glomerular basement membrane (GBM) of all glomeruli were demonstrated, suggestive of a membranous glomerulopathy, such as vacuolisation, spike formation or some ring-like structures. Podocytes, endothelial cells and mesangial areas were normal (figures 1A and 1B). Congo red staining did not reveal amyloid. Immunofluorescence revealed fine granular deposits of particularly IgG and C3 along the capillary wall in a characteristic membranous pattern (figure 2). No AA amyloid deposits were found using a monoclonal antibody. By electron microscopy, podocytes were hypertrophic and showed retraction of the foot processes (figure 3).

In some segments of the GBM, irregularly spaced subepithelial electron dense deposits accompanied by spike formation were observed. Other segments displayed a broad and abnormal GBM, incorporating many complexes that were less electron dense and resolving (figure 3). Several segments of the GBM, however, did not show any alterations or electron dense aggregates.

The patient was treated with diuretics and an angiotensin-converting enzyme inhibitor; anti-TNFα treatment and diclofenac were withdrawn (figure 4). The proteinuria eventually disappeared after three months. Interestingly, symptoms of RA relapsed in the same week that the nephrotic syndrome disappeared. Diclofenac therapy was reinstated but the proteinuria did not reappear until after reintroduction of anti-TNFα treatment. The patient was treated with prednisone 60 mg daily and cyclophosphamide 100 mg daily. Within one week, proteinuria had completely disappeared. Cyclophosphamide was stopped and prednisone quickly tapered to 10 mg daily. At this point, anti-TNFα was started again. The nephrotic syndrome did not reappear until the prednisone was reduced to a dose of 5 mg daily and subsided again after the dose was increased to 10 mg. Since December 1998, the patient is being treated with anti-TNFα in combination with 10 mg prednisone daily and has retained an excellent clinical response without proteinuria.

**DISCUSSION**

In our patient a nephrotic syndrome developed during treatment with adalimumab. Renal biopsy disclosed a membranous glomerulopathy. The clinical course strongly incriminated anti-TNFα as the culprit, since proteinuria disappeared after withdrawal of anti-TNFα treatment and reappeared upon rechallenge. Furthermore, we could exclude that the other likely candidate, the NSAID diclofenac, was the cause of the nephrotic syndrome. Membranous nephropathy has been frequently reported in patients with RA. In a series of 110 biopsies in patients with RA, membranous nephropathy was observed in 19 patients (17%). In most patients with RA and membranous nephropathy, treatment with gold salts or D-penicillamine has been incriminated as the cause. However, it has also been suggested that patients with RA may be prone to the development of membranous nephropathy, even in the absence of gold or D-penicillamine treatment. It is however unclear if in these latter patients the use of NSAIDs as causative agents was excluded, as an association also exists between NSAID use and membranous nephropathy. Our patient may thus represent a case of membranous nephropathy caused by anti-TNFα treatment. TNFα has been incriminated in glomerular diseases. *In vitro* studies...
have shown that TNFα can increase glomerular permeability, possibly by inducing oxygen radical production. Of particular interest for our case are the observations of Neale et al. These investigators have demonstrated the unique presence of TNFα in the visceral epithelial cells and in the subepithelial deposits in patients with membranous nephropathy. TNFα was not found in other forms of glomerular disease. Since membranous nephropathy is the result of an interaction between antibodies and antigens present on the surface of the glomerular epithelial cells, one can speculate that in our patient the membranous nephropathy was caused by interaction of the anti-TNFα antibodies with TNFα present on visceral epithelial cells. Infliximab, a chimeric anti-TNF monoclonal antibody, can bind to membrane-bound TNF and bring cells into apoptosis. This characteristic has, however, not yet been demonstrated for adalimumab. Also, to the best of our knowledge, TNFα is only present in the cytoplasm of glomerular epithelial cells and an increased production is only found in patients with established membranous glomerulopathy. Furthermore, we found no evidence for TNFα in the glomerular basement membrane in our patient using biotin-labelled IgG1 anti-TNFα MoAb (monoclonal antibody) or a polyclonal anti-human TNFα by indirect immunofluorescence. The clinical data point to an alternative explanation for the cause of the nephrotic syndrome. The highly selective proteinuria and the immediate and brisk response to treatment with steroids strongly suggest the presence of minimal change nephropathy. In patients with established membranous nephropathy proteinuria disappears quite slowly if at all, even during treatment with immuno-suppressive agents. We did not observe any complete remissions of proteinuria within three months after the start of treatment in 34 patients with membranous nephropathy who were treated with high-dose prednisone. Likewise, in patients with RA with membranous nephropathy caused by either gold or D-penicillamine, proteinuria disappears gradually; in most patients the maximal proteinuria is even reached one to two months after stopping the offending drug. In such patients, treatment with prednisone apparently does not influence the duration of proteinuria. The finding of subepithelial deposits certainly does not prove that the proteinuria is caused by the membranous

nephropathy. In the rat model of membranous nephropathy, injection of monoclonal antibodies against the podocyte antigen megalin results in the formation of small subepithelial deposits that are not paralleled by the development of proteinuria. Likewise, a membranous nephropathy with subepithelial deposits in the absence of proteinuria has been observed in hypertensive patients treated with captopril. Notably, in the study mentioned above in patients with RA, three patients were described with membranous nephropathy but no proteinuria. Furthermore, it is well established that in patients with membranous nephropathy the deposits can remain visible after remission has occurred, even for many years. In our patient, many deposits seem to be old, incorporated in the GBM and dissolving, suggesting that they may have been caused by the previous gold therapy. Alternatively, the deposits may have been induced by the diclofenac treatment. Experience with infliximab has shown that renal side effects are very rare. Increase of antinuclear and antidouble-stranded DNA antibodies is, however, frequent and a few cases of drug-induced systemic SLE-like syndromes have been described, although these cases did not include renal involvement. As antinuclear and antidouble-stranded DNA antibodies and other signs and symptoms of SLE were absent in our patient, our case does not represent a drug-induced SLE. As noted, the nephrotic syndrome in our patient was highly steroid sensitive but also steroid dependent. A dose of 10 mg prednisone was able to prevent the reoccurrence of proteinuria. Since a considerable number of patients who receive anti-TNF treatment are concurrently being treated with prednisone, the potential of anti-TNF to induce proteinuria may thus be masked. It remains to be established if this side effect becomes more frequent if anti-TNF treatment is instituted in patients who do not receive steroids.

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Adrenal carcinoma causing secondary amyloidosis: report of the first case in the literature

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CASE REPORT

A 53-year-old male was admitted to our hospital in December 1998 with mild abdominal pain, weakness and fatigue. On admission, his vital signs were normal. He had conjunctival pallor; breath sounds on the lower part of the left lung were diminished. The left upper quadrant of the abdomen was mildly tender to palpation. He had no lymphadenopathy and no organomegaly.

Laboratory data revealed a low haematocrit level (23.4%) and a high erythrocyte sedimentation rate (55 mm/h). Urea, creatinine, glucose, albumin levels, urinalysis, and the tumour markers CEA, CA 19.9, AFP, β-hCG, NSE were normal. Also, the steroid hormone levels, namely, testosterone, dehydroepiandrosterone, dehydroepiandrosterone sulphate, androstenedione, aldosterone, oestriadiol, 11-deoxycortisol, cortisol, 17-OH-progesterone, were within the normal ranges. Thorax CT showed masses containing necrotic hypodense areas in both lung fields, the biggest of which was 9 cm in diameter. Abdominal MRI showed a regularly contoured mass measuring 7 x 9 cm in diameter originating from the left adrenal gland and containing calcified areas. The mass was displacing the tail of the pancreas anteriorly and the spleen laterally.

A thoracic fine needle aspiration (FNA) biopsy to the biggest pulmonary mass in the left lung was conducted. The histopathological examination of the biopsy specimen showed adrenal carcinoma cells with moderate degrees of anaplasia. The FNA biopsy of the left adrenal mass also revealed the same pathology. The immunohistochemical staining of both biopsy specimens was positive for vimentin,
but negative for cytokeratin and D11. According to the adrenal carcinoma staging system proposed by MacFarlane and modified by Sullivan et al., we diagnosed the patient as stage IV adrenal carcinoma. The patient was started on treatment with mitotane 1000 mg orally, four times a day. He was discharged to come to regular follow-up visits but he was lost to follow-up.

In August 2000, 20 months after the initial admission, the patient was readmitted with complaints of dyspnoea, increasing fatigue and swelling of the legs. He stated that he had not been taking the mitotane regularly as it caused gastrointestinal upset. His blood pressure was 100/65 mmHg, pulse rate 88 beats/min and temperature 36.7°C. His conjunctivae were pale. Breath sounds were diminished on the lower parts of both lungs. He had no lymphadenopathy, no hepatomegaly and no ascites, but the spleen was enlarged 2 cm below the costal margin. He also had pretibial oedema.

Laboratory data revealed: haematocrit 0.26%, leucocytes 7,900/mm³ (with a normal differential), platelets 391,000/mm³, ESR 130 mm/h, urea 73 mg/dl, creatinine 4.1 mg/dl, total protein 4.8 g/dl and albumin 1.0 g/dl. Serum glucose, electrolytes, ALAT, ASAT, bilirubins and lipids were normal. Urinalysis revealed 4+ proteinuria and oval fat bodies in the sediment. Creatinine clearance was 15 ml/min and the protein excretion was 10 g/day. Serum protein electrophoresis revealed elevated alpha and beta globulin regions with normal gamma globulins. C₃ and C₄ complement levels, and serum and urine immunoelectrophoresis were all normal. Antinuclear antibody, anti-dsDNA, p-ANCA, c-ANCA, cryoglobulins, HBsAg, anti-HCV and anti-HIV were negative. PPD skin test (Mantoux) was also negative.

Thorax CT revealed bilateral pleural effusion with persistence of the right and bilateral pulmonary metastases containing necrotic hypodense areas, the biggest of which measured 9 cm in diameter and was on the left.

Figure 1 Thorax CT showing pleural effusion more prominent on the right and bilateral pulmonary metastases containing necrotic hypodense areas, the biggest of which measured 9 cm in diameter and was on the left.

Figure 2 Abdominal MRI showing a partially regularly contoured mass with heterogenous intensity measuring 7 x 9 cm in diameter. The mass originates from the left adrenal gland and displaces the pancreas and the spleen.
DISCUSSION

In this case, the diagnosis of adrenal carcinoma was reached by FNA biopsy, which has a high degree of accuracy and is an important tool in diagnosing adrenal masses. If the FNA biopsy of adrenal tissue is positive for malignancy, this has a positive predictive value of 100%. It has also been reported that most adrenal malignancies measure >6 cm in diameter. The survival rate of patients with stage I or II disease is better, while those with stage IV disease have the shortest survival. One study reported that 10 of 11 adrenal carcinoma patients died after 24 months. Until now, mitotane has been shown to be the only drug to have some effect in the treatment of patients with metastatic adrenal carcinoma.

Systemic amyloidosis might be associated with malignancies, the majority of which are accounted for by the immunocyte dyscrasias. Hodgkin’s disease and renal cell carcinoma are other cancers which might be associated with secondary amyloidosis. In a large autopsy series, amyloidosis was found to have an incidence of 0.4% in cancer patients, the most common underlying malignancies being multiple myeloma, Hodgkin’s disease and renal cell carcinoma. However, it is uncommon to encounter amyloidosis in other types of cancers. In a literature search, no cases of surrenal carcinoma ending in secondary amyloidosis could be cited.

Serum amyloid A (SAA) is an acute phase protein produced by the liver after stimulation from activated macrophages and SAA serves as the precursor of AA-type amyloid. Tumour cells activate macrophages, thereby triggering the formation of amyloid. Also, the patient may lack the proteolytic enzyme which breaks down SAA protein and this causes formation of AA-like fragments available for amyloid formation. In addition, it has been reported that SAA and AA proteins are polymorphic; so, some molecules might be more ‘amyloidogenic’ than others. Secondary amyloidosis is more frequent in renal cell carcinoma than other cancers because it grows relatively slowly leading to a long-term stimulation of SAA protein production. In other cancers, patients probably die before they have had time to develop secondary amyloidosis. In our case, the patient was diagnosed when he had stage IV adrenal carcinoma. We do not exactly know how much time elapsed before the first occurrence of the disease and the diagnosis of stage IV carcinoma. After diagnosis, he was prescribed mitotane, which he did not take regularly. In spite of this, he had stable disease for nearly 20 months with no regression or progression. This might have led to a longer period of inflammatory stimulus leading to secondary amyloidosis. Also, no clinical and laboratory evidence of any superimposed chronic infections or inflammatory diseases was found. In our country, one of the frequent causes of secondary amyloidosis is familial Mediterranean fever (FMF). However, our patient’s family history and clinical presentation was not compatible with FMF or familial amyloidosis.

Clinical and histological resolution of systemic amyloidosis after removal of the tumour has been reported. Our patient did not undergo surgery as it was shown to be of no use in metastatic adrenal disease. He was prescribed mitotane which he did not take regularly and there was no regression of disease activity within this period.

Here, we present the first report of a case of adrenal carcinoma, which caused secondary amyloidosis ending in nephrotic syndrome. This complication should be born in mind in long-standing cases with proteinuria that are unresponsive to therapy.

REFERENCES


Acute renal failure in patients with glomerular diseases: a consequence of tubular cell damage caused by haematuria?

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ABSTRACT

We describe three patients with acute renal failure after the onset of gross haematuria. In all patients a presumptive diagnosis of rapidly progressive glomerulonephritis was made and immunosuppressive therapy initiated. A renal biopsy was performed in two patients, which showed evidence of IgA nephropathy. Extracapillary proliferation was seen in a few glomeruli. The most notable abnormality was acute tubular necrosis with intraluminal erythrocytes and cell debris. In the third patient, who was known to have longstanding glomerular haematuria, acute tubular necrosis was considered likely after review of the urinary sediment. Despite the fact that immunosuppressive therapy was stopped, renal function rapidly returned to normal in all these patients. We feel that our patients and additional literature data demonstrate that in patients with glomerular disease a reversible acute renal failure can occur that is caused by acute tubular necrosis mediated by haematuria. Recognition of this entity will prevent unnecessary long-term immunosuppressive therapy.

INTRODUCTION

The nephritic syndrome is characterised by haematuria, oliguria, hypertension and often progressive renal failure. In most patients the underlying cause is a glomerulonephritis. Well-known examples are the extracapillary (crescentic) glomerulonephritis in patients with a systemic vasculitis or anti-GBM disease and the endocapillary proliferative glomerulonephritis after a streptococcal infection. In the former group of patients immunosuppressive treatment must be started without delay to ensure recovery of renal function. Therefore, in many patients immunosuppressive therapy is started at the moment that an extracapillary glomerulonephritis is suspected, often before renal biopsy findings are available.

It is not well known that patients with macroscopic glomerular haematuria may develop a reversible acute renal failure due to acute tubular necrosis, which is attributed to haemoglobin-mediated tubular cell injury. We present three patients to demonstrate the clinical picture.

CASE REPORT 1

A 63-year-old woman with hypertension (150/100 mmHg, ventricular response 100 beats/min) and atrial fibrillation was admitted to the hospital because of vasculitic skin lesions. During the admission she developed proteinuria and renal insufficiency. Physical examination revealed hypertension, and red and purple skin lesions. Diuresis amounted to 600 ml/day, and her urine was stained dark brown, compatible with macroscopic haematuria. The urinary sediment showed more than 50, mostly dysmorphic erythrocytes per high power field, and numerous red cell casts, which are virtually diagnostic of glomerulonephritis. Additional laboratory analysis (antinuclear antibodies (ANA), antineutrophilic cytoplasmic antibodies (ANCA), anti-GBM antibodies, cryoglobulins, and complement profile) was unremarkable. A renal biopsy disclosed a focal extracapillary glomerulonephritis. On immunofluorescence IgA deposits were seen in the mesangium and to a lesser extent in the capillary loops. These clinical and histological findings supported a diagnosis of Henoch-Schönlein
purpura with renal involvement. Given the progressive nature of the renal insufficiency, therapy with prednisone and cyclophosphamide was instituted. The clinical course (figure 1) was complicated by traumatic hip fracture and urinary tract infections. After an initial recovery of renal function the serum creatinine rose again. A second opinion was sought and the renal biopsy was revised. In the biopsy 55 glomeruli where present, 15 of which showed signs of recent extracapillary inflammation. Most remarkable, however, were signs of extensive tubular cell damage and tubular cell necrosis (figure 2A) and erythrocytes in the tubuli (figure 2B). The histological findings confirmed that the patient was suffering from a glomerulonephritis with IgA deposits, in the context of Henoch-Schönlein purpura. However, it seemed unlikely that the glomerular abnormalities could fully explain the renal insufficiency and we assumed that the acute tubular necrosis, so predominantly present, was the cause of the renal insufficiency and that there was no indication for immunosuppressive therapy. The treatment with prednisone and cyclophosphamide was stopped. The further clinical course was complicated by the development of acute congestive heart failure with further deterioration of renal function (figure 1). Eventually there was a near complete recovery of renal function.

**CASE REPORT 2**

A previously healthy, 36-year-old woman developed a sore throat and fever. One day later she consulted the family physician, who prescribed a penicillin antibiotic. The same day she noticed that the urine production had diminished and that it was stained dark brown (tea-colour). On the third day she was admitted to a hospital and because of progressive renal failure she was transferred to our hospital two days later. On admission the blood pressure was 145/95 mmHg, with a pulse rate of 64 beats/min. The pharynx was red with inflamed tonsils. There was no oedema present. Laboratory values on admission were: creatinine 729 µmol/l, urea 15.3 mmol/l, C-reactive protein 157 mg/l, anti-streptolysin titre normal, anti-DNase B was elevated, ANA negative, ANCA negative, complement C3 and C4 normal. Urinalysis showed gross haematuria, in which it was not possible to discern the morphological aspects of the erythrocytes. Renal function and urinary output progressively declined (figure 3). Renal biopsy was not feasible due to a prolonged bleeding time. Since an extracapillary glomerulonephritis as cause of the progressive renal failure could not be ruled out immunosuppressive therapy with steroids was instituted. Haemodialysis was started. After normalisation of the bleeding time a renal biopsy was performed 10 days after onset of the haematuria. In the biopsy 15 glomeruli were present, and 5 glomeruli showed...
signs of extracapillary proliferation. On immunofluorescence there were discrete mesangial IgA deposits. There was evidence of tubular damage, and in some tubules erythrocytes and cellular debris from necrotic cells were present (figures 4A and 4B). We concluded that the patient had IgA nephropathy, presenting with gross haematuria after an upper respiratory tract infection. Renal failure was thought to have developed as a consequence of tubular cell damage. Immunosuppressive therapy was rapidly decreased and stopped. The further clinical course was uneventful (figure 3) and renal function completely normalised. Outpatient follow-up showed that there was residual albeit very slight proteinuria. The urinary sediment persistently showed some dysmorphic erythrocytes.

CASE REPORT 3

A 36-year-old woman was known with haematuria from her youth. The urinary sediment at that time showed many dysmorphic erythrocytes and erythrocyte casts. Renal function was normal and there was no proteinuria or hypertension. Benign glomerular haematuria was diagnosed, possibly as a consequence of IgA nephropathy. She now presented with a three-day history of fever, a sore throat, headache, nausea and vomiting. A physician had prescribed Pheneticillin. The day after onset of the fever, her urine was stained dark and the urinary output was reduced. On physical examination she did not appear acutely ill. The blood pressure was 140/75 mmHg and the pulse 76 beats/min. Further examination was unremarkable, no oedema was present. The serum creatinine was markedly elevated (682 μmol/l). Urinary sodium was 32 mmol/l, the urinary sediment showed 20 to 50 mostly dysmorphic red blood cells per high power field with red cell casts. On ultrasound examination the kidneys were normal without outflow obstruction. Since a rapidly progressive renal failure due to extracapillary glomerulonephritis was considered possible, i.e. methylprednisolone and oral cyclophosphamide were started. On the following day the urinary sediment further revealed many granular casts and tubular epithelial cells. The urinary output seemed to increase. As the sediment was compatible with acute tubular necrosis and in view of the absence of hypertension and sodium retention, immunosuppressive therapy was stopped. The clinical course was uneventful with a complete recovery (figure 5).

DISCUSSION

We describe three patients who developed acute renal failure during a period of gross haematuria. In two patients a definite diagnosis of IgA nephropathy was made, in patient 3 a diagnosis of IgA nephropathy was very likely given the

Figure 3
Time course of renal function and diuresis in patient 2

Figure 4
Tubular cell injury in the kidney biopsy of patient 2
There are signs of cell and nuclear atypia (panel A). In some tubuli erythrocytes can be recognised between granular debris (arrows) (panel B). (Methenamine silver-staining, A 400x, B 400x.)
medical history. It is well known that patients with IgA nephropathy frequently experience periods of macroscopic haematuria that are often provoked by fever, infection or exercise. Typically there is a short interval of hours or days between the provoking factor (e.g. an upper respiratory tract infection) and the appearance of haematuria. In contrast, in postinfecitious (poststreptococcal) glomerulonephritis the interval between the infection and the occurrence of haematuria ranges from 10 to 21 days.

In patients with renal failure the presence of dysmorphic erythrocytes and red cell casts is virtually pathognomonic for the presence of glomerular lesions. It is therefore almost inevitable that the glomeruli receive the most attention on histopathological examination of the renal biopsy. In two of our patients evidence of an IgA nephropathy was found in the glomeruli. In 25 to 30% of the glomeruli, extracapillary proliferation was present. The presence of extracapillary proliferation points to serious damage of the capillary wall, often with necrosis. An extracapillary glomerulonephritis is most often associated with a vasculitis. However, it is well known that in many forms of primary glomerulonephritis, some glomeruli show extracapillary proliferation. In patients with IgA nephropathy extracapillary proliferation is found in 17% of the patients and in 5 to 50% of the glomeruli. 8 Signs of extracapillary proliferation are found more often in periods of macroscopic haematuria. It is very important that one realises that renal insufficiency per se cannot be ascribed to the presence of a small number of glomeruli with signs of extracapillary proliferation. In anti-GBM disease or vasculitis with rapidly progressive renal failure (serum creatinine >500 µmol/l) crescents are found in at least 50% and usually in more than 80% of the glomeruli. 4,5

In our patients the renal biopsies showed signs of tubular cell damage (case report 1). In case report 2 the pathological abnormalities of the tubular apparatus were less noticeable, which can be explained by the fact that the biopsy was taken late after the onset of haematuria, in the recovery phase.

In case report 3 the urinary sediment findings were highly suggestive of acute tubular necrosis. Furthermore, in this case, there were no other signs of the nephritic syndrome such as hypertension, oedema or urinary sodium retention. Acute renal failure caused by macroscopic haematuria has been described previously in patients with IgA nephropathy. 3,6 The renal failure is thought to be caused by the erythrocytes in the tubuli and attributed to either intratubular obstruction or a direct toxic effect on the tubular epithelial cells by haemoglobin or iron. Alternatively, we cannot exclude that cytokines produced by the inflammatory cells may contribute to the development of tubular cell injury. In all circumstances it is likely that volume depletion will enhance the risk of developing acute renal failure. The incidence of renal failure caused by haematuria is probably high, although it is often overlooked and not recognised. In a prospective study in which the occurrence of renal failure during episodes of macroscopic haematuria was studied, 11 of 29 patients developed renal failure. 4 The mean serum creatinine was 271 µmol/l (108-603 µmol/l), the maximal value being reached three to ten days after the onset of haematuria. Renal function normalised in all cases, although the time needed for recovery varied from 15 to 70 days. Renal biopsies taken within four days after the onset of haematuria showed considerable tubular cell damage, with erythrocyte casts in most of the tubuli. Extracapillary proliferation was noticed in 3 to 15% of the glomeruli. A striking finding was that in late biopsies, taken 12 days after the onset of haematuria, there was only mild tubular damage and that hardly any erythrocytes could be found in the tubular lumina. Other authors have largely confirmed these findings. 5,6 In some of the patients reported in the latter studies renal insufficiency was so severe that haemodialysis was necessary. The number of crescents found in renal biopsies varied between 0 and 37%. In almost all cases renal function recovered.

Acute renal failure due to haematuria has also been described in patients with other forms of glomerulonephritis. 7 Notably, in one paper acute renal failure was described in a marathon runner, who developed haematuria shortly after finishing the race. The renal biopsy did not disclose any glomerular abnormality, but erythrocyte casts filled the tubular lumina. 8

In our patients we cannot formally exclude that the short-term use of immunosuppressive therapy contributed to the improvement of renal function. However, most patients with extracapillary glomerulonephritis do not respond to short-term (days) prednisone therapy. Furthermore, even the most vigorous treatment with steroids, cyclophosphamide and plasmapheresis mostly results in a slow improvement of renal function, with clear evidence of renal failure being still present two months after the start of treatment. 9

These findings are in sharp contrast with the course of renal recovery in our patients.
In conclusion, patients with glomerular diseases and macroscopic haematuria may develop acute renal failure due to tubular necrosis. In a patient with rapidly progressive glomerulonephritis and macroscopic haematuria the absence of hypertension and oedema, and an increased urinary sodium concentration must raise suspicion to include acute tubular necrosis in the differential diagnosis. In these patients volume depletion must be vigorously treated. The recognition of this entity is of importance as it prevents the institution and maintenance of potentially harmful immunosuppressive therapy.

REFERENCES

Since 1996, Theo Elfrink (1923) has been cycling to Mehr, his favourite place just across the border in Germany, almost every day. There, he settles himself in a farmyard to draw and paint. This farmyard with its barns, animals, fences and constantly changing flora now completely determinates his work. Elfrink is an intuitive painter, always painting what he feels. Both in his drawings and paintings, he works from his heart with a style and gesture which finds its roots in ‘expressionism’.

Theo Elfrink exhibits his work widely in our country and in a number of foreign countries, and has now been honoured with the ‘Keizer Karel’ award of the city of Nijmegen. This is added to the several prizes he has already received.

This month’s printing (45 x 35 cm) is made by the ‘dry point’ technique. A very limited edition (5) of these original prints is available at a price of € 215. You can order the print at Galerie Unita, Rijksstraatweg 109, 6573 CK Beek-Ubbergen, the Netherlands or by e-mail: galerie-unita@planet.nl.