FALK SYMPOSIUM 159

IBD 2007 – Achievements in Research and Clinical Practice

Edited by
N. Tözün, Marmara University, Istanbul, Turkey
G. Mantzaris, Evangelismos Hospital, Athens, Greece
Ü. Dağlı, Yüksek İhtisas Hospital, Ankara, Turkey
J. Schölmerich, Universität Regensburg, Germany

IBD 2007 – Achievements in Research and Clinical Practice

FALK SYMPOSIUM 159

IBD 2007 – Achievements in Research and Clinical Practice

Edited by

N. Tözün
Marmara University
Istanbul
Turkey

Ü. Dağlı
Yüksek İhtisas Hospital
Ankara
Turkey

G. Mantzaris
Evangelismos Hospital
Athens
Greece

J. Schölmerich
Universität Regensburg
Regensburg
Germany

Proceedings of the Falk Symposium 159 held in Istanbul, Turkey, May 4–5 2007

Springer

Library of Congress Cataloging-in-Publication Data is available.

ISBN-13 978-1-4020-6986-4

Published by Springer,
PO Box 17, 3300 AA Dordrecht, The Netherlands

Sold and distributed in North, Central and South America
by Springer,
101 Philip Drive, Norwell, MA 02061 USA

In all other countries, sold and distributed
by Springer,
PO Box 322, 3300 AH Dordrecht, The Netherlands

Printed on acid-free paper

Printed and bound in Great Britain by MPG Books Limited, Bodmin, Cornwall.

Contents

List of principal contributors

HA Akpinar
Dokuz Eylül University School of Medicine
Department of Internal Medicine
Division of Gastroenterology
Inciraltı
35340 Izmir
Turkey

S Baratsis
Evangelismos Hospital
Surgical Clinic A
45-47 Ipsilantou str.
10675 Athens
Greece

I Bjarnason
Department of Gastroenterology
King's College Hospital
Denmark Hill
London, SE5 9PJ
UK

AF Çelik
Istanbul University
Cerrahpasa Faculty of Medicine
Department of Gastroenterology
34098 Cerrahpasa/Istanbul
Turkey

J-F Colombel
Service de Maladies de l'Appareil Digestif et de la Nutrition
CHRU Claude Huriez
59037 Lille Cedex
France

Ü Dağli
Yüksek İhtisas Hospital
Gastroenterology Division
Sıhhiye/Ankara
Turkey

G D'Haens
Imelda Gastrointestinal Clinical Research Center
Imeldalaan 9
2820 Bonheiden
Belgium

D Epstein
Division of Gastroenterology
Department of Medicine
Groote Schuur Hospital
Anzio Road
Observatory 7925 Cape Town
South Africa

A Gangl
Division of Gastroenterology and Hepatology
Department of Medicine III
Medical University of Vienna
Währinger Gürtel 18-20
1090 Vienna
Austria

P Gionchetti
Department of Internal Medicine and Gastroenterology
Policlinico S. Orsola
Via Massarenti 9
40138 Bologna
Italy

V Gross
Innere Medizin II
Klinikum St Marien Amberg
Mariahilfbergweg 7
92224 Amberg
Germany

HÖ Hamzaoğlu
Marmara University
School of Medicine
Department of Gastroenterology
Tophanelioglu Cad. 13-15
34662 Üsküdar/Istanbul
Turkey

U Klotz
Dr. Margarete Fischer-Bosch Institut für Klinische Pharmakologie
Auerbachstrasse 112
70376 Stuttgart
Germany

U Korman
Istanbul University
Cerrahpasa Medical Faculty
Department of Radiology
34300 Istanbul
Turkey

EA Kouroumalis
Department of Gastroenterology
University Hospital
PO Box 1352
71100 Heraklion
Crete, Greece

E Langholz
Department of Medical Gastroenterology F
Gentofte Hospital
University of Copenhagen
2900 Hellerup
Denmark

G Mantzaris
Evangelismos Hospital
Department of Gastroenterology
45-47 Ipsilantou Street
10675 Athens
Greece

P Marteau
AP-HP
Département Médicochirurgical de Pathologie Digestive
Hôpital Lariboisière
2 rue A Paré
75010 Paris
France

NJMcC Mortensen
Department of Colorectal Surgery
John Radcliffe Hospital
Headley Way Headington
Oxford, OX3 9DU
UK

MF Neurath
Laboratory of Immunology
I. Medical Clinic
University of Mainz
Langenbeckstr. 1
55131 Mainz
Germany

HS Odes
Department of Gastroenterology and Hepatology
Soroka Medical Center and Ben Gurion University of the Negev
PO Box 151
Beer Sheva 84101
Israel

P Rutgeerts
Department of Medicine
Division of Gastroenterology
University Hospital
Herestraat 49
3000 Leuven
Belgium

WJ Sandborn
Inflammatory Bowel Disease Clinic
Division of Gastroenterology and Hepatology
Mayo Clinic
200 First Street SW
Rochester, MN 55905
USA

BE Sands
MGH Crohn's and Colitis Center
Gastrointestinal Unit
Massachusetts General Hospital
165 Cambridge Street, 9th Floor
Boston, MA 02114
USA

J Schölmerich
Klinik und Poliklinik für Innere Medizin I
Klinikum der Universität Regensburg
93042 Regensburg
Germany

M Toruner
Department of Gastroenterology
Ankara University Medical School
Ibni - Sina Hospital
Ankara 06100
Turkey

JK Triantafillidis
Iera Odos 354
12461 Haidari Athens
Greece

B Vucelic
Division of Gastroenterology
University Hospital Rebro
Kispaticeva 12
10000 Zagreb
Croatia

List of chairpersons

S Arslan
Hacettepe University
Faculty of Medicine
Department of Gastroenterology
06100 Sıhhıye/Ankara
Turkey

C Arvanitakis
PO Box 60322
57001 Thermi/Thessaloniki
Greece

S Boyacıoğlu
MESA Hospital
Gastroenterology Division
Yasam Caddesi No. 5
06510 Söğütözü/Ankara
Turkey

D Buğra
Istanbul University
Faculty of Medicine
Department of General Surgery
Millet Caddesi
34390 Çapa/Istanbul
Turkey

M Farthing
St George's Hospital
Medical School
Department of Gastroenterology
Cranmer Terrace
London, SW17 0RE
UK

C Gasche
Medizinische Universität Wien
Innere Medizin IV
Gastroenterologie/Hepatologie
Währinger Gürtel 18-20
A-1090 Wien
Austria

H Herfarth
University of North Carolina
School of Medicine
Gastroenterology/Hepatology
Center for GI Biology and Disease
Chapel Hill, NC 27514
USA

W Jafri
Tha Aga Khan University Hospital
Department of Medicine
Gastroenterology Section
Link Building
74800 Karachi
Pakistan

D Karamanolis
Evangelismos Hospital
Department of Gastroenterology
45-47 Ipsilantou St.
10675 Athens
Greece

IE Koutroubakis
University of Heraklion
Department of Gastroenterology
PO Box 1352
71110 Heraklion
Greece

O Manoussos
25 Martiou str. no. 27
15561 Athens
Greece

A Nakos
Hippokration General Hospital
Department of Gastroenterology
114 Vas. Sophias Ave
11527 Athens
Greece

DK Podolsky
Massachusetts General Hospital
School of Medicine
GI Unit
55 Fruit Street
Boston, MA 02114-2696
USA

D Rachmilewitz
Shaare Zedek Medical Center
Department of Medicine
PO Box 3235
91031 Jerusalem
Israel

I Rozanes
Istanbul University
Faculty of Medicine
Department of Radiology
Millet Caddesi, Capa
34390 Istanbul
Turkey

RW Stockbrügger
Academisch Ziekenguis Maastricht
Afd. Interne Geneeskunde
Werkgroep Gastroent/Hepatology
Postbus 5800
6202 AZ Maastricht
The Netherlands

N Tözün
Mamara University
School of Medicine
Department of Gastroenterology
Tophaṇelioğlu Cad. 13-15
34662 Üsküdar/Istanbul
Turkey

A Tromm
Innere Medizin
Evangelisches Krankenhaus
Hattingen
Bredenscheider Str. 54
45525 Hattingen
Germany

E Tsianos
University Hospital of Ioannina
Department of Internal Medicine
PO Box 1186
45110 Ioannina
Greece

S Vermeire
Univ. Ziekenhuis Gasthuisberg
Gastroenterology Unit
Herestraat 49
3000 Leuven
Belgium

Preface

No field in medicine has been as challenging and flamboyant as that of IBD over the past ten years. From a nebulous bulk of speculation about the aetiology and pathogenesis of the disease we have smoothly sailed to specific targets to attack, although the mystery behind the pathogenesis and risk factors of the disease remains still unveiled.

Falk Symposium 159, which was held in the historic city of Istanbul, had the main goal of delineating the role of various factors leading to IBD, with special emphasis on the influence of environment in this less well-known region of the Eastern Mediterranean. Concerns about the steady rise of complicated Crohn's disease incidence in young adults and the refractory cases were raised, the dilemmas in the differential diagnoses of IBD from infectious or other causes like Behçet's disease were discussed, and the exciting contribution of today's tools to the diagnosis were underlined by distinguished speakers.

Tough cases of IBD raised an ardent debate between the speakers and the discussants and a panel of experts summarized the resulting knowledge supported with the most recent evidence-based data in the literature.

Were we enlightened or did we move from 'cocky ignorance to miserable uncertainty' as Mark Twain said about the path of education? There are still some dark corners which need to be illuminated by further work; however this symposium has certainly left its imprint on the memory of IBD enthusiasts, not only by giving impetus for future research but also by clarifying some aspects of the disease behaviour, the contribution of novel and old techniques to the diagnosis, and the reassesment of the role of surgery in IBD.

This monograph is a summary of the lectures delivered during the meeting and comprises the presentation of nearly all speakers. Regardless of how long the present knowledge will last, we already know that in IBD 'the future advances but slowly'.

Above all, what is needed for science to advance is time and patience: "Time's glory is to calm contending kings, to unmask falsehood and bring truth to light" as Shakepeare beautifully phrased.

We would like to thank all the speakers who did a sterling job by the excellence of their lectures, the quality of their contributions and the excitement they conveyed from their chairs to the audience. The interaction was great!

We would also like to thank Falk Foundation e.V., Freiburg, Germany for lending support to this meeting and to Dr Martin Falk in particular who by his generous contribution made this symposium a landmark in IBD management in Turkey and elsewhere. Finally, from the roots to the tiny little details the contribution of local partner ARIS was outstanding!

Until the next encounter, please keep warm memories from Istanbul, the city of of wisdom, dream and legends.

N. Tözün, Ü. Dağli, G. Mantzaris, J. Schölmerich

Section I
Epidemiology, risk factors and genetics

Chair: RW STOCKBRÜGGER and E TSIANOS

1
Epidemiology and genetics of inflammatory bowel disease in Turkey

Ü. DAĞLI

INTRODUCTION AND AIMS

Inflammatory bowel disease (IBD) is a chronic and relatively common disorder of uncertain aetiology. Accordingly, a considerable number of epidemiological studies over the past four decades have sought to define its geographic, temporal, demographic and ethnic trends in incidence and prevalence. Epidemiological data can provide valuable insights into the pathogenesis, associations and natural history of the disease.

Systematic epidemiological studies have not been performed in Turkey, and the incidence and prevalence of IBD are unknown. There has been no population-based study of IBD in Turkey. In 2004 the IBD Society decided to create a database to record IBD patients, which was announced to gastroenterologists in national meetings. There were 19 medical centres involved. A standard protocol was adopted for data collection and analysis, and in this chapter we report the final epidemiological findings.

METHODS

In order to investigate IBD epidemiology in Turkey, 10 areas throughout Turkey with medical centres were sampled. For inclusion the medical centre had to be a university hospital or grade A tertiary hospital, with diagnostic facilities available for high-quality endoscopy, radiology and pathology.

The medical records of hospitalized or outpatient IBD patients from 19 hospitals in these 10 areas were recorded on the website of the IBD Society (www.ibhd.org.tr) beginning in September 2004.

The diagnosis of IBD was made in accordance with previously established internationally accepted criteria[1] on the basis of clinical, endoscopic and histopathological and/or radiological evidence after exclusion of infection and other recognized causes of inflammation. The diagnosis had to be of at least 6 months' duration.

Disease involving the rectum was defined as proctitis, that below the splenic flexure as left-sided colitis, and above the splenic flexure as pancolitis. The extent of disease was defined as the greatest extent recorded at any stage in the course of the illness by colonoscopy or radiography. Crohn's disease (CD) was categorized as ileal, small bowel, large bowel, ileocaecal, and perianal disease. The majority of patients were followed up regularly.

Data collected from all patients included demographics, smoking history, level of education, occupation, family history of IBD, disease distribution and behaviour, and extraintestinal manifestations.

Prevalence and incidence was calculated from three (Ankara, İzmir and Edirne) of the 10 cities because in these three cities all gastroenterology clinics participated in this study. However, demographic and clinical data presented herein are taken from the records of all patients from the 10 cities.

Statistics

Statistical analysis was performed by SPSS 10.0 (Statistical Package for Social Sciences). Numeric variables were compared by *t*-test and nominal variables by chi-square tests. The significance level was set as 0.05.

RESULTS

Demographics

A total of 3954 cases were recorded from 19 hospitals in 10 areas in Turkey from 2004 to 2007. Diagnoses included 2938 patients with ulcerative colitis (UC) (74%), 975 with CD (25%) and 41 with indeterminate colitis (1.0%) (Figure 1). Among the patients with IBD, 1726 were female and 2187 male, with a male/female ratio of 1.27 (Table 1). The mean age at diagnosis was 34.3 $\pm$13 years for CD and 38.4$\pm$13.0 years for UC ($p = 0.001$).

A histogram of the age of the patients showed the monophasic distribution with one peak between 20 and 49 years for UC and CD (Figure 2). In addition, the median disease duration from onset to diagnosis was 1.1 years for UC, and 1.6 years for CD.

Prevalences

All patients permanently living in Ankara, İzmir and Edirne were included in the prevalence calculation. Prevalences of UC and CD as of 1 April 2007 are shown in Table 2. Records of 1991 IBD patients from five centres in Ankara, 959 IBD patients from three centres in İzmir and 150 IBD patients from a single centre in Edirne were recorded in the database.

Incidences

During the observation period 228 new patients with UC and 125 new patients with CD were diagnosed in these three cities. The mean incidence rate for UC was 4.1 and for CD was 2.6 cases per 100 000 persons per year (Table 3).

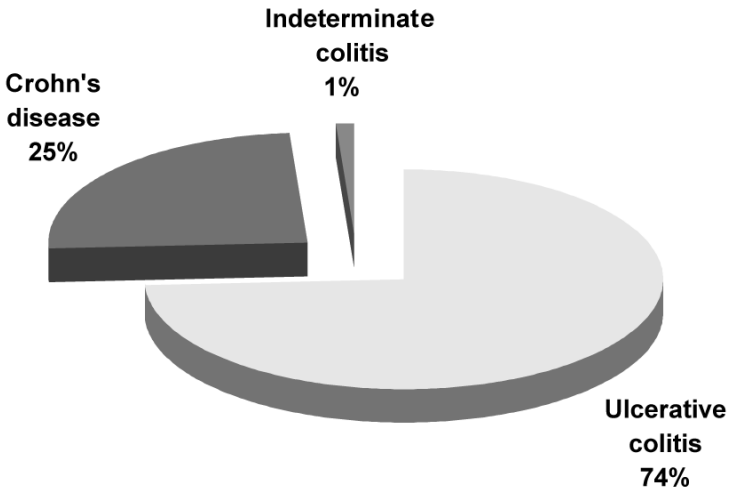

Figure 1 Distribution of patients according to the type of IBD

Table 1 Distribution of IBD patients according to gender

Patients	*Female n (%)*	*Male n (%)*	*Male/female*
Ulcerative colitis	1291 (43.9)	1647 (56.1)	1.27
Crohn's disease	435 (44.6)	540 (55.4)	1.24
IBD	1726 (44.1)	2187 (55.9)	1.27

Risk factors

A familial history of IBD was present in 4.3% of UC patients and in 4.2% (41/975) of CD patients ($p > 0.05$).

Most of the patients with UC and CD live in urban areas. In this cohort 61.5% of patients with UC and 58.6% with CD had an elementary level of education. Cigarette consumption rate in UC patients was 31.3% and in CD was 55.8% at the time of diagnosis, and the difference was statistically significant ($p = 0.001$). History of appendectomy was 9.4% in CD and 2.4% in UC and the difference was again statistically significant ($p = 0.001$). The higher appendectomy rate in CD was probably due to a false appendicitis diagnosed in misdiagnosed CD. The history of measles was approximately the same between UC and CD. The percentage of tonsillectomy was low in all patients. Amoebiasis infection was present in 9.6% of UC, and 2.1% of CD patients (Table 4) .

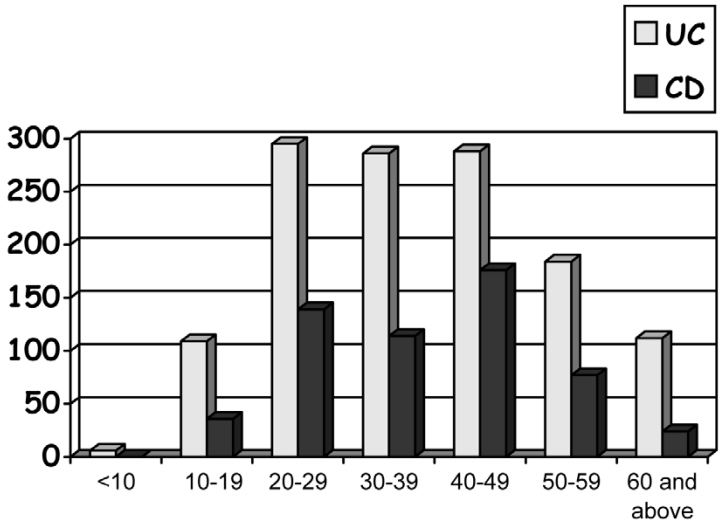

Figure 2 Distribution of age at diagnosis in UC and CD

Table 2 Prevalences of UC and CD in Ankara, İzmir and Edirne

	Patient number	*Population*	*Prevalence (10^5)*
Ankara			
IBD	1991	4 453 000	44.7
UC	1346		30.2
CD	461		10.4
İzmir			
IBD	959	3 769 000	25.4
UC	717		19
CD	190		5
Edirne			
IBD	150	772 000	39.7
UC	126		33.3
CD	9		2.4
Total			
IBD	3100	8 600 000	36.0
UC	2189		25.5
CD	660		7.7

Table 3 Incidences of UC and CD in Ankara, İzmir and Edirne

	Patient number	*Population*	*Incidences* (10^5)
Ankara			
IBD	200	4 453 000	4.5
UC	125		2.8
CD	75		1.7
İzmir			
IBD	123	3 769 000	3.7
UC	81		2.1
CD	42		1.1
Edirne			
IBD	30	772 000	3.9
UC	22		2.8
CD	8		1.0
Total			
IBD	353	8 600 000	4.1
UC	228		2.6
CD	125		1.4

Table 4 Demographics of Turkish UC and CD patients

	UC	*CD*	*p-Value*
Mean age at diagnosis (years)	38.4 ± 13.0	34.3 ± 13	0.0001
Education (%)			
Elementary	1808 (61.5)	573 (58.6)	0.043
High school	559 (19.0)	227 (23.2)	
University	512 (19.4)	177 (18.1)	
Residence			
Urban	2441 (83.1)	908 (93.0)	0.001
Rural	497 (16.9)	69 (7.0)	
Family history	126 (4.2)	41 (4.3)	0.911
Smoking (%)	920 (31.3)	544 (55.8)	0.001
Appendectomy (%)	70 (2.4)	92 (9.4)	0.001
Tonsillectomy (%)	49 (1.7)	21 (2.2)	0.321
Measles (%)	423 (14.4)	118 (12.1)	0.072
Amoebiasis (%)	282 (9.6)	20 (2.1)	0.001

Phenotypes

The predominant form of UC was left-sided colitis, which affected almost 45% of the patients. Pancolitis was present in 27% and proctitis in 28% of the studied population.

In CD patients the colon was affected in 25%, small bowel in 22%, ileocolon in 34%, and ileocaecum in 18%.

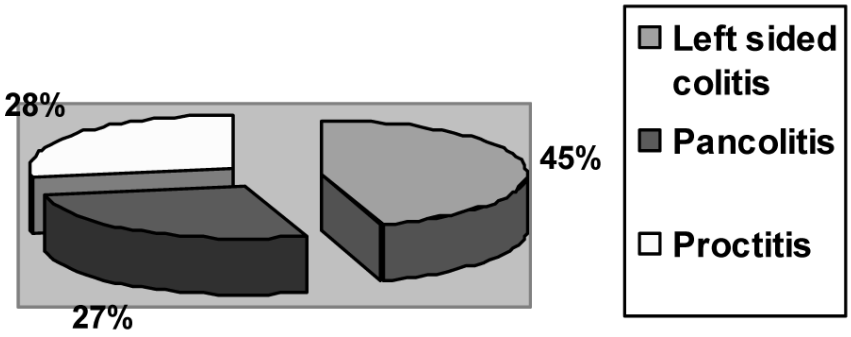

Figure 3 Distribution of UC patients according to localization

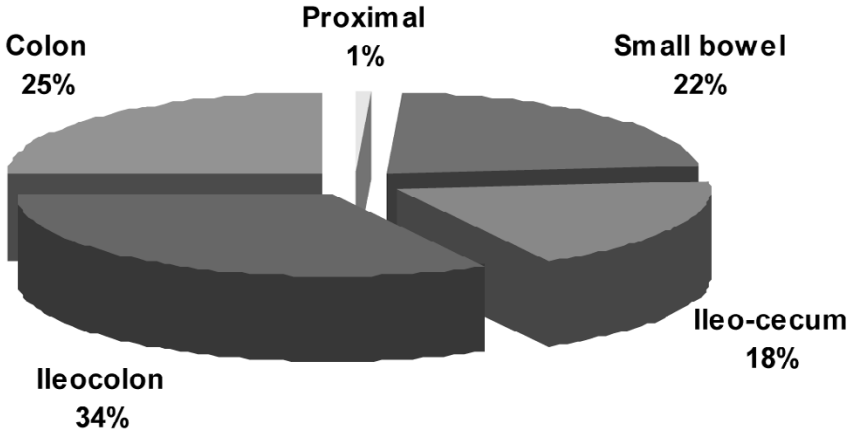

Figure 4 Distribution of CD patients according to localization

At least one extraintestinal manifestation was present in 13.8% of UC patients and in 21.7% of CD patients. The most common findings were peripheral arthropathy and sacroileiitis in both groups, followed by cholecystopathy. Primary sclerosing cholangitis was found in 24 UC patients and in nine CD patients (Tables 5 and 6).

Table 5 Extraintestinal manifestations in patients with UC

Extraintestinal complications	*n*	*%*
Absent	2534	86.2
Skin diseases	8527	20.91
Eye complications	34	1.14
Hepatobiliary	92	3.13
Musculoskeletal	155	5.27
Genitourinary system	40	1.37
Others	56	1.91

Table 6 Extraintestinal manifestations in patients with CD

Extraintestinal complications	*n*	*%*
Absent	741	78.3
Skin	30	3.1
Eye	16	1.6
Hepatobiliary	41	4.2
Musculoskeletal	108	11.1
Genitourinary system	16	1.6

Genetics of IBD in Turkey

Strong epidemiological evidence concerning a genetic contribution to the pathogenesis of IBD has stimulated efforts to identify susceptibility genes for both of its major clinical forms, CD and UC. Although UC and CD are distinct disorders there are some similarities in their clinical phenotypes. They affect both sexes equally and occur at all ages.

The occurrence of familial clustering of IBD is well established. First -degree relatives of both CD and UC patients have an estimated 10-fold increase in the risk of developing the same disease compared to the general population. The epidemiological database revealed a positive family history in 126 of 2938 UC patients (4.3%) and 41 of 975 CD patients (4.2%).

In Turkey, investigations of IBD genetic markers are still scarce. There is a study from Turkey which associated with HLA-DRB alleles with UC patients. In UC patients Uyar et al. found a positive association with the HLADRB1* 1502 allele and a negative association with the DRB1*B allele. They also found that patients with positive pANCA more often revealed a HLADRB1*0701 allele and negative patients more often revealed a HLADRB1*1502 allele[2].

There are two studies associated with polymorphism in the NOD2/CARD15 gene. In one of the studies polymorphisms in the NOD2/CARD15, NOD1/ CARD4 and ICAM-1 genes were investigated in Turkish patients with IBD and healthy control groups[3]. The genotypes of 70 patients with CD, 120 patients with UC and 106 healthy subjects were compared. In this study the three previously described CD predisposing variants of the NOD2/CARD15 gene

and the polymorphism examined in the NOD1/CARD4 and ICAM-1 genes were not found to be associated with UC and CD. Although no significant association was found between these variations and CD, the heterozygous mutant genotype for the insertion mutation at position 3020 was observed in only two of the 70 CD patients. However, the rare allele was not detected in either the UC or healthy control group[3]. In previous studies of populations in Europe and the United States the frequency of the 3020insC mutation has been reported to be 2–8%, but it was 1.4% in Turkish CD patients.

Another study on the same subject was reported by Uyar et al.[4]. Their study group consisted of 56 CD patients and 100 healthy controls. According to this study, among the three NOD2/CARD15 mutations only the G908R variant allele in CD was found to be associated with disease. The frequency of the G908R variant allele in CD cases was 8% compared with 0% in controls. No association was determined between the G908R variant and the clinical course of CD[4].

There are three studies published from Turkey regarding prothrombotic gene mutations in IBD. All three studies found that the prevalences of prothrombin G20210A gene and factor V Leiden gene mutations were statistically insignificant among CD patients and the control group[5–7]. In the last study there was a statistical difference between the proportions of the mutated allele frequencies of beta-fibrinogen-455G-A, MTHFR A1298C and ACE-1/D in IBD[7].

Although the exact mechanisms underlying IBD pathogenesis are obscure, the mucosal immune system activation and secretion of powerful inflammatory cytokines may play a role. The proinflammatory cytokines interleukin-1 (IL-1) and tumour necrosis factor alpha (TNF-α) seem to play an essential role in the inflammation. Most of the changes in inflammation can be triggered by the activities of TNF-α and IL-1 sharing many of their functions, especially those leading to the amplification of immunological and inflammatory processes. In Turkish patients with IBD, cytokine gene polymorphisms were researched[8]. The aim of that study was to examine the allelic polymorphisms that can determine the immune response levels in TNF-α, IL-1β, interleukin-1 receptor antagonist (IL-1RN) and IL-10 genes, and to investigate their roles in the inflammatory pathway in IBD. No significant differences were found in the allele and genotype frequencies of the polymorphisms in these genes between the patients with UC and healthy controls[8].

DISCUSSION/CONCLUSION

Turkey is situated between Europe and Asia; therefore, epidemiological data may be different from that of its neighbours. The prevalence of IBD in Turkey is lower than in other European countries but higher than the Asian populations data. Due to the lack of a solid population-based study these data may not represent the real prevalence of IBD in Turkey .

The peak age of IBD onset in Turkey is similar to that observed in the East and West. The majority of IBD cases are diagnosed among middle-aged patients (31–50 years), with a male predominance.

At the clinical stage the distribution and extent of disease are similar to those classically seen in both the West and the East. The most common clinical form of UC is left -sided colitis and of CD is the ileocolonic form. Most IBD patients are settled in urban areas.

The studies associated with mutations of the NOD2/CARD15 gene in Turkish CD patients are controversial. According to the results of all the genetic studies, genetic factors may not be important in Turkish IBD patients.

Appendix: Turkish IBD Study Group participants (centre and investigators)

Ankara: Dr Mehmet Bektaş, Dr Hülya Çetinkaya, Department of Gastroenterology, School of Medicine, Ankara University. Dr Serap Aslan, Department of Gastroenterology, School of Medicine, Hacettepe University. Dr Hilmi Ataseven, Dr İlhami Yüksel, Dr Işılay Nadir, Dr Serkan Uysal, Dr Başak Çakal, Dr Aysel Ülker, Dr Ülkü Dağlı, Clinics of Gastroenterology, T.C. Yüksek İhtisas Hospital. Dr Candan Tuncer, Department of Gastroenterology, School of Medicine, Gazi University. Dr Yüksel Ateş, Dr Ahmet Erdil, Department of Gastroenterology, School of Medicine, Gülhane University.

Antalya: Dr Hilmi Dikici, Dr İnci Süleymanlar, Department of Gastroenterology, School of Medicine, Akdeniz University.

Dıyarbakir: Dr Şerif Yılmaz, Dr Vedat Göral, Department of Gastroenterology, School of Medicine, Dicle University.

Edırne: Dr Ahmet Tezel, Department of Gastroenterology, School of Medicine, Trakya University.

Eskışehır: Dr Ayşegül Özakyol, Dr Tülay Sarıçam, Department of Gastroenterology, School of Medicine, Osmangazi University.

İstanbul: Dr Yekta Kişioğlu, Dr Nefise Barlas Ulusoy, Clinics of Gastroenterology, American Hospital. Dr Ali Tüzün İnce, Dr Oya Övünç, Clinics of Gastroenterology, Haydarpaşa Numune Hospital. Dr Zeynel Mungan, Dr Aykut Feryal Çelik, Department of Gastroenterology, School of Medicine, İstanbul University. Dr Hülya Över Hamzaoğlu, Department of Gastroenterology, School of Medicine, Marmara University.

İzmır: Dr Serdal Demir, Dr Belkıs Ünsal, Clinics of Gastroenterology, İzmir Atatürk Hospital. Necla Osmanoğlu, Department of Gastroenterology, School of Medicine, Ege University. Dr Hale Akbaylar, Department of Gastroenterology, School of Medicine, Dokuz Eylül University.

Mersın: Dr Orhan Sezgin, Dr Engin Altıntaş, Department of Gastroenterology, School of Medicine, Mersin University.

Trabzon: Dr Yusuf Uzun, Department of Gastroenterology, School of Medicine, Karadeniz Teknik University.

Uşak: Dr Derya Akın, Clinics of Gastroenterology, Uşak State Hospital

References

1. Lennard-Jones JE. Classification of inflammatory bowel disease. Scand J Gastroenterol Suppl. 1989;24:2–9.
2. Uyar FA, İmeryüz N, Saruhan Direskeneli et al. The distribution of HLA-DRB alleles in ulcerative colitis patients in Turkey. Eur J Immunogenet. 1998;25:293–6.
3. Özen SC, Dağlı Ü, Kılıç MY et al. NOD2/CARD15, NOD1/CARD4/, and ICAM-1 gene polymorphism in Turkish patients with inflammatory bowel disease. J Gastroenterol. 2006; 41:304–10.
4. Uyar FA, Hamzaoğlu Över H, Türe F et al. Distribution of common CARD15 variants in patients with sporadic Crohn's disease: cases from Turkey. Dig Dis Sci. 2006;51:706–10.
5. Törüner M, Erkan Ö, Soykan İ et al. Factor V Leiden, prothrombin G20210A, and MTHFR gene mutations in inflammatory bowel disease. Turk J Gastroenterol. 2004;15: 250–2.
6. Hamzaoğlu Över H, Abacı N, Türe F et al. Prothrombotic gene mutations and Crohn's disease; is there any association? Hepato-Gastroenterology. 2005;52:1467–9.
7. Yılmaz Ş, Bayan K, Tüzün Y et al. A comprehensive analysis of 12 thrombophilic mutations and related parameters in disease: data from Turkey. J Thromb Thrombolysis. 2006;22:205–12.
8. Celik Y, Dağlı U, Kılıç MY et al. Cytokine gene polymorphisims in Turkish patients with inflammatory bowel disease. Scand J Gastroenterol. 2006;41:559–65.

2
Inflammatory bowel disease in Greece: 25-years' epidemiological and clinical studies

J. K. TRIANTAFILLIDIS, P. CHERACAKIS, E. MERIKAS, G. MALGARINOS, F. GEORGOPOULOS, I. FOUSKAS, M. MYLONAKI and E. GIANNAKOULOPOULOU

INTRODUCTION

The clinical course of inflammatory bowel disease (IBD) ulcerative colitis (UC) and Crohn's disease (CD)) has been extensively studied in Greece, especially during the past few years. Quite a large number of studies were published in the form of full papers in the Greek or international literature, or as free presentations at Greek or international congresses.

In this chapter we attempt to critically synthesize the available data in order to reach some conclusions concerning the clinicoepidemiological characteristics and the course of the disease in Greece, based on a large number of patients originating from various geographical areas of Greece.

PATIENTS AND METHODS

Ulcerative colitis

We evaluated a number of clinical and epidemiological parameters (e.g. age, sex, and extent of disease, severity of attacks, surgical treatment and course of the disease – including the development of colorectal cancer, etc.) of 1606 patients with UC, who were hospitalized and followed up in teaching hospitals of the country, as well as data on patients with UC derived from papers published in the international gastroenterology literature.

So collection of data was achieved in two ways. The first was related to direct contact and communication with departments well known for their interest in the study of IBD. Doctors were asked to provide the exact number of IBD patients diagnosed and followed up in their institution, and also to provide data concerning various clinicoepidemiological characteristics, outcome and course

of the disease[1–6]. The second way was referring to the collection and elaboration of data already published in the international and Hellenic medical literature[7–72].

All patients were studied in accordance with a pre-established protocol. The extent of the disease was estimated endoscopically and histologically. Severity was graded in accordance with the criteria of Truelove and Witts.

We evaluated various clinicoepidemiological parameters such as age, sex, severity and extent of disease, duration of follow-up, need for surgical treatment, course of the disease, and colon rectal cancer development. All data related to UC are listed in references 1 to 72.

Crohn's disease

Data concerning CD were derived from relevant publications in the Greek and international literature, abstracts presented in Greek and international congresses, as well as from personal communications. All data related to CD are listed in references 93–145.

RESULTS

Ulcerative colitis (UC)

Epidemiological features

Incidence: The incidence of UC in the prefecture of Heraklion, Crete, was found to be 8.9 cases per 100 000 population[7], while in the area of Epirus (Ioannina) it was found to be 4.8 cases per 100 000 population[8]. The incidence of UC in three other places in Greece was found to be 5.4 cases per 100 000 population (Zakynthos island)[44], 11.2 cases per 100 000 population in Central Greece[45], and six cases per 100 000 population in the island of Evia[46]. The incidence of UC in most parts of the country is generally similar, with the exception of Epirus.

According to recent data the number of new cases of CD diagnosed or hospitalized during the year 2003 in five teaching hospitals of the Athens area, now tends to exceed the number of new cases with UC[72]. Another interesting point of this study was the fact that more newly diagnosed UC patients had extensive colitis. Family history for IBD at the onset of the disease was quite low and, in accordance with data from other countries, most of the patients were urban dwellers of moderate to high socioeconomic level.

Sex: Contrary to other studies from Europe and North America[73,74] an excess risk of men to acquire the disease was noticed in all studies. So in the study of Manousos et al.[7] the ratio of men to women was 1.8:1, while in the study of Triantafillidis et al.[9] the ratio of men to women was 1.14:1. Similar findings were found in all other studies[1–6,24,44,46], with the exception of two (which, however, included a small number of patients) in which no difference was noticed[6,45]. After comparing all the available data the proportion of men to women was 1.40:1.

Genetics – positive family history: In a recently published study a significant increase in the proportion of mutations in the gene MEFV was found, suggesting that this gene is probably involved in the pathogenesis of UC[59]. In another study an absence of immunohistochemical expression of the Fhit protein was noticed both in patients with UC and in patients with colon cancer developed on the ground of UC[60].

In one study a positive family history for IBD was noticed in 5.2%[2], while in the studies by Triantafillidis et al.[9] and Manousos et al.[7] the corresponding figures were 2.42% and 9.6%, respectively. These features are relatively low compared to those reported from other countries, where percentages ranging from 7.9% to 15% (and even higher) have been described[75–77].

Age: In all studies the mean age of patients fluctuated between 40 and 46 years[1–6,9,24,44]. These findings are in accordance with those reported from developed[78,79] and developing[80–83] countries.

Clinical characteristics

Extent of disease: In the study by Ladas et al.[45] referring to a population in Central Greece, 49% had proctitis, 47% left colitis and 4% total colitis. In the study by Archimandritis et al.[24] the disease was located in the left colon in 73.4%. In the same study extensive disease was noticed in 26.6% and total colitis in 15.6%. In the study by Triantafillidis et al.[9] proctitis was found in 26.2%, sigmoiditis in 20.8%, left colitis in 27.4% and extensive or total colitis in 25.6%.

Combining all data the disease was found to be located in the rectum in 24.4%, in the sigmoid colon in 25%, in the left colon in 33.1% and in the whole bowel in 29.9%[1–3,5,6] (Figure 1). These data are similar to those reported from developed[78,79] and developing[81,83] countries.

Severity of disease: The onset of the disease was of mild or moderate severity in the majority of cases. The proportions of mild, moderate and severe course of the disease are shown in Figure 2.

Proximal progression of disease: Few data are available dealing with the proximal progression of UC. In a recently published study among 256 cases, with a follow-up of 7 years, the 10-year cumulative probability of proximal disease extension in patients with proctitis and left colitis was 37% and 17%, respectively[61]. In another study, including 413 cases with a follow-up of 12 years, evolution to proximal disease was noticed in 17%, while evolution to extensive colitis was noticed in 22.5%[9]. According to the existing data the disease is progressing proximally after 5 and 10 years in 16% and 31% of patients respectively[83], reaching the percentage of 51% after 25 years[84].

Extraintestinal manifestations: At least one extraintestinal manifestation was noticed in 19.8% of a total number of 1367 patients[1,2,5,9,36,38,39,62]. Table 1 shows the percentage of extraintestinal manifestations according to the organ or system involved. The proportion of patients with extraintestinal manifestations is in accordance with the majority of descriptions[84].

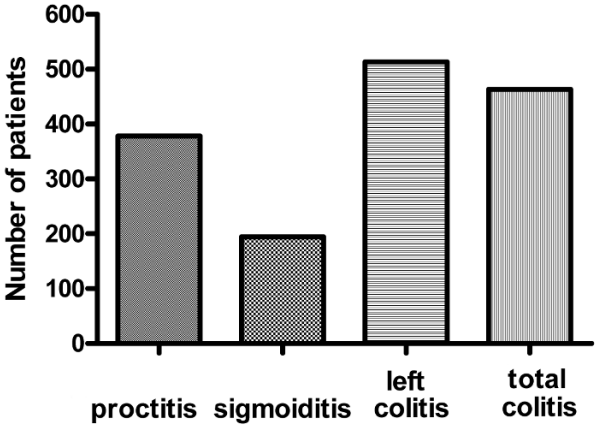

Figure 1 Location of ulcerative colitis

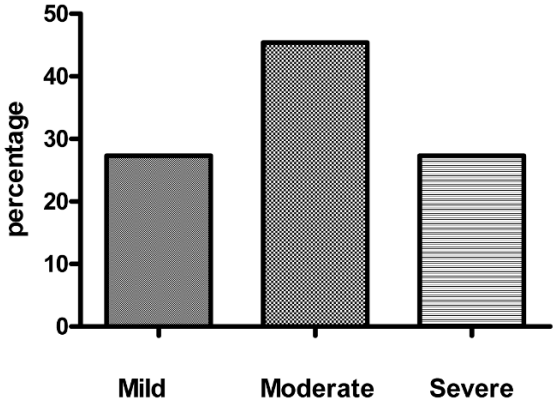

Figure 2 Severity of ulcerative colitis

Oral lesions: In a relevant study dealing with the incidence and type of oral manifestations in UC, 84.6% of patients with UC had at least one oral lesion compared with 13.3% of the normal controls[53].

Helicobacter pylori infection: A low incidence of *H. pylori* infection was also noticed in Greek patients with UC, which could be attributed to previous antibiotic treatment[63].

Table 1 Extraintestinal manifestations in patients with ulcerative colitis

Extraintestinal manifestation	*Number of patients (%)*
Joints	66/705 (9.4)
Renal	30/661 (4.5)
Liver	20/661 (3.0)
Skin disorders	21/725 (2.9)
Ocular lesions	10/661 (1.5)

Seasonal variation: Conflicting data concerning seasonal exacerbation of UC exist[31,41,42].

History of appendectomy and tonsillectomy: Appendectomy and tonsillectomy are not risk factors either increasing or decreasing the risk of development of UC[27].

Constipation: Constipation as a predominant symptom in patients with UC was described in 20.3%[43].

Coexistence with other disorders: Urinary involvement was noticed in many studies. The proportion of urinary stones fluctuated between 3.4%[2] and 12%[37]. A high proportion of allergic and atopic manifestations, as well as psoriasis and autoimmune diseases including Sjögren's syndrome, was also reported[17,37,48].

Combination with autoimmune anaemia[33,48,50], hyperthyroidism[21], varicella infection[15,16], systemic lupus erythematosus[55], Eales' disease[23], sarcoidosis[24,32], primary biliary cirrhosis[26], sponge kidney[64] and valandidiasis[28] was described.

According to recently published data, the cytomegalovirus genome was present in 31% of UC patients[65].

Conservative treatment

Antibiotics in severe attacks of UC: The absence of benefit after administration of tobramycin, metronidazole[68] and ciprofloxacin[69] in patients with a severe attack of UC was noticed in a series of patients with severe exacerbation of UC.

Surgical treatment

During the follow-up period of 7.2 ± 3.5 years, 142 out of 1563 patients with UC (9.1%) were operated-on[1–3,5,13,14,24,44]. This rate of colectomy was compartively low compared to other European or North American countries, in which a rate of 24% and 37.6% has been described[86,87]. Similar to other studies, the main indication for surgical treatment was poor response to conservative treatment. The more common surgical procedure was total colectomy with permanent ileostomy followed by ileorectal anastomosis. However, during recent years more and more patients are submitted to ileoanal–pouch anastomosis. Figure 3 shows the number of patients operated-on in the years following diagnosis of UC[14]

Table 2 Colorectal cancer (CRC) development in patients with ulcerative colitis

Author	*Year*	*UC patients*	*CRC cases*	*Percentage*
Katsaros[40]	1989	144	2	1.4
Triantafillidis[9]	1998	413	4	1.2
Karamanolis[2]	2000	248	3	1.2
Archimandritis[24]	1992	64	1	1.2
Roussomoustakaki[5]	2000	470	2	0.45
Katsanos[71]	2005	215	6	2.8
Total		1554	18	1.2

The extent of the disease is the main factor related to the severity of disease and the need for surgical intervention. In the study by Triantafillidis et al.[9] the rate of colectomy in patients with severe attacks, was 58% which is in accordance with other descriptions[85–89]. In the same study 50% of the operated patients were submitted to operation during the first 5 years after the onset of the disease, a finding similar to other descriptions (Table 2).

Course

The course of the disease was characterized by exacerbations and remissions in the majority of cases. The mortality of patients was described to be 0.9%[5], 3.2%[24], or 6.5%[9], probably reflecting the length of follow-up. Compared to data from other countries a 3.8% reduction in survival 10 years after the onset of the disease, in a study referring to Japanese patients with UC, was noticed[90]. It is of interest that the rate of suicide in Greek male patients with IBD is 10-fold higher compared to that expected in the general Greek population[70].

Colorectal cancer development: Colorectal cancer developed in 18 out of 1554 patients (1.2%). The corresponding studies are shown in Table 2. The incidence of colorectal cancer in UC patients in Greece is probably among the lowest described so far. We have no obvious explanation for that, although the adoption of a Mediterranean diet by a large proportion of the population could be the main reason. However, different behaviour of colorectal cancer in Greece could not be excluded. Another interesting point was the fact that most cancers were located in the rectosigmoid area. This observation makes the surveillance of high-risk patients easier and more convenient for both the endoscopist and the patient.

Other malignant disorders: Development of other malignant disorders was described in many studies; so eight out of 413 patients (2%) developed acute leukaemia, lung cancer, cancer of unknown origin, or brain, liver, and bladder cancer[51]. In another study four out of 470 patients (0.9%) developed malignancies, namely thymoma, as well as lung, gastric and liver cancer[5]. Finally, three out of 181 patients (1.7%) developed cancer of uterus, breast and prostate[24].

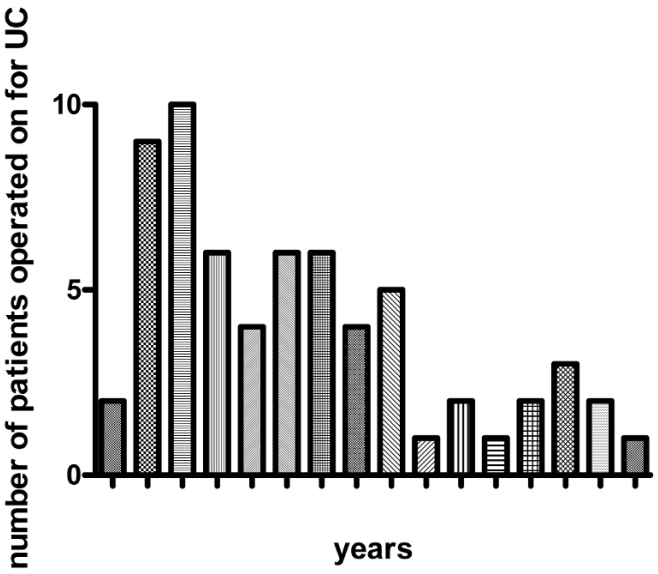

Figure 3 Number of patients operated on for ulcerative colitis in the years following diagnosis

Pregnancy and UC: In a prospective study concerning the course of gestation in Greek women with IBD it was found that IBD is accompanied by unwanted events such as premature delivery, spontaneous abortion, and reduced body weight of the newborn[91].

UC in the elderly: In the only available study[12] it was found that the course of UC was similar to that of young patients, with some exceptions such as the smaller number of operations performed, the absence of cases of colorectal cancer development and the increased mortality compared to young patients, largely due to other causes.

In a study performed in China[92], including 10 218 cases, it was found that the disease is mainly located in the left colon, the age at onset is relatively higher, and a positive family history is rare.

Based on the above-mentioned data, the clinicoepidemiological features and course of UC in Greece could be summarized as follows:

1. The epidemiological features are similar to those of certain European countries. However, an increase in the incidence of CD compared to UC would be expected in the next few years.
2. The proportion of men is greater compared to women.

3. Positive family history is present also in Greek patients with UC, although in a lesser degree compared to other developed countries.
4. Age at onset, severity of attack, and extraintestinal manifestations are similar to those of other developed countries.
5. The number of operations performed seems to be lower compared to those of other countries.
6. The incidence of colorectal cancer development is lower compared to other European and North America countries.
7. Mortality is generally low.

CROHN'S DISEASE

It is well established that the epidemiology, clinical patterns and behaviour of CD differ in various parts of the world[93]. Studies performed in Mediterranean countries such as Italy[94–95], France[96–98], Spain[99–101], former Yugoslavia[102] and Israel[103–105], as well as in countries of northern Europe[106–108], Africa[109–111], Asia[112–114], and North America[115–119], are in favour of this assumption. Moreover, it seems that, even inside countries with a genetically homogeneous population (such as Greece), differences in the incidence of disease in different parts of the country may exist[8,120–122]. As the aetiology of CD is obscure, it is possible that the different disease behaviour in various parts of the world may be related to environmental, genetic or other unknown factors. Some of the clinicoepidemiological data concerning Greek patients with CD are analysed subsequently.

Epidemiology

Similar to UC, a higher proportion of men suffering from CD was noticed in almost all studies. The rate of men to women fluctuated between 1.58:1[93] and 2.4:1[120]. However, in most other studies performed outside Greece, an inverse ratio of male to female has been observed, women being affected more often than men[97,98,112,113], and this is true for other neighbouring countries such as Italy[94,123] and Spain[101], and countries with low incidence of CD (Korea)[112]. Generally, females have been found to have a higher risk compared to males, in the order of 30%.

We have no obvious explanation for the predominance of male sex in Greece, although the small proportion of Greek women who are smokers and users of oral contraceptives could be the most logical explanation.

Age at onset and at diagnosis

In the majority of patients with CD symptoms started around the third decade of life. After the onset of symptoms, 2.4 years (mean value) are required in order to reach a definite diagnosis. In patients with exclusive small bowel involvement this time period is even higher, reaching a level of 3.6 years.

Positive family history

The percentage of familial aggregation in the Greek patients (1.3%) is much lower than that reported in other studies concerning European populations. The proportion of CD patients with a positive family history varies considerably between case-series, but usually falls in the range of 5.5 to 8%[95,124–128].

It is of interest that two families in our series had more than one member affected by CD. In the first family, among a total of seven children, four developed CD, while in the second family, among a total of four children, two developed CD[93]. A familial clustering rate of CD, almost equal to that reported in our series, was reported in a study from Germany[129].

Clinical features

The clinical features of Greek patients with CD show both similarities and differences compared with reports from other countries. So the age at onset of symptoms and the age at diagnosis were similar to those reported from developed and developing countries[94,123]. Most patients had their first attack between 21 and 30 years of age, and this finding is quite similar to that reported from Europe, Asia and North America[108,114,116].

The pattern of anatomical involvement is divided equally between the three main locations of the disease, while in most of the large series reported so far, ileocolonic involvement was seen more frequently[97]. However, in accordance with the above-mentioned data, other studies claim that the percentage of colonic CD is rising steadily[130].

Duration of symptoms

Almost 50% of patients reported that their symptoms started between 1 and 12 months before the establishment of diagnosis. In a comparatively large proportion (20%) of patients the diagnosis was made 5 or more years after the onset of symptoms[93]. No significant differences were observed between the various periods of time preceding the correct diagnosis.

Extraintestinal manifestations

The rate of extraintestinal manifestations fluctuates between 42% and 35%, a percentage quite similar (34.1%) to that reported from other Mediterranean countries, such as Spain[131]. Forty-two per cent of patients with CD developed one or more extraintestinal manifestations during the course of their disease. The most common extraintestinal manifestation was arthritis and/or arthralgias, followed by renal colic and/or nephrolithiasis.

Perianal CD

Of the 155 patients in a relevant study, 33 (21.3%) (23 male and 10 female), had evidence of either old or active perianal CD. Ileocolonic involvement was noticed in 54.5%, colonic involvement alone in 33.5% and ileal involvement

alone in 12% of these patients. Significant differences between patients with and without perianal disease were found only in ileal involvement (12% vs 39.4%, respectively). A significantly higher number of patients with perianal disease had the rectum involved, compared to patients without perianal disease (54.5% vs 16.4%, respectively). Ulceration type lesions were observed in 14 (42.4%), fistulas and/or abscesses in 26 (78.8%) and strictures in six patients (18%), respectively. The rate of perianal disease in Greek patients with CD is among the lowest reported in the relevant literature[132].

Development of colon cancer

The clinical presentation of patients with CD, who developed large bowel carcinoma, was quite severe. These patients developed symptoms and signs of acute abdomen. In two of them the diagnosis of the concurrently existing carcinoma of large bowel and CD was suspected at the operative table and was confirmed histologically on the operative specimen. It seems that the incidence, characteristics and prognosis of colorectal carcinoma complicating CD are similar to the features of cancer complicating UC, including young age and long duration of disease[133,134]. According to a relevant study 191 of the 295 cases (64.8%) of colorectal carcinomas in CD occurred in the past 7 years[135]. This finding leads to the possible conclusion that the risk of developing colorectal cancer in patients with CD has increased in recent years. In some Mediterranean countries (Israel)[136] and countries of North Europe (Sweden)[137], the incidence of colorectal cancer among patients with CD does not differ from that expected in the general population.

Surgical treatment

In a study dealing with surgical treatment of patients with CD, at least one operation was required in 79 patients (51%)[138]. Patients with large bowel involvement had a lower incidence of operations compared to the other two groups, and the difference was statistically significant.

Poor response to conservative treatment and perianal disease were the most frequent indications for surgical intervention, followed by obstructive ileus and erroneous diagnosis of acute appendicitis before diagnosis of CD.

The most frequently performed major surgical procedures were enterectomy and end-to-end anastomosis[138]. Minor surgical procedures were mainly performed for drainage of perianal abscess. The rate of perioperative complications was 14.5%. There were no perioperative deaths[93]. Twenty-one patients (28%) required operation for perineal complication of CD.

The frequency of surgical intervention, indication for surgery and type of operation performed in Greek patients fit well with data reported from North America, Western Europe[139–141] and Mediterranean countries[94,95]. Ileal or ileocaecal involvement was the most common disease pattern encountered, a finding common in almost all other reported series[139,140]. Poor response to medical treatment was the main indication for surgery, followed by perianal fistulas and bowel obstruction. The rate of emergency surgery was higher in elderly people with CD, a finding similar to that reported from other countries.

Although the true incidence of free bowel perforation in patients with CD is difficult to assess, 1–2% is the anticipated occurrence during the course of illness[142]. This percentage is relatively lower compared to that reported in Greek patients (3.2%). The rate of perioperative complications was quite low (14.8%) and almost similar (16%) to that reported elsewhere[139]. No perioperative deaths were noticed. The rate of operation for perianal disease fits well with that of current series (28% vs 25%, respectively)[139].

Outcome

In the majority of patients the disease was running with exacerbations and remissions. In a relevant study[138] 18 patients died during the follow-up period, of whom three belonged to the group of non-operated (4%) and 15 (19%) to the group of operated patients ($p < 0.0076$). The deaths noticed at the end of follow-up were due to CD itself (e.g. colorectal cancer) or to other causes (myocardial infarction, cerebral strokes, etc.). In the same study significantly more deaths were noticed among patients who were operated on compared to the non-operated patients. However, this difference could be biased because: (a) patients who were operated on were followed for a relatively longer period, thus having more chances to die; and (b) patients operated on were quite different from the non-operated group in various aspects of the underlying disease, such as the extent and severity of disease and the presence of complications.

There are no published data in Greece concerning survival of patients with CD. Studies performed in neighbouring countries such as Italy[143] and in Asian countries (Japan)[144], showed a very low disease-specific mortality (around 3% at 10 years and 5% at 20 years). However, other population-based studies from northern Europe[145] showed an increased mortality rate compared with the general population.

Based on the above-mentioned data it can be concluded that certain clinicoepidemiological characteristics of CD in Greece are similar to those reported from the developed countries of Europe, North America and the neighbouring Mediterranean countries. However, other parameters – such as the higher incidence in males, the relatively low incidence of familial clustering and the milder course of perianal disease – all underline the importance of environmental, genetic and other factors in the pathogenesis and course of the disease in different parts of the world.

Acknowledgements

We thank doctors G. Kitis, D. Karamanolis, D. Tzourmakliotis and G. Tambakologos for their permission to use data of their patients with ulcerative colitis.

References

1. Tzourmakliotis D. Clinical experience on 47 patients with ulcerative colitis. Data on file, 2000.
2. Karamanolis D. Clinical experience on 248 patients with ulcerative colitis. Data on file, 2000.
3. Tambakologos G. Clinical experience on 165 patients with ulcerative colitis. Data on file, 2000.
4. Kitis G. Clinical experience on 110 patients with ulcerative colitis. Data on file, 2000.
5. Roussomoustakaki M. Inflammatory bowel disease: experience of the Gastroenterology department of the University of Heraklion, Crete. Data on file, 2000.
6. Paspatis G. Inflammatory bowel disease in Venizelion hospital, Heraklion, Crete. Data on file, 2000.
7. Manousos ON, Giannadaki E, Mouzas IA et al. Ulcerative colitis is as common in Crete as in northern Europe: a 5-year prospective study. Eur J Gastroenterol Hepatol. 1996;8:893–8.
8. Tsianos EV, Masalas CN, Merkouropoulos M, Dalekos GN, Logan RF. Incidence of inflammatory bowel disease in northwest Greece: rarity of CD in an area where ulcerative colitis is common. Gut. 1994;35:369–72.
9. Triantafillidis JK, Emmanouilidis A, Manousos O et al. Ulcerative colitis in Greece: clinicoepidemiological data, course and prognostic factors in 413 consecutive patients. J Clin Gastroenterol. 1998;27:204–10.
10. Emmanouilidis A, Manousos O, Papadimitriou C, Triantafillidis JK. Ulcerative colitis in Greece: course and prognostic factors. Digestion. 1988;39:181–6.
11. Manousos ON, Triantafillidis JK, Emmanouilidis A, Papadimitriou D. Ulcerative colitis in Greece: epidemiologic and clinical data. Scand J Gastroenterol. 1989;24(Suppl. 170):25–6.
12. Triantafillidis JK, Emmanouilidis A, Pomonis E et al. Ulcerative colitis in the elderly: clinicoepidemiological features and follow-up study of 51 Greek patients. Hell J Gastroenterol. 1997;10:236–40.
13. Triantafillidis JK, Manousos O, Emmanouilidis A, Pomonis E, Papadimitriou C. Surgery for ulcerative colitis in Greece. XXVIII World Congress of the International College of Surgeons, Cairo, Egypt 16–21/11/1992. Proceedings, pp. 773–7.
14. Triantafillidis JK, Pomonis E, Emmanouilidis A, Merikas E, Tsitsa C, Hatjiandreou E. Surgery for ulcerative colitis in Greece: clinicoepidemiological features and long-term outcome of 69 consecutive patients. Hepatogastroenterology. 1998;45:961–8.
15. Triantafillidis JK, Cheracakis P, Papalouka E, Barbatzas Ch, Govosdis V. Severe infection by varicella virus in an adult with ulcerative colitis: favorable response to acyclovir treatment. Hell J Gastroenterol. 1995;8:71–3.
16. Mouzas IA, Greenstein AJ, Giannadaki E, Balasubramanian S, Manousos ON, Sachar DB. Management of varicella infection during the course of inflammatory bowel disease. Am J Gastroenterol. 1997;92:1534–7.
17. Triantafillidis JK, Roussou P, Manousos ON, Dadioti P, Nicolakis D. Ulcerative colitis and Sjögren's syndrome in the same patient: report of two cases and a review of the literature. Ital J Gastroenterol. 1994;26:299–302.
18. Triantafillidis JK, Cheracakis P, Zervakakis A, Theodorou M. Coexistence of hyperthyroidism and ulcerative colitis: report of 4 cases and a review of the literature. Ital J Gastroenterol. 1992;24:494–7.
19. Triantafillidis JK, Tsamakidis K, Merikas E, Nikolakis D. Incidence of atopy, breast feeding, severe childhood enteric infections and use of oral contraceptives in Greek patients with inflammatory bowel disease. Hell J Gastroenterol. 1990;3:313–18.
20. Triantafillidis JK, Tsamakidis K. Bourboulis N, Seitanidis B, Siskos P. Acute leukemia following ulcerative colitis. Am J Gastroenterol. 1987;82:1334–6.
21. Triantafillidis JK, Manoussakis K, Tsafaras Ch, Koutsorizof A. Coexistence of thyrotoxicosis and exacerbation of ulcerative colitis. Am J Gastroenterol. 1990;85:908–10.
22. Triantafillidis JK. Coexistence of hyperthyroidism and ulcerative colitis: report of 4 cases and a review of the literature. Ital J Gastroenterol. 1992;24:494–7.
23. Pomonis E, Triantafillidis JK, Tjenaki M, Emmanouilidis A. Report of Eales' disease and ulcerative colitis in the same patient. Am J Gastroenterol. 1992;87:1531–2.
24. Archimandritis A, Tzivras M, Davaris P et al. Inflammatory bowel disease in Greece: a clinical study of 85 consecutive cases. Hepatogastroenterology. 1992;39:99–102.

25. Daskalakis G, Michopoulos S, Akrivos T, Petrogiannis N, Michalas S. Fulminant ulcerative colitis during pregnancy. Hell J Gastroenterol. 1998;11:42–4.
26. Koulentaki M, Koutroubakis IE, Petinaki E et al. Ulcerative colitis associated with primary biliary cirrhosis. Dig Dis Sci. 1999;44:1953–6.
27. Koutroubakis IE, Vlachonikolis IG, Kapsoritakis A et al. Appendectomy, tonsillectomy and risk of inflammatory bowel disease: case–controlled study in Crete. Dis Colon Rectum. 1999;42:225–30.
28. Kamberoglou D, Savva S, Adraskelas N, Yannitsiotis A, Cocca E. Balantidiasis complicating a case of ulcerative colitis. Am J Gastroenterol. 1990;85:765–6.
29. Moscoutis N. Ulcerative colitis with reference to its special characteristics in Greece. Proc R Soc Med. 1969;62:261.
30. Zevgolatis C, Economopoulos P, Sakellaropoulos N. Ulcerative colitis in Greece: a study of 181 cases. Proc R Soc Med. 1969;62:261.
31. Kamberoglou D, Tzitzi D, Protopappa F, Giannitsiotis A. Seasonal variation of ulcerative colitis exacerbations [In Greek with English summary]. Hell Gastroenterol. 1991,4:295–7.
32. Triantafillidis JK, Manousos ON, Bakalos D. Hemorrhagic colitis and sarcoidosis in the same patient. Nosokomiaka Chronika. 1979;41:469–72.
33. Giannadaki E, Potamianos S, Roussomoustakaki M, Kyriakou D, Fragiadakis N, Manousos ON. Autoimmune hemolytic anemia and positive Coomb's test associated with ulcerative colitis. Am J Gastroenterol. 1997;92:1872–4.
34. Michopoulos S, Karambelas D, Pardalos E. History of appendisectomy and tonsillectomy in patients with ulcerative colitis. Hell J Gastroenterol (Suppl.). 1995;8:106.
35. Mouzas I, Skordilis P, Giannadaki E. Appendectomy protects against the development of ulcerative colitis. Hell J Gatsroenterol (Suppl.). 1995;8:109.
36. Kazouris D, Potamianos S, Tzathas X. Incidence of extraintestinal manifestations in Greek patients with inflammatory bowel disease. Hell Gastroenterol. 1989,4:EA15.
37. Triantafillidis JK, Emmanouilidis A, Manousos ON. Extraintestinal manifestations of 568 Greek patients with inflammatory bowel disease. Hell J Gastroenterol (Suppl.). 1993,6:262.
38. Pomonis E, Nikolakis D, Cherakakis P. Arthritis in Greek patients with inflammatory bowel disease. Hell J Gastroenterol (Suppl.). 1995;8:112.
39. Mouzas I, Kritikos H, Karambekios S. Sacrocoxitis in patients with inflammatory bowel disease. Hell J Gastroenterol (Suppl.). 1997;10:172.
40. Katsaros D, Barbati K, Mbarbatzas X. Ulcerative colitis and cancer. Hell Gastroenterol. 1989;4:149.
41. Karamanolis DG, Delis KC, Papatheodoridis GV, Kalafatis E, Paspatis G, Xourgias VC. Seasonal variation in exacerbations of ulcerative colitis. Hepatogastroenterology. 1997;44: 1334–8.
42. Karkotis K, Logiou M, Soutos D, Katsaros D. Seasonal variation of ulcerative colitis. Hell Gastroenterol. 1990;5:95.
43. Nasalas K, Dalekos G, Mercouropoulos N, Tsianos E. Constipation and ulcerative colitis. Hell Gastroenterol. 1991;4:82
44. Papadatou A, Ntikas E, Kalogeropoulos N, Kapsabelis P. Prevalence of inflammatory bowel disease in the island of Zakinthos. Hell J Gastroenterol (Suppl.). 1991;6:257.
45. Ladas SD, Mallas E, Giorgiotis K et al. Incidence of ulcerative colitis in central Greece: a prospective study. World J Gastroenterol. 2005;11:1785–7.
46. Kouvaras S, Xaliotis G, Diakos A. Prevalence of inflammatory bowel disease in Evia Island. Hell J Gastroenterol (Suppl.). 1995;8:110.
47. Dimou V, Nikolakis D, Pomonis E. Diseases associated with inflammatory bowel disease in Greece. Hell J Gastroenterol (Suppl.). 1993;6:261.
48. Mantzaris G, Hatzis A, Kamati N. Autoimmune disorders in patients with inflammatory bowel disease. Hell J Gastroenterol (Suppl.). 1993;6:264.
49. Emmanouilidis A, Triantafillidis JK, Manousos ON. Familial aggregation of ulcerative colitis in Greece. Hell J Gastroenterol (Suppl.). 1993;6:266.
50. Paraskevas E, Salametis N, Sabbas S. Autoimmune hemolytic anemia in ulcerative colitis. Hell J Gastroenterol (Suppl.). 1993,6:270.
51. Triantafillidis JK, Emmanouilidis A, Manousos ON. Colorectal cancer in inflammatory bowel disease. Hell J Gastroenterol (Suppl.). 1993;6:283.
52. Mantzaris G, Hatzis A, Zografos X. Treatment of resistant forms of ulcerative colitis with azathioprine. Hell J Gastroenterol (Suppl.). 1993;6:286.

53. Zervou-Valvi F, Triantafillidis JK, Karakatza I et al. Oral lesions in patients with inflammatory bowel disease. Fourth Mediterranenan Medical Congress, Limassol, Cyprus, 13-17 November 1996, Abstract book no. 056.
54. Maris Th, Heliadis M, Kokozidis G. Cyclosporin in severe ulcerative colitis. Hell J Gastroenterol (Suppl.). 1999;12:A162.
55. Koutroumbakis IE, Kritikos H, Mouzas IA et al. Association between ulcerative colitis and systemic lupus erythematosus: report of two cases. Eur J Gastroenterol Hepatol. 1998;10: 437–9.
56. Papadimitriou N, Koumentakis N, Mpakiri M. Acute leukemia in patient with ulcerative colitis. Hell J Gastroenterol (Suppl.). 1993;6:281.
57. Mantzaris G, Xatzis A, Zavos K. Ulcerative colitis and gestation. Hell J Gastroenterol (Suppl.). 1993;6:265.
58. Giannadaki E, Mouzas I, Foukakis Th. Proximal progression of ulcerative colitis patients. Hell J Gastroenterol (Suppl.). 1997;10:174.
59. Giaglis S, Mimidis K, Papadopoulos V et al. Increased frequency of mutations in the gene responsible for familial Mediterranean fever in a cohort of patients with ulcerative colitis: evidence for a potential disease-modifying effect? Dig Dis Sci. 2006;51:687–92.
60. Skopeliotu AS, Katsanos KH, Michail M, Mitselou A, Tsianos EV. Immunohistochemical expression of FHIT gene product in inflammatory bowel disease: significance and correlation with histopathological data. Eur J Gastroenterol Hepatol. 2003;15:665–73.
61. Chatzikosta C, Roussomoustakaki M, Potamianos S et al. Factors associated with disease evolution in Greek patients with inflammatory bowel disease. BMC Gastroenterol. 2006; 6:21.
62. Christodoulou DK, Katsanos KH, Kitsanou M, Stergiopoulou C, Hatzis J, Tsianos EV. Frequency of extraintestinal manifestations in patients with inflammatory bowel disease in northwest Greece and review of the literature. Dig Liver Dis. 2002;34:781–6.
63. Triantafillidis JK, Gikas A, Apostolides N, Merikas E, Mallas E, Peros G. The low prevalence of *Helicobacter* infection in patients with inflammatory bowel disease could be attributed to previous antibiotic treatment. Am J Gastroenterol. 2003;98:1213–14.
64. Triantafillidis JK, Iatrou Ch, Hatzivasiliou A, Kalogerakos S. Medullary sponge kidney and ulcerative colitis in the same patient: an extremely rare combination. Ann Gastroenterol. 2004;17:210–12.
65. Dimitroulia E, Spanakis N, Konstandinidou AE, Legakis NJ, Tsakris A. Frequent detection of cytomegalovirus in the intestine of patients with inflammatory bowel disease. Inflamm Bowel Dis. 2006;12:879–84.
66. Mantzaris GJ, Sfakianakis M, Archavlis E et al. A prospective randomized observer-blind 2-year trial of azathioprine monotherapy versus azathioprine and olsalazine for the maintenance of remission of steroid-dependent ulcerative colitis. Am J Gastroenterol. 2004;99:1122–28.
67. Christodoulou D, Katsanos K, Baltayannis G, Tzavouras N, Tsianos EV. A report on efficacy and safety of azathioprine in a group of inflammatory bowel disease patients in northwest Greece. Hepatogastroenterology. 2003;50:1021–4.
68. Mantzaris GJ, Hatzis A, Kontogiannis P, Triantaphyllou G. Intravenous tobramycin as an adjunct to corticosteroids in acute severe ulcerative colitis. Am J Gastroenterol. 1994;89: 43–6.
69. Mantzaris GJ, Petraki K, Archavlis E et al. A prospective randomized controlled trial of intravenous ciprofloxacin as an adjunct to corticosteroids in acute severe ulcerative colitis. Scand J Gastroenterol. 2001;36:971–4.
70. Triantafillidis JK, Hyphantis T, Cheracakis P, Antoniou A, Sklavaina M. Suicide as an outcome in patients with inflammatory bowel disease. Am J Gastroenterol. 2002;97:1066–8.
71. Katsanos KH, Christodoulou DK, Michael M et al. Inflammatory bowel disease-related dysplasia and cancer: a referral center study in northwestern Greece. Eur J Intern Med. 2005;16:170–5.
72. Triantafillidis JK, Kitis G, Karamanolis D, Karagiannis I, Tzourmakliotis D. Clinicoepidemiological characteristics of adult patients with inflammatory bowel disease diagnosed in 5 Greek teaching hospitals during the year 2003. Ann Gastroenterol. 2005; 18:68–72.

73. Sinclair TS, Brunt PW, Mowat NAG. Nonspecific proctocolitis in northwest Scotland: a community study. Gastroenterology. 1983;85:1–11.
74. Stowe SP, Redmond SR, Stormond JM et al. An epidemiologic study of inflammatory bowel disease in Rochester, New York; hospital incidence. Gastroenterology. 1990;98:104–10.
75. Ekbom A, Helmick C, Zack M, Adami HO. The epidemiology of inflammatory bowel disease: a large population-based study in Sweden. Gastroenterology. 1991;100:350–8.
76. Monsen U, Bronstrom O, Nordenvall B, Sorstad J, Hellers G. Prevalence of inflammatory bowel disease among relatives of patients with ulcerative colitis. Scand J Gastroenterol. 1987;22:214–18.
77. McConnell RB, Vadheim CM. Inflammatory bowel disease. In: King RA, Rotter JI, Motulsky AG, editors. The Genetic Basis of Common Diseases. New York: Oxford University Press, 1992:326–48.
78. Farmer RG. Study of family history among relatives of patients with inflammatory bowel disease. Scand J Gastroenterol. 1989,24(Suppl. 170):64–5.
79. Langholz E, Munkholm P, Davidson M, Binder V. Course of ulcerative colitis: analysis of change of disease activity over years. Gastroenterology. 1994;107:3–11.
80. Farmer RG, Easley KA, Rankin GB. Clinical patterns, natural history and progression of ulcerative colitis. A long-term follow-up of 1116 patients. Dig Dis Sci. 1993;38:1137–46.
81. Kusakcioglou O, Kusakcioglou A, Feriha OZ. Idiopathic ulcerative colitis in Instanbul: clinical review of 204 patients. Dis Colon Rectum. 1979;22:350–5.
82. Mir-Madzlessi SH, Forouzandeh B, Ghadimi R. Ulcerative colitis in Iran: a review of 112 cases. Am J Gastroenterol. 1985;80:862–6.
83. Vucelic B, Korac B, Sentic M et al. Ulcerative colitis in Zagreb, Yugoslavia: incidence and prevalence 1980–1989. Int J Epidemiol. 1991;20:1043–7.
84. Duphare H, Misra SC, Patnaik PK, Mathur M, Tandon RK. Spectrum of ulcerative colitis in India. J Clin Gastroenterol. 1994;18:23–6.
85. Monsen U, Sorstad J, Hellers G, Johanson C. Extracolonic diagnoses of ulcerative colitis: an epidemiological study. Am J Gastroenterol. 1990;85:711–16.
86. Ahsgren L, Jonsson B, Stenling R, Rutegard J. Prognosis after early onset of ulcerative colitis. A study of unselected patient population. Hepatogastroenterology. 1993;40:467–70.
87. Ayres RC, Gillen CD, Walmsley RS, Allan RN. Progression of ulcerative proctosigmoitis: incidence and factors influencing progression. Eur J Gastroenterol Hepatol. 1996;8:555–8.
88. Langholz E, Munholm P, Davidsen M, Nielsen OH, Binder V. Changes in extent of ulcerative colitis: a study on the course and prognostic factors. Scand J Gastroenterol. 1996;31:260–6.
89. Jarnerot G, Rolny P, Sandberg-Gertzen H. Intensive intravenous treatment of ulcerative colitis. Gastroenterology. 1985;89:1005–13.
90. Hiwatashi N, Yao T, Watanabe H et al. Long-term follow-up study of ulcerative colitis in Japan. J Gastroenterol. 1995;30(Suppl. 8):13–16.
91. Triantafillidis JK, Malgarinos G, Gikas A et al. Pregnancy and inflammatory bowel disease in Greece: a prospective study of seven cases in a single hospital setting. Ann Gastroenterol. 2007 (In press).
92. Xue-Liang J, Hui-Fei C. An analysis of 10218 ulcerative colitis cases in China. World J Gastroenterol. 2002;8:158–61.
93. Triantafillidis JK, Emmanouilidis A, Manousos O, Nikolakis D, Kogevinas M. Clinical patterns of Crohn's disease in Greece: a follow-up study of 155 cases. Digestion. 2000;61: 121–8
94. Corrao G, Tragnone A, Kaprilli R et al. Risk of inflammatory bowel disease attributable to smoking, oral contraception and breast feeding in Italy: a nationwide case–control study. Cooperative investigators of the Italian Group for the study of the colon and rectum. Int J Epidemiol. 1998,27:397–404.
95. Vannozzi G, Fontana R, Milla M et al. Disease history in 382 Italian patients with Crohn's disease. Ital J Gastroenterol Hepatol. 1997;29:525–32.
96. Latour P, Louis E, Belaiche J. Incidence of inflammatory bowel disease in the area of Liege: a 3 years prospective study (1993–1996). Acta Gastroenterol Belg. 1998;61:410–13.
97. Beaugerie L, Lamy P, Ganne N et al. Morbid associations in Crohn's disease. Study of a series of 832 patients. Presse Med. 1997;26:892–4.

98. Gower-Rousseau C, Salomez JL, Dupas JL et al. Incidence of inflammatory bowel disease in northern France (1988–1990) Gut. 1994;35:1433–8.
99. Mate-Jimenez J, Munoz S, Vicent D, Pajares JM. Incidence and prevalence of ulcerative colitis and Crohn's disease in urban and rural areas of Spain from 1981 to 1988. J Clin Gastroenterol. 1994;18:27–31.
100. Martinez-Salmeron JF, Rodrigo M, de Teresa J et al. Epidemiology of inflammatory bowel disease in the province of Granada, Spain: a retrospective study from 1979 to 1988. Gut. 1993;34:1207–9.
101. Ruiz V. Crohn's disease in Galicia, Spain. Scand J Gastroenterol Suppl. 1989;170:29–31.
102. Vucelic B, Korac B, Sentic M et al. Epidemiology of Crohn's disease in Zagreb, Yugoslavia: a ten-year prospective study. Int J Epidemiol. 1991;20:216–20.
103. Odes HS, Locker C, Neumann L et al. Epidemiology of Crohn's disease in Southern Israel. Am J Gastroenterol. 1994;89:1859–62.
104. Odes HS, Fraser D, Krugliak P, Fenyves D, Fraser GM, Sperber AD. Inflammatory bowel disease in the Beduin Arabs of southern Israel: rarity of diagnosis and clinical features. Gut. 1991;32:1024–6.
105. Odes HS, Fraser D, Krawiec J. Inflammatory bowel disease in migrant and native Jewish population of southern Israel. Scand J Gastroenterol (Suppl.). 1989;170:36–8.
106. Shivananda S, Lennard-Jones J, Logan R et al. Incidence of inflammatory bowel disease across Europe: is there a difference between north and south? Results of the European Collaborative Study on Inflammatory Bowel Disease (EC-IBD). Gut. 1996;39:690–0.
107. Steinhardt HJ, Loeschke K, Kasper H, Holtermuller KH, Schafer H. European cooperative Crohn's disease study (ECCDS): clinical features and natural history. Digestion. 1985;31: 97–108.
108. Fielding JF. Clinical features of Crohn's disease in Ireland. Am J Gastroenterol. 1986;81: 524–8.
109. Binder V, Hendriksen C, Kreiner S. Prognosis of Crohn's disease – based on the results from a regional patient group from the county of Copenhagen. Gut. 1985;26:146–50.
110. Kuruvilla MJ, Chhallani CR, Rajagopal AK, Salem-Rakas F. Major causes of intestinal obstruction in Libya. Br J Surg. 1987;74:314–15.
111. Al-Nakib B, Radhakrishnan S, Jacob GS, Al-Liddawi H, Al-Ruwaih A. Inflammatory bowel disease in Kuwait. Am J Gastroenterol. 1984;79:191–4.
112. O'Keefe EA, Wright JP, Froggatt J, Zabow D. Medium-term follow-up of Crohn's disease in Cape Town. S Afr Med J. 1989;76:139–41.
113. Mengesha B, Johnson O, Taye M, Gemetchu T. Crohn's disease: report of seven cases from Ethiopia. East Afr Med J. 1997;74:397–9.
114. Yoon CM, Kim SB, Park IJ et al. Clinical features of Crohn's disease in Korea. Gastroenterol Jpn. 1988;23:576–81.
115. Teh LB, Ng HS, Ho MS, Seah CS. Crohn's disease. a diagnostic rarity in Singapore. Ann Acad Med Singapore. 1987;16:480–7.
116. Okada M, Sakurai T, Yao T, Iida M, Okuda N, Maeda K. Clinical course and long-term prognosis of Crohn's disease in Japan. J Gastroenterol. 1994;29:406–14.
117. Simsek H, Schuman BM. Inflammatory bowel disease in 64 black patients: analysis of course, complications and surgery. J Clin Gastroenterol. 1989;11:294–8.
118. Farmer RG, Whelan G, Fazio VW. Long-term follow-up of patients with Crohn's disease: relationship between the clinical patterns and prognosis. Gastroenterology. 1985;88:1818–25.
119. Meyers S, Janowitz HD. Natural history of Crohn's disease: an analytical review of the placebo lesson. Gastroenterology. 1984;87:1189–92.
120. Mekhjian HS, Switz DM, Melnyk CS, Rankin GB, Brooks RK. Clinical features and natural history of Crohn's disease (National Cooperative Crohn's disease study). Gastroenterology. 1979;77:898–906.
121. Harper PH, Fazio VW, Lavery IC et al. The long-term outcome of Crohn's disease. Dis Colon Rectum. 1987;30:174–9.
122. Manousos ON, Koutroumbakis I, Potamianos S, Roussomoustakaki M, Gourtsoyiannis N, Vlachonikolis IG. A prospective epidemiologic study of Crohn's disease in Heraklion, Crete. Incidence over a 5-year period. Scand J Gastroenterol. 1996;31:599–603.

123. Cottone M, Cipolla C, Orlando A, Oliva L, Aiala R, Puleo A. Epidemiology of Crohn's disease in Sicily: a hospital incidence study from 1987 to 1989. The Sicilian Study Group of Inflammatory Bowel Disease. Eur J Epidemiol. 1991;7:636–40.
124. Gollop JH, Phillips SF, Melton LJ, Zinmeister AR. Epidemiologic aspects of Crohn's disease: a population based study in Plimsted County, Minnesota, 1943–1982. Gut. 1988; 29:49–56.
125. Ekbom A, Helmick C, Zack M, Adami HO. The epidemiology of inflammatory bowel disease: a large, population based study in Sweden. Gastroenterology. 1991;100:350–8.
126. Binder V, Orholm M. Familial occurence and inheritance studies in inflammatory bowel disease. Neth J Med. 1996;48:53–6.
127. Zlotogora J, Zimmerman J, Rachmilewitz D. Prevalence of inflammatory bowel disease in family members of Jewish Crohn's disease patients in Israel. Dig Dis Sci. 1991;36:471–5.
128. Grandbastien B, Peeters M, Franchimont D et al. Anticipation in familial Crohn's disease. Gut. 1998;42:170–4.
129. Stetter M, Schuster E, Knoflach P. Langzeitverlauf des morbus Crohn. Z Gastroenterol. 1992;30:454–8.
130. Lapidus A, Bernell O, Hellers G, Loflerg R. Clinical course of colorectal Crohn's disease: a 35-year follow-up study of 507 patients. Gastroenterology. 1998;114:1151–60.
131. Urighen CM, Losana P Jr, Suarez A, Gonzalez MA. The extraintestinal manifestations of Crohn's disease: our experience. Rev Esp Enferm Dig. 1991;80:169–72.
132. O'Donoghue DP, Hyland JMP. Perianal Crohn's disease. Eur J Gastroenterol Hepatol. 1997;9:235–36.
133. Savoca PE, Ballantyne GH, Cahow CE. Gastrointestinal malignancies in Crohn's disease: a 20-year experience. Dis Colon Rectum. 1990;33:7–11.
134. Ribeiro MB, Greenstein AJ, Sachar DB et al. Colorectal carcinoma in Crohn's disease. Ann Surg. 1996;223:186–93.
135. Rubio CA, Befrits R. Colorectal adenocarcinoma in Crohn's disease: a retrospective histologic study. Dis Colon Rectum. 1997;40:1072–8.
136. Fireman Z, Grossman A, Lilos P et al. Intestinal cancer in patients with Crohn's disease. A population study in Central Israel. Scand J Gastroenterol. 1989;24:346–50.
137. Persson PG, Karlen P, Bernell O et al. Crohn's disease and cancer: a population-based cohort study. Gastroenterology. 1994;107:1675–9.
138. Triantafillidis JK, Emmanouilidis A, Nokolakis D. Surgery for Crohn's disease in Greece: a follow-up study of 79 cases. Hepatogastroenterology. 2001;48:1072–7.
139. Hurst RD, Molinari M, Chung TP, Rubin M, Michelassi F. Prospective study of the features, indications, and surgical treatment in 513 consecutive patients affected by Crohn's disease. Surgery. 1997;122:661–7.
140. Fazio VW, Wu JS. Surgical therapy for Crohn's disease of the colon and rectum. Surg Clin N Am. 1997;77:197–210.
141. Pastore RL, Wolff BG, Hodge D. Total abdominal colectomy and ileorectal anastomosis for inflammatory bowel disease. Dis Colon Rectum. 1997;40:1455–64.
142. Roy MK, Delicata RJ, Carey PD. Crohn's disease presenting with acute colonic perforation. Postgrad Med J. 1997;73:645–6.
143. Vannozzi G, Fontana R, Milla M et al. Disease history in 382 Italian patients with Crohn's disease. Ital J Gastroenterol Hepatol. 1997;29:525–32.
144. Iida M, Yao T, Okada M. Long-term follow-up study of Crohn's disease in Japan. R esearch committee of inflammatory bowel disease in Japan. J Gastroenterol. 1995;30 (Suppl. 8):17–19.
145. Persson PG, Bernell O, Leijonmarck CE, Farahmand BY, Hellers G, Ahlbom A. Survival and cause-specific mortality in inflammatory bowel disease: a population-based cohort study. Gastroenterology. 1996;110:1339–45.

3
Inflammatory bowel disease in southern Europe

B. VUCELIC and S. CUKOVIC-CAVKA

INTRODUCTION

Crohn's disease (CD) and ulcerative colitis (UC), two major forms of idiopathic inflammatory bowel disease (IBD), are precipitated by a complex interaction of genetic, environmental and immunoregulatory factors. They share many epidemiological and clinical characteristics. IBD is traditionally considered to be common in the Western world, with a sharp increase of incidence since the early 1950s. In contrast, until the past decade, low incidence and prevalence rates have been reported from other parts of the world including eastern Europe, South America, Asia and the Pacific region. Within Europe a north–south gradient of IBD incidence was observed until the 1990s, but with some exceptions, notably the island of Crete[1]. Recent trends indicate a change in the epidemiology of IBD[2]. Previously low-incidence areas are now reporting a progressive rise in incidence. In contrast, the incidence remains stable or slightly increased in western Europe and North America, with decreasing incidence rates for UC[2]. The important observation is an increasing incidence of IBD in younger age groups, with childhood IBD accounting for nearly 30% in some studies[3]. Among childhood IBD, early-onset IBD is characterized by a predominance of colonic involvement, more extensive disease and a high positive family history. The question to be answered is whether these changes are real or are results of better diagnostic practices and increased awareness of the disease. The available epidemiological studies should be scrutinized in order to detect possible flaws in methodology and accuracy due to well-known problems in the epidemiological studies of IBD: (1) lack of diagnostic gold standard criteria and complexity and expense of diagnostic work-up causing problems in reporting incidence of these diseases, particularly in less developed countries; (2) most epidemiological studies come from the large referral centres and may therefore be biased towards reporting more advanced forms of disease, underestimating the true incidence; (3) frequent misclassification of IBD phenotypes might be due either to the true natural course of disease (reassignment of a diagnosis may be as high as 10% in the first 2 years after diagnosis) or due to methodological

problems; (4) differences in health systems in various countries. However, in spite of these methodological limitations, data indicate the distinct temporal and geographic trends mentioned earlier. It is of interest that the pattern of a sharp rise in the incidence of CD observed from the mid-1950s to the early 1970s followed by stabilization of the rate since 1980s in developed countries of northern Europe is now repeating itself with time delay in southern Europe. The same holds true for UC. The commonly discussed north–south gradient is increasingly disappearing in Europe due to the stabilization of incidence rates in the north and increasing rates in the south.

Genetic studies in IBD are becoming more important every day, not only because of the opportunity to clarify the pathophysiology of the inflammatory process, but also due to the fact that IBD occurrence is shifting more and more towards younger age groups with significantly lower influence of environmental factors. Most available data concern the NOD2/CARD15 gene, an important IBD susceptibility gene identified among several mapped loci. Three single-nucleotide polymorphisms (SNP) (R702W, G908R and L1007fs) have shown significant association with CD, but not with UC, in different Caucasian populations. Polymorphisms in the carnitine organic cation transporter cluster (OCTN1/2) on chromosome 5q31 and mutations in disc large gene 5 (DLG5) on the long arm of chromosome 10 (10q23) have also been reported to be associated with CD.

The aim of this chapter is to review the epidemiological and genetic data from southern Europe, particularly from four countries – Croatia, Greece, Italy and Spain – in view of general observations mentioned earlier.

IBD IN CROATIA

The available epidemiological data on IBD in Croatia are presented in Tables 1 and 2. First epidemiological data on IBD were published in 1991, when IBD Group Zagreb reported results of a 10-year prospective population-based study (1980–1989) on UC and CD done in Zagreb County (population 1 175 000). The study on UC revealed the mean annual incidence rate of 1.5 per 100 000

Table 1 Incidence rates of ulcerative colitis in Croatia

Zagreb	0.9	1980
	2.0	1989
Rijeka	2.7	1995
	4.6	2000–2004

Table 2 Incidence rates of Crohn's disease in Croatia

Zagreb	0.4	1980
	1.3	1989
Rijeka	3.5	1994
	6.5	2000–2004

with an incidence rate of 0.9 at the beginning of the study and 2.0 in the last year of the study[4]. The prevalence rate estimated per 100 000 inhabitants, based on the July 1985 official census, was 21.4. Data on smoking habits showed higher smoking rates among the general population (51.4%) than among IBD patients. Among UC patients there were 11.3% smokers and 19.2% ex-smokers. The study on CD showed a mean annual incidence rate of 0.7 per 100 000 with an incidence rate of 0.4 in 1980 and 2.0 in 1989[5]. Regarding smoking, there were 35.8% smokers and 13.2% ex-smokers among the CD patient group, a lower figure than in the general population but higher than in the UC patient group. These two studies showed lower incidence rates of IBD in Zagreb compared with northern and western Europe at the time.

The trend of an increasing incidence of IBD in Croatia was recognized in the preliminary studies from the Rijeka County, showing an incidence rate of 2.7 for UC in 1995 and an incidence rate of 3.5 for CD in 1994. A recent prospective 5-year (2000–2004) population-based study from Rijeka County (305 505 inhabitants) revealed mean annual crude incidence rates of 4.6 for UC and of 6.5 for CD[6]. These data show that the incidence of CD in Croatia is now comparable with the CD incidence in northern Europe, indicating a loss of the north–south gradient.

The study on NOD2/CARD15 mutations in Croatian patients with CD was reported in 2006[7]. That study demonstrated that mutations in this gene are implicated in susceptibility to CD in the Croatian population. Phenotypic association showed a younger age at diagnosis and a higher need for surgery in patients carrying NOD2/CARD15 mutations. However, the prevalence is somewhat lower compared to other reports, probably due to a more prominent colonic inflammation.

IBD IN GREECE

The epidemiological data on IBD in Greece generated from the population-based studies are presented in Tables 3 and 4. Two areas of Greece were part of the EC-IBD study, Ioannina in the north and Heraklion in the south of the country[1]. The data dispute the concept of a north–south gradient, particularly due to the high incidence rates in Heraklion (island of Crete)[8]. It is also of interest to report the data from the prospective population-based study (1990–1994) on UC from the semirural prefecture of Trikala in central Greece (population 139 000)[9]. This reported the mean incidence rate of 11.2 per 100 000 inhabitants, quite comparable to the incidence in northern Europe.

The initial data on genetics in IBD came from Greece 10 years ago. Papasteriades and co-workers found associations between HLA genes (HLA-A, B) and UC. A severe course of the disease was associated with HLA-B13, whereas extensive colitis was associated with HLA-A11 and B7[10]. In 2004 the Molecular Carcinogenesis Group from the University of Athens confirmed that the NOD2/CARD15 mutations were risk factors for CD in Greece. They also found some genotype–phenotype associations between NOD2 polymorphisms and earlier age of onset and ileal location of disease[11]. Data regarding the association between polymorphisms in the Toll-like receptor 4, CD14, and

Table 3 Incidence of ulcerative colitis in Greece

Ioannina	4.0	1982–1991
Central Greece	11.2	1990–1994
Heraklion	9.4	1990–1994

Table 4 Incidence of Crohn's disease in Greece

Ioannina	0.3	1982–1991
	0.9	1991–1993
Heraklion	1.9	1990
	3.8	1994

NOD2/CARD15 and IBD in the Greek population indicated that coexistence of a mutation in either the TLR4 or CD14 gene, and in NOD2/CARD15, was associated with an increased susceptibility to CD compared to UC, and to the development of either CD or UC compared to healthy controls[12]. The study on the SNP of OCTN1, OCTN2, and DLG5 genes in Greek patients with CD suggested that the 1672T variant of the OCTN1 gene and the –207C variant of the OCTN2 gene represent risk factors for CD in the Greek population[13].

IBD IN ITALY

Italy has always been a very interesting area for epidemiological studies of IBD due to its diversity, with highly industrialized areas in the north belonging to the 'classical' western Europe in epidemiological terms, and with the more rural southern part belonging to Mediterranean Europe. As indicated in Tables 5 and 6, the majority of data are from the northern part of Italy, confirming the general epidemiological pattern described in the introductory part of this chapter. The review paper by Cottone et al. showed that the incidence of IBD is similar throughout Italy, with incidence rates for UC ranging from 3.4 to 10.5 and for CD from 1.9 to 6.6[14]. Incidence rates for IBD are therefore very similar to those of northern Europe. The time trends indicate the increase of incidence rates in both diseases.

Research on the genetics of IBD has been very active in Italy. A research group from Torino confirmed the association of the CARD15 genotype with behaviour and location of CD in an Italian population[15]. An IG-IBD study showed that Italian patients with familial or sporadic CD carrying at least one major variant of CARD15 had an aggressive course of the disease. This group did not find any difference in SNP frequency between familial and sporadic cases[16,17]. Recently, in a large cohort of Italian paediatric patients with IBD, it was confirmed that polymorphisms of CARD15 were significantly associated with CD, but no association with the DLG5 variant was found. Also, homozygosis for both OCTN1/2 variants was more common in CD patients,

Table 5 Incidence of ulcerative colitis in Italy

Florence	4.0	1978–1987
	9.6	1990–1992
Bologna	5.0	1986–1989
Lombardia	7.0	1990–1994
Italy	3.4–10.5	1999

Table 6 Incidence of Crohn's disease in Italy

Florence	1.5	1978–1987
	3.4	1990–1992
Bologna	2.7	1986–1989
Lombardia	3.4	1990–1994
Palermo	2.7	1987–1989
Italy	1.9–6.6	1999

which associated OCTN polymorphisms with a paediatric onset of CD. Regarding genotype–phenotype correlations, that study showed that CARD15 variants were slightly more frequent in patients with ileal disease, while OCTN and DLG5 polymorphisms were associated with the penetrating type of the disease[18]. However, a major role in adult CD in Italy has been confirmed for three SNP in the CARD15 gene[19,20]. In the Ferraris et al. study polymorphisms in the SLC22A4, SLC22A5, and DLG5 genes might have a minor contribution to IBD susceptibility[19]. The same study confirmed that the major CARD15 polymorphisms were associated with early-onset CD and with ileal disease, also suggesting an interesting association with very early-onset UC[19].

IBD IN SPAIN

There is a plethora of data on the epidemiology of IBD in Spain, as presented in Tables 7 and 8. The data represent the whole spectrum, from large urban areas to rural areas, from the north to the south of the country. Information on incidence rates in two large cities, Madrid and Barcelona, was obtained in the 1980s, showing lower incidence rates than in the corresponding large cities in northern Europe at the time. The representative data from smaller areas of Spain, obtained more recently, show trends detected in other areas of southern Europe. The study from Pamplona area conducted during the period from1983 to 1993 reported a mean incidence of 3.8 for UC and of 2.5 for CD[21]. The weaknesses of the study were its retrospective nature and the inclusion of only hospitalized patients. Another retrospective study was conducted in Castellon (Valencia) in the period 1992–1996; it showed an incidence rate for UC of 6.8 and for CD of 1.9[22]. A study on CD from Asturias performed in the 1970s shows a significantly lower incidence rate than the recent prospective

Table 7 Incidence of ulcerative colitis in Spain

Barcelona	0.6	1978–1987
Madrid	2.4	1983–1988
Pamplona	3.8	1983–1993
Castellon	6.8	1992–1996
Oviedo	9.1	2001–2003
Huelva	5.2	1996–2003
Spain	3.8	(0.6–8.0) until January 2000

Table 8 Incidence of Crohn's disease in Spain

Barcelona	0.4	1978–1987
Madrid	1.3	1983–1988
Pamplona	2.5	1983–1993
Castellon	1.9	1992–1996
Asturias	0.8	1975–1979
Oviedo	7.5	2001–2003
Huelva	6.6	1996–2003
Spain	1.9	(0.4–5.5) until January 2000

population-based study from Oviedo[23]. An interesting study comes from the province of Huelva. The authors carried out a long-term study, with the initial phase (1980–1996) being retrospective and the final phase (1996–2003) being prospective. The area studied is relatively small (78 000 inhabitants). The mean incidence rate for UC was 5.2 during the prospective part of the study. The mean incidence rate for CD was 6.6 during the prospective phase of the study[24]. The review article on epidemiology of IBD in Spain comments that both UC and CD have shown important changes of incidence during recent decades[25]. The reported incidence rates for UC in Spain range from 0.6 to 8, with a mean value of 3.8. Reported incidence rates for CD in Spain range from 0.4 to 5.5, with a mean value of 1.9. Many studies have shown an increase of incidence in IBD in Spain over time, with the increase being more evident in cases of CD. In general, IBD is no longer considered a rare disease in Spain, but it has become relatively frequent once again, as in other southern European countries, showing the disappearance of the north–south European gradient.

Genetic analysis of population from Madrid confirmed that NOD2/CARD15 gene was associated with ileal CD in Spanish patients[26]. The report from Galicia showed a lower frequency of NOD2/CARD15 mutations in the local population[27]. Another study in a Spanish population confirmed that NOD2/CARD15 mutations and HLA-DRB1*07 confer susceptibility to the ileal CD, whereas HLA-DRB1*0103 was associated with the colonic location of the disease[28]. The same group reported that interleukin-10 (IL-10) polymorphisms contribute to susceptibility to CD in Spain[29]. Martinez et al. reported the association of the OCTN genes with CD in Spanish patients[30]. A study on the MDR1 (multidrug resistance) gene showed that the C3435 allele was associated with susceptibility to CD and the 3435T allele to UC in the

Spanish population studied[31]. Recent data from a Madrid population also confirmed genetic basis of the heterogeneity of clinical forms of CD[32]. That analysis showed that, in CD patients carrying at least one NOD2/CARD15 mutation, the –1082G allele was associated with ileocolonic disease and the IL-10G14 microsatellite allele was associated with previous appendectomy and smoking at diagnosis.

CONCLUSIONS

The main aim of this chapter was to analyse the epidemiological situation in southern Europe and to review the genetic data generated in the studies conducted in the same region. Epidemiological studies from all four countries analysed in this chapter show the same thing: the incidence of IBD, particularly of CD, is increasing in the entire region and approaching incidences in northern Europe. There is a gradual disappearance of the earlier-observed north–south gradient, with lessening differences between urban and rural areas in the region. Since the genetic data from the region did not detect any specific genetic differences compared with other parts of Europe, southern Europe is fast becoming very similar environmentally to the rest of Europe.

References

1. Shivananda S, Lennard-Jones J, Logan R et al. Incidence of inflammatory bowel disease across Europe: is there a difference between the north and south? Results of the European Collaborative Study on Inflammatory Bowel Disease (EC-IBD). Gut. 1996;39:690–7.
2. Lakatos PL. Recent trends in the epidemiology of inflammatory bowel diseases: up or down? World J Gastroenterol. 2006;12:6102–8.
3. Ravikumara M, Sandhu BK. Epidemiology of inflammatory bowel diseases in childhood. Indian J Pediatr. 2006;73:717–21.
4. Vucelic B, Korac B, Sentic M et al. Ulcerative colitis in Zagreb. Incidence and prevalence 1980–1989. Int J Epidemiol. 1991;20:1043–7.
5. Vucelic B, Korac B, Sentic M et al. Epidemiology of Crohn's disease in Zagreb: a ten years prospective study. Int J Epidemiol. 1991;20:216–20.
6. Sincic B, Vucelic B, Persic M et al. Incidence of inflammatory bowel diseases in Primorsko-goranska County, Croatia, 2000–2004: a prospective population-based study. Scand J Gastroenterol. 2006;41:437–44.
7. Cukovic-Cavka S, Vermeire S, Hrstic I et al. NOD2/CARD15 mutations in Croatian patients with Crohn's disease: prevalence and genotype–phenotype relationship. Eur J Gastroenterol Hepatol. 2006;18:895–9.
8. Vlachonikolis IG, Koutroubakis IE, Manousos ON. Epidemiology of inflammatory bowel diseases in Heraklion, Crete. Scand J Gastroenterol. 2001;36:2–15.
9. Ladas SD, Mallas E, Giorgiotis K et al. Incidence of ulcerative colitis in Central Greece: a prospective study. World J Gastroenterol. 2005;11:1785–7.
10. Papasteriades C, Spiliadis C, Emmanouilidis A et al. Histocompatibility antigens (HLA-A, B) and ulcerative colitis in a Greek population. Digestion. 1986;33:229–32.
11. Gazouli M, Zacharatos P, Mantzaris GJ et al. Association of NOD2/CARD15 variants with Crohn's disease in a Greek population. Eur J Gastroenterol Hepatol. 2004;16:1177–82.
12. Gazouli M, Mantzaris G, Kotsinas A et al. Association between polymorphisms in the Toll-like receptor 4, CD14, and CARD15/NOD2 and inflammatory bowel disease in the Greek population. World J Gastroenterol. 2005;11:681–5.

13. Gazouli M, Mantzaris G, Archimandritis AJ et al. Single nucleotide polymorphisms of OCTN1, OCTN2, and DLG5 genes in Greek patients with Crohn's disease. World J Gastroenterol. 2005;11:7525–30.
14. Cottone M, Matorana G, Di Mitri R et al. Epidemiology of inflammatory bowel disease in Italy. Ital J Gastroenterol Hepatol. 1999;31:503–7.
15. Giachino D, van Duist MM, Regazzoni S et al. Analysis of the CARD15 variants R702W, G908R and L1007fs in Italian IBD patients. Eur J Hum Genet. 2004;12:206–12.
16. Annese V, Palmieri O, Latiano A et al. Frequency of NOD2/CARD15 variants in both sporadic and familial cases of Crohn's disease across Italy. An Italian Group for Inflammatory Bowel Disease Study. Dig Liver Dis. 2004;36:121.
17. Annese V, Lombardi G, Perri F et al. Variants of CARD15 are associated with an aggressive clinical course of Crohn's disease – an IG–IBD Study. Am J Gastroenterol. 2005;100:84–92.
18. Cucchiara S, Latiano A, Palmieri O et al. Role of CARD15, DLG5 and OCTN genes polymorphisms in children with inflammatory bowel diseases. World J Gastroenterol. 2007; 13:1221–9.
19. Ferraris A, Torres B, Knafelz D et al. Relationship between CARD15, SLC22A4/5, and DLG5 polymorphisms and early-onset inflammatory bowel diseases: an Italian multicentric study. Inflamm Bowel Dis. 2006;12:355–61.
20. Bianchi V, Maconi G, Ardizzone S et al. Association of NOD2/CARD15 mutations on Crohn's disease phenotype in an Italian population. Eur J Gastroenterol Hepatol. 2007;19: 217–23.
21. Arin Letamendia A, Burusco Paternain MJ, Borda Celaya F et al. Epidemiological aspects of inflammatory bowel diseases in the Pamplona area. Rev Esp Enferm Dig. 1999;91:769–76.
22. Monferrer Guardiola R, Martin Jimenez JA, Pedraza Sanz RG et al. Incidence of inflammatory bowel diseases in the 02 health area of Castellon (1992–1996). Rev Esp Enferm Dig. 1999;91:33–46.
23. Rodrigo L, Riestra S, Nino P et al. A population-based study on the incidence of inflammatory bowel diseases in Oviedo (Northern Spain). Rev Esp Enferm Dig. 2004;96: 291–6.
24. Garrido A, Martinez MJ, Ortega JA et al. Epidemiology of chronic inflammatory bowel disease in the northern area of Huelva. Rev Esp Enferm Dig. 2004;96:687–94.
25. Pajares JM, Gisbert JP. Epidemiology of inflammatory bowel diseases in Spain. A systematic review. Rev Esp Enferm Dig. 2001;93:9–20.
26. Mendoza JL, Murillo LS, Fernandez L et al. Prevalence of mutations of the NOD2/ CARD15 gene and relation to phenotype in Spanish patients with Crohn's disease. Scand J Gastroenterol. 2003;38:1235–40.
27. Nunez C, Barreiro M, Dominguez-Munoz JE et al. CARD15 mutations in patients with Crohn's disease in a homogenous Spanish population. Am J Gastroenterol. 2004;99:450–6.
28. Fernandez L, Mendoza JL, Martinez A et al. IBD1 and IBD3 determine location of Crohn's disease in the Spanish population. Inflamm Bowel Dis. 2004;10:715–22.
29. Fernandez L, Martinez A, Mendoza JL et al. Interleukin-10 polymorphisms in Spanish patients with IBD. Inflamm Bowel Dis. 2005;11:739–43.
30. Martinez A, Martin MC, Mendoza JL et al. Association of the organic cation transporter OCTN genes with Crohn's disease in the Spanish population. Eur J Hum Genet. 2006;14: 222–6.
31. Urcelay E, Mendoza JL, Martin MC et al. MDR1 gene: susceptibility in Spanish Crohn's disease and ulcerative colitis patients. Inflamm Bowel Dis. 2006;12:33–7.
32. Mendoza JL, Urcelay E, Lana R et al. Polymorphisms in interleukin-10 gene according to mutations of NOD2/CARD15 gene and relation to phenotype in Spanish patients with Crohn's disease. World J Gastroenterol. 2006;12:443–8.

4
Comparison to North and Central Europe

E. LANGHOLZ

INTRODUCTION

The purposes of epidemiological research are to describe occurrence and disease-specific features, to look for variations in occurrence both in time and geography, to identify possible common risk factors and analyse all these factors in order to provide clues to the aetiology of a disease. Since the disease occurrence is often multifactorially determined it is necessary that both environmentally and genetic factors are investigated.

The chronic inflammatory bowel diseases (IBD) Crohn's disease (CD) and ulcerative colitis (UC) are diseases with unknown aetiology. A number of risk factors have been implicated ranging from a positive family history, residence in an urban area in an industrialized country, Caucasian race, to tobacco consumption[1–4].

It has become more and more evident that genetic factors also play an important role. This has been shown in family studies and twin studies with demonstrations of higher risks of IBD in relatives to patients with IBD and greater concordance in monozygotic as compared to dizygotic twins[1,2].

The common hypothesis is that an external stimulus may cause an exaggerated immune response in a genetically susceptible person.

The best epidemiological approach to examine this is by undertaking population-based studies free of most sources of bias that otherwise could hamper the results, combined with twin studies that can provide data both on the genetic influence and on external factors depending on the methodology of research.

During recent decades we have seen a steady increase in the incidence of IBD, especially CD and to a lesser extent in UC, pointing especially to environmental factors as responsible for the rise in incidence, since genetically determined diseases have a stable occurrence. In 2001 it was discovered that mutations in the CARD15 gene contribute to CD susceptibility; since that time the correlations and impact of genetics have been studied intensely[5].

INCIDENCE AND PREVALENCE

Most incidence studies have been reported from Scandinavia, and northern Europe, the USA and Israel, but IBD is present all over the world, when looked for. IBD is apparently more common in the northern part of the world and among Caucasians. It appears equally in men and women. A familial occurrence exists. Peak incidences are reported in young adults. In long-term studies results are divering regarding secular trends; some report an increase in incidence, others a stable or falling incidence. However, it seems from the literature that the incidence of UC tended to level off at the end of the century, whereas the incidence of CD was still increasing in most countries.

However, the most recent data from Scandinavia suggest a further increase in the incidence of IBD. Earlier studies from Copenhagen[6,7] reported a stable incidence of UC of approximately $9.2/10^5$ and a steadily increasing incidence of CD of $4.1/10^5$, while the most recent update for the period 2003–2005 showed a further increase to $8.6/10^5$ for CD and $13.4/10^5$ for UC[8]; the last study, however, was of a shorter duration. The same trend is seen in Stockholm, where the incidence of CD nearly doubled from $4.9/10^5$ in the 1980s to $8.3/10^5$ in the period 1990–2001[9].

Studies from southern Europe have previously reported incidence values much lower than those mentioned above[10–15]; however, these studies were retrospective in design and thereby flawed. In contrast a European multicentre study simultaneusly employing suitable epidemiological methods, i.e. prospective registration and rigorous case ascertainment procedures, in 20 areas of 12 European countries found comparable incidence values between northern and southern Europe[16].

The European incidence rate overall was $10.4/10^5$ for UC (northern centres 11.8 vs. 8.7 in southern centres) and $5.6/10^5$ (northern centres 7.0 vs 3.9 in southern centres). This illustrates that the differences in incidence is much less than previously anticipated; in fact it was shown in the study that the best correlation to incidence was not geography, but rather the size of GNP.

The prevalence rates are still higher in northern Europe compared to southern Europe, but it is important to realize which factors are determining the prevalence. Prevalence is determined by incidence, prognosis, and observation time. Factors reducing the prevalence are high mortality and emigration rates. The hypothetical 'true' prevalence for a disease with a good prognosis and a stable incidence can be calculated as the cumulative incidence multiplied by the mean disease duration, provided that migration rates are balanced. For IBD in Europe this would point to a 'true' prevalence of $680/10^5$ once a steady state is reached.

The low prevalene rates earlier reported from southern Europe probably mirror a previous lesser awareness of IBD, and the absence of solid population-based studies with adequate case ascertainment procedures. The European Collaborative Study on Inflammatory Bowel Diseases (ECIBD) may have contributed to the establishment of better epidemiological studies, and it is anticipated that the next decades will bring prevalence rates of the same magnitude, or they will show the same trend in increasing prevalence as in Scandinavia.

RISK FACTORS

When looking at risk factors the most consistent relationship is found between IBD and cigarette smoking, with a decreased risk of UC in current smokers (OR <1). It also appears that former smokers are more likely to develop UC than those who have never smoked. Cigarette smoking may also influence the disease course of UC with improvement of symptoms and a reduced risk of colectomy[17].

In contrast to UC cigarette smoking is a risk factor for developing CD. Smokers were found in a meta-analysis to be twice more likely to develop CD. Smoking also influences the disease course. Smokers with CD are more likely to have ileal disease than colonic disease[18] and more likely to develop stenosing or fistulizing disease[19]. Continued smoking increases the risk of postoperative recurrence[20].

Another consistently found risk factor is appendectomy, which appears to be protective for development of UC. A large meta-analysis showed that appendectomy was associated with a nearly 70% risk reduction of subsequent UC. Appendectomy seems to increase the risk of subsequent CD. The mechanisms whereby appendectomy protects against UC, but increases the risk of CD, are unknown.

Other risk factors suggested to be responsible for the development of IBD include the use of oral contraceptives, but results are inconclusive. Dietary antigens have also been examined, but despite numerous studies no consensus has emerged, and studies are difficult to interpret due to methodological problems, i.e. recall bias. Perinatal and childhood factors such as lack of breastfeeding, domestic hygiene, perinatal infections or frequent infections in childhood have also been suggested. Other studies have suggested the absence of infections in childhood, but conflicting results have occurred, so in conclusion the strongest risk factors identified so far are a family history of IBD, cigarette smoking and appendectomy.

GENETICS

A genetic aetiology for IBD has been suspected, supported by findings of familial clustering and a greater concordance in monozygotic twins compared to dizygotic[21,22]. The discovery of mutations in CARD15 as an independent risk factor for development of CD has later been confirmed in many studies[5]. In white Americans and Europeans CARD15 mutations are found in 10–20% of controls and in up to 30–40% of CD patients, but mutations are much rarer, or are absent, in other populations[23].

CARD15 mutations seem to confer an increased risk for development of ileal CD, early-onset CD, stricturing CD and CD complicated by internal fistulas[23].

The ECIBD study showed some variations in the incidence of IBD across Europe, although much smaller than anticipated. In a follow up study 10 years later multiple risk factors were assessed among those also the prevalence of different genetic markers in the different participating population-based

cohorts from northern and southern Europe[24]. In that study the prevalence of mutations in CARD15 varied across Europe with a significantly lower prevalence in Scandinavia compared to central and southern Europe. No correlation between the incidence of IBD and the prevalence of mutations in CARD15 was found, and there was no European north to south gradient.

Patients were recruited from 10 centres in eight European countries (Norway, Denmark, The Netherlands, Italy, Spain, Portugal, Greece and Israel). For the total cohort the prevalence of having a mutation in the CARD15 gene was 23.9% for CD, 9.6% for UC and 14.4% for healthy controls. No difference in prevalence between north and south was found. When looking at Scandinavian centres CD patients and controls had a significantly lower prevalence compared to the rest of Europe, 6% vs 11% (OR 2.8, 95% CI 0.14–0.59).

The excess rate of CD in patients with a CARD15 mutation expressed as the population attributable risk percentage (PAR%) was 11.2 for the total, cohort ranging from –13.7 to 44.6%. Overall PAR% was lower in the north compared to south; 6.8% compared to 19.6%. The overall PAR% is low, indicating that the mutations in CARD15 do not play a major role for the susceptibility in CD.

CONCLUSIONS

The previously suggested north–south gradient in occurrence of IBD is less than previously thought. A multicentre European study simultaneusly employing suitable epidemiological methods, i.e. prospective registration and rigorous case ascerainment procedures, in 20 areas of 12 European countries found comparable incidence values between northern and southern Europe.

The disease occurrence is determined both by environmental and to a lesser degree genetic factors.

The prevalence of mutations in CARD15 varies across Europe with a significantly lower prevalence in Scandinavia compared to central and southern Europe. No correlation between the incidence of IBD and the prevalence of mutations in CARD15 was found.

References

1. Orholm M, Munkholm P, Langholz E et al. Familial occurrence of inflammatory bowel disease. N Engl J Med. 1991;324:84–8.
2. Orholm M, Binder V, Sørensen TIA et al. Concordance of inflammatory bowel disease among Danish twins. Results of a nationwide study. Scand J Gastroenterol. 2000;35:1075–81.
3. Probert CS, Jayanthi V, Hughes AO et al. Prevalence and family risk of ulcerative colitis and Crohn's disease: an epidemiological study among Europeans and south Asians in Leicestershire. Gut. 1993;34:1547–51.
4. Sandler RS. Epidemiology. In: Targan SR, Shanahan F, editors. Inflammatory Bowel Disease: From Bench to Bedside. Baltimore, Williams and Wilkins, 1994:5–30.
5. Hugot JP, Chamaillard M, Zouali H et al. Association of NOD2 leucine-rich repeat variants with susceptibility to Crohn's disease. Nature. 2001;411:599–603.
6. Langholz E, Munkholm P, Nielsen OH et al. Incidence and prevalence of ulcerative colitis in Copenhagen county from 1962 to 1987. Scand J Gastroenterol. 1991;26:1247–56.

7. Munkholm P, Langholz E, Nielsen OH et al. Incidence and prevalence of Crohn's disease in the county of Copenhagen, 1962–87: a sixfold increase in incidence. Scand J Gastroenterol. 1992;27:609–14.
8. Vind I, Riis L, Jess T et al. Increasing incidences of inflammatory bowel disease and decreasing surgery rates in Copenhagen City and County, 2003–2005: a population-based study from the Danish Crohn's Colitis Database. Am J Gastroenterol. 2006;101:1274–82.
9. Lapidus A. Crohn's disease in Stockholm County during 1990–2001: an epidemiological update. World J Gastroenterol. 2006;12:75–81.
10. Vucelic B, Korac B, Sentic M et al. Ulcerative colitis in Zagreb, Yugoslavia: incidence and prevalence 1980–1989. Int J Epidemiol. 1991;20:1043–7.
11. Trallori G, d'Albasio G, Palli D et al. Epidemiology of inflammatory bowel disease over a 10-year period in Florence (1978–1987). Ital J Gastroenterol. 1991;23:559–63.
12. Tragnone A, Hanau C, Bazzocchi G et al. Epidemiological characteristics of inflammatory bowel disease in Bologna, Italy – incidence and risk factors. Digestion. 1993;54:183–8.
13. Martinez-Salmeron JF, Rodrigo M, De Teresa J et al. Epidemioogy of inflammatory bowel diseases in the province of Granada, Spain – a retrospective study from 1979 to 1988. Gut. 1993;34:1207–9.
14. Maté-Jimenez J, Muñoz S, Vicent D et al. Incidence and prevalence of ulcerative colitis and Crohn's disease in urban and rural areas of Spain from 1981 to 1988. J Clin Gastroenerol. 1994;18:27–31.
15. Tsianos EV, Masalas CN, Merkouropoulos M et al. Incidence of inflammatory bowel disease in north west Greece: rarity of Crohn's disease in an area where ulcerative colitis is common. Gut. 1994;35:369–72.
16. Shivananda S, Lennard-Jones J, Logan R et al. Incidence of inflammatory bowel disease across Europe: is there a difference between north and south? Results of the European Collaborative Study on Inflammatory Bowel Disease (ECIBD). Gut. 1996;39:690–7.
17. Boyko EJ, Perera DR, Koepsell TD et al. Effects of cigarette smoking on the clinical course of ulcerative colitis. Scand J Gastroenterol. 1988;23:1147–52.
18. Russel MG, Volovics A, Schoon EJ et al. Inflammatory bowel disease: is there any relation between smoking status and disease presentation? Inflamm Bowel Dis. 1998;4:182–6.
19. Picco MF, Bayless TM. Tobacco consumption and disease duration are associated with fistulising and stricturing behaviors in the first 8 years of Crohn's disease. Am J Gastroenterol. 2003;98:363–8.
20. Cosnes J, Carbonnel F, Carrat F et al. Effects of current and former cigarette smoking on the clinical course of Crohn's disease. Aliment Pharmacol Ther. 1999;13:1403–11.
21. Orholm M, Binder V, Sørensen TIA et al. Concordance of inflammatory bowel disease among Danish twins. Results of a nationwide study. Scand J Gastroenterol. 2000;35:1075–81.
22. Halfvarson J, Bodin L, Tysk C et al. Inflammatory bowel disease in a Swedish twin cohort: a long-term follow up of concordance and clinical characteristics. Gastroenterology. 2003;124:1767–73.
23. Economou M, Trikalinos TA, Loizou KT et al. Differential effects of NOD2 variants on Crohn's disease risk and phenotype in diverse populations: a metaanalysis. Am J Gastroenterol. 2004;99:2393–404.
24. Riis L, Vind I, Vermeire S et al. The prevalence of genetic and serologic markers in an unselected European population based cohort of IBD patients. Inflamm Bowel Dis. 2007;13:24–32.

Section II
Bacterial and viral infections mimicking inflammatory bowel disease

Chair: W JAFRI and M FARTHING

5
Ileocolonic tuberculosis: a diagnostic challenge

D. EPSTEIN

INTRODUCTION

Gastroenterologists practising in the developing world face high burdens of enteric infections, epidemic tuberculosis (TB) and an increasing incidence of inflammatory bowel disease (IBD). One of the challenges in this environment is diagnosing intestinal tuberculosis (ITB) and the literature is replete with case reports of ITB being mistaken for Crohn's disease (CD).

Although this problem frequently occurs in developing countries, global migration and the emergence of drug-resistant strains of TB in a number of countries around the world make this relevant to gastroenterologists from both low- and high-income countries.

This chapter discusses the approach to diagnosing colonic tuberculosis (and ITB generally) and how to distinguish this condition from CD. A diagnostic and therapeutic algorithm for cases in which diagnostic uncertainty exists is also proposed (Figure 1).

CD and TB share a multifaceted relationship. First they share a number of pathogenic similarities. Both are chronic granulomatous conditions characterized by a Th1 cytokine response with production of interferon gamma, and interleukins IL-12 and IL-23[1,2]. Impairments in innate immunity are also found in both conditions[3,4]. Host bacterial interactions play an important role in both diseases, and in TB only 5–10% of infected people will develop active disease[5]. Furthermore, steroids have been used in both conditions to treat inflammatory reactions. Secondly both ITB and CD have protean clinical manifestations and share many phenotypic similarities. Lastly treatment of CD, particularly in high TB prevalence environments, can be complicated by the development of active TB.

TB is the most important infectious cause of morbidity and mortality in adults. In 2004 8.9 million new cases and 1.7 million deaths due to TB were reported, the majority of cases from sub-Saharan Africa and Asia[6]. Extrapulmonary TB occurs in 10–15% of cases but has increased dramatically in recent years due to the HIV epidemic. The emergence of multi-drug-resistant (MDR) and more recently extensively drug-resistant (XDR) strains of

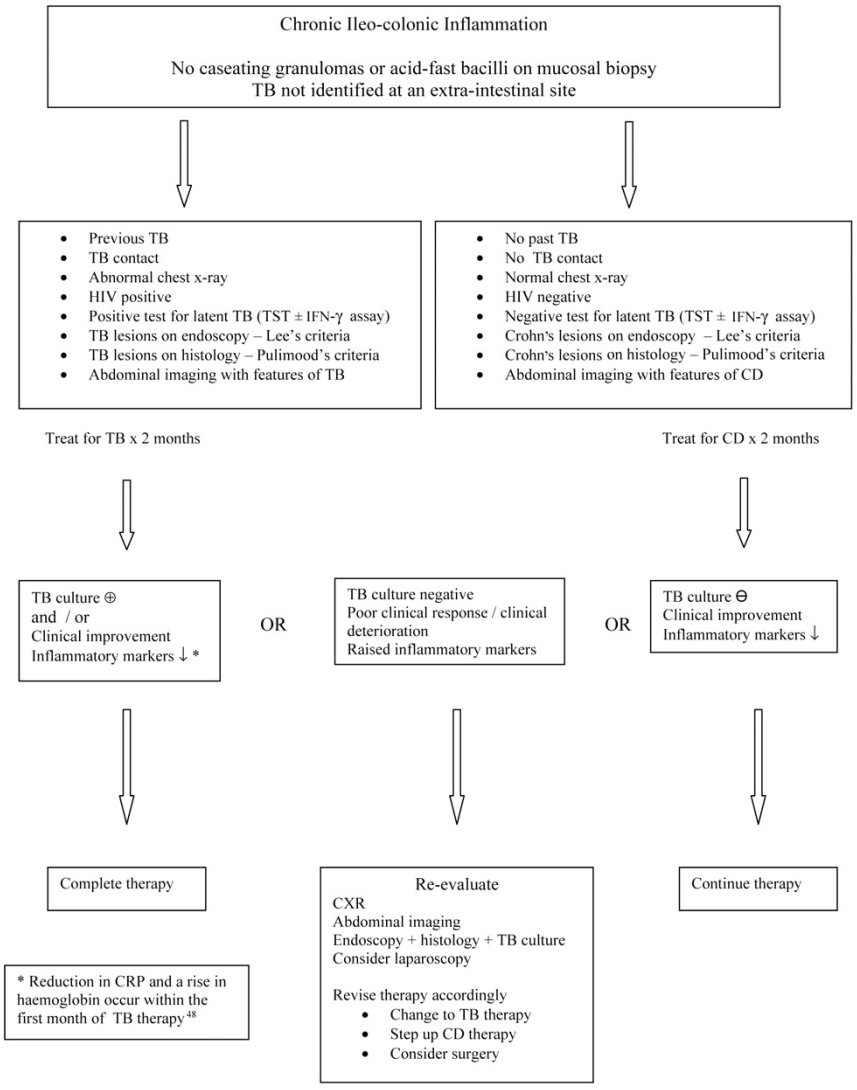

Figure 1 Management algorithm: Crohn's disease vs intestinal tuberculosis[50]

tuberculosis[7] has implications for global public health as these cases are being identified in both developing and developed countries[8]. Gastrointestinal TB is often under-recognized. A study from South Africa found 46% of patients with cavitating pulmonary TB (PTB) had features of ITB when the gastrointestinal tract was examined[9]. This is supported by autopsy data from India, which found that ITB often goes clinically undetected in patients with PTB[10].

HIV increases the risk of developing active TB. In South Africa 36% of people with clinically advanced HIV will develop TB per annum[11]. Tuberculosis in the HIV-infected patient tends to disseminate and manifests in the abdomen as ascites, abdominal lymphadenopathy and hepatosplenic disease. These patients are often smear negative for acid-fast bacilli. ITB with a Crohn's-like phenotype is usually seen in the HIV-negative patient with a robust immune response and contained infection. In contrast to TB the epidemiology of IBD is far less dramatic; nevertheless more cases are being reported from developing countries[12–14].

A common clinical dilemma is the patient with chronic ileocolonic inflammation who resides in a high TB prevalence environment. Often the chest radiograph is normal and endoscopy, histology and abdominal imaging are non-diagnostic. A decision to treat a patient for CD with steroids, immunomodulators or biologicals cannot be taken lightly, as a missed diagnosis of ITB may have disastrous consequences.

CLINICAL FEATURES OF INTESTINAL TUBERCULOSIS

ITB affects young and middle-aged people in developing countries with no distinguishing demographics. Symptoms develop insidiously and may be present for several years before a diagnosis is made. Furthermore ITB has been identified in patients undergoing colonoscopy who were either well or had trivial symptoms[15]. Patients with ITB present with constitutional symptoms, symptoms of ileocolitis or an abdominal mass. More acute presentations with bowel perforation or obstruction may also occur. Rarely ITB may present with malabsorption and a protein-losing enteropathy.

Smoking, a well-known CD association, is also more common in PTB[16,17], but no direct relationship with ITB has been identified.

A number of CD-related extraintestinal manifestations can be mimicked by TB. TB can directly involve a number of extrapulmonary sites such as the joints[18], soft tissues[19], skin[20] and eyes[21]. A number of immune-mediated phenomena are also seen in TB, such as a reactive arthritis (Poncet's disease)[18], conjunctivitis (phlyctenular conjunctivitis)[21] and erythema nodosum[20]. Thromboembolic disease, well recognized in IBD, is also associated with TB[22,23].

Perianal fistulas are a clinical hallmark of CD; however, these are well described in ITB. A surgical series from South Africa found that 17% of patients with perianal fistulas had TB[24]. Extrapulmonary TB is often associated with a normal chest radiograph and similarly in ITB less than 50% of cases will have evidence of TB on chest radiograph[25–27].

Tuberculin skin testing has been used for over a century as a diagnostic test for TB; however, a positive test may reflect latent infection, previous bacille Calmette-Guérin (BCG) vaccination or exposure to non-tuberculous mycobacteria. Conversely a negative test can occur with disseminated or primary TB and HIV co-infection. Studies from the 1960s indicate that anergy to skin testing may also be a feature of untreated CD[28].

Anaemia, leukocytosis, thrombocytosis and elevated inflammatory markers are seen in both ITB and CD and have no discriminatory value in diagnosis.

RADIOLOGY IN THE DIAGNOSIS OF ITB

Barium contrast studies are useful in demonstrating luminal detail and characterizing the inflammatory and cicatrizing lesions of both ITB and CD, but are seldom diagnostic. Cross-sectional imaging such as ultrasound, computed tomography scan and magnetic resonance imaging are useful in evaluating the bowel wall changes and other abdominal features such as mesenteric attachments, lymph nodes and other organs. However, if ITB is not associated with ascites, extensive lymphadenopathy or hepatosplenic lesions, then differentiating CD from ITB is difficult. Intestinal lesions in combination with features of primary sclerosing cholangitis, sacro-iliitis, non-alcoholic fatty liver disease and gallstones would favour a diagnosis of CD. Fibrofatty proliferation of the mesentery ('creeping fat') is well recognized in CD, but has also been described in a large ITB surgical series from India[29].

ENDOSCOPY IN THE DIAGNOSIS OF ITB

ITB can take a variety of forms in terms of extent, morphology and complications[26,30–34]. The ileocaecum is the most commonly involved area with varying degrees of contiguous colon involvement. However, a variety of combinations of colonic, small bowel and upper GI disease can occur. Lesions may be diffuse, for example a pancolitis pattern similar to ulcerative colitis, or discrete. Solitary lesions and skip lesions are also well described. The morphology of ITB lesions is also variable, with ulcers being the most common finding but hypertrophic lesions (mass lesions, nodules, pseudopolyps, pseudoshelves) and strictures can also occur. As with ITB combinations of these lesions are also found in IBD.

Aoki and colleagues published one of the earliest descriptions of colonic TB in 1975[35]. Ulcers with transverse orientation, ileocaecal destruction and inflammatory polyps were described. Since then these and other lesions have been reported; however, there is considerable overlap with lesions found in CD. Lee and colleagues[36], in a prospective study, compared patients with colonic TB and CD and devised a scoring system based on four features of ITB (transverse ulcers, pseudopolyps, involvement of fewer than four segments, and a patulous ileocaecal valve) and four features of CD (anorectal lesions, longitudinal ulcers, aphthous ulcers, and cobblestone appearance). Using this methodology a positive predictive value for CD of 94.9% and a positive predictive value of 88.9% for ITB was achieved.

Table 1 Prevalence (%) of selected histological parameters in patients with intestinal tuberculosis (ITB) and Crohn's disease (CD): a comparison of three similar studies[50]

	Pulimood et al.[41] *Southern India*		*Pulimood et al.*[40] *Southern India*		*Kirsch et al.*[39] *Cape Town, South Africa*	
	ITB (n = 20)	*CD (n = 20)*	*ITB (n = 33)*	*CD (n = 31)*	*ITB (n = 18)*	*CD (n = 25)*
Caseous necrosis	40	0	36	0	22	0
Confluent granulomas	60	0	42	3	50	0
$\geqslant 5$ granulomas/biopsy site	40	0	45	0	44	24
$\geqslant 10$ granulomas/biopsy site	–	–	–	–	33	0
Large granulomas	*Diameter* > 200 μm		*Diameter* > 400 μm		*Area* > 0.05 *mm*^{2}	
	90	5	51	0	67	8
Submucosal granulomas	45	5	39	6	44	12
Ulcers lined by bands of epithelioid histiocytes	45	5	61	0	61	8
Disproportionate submucosal inflammation	65	5	–	–	67	10
Architectural distortion distant to granulomatous inflammation	–	–	0	62	–	–

HISTOLOGICAL DIAGNOSIS OF ITB

Confirming a diagnosis of ITB is difficult in surgically resected specimens as extrapulmonary TB tends to be paucibacillary. In the context of superficial endoscopic mucosal biopsies the finding of pathognomonic histological lesions, i.e. acid-fast bacilli and/or caseating granulomas, is uncommon (less than 30%)[37–41]. Similarly a positive TB culture is unusual. However, a number of histological features other than acid-fast bacilli and caseating granulomas can be used to identify the TB patient. These include: large granulomas, confluent granulomas, submucosal granulomas, disproportionate submucosal inflammation and ulcers lined by bands of epithelioid histiocytes. A summary of three studies[39–41] is presented in Table 1.

Nucleic acid amplification tests targeting the *Mycobacterium tuberculosis* complex have high specificity but variable sensitivity in the diagnosis of TB. Several studies have looked at the role of TB polymerase chain reaction in the diagnosis of ITB. Four retrospective studies using formalin-fixed specimens are diagnostic in 22% to 75% of confirmed ITB cases[32,37,38,42].

NEW TOOLS FOR THE DIAGNOSIS OF TB

A number of new diagnostic tests for TB have been developed in recent years. These include the interferon gamma release assays, skin patch test, antibody tests, antigen recognition tests and rapid culture systems[43,44]. Although promising, these modalities have been developed and tested in populations with low TB prevalence. Their performance in populations with high rates of TB, co-infection with HIV and co-infection with helminths, which may alter the immune milieu, remains to be seen. These modalities also need to be functional within the constraints of a resource-poor environment. Furthermore their value in diagnosing ITB remains unknown.

NEW TOOLS IN THE DIAGNOSIS OF IBD

Genetic testing and serology are emerging modalities for the diagnosis of IBD in 'traditional' Western IBD populations. The three common NOD2/CARD15 mutations, however, are not associated with CD in IBD patients from Asia and South Africa[45]. Intuitively it would seem that NOD2 mutations would predispose to TB; however, a study of PTB patients in West Africa[46] found no evidence of these mutations in PTB patients or controls. Furthermore an unpublished study by Gill Watermeyer, from our unit in Cape Town, found no evidence of NOD2 mutations in patients with ITB.

Antibodies to the ubiquitous yeast *Saccharomyces cerevisiae* (ASCA) are specific for the diagnosis of CD in Caucasian, Chinese and Japanese IBD patients. A study from India[47] compared IgA and IgG ASCA in patients with CD, ulcerative colitis (UC) and ITB, as well as healthy controls. ASCA was positive in a significant number of both CD and ITB patients when compared to both UC patients and controls. This study suggests that ASCA has poor

discriminatory value between CD and ITB in a high TB prevalence environment. This study also raises the intriguing question as to why so many ITB patients are positive for ASCA. Hypotheses include increased intestinal permeability in ITB or structural homology between yeast oligomannanosides and mycobacterial lipoarabinomannan. The latter would incriminate *Mycobacterium avium* ssp. *paratuberculois* again as the causative agent in CD.

CONCLUSIONS

The diagnosis of ITB should be based on careful clinical evaluation, in particular cautious interpretation of extraintestinal signs, imaging, systematic endoscopic evaluation and histological evaluation according to criteria described by Pulimood et al.[41]. If a decision is made to treat for TB then objective responses to therapy should be seen within 8 weeks of therapy[48].

Walsh noted that 'It is impossible to diagnose abdominal tuberculosis with any degree of certainty, since the disease mimics many other abdominal conditions and histological confirmation may be equivocal'[49]. This was published in 1909, and despite the many advances over the past 100 years the diagnosis of ITB remains a challenge.

References

1. Raj A. Immunology of tuberculosis. Indian J Med Res. 2004;120:213–32.
2. Podolsky DK. Inflammatory bowel disease. N Engl J Med. 2002;347:417–29.
3. Comalada M, Peppelenbosch MP. Impaired innate immunity in Crohn's disease. Trends Mol Med. 2005;12:397–9.
4. Korzenik JR, Dieckgraefe BK, Valentine JF et al. Sargramostim for active Crohn's disease. N Engl J Med. 2005;352:2193–201.
5. Harries AD, Dye C. Tuberculosis. Ann Trop Med Parasitol. 2006;100:415–31.
6. Dye C. Global epidemiology of tuberculosis. Lancet. 2006;367:938–40.
7. Gandhi NR, Moll A, Sturm AW et al. Extensively drug-resistant tuberculosis as a cause of death in patients co-infected with tuberculosis and HIV in a rural area of South Africa. Lancet. 2006;368:1575–80.
8. Raviglione MC, Smith IM. XDR tuberculosis – implications for global public health. N Engl J Med. 2007;356:656–9.
9. Pettengell KE, Larsen C, Garb M et al. Gastrointestinal tuberculosis in patients with pulmonary tuberculosis. Q J Med. 1990;74:303–8.
10. Sharma MP, Bhatia V. Abdominal tuberculosis. Indian J Med Res. 2004;120:305–15.
11. Boulle A, Zweigenthal V, Hildebrand K et al. Incidence of tuberculosis pre- and post-ART in a setting of high tuberculosis-HIV comorbidity. Poster MoPeB 3239 9th International Workshop in HIV Observational Databases, Budapest, 2005.
12. Ouyang Q, Tandon R, Goh K-L et al. The emergence of inflammatory bowel disease in the Asian Pacific region. Curr Opin Gastroenterol. 2005;21:408–13.
13. Wright JP, Froggatt J, O'Keefe EA et al. The epidemiology of inflammatory bowel disease in Cape Town 1980–1984. S Afr Med J. 1996;70:10–15.
14. Lakatos PL. Recent trends in the epidemiology of inflammatory bowel disease: up or down? World J Gastroenterol. 2006;14:6102–8.
15. Sato S, Yao K, Yao T et al. Colonoscopy in the diagnosis of intestinal tuberculosis in asymptomatic patients. Gastrointest Endosc. 2004;59:362–8.
16. den Boon S, van Lill SWP, Borgdorff MW et al. Association between smoking and tuberculosis infection: a population survey in a high tuberculosis incidence area. Thorax. 2005;60:555–7.

17. Chiang C-Y, Slama K, Enarson DA. Associations between tobacco and tuberculosis. Int J Tuberc Lung Dis. 2007;11:258–62.
18. Franco-Paredes C, Diaz-Borjon A, Senger MA et al. The ever expanding association between rheumatologic disease and tuberculosis. Am J Med. 2006;119:470–7.
19. Abdelwahab IF, Bianchi S, Martinoli C et al. Atypical extraspinal musculoskeletal tuberculosis in immunocompetent patients. Part II: tuberculous myositis, tuberculous bursitis and tuberculous tenosynovites. Can Assoc Radiol J. 2006;57:278–86.
20. Umapathy KC, Begum R, Ravichandran G et al. Comprehensive findings on clinical, bacteriological, histopathological and therapeutic aspects of cutaneous tuberculosis. Trop Med Int Health. 2006;11:1521–8.
21. Kurup SK, Chan C-C. *Mycobacterium*-related ocular inflammatory disease: diagnosis and management. Ann Acad Med Singapore. 2006;35:203–9.
22. Robson SC, White NW, Aronson I et al. Acute phase response and the hypercoagulable state in pulmonary tuberculosis. Br J Haematol. 1996;93:943–9.
23. White NW. Venous thrombosis and rifampicin. Lancet. 1989;2:434–5.
24. Terreblanche J. Fistula in ano: a five year survey at Groote Schuur Hospital and a review of the literature. S Afr Med J. 1964:403–8.
25. Singh V, Kumar P, Kamal J et al. Clinicocolonoscopic profile of colonic tuberculosis. Am J Gastroenterol. 1996;3:565–8.
26. Alvares JF, Devarbhavi H, Makhija P et al. Clinical, colonoscopic, and histological profile of colonic tuberculosis in a tertiary hospital. Endoscopy. 2005;37:351–6.
27. Patel N, Amarapurkar D, Agal S et al. Gastrointestinal luminal tuberculosis: establishing a diagnosis. J Gastroenterol Hepatol. 2004;19:1240–6.
28. Verrier Jones J, Housley J, Ashurst PM et al. Development of delayed hypersensitivity to dinitrochlorobenzene in patients with Crohn's disease. Gut. 1969;10:52–6.
29. Bhansali SK. Abdominal tuberculosis: experience with 300 cases. Am J Gastroenterol. 1977;67:324–37.
30. Bhargava DK, Kushwaha AKS, Dasarathy S et al. Endoscopic diagnosis of segmental colonic tuberculosis. Gastrointest Endosc. 1992;38:571–4.
31. Kalvaria I, Kottler RE, Marks IN. The role of colonoscopy in the diagnosis of tuberculosis. J Clin Gastroenterol. 1988;10:516–23.
32. Kim KM, Lee A, Choi KY et al. Intestinal tuberculosis: clinicopathologic analysis and diagnosis by endoscopic biopsy. Am J Gastroenterol. 1998;93:606–9.
33. Nagi B, Sodhi KS, Kochhar R et al. Small bowel tuberculosis: enteroclysis findings. Abdom Imaging. 2004;29:335–40.
34. Shah S, Thomas V, Mathan M et al. Colonoscopic study of 50 patients with colonic tuberculosis. Gut. 1992;33:347–51.
35. Aoki G, Nagasako K, Nakae Y et al. The fibercolonoscopic diagnosis of intestinal tuberculosis. Endoscopy. 1975;7:113–21.
36. Lee YJ, Yang SK, Byeon JS et al. Analysis of colonoscopic findings in the differential diagnosis between intestinal tuberculosis and Crohn's disease. Endoscopy. 2006;38:592–7.
37. Amarapurkar DN, Patel ND, Amarapurkar AD et al. Tissue polymerase chain reaction in diagnosis of intestinal tuberculosis and Crohn's disease. J Assoc Phys India. 2004;52:863–7.
38. Gan HT, Chen YQ, Ouyang Q et al. Differentiation between intestinal tuberculosis and Crohn's disease in endoscopic biopsy specimens by polymerase chain reaction. Am J Gastroenterol. 2002;97:1446–51.
39. Kirsch R, Pentecost M, Hall Pde M et al. Role of colonoscopic biopsy in distinguishing between Crohn's disease and intestinal tuberculosis. J Clin Pathol. 2006;59:840–4.
40. Pulimood AB, Peter S, Ramakrishna B et al. Segmental colonoscopic biopsies in the differentiation of ileocolic tuberculosis from Crohn's disease. J Gastroenterol Hepatol. 2005;20:688–96.
41. Pulimood AB, Ramakrishna BS, Kurian G et al. Endoscopic mucosal biopsies are useful in distinguishing granulomatous colitis due to Crohn's disease from tuberculosis. Gut. 1999; 45:537–41.
42. Balamurugan R, Venkataraman S, John KR et al. PCR amplification of the IS6110 insertion element of *Mycobacterium tuberculosis* in fecal samples from patients with intestinal tuberculosis. J Clin Microbiol. 2006;44:1884–6.
43. Pai M, Kalantri S, Dheda K. New tools and emerging technologies for the diagnosis of tuberculosis: Part I. Latent tuberculosis. Expert Rev Mol Design. 2006;6:413–22

44. Pai M, Kalantri S, Dheda K. New tools and emerging technologies for the diagnosis of tuberculosis. Part II. Active tuberculosis and drug resistance. Expert Rev Mol Design. 2006;6:423–32.
45. Zaahl MG, Winter T, Warnich L et al. Analysis of the three common mutations in the CARD15 gene (R702W, G908R, 1007fs) in South African coloured patients with inflammatory bowel disease. Moll Cell Probes. 2005;19:278–81
46. Stockton JC, Howson JM, Awomoyi AA et al. Polymorphisms in NOD2, Crohn's disease and susceptibility to pulmonary tuberculosis. FEMS Immunol Med Microbiol. 2004;41: 157–60.
47. Makharia GK, Sachdev V, Gupta R et al. Anti-*Saccharomyces cerevisiae* antibody does not differentiate between Crohn's disease and intestinal tuberculosis. Dig Dis Sci. 2007;52:33–9.
48. Lawn SD, Obeng J, Acheampong JW et al. Resolution of the acute-phase response in West African patients receiving treatment for pulmonary tuberculosis. Int J Tuberc Lung Dis. 2000;4:340–4.
49. Walsh J. Diagnosis of intestinal tuberculosis. Trans Natl Assoc Prev Tuberc (London). 1909;5:217–22.
50. Epstein D, Watermeyer G, Kirsch R. Review: Diagnosis and management of Crohn's disease in populations with high risk rates for tuberculosis. Aliment Pharmacol Ther. 2007 (In press).

6
Bacterial and viral infections mimicking inflammatory bowel diseases

M. TÖRÜNER

Inflammatory bowel disease (IBD) – ulcerative colitis (UC) and Crohn's disease (CD) – is a group of chronic diseases involving the gastrointestinal system, characterized by inflammation in bowel and sometimes associated with extra-intestinal features. IBD is characterized by clinical symptoms such as bloody diarrhoea, passage of mucus, abdominal pain, fever, weight loss, etc., depending on the specific disease, either UC or CD.

There are several disorders which can simulate IBD, including infectious colitis (bacterial, viral and parasites), malignant diseases, radiation-induced colitis, non-steroidal anti-inflammatory drug-induced colitis, diverticulitis and some motility disorders such as irritable bowel syndrome.

In recent years (due to the more common use of immunomodulators in treatment of IBD and the entrance of biologic agents into IBD treatment schemes such as tumour necrosis factor alpha inhibitors) differential diagnosis between infectious colitis simulating IBD and IBD itself has become more crucial.

In this chapter bacterial and viral infections mimicking inflammatory bowel diseases will be reviewed.

INFECTIONS

Intestinal tuberculosis

Intestinal tuberculosis is a rare but increasing infection in western communities which is caused by acid-fast bacilli – *Mycobacterium tuberculosis*. It is seen more commonly in immigrants and immunocompromised patients. Diagnosis of intestinal tuberculosis is nearly always challenging, especially in the absence of pulmonary tuberculosis. Microorganisms could be transmitted by swallowing the infected sputum, haematogenous spread from active pulmonary disease or milliary tuberculosis, ingestion of contaminated milk or

food and contagious spread from adjacent organs. The most common macroscopic appearance of intestinal lesions is ulcerative (multiple ulcers). Other lesions which might be seen are hypertrophic and ulcerohypertrophic. Symptoms of intestinal tuberculosis include night fever, diarrhoea, abdominal pain, right quadrant mass, etc. The ileocaecal area is the most commonly involved part of the intestine. Differential diagnosis between tuberculosis and IBD is so important because, if tuberculosis is suspected, empirical anti-tuberculosis treatment should be started, especially when immunomodulatory and immunosuppressive drugs are to be initiated for IBD. Differential diagnosis between these two diseases is quite difficult because of similarities in clinical symptoms and some similarities in diagnostic test results. The diagnostic procedure of choice for intestinal tuberculosis is colonoscopy and biopsy. Appropriately stained slides should be carefully examined for acid-fast rods; culture of a biopsy specimen might increase the diagnostic yield. The culture for *M. tuberculosis* is reliable but results take 4–6 weeks. Polymerase chain reaction analysis of biopsy specimens for *M. tuberculosis* is found to be more sensitive than direct microscopy and culture. Makharia et al.[1] have investigated the role of serological markers such as anti-*Saccharomyces cerevisiae* IgA (ASCA IgA) and IgG (ASCA IgG) in differential diagnosis between UC, CD and intestinal tuberculosis. ASCA IgA was positive in 28%, 33.9% and 43.3% of patients, respectively and ASCA IgG was positive in 24%, 50.8% and 46.6% of patients, respectively[1]. Differences between these three groups did not reach statistical significance. Several studies have demonstrated the usefulness of colonoscopic biopsies in differential diagnosis between CD and intestinal tuberculosis. Pulimood et al. have identified five factors favouring intestinal tuberculosis, including large granulomas, more than four sites of granulomatous inflammation, caseation, band of epithelioid histiocytes and granulomatous inflammation in the caecum. In the same study, factors including non-tuberculous granulomas, mucosal changes distant to sites with granulomas, focal crypt-related inflammation and granulomas in sigmoid or rectum were found to favour CD[2]. Kirsch et al. have evaluated clinical and histopathological findings to differentiate CD and tuberculosis. In this study a positive chest X-ray for tuberculosis, confluent granulomas, more than 10 granulomas per biopsy site, caseous necrosis, ulcers lined by conglomerate epithelioid histiocytes and disproportionate submucosal inflammation were found more commonly in patients with intestinal tuberculosis, whereas perianal fistulas and extraintestinal manifestations were found more commonly in patients with CD[3]. In another study, performed by Lee et al., anorectal lesions, longitudinal ulcers, aphthous ulcers and cobblestone appearance were more frequent in CD patients, whereas involvement of fewer than four segments, a patulous ileocaecal valve, transverse ulcers, scars or pseudopolyps were more frequent in patients with intestinal tuberculosis[4].

The treatment for intestinal tuberculosis consists of anti-tuberculosis therapy including isoniazid, rifampicin, pyrazinamide and either streptomycin or ethambutol, in addition to surgical treatment, which should be reserved for complications.

Clostridium difficile Infection

Clostridium difficile is a Gram-positive spore-forming rod which is present naturally in the environment, more commonly on contaminated hospital surfaces. Normally there is a resistance to *C. difficile* colonization; however, antibiotic use which disturbs the natural flora decreases the resistance to *C. difficile* colonization. After colonization, several factors determine (mainly host's immune system) whether a person becomes ill or not. This microorganism produces three types of toxin; toxin A, toxin B and binary toxin. Toxin B is 1000 times more cytotoxic than toxin A. Toxins A and B induce fluid secretion, apoptosis of intestinal epithelial cells and a marked inflammation. Clinical symptoms of *C. difficile* infection include diarrhoea, which is mostly watery and rarely bloody, fever and crampy abdominal pain. There are several risk factors for development of *C. difficile*-associated diarrhoea; increasing age, severe underlying disease, non-surgical gastrointestinal procedures, presence of a nasogastric tube, receiving anti-ulcer medication, stay on intensive-care unit, multiple antibiotic use and long hospital stay. Cytotoxicity assay is gold standard procedure for diagnosis. It requires 48 h to obtain an accurate result and it is more sensitive than enzyme immunoassay in detecting toxin B. Other diagnostic tests are enzyme immunoassay and colonoscopy. With enzyme immunoassay, detection of toxin A and B or toxin A alone is possible. The sensitivity of this test reaches to 80% per stool if three examinations are performed. In colonoscopy, pseudomembranes are the characteristic features. In addition, non-specific features such as erythema, oedema, friability, and non-specific colitis with small ulcerations could be seen during the colonoscopic procedure.

C. difficile infection is one of the infections which should be investigated in refractory IBD. Rodemann et al. have found an increased incidence of *C. difficile* infection in both CD (9.5–22.3/1000) and UC (18.4–57.6/1000) between 1998 and 2004[5]. Another study which is supporting the evidence of increased frequency of *C. difficile* infection in IBD has showed that the rate of *C. difficile* infection increased from 1.8% in 2004 to 4.6% in 2005[6]. Colonic involvement and maintenance immunomodulator use are independent risk factors for *C. difficile* infection and it is more often contracted infection as an outpatient (76%)[6].

Treatment is not always essential in patients with toxin positivity unless evidence of colitis (leukocytosis, endoscopic findings, etc.) or severe diarrhoea is present, or if a patient has persistent diarrhoea despite cessation of the offending antibiotic treatment or the presence of a need to continue antibiotic therapy to treat the underlying infection. For treatment the drug of choice is metranidazole (500 mg t.i.d. for 10–14 days). Vancomycin (alternative first-line drug, 125 mg q.i.d. for 10–14 days), fucidic acid (500 mg t.i.d. for 10 days) or nitazoxanide (500 mg b.i.d. for 10 days) are alternative drugs in treatment.

Cytomegalovirus infection

Human cytomegalovirus (CMV) is a member of the herpes virus family. CMV is present in a latent state in most people[7] whereas CMV-induced ulceration of

the bowel is frequently observed during disseminated viraemia in immunocompromised patients[8,9]. It has been believed that CMV is either acquired at birth or transmitted through sexual or parental exposure. Antigenic stimuli or immune suppression leads to CMV activation; if this happens in the bowel wall, the condition is then called gastrointestinal CMV disease. Clinical manifestations of CMV colitis include fever, abdominal pain, diarrhoea, bloody diarrhoea and haematochezia. Colonoscopy reveals patchy erythema, deep ulcers, and multiple mucosal erosions which occur mostly in the right colon[10]. Cytoplasmic inclusions might be seen in haematoxylin and eosin staining by 10% to 87%[11], sensitivity increases to 93% when immunohistochemistry is performed[12]. Serological tests such as CMV IgM and CMV IgG, as well as CMV culture, can be used as diagnostic tests which have low sensitivity. The CMV antigen test detects late structural protein pp65 produced in leukocytes through staining with immunofluorescent pp65-specific monoclonal antibodies. This test is semi-quantative, so results are somewhat subjective[12]. Polymerase chain reaction testing for CMV has been used more frequently in recent years. Plasma or whole blood can be used for PCR testing. Cut-off values for PCR still remain unclear. CMV colitis does not occur only in immunosuppressed patients. A meta-analysis covering the years between 1980 and 2003 has identified 44 immunocompetent patients with CMV infections. In this group of patients female patients survived better, younger patients (<55 years) survived better, and patients treated with antivirals survived better than patients who had colectomy[13].

In the case of IBD flare or steroid-refractory IBD one should consider CMV infection. There are several reports indicating frequent CMV infections in IBD patients. Maconi et al. showed CMV infection in 22% of resected colon materials of IBD patients[14]. Cottone et al. have identified CMV infection in 18% of steroid-refractory patients[15]. CMV infection does not occur only in corticosteroid-refractory IBD patients. In a prospective study including 42 consecutive IBD patients, CMV infection prevalence has been found as 21% (9/42), whereas it was 33% (4/12) in the corticosteroid-refractory group[16].

There is no consensus as to whether to treat CMV infection in IBD patients or not[16,17]. Ganciclovir (5 mg/kg every 12 h for 3–5 days), oral valganciclovir and foscarnet (90 mg/kg intravenously, every 12 h for 2–3 weeks) are drugs of choice in the treatment of CMV infections.

Campylobacter jejuni infection

Campylobacter jejuni is a motile, non-spore-forming, comma-shaped, Gram-negative rod. Transmission via direct contact with infected animals and faecal–oral person-to-person transmission have been reported. The sites of intestinal inflammation include jejunum, ileum and colon. This microorganism accounts for 3.2–30% of cases of enterocolitis in the general population[18,19]. Clinical features include fever, crampy abdominal pain, watery or bloody diarrhoea, toxic megacolon in very severe disease, enlarged mesenteric lymph nodes, reactive arthritis (HLA-B27 positive) and Gullian–Barré syndrome (1:2000 cases). The prevalence of *Campylobacter* infection in relapsing IBD has been found to be between 1.5% and 4.5%[19–21]. Diagnosis is made using either direct

examination of fresh stool, which shows characteristic darting microorganisms, or stool and blood culture for *C. jejuni* using microaerobic conditions.

Erythromycin (500 mg b.i.d. for 5–7 days) is the drug of choice for treatment. Azithromycin and fluoroquinolones are also effective, but increasing tolerance to fluoroquinolones is a major concern.

Yersinia enterocolitica infection

Yersinia enterocolitica is a pleomorphic Gram-negative bacillus which can be acquired by oral ingestion or direct inoculation (i.e. blood transfusion, etc.). Once the organism has invaded, *Y. enterocolitica* localizes in lymphoid tissue within the gut wall and in regional mesenteric lymph nodes. Symptoms of *Yersinia* infection include fever, right quadrant pain, vomiting, bloody or watery diarrhoea and sore throat. Sometimes infections can mimic acute appendicitis. As diagnostic tools, stool culture, agglutination assays, ELISA and immunoblotting can be used[22].

The prevalence of *Y. enterocolitica* infection in IBD was found as 8/73 = 11%; in UC it was 5/53 = 9.4% and in CD it was 3/20 = 15%[23].

Treatment of acute, non-complicated yersiniosis is not beneficial; most cases of *Yersinia* enterocolitis do not require treatment. Treatment of an individual case should be judged clinically because of clinical severity or the underlying condition of the patient (e.g. immunocompromised patients). Drugs of choice are fluoroquinolones such as ciprofloxacin (500 mg twice daily) or trimethoprimsulphamethoxazole for paediatric patients (TMP 8 mg/kg per day and SMX 40 mg/kg per day in two divided doses).

CONCLUSIONS

Ulcerative colitis and Crohn's disease account for only a small proportion of the number of patients with inflammatory colitis. The majority of inflammatory colitis cases are due to infectious causes, and these infectious causes can mimic IBD or trigger the activation of IBD. Differential diagnosis between IBD and infectious colitis is important to treat patients adequately.

References

1. Makharia GK, Sachdev V, Gupta R, Lal S, Pandey RM. Anti-*Saccharomyces cerevisiae* antibody does not differentiate between Crohn's disease and intestinal tuberculosis. Dig Dis Sci. 2007;52:33–9.
2. Pulimood AB, Peter S, Ramakrishna B et al. Segmental colonoscopic biopsies in the differentiation of ileocolic tuberculosis from Crohn's disease. J Gastroenterol Hepatol. 2005;20:688–96.
3. Kirsch R, Pentecost M, Hall M, Epstein DP, Watermeyer G, Friederich PW. Role of colonoscopic biopsy in distinguishing between Crohn's disease and intestinal tuberculosis. J Clin Pathol. 2006;59:840–4.
4. Lee YJ, Yang SK, Byeon JS et al. Analysis of colonoscopic findings in the differential diagnosis between intestinal tuberculosis and Crohn's disease. Endoscopy. 2006;38:592–7.

5. Rodemann JF, Dubberke ER, Reske KA, da Seo H, Stone CD. Incidence of *Clostridium difficile* infection in inflammatory bowel disease. Clin Gastroenterol Hepatol. 2007;5:339–44.
6. Issa M, Vijayapal A, Graham MB et al. Impact of *Clostridium difficile* on inflammatory bowel disease. Clin Gastroenterol Hepatol. 2007;5:345–51.
7. Surawicz CM, Myerson D. Self-limited cytomegalovirus colitis in immunocompetent individuals. Gastroenterology. 1988;94:194–9.
8. Goodgame RW. Gastrointestinal cytomegalovirus disease. Ann Intern Med. 1993;119:924–35.
9. Kaplan B, Meier-Kriesche HU, Jacobs MG et al. Prevalence of cytomegalovirus in the gastrointestinal tract of renal transplant recipients with persistent abdominal pain. Am J Kidney Dis. 1999; 34:65–8.
10. Ljungman P, Griffiths P, Paya C et al. Definitions of cytomegalovirus infection and disease in transplant recipients. Clin Infect Dis. 2002;34:1094–7.
11. Streetz KL, Buhr T, Wedemeyer H et al. Acute CMV-colitis in a patient with a history of ulcerative colitis. Scand J Gastroenterol. 2003;38:119–22.
12. Kambham N, Vij R, Cartwright CA, Longacre T. Cytomegalovirus infection in steroid-refractory ulcerative colitis: a case–control study. Am J Surg Pathol. 2004;28:365–73.
13. Galiatsatos P, Shrier I, Lamoureux E, Szilagyi A. Meta-analysis of outcome of cytomegalovirus colitis in immunocompetent hosts. Dig Dis Sci. 2005;50:609–16.
14. Maconi G, Colombo E, Zerbi P et al. Prevalence, detection rate and outcome of cytomegalovirus infection in ulcerative colitis patients requiring colonic resection. Dig Liver Dis. 2005;37: 418–23.
15. Cottone M, Pietrosi G, Martorana G et al. Prevalence of cytomegalovirus infection in severe refractory ulcerative and Crohn's colitis. Am J Gastroenterol. 2001;96:773–5.
16. Criscuoli V, Casa A, Orlando A et al. Severe acute colitis associated with CMV: a prevalence study. Dig Liver Dis. 2004;36:818–20.
17. Papadakis KA, Tung JK, Binder SW et al. Outcome of cytomegalovirus infections in patients with inflammatory bowel disease. Am J Gastroenterol. 2001;96:2137–42.
18. Saenz Y, Zarazaga M, Lantero M, Gastanares MJ, Baquero F, Torres C. Antibiotic resistance in *Campylobacter* strains isolated from animals, foods, and humans in Spain in 1997–1998. Antimicrob Agents Chemother. 2000; 44:267–71.
19. Boyanova L, Gergova G, Spassova Z et al. *Campylobacter* infection in 682 Bulgarian patients with acute enterocolitis, inflammatory bowel disease, and other chronic intestinal diseases. Diagn Microbiol Infect Dis. 2004;49:71–4.
20. Weber P, Koch M, Heizmann WR, Scheurlen M, Jenss H, Hartmann F. Microbic superinfection in relapse of inflammatory bowel disease. J Clin Gastroenterol. 1992;14:302–8.
21. Geboes K. Crohn's disease, ulcerative colitis or indeterminate colitis – how important is it to differentiate? Acta Gastroenterol Belg. 2001;64:197–200.
22. Tuohy AM, O'Gorman M, Byington C, Reid B, Jackson WD. *Yersinia* enterocolitis mimicking Crohn's disease in a toddler. Pediatrics. 1999;104:e36.
23. Saebo A, Vik E, Lange OJ, Matuszkiewicz L. Inflammatory bowel disease associated with *Yersinia enterocolitica* O:3 infection. Eur J Intern Med. 2005;16:176–82.

7
Inflammatory bowel disease coexistent with viral hepatitis and HIV

E. A. KOUROUMALIS

INTRODUCTION

Inflammatory bowel disease (IBD) may coexist with viral infections or may be complicated by the appearance of viral infections at a later stage. A further potentially dangerous event is the effect of agents used for treatment of IBD. Viral hepatitis is one of the most common viral infections worldwide. Although many viruses may cause either acute or chronic hepatocellular damage, hepatitic B virus (HBV) and hepatitic C virus (HCV) are the main viral liver infections. On the other hand, human immunodeficiency virus (HIV) might be a potential serious complication of IBD or it might also be influenced by the use of immunosuppressives or biological treatments of IBD. A review of the current status of the influence of HBV, HCV and HIV infections on the management of IBD will be presented in this chapter.

HBV INFECTION AND IBD

There is no evidence that the presence of HBV might influence the natural course of either Crohn's disease (CD) or ulcerative colitis (UC). However, CD patients are possibly at an increased risk of HBV (and HCV) infection due to surgical and endoscopic procedures[1]. Treatment modalities of IBD might potentially influence the natural course of HBV infection.

Corticosteroids

HBV contains a glucocorticosteroid-responsive element that stimulates viral replication and transcriptional activity[2].

As early as 1980 it was shown that administration of corticosteroids significantly increased viral replication, a finding repeatedly confirmed and recently reviewed[3]. Moreover, on corticosteroid withdrawal, flares of HBV hepatitis with increased aminotransferases have been described, and decompensation of cirrhosis has been reported in formerly stable cirrhotic

patients[3–6]. However, the problem may not be so serious in IBD patients infected with HBV, probably due to the low dosage of steroids used[7].

In patients with IBD and HBV co-infection, if corticosteroids are to be used, early antiviral treatment with nucleoside analogues is mandatory. Interferon alpha is unlikely to be as effective because of its relatively slow onset of action and limited antiviral potency[8]. Lamivudine is the drug of choice[9]. Controlled clinical trials of preemptive treatments are not available but are most probably expected to be effective[3]. Adeforir or entecavir are also possible candidates for preemptive treatment but no data are available at present.

Azathioprine

Data so far indicate that administration of azathioprine is safe as there is no effect on underlying liver disease in HBV infection[7]. However, treatment with azathioprine in rheumatoid arthritis was associated with a flare-up of liver disease[10]. Careful monitoring of HBV viral load should therefore be done when azathioprine (and hence 6-mercaptopurine) is being used in IBD with coexistent HBV.

Methotrexate

Evidence comes again from rheumatoid arthritis patients, where a flare-up of HBV infection has been reported on discontinuation of the drug[11–13].

Infliximab

Widespread use of infliximab (and of other anti-tumour necrosis factor alpha (TNF-α) biologicals) in the treatment of patients with IBD requires careful consideration of a possible detrimental effect of this drug on the natural course of an underlying HBV infection. There is strong evidence that TNF-α synergizes with interferons in suppressing HBV replication[14] and its presence is essential in clearing HBV and other viruses[15–19]. Clinical reports are controversial. There have been case studies in which no effect of infliximab on HBV replication was proven[20,21].

In rheumatoid arthritis patients on infliximab, the disappearance of HBV DNA and improvement of renal amyloidosis has recently been reported[22].

In a series of patients with IBD treated with infliximab, the replication of Epstein–Barr virus has recently been studied. No evidence of EBV replication was identified[15,23]. However, case reports of reactivation of HBV in CD treated with infliximab have also been published. Two severe HBV flare- ups have been reported by Esteve et al.[24], and confirmed by later reports[25]. An additional case of HBV reactivation was reported in a CD patient treated with infliximab and methotrexate[26].

Recently a case of subfulminant hepatitis B flare-up was reported in an HBsAg-positive patient with CD. The patient was effectively treated with lamivudine[27]. Additionally, CD patients on lamivudine showed no evidence of HBV reactivation[21], a finding recently verified[28].

Therefore, the current recommendation favours preemptive treatment with lamivudine of HBV carriers with CD before administration of infliximab[24,29,30].

HCV INFECTION AND IBD

HCV infection might be a serious problem in IBD patients. Prevalence of HCV has been reported to be higher in IBD patients in Europe, compared with blood donors[1,31].

Interferon α and IBD

There is limited information on the effect of interferon α used for HCV treatment on the natural course of CD. Evidence suggests that there is no influence on CD disease course in HCV patients treated with interferon α[7]. Moreover, in a controlled trial response rates after interferon monotherapy were similar in HCV patients with or without IBD. It should be noted, however, that all IBD patients were in remission when studied[32].

Corticosteroids

Conflicting results have been reported on the effect of steroids on HCV replication. An increase of HCV viraemia was observed similar to that with HBV infection[33]. Recently, however, bolus administration of steroids in renal transplant patients had no effect on viral load, but it is of interest that the histological activity index was higher than in controls[34].

Azathioprine

As in the case of HBV, azathioprine has no effect on underlying liver disease in HCV chronic hepatitis[7]. Although *in vitro* azathioprine may show antiviral activity against HCV[35], clinical studies in HCV-positive renal transplant patients receiving azathioprine failed to show any effect on HCV viral replication[36].

Cyclosporin

Cyclosporin inhibits HCV replication in a dose-dependent manner and shows additive effects with interferon[37]. However, clinical studies have provided conflicting results. Beneficial effects on viral replication have been both reported[38] and denied[39]. In general, however, existing evidence suggests that cyclosporin can be used without problems in IBD patients with coexistent HCV infection.

Infliximab

Current evidence suggests that TNF-α has a different effect in HCV patients compared to HBV infection. In contrast to HBV, TNF-α might lead to increased necroinflammation in HCV-infected patients[40]. Clinical evidence on the use of infliximab in HCV infection comes from rheumatoid arthritis patients. In the largest published series of 24 patients with rheumatoid arthritis and HCV there was no effect of infliximab treatment on HCV viral load or liver disease[41]. All other series with small numbers of patients also agree[7,42–44], including case reports with IBD.

Recently the use of adalimumab also showed no effect on HCV viral load in HCV rheumatoid arthritis patients[28]. These results have been confirmed in other clinical studies using another anti-TNF-α agent, etarnecept. Although etarnecept is not active in IBD patients, its use in rheumatoid arthritis and HCV infection showed no evidence of either viral replication or exacerbation of liver disease[34,45,46]. Therefore the current recommendation is that anti-TNF-α biologicals can be safely used in IBD patients co-infected with HCV[30].

HIV INFECTION AND IBD

Effect of HIV infection on IBD

There have been several case reports indicating that, when CD^4 lymphocytes drastically drop as a result of HIV infection, there is a dramatic remission of IBD inflammatory activity[47–49]. However, as is usually the case, opposite reports have been published; despite a severe CD^4 drop, active CD persisted and even deteriorated[50,51].

HIV infection mimicking IBD

Diarrhoea and abdominal pain are usual in HIV infection as well as large bowel ulcers as a result of opportunistic infections[52]. Indeed, HIV-associated cytomegalovirus colitis may mimic IBD[57], while *Salmonella*, *Campylobacter* and *Shigella* HIV-associated colitis and ileitis may erroneously be diagnosed as CD[58]. Adenovirus infection is common in HIV-associated diarrhoea as well as in CMV HIV-associated colitis[52]. The opposite may also be true. CD mimicking genital pyoderma gangrenosum in an HIV-infected patient has recently been reported[59]. Moreover, direct HIV lesions appearing as idiopathic ulcers of oesophagus and rectum can lead to confusion of HIV infection with CD[53–55]. Kaposi's sarcoma of the large bowel may be confused with HIV UC[56].

Treatment of concomitant HIV and IBD

Corticosteroids

Corticosteroids may induce Kaposi's sarcoma[60–62] or alternatively Kaposi's sarcoma may be associated with UC usually due to HHV-8 virus[63–65]; therefore corticosteroids are better avoided in IBD patients with HIV.

Infliximab

Elevated TNF-α is found in all stages of HIV infection. Three controlled clinical trials on the use of anti-TNF-α agents, in HIV-infected patients, have been published; they all demonstrated that a significant reduction of serum TNF-α levels was not associated with any effect on CD^4 cells or on plasma HIV RNA levels[66–68].

Recently a case report of concomitant CD and HIV infection treated with infliximab appeared. Complete clinical and endoscopic remission with fistula closure was achieved[69], but HIV infection had been controlled with HAART in this patient. Therefore the question of CD control in active HIV remains unanswered. Further indirect evidence that infliximab is probably safe in HIV infection comes from another study in which infliximab did not activate replication of many lymphotropic herpes viruses, including CMV, HHV-6, HHV-7, HHB-8 and EBV[70].

Thalidomide

This is a drug with potential interest for both HIV infection and CD. Oral aphthous ulcers in HIV-infected patients were effectively treated with oral 200 mg/day thalidomide with 90% complete or partial resolution[71]. The drug was also 73% effective in aphthous ulcers of the oesophagus.

HIV-associated wasting has also favourably responded to thalidomide treatment[72,73].

Thalidomide effectively decreases proinflammatory cytokines[74] and has been used in two open-label studies in refractory CD. There was also a 70% overall response with CDAI decrease with favourable response in fistula remission[75,76]. The subject has recently been reviewed[77].

CONCLUSIONS

Data on the problem of HBV, HV and HIV infection in IBD patients are limited. Most information, particularly on the use of biologicals in these patients, comes from extrapolation of data on rheumatoid arthritis. Based on the current evidence, corticosteroids should be used after preemptive treatment with nucleoside analogues in IBD patients with HBV and used with care in HCV patients, while best avoided in HIV.

Azathioprine, and particularly cyclosporin, are probably safe in HBV and HCV, with cyclosporin even beneficial in these infections. Infliximab should be used with preemptive nucleoside analogues in HBV, while it seems safe in HCV and HIV. Clearly more studies are required.

References

1. Biancon L, Pavia M, Del Vecchio Blanco G et al. and the GISC. Hepatitis B and C virus infection in Crohn's disease. Inflamm Bowel Dis. 2001;7:287–94.
2. Chou CK, Wang LH, Lin HM, Chi CW. Glucocorticoid stimulates hepatitis B viral gene expression in cultured human hepatoma cells. Hepatology. 1992;16:13–18.
3. Perrillo RP. Acute flares in chronic hepatitis B: the natural and unnatural history of an immunologically mediated liver disease. Gastroenterology. 2001;120:1009–22.
4. Sheen IS, Liaw YF, Lin SM, Chu CM. Severe clinical rebound upon withdrawal of corticosteroids before interferon therapy: incidence and risk factors. J Gastroenterol Hepatol. 1996;11:143–7.
5. Buti M, Esteban R, Esteban JI, Gonzales A, Guardia J. Severe hepatic failure after ARA-A prednisolone for chronic type B hepatitis. Gastroenterology. 1987;92:274–75.
6. Laskus T, Slusarczyk J, Cianciara J, Loch T. Exacerbation of chronic active hepatitis B after short term corticosteroid therapy resulting in fatal liver failure. Am J Gastroenterol. 1990; 85:1414–17.
7. Biancone L, DelVecchio Blanco G, Pallone F, Castiglione F, Bresci G, Sturniolo G, Immunomodulatory drugs in Crohn's disease patients with hepatitis B or C virus infection. Gastroenterology. 2002;122:593–4.
8. Khalidi M, Perillo RP. Interferon therapy of hepatitis B. Clin Liver Dis. 1999;3:363–87.
9. Shibolet O, Ilan Y, Gillis S, Hubert A, Shouval D, Safadi R. Lamivudine therapy for prevention of immunosuppressive-induced hepatitis B virus reactivation in hepatitis B surface antigen carriers. Blood. 2002;100:391–6.
10. Mok MY, Ng WL, Yuen MF, Wong RW, Lau CS. Safety of disease modifying anti-rheumatic agents in rheumatoid arthritis patients with chronic viral hepatitis. Clin Exp Rheumatol. 2000;18:368–78.
11. Flowers MA, Healthcote J, Wanless IR et al. Fulminant hepatitis as a consequence of reactivation of hepatitis B virus infection after discontinuation of low-dose methotrexate therapy. Ann Intern Med. 1990;112:381–2.
12. Ito S, Nakazono K, Murasawa A et al. Development of fulminant hepatitis B (precore variant mutant-type) after the discontinuation of low-dose methotrexate therapy in a rheumatoid arthritis patient. Arthritis Rheum. 2001;44:339–42.
13. Narvaez J, Rodriguez-Moreno J, Martinez-Aguila MD, Clavaguera MT. Severe hepatitis linked to B virus infection after withdrawal of low-dose methotrexate therapy. J Rheumatol. 1998;25:2037–8.
14. Kasahara S, Ando K, Saito K et al. Lack of tumour necrosis factor alpha induces impaired proliferation of hepatitis B virus specific T lymphocytes. J Virol. 2003;77:2469–76.
15. Feduchi F, Alonso MA, Carasco L. Human gamma interferon and tumour necrosis factor exert a synergistic blockade on the replication of herpes simplex virus. J Virol. 1989;63: 1354–9.
16. Lucin P, Jonjic S, Messerle M, Polic B, Hengel H, Koszinowski UH. Late phase inhibition of murine cytomegalovirus replication by synergistic action of interferon gamma and tumour nerosis factor. J Gen Virol. 1994;75:101–10.
17. Schlaak JF, Tully G, Lohr HF, Gerken G, Meyer zum Buschenfelde KH. HBV-specific immune defect in chronic hepatitis B is correlated with a dysregulation of pro- and anti-inflammatory cytokines. Clin Exp Immunol. 1999;115:508–14.
18. Wong GH, Goedel DV. Tumour necrosis factor alpha and beta inhibit virus replication and synergize with interferons. Nature. 1986;323:819–22.
19. Guidotti LG, Ishikawa T, Hobbs MV, Matzke B, Schreiber R, Chisari FV. Intracellular inactivation of the hepatitis B virus by cytotoxic T lymphocytes. Immunity. 1996;4:25–36.
20. Oniankitan O, Duvoux C, Challine D et al. Infliximab therapy for rheumatic diseases in patients with chronic hepatitis B or C. J Rheumatol. 2004;31:107–9.
21. del Valle Garcia-Sanchez M, Gomez-Camacho F, Poyato-Gonzalez A, Inglesias-Flores EM, deDios-Vega JF, Sancho-Zapatero R. Infliximab therapy in a patient with Crohn's disease and chronic hepatitis B virus infection. Inflamm Bowel Dis. 2004;10:701–2.
22. Anelli MG, Torres DD, Manno C et al. Improvement of renal function and disappearance of hepatitis B virus DNA in a patient with rheumatoid arthritis and renal amyloidosis following treatment with infliximab. Arthritis Rheum. 2005;52:2519–20.

23. Reijasse D, Le Pendeven C, Cosnes J et al. Epstein–Barr virus viral load in Crohn's disease: effect of immunosuppressive therapy. Inflamm Bowel Dis. 2004;10:85–90.
24. Esteve M, Saro C, Gonzalez-Huix F, Suarez F, Forné M, Viver JM. Chronic hepatitis B reactivation following infliximab therapy in Crohn's disease patients: need for primary prophylaxis. Gut. 2004;53:1363–5.
25. Ueno Y, Shimamoto M, Miyanaka Y et al. Infliximab therapy for Crohn's disease in a patient with chronic hepatitis B. Dig Dis Sci. 2005;50:163–6.
26. Ostuni P, Botsios C, Punzi L, Sfriso P, Todesco S. Hepatitis B reactivation in a chronic hepatitis B surface antigen carrier with rheumatoid arthritis treated with infliximab and low dose methotrexate. Ann Rheum Dis. 2003;62:686–7.
27. Milloning G, Kerm M, Ludwiczek D, Nachbaur K, Vogel W. Subfulminant hepatitis B after infliximab in Crohn's disease: need for HBV screening? World J Gastroenterol. 2006;12: 974–6.
28. Roux CH, Brocq O, Breuil V, Albert C, Euller-Ziegler L. Safety of anti-TNF alpha therapy in rheumatoid arthritis and spondylarthropathies with concurrent B or C chronic hepatitis. Rheumatology. 2006;45:1294–7.
29. Michel M, Duvoux C, Hezode C, Chergui D. Fulminant hepatitis after infliximab in a patient with hepatitis B virus treated for an adult onset Still's disease. J Rheumatol. 2003; 30:1624–5.
30. Desai SB, Furst DE. Problems encountered during anti-tumour necrosis factor therapy. Best Pract Res Clin Rheumatol. 2006;20:757–90.
31. Longo F, Hebuterne X, Tran A et al. Prevalence of hepatitis C in patients with chronic inflammatory bowel disease in the region of Nice and evaluation of risk factors. Gastroenterol Clin Biol. 2000;24:77–81.
32. Bargiggia S, Thorburn D, Anderloni A et al. Is interferon-alpha safe and effective for patients with chronic hepatitis C and inflammatory bowel disease? A case–control study. Aliment Pharmacol Ther. 2005;22:209–15.
33. Magrin S, Craxi A, Fabiano C et al. Hepatitis C viremia in chronic liver disease: relationship to interferon-alpha or corticosteroid treatment. Hepatology. 1994;19:273–9.
34. Barcena R, Oton E, Barreales M, Castillo M, Blesca C. Scarce influence of corticosteroid boluses on long-term viral load and liver histology in transplanted patients with recurrent hepatitis C. Transplant Proc. 2006;38:2502–4.
35. Stangl JR, Carrol KL, Illichmann M, Striker R. Effect of antimetabolite immunosuppresants on Flaviviridae, including hepatitis C virus. Transplantation. 2004;77: 562–7.
36. Melon S, Galarraga MC, Villar M et al. Hepatitis C virus reactivation in anti-hepatitis C virus-positive renal transplant recipients. Transplant Proc. 2005;37:2083–5.
37. Firpi RJ, Zhu H, Morelli G et al. Cyclosporine suppresses hepatitis C virus *in vitro* and increases the chance of sustained virological response after viral liver transplantation. Liver Transplant. 2006;12:51–7.
38. Loho R, Turlin B, de Lajarte-Thirouard AS et al. Improved liver function and decreased hepatitis C viral load after tacrolimus was replaced by cyclosporine. Transplant Proc. 2005; 37:2871–2.
39. Oton E, Barcena R, Castillo M et al. Hepatitis C virus recurrence after liver transplantation: influence of immunosuppressive regimens on viral load and liver histology. Transplant Proc. 2006;38:2499–501.
40. Chuang E, del Vecchio A, Smolinski S, Song XY, Sarisky R. Biomedicines to reduce inflammation but not viral load in chronic HCV – what's the sense? Trends Biotechnol. 2004;22:517–23.
41. Peterson JR, Hsu FC, Simkin PA, Wener MH. Effect of tumour necrosis factor alpha antagonists on serum transaminases and viraemia in patients with rheumatoid arthritis and chronic hepatitis C infection. Ann Rheum Dis. 2003;62:1078–82.
42. Campbell S, Ghosh S. Infliximab therapy for Crohn's disease in the presence of chronic hepatitis C infection. Eur J Gastroenterol Hepatol. 2001;13:191–2.
43. Holtmann MH, Galle PR, Neurath MF. Treatment of patients with Crohn's disease and concomitant chronic hepatitis C with a chimeric monoclonal antibody to TNF. Am J Gastroenterol. 2003;98:504–5.
44. Parke FA, Reveille JD. Anti-tumour necrosis factor agents for rheumatoid arthritis in the setting of chronic hepatitis C infection. Arthritis Rheum. 2004;51:800–4.

45. Magliocco MA, Gottlieb AB. Etarnecept therapy for patients with psoriatic arthritis and concurrent hepatitis C infection: report of 3 cases. J Am Acad Dermatol. 2004;51:580–4.
46. Zein NN. Etarnecept as an adjuvant to interferon and ribavirin in treatment naœve patients with chronic hepatitis C virus infection: a phase 2 randomized double blind placebo controlled study. J Hepatol. 2005;42:315–21.
47. James SP. Remission of Crohn's disease after human immunodeficiency virus infection. Gastroenterology. 1988;95:667–9.
48. Yoshida EM, Chan NH, Herrick RA et al. Human immunodeficiency virus infection, the acquired immunodeficiency syndrome and inflammatory bowel disease. J Clin Gastroenterol. 1996;23:24–8.
49. Pospai D, Rene E, Fiasse R et al. Crohn's disease stable remission after human immunodeficiency virus infection. Dig Dis Sci. 1998;43:412–19.
50. Christ AD, Sieber CC, Cathomas G, Gyr K. Concomitant active Crohn's disease and the acquired immunodeficiency syndrome. Scand J Gastroenterol. 1996;31:733–5.
51. Lantenbach E, Lichtenstein GR. Human immunodeficiency virus infection and Crohn's disease: the role of CD^4 cell in inflammatory bowel disease. J Clin Gastroenterol. 1997;25: 456–9.
52. Orenstein JM, Dieterich DT. The histopathology of 103 consecutive colonoscopy biopsies from 82 symptomatic patients with acquired immunodeficiency syndrome. Arch Pathol Lab Med. 2001;125:1042–6.
53. Kotler DD, Reka S, Orenstein JM et al. Chronic idiopathic esophageal ulceration in the acquired immunodeficiency syndrome. J Clin Gastroenterol. 1992;15:284–5.
54. Wilcox CM, Schwartz DA. Idiopathic anorectal ulceration in patients with human immunodeficiency virus infections. Am J Gastroenterol. 1994;89:599–604.
55. Bach M, Howell DA, Valenti AJ et al. Aphthous ulceration of the gastrointestinal tract in patients with the acquired immunodeficiency syndrome. Ann Intern Med. 1990;112:465–7.
56. Weber JN, Carmichael DJ, Boylston A et al. Kaposi's sarcoma presenting as ulcerative colitis and complicated by toxic megacolon. Gut. 1985;26:295–300.
57. Roskell DE, Hyde GM, Campbell AP et al. HIV associated cytomegalovirus colitis as a mimic of inflammatory bowel disease. Gut. 1995;37:148–50.
58. Nelson MR, Shanson DC, Hawkins DA et al. *Salmonella*, *Campylobacter* and *Shigella* in HIV-seropositive patients. AIDS. 1992;6:1495–8.
59. Aviles-Izquierdo JA, Suarez-Fernandez R, Lazaro-Ochaita P, Longo-Imedio MI. Metastatic Crohn's disease mimicking genital pyoderma gangrenosum in an HIV patient. Acta Derm Venereol. 2005;85:60–2.
60. Frances C. Kaposi's sarcoma after renal transplantation. Nephrol Dial Transplant. 1998; 13:768–73.
61. Trattner A, Hodak E, David M et al. The appearance of Kaposi's sarcoma during corticosteroid therapy. Cancer. 1993;72:1779–83.
62. Wijveen AC, Person H, Bjork S et al. Disseminated Kaposi's sarcoma – full regression after withdrawal of immunosuppressive therapy: report of a case. Transplant Proc. 1987;?:3735–6.
63. Thompson GB, Pemberfon JH, Morris S et al. Kaposi's sarcoma of the colon in a young HIV-negative man with chronic ulcerative colitis. Report of a case. Dis Colon Rectum. 1989;32:73–6.
64. Tedesco M, Benevolo M, Frezza F et al. Colorectal Kaposi's sarcoma in an HIV-negative male in association with ulcerative rectocolitis: a case report. Anticancer Res. 1999;19: 3045–8.
65. Bursics A, Morway K, Abraham K et al. HHV-8 positive, HIV negative disseminated Kaposi's sarcoma complicating steroid dependent ulcerative colitis: a successfully treated case. Gut. 2005;54:1049–50.
66. Walker RE, Spooner KM, Kelly G et al. Inhibition of immunoreactive tumour necrosis factor-alpha by a chimeric antibody in patients infected with human immunodeficiency virus type 1. J Infect Dis. 1996;174:63–8.
67. Sha BE, Valdez H, Gelman RS et al. Effect of etarnecept on interleukin-6, tumour necrosis factor alpha and markers of immune activation in HIV infected subjects receiving interleukin 2. AIDS Res Hum Retroviruses. 2002;18:661–5.

68. Wallis RS, Kyambadde P, Johnson JL et al. A study of safety, immunology, virology and microbiology of adjunctive etarnecept in HIV-1 associated tuberculosis. AIDS. 2004;18: 257–64.
69. Beltran B, Nos P, Bastida B, Iborra M, Hoyos M, Ponce J. Safe and effective application of anti-TNFα in a patient infected with HIV and concomitant Crohn's disease. Gut. 2006;54: 1670–1.
70. Torre-Cisneros J, del Castillo M, Caston JJ, Castro MC, Perez V, Collantes E. Infliximab does not activate replication of lymphotropic herpes viruses in patients with refractory rheumatoid arthritis. Rheumatology. 2005;44:1132–5.
71. Jacobson JM, Spritzler J, Fox L et al., for the National Institute of Allergy and Infectious Diseases AIDS Clinical Trials Group. Thalidomide for the treatment of aphthous ulcers in patients with human immunodeficiency virus infection. J Infect Dis. 1999;180:61–7.
72. Kaplan G, Thomas S, Fieter DS et al. Thalidomide for the treatment of AIDS-associated wasting. AIDS Res Hum Retroviruses. 2000;16:1435–55.
73. Reyes-Teran G, Sierra-Madevo JG, Martinez del Cerro V et al. Effects of thalidomide on HIV-associated wasting syndrome: a randomized double-blind, placebo controlled clinical trial. AIDS. 1996;10:1501–7.
74. Bauditz J, Wedel S, Lochs H. Thalidomide reduces tumour necrosis factor a and interleukin 12 production in patients with active chronic Crohn's disease. Gut. 2002;50:196–200.
75. Ehrenpreis ED, Kane SV, Cohen LB et al. Thalidomide therapy for patients with refractory Crohn's disease: an open label trial. Gastroenterology. 1999;117:1271–7.
76. Vasiliauskas EA, Kam LY, Abreu-Martin MT et al. An open-label pilot study of low-dose thalidomide in chronically active steroid-dependent Crohn's disease. Gastroenterology. 1999;117:1278–87.
77. Matthews SJ, McCoy C. Thalidomide: a review of approved and investigational uses. Clin Ther. 2003;25:342–93.

Section III
Diagnostic standards and developments in imaging

Chair: H HERFARTH and I ROZANES

8
Endoscopic and histological grading in inflammatory bowel disease

G. D'HAENS

INTRODUCTION

In order to allow the clinician to select the optimal therapy for each individual patient with inflammatory bowel disease (IBD), it is important to assess not only the location of the disease, but also its severity. This can be done with endoscopic examinations and with histological assessment of biopsy samples. This chapter will deal with the 'grading of IBD lesions' and its relevance for clinical practice.

CROHN'S DISEASE

Typical lesions in active ileocolonic Crohn's disease (CD) include aphthous ulcerations, deep irregular ulcerations, 'punched-out' ulcers, longitudinal ulcerations, cobblestoning, discontinuous involvement (86%), rectal sparing (25%), luminal narrowing and fistulas[1]. In upper gastrointestinal CD, lesions are invariably accompanied by small bowel and/or colonic disease. Upper gastrointestinal CD occurs according to prospective studies in 17–75% of both symptomatic and asymptomatic patients, in retrospective studies in 0.5–13%[2]. Oral lesions are more frequent than gastroduodenal (1.8–4.5%) and oesophageal lesions (1.8%). In the oesophagus aphthous ulcers, punched-out ulcers, erosions and strictures can occur[3].

Three methods are available to measure the extent and severity of Crohn's lesions in ileal, ileocolonic and colonic CD: the Crohn's Disease Endoscopic Index of Severity (CDEIS), with scores ranging from 0 to 44 (higher scores representing more severe disease)[4], the Simple Endoscopic Index for Crohn's Disease (SES-CD)[5] and the Rutgeerts' score for postoperative recurrence of CD in the neoterminal ileum[6]. This score (i0 to i4) correlates with the clinical behaviour of CD in the future.

With recent drug development it has become apparent that it is now possible to 'heal' mucosal lesions in CD, and this has become a genuine endpoint for the evaluation of drugs in clinical trials. Whereas 5-aminosalicylic acid (5-ASA)

and corticosteroids only induce a limited degree of healing, immunomodulators such as azathioprine and biologic agents such as infliximab have been show to be more effective in this regard[7,8]. More importantly, healing with infliximab has been associated with a significant reduction in hospitalizations and need for surgery[9]. The consensus published by the International Organization of Inflammatory Bowel Diseases (IOIBD) stated that the CDEIS is to be used as a secondary endpoint in studies looking at inflammatory activity and the Rutgeerts' score in studies for postoperative recurrence[10].

ULCERATIVE COLITIS

The typical endoscopic features of ulcerative colitis (UC) include continuous involvement (which can be disturbed by topical treatment), erythema, friability, granularity, microulcerations, shallow ulcerations and sometimes the presence of an isolated area of inflammation around the appendiceal orifice, a so-called 'caecal patch'[1,11].

In quiescent UC, on the other hand, typical lesions are an attenuated vascular pattern or loss of vascular pattern, pseudopolyps and mucosal bridging and rarely stricture formation.

In UC a full ileocolonoscopy is to be recommended at diagnosis, whereas a flexible sigmoidoscopy is sufficient for follow-up. No bowel preparation is needed in patients with active symptoms. Caution is needed in fulminant colitis, since the endoscopy can induce a toxic megacolon. The presence of deep ulcers in spite of anti-inflammatory therapy is a poor prognostic sign. Also, it is recommended to take biopsies at relapse in order to exclude cytomeglovirus, *Clostridium difficile* or other infections[12].

Several endoscopic scores have been developed for the evaluation of drug effects in active UC. They are useful since endoscopic improvement lags behind symptom improvement and endoscopic healing is an endpoint that is aimed at.

The Baron Endoscopic score for UC includes the following activity variables: 0 = normal: mat mucosa, ramifying vascular pattern clearly visible throughout, no bleeding spontaneous or to light touch; 1 = abnormal but not haemorrhagic (between 0 and 2); 2 = moderately haemorrhagic: bleeding to light touch but no spontaneous bleeding; 3 = severely haemorrhagic; 4 = spontaneous bleeding. The problem is that the Baron score does not include a description of 'ulcers'. The interobserver variation is the highest for 'graded' variables (e.g. 'redness'). In fact, the score was developed for mild and moderate cases, with best agreement for friability (bleeding to light touch) and spontaneous bleeding and the lowest agreement for granularity[13].

Mucosal healing is indeed an important endpoint in UC: active UC is associated with a higher likelihood of dysplasia/cancer. In addition, endoscopy correlates well with histology and 'histological healing' predicts a longer 'time to relapse'. In the ACT trials, patients had a 4-fold higher likelihood of clinical remission at week 30 (43.8% vs 9.5%) if healing (score 0) was present at week 8[14].

DIFFERENTIAL DIAGNOSIS

The differential diagnosis between the two classic types of IBD is in most patients rather easy. Crohn's disease is characterized by cobblestoning, aphthous ulcers, deep serpiginous ulcers, rectal sparing, anal lesions and stenosis or ulceration of the ileocaecal valve. To the contrary, UC typically has a continuous involvement, erosions/microulcerations, loss of vascular pattern, rectal involvement in practically all cases and an ileocaecal valve that is rather patulous and free of ulceration[15]. However, in approximately 10% of IBD patients this distinction cannot be made at the time of diagnosis.

HISTOLOGY

As stated above, CD can affect the entire gastrointestinal tract. The histopathological features, however, are similar in the different intestinal segments. Even in areas that appear normal on endoscopy, signs of inflammation can be present. Since CD is a transmural disease, mucosal biopsies do not necessarily reflect the severity of inflammation in the entire bowel wall. Deeply situated lesions are often detected only in surgical resection specimens[16,17].

Early lesions of CD occur in a background of normal mucosa. These 'focal lesions' can have different appearances: 'summit lesions', with damaged small capillaries and loss of epithelial cells, epithelial patchy necrosis, mucosal microulcerations (loss of up to six cells), aphthoid ulcers and mountain peak ulcer (located at the base of crypts)[18].

Established Crohn's lesions consist of epithelial alterations and inflammatory changes. Epithelial damage includes cytological changes of damage and repair, architectural changes and metaplastic changes. The inflammatory response can be characterized based upon intensity, composition and distribution of the infiltrate. The correlation between histological changes and clinical symptoms or response to treatment in CD is rather poor, although anti-TNF therapy has been associated with disappearance of neutrophilic leukocytes[8]. For studies such as the anti-tumour necrosis factor (TNF) study, a histological severity score was developed but not yet validated. Nonetheless, histology is recommended for exploratory studies with new therapeutic agents[10].

In UC, unlike in Crohn's disease, UC endoscopic (and histological) scores are absolutely recommended in the assessment of disease activity and effects of drug therapy. It was clearly demonstrated by Riley et al. that in patients with histological remission (no neutrophils present in the biopsies) the likelihood for clinical relapse was significantly reduced[19].

CONCLUSION

In conclusion, the use of endoscopic and to a certain extent also histological assessment has entered clinical trials and gradually also daily clinical practice. In UC, the endoscopic appearance is of paramount importance to guide therapeutic interventions. In CD it remains to be demonstrated whether endoscopic healing should become a genuine goal of treatment.

References

1. Pera A, Bellando P, Caldera D. Colonoscopy in inflammatory bowel disease. Diagnostic accuracy and proposal of an endoscopic index. Gastroenterology. 1987;92:181–5.
2. Witte AMC, Veenendaal RA, van Hogezand RA, Verspaget HW, Lamers CBHW. Crohn's disease of the upper gastrointetinal tract: the value of endoscopic examination. Scand J Gastroenterol. 1998;225:S100–5.
3. D'Haens G, Rutgeerts P, Geboes K. Esophageal Crohn's disease: three patterns of evolution. Gastrointest Endosc. 1994;40:296–300.
4. Mary JY, Modigliani R; Groupe d'Etudes Thérapeutiques des affections inflamatoires du tube digestif (GETAID). Development and validation of an endoscopic index of the severity for Crohn's disease: a prospective multicenter study. Gut. 1989;30:983–9.
5. Daperno M, D'Haens G, Van Assche G et al. Development and validation of a new, simplified endoscopic activity score for Crohn's disease: the SES-CD. Gastrointest Endosc. 2004;60:505–12.
6. Rutgeerts P, Geboes K, Vantrappen G et al. Predictability of the postoperative course of Crohn's disease. Gastroenterology. 1990;99:956–62.
7. D'Haens G, Geboes K, Rutgeerts P. Endoscopic and histologic healing of Crohn's ileocolitis with azathioprine. Gastrointest Endosc. 1999;50:667–71.
8. D'Haens G, van Deventer S, Van Hogezand R et al. Endoscopic and histological healing with infliximab antibodies in Crohn's disease: a European multicenter trial. Gastroenterology. 1999;116:1029–34.
9. Rutgeerts P, Diamond RH, Bala M et al. Scheduled maintenance treatment with infliximab is superior to episodic treatment for the healing of mucosal ulceration associated with Crohn's disease. Gastrointest Endosc. 2006;63:433–42.
10. Sandborn WJ, Feagan BG, Hanauer SB et al. A review of activity indices and efficacy endpoints for clinical trials of medical therapy in adults with Crohn's disease. Gastroenterology. 2002;122:512–30.
11. D'Haens G, Geboes K, Peeters M, Baert F, Ectors N, Rutgeerts P. Patchy cecal inflammation associated with distal ulcerative colitis: a prospective endoscopic study. Am J Gastroenterol. 1997;92:1275–9.
12. D'Haens G, Sandborn WJ, Feagan BG et al. A review of activity indices and efficacy end points for clinical trials of medical therapy in adults with ulcerative colitis. Gastroenterology. 2007;132:763–86.
13. Baron JH, Connell AM, Lennard-Jones JE. Variation between observers in describing mucosal appearances in proctocolitis. Br Med J. 1964;1:89–92.
14. Rutgeerts P, Sandborn WJ, Feagan BG et al. Infliximab for induction and maintenance therapy for ulcerative colitis. N Engl J Med. 2005;353:2462–76.
15. Waye JD. The role of colonoscopy in the differential diagnosis of inflammatory bowel disease. Gastrointest Endosc. 1977;23:150–4.
16. Rotterdam H, Korelitz BI, Sommers SC. Microgranulomas in grossly normal rectal mucosa in Crohn's disease. Am J Clin Pathol. 1977;67:550–4.
17. Potzi R, Walgram M, Klozner H, Gangl A. Diagnostic significance of endoscopic biopsy in Crohn's disease. Endoscopy. 1989;21:60–2.
18. Rutgeerts P, Geboes K. Crohn's disease and pre-aphthoid lesions. Lancet. 1993;341:1443–4.
19. Riley SA, Mani V, Goodman MJ et al. Microscopic activity in ulcerative colitis: what does it mean? Gut. 1991;32:174–8.

9
Conventional imaging

U. KORMAN

INTRODUCTION

Between 1994 and 2006 we performed 4000 conventional enteroclysis (CE) procedures to achieve primary radiological diagnosis of different neoplastic or non-neoplastic small bowel diseases. In our series 340 (34%) of the last 1000 pathological cases were inflammatory bowel disease (IBD). After cases of partial intestinal obstructions by adhesions, this was a second-largest pathological group. Crohn's disease (CD) was the most common IBD.

CD is an ongoing disorder that causes inflammation of the gastrointestinal (GI) tract. CD can affect any area of the GI tract, from the mouth to anus, but it most commonly affects the lower part of the small bowel; for example, 28% of all CD cases involve the terminal ileum alone; 35% of all cases involve ileum and large bowel; 1–4% stomach and duodenum; and 0.2% of cases affect the oesophagus[1,2].

In recent years there have been important improvements in different technologies and procedures to evaluate small bowel diseases. Despite developed endoscopic procedures a radiological work-up is still attempted first. For initial examination, including the evaluation of mucosal pathologies, morphological changes, luminal and fuctional abnormalities, conventional enteroclysis (CE) is the gold standard[3–6].

DIAGNOSTIC ROLES OF CE

The main role of CE is the determination of subtypes of the disease. The disease is characterized by inflammatory, fistulizing and stenosing (Figure 1) types. Periodical assessment of inflammatory activity influences treatment planning because it predicts the outcome of the surgical intervention and the results of medical treatment. Stenosing disease usually benefits from surgical therapy, the inflammatory disease from medical treatment, and fistulizing disease from both treatments[7,8].

The other important role of CE is determination of the stage of disease (Figure 2). A classification system is presented that is based upon the degree of involvement. Stage 1 lesions are early lesions including fold thickening,

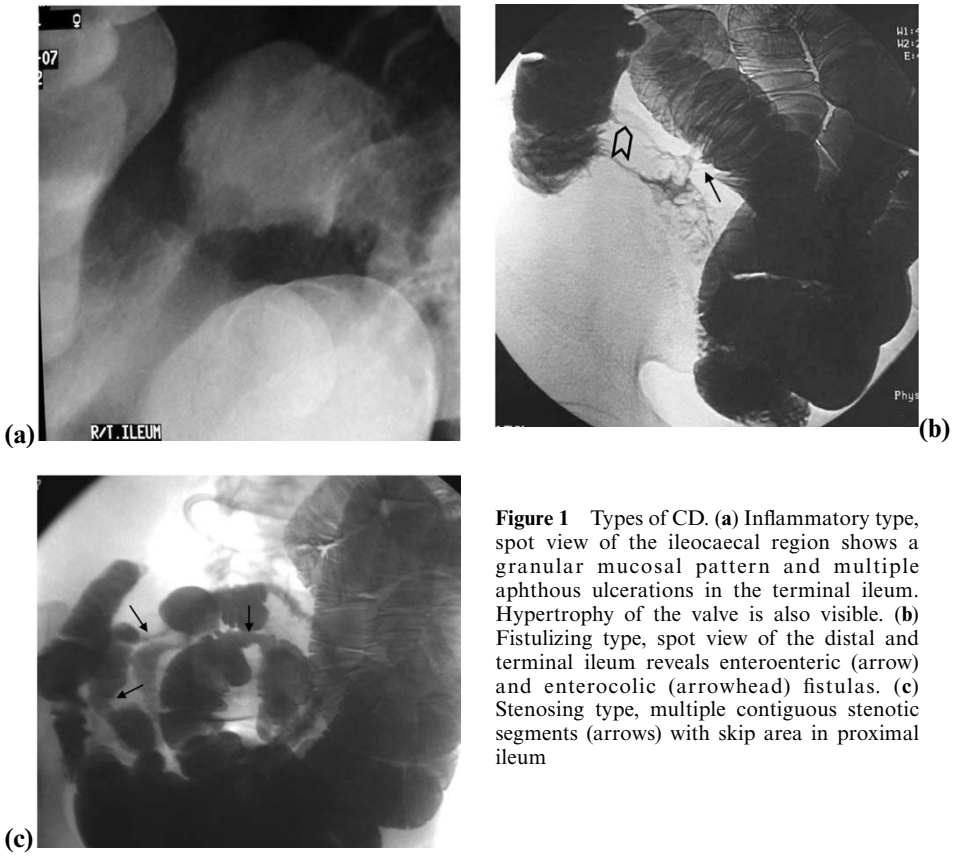

Figure 1 Types of CD. (**a**) Inflammatory type, spot view of the ileocaecal region shows a granular mucosal pattern and multiple aphthous ulcerations in the terminal ileum. Hypertrophy of the valve is also visible. (**b**) Fistulizing type, spot view of the distal and terminal ileum reveals enteroenteric (arrow) and enterocolic (arrowhead) fistulas. (**c**) Stenosing type, multiple contiguous stenotic segments (arrows) with skip area in proximal ileum

aphthous ulcerations, and coarse granularity of the villi. The small bowel wall retains normal contractility. Stage 2 lesions consist of intermediate lesions including nodular fold thickening, mesenteric border ulceration, shortening and rigidity with scalloping of the uninvolved antimesenteric border. The bowel wall is moderately thickened. Stage 3 lesions include advanced stages of transmural disease indicating the ulceronodular pattern in a stiffened and narrowed segment with more pronounced wall thickening and the possible development of stricture[2,8,9].

Depicting the length of involvement and skip areas (Figure 3) is another role of CE which helps to determine treatment. It is generally agreed that the disease remains confined to that length of bowel shown to have been involved at the time of the initial radiological examination[1,2,8,9].

Stenosis of the small bowel lumen occurs frequently in CD, and results from a combination of fibrosis, inflammatory thickening, and spasm. Luminal

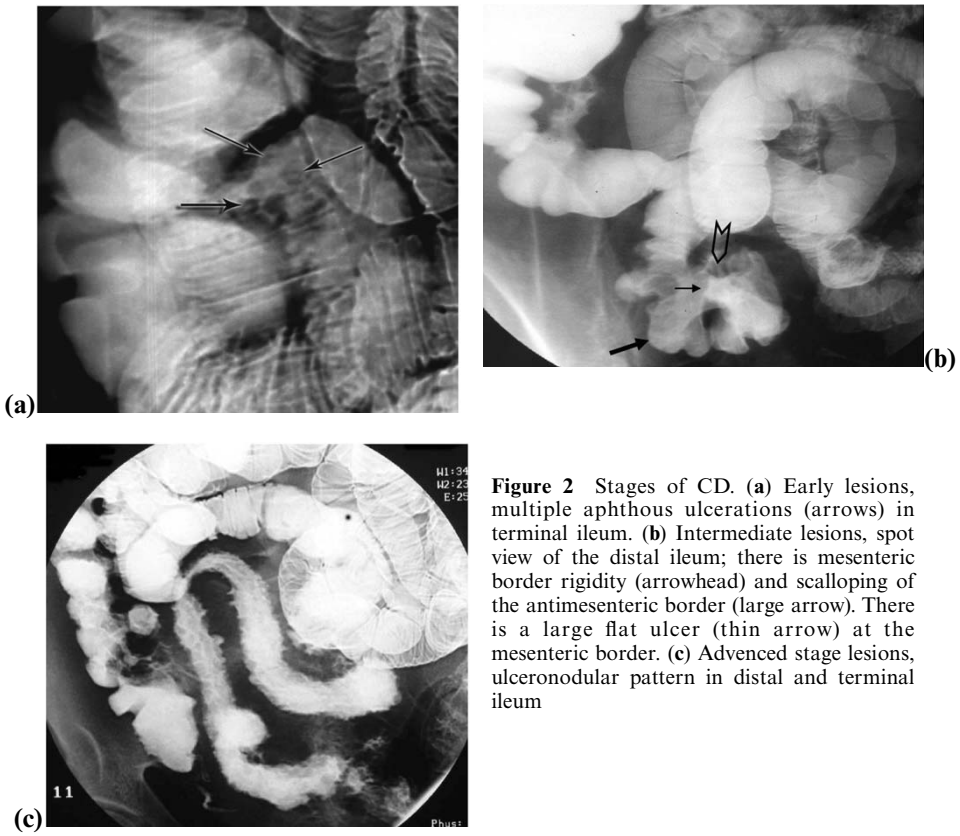

Figure 2 Stages of CD. (**a**) Early lesions, multiple aphthous ulcerations (arrows) in terminal ileum. (**b**) Intermediate lesions, spot view of the distal ileum; there is mesenteric border rigidity (arrowhead) and scalloping of the antimesenteric border (large arrow). There is a large flat ulcer (thin arrow) at the mesenteric border. (**c**) Advenced stage lesions, ulceronodular pattern in distal and terminal ileum

narrowing can be manifested by the 'string sign' that represents intense irritability and spasm usually associated with extensive ulceration[9]. The organic reality of the string sign is best revealed during the lumen-distending phase of CE. The ability of CE to challenge the distensibility of the intestinal wall may make it possible to confidently demonstrate the unchanging narrowed pattern of a true fibrous stricture.

Motor disturbances, either hypermotility or hypomotility, are results of the inflammatory process and can be evaluated by CE.

Complications of CD cover a wide spectrum including strictures, abscesses or phlegmons, fistulas, perforations, Crohn's carcinoma, and giant fibroid polyp. Small bowel obstruction (Figure 4) is a frequent problem once the disease advances into the stenotic stage. High-grade obstruction is unusual, even in the presence of multiple strictures. If surgery is needed because of recurrent obstructive episodes and inability to discontinue steroid therapy, preoperative radiological assessment of the extent of disease can serve as a useful road map for the surgeon. Equally relevant is the demonstration or exclusion of more proximal skip lesions. Both purposes are optimally served by

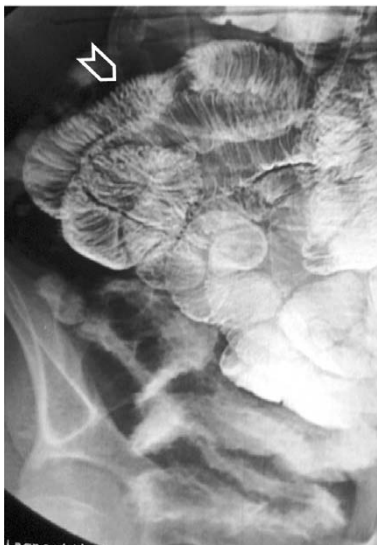

Figure 3 Long segment involvement and skip area. CE shows ulceronodular pattern at a long segment of distal and terminal ileum. There is also a skip area (arrowhead) at the distal jejunum identified by thickened plicaes and aphthous ulcerations

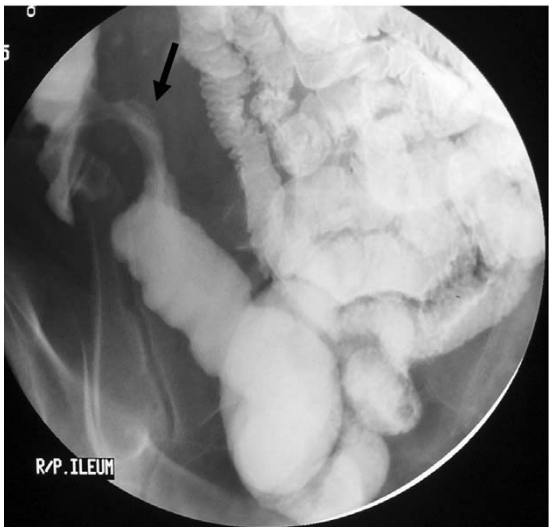

Figure 4 Small bowel obstruction. CE demonstrates a tight stricture in the terminal ileum (arrow), associated with significant obstruction

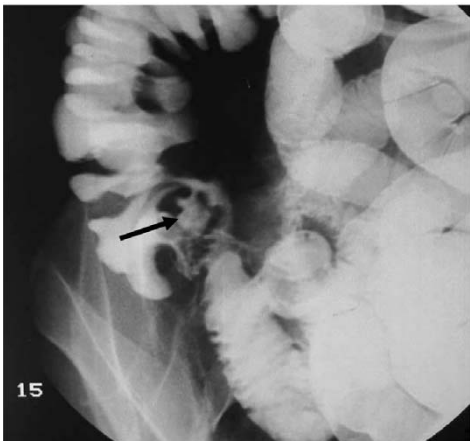

Figure 5 Abscess formation. CE depicts the paracaecal abscess cavity (arrow) filling from the terminal ileum

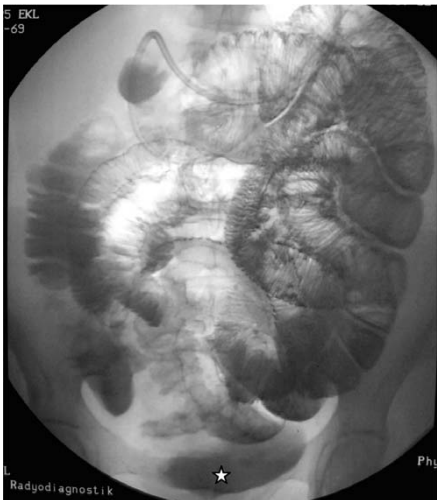

Figure 6 Ileovesical fistulas. CE visualizes advanced-stage findings of CD in the distal ileum adjacent to the bladder, also filling of the bladder (*) with contrast material

CE[9,10]. Abscess formation (Figure 5) occurs in approximately 15–20% of patients with CD. Abscesses are often a consequence of transmural disease and subsequent sinus tracts, but may also occur as a postsurgical complication. Transmural extension of fissures or ulcers is responsible for the formation of fistulas and sinus tracts. Fistulas (Figure 6) and sinus tracts are highly characteristic of advanced CD and are often multiple[8]. Deep fissures usually reach the serosal surface slowly enough to permit walling off by adhesions to a neighbouring viscus, or by the parietal peritoneum or omentum, thus

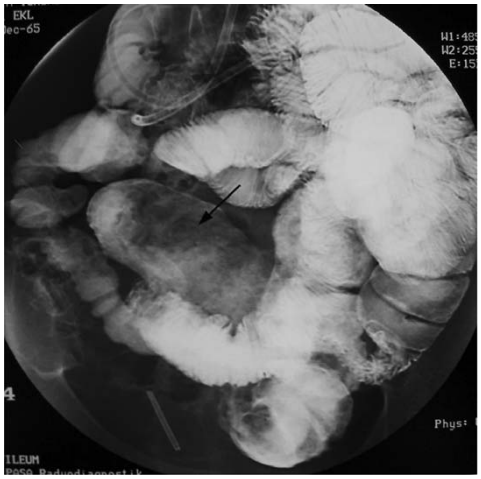

Figure 7 Closed loop perforation due to CD. CE depicts a large and well-outlined cavity (arrow) filling from the inflamed distal ileal segment

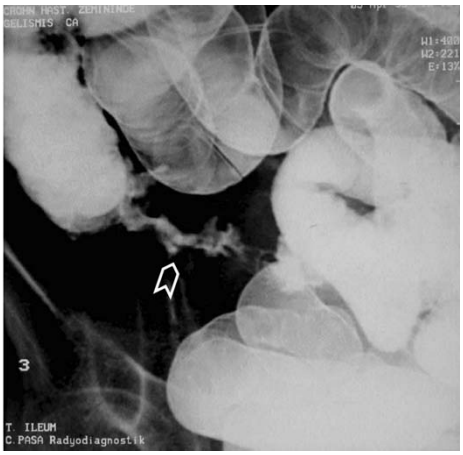

Figure 8 Crohn's carcinoma. A patient with long-standing CD. Spot view of the ileocaecal region demonstrates highly narrowed terminal ileum (arrowhead) with irregular borders

preventing a free perforation (Figure 7). Perforation may be secondary to a more acute penetration of the bowel wall by ulceration, fissuring, necrosis, or the rupture of an intra-abdominal abscess[8,9]. The rarity of small bowel adenocarcinoma complicating small bowel CD (Figure 8) makes it impossible to assess the exact magnitude of the risk. Although no exact numbers are available, it is evident that, when compared with the general population, an increased risk of small bowel cancer exists in CD of many years duration, especially situated in bypassed bowel loops[11]. Giant fibroid polyp (Figure 9) is a rare complication of CD and can be detected by CE[12].

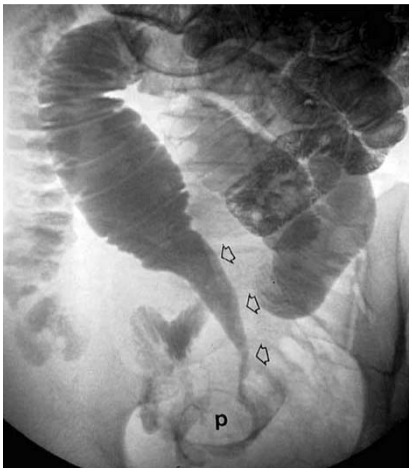

Figure 9 Rare complication of CD: giant fibroid polyp. CE shows intussusception in the distal ileum due to a large polyp. There is a beak-shaped tapering of intussusception (arrowheads) and a giant polyp (p) just adjacent to its distal end

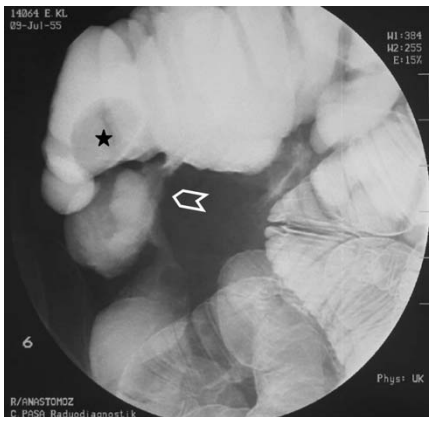

Figure 10 Recurrence after surgical treatment. Spot view reveals recurrence in both sides of the anastomosis; a stricture in the neoterminal ileum (arrowhead), and an inflammatory polyp (*) in the colonic side

CD is clinically characterized by periods of exacerbation separated by remissions. CE has an important role for follow-up evaluations of the disease. The majority of cases show progression. Surgical intervention in CD carries the likelihood of recurrence with extension into previously uninvolved areas. The early radiographic findings in recurrent disease of the neoterminal ileum are diffuse fold thickening and aphthous ulcerations. Recurrences mostly involve the small bowel immediately proximal to the anastomosis with the colon (Figure 10). On the other hand it is possible to evaluate feasible postsurgical complications by CE[2,8,9].

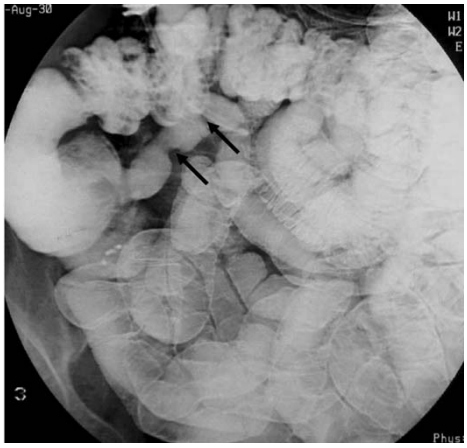

Figure 11 Intestinal tuberculosis. CE shows two contiguous hourglass deformities (arrows) in the distal and terminal ileum. The configuration of the ascending colon and caecum is dismorphic due to asymmetric narrowing

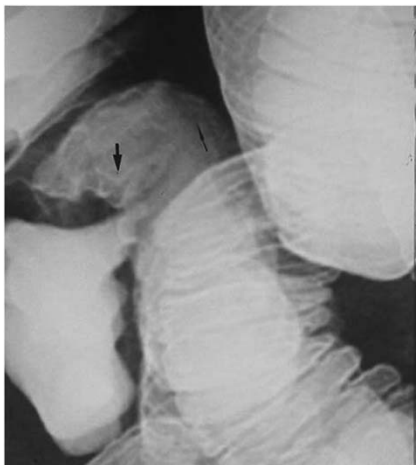

Figure 12 Behçet's disease. Spot view of the terminal ileum; aphthous (thin arrow) and linear (large arrow) ulcerations. Similar findings with early stage of CE

Some other disorders can clinically and radiologically mimic CD. In intestinal tuberculosis, single or multiple short concentric strictures and prestenotic dilations, 'hour-glass appearance' (Figure 11), are characteristic findings on CE. Asymmetric caecal narrowing and ascending colon involvement are other findings of intestinal tuberculosis[13,14]. The rate of GI involvement in Behçet's disease varies in different populations, being more common in Japan (50–60%), and less common in Mediterranean countries (0–5%). Behçet's disease manifests as ulcers prominent at the terminal ileum and

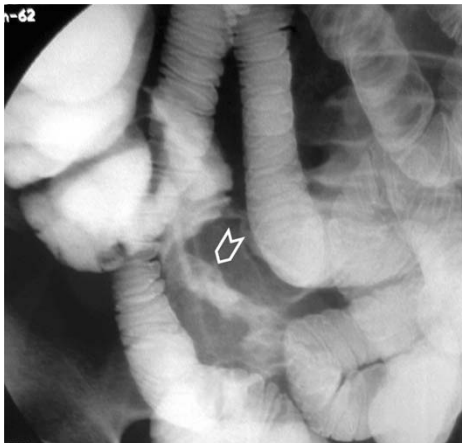

Figure 13 An infiltrative form of intestinal lymphoma mimicking stenosing CD. CE shows a short stenotic ileal segment (arrowhead) adjacent to the terminal ileum

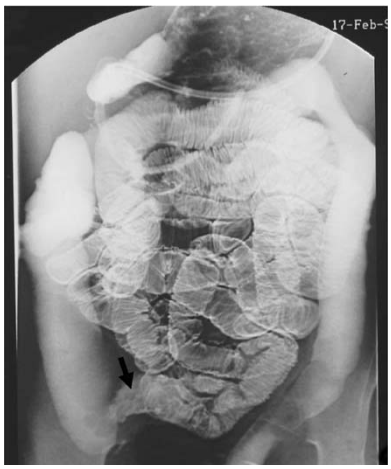

Figure 14 Backwash ileitis. CE demonstrates terminal ileitis (arrow) and pancolitis

ileocaecal region like CD (Figure 12). The EC findings in Behçet's disease are usually mild when compared with those seen in CD[15]. Infectious ileitis can radiologically mimic non-stenotic CD. The radiological features are mucosal fold thickening; nodular defects; mucosal ulcerations varying from aphthous ulcers to larger, oval, or longitudinal shapes; and the terminal ileum as the favoured site of involvement[16]. The infiltrative form of lymphoma (Figure 13) can occasionally mimic stenotic CD. Deep ulceration, excavation into the mesentery, skip lesions, and frequent involvement of the terminal ileum are shared by two conditions. Lymphoma lacks the hyperperistalsis and segmental

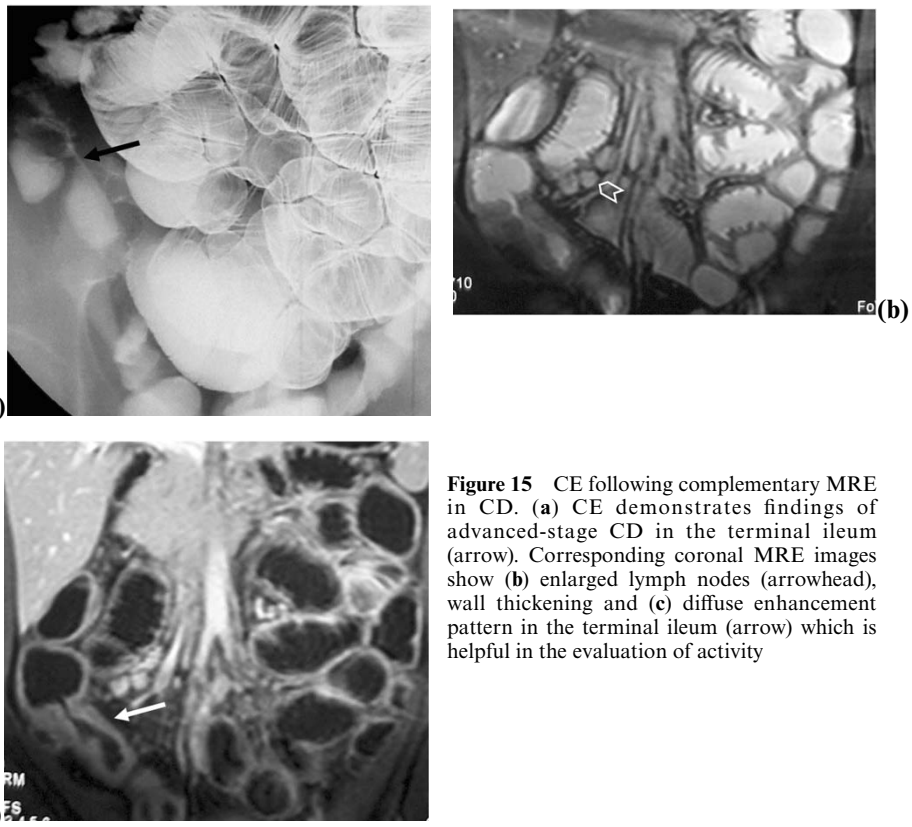

Figure 15 CE following complementary MRE in CD. (**a**) CE demonstrates findings of advanced-stage CD in the terminal ileum (arrow). Corresponding coronal MRE images show (**b**) enlarged lymph nodes (arrowhead), wall thickening and (**c**) diffuse enhancement pattern in the terminal ileum (arrow) which is helpful in the evaluation of activity

spasm of CD and rarely causes stenosis[8]. Ischaemia of the small intestine, if shown by barium study, can closely resemble CD. However, rapid changes in appearance are the hallmark of ischaemia. Miscellaneous conditions such as back-wash ileitis of ulcerative colitis (Figure 14), radiation enteropathy, metastatic disease of mesenteric borders and the ileocaecal area, adenocarcinoma of the ileum, appendiceal abscesses or pelvic inflammatory disease may also mimic CD[8,9,17]. It should be reassuring to know that, in the great majority of patients, the differential diagnosis of CD is not difficult. Repeat CE, examination of the colon, magnetic resonance imaging, computed tomography, or endoscopy may all have to be employed to reach a definitive diagnosis in an unusual situation.

Although CE provides indirect findings concerning the wall and perienteric structures, complementary imaging is often needed in cases in which the pathological changes go beyond the small bowel wall[18]. CE provides mandatory optimal luminal dilation and mural distension for following complementary magnetic resonance enteroclysis (MRE). CE provides information about mucosal pathologies, morphological changes, and

functional abnormalities[19]. On the other hand MRE provides complementary data about mural and perivisceral pathologies, and also activation[19,20] (Figure 15). All this information and complementary data compose all components of CD.

CONCLUSIONS

In diagnostic procedures the small bowel is the most difficult part of the GI tract. Despite recent important technological improvements, endoscopic evaluation of small bowel has some limitations. CD is the most common IBD affecting the small bowel. Like clinical findings, CE findings are also numerous, and depend upon the stage and type of the disease. CE can detect all the main mucosal changes, the inner profile of the wall and also functional disturbances of CD. In CD the sensitivity of CE is 85.4% and the specificity is 76.9%. In initial diagnostic imaging CE is still the gold standard. CE findings answer five main questions important for a therapeutic approach:

1. Type of CD
2. Stage of CD
3. Length of involvement
4. Complications
5. Differential diagnosis.

In follow-up of CD we recommend CE to confirm clinical suspicion of:

1. Progression
2. Recurrence
3. Postsurgical complications.

Conversely CE provides indirect findings about pathological changes of outer wall layers and perienteric tissues. We recommend a combination of CE with complementary MRE in advanced CD cases to depict:

1. Complications
2. Mural and extramural extension
3. Activation of the CD.

References

1. Engelholm L, De Toeuf J, Herlinger H, Maglinte DD. Crohn's disease of the small bowel. In: Herlinger H, Maglinte DD, editors. Clinical Radiology of the Small Intestine. Philadelphia, Saunders, 1989:295–34.

2. Reeders JWA, Derchi LE. Radiology of Crohn's disease. In: Gourtsoyiannis NC, editor. Radiological Imaging of the Small Intestine. Berlin: Springer, 2002:299–39.
3. Liangpunsakul S, Chaladalawada V, Rex DK, Maglinte D, Lappas J. Wireless capsule endoscopy detects small bowel ulcers in patients with normal results from state of the art enteroclysis. Am J Gastroenterol. 2003;98:1295–8.
4. Hara AK, Leighton JA, Sharma VK, Fleisher DE. Small bowel: preliminary comparison of capsule endoscopy with barium study and CT. Radiology. 2004;230:260–5.
5. Hara AK. Capsule endoscopy: the end of the barium small bowel examination? Abdom Imaging. 2005;30:179–83.
6. Crillo LC, Camera L, Della Noce M et al. Accuracy of enteroclysis in Crohn's disease of the small bowel: a retrospective study. Eur Radiol. 2000;10:1894–98.
7. Sachar DB, Andrew HA, Farmer RG et al. Proposed classification of patients subgroups in Crohn's disease. Gastroenterology. 1992;5:141–54.
8. Frederick MK, Herlinger H. Crohn's disease. In: Herlinger H, Maglinte DT, Birnbaum BA, editors. Clinical Imaging of Small Intestine. New York: Springer-Verlag, 1999:259–91.
9. Herlinger H, Caroline DF. Crohn's disease of the small bowel. In: Gore RM, Levine MS, editors. Textbook of Gastrointestinal Radiology, 2nd edn. Philadelphia: Saunders, 2000: 726–46.
10. Maglinte DDT, Chernish SM, Kelvin FM et al. Crohn's disease of the small intestine: accuracy and relevance of enteroclysis. Radiology. 1992;184:541–45.
11. Shenderey RL, Thomson N, Mansfield JC, Rees C. Adenocarcinoma as a complication of small bowel Crohn's disease. Eur J Gastroenterol Hepatol. 2005;17:1255–7.
12. Korman U, Kuruoglu S, Haider S. Rare complication of intestinal Crohn's disease: giant fibroid polyp. Eur Radiol. 2000;10:435–37.
13. Engin G, Balk E. Imaging findings of intestinal tuberculosis. J Comput Assist Tomogr. 2005;29:37–41.
14. Vanhoenacker FM, De Backer AI, Op de BB et al. Imaging of gastrointestinal and abdominal tuberculosis. Eur Radiol. 2004;14(Suppl. 3):E103–15.
15. Korman U, Cantasdemir M, Kuruoglu S et al. Enteroclysis findings of intestinal Behçet disease: a comparative study with Crohn disease. Abdom Imaging. 2003;28:308–12.
16. Tuohy AM, O'Gorman M, Byington C et al. *Yersinia* enterocolitis mimicking Crohn's disease in a toddler. Pediatrics. 1999;104:36–9.
17. Nolan DJ. Enteroclysis of non-neoplastic disorders of small intestine. Eur Radiol. 2000;10: 342–53.
18. Maglinte DD. Small bowel imaging: a rapid changing field and a challenge to radiology. Eur Radiol. 2006;16:967–71.
19. Korman U, Kuruoglu S, Ogut G. Conventional enteroclysis with complementary MR enteroclysis: a combination of small bowel imaging. Abdom Imaging. 2005;30:1–13.
20. Koh DM, Miao Y, Chinn RJS et al. MR imaging evaluation of the activity of Crohn's disease. Am J Roentgenol. 2001;177:1325–32.

Section IV
Cases and controversies

Chair: D RACHMILEWITZ and N TÖZÜN

10
State-of-the-Art Lecture: What is the role of surgery for inflammatory bowel disease – State of the art 2007

B. SINGH and N. J. McC. MORTENSEN

INTRODUCTION

This chapter highlights some of the recent advances in surgery in the management of patients with inflammatory bowel disease (IBD).

ILEAL POUCH–ANAL ANASTOMOSIS

Whilst ileal pouch–anal anastomosis (IPAA) has become a well-established procedure for patients with ulcerative colitis (UC), who have undergone a colectomy and wish to avoid a permanent stoma, it is associated with significant morbidity. The number of pouches performed worldwide has increased to over 30 000 since the seminal paper which introduced this concept by Parks and Nicholls in 1978[1].

Several points have been addressed in recent years regarding the techniques involved in pouch surgery. Studies have shown that there is no difference between close rectal dissection and total mesorectal dissection with regard to sexual dysfunction following pelvic dissection to remove the rectal stump prior to pouch formation[2].

This procedure has traditionally been performed by the open technique; however, two centres have reported their own experience with laparoscopic surgery for IPAA. Larson et al. compared 100 laparoscopic IPAA with 200 case-matched open IPAA[3]. The laparoscopic procedure resulted in a reduced hospital stay, opiate usage and return to normal diet. In the follow-up period of 90 days there was no difference in readmission rates between the two groups but the re-operation rate was significantly higher in the open group. Interestingly 25% of the laparoscopic procedures were hand-assisted. However, a disadvantage of the laparoscopic procedure is the longer

operation time which does reduce with surgical experience. Similar results have also been reported by Ouaissi et al.[4] In this smaller study the authors highlighted the fact that laparoscopic colectomy could safely be performed in patients with severe colitis. An IPAA was subsequently undertaken once the patient had recovered from the acute episode and allowing time to optimize nutrition. A meta-analysis looking at 168 patients who underwent a laparoscopic IPAA also showed a shorter hospital stay, but a similar incidence of postoperative complications compared to the open procedure[5].

CUFFITIS

The region of epithelium between the columnar epithelium of the colon and the squamous epithelium of the anal canal is referred to as the anal transitional zone (ATZ). This region is composed of transitional epithelium. In patients undergoing an IPAA two surgical techniques are commonly employed. The first is a handsewn technique in which a mucosectomy is performed to remove the ATZ and columnar epithelium to allow a handsewn IPAA to be performed. Alternatively the ATZ and a cuff of columnar mucosa are retained to enable a double-stapled IPAA to be performed. The latter is far and away the most commonly performed procedure for IPAA. Advocates of the double-stapled technique argue that the ATZ has an important sensory function and its preservation is important for pouch function. Furthermore preservation of the ATZ and the columnar cuff requires minimal disruption to anal anatomy compared to the retraction required to perform a mucosectomy. This is also thought to lead to better pouch function and to reduce the risk of pouch sepsis.

However, an obvious disadvantage of a columnar cuff is that it retains diseased mucosa and can therefore be a source of subsequent inflammation (cuffitis) or cancer change. A mucosectomy is therefore advocated in patients with ulcerative colitis and a coexisting rectal carcinoma or underlying dysplasia, as well as in patients with familial adenomatous polyposis. However, these indications are relative and an argument can be made for long-term surveillance of the columnar cuff with the advantage of better functional results.

Cuffitis commonly presents with frank bleeding[6] It can be also be associated with extraintestinal manifestations of IBD. A scoring system has been proposed by Thompson-Fawcett et al.[7] for the histological appearances of cuffitis; however, a more detailed scoring system, the Cuffitis Activity Index, combines symptoms, histology and endoscopic appearances[8]. The diagnosis can be made in the clinic or at endoscopy with characteristic inflammation confined to the columnar cuff. A biopsy will be able to give histological confirmation. Treatment for cuffitis is local administration of a 5-aminosalicylate compound or steroids[8]. In contrast to pouchitis inflammation does not respond to antibiotic treatment. The long-term complications of cuffitis are stricture formation and, in theory, dysplasia. If medical therapy fails then a mucosectomy can be performed with pouch advancement to create a new IPAA. The latter technique has been used in patients with dysplasia in the columnar cuff [9].

Although it would seem that cuffitis is a consequence of the double-stapled technique, which retains a cuff of diseased columnar epithelium, cuffitis has been described in patients who have had a mucosectomy. Heppell et al.[10] reported small islets of rectal mucosa in two out of eight patients who underwent a mucosectomy. Whilst an incomplete mucosectomy is a possibility they believed regeneration of rectal mucosa to be the most likely cause. Cuffitis is more likely to occur in patients with a double-stapled IPAA compared to patients who have had a mucosectomy. Regeneration of rectal or an incomplete mucosectomy can also predispose to cuffitis, though with a much lower incidence.

TERMINAL ILEAL CROHN'S DISEASE

Whilst laparoscopic surgery has not been a new development it has begun to play an important role in the management of patients with IBD. This technique offers several advantages compared to open surgery. First the operative technique involves less manipulation of bowel, which is believed to lead to a reduced postoperative ileus with early resumption of a normal diet. Furthermore, whilst laparoscopic procedures certainly offer a better cosmetic result, a smaller incision also leads to a reduction in postoperative pain and to earlier mobilization. A long-term advantage of laparoscopic surgery is believed to be a reduction in the incidence of adhesions, which would in turn lead to fewer re-admissions for small bowel obstruction and ultimately a reduced incidence for re-operation.

A recent meta-analysis by Tilney et al. looked at 15 studies comparing open versus laparoscopic surgery for ileocaecal CD from 1995 to 2004[5]. The total number of cases was 783 with a conversion rate of approximately 7%. This meta-analysis showed that laparoscopic surgery offered a shorter hospital stay and an earlier return of gut function. More recently a randomized study was performed to further address the perceived benefits of laparoscopic resection. Maartense et al. looked at 60 patients who underwent either open or laparoscopic ileocaecal resection[11]. The surgeons in this study performed an extracorporeal handsewn anastomosis through a 4–5 cm midline incision. The conversion rate was 10%. Whilst operating time was longer for laparoscopic surgery, probably a feature of the learning curve, it was associated with a significantly shorter hospital stay. Furthermore morbidity (major and minor) was 10% in the laparoscopic group versus 33% in the open group. Interestingly the cost of laparoscopic surgery was significantly less than the open procedure. This is most likely due to the avoidance of expensive stapling devices. Surprisingly the authors of this study did not show any differences in the quality of life between the two groups. Whilst laparoscopic surgery may be feasible it is not appropriate for all patients with ileocaecal CD. Alves et al. showed that a prolonged medical course or the intraoperative findings of an abscess or fistula were associated with an increased risk of conversion[12]. In the presence of an abscess the general principle is to drain the collection, ideally via the percutaneous route[13]. Therefore the ideal candidate for laparoscopic surgery would be someone with a relatively short, uncomplicated history of ileocaecal CD and without evidence of an inflammatory mass/abscess or fistula.

Whilst the short-term results of laparoscopic surgery are clearly beneficial there are few studies which have addressed the long-term sequelae. Bergamaschi et al. found a reduced incidence of bowel obstruction in the laparoscopic group (11.1%) compared to open resection (35.4%) at 5 years following initial surgery[14]. However, Lowney et al. were unable to show that laparoscopic ileocaecal resection was associated with a lower recurrence rate[15]. Lawes et al. have also shown that patients who have undergone initial laparoscopic surgery for CD are more likely to have a further successful laparoscopic procedure in the event of recurrence[16]. It is likely we will see an increasing role for laparoscopic surgery in the management of uncomplicated CD.

MANAGEMENT OF CROHN'S STRICTURES

Alternative strategies for the management of Crohn's strictures include balloon dilation and stent insertion. The latter seems an attractive option, especially in patients with poor nutrition who would benefit from optimization prior to surgical resection[17]. In contrast balloon dilation has been used for large and small bowel Crohn's disease as well as strictures at a previous ileocolic anastomosis[18,19]. The size of stricture which is amenable to dilation is 2–3 cm and the procedure can be used in the presence of acute inflammation at the stricture site. The long-term recurrence was reduced in patients who also underwent steroid injection into the four quadrants of the stricture[20]. This technique has the advantage of avoiding intestinal resection and can be repeated. However, a disadvantage of dilation is that it cannot be used for the treatment of long segment Crohn's strictures and there is also a risk of perforation at the site of dilation.

Surgical strictureplasty now has an accepted place in the management of Crohn's stricture. Whilst the most used strictureplasty is the Heineke–Mikulicz it is not ideal in dealing with long strictures separated by short segments of normal bowel. These can be managed by surgical excision, but the amount of small bowel involved predisposes to short bowel syndrome. An alternative is a novel procedure described by Michelassi, which is a side-to-side isoperistaltic strictureplasty[21].

COMBINED THERAPY FOR PERIANAL CROHN'S DISEASE

The management of patients with perianal Crohn's disease fistulas has been modified with the introduction of potent immunological agents such as infliximab. This is a complex condition in which medical or surgical treatments alone are not always successful. However, following drainage of any underlying abscess a complex perianal fistula may be successfully drained by a long-term seton. The latter is commonly made of silastic and inserted along the fistula tract. However, this treatment on its own may not successfully heal the fistula. In contrast seton drainage and combined treatment with infliximab, followed by maintenance therapy with azathioprine and methotrexate, led to healing in up to 67% of cases[22]. Talbot et al. reported complete healing in 47% of patients with the remainder of patients having a

partial response[23]. A novel approach is administration of infliximab directly into the fistula tract with healing reported in 67% of patients[24]. However, in patients with extensive perianal Crohn's disease a laparoscopic defunctioning stoma can also be created to further optimize perianal wound healing[25].

ACUTE COLITIS

Surgery also has an important role in the management of acute colitis. There are a number of causes of colitis, the most frequent being inflammatory bowel disease, comprising ulcerative colitis (UC) and Crohn's disease (CD). In the acute setting the management of colitis is often confined to that of UC, but other causes of acute colitis should not be overlooked, including bacterial, viral, parasitic, ischaemic and drug-induced.

The surgical management of acute colitis cannot be described in isolation but goes hand in hand with the medical management of this condition. The acute presentation of bloody diarrhoea should initiate routine blood tests, stool culture, plain radiographs and a rigid sigmoidoscopy. The severity of an acute attack of UC can be determined by the Truelove and Witts classification[26]. Stool culture should be sent to exclude a bacterial and parasitic cause. It is also important to send stool for *Clostridium difficile* toxin to exclude pseudomembranous colitis.

A sigmoidoscopy is useful as it will reveal rectal inflammation. Furthermore a biopsy will be able to exclude cytomegalovirus (CMV) infection. This is very important as successful medical management of CMV colitis obviates the need for surgery. The findings of a toxic megacolon or mucosal islands on an abdominal X-ray are indicative in three-quarters of patients of potential failure of medical management and the likelihood of requiring surgery[27]. Travis et al. have described predictors for surgery[28]; they noted that a stool frequency greater or equal to eight or a C-reactive protein greater than 45 on day 3 during admission is associated with an 85% chance of requiring surgery.

The mainstay of medical treatment has been intravenous steroids. However, approximately 60% of patients have been reported to be resistant to this therapy. The alternative possibilities, which are referred to as second-line treatment, include the use of cyclosporin or a monoclonal antibody to tumour necrosis factor alpha (infliximab). The former can be used as an intravenous or oral preparation; furthermore cyclosporin does not increase the risk of perioperative complications[29]. However, whilst cyclosporin is effective in the treatment of acute severe UC, infliximab has been shown to be effective only for moderate UC[30,31]. In addition the longer half-life of infliximab compared to cyclosporin, days compared to hours respectively, can potentially render a patient immunosuppressed following surgery. This in theory can lead to increased postoperative morbidity, especially infective complications. Whilst a combined regime of cyclosporin and infliximab may seem attractive, the risk of profound immunosuppression is considerable; therefore infliximab would seem appropriate in the management of patients with moderate UC who would be predicted as having a minimal risk for surgery. The critical point to highlight is that the decision for surgery is not made unilaterally by the colorectal surgeon but as a consensus between surgeon and gastroenterologist. Patients predicted

as having a severe attack of colitis should have a colorectal surgeon involved from the onset. The decision for surgery should not be delayed, especially if there is a risk of perforation, as early surgery ensures better results.

Indications for a colectomy include acute colitis, chronic disease which has failed medical therapy and malignancy. The former accounts for approximately 40% of cases[32]. Absolute indications for surgery are perforation, massive bleeding and toxic megacolon. Once a decision has been made to operate it is important to involve the stomatherapist because one of the major concerns following surgery is stoma management. This also allows the opportunity to correctly site the stoma, especially as it is notoriously difficult once the patient is anaesthetized and supine. The patient is positioned in the modified Lloyd Davies position. Broad-spectrum antibiotics are routinely given as patients with acute colitis and on immunosuppression have an increased risk of wound infection. Mechanical bowel preparation is avoided due to the risk of perforation. Before surgery is commenced a rectal catheter is inserted to ensure that the rectum is decompressed, and also for irrigation of the rectal stump at the end of the surgical procedure. If one has not been placed preoperatively a urinary catheter is also inserted.

The surgical option of choice is a subtotal colectomy and ileostomy. The general principles of surgery are to remove the source of inflammation and avoid an anastomosis. These patients can be severely malnourished and the combination of high-dose steroids and immunosuppressants also makes them susceptible to both infection and anastomotic leak.

The surgical approaches include either a classical midline laparotomy or more recently a laparoscopic procedure[25]. There have been several studies which have compared the two techniques. The advantage of laparoscopic surgery is the obvious cosmetic result. Furthermore reduced tissue handling translates into a shorter time for bowel function to return and hence restoration of a normal diet. In the long term laparoscopic surgery should lead to a reduced incidence of adhesion formation[33]. There have been a few studies which have compared the two techniques. Dunker et al. compared 32 patients who had an open colectomy with 10 who underwent laparoscopic surgery[34]. In the latter technique the colon was removed via a pfannensteil incision. Neither study stated the number of cases in which the rectum was decompressed by a Foley catheter or as a mucous fistula. Patients with UC accounted for 80% of the laparoscopic group but 56% of the open group. The length of operative time was longer in the laparoscopic group but this was associated with a significantly shorter hospital stay (14.6 days in the laparoscopic vs 18 days in the open group). However, a criticism of this study is that the average length of hospital stay was significantly longer than is currently anticipated for open surgery. This may in part be due to the data being collected from 1996 to 1999. The recent trend for fast-track surgery has significantly reduced hospital stays even for an open procedure, which can average 5 days. Though the incidence of complications was similar between the two groups, there was a higher incidence of re-laparotomy in the open group. Marcello et al. compared 19 patients who underwent laparoscopic colectomy with 29 patients having an open procedure[35]. The rectal stump was left attached to the subcutaneous tissue. This study reported that laparoscopic surgery was associated with a

faster return of bowel function which correlated with a shorter hospital stay. However, as with the study by Dunker et al., the operative time was significantly longer in the laparoscopic procedure.

Several institutions have also reported their retrospective experience with laparoscopic colectomy[4,36]. One of the largest series to date is that of Marohn et al., in which 65 patients underwent laparoscopic colectomy, of whom 85% had UC[37]. In this study the rectal stump was closed and left intraperitoneally. Hospital stay was a mean of 4.3 days with a complication rate of 12%. Compared to the previously described studies the operative time was the longest with a mean of 444 min.

A subtotal colectomy and an end ileostomy removes the diseased colon and the patient usually makes a straightforward recovery despite retaining the rectal stump. Management of the rectum includes either leaving it long and intraperitoneally or short and in the pelvis, making a mucous fistula or leaving a closed rectal stump in the subcutaneous plane. The former, though leaving a minimal amount of diseased bowel, has the potential risk of resulting in a perforated rectal stump, which can be life-threatening. In contrast, if a subcutaneous stump leaks it leads to a wound discharge and formation of a mucous fistula which can easily be dealt with. Furthermore, during subsequent surgery for formation of an ileo–anal pouch or completion proctectomy a subcutaneous stump is easily located compared to an intraperitoneal one. A mucous fistula effectively decompresses the rectal stump from the onset.

Historical studies published from St Marks have shown that pelvic dissection in the acute setting is associated with an increased mortality for proctocolectomy of 9% compared to fewer than 2% for a subtotal colectomy[38]. The classic description of a subtotal colectomy involves formation of a left iliac fossa mucous fistula[39]. However, a major disadvantage of this technique is that it creates a second stoma with the associated problems, surgical and psychological, involved in its management. Albrechtsen et al. performed an emergency colectomy with ileostomy and mucous fistula in 120 patients with UC[40]. The mucous fistula was placed at the lower end of the wound. A further 12 patients with acute UC underwent an ileo–rectal anastomosis or a proctocolectomy. The complication rate for the individual procedure is not stated; the overall complications were wound sepsis/dehiscence of 24% and intra-abdominal abscess of 8%. Furthermore an ileo-rectal anastomosis is potentially dangerous, especially with the risk of anastomotic leakage, in the acute setting[41]. A novel approach is to place the mucous fistula in the right iliac fossa, at the same site as the ilesotomy[42].

In 1985 Motson and Manche described a technique in which a closed rectal stump was left at the lower aspect of the abdominal wound[43]. In our institution we also routinely practise this approach. A closed subcutaneous rectal stump avoids a second stoma. Ng et al. reported our experience with 32 patients who underwent a subtotal colectomy and closed subcutaneous rectal stump[44]. Wound infection occurred in 6% and pelvic sepsis in 3%. The choice for closure of the rectal stump was either stapled or handsewn, or in some cases both were employed. However, there have been only a handful of studies which have compared the different techniques for management of the rectal stump. Carter et al. compared a closed subcutaneous rectal stump with a mucous

fistula and a short pelvic rectal stump[45]. In this study the closed subcutaneous rectal stump was placed in the left iliac fossa. The method of closure of the stump, stapled or handsewn, had no association with pelvic sepsis or wound infection. Pelvic sepsis occurred in 4% of patients with a closed subcutaneous rectal stump, 7% with a mucous fistula and in 12% with a pelvic rectal stump, no differences being significant. Furthermore, patients were more symptomatic following a pelvic rectal stump compared to a mucous fistula or a closed subcutaneous rectal stump, with rectal bleeding occurring in 41% of the former compared to 27% in both the latter cases.

All these patients subsequently had an ileo–anal pouch with dissection being the most difficult in patients who had a pelvic rectal stump. In two of the latter group the rectum was perforated. Following a pouch formation sexual dysfunction was reported only in the group with a pelvic rectal stump. The authors reasoned that, with a short inflamed rectal stump, there was a higher propensity for adhesion formation. However, pelvic sepsis can also occur even with a closed subcutaneous rectal stump. This may be related to an inadequate length of colon resulting in a stump which may have an impaired blood supply and also be under tension.

Trickett et al. also compared their experience with a subcutaneous rectal stump versus an intraperitoneal stump[46]. Patients with a subcutaneous rectal stump had a significantly shorter hospital stay. This was associated with a pelvic sepsis rate of 0% and 7%, respectively. Furthermore, the incidence of wound infection was similar between the two groups. However, a major determinant of pelvic sepsis is clearly the length of the rectal stump. McKee et al. reported on a retrospective study comparing long and short rectal stumps[47]. Pelvic sepsis occurred in 1.9% of patients with a long stump compared to 33.3% in patients with a short rectal stump. Therefore an alternative to a subcutaneous closed rectal stump is an intraperitoneal stump adequately decompressed by a Foley catheter. Indeed Karch et al. reported no leakage from the closed rectal stump following routine insertion of a rectal catheter[48]. However, to date there have been no studies comparing the various surgical options for management of the rectal stump with routine use of a rectal catheter.

CONCLUSIONS

We have seen the steady introduction of laparoscopic surgery for IBD. The wider use of immunological agents in IBD has led to a reassessment of the timing of surgery. As highlighted by the treatment of acute colitis, optimal management involves a combined approach with both gastroenterologists and colorectal surgeons in a continuous clinical dialogue over the best treatment for the IBD patient.

References

1. Parks AG, Nicholls RJ. Proctocolectomy without ileostomy for ulcerative colitis. Br Med J. 1978;2:85–8.
2. Lindsey I, George BD, Kettlewell MG, Mortensen NJ. Impotence after mesorectal and close rectal dissection for inflammatory bowel disease. Dis Colon Rectum. 2001;44:831–5.

3. Larson DW, Cima RR, Dozois EJ et al. Safety, feasibility, and short-term outcomes of laparoscopic ileal pouch–anal anastomosis: a single institutional case-matched experience. Ann Surg. 2006;243:667–70; discussion 70–2.
4. Ouaissi M, Alves A, Bouhnik Y, Valleur P, Panis Y. Three-step ileal pouch–anal anastomosis under total laparoscopic approach for acute or severe colitis complicating inflammatory bowel disease. J Am Coll Surg. 2006;202:637–42.
5. Tilney HS, Constantinides VA, Heriot AG et al. Comparison of laparoscopic and open ileocecal resection for Crohn's disease: a metaanalysis. Surg Endosc. 2006;20:1036–44.
6. Shen B, Fazio VW, Remzi FH, Lashner BA. Clinical approach to diseases of ileal pouch–anal anastomosis. Am J Gastroenterol. 2005;100:2796–807.
7. Thompson-Fawcett MW, Mortensen NJ, Warren BF. 'Cuffitis' and inflammatory changes in the columnar cuff, anal transitional zone, and ileal reservoir after stapled pouch–anal anastomosis. Dis Colon Rectum. 1999;42:348–55.
8. Shen B, Lashner BA, Bennett AE et al. Treatment of rectal cuff inflammation (cuffitis) in patients with ulcerative colitis following restorative proctocolectomy and ileal pouch–anal anastomosis. Am J Gastroenterol. 2004;99:1527–31.
9. Remzi FH, Fazio VW, Delaney CP et al. Dysplasia of the anal transitional zone after ileal pouch–anal anastomosis: results of prospective evaluation after a minimum of ten years. Dis Colon Rectum. 2003;46:6–13.
10. Heppell J, Weiland LH, Perrault J, Pemberton JH, Telander RL, Beart RW Jr. Fate of the rectal mucosa after rectal mucosectomy and ileoanal anastomosis. Dis Colon Rectum. 1983;26:768–71.
11. Maartense S, Dunker MS, Slors JF et al. Laparoscopic-assisted versus open ileocolic resection for Crohn's disease: a randomized trial. Ann Surg. 2006;243:143–9; discussion 50–3.
12. Alves A, Panis Y, Bouhnik Y et al. Factors that predict conversion in 69 consecutive patients undergoing laparoscopic ileocecal resection for Crohn's disease: a prospective study. Dis Colon Rectum. 2005;48:2302–8.
13. Mortensen NJ. Abscess and Crohn's disease – early surgery or percutaneous drainage? (a British view). Dig Liver Dis. 2006;38:666–7.
14. Bergamaschi R, Pessaux P, Arnaud JP. Comparison of conventional and laparoscopic ileocolic resection for Crohn's disease. Dis Colon Rectum. 2003;46:1129–33.
15. Lowney JK, Dietz DW, Birnbaum EH, Kodner IJ, Mutch MG, Fleshman JW. Is there any difference in recurrence rates in laparoscopic ileocolic resection for Crohn's disease compared with conventional surgery? A long-term, follow-up study. Dis Colon Rectum. 2006;49:58–63.
16. Lawes DA, Motson RW. Avoidance of laparotomy for recurrent disease is a long-term benefit of laparoscopic resection for Crohn's disease. Br J Surg. 2006;93:607–8.
17. Bickston SJ, Foley E, Lawrence C, Rockoff T, Shaffer HA Jr, Yeaton P. Terminal ileal stricture in Crohn's disease: treatment using a metallic enteral endoprosthesis. Dis Colon Rectum. 2005;48:1081–5.
18. Singh VV, Draganov P, Valentine J. Efficacy and safety of endoscopic balloon dilation of symptomatic upper and lower gastrointestinal Crohn's disease strictures. J Clin Gastroenterol. 2005;39:284–90.
19. Thomas-Gibson S, Brooker JC, Hayward CM, Shah SG, Williams CB, Saunders BP. Colonoscopic balloon dilation of Crohn's strictures: a review of long-term outcomes. Eur J Gastroenterol Hepatol. 2003;15:485–8.
20. Brooker JC, Beckett CG, Saunders BP, Benson MJ. Long-acting steroid injection after endoscopic dilation of anastomotic Crohn's strictures may improve the outcome: a retrospective case series. Endoscopy. 2003;35:333–7.
21. Michelassi F. Side-to-side isoperistaltic strictureplasty for multiple Crohn's strictures. Dis Colon Rectum. 1996;39:345–9.
22. Topstad DR, Panaccione R, Heine JA, Johnson DR, MacLean AR, Buie WD. Combined seton placement, infliximab infusion, and maintenance immunosuppressives improve healing rate in fistulizing anorectal Crohn's disease: a single center experience. Dis Colon Rectum. 2003;46:577–83.
23. Talbot C, Sagar PM, Johnston MJ, Finan PJ, Burke D. Infliximab in the surgical management of complex fistulating anal Crohn's disease. Colorectal Dis. 2005;7:164–8.

24. Poggioli G, Laureti S, Pierangeli F et al. Local injection of infliximab for the treatment of perianal Crohn's disease. Dis Colon Rectum. 2005;48:768–74.
25. Casillas S, Delaney CP. Laparoscopic surgery for inflammatory bowel disease. Dig Surg. 2005;22:135–42.
26. Truelove SC, Witts LJ. Cortisone in ulcerative colitis; final report on a therapeutic trial. Br Med J. 1955;2:1041–8.
27. Lennard-Jones JE, Ritchie JK, Hilder W, Spicer CC. Assessment of severity in colitis: a preliminary study. Gut. 1975;16:579–84.
28. Travis SP, Farrant JM, Ricketts C et al. Predicting outcome in severe ulcerative colitis. Gut. 1996;38:905–10.
29. Hyde GM, Jewell DP, Kettlewell MG, Mortensen NJ. Cyclosporin for severe ulcerative colitis does not increase the rate of perioperative complications. Dis Colon Rectum. 2001; 44:1436–40.
30. Hommes D, Baert F, van Assche G et al. Management of recent onset Crohn's disease: a controlled, randomised trial comparing step-up and top-down therapy. Gastroenterology. 2005;129:371.
31. Jarnerot G, Hertervig E, Friis-Liby I et al. Infliximab as rescue therapy in severe to moderately severe ulcerative colitis: a randomized, placebo-controlled study. Gastroenterology. 2005;128:1805–11.
32. Oresland T. Review article: Colon-saving medical therapy vs. colectomy in ulcerative colitis – the case for colectomy. Aliment Pharmacol Ther. 2006;24(Suppl. 3):74–9.
33. Kessler H. Laparoscopic surgery in inflammatory bowel disease: is the future already here? Curr Opin Gastroenterol. 2006;22:391–5.
34. Dunker MS, Bemelman WA, Slors JF, van Hogezand RA, Ringers J, Gouma DJ. Laparoscopic-assisted vs open colectomy for severe acute colitis in patients with inflammatory bowel disease (IBD): a retrospective study in 42 patients. Surg Endosc. 2000;14:911–14.
35. Marcello PW, Milsom JW, Wong SK, Brady K, Goormastic M, Fazio VW. Laparoscopic total colectomy for acute colitis: a case–control study. Dis Colon Rectum. 2001;44:1441–5.
36. Bell RL, Seymour NE. Laparoscopic treatment of fulminant ulcerative colitis. Surg Endosc. 2002;16:1778–82.
37. Marohn MR, Hanly EJ, McKenna KJ, Varin CR. Laparoscopic total abdominal colectomy in the acute setting. J Gastrointest Surg. 2005;9:881–6; discussion 7.
38. Hawley PR. Emergency surgery for ulcerative colitis. World J Surg. 1988;12:169–73.
39. Lee EC, Truelove SC. Proctocolectomy for ulcerative colitis. World J Surg. 1980;4:195–201.
40. Albrechtsen D, Bergan A, Nygaard K, Gjone E, Flatmark A. Urgent surgery for ulcerative colitis: early colectomy in 132 patients. World J Surg. 1981;5:607–15.
41. Jones PF, Bevan PG, Hawley PR. Ileostomy or ileorectal anastomosis for ulcerative colitis? Br Med J. 1978;1:1459–63.
42. Alves A, Panis Y, Bouhnik Y, Maylin V, Lavergne-Slove A, Valleur P. Subtotal colectomy for severe acute colitis: a 20-year experience of a tertiary care center with an aggressive and early surgical policy. J Am Coll Surg. 2003;197:379–85.
43. Motson RW, Manche AR. Modified Hartmann procedure for acute ulcerative colitis. Surg Gynecol Obstet. 1985;160:462–3.
44. Ng RL, Davies AH, Grace RH, Mortensen NJ. Subcutaneous rectal stump closure after emergency subtotal colectomy. Br J Surg. 1992;79:701–3.
45. Carter FM, McLeod RS, Cohen Z. Subtotal colectomy for ulcerative colitis: complications related to the rectal remnant. Dis Colon Rectum. 1991;34:1005–9.
46. Trickett JP, Tilney HS, Gudgeon AM, Mellor SG, Edwards DP. Management of the rectal stump after emergency sub-total colectomy: which surgical option is associated with the lowest morbidity? Colorectal Dis. 2005;7:519–22.
47. McKee RF, Keenan RA, Munro A. Colectomy for acute colitis: is it safe to close the rectal stump? Int J Colorectal Dis. 1995;10:222–4.
48. Karch LA, Bauer JJ, Gorfine SR, Gelernt IM. Subtotal colectomy with Hartmann's pouch for inflammatory bowel disease. Dis Colon Rectum. 1995;38:635–9.

11
State-of-the-Art Lecture: The changing face of inflammatory bowel disease over recent decades

P. RUTGEERTS, S. VERMEIRE and G. VAN ASSCHE

INTRODUCTION

The chronic idiopathic inflammatory bowel diseases (IBD), Crohn's disease (CD) and ulcerative colitis (UC), are diseases that have become a real challenge to health in all developed and many developing countries over the world. Although CD was first described in detail in 1932 there have been cases reported in the latter half of the 19th century and certainly UC was already well known. The statement that IBD is associated with modern society implies only that an important increase in prevalence is observed overall and in westernized societies in particular. In this chapter we will review the changes that have occurred in epidemiology and in therapy in recent decades.

EPIDEMIOLOGY AND AETIOPATHOGENESIS

Racial differences observed in the past in the incidence and prevalence of IBD had probably more to do with the risk of exposure to environmental factors triggering the disease than with differences in genetic background. The highest incidence and prevalence rates of IBD are at present observed in Manitoba (Canada) and are the highest among non-Aboriginal persons, persons with high socioeconomic status, persons with the lowest rates of enteric infection and the highest rates of multiple sclerosis[1]. In Manitoba the incidence of CD seems to stabilize whereas the incidence of UC is gradually decreasing.

In general the north–south gradient (much lower incidence and prevalence in the south as compared with the north) observed in early epidemiological studies is gradually disappearing, as is the west–east gradient. In the south and the east incidence and prevalence rates are catching up with those in western Europe and North America, with a lag time of about 20 years.

In areas with low incidence IBD cases tend to be UC and very rarely CD. With increasing IBD incidence the proportion of cases of CD is rising.

Immigrants from areas with low IBD incidence moving into areas with high prevalence have an increasing chance of developing IBD over time. In areas with high prevalence there is a strikingly high occurrence of familial clustering, suggesting common exposure and shared susceptibility. In some of these areas, such as northern France and Belgium, cases of CD have started to outnumber UC. The course of IBD seems more aggressive in areas with high prevalence than in areas with low prevalence.

IBD slowly becomes a low-grade pandemic, and it is striking that the disease phenotypes for both CD and UC are completely similar all over the world. The predominant ileal and ileocolic location, the occurrence of perianal disease, the different types of disease behaviour, the lesions as visualized with different imaging techniques are found from northern Canada to South Africa, from Argentina to Tokyo. The same is true for UC, where the superficial diffuse spread from the anal verge up is universal. This suggests that the cause of IBD is probably not a sophisticated coexistence of abnormalities but rather a staightforward cause.

It probably still remains true that the two diseases are mutually exclusive, and that a patient either suffers from CD or from UC. It is a general belief that no patient has both, or develops one disease after the other, although an entity such as 'UC-like CD' is a puzzling finding.

There is now compelling evidence that genetic susceptibility to CD is characterized by disturbed innate immunity in the gut[2]. It is not clear whether disturbances in the innate immunity pathways can also be acquired leading to exposure of the mucosa to bacterial products and an inappropriately activated adaptive immune system. The interplay between genetic background and changed environment is probably critical, but is difficult to investigate. Changes in the environment associated with westernization are so profound and diverse that is very difficult to identify possible causative factors. Smoking remains the only environmental factor modulating both CD and UC, although in opposite directions. Appendicectomy has been found to protect against UC[3].

Although no single pathogen causing IBD has been identified, molecular studies of the faecal flora show quantitative changes of the different species and loss of diversity. It appears that changes in the gut flora occur over time in subjects exposed to a western diet. How this predisposes to the development of IBD is not clear.

TREATMENT OF IBD

Although cure of IBD is still far away, much progress has been made concerning its treatment. Population-based studies have shown that glucocorticosteroids do not maintain remission of IBD, do not change the outcome of the disease and have a deleterious effect long term[4]. In most IBD centres where many patients with IBD are treated the use of steroids has been reduced over time and, if used, patients are exposed for short periods of time, and more topically acting steroids and less systemic steroids are used. Although immunosuppression with azathioprine/6-mercaptopurine or methotrexate has been used more widely in the recent decade than in the early 1990s this use has

not yet decreased the need for surgery[5] despite the fact that azathioprine and methotrexate are efficacious in maintaining remission and inducing mucosal healing in CD. The beneficial effects of immunosuppression have been less convincingly demonstrated in UC than in CD. It is clear that these drugs are mostly initiated too late in the course of the diseases at a time when complications have already arisen, and when surgery cannot be avoided. The greatest breakthrough in therapy has been the introduction of the monoclonal antibody to tumour necrosis factor (TNF). This therapy has had a great impact on the quality of life of Crohn's patients, and the need for corticosteroid use has decreased. Moreover the need for surgery was decreased with these treatments in controlled trials[6,7]; this has, however, to be confirmed in the real-life situation. Surgery in Crohn's disease overall surely has become more conservative. Extensive resections have been replaced by short segmental resections and strictureplasties using different techniques. As a consequence there are less intestinal cripples in the CD population than 10–15 years ago.

There was much doubt, based on animal models of IBD, that anti-TNF strategies would be beneficial in the treatment of UC; the disease was considered to be a Th2 helper disease where not TNF-α but interleukins IL-5 and IL-13 were the main deleterious cytokines. The ACT studies, however, have shown that infliximab may have the same efficacy in the treatment of UC as in CD[8]. Infliximab was even shown to be very effective in rescuing pre-surgical patients with steroid-refractory acute colitis[9]. The impact of better therapy for UC on the need for surgery overall is not yet proved, and we await the results of longitudinal studies. Most series on colectomy with restorative ileo-anal pouch report on suboptimal quality of life, with a high rate of complications, especially of pouchitis, in the long term; therefore a medical alternative for this procedure is greatly needed.

With the introduction of biological therapies our treatment goals have changed in recent years. We now aim at induction and maintenance of remission without steroids, maintained mucosal healing and avoidance of complications, strictures and perforation and improvement of the long-term outcome of IBD.

There is still need for improved therapy, however. About 30% of patients do not respond to anti-TNF therapy and about 10% of patients who respond early to the drug lose response per treatment year. It is critical to develop alternative biologicals. Candidate drugs are the selective anti-migration (SAM) strategies, anti-IL-12 among others. We have to learn from the failures. Although stimulation of the innate immunity is an attractive strategy, and despite early promising data, a recent large phase 3 trial has not shown benefit for GM-CSF. It is possible that stimulation of the innate immune system might be efficacious especially in the early phases of IBD or in patients with proven defects in innate defence. Early CD may be quite different from late disease concerning the immunological insult to the bowel.

There are indeed preliminary data showing that the early introduction of aggressive therapy in CD is able to change the long-term outcome of the disease, and this allows us to treat active disease without using steroids[10]. The step-up top-down study shows that aggressive therapy in patients who are naive to steroids, immunosuppression and biologicals is more effective than the

classical treatment pyramid in inducing remission and avoiding steroids over a period of 2 years. With this strategy improved bowel healing is also achieved.

There is also evidence that biological therapy works better in the paediatric population than in adults[11]. At present the combination of infliximab with concomitant immunosuppression is the most effective approach. In about 50% of patients infliximab can be stopped after being used as a bridge to full immunosuppression efficacy[12]. On the other hand azathioprine can be discontinued in patients treated for 6 months with the combination of infliximab and azathioprine, and who had formerly failed azathioprine monotherapy, without increased risk for relapse. This approach should also be studied using other biological treatments.

Aggressive treatment also allows healing of the bowel mucosa quickly, and maintenance of healing. There is evidence that induction of mucosal healing is necessary to improve the long-term outcome of the disease.

A proportion of patients, however, have rather indolent disease, and in these patients the use of aggressive therapy early may be harmful. At present it is difficult to identify which patients will do well over time without needing aggressive therapy that is associated with potential severe side-effects. Therefore we need predictive factors for disease evolution, and at present these are lacking.

There is a great need for development of biomarkers or other predictors for response to the different medications. In this respect our clinical outcome parameters lack sensitivity. We have to rely more on the extent of mucosal ulcerations as the most objective parameter of disease activity, although less invasive parameters such as calprotectin may also be of great value.

It may be expected that therapy will greatly improve further in the coming years. Can we identify disease-modifying therapy in IBD similar to rheumatoid arthritis? In RA cessation of joint damage can be achieved with the combination of methotrexate and different biologicals. This effect is more important when this therapy is started early in the course of the disease.

CONCLUSIONS

It is clear that, although IBD has become a big health problem in our societies, these diseases can now be treated much more efficaciously than 10 years ago There is a great need for intensive basic and clinical research to improve our understanding of IBD and to further improve our therapies.

References

1. Green C, Elliot L, Beaudoin C, Bernstein CN. A population-based ecologic study in inflammatory bowel disease searching for etiologic factors. Am J Epidemiol. 2006;146: 615–23.
2. Wehkamp J, Stange EF. A new look at Crohn's disease: breakdown of the mucosal antibacterial defense. Ann NY Acad Sci. 2006;1072:321–31.
3. Rutgeerts P, D'Haens G, Hiele M, Geboes K, Vantrappen G. Appendectomy protects against ulcerative colitis. Gastroenterology. 1994;106:1251–3.
4. Munkholm P, Langholz E, Davidsen M, Binder V. Frequency of glucocorticoid resistance and dependency in Crohn's disease. Gut. 1994;35:360–2.

5. Cosnes J, Nion-Larmurier I, Beaugerie L, Afchain P, Tiret E, Gendre JP. Impact of the increasing use of immunosuppressants in Crohn's disease on the need for intestinal surgery. Gut. 2005;54:237–41.
6. Rutgeerts P, Feagan BG, Lichtenstein GR et al. Comparison of scheduled and episodic treatment strategies of infliximab in Crohn's disease. Gastroenterology. 2004;126:402–13.
7. Lichtenstein GR, Yan S, Bala M, Blank M, Sands BE. Infliximab maintenance treatment reduces hospitalisation surgeries and procedures in fistulising Crohn's disease. Gastroenterology. 2005;128:862.
8. Rutgeerts P, Sandborn WJ, Feagan BG et al. Infliximab for induction and maintenance therapy for ulcerative colitis. N Engl J Med. 2005;353:2462–76.
9. Jarnerot G, Hertervig E, Friis-Liby I et al. Infliximab as rescue therapy in severe to moderately severe ulcerative colitis: a randomized, placebo-controlled study. Gastroenterology. 2005;128:1805–11.
10. Hommes D, Baert F, Van Assche G et al. Management of recent onset Crohn's disease: A controlled randomized trial comparing step-up and top-down therapy. Gastroenterology. 2005;129:371.
11. Hyams J, Crandall W, Kugathasan S et al.; REACH Study Group. Induction and maintenance infliximab therapy for the treatment of moderate-to-severe Crohn's disease in children. Gastroenterology 2007;132:863–73.
12. Lemann M, Mary JY, Duclos B et al.; Groupe d'Etude Therapeutique des Affections Inflammatoires du Tube Digestif (GETAID). Infliximab plus azathioprine for steroid-dependent Crohn's disease patients: a randomized placebo-controlled trial. Gastroenterology. 2006;130:1054–61.

Section V
Laboratory markers and other tools

Chair: S ARSLAN and S VERMEIRE

12
Antibodies: useful tools or pathophysiology markers?

J. BRANCHE, M. CHAMAILLARD and J.-F. COLOMBEL

INTRODUCTION

The literature on serological markers for inflammatory bowel diseases (IBD) dates back to the 1960s when Broberger and Perlmann reported autoantibodies against antigen derived from colon in serum of patients with ulcerative colitis (UC). Since the description of an association between antineutrophil cytoplasmic antibodies (ANCA) and ulcerative colitis (UC) and between anti-*Saccharomyces cerevisiae* mannan antibodies (ASCA) and Crohn's disease (CD), a resurgence of interest in IBD-related antibodies has occurred. New markers directed against microbial antigens and glycans have recently emerged offering the clinician an actual IBD serological panel. Nevertheless, as reviewed in this chapter, the clinical utility of serological markers as diagnostic tools, and in disease monitoring and stratification, is still debated. Other areas of current interest concern their role as genetic marker of susceptibility in families and their pathogenetic implications.

THE IBD SEROLOGICAL PANEL (TABLE 1)

The two serological tests most widely studied are pANCA and ASCA. pANCA stands for antineutrophil cytoplasmic antibody with a perinuclear staining pattern at indirect immunofluorescence (reviewed in refs 1 and 2). In IBD the most frequent fluorescence pattern is actually perinuclear and diffusing towards the cytoplasmic area of neutrophils, so-called pANCA with diffuse cytoplasmic staining, pANCA with 'snowdrift-like appearance', 'x-ANCA', or 'atypical ANCA'. According to the International Consensus the name for these staining patterns should be 'atypical ANCA'. Nevertheless, in the literature, the ANCA pattern associated with IBD is always termed 'pANCA'. ASCA stands for anti-*Saccharomyces cerevisiae* antibody. The serological response mainly concerns sequences of mannose residues expressed in the cell wall mannan of *S. cerevisiae.* Although there are few data suggesting an advantage to performing separate testing for immunoglobulins IgG and IgA ASCA[3], most companies

Table 1 Prevalence of antibodies in IBD[1,2,7,8,11,28]

	pANCA	*ASCA*	*PAB*	*Anti-OmpC*	*Anti-I2*	*Anti-flagellin*
CD	45–82%	48–69%	27–39%	55%	50%	50%
UC	2–28%	5–15%	3–23%	2%	10%	6%
Healthy controls	2.5%	5%	0%	1.3%	10%	8%

provide separate assays. ANCA have been detected in the sera from 45% to 82% of patients with UC and in 2–28% of patients with CD (reviewed in refs 1 and 2). In contrast, ASCA are found in 39–69% of CD patients but only in 5–15% of UC patients (reviewed in refs 1 and 2). It is important to note that pANCA and ASCA detection is not standardized; therefore tests may perform differently.

Among the oligomannose repertoire of the *S. cerevisiae* mannan, the ASCA major epitopes have been shown to be a mannotriose α1-3 Man α1-2 Man α1-2 Man (M3) and a mannotetraose α1-3 Man α1-2 Man α1-2 Man α1-2 Man (M4)[4]. Using oligomannosidic epitopes constructed chemically – synthetic (Σ) oligomannosides – as antigens a new ASCA test ($ASCA_2$) directed against the M3 mannotriose has been recently developed and is currently being evaluated in IBD[5]. If the exact immunogen for ASCA is still unknown, Standaert-Vitse et al. recently identified *Candida albicans* as one of several immunogens for ASCA. *C. albicans* was found to express the major ASCA epitopes on several cell wall molecules and overexpression of ASCA epitopes by *C. albicans* was shown to be triggered by growth conditions of the yeast. Specific overexpression of the ASCA epitope is triggered by pathogenic development of *C. albicans* in human tissues, supporting the intense specific ASCA response observed during human candidiasis[6].

Pancreatic antibodies (PAB) have been reported in patients with IBD using indirect immunofluorescence. No standardized protocol is available for their detection. Two staining patterns could be distinguished, namely an intracellular and extracellular pattern of PAB. PAB are present in 27–39% of CD sera, compared with less than 5% in UC, although a recent study from Belgium found a much higher 23% prevalence in patients with UC[7].

Chronic intestinal inflammation, as seen in IBD, results from an aberrant mucosal immune response to the microbiota of the gastrointestinal (GI) tract in genetically susceptible individuals. Immune responsiveness to several microbial-specific microbial antigens in patients with CD and UC has been described by Targan and colleagues. OmpC is the outer membrane porin C of *Escherichia coli.* I2 is a fragment of bacterial DNA which has been cloned from lamina propria mononuclear cells in active CD. This sequence has been shown to be associated with *Pseudomonas fluorescens*. In CD reported seroprevalence is 55% for anti-OmpC and 50% for anti-I2[8]. Linskens et al. used several strains of Gram-positive anaerobic coccoid rods in an agglutination test[9]. The agglutinating antibodies to coccoid rods were mostly of IgG isotype. A sensitivity of 52% for CD was reported. More recently, Lodes et al. used

serological expression cloning in colitic C3H/HeJBir mice to identify commensal bacterial proteins that could contribute to the pathogenesis of IBD[10]. The dominant antigens identified were flagellins. A T cell line specific for one of those flagellins (CBir1) induced colitis when transferred into naive SCID mice. A serum response to flagellin CBir1 was detected in 50% of CD patients vs 6% of UC patients and 8% of controls[11].

Based on the existence of CD-specific antibodies against sugars, such as ASCA, the presence of several other glycan antibodies in IBD was recently evaluated[12]. Glycans are predominant surface components with glycosidic bonds including sugars (polysaccharides or carbohydrates), which can be found on micro-organisms, immune cells and erythrocytes. Three novel antiglycan antibodies have been described, designated antilaminaribioside carbohydrate IgG antibodies (ALCA), antichitobioside carbohydrate IgA antibodies (ACCA) and anti-mannobioside carbohydrate IgG anibodies (AMCA). Laminaribioside is the building block of laminarin, a polysaccharide of the β-1-3-glucan family. Chitobioside is a component of chitin, a polymer of *N*-acetyl-β-D-glucosamine, a major component of the insect cuticle as well as the cell walls of infectious pathogens such as bacteria and yeast. Mannobioside, a dimer of α-1,3-linked mannose, is a component of mannan from pathogenic fungi and yeast. In the initial work by Dotan et al. ALCA and ACCA were positive in 27% and 25% of the CD cohort; in 4% and 5% of the UC cohort; in 9% and 9% of the other-than-IBD GI diseases cohort; and in 2% and 12% of healthy control patients, respectively[12]. Recent results of a larger study in a Belgian cohort showed that such serological markers provide a panel which could complement ASCA for disease diagnosis. Even if ALCA IgG, ACCA IgA and AMCA IgG were specific for CD, their sensitivity was poor (CD vs UC: 17.7%, 20.7% and 28.1%, respectively) compared to ASCA, suggesting a need for further prospective studies in IBD[13,14].

In conclusion, the IBD serological panel is rapidly expanding. New serological tests may soon be commercially available, either alone or in combination.

SEROLOGICAL MARKERS AND IBD DIAGNOSIS

The clinical value of pANCA or ASCA testing in patients presenting with non-specific GI symptoms is limited because of inadequate sensitivity. ANCA positivity has been observed in other colitides, such as eosinophilic and collagenous colitis. The specificity of ASCA seems to be higher, but ASCA positivity has been observed in patients with Behçet's disease, primary biliary cirrhosis, autoimmune hepatitis and coeliac disease in which up to 43% of patients have been reported positive in some series[15]. The use of serological markers in routine screening in adults is not recommended[16]. It has been claimed that the conclusion may be different in children. Dubinsky et al. have proposed a sequential diagnostic testing strategy based on serological markers in order to facilitate diagnosis of IBD in children and to avoid unnecessary evaluations[17]. This approach was suggested to be cost-effective[18]; however, two recent studies in children came to the conclusion that, as in adults, the low

sensitivity of serological markers reduces their value in screening and precludes their ability to replace traditional studies when evaluating a patient with suspected IBD[19,20]. In the study by Khan et al.[19] one-half of children with a new diagnosis of CD were not identified by serology. Furthermore children who were ASCA positive were also positive with routine laboratory tests. Finally, taking into consideration the presence of rectal bleeding, routine laboratory tests and serology, only 76% of children with CD would have been identified as possibly having IBD prior to an endoscopic procedure. Negative serology should thus not preclude an endoscopic procedure in symptomatic children.

What about the distinction between CD and UC, and the role of serological markers in indeterminate colitis (IC)? The most specific serological test to distinguish CD from UC is the combination of ASCA and pANCA. The CD-associated serological pattern is ASCA+/pANCA–; conversely the UC-associated pattern is pANCA+/ASCA–. Several independent studies found that these combinations had positive predictive values of 77–96% for differentiating CD from UC[1,2]. Using likelihood ratios, patients who are pANCA positive and ASCA negative are 19 times more likely to have UC, whereas patients who are ASCA positive and pANCA negative are 16 times more likely to have CD[21]. It should be remembered, however, that these estimates are based on retrospective studies mostly performed in referral centre populations. The results of the only prospective study which assessed the usefulness of serological markers in IC were indeed less appealing[22]. Ninety-seven patients with an initial diagnosis of IC were analysed for ANCA and ASCA. After a mean of 1 year follow-up a definitive diagnosis was reached in 31/97 (32%) patients (Table 2). ASCA+/ANCA– predicted CD in 80% of IC patients whereas ASCA–/ANCA+ was predictive for UC in 64% (Table 3). Nevertheless, 48.5% of IC patients did not have antibodies against ASCA or ANCA, thus limiting the clinical utility of serological testing. Interestingly, the majority of these patients remain IC during their further clinical course, perhaps reflecting a distinct clinicoserological entity. In summary, the serological tests may serve as an adjunct to the clinical workup in IC but they should not be used alone in particular to determine the appropriateness of restorative proctocolectomy in patients needing surgery[23].

Table 2 Results of ASCA and ANCA in a prospective study of patients with indeterminate colitis[22]

	n	*Crohn's disease* *n (%)*	*Ulcerative colitis* *n (%)*	*Indeterminate colitis* *n (%)*
ASCA+/ANCA–	26 (26.8)	8 (30.8)	2 (7.7)	16 (61.5)
ASCA–/ANCA+	20 (20.6)	4 (20)	7 (35)	9 (45)
ASCA+/ANCA+	4 (4.1)	2 (50)	1 (25)	1 (25)
ASCA–/ANCA–	47 (48.5)	3 (6.4)	4 (8.5)	40 (85.1)
Total	97 (100)	17 (17.5)	14 (14.4)	66 (68.1)

Table 3 Sensitivity, specificity, positive predictive value (PPV) and negative predictive value (NPV) of the combination of ASCA and ANCA in a prospective study of patients with indeterminate colitis[22]

	Diagnosis	*Sensitivity*	*Specificity*	*PPV*	*NPV*
ASCA+/ANCA-	CD	8/12 (66.7%)	7/9 (77.8%)	8/10 (80%)	7/11 (63.6%)
ASCA-/ANCA+	UC	7/9 (77.8%)	8/12 (66.7%)	7/11 (63.6%)	8/10 (80%)

Whether addition of other tests would improve the clinical utility of serology is still unsettled. A follow-up study of the prospective cohort of IC patients showed no additive value of anti-OmpC and anti-I2 to ASCA and pANCA[24], whereas ASCA2 the test allowed correct re-classification of three CD patients[5]. The new anti-flagellin antibodies could in the near future be especially relevant in IC since they are associated with colonic CD, independent from ASCA and may allow us to differentiate pANCA+ CD patients from pANCA+ UC patients[11].

SEROLOGICAL MARKERS AND IBD STRATIFICATION

Serological markers have been further used to categorize subgroups of patients with IBD.

Associations between pANCA and UC phenotypes have remained inconsistent. In particular, a long-term follow up in 102 UC patients showed no association between pANCA and pouchitis[25]. pANCA in CD has been associated with a UC-like phenotype. A recent analysis of pANCA and ASCA, according to the Vienna classification, found late-onset disease and inflammatory disease type to be associated with pANCA[26].

The strongest phenotypic association of ASCA in CD with small bowel rather than colonic disease has been confirmed. A group from Edinburgh described a strong association between ASCA and progression type from purely inflammatory to stricturing and penetrating disease with a more severe phenotype and requirement for surgery[27].

Using cluster analysis Landers et al. have shown that CD patients can have a loss of tolerance to specific bacterial antigens and autoantigens and can be separated into four groups depending on their antibody response patterns: ASCA, OmpC/I2, pANCA or no/low response[8]. Stratification of patients based on the presence of the markers and level of the antibody responses was demonstrated in a subsequent study from the same group[28]. Anti-I2 was associated with fibrostenosis and small bowel surgery, OmpC with internal penetrating behaviour. ASCA was associated with small bowel disease, fibrostenosis, internal perforating disease, and small bowel surgery and was negatively associated with UC-like behaviour. pANCA was associated with UC-like disease and was negatively associated with small bowel disease, fibrostenosis and small bowel surgery. Seventy-two per cent of patients

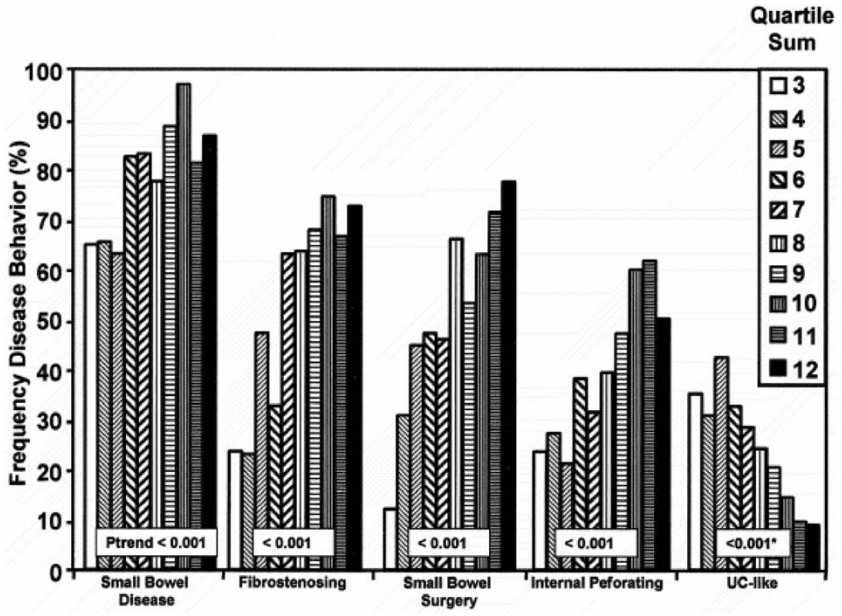

Figure 1 Frequency of Crohn's disease behaviour according to antibody reactivity (from ref. 28). Quartile sum analysis (sum of quartile scores for anti-I2, anti-OmpC and ASCA) was performed to evaluate the association between the combination of the level of antibodies and disease characteristics for any individual patient. By adding individual quartile scores for each antibody, a quartile sum score (range 3–12) was achieved that represented the cumulative immune response towards all three microbial antigens. Patients with increasing scores have an increasing likelihood of small bowel, fibrostenotic, and perforating disease and an increasing need for small bowel surgery, and a decreasing frequency of UC-like phenotype

positive for I2, OmpC and ASCA had undergone small bowel surgery compared with 23% of patients without reactivity. An association with the level of antibody production was demonstrated. Levels of ASCA, I2 and OmpC were broken down by quartile. By adding individual quartile scores for each serological marker a quartile sum score was then calculated. As shown in Figure 1, patients with increasing quartile sum scores were more likely to have small bowel disease, fibrostenotic and internal perforating disease with an increasing need for small bowel surgery and a decreasing frequency of UC-like phenotype. Complementary data from an independent European population confirmed, using the same methodology, that the presence and magnitude of serological responses was critical in determining CD phenotype and severity[29]. Disease type progressed from inflammatory to either stricturing or penetrating in 12.5% of patients who had total antibody responses in the lowest third of the cohort compared with 80% of patients with responses in the highest third. Similarly, 25% of patients with responses in the lowest third required surgery compared with 80% with responses in the highest third. In

summary, high-level immune responses towards microbial antigens are associated with more severe, complicated CD phenotypes. These associations are independent from NOD2/CARD15 variant carriage rate[27,28]. More recently, Ferrante et al. evaluated a wide range of antibodies directed towards anti-glycans (gASCA, ALCA, ACCA and AMCA) and Omp in 1225 adult IBD patients[13]. Consistently, patients with increasing numbers of CD-associated serological markers experienced more abdominal surgery, ileal involvement and complicated behaviour. Furthermore, high titres of antibodies towards gASCA, AMCA and Omp may reflect longer disease duration[13,14].

SEROLOGICAL MARKERS AND IBD MONITORING AND MANAGEMENT

The relationship between the presence of ANCA and IBD activity remains controversial. Nevertheless, most studies do not support a relationship between the presence or titre of ANCA and UC activity. Hence, in contrast with systemic vasculitides, serial measurement of ANCA titres in IBD is not useful for follow-up of disease activity and prediction of relapses. The presence of ASCA in CD is generally stable over time and is independent of CD activity and duration. A follow-up study in 25 patients with active CD showed no influence of mesalamine. ASCA levels were decreased by prednisolone but ASCA status remained stable[30]. In one paediatric study ASCA titres correlated significantly with disease activity and a significant reduction of ASCA was observed when clinical remission was achieved, suggesting an additional use for the follow-up of CD children[31]. This was not confirmed, however, by Desir et al., who concluded from a prospective study in 61 CD children that variability in ASCA titres over time was limited, with an inability of serial measurements to predict clinical outcomes[32].

The association between responses to microbial antigens and CD severity and the need for eraly early surgery makes these markers a potentially important prognostic tool. Awareness of a particular patient's susceptibility to disease progression may be an indication for more aggressive medical therapy. However, these findings must be confirmed in prospective studies evaluating the presence and levels of antibody responses and the development of CD complications and the need for surgery[28]. To evaluate the predictive and prognostic value of serological markers in IBD, a prospective study enrolling 196 paediatric CD patients showed that the presence and magnitude of immune responses influence disease behaviour. More interestingly, while seronegative patients remained complication-free, disease of seropositive patients progressed to internal penetrating and/or stricturing disease[33].

One attractive field of development for serological markers may be the prediction of response to therapy. A higher clinical response to infliximab has been associated with the presence of 'speckled' ANCA, while lack of response was associated with pANCA[34]. Esters et al. assessed the value of serological markers for prediction of response to infliximab in 279 patients with CD[35]. There was no overall relationship between ASCA or ANCA and response to therapy. Lower response rates, although not significant, were observed in

patients with refractory disease carrying the ANCA+/ASCA– combination. These observations need to be confirmed in independent series. In UC Fleshner et al. demonstrated that 56% of patients with a high level of pANCA prior to surgery are strongly associated with the development of pouchitis, compared with 20% of patients with low pANCA or no pANCA expression[36]. However, in the study by Aisenberg et al. only 1/102 patients was found to have a high titre of pANCA[25]. In a study of 27 patients with CD needing either an ileostomy or a colostomy for refractory proctocolitis or perianal disease, presence of anti-I2 antibodies was predictive of a clinical response to fecal diversion[37]. This observation supports the hypothesis that patients reactive to particular microbial antigens may respond to manipulation of bacterial flora.

SEROLOGICAL MARKERS AND PREDICTION OF IBD

Serum autoantibodies, which appear long before the onset of clinical disease, are a characteristic feature of autoimmune diseases[38], as emphasized by two recent studies. Nielen et al., studying patients with rheumatoid arthritis, found antibodies for IgM rheumatoid factor or anticyclic-citrullinated-peptide in serum samples take a median of 4.5 years before disease onset[39]. Arbuckle et al., using the US Armed Serum Repository, identified 132 military personnel diagnosed with systematic lupus erythematosus in whom antinuclear antibodies and anti-Ro appeared as early as 10 years before the onset of the disease[40]. Similarly, by using data from the Israeli Defense Force Military Corps Serum Repository, Israeli et al. determined ASCA and pANCA serological status before onset of the disease[41]. Whereas none of the controls was ASCA positive, ASCA precedes clinical diagnosis by about 38 months in one-third of CD patients, indicating a 'preclinical' value of ASCA in CD. Regarding other GI diseases, ASCA was present in seven out of 62 patients with coeliac disease before the onset of any symptom[42]. Another important reason to measure serum antibodies in IBD might thus be to detect predisposition to the disease and even subclinical IBD. Increased seroprevalence for several antimicrobial antibodies, notably ASCA[1,2,43] and anti-OmpC[44], have been consistently found with an increased frequency in first-degree relatives of patients with IBD[1,2]. However, no evolution to IBD has yet been observed in family members of IBD patients who were previously found to have ASCA.

SEROLOGICAL MARKERS AND IBD PATHOPHYSIOLOGY

The current theory in IBD is that chronic intestinal inflammation is the result of an aberrant immunological response to commensal bacteria within the gut lumen. Antibody responses towards microbial antigens may reflect the interplay between a genetically susceptible host and relevant micro-organisms. Data from experimental models of IBD have suggested that there is a relatively small number of immunodominant antigens stimulating the pathogenic T cell response and that there is an association between specific bacteria and the

disease location and severity[28]. However, the complexity of the intestinal microflora has posed a significant challenge to their identification. Seroreactivity against I2, an antigen derived from a *Pseudomonas* species present within the intestinal microflora, and against flagellin, an antigen present on most motile bacteria in the gut, points to a specific role of this bacterial antigen in the pathogenesis of IBD[10,11,28]. Interestingly, CBir1 is an immunodominant antigen in mouse colitis. CBir1 is thus the first bacterial antigen involved in experimental colitis which demonstrates a similar aberrant immune response in patients with CD.

Different hypotheses concerning the origin of ASCA in IBD can be proposed. They must take into account the widespread distribution of oligomannosides shared by many molecules present in a wide variety of organisms. The presence of IgG ASCA-reactive antigens in the granulomas of bowel resections and on infiltrating lymphocytes and neutrophils in inflamed tissue from CD patients suggests the presence of the immunogen in the lesions[45]. However, it is not clear whether ASCA detected by using *S. cerevisiae* are generated by this microbe, or whether they represent cross-reactivity with other(s), yet-unidentified, microorganism(s) carrying similar mannose structures[4]. Among these, and besides dietary yeasts, other yeast species which are frequent and natural commensals of the GI tract may also represent immunogen candidates, such as *Candida albicans*, recently identified as immunogen for ASCA[6]. Whatever the source of stimulation, endogenous and/or exogenous, it is likely that CD patients are mounting, as well as that described for bacterial antigens, an abnormal immune reaction to yeasts. It can be hypothesized that a lack of tolerance to *C. albicans* could lead to ASCA formation and persistence in a subset of CD patients genetically predisposed. An increased permeability in CD might lead to an increased exposure of yeast antigens to immune reactive cells[6]. However, no association was found between ASCA titres and small intestinal permeability, as measured by ^{51}Cr/EDTA or lactulose/mannitol test[46,47].

CONCLUSIONS

Despite the popularity of antibody testing in the gastroenterology community, evidence that it should affect clinical decision-making is still preliminary. Serological markers are not sensitive enough to be used for IBD screening in the general population. The therapeutic decision in patients with IC must not rely solely on serological testing. Although a high titre of pANCA in UC and of antibodies against microbial antigens in CD suggests a worse outcome, prospective data are necessary before this can be used to guide clinical decisions. Recent observations have reinforced the potential of serological markers for clustering of CD patients into more homogeneous groups based on antibody responses. Ongoing studies looking at correlations between serological markers and phenotypes, genotypes and responses to treatment would lead to a clearer understanding of the pathophysiology of different subsets of IBD.

References

1. Reumaux D et al. Serological markers in inflammatory bowel diseases. Best Pract Res Clin Gastroenterol. 2003;17:19–35.
2. Colombel JF et al. Serodiagnostics in IBD. In: Bernstein CN, editor. The Inflammatory Bowel Disease Yearbook 2003. London: Remedica, 2003:63–78.
3. Klebl FH et al. Optimising the diagnostic value of anti-*Saccharomyces cerevisiae*-antibodies (ASCA) in Crohn's disease. Int J Colorectal Dis. 2004;19:319–24.
4. Sendid B et al. Specific antibody response to oligomannosidic epitopes in Crohn's disease. Clin Diagn Lab Immunol. 1996;3:219–26.
5. Vandewalle P et al. A new ASCA test based on synthetic oligomannosides epitopes complements ASCA detection in both Crohn's disease and indeterminate colitis. Gastroenterology. 2004;126:A112.
6. Standaert-Vitse A et al. *Candida albicans* is an immunogen for anti-*Saccharomyces cerevisiae* antibody markers of Crohn's disease. Gastroenterology. 2006;130:1764–75.
7. Joossens S et al. Pancreatic autoantibodies in inflammatory bowel disease. Inflamm Bowel Dis. 2004;10:771–7.
8. Landers CJ et al. Selected loss of tolerance evidenced by Crohn's disease-associated immune responses to auto- and microbial antigens. Gastroenterology. 2002;123:689–99.
9. Linskens RK et al. Evaluation of serological markers to differentiate between ulcerative colitis and Crohn's disease: pANCA, ASCA and agglutinating antibodies to anaerobic coccoid rods. Eur J Gastroenterol Hepatol. 2002;14:1013–18.
10. Lodes MJ et al. Bacterial flagellin is a dominant antigen in Crohn disease. J Clin Invest. 2004;113:1296–306.
11. Targan S et al. Antibodies to a novel flagellin (CBir1) define a unique serologic response in Crohn's disease. Gastroenterology. 2004;126:A112.
12. Dotan I et al. Antibodies against laminaribioside and chitobioside are novel serologic markers in Crohn's disease. Gastroenterology. 2006;131:366–78.
13. Ferrante M et al. New serological markers in inflammatory bowel disease are associated with complicated disease behaviour. Gut. 2007. April 24 (Epub ahead of print).
14. Peyrin-Biroulet L et al. IBD serological panels: facts and perspectives. Inflamm Bowel Dis. 2007. July 16 (Epub ahead of print).
15. Damoiseaux JG et al. Diagnostic value of anti-*Saccharomyces cerevisiae* and antineutrophil cytoplasmic antibodies for inflammatory bowel disease: high prevalence in patients with celiac disease. J Clin Immunol. 2002;22:281–8.
16. Escher JC. Serologic tests are helpful in managing but not in diagnosing IBD. Inflamm Bowel Dis. 2002;8:230–321.
17. Dubinsky MC et al. Clinical utility of serodiagnostic testing in suspected pediatric inflammatory bowel disease. Am J Gastroenterol. 2001;96:758–65.
18. Dubinsky MC et al. Suspected inflammatory bowel disease – the clinical and economic impact of competing diagnostic strategies. Am J Gastroenterol. 2002;97:2333–42.
19. Khan K et al. Role of serology and routine laboratory tests in childhood inflammatory bowel disease. Inflamm Bowel Dis. 2002;8:325–9.
20. Gupta SK et al. Comparison of serological markers of inflammatory bowel disease with clinical diagnosis in children. Inflamm Bowel Dis. 2004;10:240–4.
21. Panaccione R, Sandborn WJ. Is antibody testing for inflammatory bowel disease clinically useful? Gastroenterology. 1999;116:1001–2; discussion 1002–3.
22. Joossens S et al. The value of serologic markers in indeterminate colitis: a prospective follow-up study. Gastroenterology. 2002;122:1242–7.
23. Plevy S. Do serological markers and cytokines determine the indeterminate? J Clin Gastroenterol. 2004;38(5 Suppl.):S51–6.
24. Joossens S et al. Anti-outer membrane of porin C and anti-I2 antibodies in indeterminate colitis. Gut. 2006;55:1667–9.
25. Aisenberg J et al. Are pANCA, ASCA, or cytokine gene polymorphisms associated with pouchitis? Long-term follow-up in 102 ulcerative colitis patients. Am J Gastroenterol. 2004; 99:432–41.
26. Klebl FH et al. Association of perinuclear antineutrophil cytoplasmic antibodies and anti-*Saccharomyces cerevisiae* antibodies with Vienna classification subtypes of Crohn's disease. Inflamm Bowel Dis. 2003;9:302–7.

27. Walker LJ et al. Anti-*Saccharomyces cerevisiae* antibodies (ASCA) in Crohn's disease are associated with disease severity but not NOD2/CARD15 mutations. Clin Exp Immunol. 2004;135:490–6.
28. Mow WS et al. Association of antibody responses to microbial antigens and complications of small bowel Crohn's disease. Gastroenterology. 2004;126:414–24.
29. Arnott ID et al. Sero-reactivity to microbial components in Crohn's disease is associated with disease severity and progression, but not NOD2/CARD15 genotype. Am J Gastroenterol. 2004;99:2376–84.
30. Teml A et al. Anti-*Saccharomyces cerevisiae* antibodies: a stable marker for Crohn's disease during steroid and 5-aminosalicylic acid treatment. Am J Gastroenterol. 2003;98:2226–31.
31. Canani RB et al. Effects of disease activity on anti-*Saccharomyces cerevisiae* antibodies. Implications for diagnosis and follow-up of children with Crohn's disease. Inflamm Bowel Dis. 2004;10:234–9.
32. Desir B et al. Utility of serum antibodies in determining clinical course in pediatric Crohn's disease. Clin Gastroenterol Hepatol. 2004;2:139–46.
33. Dubinsky MC et al. Serum immune responses predict rapid disease progression among children with Crohn's disease: immune responses predict disease progression. Am J Gastroenterol. 2006;101:360–7.
34. Taylor KD et al. ANCA pattern and LTA haplotype relationship to clinical responses to anti-TNF antibody treatment in Crohn's disease. Gastroenterology. 2001;120:1347–55.
35. Esters N et al. Serological markers for prediction of response to anti-tumor necrosis factor treatment in Crohn's disease. Am J Gastroenterol. 2002;97:1458–62.
36. Fleshner PR et al. High level perinuclear antineutrophil cytoplasmic antibody (pANCA) in ulcerative colitis before colectomy predicts the development of chronic pouchitis after ileal pouch–anal anastomosis. Gut. 2001;49:671–7.
37. Spivak J et al. Antibodies to I2 predict clinical response to fecal diversion in Crohn's disease. Gastroenterology. 2004;126:A69.
38. Scofield RH. Autoantibodies as predictors of disease. Lancet. 2004;363:1544–6.
39. Nielen MM et al. Specific autoantibodies precede the symptoms of rheumatoid arthritis: a study of serial measurements in blood donors. Arthritis Rheum. 2004;50:380–6.
40. Arbuckle MR et al. Development of autoantibodies before the clinical onset of systemic lupus erythematosus. N Engl J Med. 2003;349:1526–33.
41. Israeli E et al. Anti-*Saccharomyces cerevisiae* and antineutrophil cytoplasmic antibodies as predictors of inflammatory bowel disease. Gut. 2005;54:1232–6.
42. Granito A et al. Anti-*Saccharomyces cerevisiae* antibodies (ASCA) in coeliac disease. Gut. 2006;55:296.
43. Sendid B et al. Anti-*Saccharomyces cerevisiae* mannan antibodies in familial Crohn's disease. Am J Gastroenterol. 1998;93:1306–10.
44. Mei L et al. Familial expression of anti-*Escherichia coli* outer membrane porin C in relatives of patients with Crohn's disease. Gastroenterology. 2006;130:1078–85.
45. Oshitani N et al. Cross-reactivity of yeast antigens in human colon and peripheral leukocytes. J Pathol. 2003;199:361–7.
46. Vermeire S et al. Anti-*Saccharomyces cerevisiae* antibodies (ASCA), phenotypes of IBD, and intestinal permeability: a study in IBD families. Inflamm Bowel Dis. 2001;7:8–15.
47. Harrer M et al. Do high serum levels of anti-*Saccharomyces cerevisiae* antibodies result from a leakiness of the gut barrier in Crohn's disease? Eur J Gastroenterol Hepatol. 2003; 15:1281–5.

13
How to diagnose Behçet's and intestinal Behçet's disease?

A. F. ÇELIK, Ö. N. PAMUK, M. MELIKOĞLU and H. YAZICI

WHAT IS BEHÇET'S DISEASE?

Behçet's disease (BD) was first described by Hulusi Behçet, as a triad consisting of aphthous stomatitis, genital ulcers and uveitis[1]. It was later shown that many organs, including joints and other systems, e.g. pulmonary and neurological systems, can be affected[2].

Although generally considered as a vasculitis there can be lesions, e.g. in the central nervous system, in which a clear-cut vascular disease cannot be discerned. There is no distinctive histological characteristic.

The underlying cause is unknown, but it seems to be a multifactorial disease in which genetic components and external factors jointly influence the course and clinical expression of disease[3,4]. Histologically, $CD4^+$ T lymphocytes seem to be major cell type in inflammatory infiltrates[5] and peripheral T cells have a predominant Th1-type cytokine pattern[6,7], suggesting a role of T cell-mediated immune response in the pathogenesis.

PREVALENCE OF BD

The prevalence of BD is relatively higher in Asia, especially around the ancient trade line known as the 'Silk Route'. Turkey, as a bridge between Asia and Europe, has the highest prevalence (420/100 000)[8]. BD is also common in the Middle East, Korea and Japan[9,10], but rare in Europe, the UK[11], and the USA[12]. In addition to the most common presentation with mucocutaneous lesions, in which oral and genital ulcers are present along with folliculitis and sometimes arthritis, the major morbidities are ocular and nervous system involvement[13] in addition to vascular disease. Pulmonary artery aneurysms, with thrombotic events, especially cause mortality[14].

DEDICATED CENTRE EXPERIENCE ON BD AND GASTROINTESTINAL BD (GIBD)

In the past 30-year period more than 7000 BD cases were registered in our dedicated multidisciplinary Behçet centre. In this centre the patients are under the care of rheumatologists, ophthalmologists, dermatologists, neurologists, vascular surgeons and gastroenterologists.

For more than 7 years now our GI department has also been running a regular inflammatory bowel disease (IBD) clinic under a senior gastroenterologist. Currently we have 23 registered GIBD patients; of these 17 were diagnosed at this centre. The remaining six patients with BD had been referred to our unit from peripheral hospitals located either in or outside Istanbul with their GIBD diagnosis already made. Therefore, as a dedicated centre our yearly GIBD hospital incidence is 23/7 = 3.3 patients per year. According to this hospital incidence the prevalence of GIBD in our centre is less than 1%, which is in accordance with the prevalence of our recent file search of a group of BD patients younger than 18 years old[15] and a previous controlled study[16].

HOW TO DIAGNOSE BD

In diagnosis of BD the presence or a history of oral aphthae (OA) is almost a must. The International Behçet's Disease Study Group classification criteria for BD are shown in Table 1[17]. Although classification criteria of BD are sufficient to accurately classify most of the cases, some rare cases, especially those without OA, or cases without genital ulcers or lack of genital scars, may create problems in diagnosis. A positive pathergy test, which is an assessment of inflammatory skin reaction to needle-prick, may help to complete the diagnostic criteria. Since OA are also commonly seen in the normal population, to ascribe to BD one should not forget that, according to the criteria, the frequency of OA should be more than three times a year with a tendency to persist longer than usual.

OA do not seem to have any relation to GI involvement. First, in contrast to the almost ubiquitous presence of OA, GIBD is rare. While OA appear and disappear, ulcerations in GIBD persist and often show Crohn's disease (CD)-like chronicity. In our retrospective analysis of 35 Behçet's patients the upper endoscopic examinations along the GI canal distal to the oral cavity, down to the upper jejunum, we could not find any case in whom there was an ulcer, which could not be explained by other factors (unpublished observations).

It has been shown that cessation of smoking may cause flares in OA[18] and a nicotine patch[19] reduces its frequency and severity. However, smoking is one of the well-known causes which increase CD activity[20]. This effect, however, is reversed for ulcerative colitis (UC)[21]. As stressed above, OA and GIBD activities do not seem to act together; therefore, although there is no study on the effect of smoking on GIBD, the similar GI location[16] and morphology compared with CD, lead us to believe that smoking may play a role in GIBD activity.

Table 1 International Study Group criteria for the classification of BD[17]

Recurrent oral ulceration	Minor aphthous, or herpetiform ulceration, observed by physician or patient, which recurred at least three times in one 12-month period.
Plus two of:	
Recurrent genital ulceration	Aphthous ulceration or scarring, observed by physician or patient.
Eye lesions	Anterior uveitis, posterior uveitis, or cells in vitreus on slit-lamp examination, or retinal vasculitis observed by ophthalmologist.
Skin lesions	Erythema nodosum observed by physician or patient, pseudofolliculitis, or papulopustular lesion or acneiform nodules in post-adolescent patients not on corticosteroid treatment.
Positive pathergy test	Read by a physician at 24–48 h.

Genital ulcers (GU) or scar formation are strong indicators for the presence of BD; however, like other manifestations of BD, GU have a tendency to be more frequent in the early phase of the disease. This frequency diminishes with time and GU may disappear without leaving scar formation. Therefore, in the relatively late phase of the disease, only the presence of scar formation in the genital area is helpful for diagnosis. Nevertheless, genital scars may be easily missed by the inexperienced eye.

HOW COMMON IS GIBD IN BD-PREVALENT AND NON-PREVALENT COUNTRIES?

Although non-specific gastrointestinal symptoms are common in BD[16], clinically important and BD-related GI involvement is a rare event[22]. The frequency of GIBD varies widely in different countries (Table 2). While some early studies from Japan[23] reported high frequencies (50%), surprisingly, a figure of less than 1% was reported from a country where BD is endemic, i.e. Turkey[15]. Although there are reported high frequency rates, the majority indicates less than 10% frequency [11,15,16,24–33].

Why is GIBD a rare event in a country endemic for BD, such as Turkey? Why it is very common in another endemic region, e.g. Asia? Is this connected to the prevalence of IBD in these regions? There are no published reports on the prevalence of IBD in Turkey. However, according to a single large survey involving 17 000 people[34], involvement and the recent assessment of the IBD registry of the Turkish Society of Inflammatory Bowel Diseases (Epidemiology of Inflammatory Bowel Disease in Turkey, Ülku Dağli, see related chapter in this book), it is no less than 36/100.000. This moderate prevalence rate suggests that IBD is not very uncommon in Turkey; therefore, we consider that this discordance may partly arise from misdiagnosis related to an overestimation of GIBD in Asia.

Table 2 Frequency of BD gastrointestinal involvement in various countries

Reference	*Country*	*Year*	*n*	*GI involvement (%)*
Yamamoto et al.[24]	Japan	1972	2031	25
Eun et al.[25]	Korea	1984	114	5.3
Dilsen et al.[26]	Turkey	1996	496	5
Yurdakul et al.[16]	Turkey	1997	1000	0.7
Kaklamani et al.[27]	Greece	1998	64	3
Bang et al.[28]	Korea	2001	1155	4
Bang et al.[29]	Korea	2001	3497	7.3
Chang et al.[30]	Korea	2002	73	15
Tursen et al.[31]	Turkey	2003	2313	1.4
Seyahi et al.[15]	Turkey	2003	121	0.8
Chamberlain et al.[11]	UK	1977	32	10
O'Duffy et al.[32]	USA	1971	10	30
Jankowski et al.[33]	UK	1992	15	40

None of the studies listed in Table 2 has been specifically designed to discern the prevalence of GIBD. The reports quoted are from different disciplines and thus are biased towards the referred discipline. This is one of the reasons why the prevalence of GIBD can vary even in the studies that arise from the same region. Since some recent reports indicate a relatively low number of GIBD cases in Asia[24,25] it is unclear whether GIBD is as common as has been claimed in earlier reports[23].

In the countries where BD prevalence is low there is a potential for a type of referral bias. Intestinal involvement may not receive so much attention, unless sporadic cases present with severe inflammation, intestinal ulcers and bleeding. As a GI specialist if you see BD sporadically in a country with low prevalence, and come across with GIBD as a rare event, and/or when you diagnose GIBD mostly in severe cases and cases with complications, you may miss a much more mucocutaneous form of BD, which is the most common and mild form of the disease. Obviously missing these patients in a cumulative assessment would relatively increase GIBD prevalence among the BD group, and this would make GIBD prevalence much higher than expected in low prevalent countries such as the USA[32] and the UK[11].

Different prevalence rates may also reflect differences in design and methodology of the quoted papers. In this regard surveys conducted in multidisciplinary clinics, such as ours[16], we maintain, would give less biased results. In this particular study the frequency and history of diarrhoea were generally similar in patients with BD and controls. In addition, in none of the BD patients were diarrhoea and histological examination inflammatory in origin.

GIBD: DIAGNOSTIC TOOLS

Although a supportive role of radiology cannot be ignored, endoscopic examination is the gold standard for diagnosis of GIBD. However, macroscopic and microscopic details of gastrointestinal involvement are not disease-specific, and easily confused with other IBD such as CD and UC.

In a population of BD patients in which arthritis and arthralgia-related non-steroidal anti-inflammatory drugs (NSAID) use is common, even colonoscopic diagnosis of GIBD would not be good enough without controlling other factors, such as NSAID use[35,36] comorbid pathologies, and repeating the colonoscopy under these controlled circumstances.

In our Behçet's centre within a 7-year period the number of patients who suffered from abdominal complaints and having proved intestinal ulcerations with colonoscopy, was 30. However, detailed history-taking and repeating of colonoscopies within a 3-month period, indicated that nearly one-third of BD patients with colonoscopic lesions did not have real intestinal involvement of BD. They suffered rather from NSAID-related enterocolitis, or intestinal tuberculosis, antibiotic-associated haemorrhagic colitis (unpublished data). We therefore recommend repeating colonoscopy within 3–6 months after stopping NSAID and other related drugs.

In addition, most of the above-mentioned prevalence figures (Table 2) do not have endoscopic confirmation, and the methodology in many others is retrospective and not uniform. In a radiology-based study enteroclysis findings in Turkish patients with GIBD showed that the intestinal ulcers were usually shallow, multiple, and generally localized to the terminal ileum[37]. This contrasted with the findings in another recent colonoscopic study from Korea in which usually single, large, and deep ulcers with distinct borders were described[38]. Therefore, the straightforward comment is the possibility of regional differences between Turkey and Far East Asia, for the type of bowel disease as well as for the frequency. However, according to our cohort of BD patients with intestinal involvement (unpublished data) 15 out of 23 GIBD patients (70%) had been found to have single, large ulcers at endoscopy as previously described by the Korean group. This observation leads us to consider that the method of assessing the signs of inflammation in the bowel (radiological versus endoscopic or both) is quite important.

As emphasized above, in some studies, even GI symptoms have been taken as indicators of GIBD where prevalences reach up to 50–60%[23]. At the beginning of the 1970s when use of flexible endoscopic procedures spread worldwide, the prevalence of GIBD in Asia, especially in the pioneer countries of endoscopic procedures, was found to be considerably lower than before[24,25]. However, even this prevalence rate was not free from the risk of referral bias and the risk of diagnosing any irrelevant mucosal lesion as GIBD by endoscopic assessment. So, through endoscopy more accurate morphological diagnosis appeared with the risk of misclassification of a non-relevant mucosal lesion as GIBD.

On the other hand, one may consider that endoscopy might also make us aware of some cases of asymptomatic GIBD. However, as we know from IBD clinics, endoscopically documented asymptomatic involvement is rather mild and may heal without any sign of GI disease. So, as practising physicians, we

may decide not to be very interested in discerning asymptomatic BD patients who have only endoscopic mucosal evidence of GI involvement.

In IBD diagnosis histology is a non-specific diagnostic tool. Except for rare cases of caseous granuloma formation, which may indicate intestinal tuberculosis, mucosal punch biopsies may only be helpful in demonstrating acute or chronic inflammation, mainly in the mucosa and rarely in the submucosa. Although granuloma formation mostly but not necessarily indicates CD, sensitivity of mucosal punch biopsies for granuloma is low. Among CD patients granuloma formation is seen in around 25–30% in full-thickness intestinal biopsies[39,40], and its much less than this in mucosal punch biopsies. Unless it is caseous, specificity of granuloma formation is arguable, since some non-CD cases with granuloma, including GIBD, have been reported[41].

OTHER GI INVOLVEMENT

Except for relatively common hepatic venous vascular occlusion by thrombosis, Budd–Chiari syndrome, other intestinal and extraintestinal involvement such as pancreatitis is rare, and it is difficult to say that they are causally related to BD[42].

DIFFERENTIAL DIAGNOSIS

Similar to CD and intestinal tuberculosis (IT), GIBD is most commonly seen in the ileocaecal area[16,42]. Diagnosis of GIBD needs macroscopically documented inflammation in a patient with BD.

Furthermore, as with macroscopic study, microscopic details of inflammation are also not specific for GIBD and can easily be confused with other IBD, such as CD, UC, IT and some potential inflammatory causes such as NSAID use.

Similarities and dissimilarities between CD and GIBD (Table 3) may create diagnostic difficulty. Extraintestinal involvement such as arthropathy (5–20%), and ophthalmological manifestations (1.6–4.6%) are relatively rare in IBD[43],

Table 3 Comparison of GIBD and CD (similarities and dissimilarities)

Features	*GIBD*	*CD*
Non-specific histology	Yes	Yes
Major vascular involvement	Yes	No
Serious eye event	Yes	No
Central nervous system involvement	Yes	Yes
Inflammatory morphology	Yes	Yes
Ileocaecal involvement	Yes	Yes
CARD15/NOD susceptibility	No	Yes
Positive ASCA	No?	Yes

Table 4 Frequencies of Behçet's clinical features in UC and CD patients

	Crohn's Disease (n = 93)	*Ulcerative colitis (n = 130)*	*p-Value*
Oral ulcer >3/year	20/93 (20%)	32/130 (25%)	n.s.
Positive pathergy	10/93 (10%)	11/130 (9%)	n.s.
Uveitis	2/93 (2%)	2/130 (2%)	n.s.
Arthritis	3/93 (3%)	2/130 (2%)	n.s.
Nodular lesion	2/93 (2%)	3/130 (2%)	n.s.
Pustule	22/93 (24%)	23/130 (18%)	n.s.
Genital ulcer	4/93 without scar	0/130	–
No. of patients fulfilling the Behçet's criteria	0/93	1/130 (1%)	n.s.

n.s., not significant.

while it is much more common in BD (50%)[44]. Although there are as yet no controlled studies, it is generally regarded that, with more perforation and bleeding, GIBD has a more guarded prognosis when compared to CD or UC[45].

In a recent work (Table 4), we applied BD diagnostic criteria to our group of CD and UC patients. Although around 20–25% oral aphthae and 10% pathergy positivity were present in CD and UC groups, except for two patients in the UC group none of the patients fulfilled the diagnostic criteria for BD[46]. When compared with IBD the discriminatory value of BD diagnostic criteria is fairly high. A drawback of this study was the rather limited number of patients with CD or UC.

CARD15/NOD2

At least two separate papers failed to indicate an association with CARD15/NOD2 variants in Behçet's patients from Turkey and the UK[47,48]. Nevertheless, whether this is also true for GIBD has not yet been assessed. Studies similarly reporting no such association with CARD15/NOD2 mutation among Turkish CD patients[49] may simply indicate that CARD15/NOD2 is not the major genetic mutation which may be held responsible from CD-like IBD.

ANTI-*SACCHAROMYCES CEREVISIAE* ANTIBODY (ASCA)

There is conflicting evidence regarding the presence of ASCA in GIBD. Table 5 shows comparisons of the ASCA values in CD, BD, and GIBD in the available literature[50–52]. In a recent report from India, where tuberculosis is endemic, it has been shown that ASCA is 40% positive also in intestinal tuberculosis[53]. The results suggest that ASCA might be a non-specific intestinal inflammatory indicator; therefore its clinical value in differential diagnosis of bowel inflammations is not yet clear.

Table 5 Comparison of available ASCA literature in CD, BD and GIBD

		Any ASCA positivity (IgG or IgA)				*For CD*	
	Method	*Controls*	*BD*	*GIBD*	*CD*	*Sensitivity*	*Specificity*
Krause et al.[50]	ELISA	1/10 (10%)	13/27 (48%)	–	–	n.a.	n.a.
Fresko et al.[51]	ELISA	3/21 (14%)	18/85 (21%)	2/7 (28%)	15/24 (62%)	60%	62%
Choi et al.[52]	IFA	4/45 (8.8%)	1/30 (3%)	47/106 (44%)	–	53%	67%

DISEASE FOLLOW-UP

'Disease burden' of BD is usually confined to the early years of its course, and in many patients the disease 'burns out'[13]. Although during the follow-up a small number of CD patients may turn into the fibrostenotic type without any inflammatory activity, in general CD is a chronic, persistent and progressive disease. However, whether GIBD behaviour is similar to CD in this regard remains to be formally studied.

TREATMENT

Treatment modalities in GIBD are no different from those of IBD. Because of the relatively low prevalence of GIBD, placebo-controlled studies are lacking, and we mostly have to rely on case-based or small-numbered open-label limited observations.

Even open-label studies in GIBD are not available; however, it is known that there are morphological, histological and clinical similarities between IBD and GIBD. Therefore in the treatment of GIBD we have to rely on information obtained from IBD. Commonly the outcome of the treatment is mainly judged on an arbitrarily chosen subjective clinical symptomatology. As in IBD treatment protocols, 5-aminosalicylic acid (5-ASA), immunosuppressives, and biological agents, either used singly or in combination, are the three main steps of treatment in GIBD[42,54]. Except for patients with mild clinical activity and/or low colonoscopic score, we do not use 5-ASA in our IBD clinic; the same applies to our GIBD cases. As in CD and UC, mainly azathioprine (AZA), and in cases who cannot tolerate AZA, or also having arthritis, methotrexate are two drugs as immunosuppressive in GIBD treatment. As a third step, in addition to immunosuppressive treatment, we can use the anti-tumour necrosis factor infliximab[55–57] or thalidomide[58] for some patients who do not respond to immunosuppressives.

References

1. Behçet H. Über rezidivierende aphthöse, durch ein Virus Verursachte Geschwüre am Mund, am Auge und an den Genitalien. Dermatol Wochenschr. 1937;105:1152–7.
2. Shimizu T, Ehrlich GE, Inaba G et al. Behçet's disease (Behçet's syndrome). Semin Arthritis Rheum. 1979;8:223–60.
3. Yazici H, Fresko I, Yurdakul S. Behçet's syndrome: disease manifestations, management, and advances in treatment. Nat Clin Pract Rheumatol. 2007;3:148–55.
4. Direskeneli H. Autoimmunity vs autoinflammation in Behçet's disease: do we oversimplify a complex disorder? Rheumatology (Oxford). 2006;45:1461–5.
5. Ben Ahmed M, Houman H, Miled M, Dellagi K, Louzir H. Involvement of chemokines and Th1 cytokines in the pathogenesis of mucocutaneous lesions of Behçet's disease. Arthritis Rheum. 2004;50:2291–5.
6. Frassanito MA, Damacco R, Cafforio P, Dammacco F. Th1 polarization of the immune response in Behçet's disease: a putative pathogenetic role of interleukin-12. Arthritis Rheum. 1999;42:1967–74.
7. Melikoglu M, Uysal S, Krueger JG et al. Characterization of the divergent wound-healing responses occurring in the pathergy reaction and normal healthy volunteers. J Immunol. 2006;177:6415–21.

8. Azizlerli G, Köse AA, Saryca R et al. Prevalance of Behçet's disease in İstanbul, Turkey. Int J Dermatol. 2003;42:803–6.
9. Saylan T, Mat C, Fresko İ, Melikoğlu M. Behçet's disease in the Middle East. Clin Dermatol. 1999;17:209–23.
10. Marshall SE. Behçet's disease. Best Prac Res Clin Rheumatol. 2004;18:291–311.
11. Chamberlain MA. Behçet's syndrome in 32 patients in Yorkshire. Ann Rheum Dis. 1977;36:491–9.
12. O'Duffy JD. Summary of international symposium on Behçet's disease. J Rheumatol. 1978;5:229–33.
13. Kural-Seyahi E, Fresko I, Seyahi N et al. The long-term mortality and morbidity of Behçet syndrome. A 2-decade outcome durvey of 387 patients followed at a dedicated center. Medicine (Baltimore). 2003;82:60–76.
14. Hamuryudan V, Yurdakul S, Moral F et al. Pulmonary arterial aneurysms in Behçet's syndrome: a report of 24 cases. Br J Rheumatol. 1994;33:48–51.
15. Seyahi E, Ozdogan H, Yurdakul S et al. The outcome of children with Behçet's syndrome. Clin Exp Rheumatol. 2004;22(Suppl. 34):116a.
16. Yurdakul S, Tüzüner N, Yurdakul I, Hamuryudan V, Yazici H. Gastrointestinal involvement in Behçet's syndrome: a controlled study. Ann Rheum Dis. 1996;55:208–10.
17. International Study Group for Behçet's Disease. Criteria for diagnosis of Behçet's disease. Lancet. 1990;335:1078–80.
18. Soy M, Erken E, Konca K, Ozbek S. Smoking and Behçet's disease. Clin Rheumatol. 2000;19:508–9.
19. Kaklamani VG, Markkomichelakis N, Kaklamanis PG. Could nicotine be benefical for Behçet's disease. Clin Rheumatol. 2002;21:341–2.
20. Cottone M, Rossellini M, Orlanda A et al. Smoking habits and reccurence in Crohn's disease. Gastroenterology. 1994;106:643–8.
21. Jick H, Walker AM. Cigarette smoking and ulcerative colitis. N Engl J Med. 1983;308:261–3.
22. Chajec T, Fainaru M. Behçet's disease: report of 41 cases and review of the literature. Medicine. 1975;54:179–96.
23. Oshima Y, Shimizu T, Yokohari R et al. Clinical studies on Behçet's syndrome. Ann Rheum Dis. 1963;22:36–45.
24. Yamamoto S, Toyokawa H, Matsubara J et al. A nation-wide survey of Behçet's disease in Japan. 1. Epidemiological survey. Jpn J Ophthalmol. 1974;18:282–90.
25. Eun HC, Chung H, Choi SJ. Clinical analysis of 114 patients with Behçet's disease. J Korean Med Assoc. 1984;27:933–9.
26. Dilsen N, Konice M, Aral O et al. Risk factors of vital organ involvement in Behçet's disease. In: Weschler B, Godeau F, editors. Behçet's Disease. Proceedings of the Sixth International Conference on Behçet's Disease. Amsterdam: Excerpta Medica, 1993:165–9.
27. Kaklamani VG, Vaiopoulos G, Kaklamanis PG. Behçet's disease. Semin Arthritis Rheum. 1998;27:197–217.
28. Bang D, Yoon KH, Chung HG, Choi EH, Lee ES, Lee S. Epidemiological and clinical features of Behçet's disease in Korea. Yonsei Med J. 1997;38:428–36.
29. Bang D, Lee JH, Lee ES et al. Epidemiologic and clinical survey of Behçet's disease in Korea: the first multicenter study. J Korean Med Sci. 2001;16:615–18.
30. Chang HK, Kim JW. The clinical features of Behçet's disease in Yongdong districts: analysis of a cohort followed from 1997 to 2001. J Korean Med Sci. 2002;17:784–9.
31. Tursen U, Gurler A, Boyvat A. Evaluation of clinical findings according to sex in 2313 Turkish patients with Behçet's disease. Int J Dermatol. 2003;42:346–51.
32. O'Duffy JD, Carney A, Deodhar S. Behçet's disease: report of 10 cases, 3 with new manifestation. Ann Intern Med. 1971;75:561–70.
33. Jankowski J, Crombie I, Jankowski R. Behçet's syndrome in Scotland. Postgrad Med J. 1992;68:566–70.
34. Cakir N, Pamuk ON, Dervis E et al. The prevalance of rheumatologic diseases in Havsa: the initial evaluation. Annual European Congress of Rheumatology, Vienna, Austria. Ann Rheum Dis. 2005;64(Suppl. III):551.
35. Puspok A, Kiener HP, Oberhuber G. Clinical, endoscopic, and histologic spectrum of nonsteroidal anti-inflammatory drug-induced lesions in the colon. Dis Colon Rectum. 2000;43:685–91.

36. Laine L, Connors LG, Reicin A et al. Serious lower gastrointestinal clinical events with nonselective NSAID or Coxib use. Gastroenterology. 2003;124:288–92.
37. Korman U, Cantasdemir M, Kurugoglu S et al. Enteroclysis findings of intestinal Behçet disease: a comparative study with Crohn disease. Abdom Imaging. 2003;28:308–12.
38. Kim JS, Lim SH, Choi IJ et al. Prediction of the clinical course of Behçet's colitis according to macroscopic classification by colonoscopy. Endoscopy. 2000;32:635–40.
39. Ramzan NN, Leighton JA, Heigh RI, Shapiro MS. Clinical significance of granuloma in Crohn's disease. Inflamm Bowel Dis. 2002;8:168–73.
40. Pulimood AB, Ramakrishna BS, Kurian G et al. Endoscopic mucosal biopsies are useful in distinguishing granulomatous colitis due to Crohn's disease from tuberculosis. Gut. 1999;45:537–41.
41. Fukuda Y, Watanabe I. Pathological studies on intestinal Behçet's (entero-Behçet's) disease. In: Dilşen N, Koniçe M, Övül C, editors. Behçet's Disease. Proceedings of an international symposium on Behçet's disease. London: Royal Society of Medicine Services, 1986:289.
42. Celik AF, Hatemi I. Gastrointestinal involvement of Behçet's syndrome. Turkiye Klinikleri J Int Med Sci. 2005;1:48–54.
43. Su CG, Judge TA, Leichtenstein GR. Extraintestinal manifestations of inflammatory bowel disease. Gastroenterol Clin N Am. 2002;31:307–21.
44. Yurdakul S, Yazici H, Tuzun Y et al. The arthritis of Behçet's disease: a prospective study. Ann Rheum Dis. 1983;42:505–15.
45. Chou SJ, Chen TK, Jan HC, Lou MA, Liu YM. Intestinal perforations in Behçet's disease. J Gastrointest Surg. 2007;11:508–14.
46. Hatemi I, Hatemi G, Celik A et al. İnflamatuvar barsak hastalığında Behçet sendromu sıklı ğı. Romatoloji Kongre Kitabı. 2006, İzmir, 115:127.
47. Uyar FA, Saruhan-Direskeneli G, Gül A. Common Crohn's disease-predisposing variants of the CARD15/NOD2 gene are not associated with Behçet's disease in Turkey. Clin Exp Rheumatol. 2004;22(Suppl. 34):S50–2.
48. Ahmad T, Zhang L, Gogus F et al. CARD15 polymorphisms in Behçet's disease. Scand J Rheumatol. 2005;34:233–7.
49. Ozen SC, Dagli U, Kilic MY et al. NOD2/CARD15, NOD1/CARD4, and ICAM-1 gene polymorphisms in Turkish patients with inflammatory bowel disease. J Gastroenterol. 2006;41:304–10.
50. Krause I, Monselise Y, Milo G, Weinberger A. Anti-*Saccharomyces cerevisiae* antibodies – a novel serologic marker for Behçet's disease. Clin Exp Rheumatol. 2002;20(Suppl. 26):S21–4.
51. Fresko I, Ugurlu S, Ozbakir F et al. Anti-*Saccharomyces cerevisiae* antibodies (ASCA) in Behçet's syndrome. Clin Exp Rheumatol. 2005;23(Suppl. 38):S67–70.
52. Choi CH, Kim TI, Kim BC et al. Anti-*Saccharomyces cerevisiae* antibody in intestinal Behç et's disease patients: relation to clinical course. Dis Colon Rectum. 2006;49:1849–59.
53. Govid KM, Sachdev V, Gupta R, Lal S, Pandey RM. Anti-*Saccharomyces cerevisiae* antibody does not differentiate between Crohn's disease and intestinal tuberculosis. Dig Dis Sci. 2007;52:33–9.
54. Naganuma M, Iwao Y, Inoue N et al. Analysis of clinical course and long-term prognosis of surgical and nonsurgical patients with intestinal Behçet's disease. Am J Gastroenterol. 2000;95:2848–51.
55. Hassard PV, Binder SW, Nelson V, Vasiliauskas EA et al. Anti-tumor necrosis factor monoclonal antibody therapy for gastrointestinal Behçet's disease. Gastroenterology. 2001;120:995–9.
56. Travis SP, Czajkowski M, McGovern DP et al. Treatment of intestinal Behçet's syndrome with chimeric tumor necrosis factor alpha antibody. Gut. 2001;49:725–8.
57. Lee JH, Kim TN, Choi ST et al. Remission of intestinal Behçet's disease treated with anti-tumor necrosis factor alpha monoclonal antibody (infliximab). Korean J Intern Med. 2007;22:24–7.
58. Celik AF, Seyahi EK, Coskun S et al. Thalidomide in azathioprine resistant gastrointestinal (GI) Behçet's syndrome: 3 cases. Clin Exp Rheumatol. 2004;22(Suppl. 34).

14
Stool tests: are they useful?

A. LOGANAYAGAM and I. BJARNASON

INTRODUCTION

Most gastroenterologists aspire to diagnose and manage their inflammatory bowel disease (IBD) patients *par excellence*. Fortunately this is not difficult today with the availability of pan-intestinoscopy and biopsy and a standardized treatment algorithm relying on surgeons to save the day when medical treatment fails or complications set in. There are nevertheless some specific problems and challenges that face gastroenterologists and their patients. The most obvious one is the sheer number of patients referred for differentiating between the irritable bowel syndrome (IBS) and IBD. Many of these will undergo some form of invasive endoscopy or radiological imaging. While this may ensure job security for gastroenterologists, service providers are increasingly demanding a more rapid and streamlined diagnostic service at a lower cost. Secondly, once the diagnosis has been made, the main concern is the imminence of the clinical relapse, which is the main factor contributing to the low quality of life in IBD patients[1,2]. Gastroenterologists are notoriously bad at predicting clinical relapse, which translates to the indiscriminate use of 5-aminosalicylic acid preparations during periods of quiescent disease while other patients may be subjected to immune suppression with its attendant side-effects[3].

While endoscopy in many ways provides an ideal method for diagnosis it is self-evident that it only provides a static morphological picture of the intestine. However, the gastrointestinal tract is a complex organ with an extraordinarily varied biological function. Because morphology and function may go hand in hand many clinical scientists have tried to devise simple functional tests which might indicate the presence of disease when impaired. The potential use of such tests, if of adequate sensitivity, is that they might be used for non-invasive diagnostic screening, they may provide the cause or mechanism for clinical symptoms (e.g. malabsorption in a wasted patient) and they may predict the natural history of the disease, which is especially relevant to IBD patients[4].

Intestinal function tests have come and gone. Many have been discredited by sloppy clinicians who do not understand the complexities and influence of test dose composition on the sensitivity and specificity of the tests (e.g. intestinal permeability testing and D-xylose absorption), let alone the importance of

careful marker analyses[5]. However, perhaps the most important factor for the lack of recognition of non-invasive gastrointestinal testing is excessive reliance on endoscopy teaching in many gastroenterology-training programmes. Nevertheless our non-gastroenterology colleagues appear to have grasped the fact that morphology and function yield complementary information. Hence neurologists seek cerebrovascular fluid, respiratory physicians sputum, rheumatologists synovial fluid and nephrologists urine for analyses, in addition to imaging methods. By analogy gastroenterologists might be expected to examine faeces. This may not be the case in practice, but where do we currently stand in the use of faecal tests in IBD?

HISTORY OF FAECAL TESTS

Ancient Egyptians, with their desire for eternal life in a healthy body, were the first civilizations to document the practice of macroscopic coproscopy by a *swnw* (physician)[6]. This practice reached its peak in the 18th and 19th centuries[7]. Inspection of stool it is still widely practised in certain cultures and by nutritionists while the gastroenterologist relies on it only rarely in order to document worm infestations, fat malabsorption or intestinal bleeding. Microscopy of stools is an essential component of a work-up for infectious diarrhoea[8,9] and molecular amplification of specific gene products is a rapidly evolving method[10], which holds the promise of non-invasive diagnosis of colorectal cancer. The 1950s–1970s saw the introduction of a variety of radioisotopic tests whereby gastrointestinal function could be inferred from faecal analyses of the radioisotope (e.g. ^{51}chromium labelled red cells (for bleeding), ^{51}chromium labelled albumen (for intestinal protein loss), ^{111}indium white cells (for inflammation) which were uniquely sensitive and accurate, but impractical for routine use. Ingenious clinical biochemists developed techniques to measure stool osmolarity and faecal reducing substances (mostly requested by paediatricians), faecal fat and blood, etc. However, with the widespread availability of radioimmunoassay and enzyme-linked immunoabsorbent assay (ELISA) it became possible to analyse specific proteins on an industrial scale in faeces. An offspring of this were assays of neutrophil selective proteins, which allow a quantitative assessment of intestinal inflammation. This is highly relevant to gastroenterologists because almost all intestinal diseases are associated with an inflammatory reaction.

ASSESSING INTESTINAL INFLAMMATION

Microscopy of stools in search of inflammatory cells was once in wide use, but the method lacks sensitivity, especially for more proximal disease as intestinal bacteria degrade the white cells. The measurement of key components of the inflammatory cascade, such as tumour necrosis factor, were also used as a measure of intestinal inflammation, but these substances are also unfortunately subjected to bacterial degradation[11,12], limiting the practical uses of the tests.

The gold standard for quantitative assessment of intestinal inflammation came with the validation and application of the ^{111}indium white cell technique[13,14]. The technique involved isolation of patients' own neutrophils, labelling with ^{111}indium, injection of the labelled cells and a 4-day faecal collection for analyses and calculation of ^{111}indium activity. The beauty of the method is that the neutrophils are selectively labelled, the ^{111}indium was firmly bound to the cellular DNA and on entering the intestine there is no significant absorption of the radioisotope even when the cells are disrupted. The faecal excretion of ^{111}indium over 4 days provided a quantitative measure of intestinal inflammation which correlated with histopathological indices of inflammatory activity in patients with ulcerative colitis and clinical disease activity in Crohn's disease[13–16]. Virtually all patients with active IBD had faecal excretion of ^{111}indium way above controls, and the inflammatory activity decreased with successful treatment. It was suggested that the technique could be used to discriminate, with an accuracy approaching 100%, between patients with IBD and IBS at the first outpatient visit. However, the method was not disease-specific (it is specific for intestinal inflammation). Indeed it was used to define a number of enteropathies (non-steroidal anti-inflammatory drugs, alcohol, chronic renal failure, hypogammaglobulinaemia, HIV-AIDS, etc.) where none was suspected or impossible to demonstrate by other techniques[17–19]. ^{99m}Tc labelling of white cells[20] and E-selectin scanning[21] provided similar quality intestinal images, but again the cost and radiation precluded their widespread use.

FAECAL MARKERS

Any new method for assessing intestinal function by analyses of faecal markers needs to compare its sensitivity, specificity and practicability with that of the golden standard, namely the ^{111}indium white cell technique. The next development was to assess neutrophil-specific proteins in the stool, which would relate quantitatively to the degree of intestinal inflammation. The most important quality that such proteins must have is that they resist bacterial degradation and are not reabsorbed from the gastrointestinal tract. Calprotectin is certainly resistant to degradation[22] whilst the data for lactoferrin, neutrophil-specific elastase and myeloperoxidase are more controversial[23].

Most of the studies of clinical data on these markers have involved measurement of calprotectin, which serves as a blueprint for the other markers. Calprotectin accounts for up to 50% of the neutrophil cytosol protein and is easily measured in faeces by a commercially available ELISA in as little as 200 mg of stool. Lactoferrin resides within cytoplasmic granules within the neutrophils and can be assayed by radioimmunoassay or ELISA.

The faecal calprotectin technique has been extensively validated. There is some day-to-day variation of the concentrations and there is small intra-stool variability[24,25]. Faecal calprotectin concentrations correlate significantly with the 4-day faecal excretion of ^{111}indium white cells in patients with IBD (Figure 1)[24,26,27] and with histopathological indices of inflammation in ulcerative

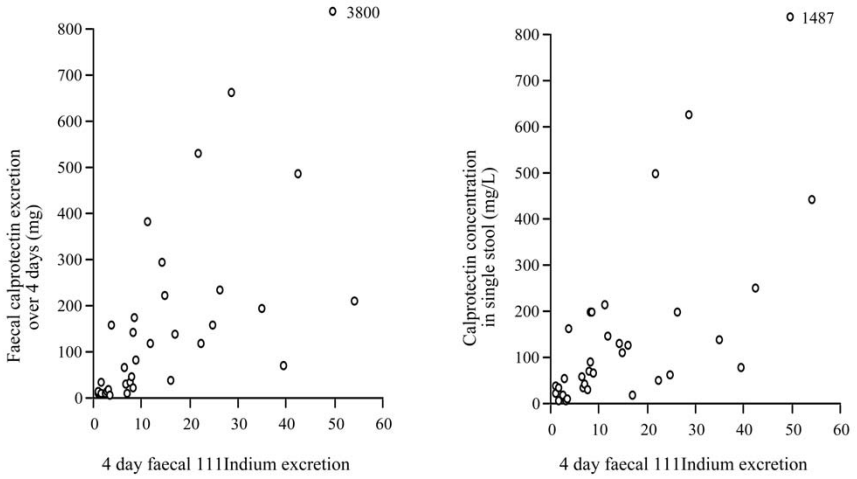

Figure 1 Correlation between faecal calprotectins and the 4-day faecal excretion of ^{111}indium labelled white cells in patients with Crohn's disease. The left graph shows that the total amount of calprotectin excreted over 4 days correlates significantly with the 4-day excretion of the labelled white cells. The right graph shows that the correlation holds when single stool calprotectin concentrations are used to assess intestinal inflammation

colitis[28]. The validation of lactoferrin has not been as extensive, but faecal values of lactoferrin and calprotectin correlate significantly and the methods are purported to have comparable sensitivity for the detection of inflammation[29–32] although calprotectin appears to outperform lactoferrin at times[23,33].

USES IN IBD

The use of the faecal markers in patients with IBD relates to at least four distinctively different clinical situations.

Diagnostic screen

Active IBD is defined on symptomatology and laboratory investigation, which reflect activation of the acute inflammatory response whereby the neutrophils are the final common effector cell. Because of the intestinal bacteria almost all colonic diseases are associated with an inflammatory reaction. However, the inflammation in active IBD is an order of magnitude greater than most other intestinal diseases[17].

A number of studies show that faecal calprotectin is vastly increased in active IBD as compared with healthy controls. Indeed no other laboratory parameter, apart from the ^{111}indium white cell faecal excretion, is as sensitive for the detection of Crohn's disease.

In the UK 30–40% of all the referrals of general practitioners to hospital-based gastroenterologists are diagnosed as IBS while 5–10% turn out to have IBD. IBS and IBD patients share many symptoms. However, although the Rome criteria are a surprisingly good guide for the diagnosis of IBS, most gastroenterologists proceed to and rely on laboratory tests to aid in the differential diagnosis. These tests include intestinal imaging (endoscopy or radiology) which come at a great cost and inconvenience to patients and are certainly not without risk. The question is then whether assessing intestinal inflammation can be used in the initial work-up of such patients in order to discriminate between patients with IBS, who will not benefit from invasive investigation, and those with IBD who require these tests for diagnosis.

One study in over 225 patients showed that a cut-off of three-fold above the normal upper limit of normal had a 100% sensitivity and 94% specificity for this purpose[24]. Other studies show similar sensitivity[23,34–39]. Tibble et al. addressed this issue in greater detail in a study of 602 patients referred by general practitioners to King's College Hospital with various lower intestinal symptoms[40]. In most cases the requests raised the possibility of IBD in patients thought to have IBS. Gastroenterologists who used their own conventional diagnostic methods saw these patients; however, all the patients also filled out a symptom questionnaire (Rome) and underwent full blood counts, routine biochemical screening, erythrocyte sedimentation rate (ESR), C-reactive protein (CRP) and faecal calprotectin (the Rome and calprotectin results were not available to the gastroenterologists).

The calprotectin results in these patients are shown in Figure 2. Most noticeable is the fact that the vast majority of patients with 'organic' disease have increased faecal calprotectin concentrations. Secondly 85% of patients with IBS have normal values. Given a patient who had positive Rome criteria and normal calprotectin there is such an overwhelming likelihood of the patient having IBS that the authors suggested that no invasive imaging procedure was necessary in these cases. Adhering to these suggestions saves over £200 000 on imaging per gastroenterologist at King's College Hospital each year. Colonoscopy waiting times for those who do not adhere to these guidelines are about 18 weeks whilst the rest of us have a 2-week wait.

The finding that perhaps 15% of patients diagnosed with IBS have a degree of intestinal inflammation is also of interest and in keeping with other research[41,42]. The question of whether these patients should be treated differently from the IBS patient who does not have inflammation awaits study.

Given the above findings, why is the faecal calprotectin method not in greater use? Perhaps the concern of gastroenterologists is that some patients with organic intestinal disease will be incorrectly diagnosed if excess reliance is placed upon the calprotectin results. More importantly, however, is their worry about job security if they reduce the demand for their colonoscopy services. A potential drop in income clearly plays an additional role, especially to those who prioritize status and wealth over patient welfare.

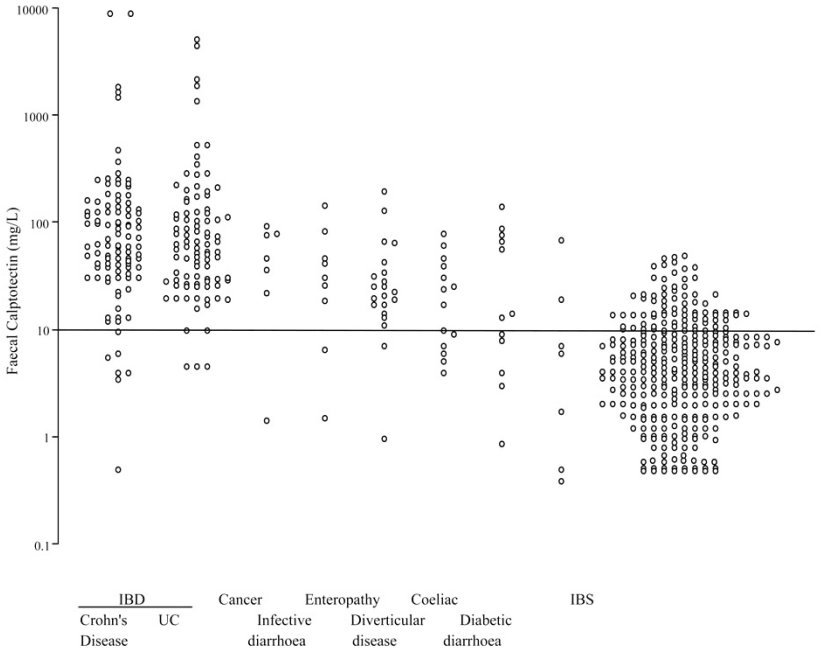

Figure 2 Faecal calprotectin concentrations in 602 consecutive patients referred to a gastroenterology outpatient clinic and their final diagnosis. The figure shows that most patients with organic intestinal disease have increased intestinal inflammation. Most patients (85%) with the irritable bowel syndrome have calprotectin values within the control range

Predicting clinical relapse of IBD

Once IBD is diagnosed the treatment involves induction and subsequently maintenance of remission based largely on clinical disease activity indices[43–45] and the physician's global assessment of well-being. Once a clinical remission is achieved the problem facing the patient and physicians alike is how long this remission will last. Until recently there were no sensitive and specific indicators of imminent relapse. There are nevertheless some clinical parameters and blood tests (ESR, orosomucoid, CRP, platelet, and white cell counts, interleukin 6 (IL-6), tumour necrosis factor alpha (TNF-α) and IL-1)[46–50], which reflect the systemic consequences of inflammation, that have been used as predictors and/or markers of clinical relapse of IBD. However, their use has in general been disappointing, due to the fact that they are non-specific, affected by a variety of non-intestinal diseases[51] and most importantly do not measure intestinal inflammation directly. Although sigmoidoscopy can predict imminent relapse of ulcerative colitis this is not a practical way of predicting relapse. The situation is more complex in Crohn's disease where there is a major discrepancy between clinical symptoms and macroscopic appearances.

An interesting finding in Crohn's disease is the universal agreement that normal intestinal permeability in asymptomatic patients heralds a good short-term (6–12 months) prognosis (90% predictive value) while increased intestinal permeability is predictive (65% predictive value) of a clinical relapse[52–57]. However, these tests are not widely available, marker analyses are demanding, the tests differ in test dose composition and sensitivity and there is an inconvenience factor for the patient having to carry out the test at home which has limited their use[4,5]. Furthermore the tests are not useful in patients with ulcerative colitis.

Faecal calprotectins outperform any other method for predicting clinical relapse in patients with both ulcerative colitis and Crohn's disease. Tibble et al. showed in 81 patients with asymptomatic IBD that a five-fold elevation of calprotectin had a 90% sensitivity and 83% specificity for predicting clinical relapse of disease[58]. Using a similar analysis with somewhat different cut-off levels Costa et al. reached a similar conclusion[59]. Lastly high faecal calprotectins in asymptomatic patients following surgery are a sensitive indicator for early recurrence of disease[60]. The implications of these studies are clear. Clinical practice dictates that the vast majority of asymptomatic IBD patients are on long-term 5-aminosalicylic acids with a view of preventing the time to and severity of clinical relapse. Most of these patients do not benefit from this treatment, and they can be identified by the calprotectin method. It remains to be seen if the same goes for patients on long-term azathioprine. Secondly, raised calprotectin levels in asymptomatic patients offer the possibility of treating these patients before the disease gets out of control. The time seems to have arrived for gastroenterologists to take lead from rheumatologists and start treating the inflammatory component of IBD in order to alter the natural history of the disease rather than waiting for the clinical relapse with all its attendant morbidity and slight mortality.

Assessing response to treatment

Medical treatment of IBD is simple and can be carried out according to the various algorithms. All of these have a fixed treatment protocol and are critically dependent on symptomatic responses. The dose of drugs and length of treatment are often empirically derived with a significant number of patients re-experiencing symptoms when drug dosage is reduced. The possibility is that symptoms improve prior to mucosal healing, which should be the objective of treatment.

There is relatively sparse data on faecal calprotectins during treatment of acute IBD. However, in a recent study by Roseth et al., infliximab treatment in naive patients (n = 14) was followed up with temporal assessments of faecal calprotectin, at time 0, week 4 and 8[61]. At time 0 the median calprotectin level was 2200 mg/kg, at week 4 it was 250 mg/kg and at week 8 it was 50 mg/kg. The CRP levels were 16, 3 and 3, respectively ($n < 10$ mg/L).

Interestingly, CRP was normal at week 4, in contrast to the significantly elevated calprotectin at 250 mg/kg, further confirming that CRP is not able to detect smouldering inflammation in patients with Crohn's disease. Furthermore, half the patients had normal calprotectin at week 8, which is a strong suggestion of mucosal healing.

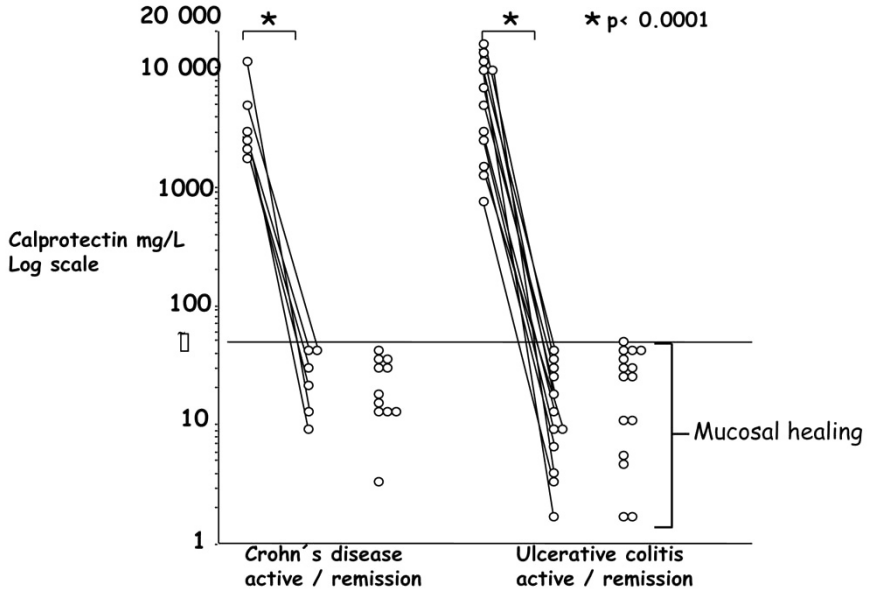

Figure 3 Calprotectin levels in IBD patients with active disease and during mucosal healing. The figure shows high calprotectin levels in active disease and normalization with near-complete healing

In a study using normalization of calprotectin in patients with Crohn's disease or ulcerative colitis (Figure 3) as the sole inclusion criterion, 45/45 patients had complete mucosal healing, and 38/45 had histological healing; seven patients had some minor infiltration of white cells not affecting the crypts or the epithelium[61].

Research potential

Having access to a non-invasive specific quantitative test to assess intestinal inflammation it would seem that the use of these tests for research and discovery would be limited only by the ingenuity of the clinical scientist. The small bowel has until now been relatively inaccessible for research, but with the advent of these tests there has been an explosion of data, and much remains to be done. One of the most fascinating aspects of gastrointestinal research is the interaction between the intestinal barrier function and development of intestinal inflammation. The basic idea is that the intestinal barrier separates the organism from the luminal intestinal contents, which contain a very high concentration of digestive enzymes and bacteria, both of which may contribute to intestinal damage when exposed to the mucosa. Previous research has indeed shown that the permeability changes can be the cause or consequence of the

inflammation and vice-versa[17]. In a comprehensive summary of available data it is shown that almost all small intestinal diseases are characterized by nearly equal quantitative changes in intestinal permeability and inflammation[17]. This is irrespective of whether agents that destroy the intestinal barrier, agents that increase the aggressiveness of luminal contents, infections or immune deficiency, bring about the intestinal damage. This shows that the body reacts to the intestinal contents in a restricted unified manner. Furthermore it emphasizes that currently available functional tests cannot be used for diagnosis. However, it is equally evident that almost all small bowel diseases are and will be associated with similar impairments in complications (such as intestinal bleeding and protein loss) with minor variations in frequency and severity. The only small bowel disease that does not seem to follow this pattern is small bowel Crohn's disease which is associated with an inflammatory response that appears to be disproportionate to the permeability changes (Table 1). The reasons for this are likely to be a key for the elucidating the pathogenesis of the disease.

An example of the research potential that the faecal calprotectin test has comes from family studies of patients with IBD. It has been recognized for a long time that a small number of first-degree relatives of patients with Crohn's disease have increased intestinal permeability[62–65]. This suggested that increased small intestinal permeability might play an aetiological or pathophysiological role in the development of the disease as opposed to the clinical relapse. Realizing that these tests are relatively insensitive for localized small bowel disease Thjodleifsson, in collaboration with deCODE Genetics in Iceland, measured faecal calprotectin levels in 151 of 220 (58%) asymptomatic

Table 1 Permeability and inflammatory changes in various conditions

	Permeability	*Inflammation*
NSAID	2–5	3–5
Alcohol	2–5	3
Renal failure	2–3	5
Radiation	2–3	3
Cytotoxic drugs	2–4	3
Giardia	2	2
Salmonella	2–5	8
Gastroenteritis	2–5	3
Rotavirus	2–6	2
Cystic fibrosis	3–5	2–3
Necrotizing enterocolitis	2–3	2
Diabetic diarrhoea	2–3	2
Coeliac disease	2–5	2
Crohn's disease	2–3	2–4
Active Crohn's disease	2–5	15–20
HIV-AIDS	2–10	2–4
Hypogammagobulinaemia	3	7
Pouchitis	2	2–5

Intestinal permeability and inflammation have been measured in these conditions.

The overall results show that there is a uniform increase (2–5-fold) in intestinal permeability and inflammation apart from the inflammation in active Crohn's disease, which is an order of magnitude greater than the other enteropathies.

first-degree relatives of 49 patients with Crohn's disease in Iceland[66]. Forty-nine per cent had raised faecal calprotectin levels, showing subclinical intestinal inflammation. Moreover the inheritance pattern conformed to an additive trait (variant component analyses). This is the first functional abnormality found in Crohn's disease patients that conforms to a recognizable inheritance pattern. It is suggested that a number of genes interact in as yet unknown manner, and that this results in asymptomatic intestinal inflammation[66]. Furthermore it is suggested that an environmental factor is responsible for translating this state to the full clinical syndrome of Crohn's disease.

Even more interesting is the suggestion that patients with idiopathic ankylosing spondylitis have ileal inflammation, which is difficult to distinguish from Crohn's ileitis. Mielants et al. have been instrumental in documenting this asymptomatic ileitis, and suggest that it represents subclinical Crohn's disease[67–71]. What is so remarkable is that, when Bjarnason et al. studied 124 of 213 (58%) asymptomatic first-degree relatives of 47 patients with ankylosing spondylitis in Iceland, 41% of them had the same increase in faecal calprotectin, and that the inheritance pattern was identical to the Crohn's disease relatives[72]. Furthermore the asymptomatic relatives with the highest calprotectin levels had computerized tomography changes suggestive of early ankylosing spondylitis (suggesting that the intestinal inflammation plays a pathogenetic role in the spondylarthropathy. Finally, in one of the most comprehensive analyses of its kind, involving deCODE's databases that contain genealogical information on all people in Iceland during the past 11 centuries, all living Icelanders with ankylosing spondylitis (n = 205) and Crohn's disease (n = 1184) it is shown that patients with ankylosing spondylitis and Crohn's disease are significantly more related (relative risk 3.3) than 10 000 matched random controls, further suggesting that these two diseases are genetically similar[72].

CONCLUSIONS

Much work remains to be done in order to position intestinal function tests within the working environment of the gastroenterologist. Nevertheless we are in possession of tests that have the potential to revolutionize our approach to the treatment of patients with IBD. The faecal calprotectin and other neutrophil-specific tests are only the first wave of techniques that allow non-invasive assessment of specific and selective cellular components of the intestinal inflammatory cascade. Already these methods are useful for a variety of purposes, outlined above, but it is likely that in the future it will be possible to estimate the participation of other cells. For instance mast cells and eosinophils carry enzymes (e.g. tryptase and chymase) that are specifically associated with these cells and, depending on the stability, it may be possible to assay these in faeces. The long-term objective is a fully automatic faecal sample assay method that provides specific information on the activity of acute inflammation (neutrophils), chronic inflammation (T cells) and allergy (mast cells).

References

1. Irvine EJ, Feagan B, Rochon J et al. Quality of life: a valid and reliable measure of therapeutic efficacy in the treatment of inflammatory bowel disease. Gastroenterology. 1994;106:287–96.
2. Bernklev T, Jahnsen J, Henriksen M et al. Relationship between sick leave, unemployment, disability, and health-related quality of life in patients with inflammatory bowel disease. Inflamm Bowel Dis. 2006;12:402–12.
3. Timmer A, McDonald JW, Macdonald JK. Azathioprine and 6-mercaptopurine for maintenance of remission in ulcerative colitis. Cochrane Database Syst Rev. 2007: CD000478.
4. Bjarnason I, Macpherson AJM, Hollander D. Intestinal permeability: an overview. Gastroenterology. 1995;108:1566–81.
5. Bjarnason I, Menzies IS. Causes and consequences of altered gut permeability. In: Brostoff J, Challacombe S, editors. Food Allergy and Intolerance, 2nd edn. London: Saunders, Elsevier Science, 2002:241–57.
6. Nunn JF. Ancient Egyptian Medicine. London: British Museum Press, 1996.
7. Porter R. Antiquity. In: Porter R, editor. The Greatest Benefit to Mankind. London: Harper-Collins, 1997:44–82.
8. Harris JC, Dupont HL, Hornick RB. Fecal leukocytes in diarrheal illness. Ann Intern Med. 1972;76:697–703.
9. Huicho L, Sanchez D, Contreras M et al. Occult blood and fecal leukocytes as screening tests in childhood infectious diarrhea: an old problem revisited. Pediatr Infect Dis J. 1993; 12:474–7.
10. Dong SM, Traverso G, Johnson C et al. Detecting colorectal cancer in stool with the use of multiple genetic targets. J Natl Cancer Inst. 2001;93:858–65.
11. Sharpstone D, Rowbottom A, Lepper M, Nelson M, Gazzard BG. Faecal tumour necrosis factor-alpha in HIV-related diarrhoea. AIDS. 1996;10:989–94.
12. Lettesjo H, Hansson T, Peterson C et al. Detection of inflammatory markers in stools from patients with irritable bowel syndrome and collagenous colitis. Scand J Gastroenterol. 2006;41:54–9.
13. Saverymuttu SH, Peters AM, Lavender JP, Pepys MB, Hodgson HJ, Chadwick VS. Quantitative faecal indium-111 labelled leucocyte excretion in assessment of disease activity in Crohn's disease. Gastroenterology. 1983;85:1333–9.
14. Saverymuttu SH, Camilleri M, Rees H, Cavender TP, Hodgson HJF. Indium-111 granulocyte scanning comparison with colonoscopy, histology and faecal indium-111 excretion in assessing disease extent and activity in colitis. Gastroenterology. 1986;90: 1121–8.
15. Saverymuttu SH. Clinical remission in Crohn's disease – assessment using faecal Indium-111 granulocyte excretion. Digestion. 1986;33:74–9.
16. Saverymuttu SH, Peters AM, Hodgson HJ, Chadwick VS, Lavender JP. Indium-111 autologous leucocyte scanning: comparison with radiology for imaging the colon in inflammatory bowel disease. Br Med J. 1982;285:255–7.
17. Bjarnason I, Takeuchi K, Bjarnason A, Adler SN, Teahon K. The G.U.T. of gut. Scand J Gastroenterol. 2004;39:807–15.
18. Bjarnason I, Sharpstone D, Francis N et al. Intestinal inflammation, ileal structure and function in HIV. AIDS. 1996;10:1385–91.
19. Bjarnason I, Maiden L, Takeuchi K. Testing the gut and its function – faecal samples – breath tests and more? In: Adler G, Blum AL, Blum HE et al., editors. Gastroenterology Yesterday, Today, Tomorrow: A Review and Preview. Falk Symposium 144. Dordrecht: Springer, 2005:253–66.
20. Becker W, Schoman E, Fischbach W, Borner W, Gruner KR. Comparison of the 99Tcm-HMPAO and 111In-oxine labelled granulocytes in man: first clinical results. Nucl Med Commun. 1988;9:435–47.
21. Bleeker-Rovers CP, Boerman OC, Rennen HJ, Corstens FH, Oyen WJ. Radiolabeled compounds in diagnosis of infectious and inflammatory disease. Curr Pharm Des. 2004; 10:2935–50.

22. Roseth AG, Fagerhol MK, Aadland E, Schjonsby H. Assessment of the neutrophil dominating calprotectin in feces. A methodologic study. Scand J Gastroenterol. 1992;27: 793–8.
23. Silberer H, Kuppers B, Mickisch O et al. Fecal leukocyte proteins in inflammatory bowel disease and irritable bowel syndrome. Clin Lab. 2005;51:117–26.
24. Tibble J, Teahon K, Thjodleifsson B et al. A simple method for assessing intestinal inflammation in Crohn's disease. Gut. 2000;47:506–13.
25. Tibble J, Sigthorsson G, Foster R, Scott D, Roseth A, Bjarnason I. Faecal calprotectin: a simple method for the diagnosis of NSAID-induced enteropathy. Gut. 1999;45:362–6.
26. Roseth AG, Schmidt PN, Fagerhol MK. Correlation between faecal excretion of Indium-111-labelled granulocytes and calprotectin, a granulocyte marker protein, in patients with inflammatory bowel disease. Scand J Gastroenterol. 1999;34:50–4.
27. Gaya DR, Lyon TD, Duncan A et al. Faecal calprotectin in the assessment of Crohn's disease activity. Q J Med. 2005;98:435–41.
28. Roseth AG, Aadland E, Jahnsen J, Raknerud N. Assessment of disease activity in ulcerative colitis by faecal calprotectin, a novel granulocyte marker protein. Digestion. 1997;58:176–80.
29. Larsen A, Hovdenak N, Karlsdottir A, Wentzel-Larsen T, Dahl O, Fagerhol MK. Faecal calprotectin and lactoferrin as markers of acute radiation proctitis: a pilot study of eight stool markers. Scand J Gastroenterol. 2004;39:1113–18.
30. Langhorst J, Elsenbruch S, Mueller T et al. Comparison of 4 neutrophil-derived proteins in feces as indicators of disease activity in ulcerative colitis. Inflamm Bowel Dis. 2005;11: 1085–91.
31. D'Inca R, Dal Pont E, Di Leo V et al. Calprotectin and lactoferrin in the assessment of intestinal inflammation and organic disease. Int J Colorectal Dis. 2007;22:429–37.
32. Scarpa M, D'Inca R, Basso D et al. Fecal lactoferrin and calprotectin after ileocolonic resection for Crohn's disease. Dis Colon Rectum. 2007;50:861–9.
33. Wildt S, Nordgaard-Lassen I, Bendtsen F, Rumessen JJ. Metabolic and inflammatory faecal markers in collagenous colitis. Eur J Gastroenterol Hepatol. 2007;19:567–74.
34. Carroccio A, Iacono G, Cottone M et al. Diagnostic accuracy of fecal calprotectin assay in distinguishing organic causes of chronic diarrhea from irritable bowel syndrome: a prospective study in adults and children. Clin Chem. 2003;49:861–7.
35. Bunn SK BW, Main MJ, Grav ES, Olson S, Golden B. Fecal calprotectin as a measure of disease activity in childhood inflammatory bowel disease. J Pediatr Gastroenterol Nutr. 2001;32:171–7.
36. Costa F, Mumolo MG, Bellini M et al. Role of faecal calprotectin as non-invasive marker of intestinal inflammation. Dig Liver Dis. 2003;35:642–7.
37. Dolwani S, Metzner M, Wassell JJ, Yong A, Hawthorne AB. Diagnostic accuracy of faecal calprotectin estimation in prediction of abnormal small bowel radiology. Aliment Pharmacol Ther. 2004;20:615–21.
38. Canani RB, de Horatio LT, Terrin G et al. Combined use of noninvasive tests is useful in the initial diagnostic approach to a child with suspected inflammatory bowel disease. J Pediatr Gastroenterol Nutr. 2006;42:9–15.
39. Bremner A, Roked S, Robinson R, Phillips I, Beattie M. Faecal calprotectin in children with chronic gastrointestinal symptoms. Acta Paediatr. 2005;94:1855–8.
40. Tibble J, Sigthorsson G, Foster R, Fagerhol M, Forgacs I, Bjarnason I. The use of surrogate markers of intestinal inflammation and symptom based criteria to distinguish organic from non-organic disease. Gastroenterology. 2002;123:450–60.
41. Liebregts T, Adam B, Bredack C et al. Immune activation in patients with irritable bowel syndrome. Gastroenterology. 2007;132:913–20.
42. Spiller RC. Inflammation as a basis for functional GI disorders. Best Pract Res Clin Gastroenterol. 2004;18:641–61.
43. Harvey RF, Bradshaw JM. A simple index of Crohn's disease activity. Lancet. 1980;1:514.
44. Best WR, Becktel JM, Singleton JW, Kern R. Development of a Crohn's disease activity index. Gastroenterology. 1976;70:439–44.
45. Truelove SC, Witts LJ. Cortisone in ulcerative colitis; final report on a therapeutic trial. Br Med J. 1955;2:1041–8.

46. Louis E, Belaiche J, van Kemseke C et al. A high serum concentration of interleukin-6 is predictive of relapse in quiescent Crohn's disease. Eur J Gastroenterol Hepatol. 1997;9: 939–44.
47. Wright JP, Young GO, Tigler Wybrandi N. Predictors of acute relapse of Crohn's disease. A laboratory and clinical study. Dig Dis Sci. 1987;32:164–70.
48. Kjeldsen J, Lauritsen K, De Muckadell OB. Serum concentrations of orsomucoid improved decision-making for tapering prednisolone therapy in patients with active inflammatory bowel disease? Scand J Gastroenterol. 1997;32:933–41.
49. Schreiber S, Nikolaus S, Hampe J et al. Tumour necrosis factor a and interleukin 1b in relapse of Crohn's disease. Lancet. 1999;353:459–61.
50. Timmer A, Sutherland LR, Martin F. Oral contraceptive use and smoking are risk factors for relapse in Crohn's disease. The Canadian Mesalamine for Remission of Crohn's Disease Study Group. Gastroenterology. 1998;114:1143–50.
51. Hodgson HJF. Laboratory markers of inflammatory bowel disease. In: Allan RN, Rhodes JM, Haunauer SB, Keighly MRB, Alexander-Williams J, Fazio FW, editors. Inflammatory Bowel Disease, 3rd edn. New York: Churchill Livingstone, 1997:329–34.
52. Teahon K, Smethurst P, Macpherson AJ, Levi AJ, Menzies IS, Bjarnason I. Intestinal permeability in Crohn's disease and its relation to disease activity and relapse following treatment with elemental diet. Eur J Gastroenterol and Hepatol. 1993;5:79–84.
53. Wyatt J, Vogelsang H, Hubl W, Waldhoer T, Lochs H. Intestinal permeability and the predictor of relapse in Crohn's disease. Lancet. 1993;341:1437–9.
54. Arnott ID, Kingstone K, Ghosh S. Abnormal intestinal permeability predicts relapse in inactive Crohn disease. Scand J Gastroenterol. 2000;35:1163–9.
55. D'Inca R, Di Leo V, Corrao G et al. Intestinal permeability test as a predictor of clinical course in Crohn's disease. Am J Gastroenterol. 1999;94:2956–60.
56. Jorgensen J, Ranlov PJ, Bjerrum PJ, Diemer H, Bisgaard K, Elsborg L. Is an increased intestinal permeability a valid predictor of relapse in Crohn disease? Scand J Gastroenterol. 2001;36:521–7.
57. Hilsden RJ, Meddings JB, Hardin J, Gall DG, Sutherland LR. Intestinal permeability and postheparin plasma diamine oxidase activity in the prediction of Crohn's disease relapse. Inflamm Bowel Dis. 1999;5:85–91.
58. Tibble J, Sigthorsson G, Fagerhol M, Bjarnason I. Surrogate markers of intestinal inflammation are predictive for relapse in patients with inflammatory bowel disease. Gastroenterology. 2000;119:15–22.
59. Costa F, Mumolo MG, Ceccarelli et al. Calprotectin is a stronger predictive marker of relapse in ulcerative colitis than in Crohn's disease. Gut. 2005;54:364–8.
60. Orlando A, Modesto I, Castiglione F et al. The role of calprotectin in predicting endoscopic post-surgical recurrence in asymptomatic Crohn's disease: a comparison with ultrasound. Eur Rev Med Pharmacol Sci. 2006;10:17–22.
61. Roseth AG, Aadland E, Grzyb K. Fecal calprotectin: a predictor of mucosal healing in patients with inflammatory bowel disease. Scand J Gastroenterol. 2004;39:1017–20.
62. Hollander D. Crohn's disease – A permeability disorder of the tight junctions? Gut. 1988; 26:1621–4.
63. Katz KD, Hollander D, Vadheim CM et al. Intestinal permeability in patients with Crohn's disease and their healthy relatives. Gastroenterology. 1989;97:927–31.
64. May GR, Sutherland LR, Meddings JB. Is small intestinal permeability really increased in relatives of patients with Crohn's disease? Gastroenterology. 1993;104:1627–32.
65. Teahon K, Smethurst P, Levi AJ, Menzies IS, Bjarnason I. Intestinal permeability in patients with Crohn's disease and their first degree relatives. Gut. 1992;33:320–3.
66. Thjodleifsson B, Sigthorsson G, Cariglia N et al. Subclinical intestinal inflammation: an inherited abnormality in Crohn's disease relatives? Gastroenterology. 2003;124:1728–37.
67. Mielants H, Veys EM, Cuvelier C, De Vos M. Subclinical involvement of the Gut in undifferentiated spondylarthropathies. Clin Exp Rheumatol. 1989;7:499–504.
68. Cuvelier C, Barbatis C, Mielants H, De Vos, Veys E, Roels H. The histopathology of intestinal inflammation in reactive arthritis. Gut. 1987;28:394–402.
69. Mielants H, Veys EM, Cuvelier C et al. The evolution of spondylarthropathies in relation to gut histology. II. Histological aspects. J Rheumatol. 1995;22:2273–8.

70. Mielants H, Veys EM, Goemaere S, Cuvelier C, De Vos M. A prospective study of patients with spondylarthropathy with special reference to HLA-B27 and gut histology. J Rheumatol. 1993;20:1353–8.
71. Mielants H, Veys EM, Cuvelier C et al. The evolution of spondylarthropathies in relation to gut histology. III. Relation between gut and joints. J Rheumatol. 1995;22:2279–84.
72. Bjarnason I, Helgason KO, Geirsson AJ et al. Sub-clinical intestinal inflammation sacroiliac changes in first-degree relatives of patients with ankylosing spondylitis. Gastroenterology. 2003;125:1598–605.

Section VI
Features of drugs used in inflammatory bowel disease

Chair: A NAKOS and DK PODOLSKY

15
Features of drugs used in inflammatory bowel disease: 5-aminosalicylic acid (mesalazine)

U. KLOTZ

INTRODUCTION

It is generally accepted that chronic inflammatory bowel disease (IBD) is not caused by a single defect but a multiplicity of environmental (e.g. microbial), genetic and (innate) immunological factors. Currently it is controversially discussed whether intervention in a specific pathway should be followed or whether a more general approach (by 'dirty' drugs) should be continued[1,2].

Despite a better understanding of the pathogenesis of IBD medical therapy is still concentrating on treating signs and symptoms of the inflammatory conditions and maintaining the disease in remission. The development of biological agents, e.g. specific antibodies targeting tumour necrosis factor (TNF-α), could not replace traditional agents such as 5-aminosalicylic acid (5-ASA; mesalazine) which still represents a drug of first choice in the treatment of mild to moderate active IBD, in particular ulcerative colitis (UC), and maintaining these patients in remission[3–5].

PLACE OF 5-ASA IN THERAPY OF IBD

There is general consensus that the well-tolerated 5-ASA provides beneficial effects in the management of UC[4]. In regard to the optimal dosing there is increasing evidence that dose escalation from 1.2 g/day to 2.4 g/day or 4.8 g/day can achieve a better overall improvement and a more delayed relapse[5–7]. Concerning the management of Crohn's disease (CD) the role of 5-ASA is less clear and discussed controversially[8,9]. It would be very helpful if (non-)response to treatment or the risk for relapse could be predicted by biomarker(s), a topic which should be more extensively studied[10]. According to recent consensus conferences Table 1 summarizes the current place of 5-ASA in the management of IBD[11–13].

Table 1 Place of 5-ASA in the management of IBD*

Ulcerative colitis (UC)	*Crohn's disease (CD)*
First choice in mild to moderate active UC	The benefit of 5-ASA is limited in active CD
If remission has been achieved medically, maintenance with 5-ASA should be preferred	If remission has been achieved medically, maintenance with 5-ASA is a treatment option
For patients in remission on 5-ASA treatment should be extended for 1 year at least	For patients in remission on 5-ASA cessation of treatment may be considered after 2 years of full remission
5-ASA can reduce risk for colon cancer	
According to guidelines of German and Austrian Consensus Conferences published in Z Gastroenterol. 2004;42:979–83 and 2006;44:525–38	ECCO consensus statements published in Gut. 2006;55(Suppl. I):I16–35

*According to guidelines of the IBD section of the British Society of Gastroenterology, published in Gut. 2004;53(Suppl. V):V1–16.

During the past few years a new indication for 5-ASA became apparent. UC (and maybe also CD) is associated with an increased risk of colorectal carcinoma. Based on several observational or epidemiological studies it could be shown for patients with UC that regular intake of 5-ASA can reduce the risk of developing colorectal cancer by about 50%. For this chemopreventive effect a daily dose of at least 1.2 g 5-ASA is probably needed[14–19].

PHARMACOLOGICAL FEATURES OF 5-ASA

As in IBD local inflammatory processes in the intestinal (sub-)mucosa of the small and large bowel are involved, the therapeutic principle 5-ASA should be targeted to the affected areas. For this reason various modified-release formulations for oral intake, as well as suppositories, enemas and foam for rectal application, have been developed[20–23]. Following oral or rectal administration the released 5-ASA is taken up by the epithelial cells of the gut. During its absorption 5-ASA is already partly presystemically acetylated in the intestinal wall by N-acetyltransferase 1 to the major, inactive metabolite N-Ac-5-ASA. Subsequently 5-ASA is further acetylated during its first passage through the liver. N-Ac-5-ASA is finally eliminated by glomerular filtration and active tubular secretion into the urine (24–41% of the dose). Unabsorbed 5-ASA, as well as drug and N-Ac-5-ASA secreted back into the gut lumen[24], are also excreted with the faeces (35–55% of the dose). In Table 2 the pharmacokinetic properties of 5-ASA have been summarized.

Selection among the various formulations of 5-ASA should be guided by the proximal extent of the disease[22]. Enemas distribute from the rectum and sigma up to the transverse colon (about 25% of the dose is absorbed). With foam 5-ASA will reach the proximal sigmoid colon. When given as suppositories 5-ASA will be delivered only to the rectosigmoid region (absorption rate about

Table 2 Pharmacokinetic properties of 5-ASA

Parameter (ranges)	*5-ASA*	*Ac-5-ASA*
$t_{\frac{1}{2}}$ (h)	0.5–2.4*	1.3–11
CL (ml/min)	300–690*	200–300 (renal *CL*)
Dialysance (ml/min)	–	60–100
Protein binding (%)	43	78
V (L/kg)	0.3–0.5	–
Renal excretion (% of dose)	6–13	24–41
Faecal excretion (% of dose)	13–28	15–25
Biliary excretion (% of dose)	0.01–0.75 (total 5-ASA)	
Excretion into breast milk (mg in 120–200 ml)	0.01–0.02	1.5–3.6

*Dose dependent (nonlinear pharmacokinetics); $t_{\frac{1}{2}}$: elimination half-life; *CL*: total (systemic) clearance; *V*: apparent volume of distribution.

13% of the dose). Apparently the local (mucosal) concentrations of 5-ASA are determinants of its clinical response[25–27] and combining rectal and oral administration of 5-ASA will achieve several-fold higher target levels in the colon and rectum than oral therapy alone[28] and combining 2 g b.i.d. of oral 5-ASA with a 1 g enema at bedtime resulted in a superior therapy in patients with extensive active UC[29].

More recently some new delivery systems for 5-ASA have been developed. Enteric-coated micropellet formulations have been designed to optimize drug delivery to the ileocaecal region[30]. The highest clinical remission rates (66%) were achieved in patients with active UC with a dosage of 1.0 g three times a day[31] and 5-ASA pellets were as effective as 5-ASA tablets in a comparative study[32]. Likewise, multi-matrix (MMx) tablets consist of a gastroresistant coated core of 5-ASA incorporated in microparticles of a lipophilic matrix dispersed within a hydrophilic matrix. The release of 5-ASA is slow and gradual (80% of the absorption occurs in the colon) and with 1.2 g 5-ASA t.i.d. clinical remission was 60% after 8 weeks in patients with UC[33]. In a small trial lower 8-week remission rates (0%, 31%, 18%) have recently been reported for MMx-mesalazine doses of 1.2, 2.4 and 4.8 g/day, respectively[34].

The intestinal release pattern of the various 5-ASA preparations has been extensively studied by pharmacokinetic and scintigraphic approaches. In general the median systemic exposure to 5-ASA ranging from about 30% to 50% is comparable for all oral formulations[35] and further advances in drug delivering/targeting could optimize patient compliance[36], which is still an important determinant of clinical outcome[37].

In addition, in experimental colitis models some new prodrugs have been investigated in which 5-ASA is conjugated either with NO-donators[38], a platelet-activating factor antagonist[39] or taurine[40]. In the future such compounds with 'dual' activity might increase response rates.

Concerning the mode of action of 5-ASA several effects have been observed. The 'polypotent' agent might act by blocking the production of proinflammatory prostaglandins and leukotrienes, by impairing production/release of interleukin 1/8 and TNF-α, by inhibiting chemotaxis and adhesion of inflammatory cells, by inhibiting the activation of the nuclear factor (NF)-

κB, by scavenging free (toxic) oxygen radicals[41], by inducing changes in apoptosis and proliferation[42] or by promotion of intestinal epithelial wound healing[43]. Based on the experimental evidence it seems that inhibition of the activation of NF-κB [44,45] and the radical scavenging properties of 5-ASA[46,47] are somewhat favoured. More recently it is believed that 5-ASA acts by activating a class of nuclear receptors involved in the control of inflammation, cell proliferation, apoptosis and metabolic function or of the γ-form of peroxisome proliferator-activated receptors (PPAR-γ). The receptors are expressed at high levels in colon epithelial cells, where their expression appears to be at least partly stimulated by gut bacteria[48]. In respect to the chemopreventive potential of 5-ASA it was shown that 5-ASA affects cell cycle progression in colorectal cells, which increases the maintenance of genomic stability and counteracts carcinogenesis[49]. Thus, based on the most likely mode of action, the development of novel pro-drugs of 5-ASA might be a rational approach for future research.

SAFETY OF 5-ASA

Compared to its preceding drug sulphasalazine (SZ) 5-ASA exhibits a lower toxicity and is much better tolerated by patients[50]. For these reasons 5-ASA has replaced the azo-compound. The incidence of allergic reactions (mostly due to the sulpha-component of SZ) is about 10-fold lower for 5-ASA compared to SZ. In addition, spontaneously reported serious adverse events are significantly more frequent with SZ (odds ratio 1.31; 95% CI 1.22–1.40) than with 5-ASA[51]. In a review of fully published randomized trials it was concluded that frequencies of adverse reactions or withdrawal due to adverse events of patients treated with 5-ASA were comparable to those in placebo-treated patients and lower than those in SZ-treated patients[52].

Based on animal experiments the kidney was regarded as a potential target for toxicity. In extensive controlled clinical studies any changes observed in the excretion of renal marker proteins in patients with IBD could be attributed to the disease activity rather than to treatment with 5-ASA[53–56]. This has been confirmed in large epidemiological studies. According to the UK general practice research database the incidence of renal disease in adult patients with IBD not taking 5-ASA was higher (0.25 cases per 100 patients per year) than in IBD patients on 5-ASA, which affected only 0.17 cases[57]. Based on data from a postal questionnaire to 1298 British gastroenterologists and 290 consultants of the Renal Association one case with 5-ASA-induced impairment in kidney function in 4000 patients per year was retrospectively estimated[58]. In 153 outpatients with CD mean exposure to 5-ASA was 8.6 years and the cumulative dose of 5-ASA amounted to 9 kg. Within 11 years a mean decline in endogenous creatinine clearance of 0.3 ml/min per year could be assessed, which did not exceed the expected age-related decline[59]. Thus, any concern in terms of renal toxicity is not justified by published data. However, monitoring of kidney function during long-term therapy with high-dose 5-ASA appears advisable, especially in patients with pre-existing renal impairment or when other potentially nephrotoxic drugs are taken concomitantly.

CONCLUSIONS

As IBD seems to be caused by multiple factors polypotent ('dirty') drugs such as 5-ASA still offer a rational and safe approach for treatment, especially as 5-ASA has demonstrated a very low potential for adverse events. The clinical efficacy of 5-ASA could be improved by optimizing its targeted delivery. Topical treatment with an ointment and spray has recently been applied successfully in a paediatric patient with oral and pharyngeal CD[60]. Furthermore, comfortable dosing schedules could increase patients' compliance, which presently is less than 80% in 43% of outpatients receiving delayed-release preparations of 5-ASA[61]. By intensifying research for predictive biomarkers[10] identification of subgroups of (non)-responders to 5-ASA (and other agents) could also contribute to a more effective treatment of IBD.

In conclusion, the polypotent 5-ASA still represents a first-line drug for the management of IBD, particularly UC. In numerous studies 5-ASA has proven its clinical efficacy and safety. Its topical action and pharmacological features have been well described.

Acknowledgements

This work was supported by the Robert Bosch Foundation, Stuttgart (Germany). The secretarial help of Mrs. U. Hengemühle is greatly appreciated.

References

1. Fiocchi C. The multiple components of inflammatory bowel disease pathogenesis: should we invest in all of them or should we pick and choose? Curr Opin Gastroenterol. 2005;21:399–400.
2. Sandborn WJ. What's new: innovative concepts in inflammatory bowel disease. Colorect Dis. 2006(Suppl. I):3–9.
3. Hanauer SB. New lessons: classic treatments, expanding options in ulcerative colitis. Colorect Dis. 2006;8(Suppl. I):20–4.
4. Bergman R, Parkes M. Systematic review: the use of mesalazine in inflammatory bowel disease. Aliment Pharmacol Ther. 2006;23:841–55.
5. Hanauer SB. Review article: High-dose aminosalicylates to induce and maintain remission in ulcerative colitis. Aliment Pharmacol Ther. 2006;24(Suppl. 3):37–40
6. Hanauer SB, Sandborn WJ, Kornbluth A et al. Delayed-release oral mesalamine at 4.8 g/day (800 mg/tablet) for the treatment of moderately active ulcerative colitis: the ASCEND II trial. Am J Gastroenterol. 2005;100:2478–85.
7. Paoluzi OA, Iacopini F, Pica R et al. Comparison of two different daily dosages (2.4 vs. 1.2 g) of oral mesalazine in maintenance of remission in ulcerative colitis patients: 1-year follow-up study. Aliment Pharmacol Ther. 2005;21:1111–19.
8. Hanauer SB. The case for using 5-aminosalicylates in Crohn's disease. Inflamm Bowel Dis. 2005;11:609–12.
9. Stange EF. The case against using 5-aminosalicylates in Crohn's disease. Inflamm Bowel Dis. 2005;11:613–15.
10. Pardi DS, Sandborn WJ. Predicting relapse in patients with inflammatory bowel disease: what is the role of biomarkers? Gut. 2005;54:321–2
11. Hoffmann JC, Zeitz M, Bischoff SC et al. Diagnostik und Therapie der Colitis ulcerosa: Ergebnisse einer evidenzbasierten Konsensuskonferenz der Deutschen Gesellschaft für Verdauungs- und Stoffwechselkrankheiten zusammen mit dem Kompetenznetz chronisch entzündlicher Darmerkrankungen. Z Gastroenterol. 2004;42:979–83.

12. Dejaco C, Haas T, Kirchgatterer A et al. Aminosalicylate und Steroide in der Behandlung von chronisch entzündlichen Darmerkrankungen – Konsensuspapier der Arbeitsgruppe für chronisch entzündliche Darmerkrankungen der ÖGGH. Z Gastroenterol. 2006;44:525-38.
13. Travis SPL, Stange EF, Lémann M et al. European evidence based consensus on the diagnosis and management of Crohn's disease: current management. Gut. 2006;55(Suppl. I):I16–35.
14. Velayos FS, Terdiman JP, Walsh JM. Effect of 5-aminosalicylic use on colorectal cancer and dysplasia risk: a systemic review and metaanalysis of observational studies. Am J Gastroenterol. 2005;100:1345–53.
15. van Staa TP, Card T, Logan RF, Leufkens HGM. 5-Aminosalicylate use and colorectal cancer risk in inflammatory bowel disease: a large epidemiological study. Gut. 2005;54:1573–8.
16. Giannini EG, Kane SV, Testa R, Savarino V. 5-ASA and colorectal cancer chemoprevention in inflammatory bowel disease: can we afford to wait for 'best evidence'? Dig Liver Dis. 2005;37:723–31.
17. Meinkholm P, Loftus Jr EV, Reinacker-Schicke A et al. Prevention of colorectal cancer in inflammatory bowel disease: value of screening and 5-aminosalicylates. Digest. 2006;73:11–19.
18. Stange EF. Review article: The effect of aminosalicylates and immunomodulation on cancer risk in inflammatory bowel disease. Aliment Pharmacol Ther. 2006;24(Suppl. 3):64–7.
19. Rubin DT, Losavio A, Yadron N et al. Aminosalicylate therapy in the prevention of dysplasia and colorectal cancer in ulcerative colitis. Clin Gastroenterol Hepatol. 2006;4:1346–50.
20. Klotz U, Schwab M. Topical delivery of therapeutic agents in the treatment of inflammatory bowel disease. Adv Drug Deliv Rev. 2005;57:267–79.
21. Klotz U. Colonic targeting of aminosalicylates for the treatment of ulcerative colitis. Dig Liver Dis. 2005;37:381–8.
22. Gionchetti P, Rizello F, Morselli C, Tambasco R, Campieri M. Review article. Aminosalicylates for distal colitis. Aliment Pharmacol Ther 2006;24(Suppl. 3):41–4.
23. Schwab M, Klotz U. Pharmacokinetic considerations in the treatment of inflammatory bowel disease. Clin Pharmacokinet. 2001;40:723–51.
24. Xin H, Schwab M, Klotz U. Transport studies with 5-aminosalicylate. Eur J Clin Pharmacol. 2006;62:871–5.
25. Frieri G, Pimpo MT, Andreoli A et al. Prevention of postoperative recurrence of Crohn's disease requires adequate mucosal concentrations of mesalazine. Aliment Pharmacol Ther. 1999;13:557–62.
26. Frieri G, Giacomelli G, Pimpo M et al. Mucosal 5-aminosalicylic acid concentrations inversely correlate with severity of colonic inflammation in patients with ulcerative colitis. Gut. 2000;47:410–14
27. Naganuma M, Iwao Y, Ogata H et al. Measurement of colonic mucosal concentrations of 5-aminosalicylic acid is useful for estimating its therapeutic efficacy in distal ulcerative colitis: comparison of orally administered mesalamine and sulfasalazine. Inflamm Bowel Dis. 2001;7:221–5.
28. Frieri G, Pimpo MT, Palumbo GC et al. Rectal and colonic mesalazine concentrations in ulcerative colitis: oral vs. oral plus topical treatment. Aliment Pharmacol Ther. 1999;13:1413–17.
29. Marteau P, Propert CS, Lindgren S et al. Combined oral and enema treatment with Pentasa (mesalazine) is superior to oral therapy alone in patients with extensive mild/moderate active ulcerative colitis: a randomized, double blind, placebo controlled stuy. Gut. 2005;54:960–5.
30. Brunner M, Greinwald R, Kletter K et al. Gastrointestinal transit and release of 5-aminosalicylic acid from 153 Sm-labelled mesalazine pellets vs. tablets in male healthy volunteers. Aliment Pharmacol Ther. 2003;17:1163–9.
31. Kruis W, Bar-Meir S, Feher J et al. The optimal dose of 5-aminosalicylic acid in active ulcerative colitis: a dose finding study with newly developed mesalamine. Clin Gastroenterol Hepatol. 2003;1:36–43.

32. Marakhouski Y, Fixa B, Holoman J et al. A double-blind dose escalating trial comparing novel mesalazine pellets with mesalazine tablets in active ulcerative colitis. Aliment Pharmacol Ther. 2005;21:133–40.
33. Prantera C, Viscido A, Biancone L et al. A new oral delivery system for 5-ASA: preliminary clinical findings for MMx. Inflamm Bowel Dis. 2005;11:421–7.
34. D'Haens G, Hommes D, Engels L et al. Once daily MMX mesalazine for the treatment of mild-to-moderate ulcerative colitis: a phase II, dose-ranging study. Aliment Pharmacol Ther. 2006;24:1087–97.
35. Sandborn WJ, Hanauer SB. Systematic review: the pharmacokinetic profiles of oral mesalazine formulations and mesalazine pro-drugs used in management for ulcerative colitis. Aliment Pharmacol Ther. 2003;17:29–42.
36. Cohen RD. Review article: Evolutionary advances in the delivery of aminosalicylates for the treatment of ulcerative colitis. Aliment Pharmacol Ther. 2006;24:465–74.
37. López-Sanromán A, Bermejo F. Review article: How to control and improve adherence to therapy in inflammatory bowel disease. Aliment Pharmacol Ther. 2006;24(Suppl. 3):45–9
38. Wallace JL. Nitric oxid-releasing mesalamine: potential utility for treatment of inflammatory bowel disease. Dig Liver Dis. 2003;35(Suppl. 2):S35–40.
39. Gálvez J, Garrido M, Rodriguez-Cabezas ME et al. The intestinal anti-inflammatory activity of UR-12746S on reactivated experimental colitis is mediated through downregulation of cytokine production. Inflamm Bowel Dis. 2003;9:363–71.
40. Jung Y, Kim H-H, Kim H et al. Evaluation of 5-aminosalicyltaurine as a colon-specific prodrug of 5-aminosalicylic acid for treatment of experimental colitis. Eur J Pharm Sci. 2006;28:26–33.
41. MacDermott RP. Progress in understanding the mechanisms of action of 5-aminosalicylic acid. Am J Gastroenterol. 2000;95:3343–5.
42. Reinacker-Schick A, Seidensticker F, Petrasch S et al. Mesalazine changes apoptosis and proliferation in normal mucosa of patients with sporadic polyps of the large bowel. Endoscopy. 2000;32:245–54.
43. Baumgart DC, Vierziger K, Sturm A et al. Mesalamine promotes intestinal epithelial wound healing *in vitro* through a TGF-beta-independent mechanism. Scand J Gastroenterol. 2005;40:958–64.
44. Bantel H, Berg C, Vieth M et al. Mesalazine inhibits activation of transcription factor NF-κB in inflammed mucosa of patients with ulcerative colitis. Am J Gastroenterol. 2000;95:3452–7.
45. Gan H-T, Chen Y-Q, Quyang Q. Sulfasalazine inhibits activation of nuclear factor-κB in patients with ulcerative colitis. J Gastroenterol Hepatol. 2005;20:1016–24.
46. Allgayer H. Review article: Mechanism of action of mesalazine in preventing carcinoma in inflammatory bowel disease. Aliment Pharmacol Ther. 2003;18(Suppl. 2):10–14.
47. López-Alarcón C, Rocco C, Lissi E et al. Reaction of 5-aminosalicylic acid with peroxyl radicals: protection and recovery by ascorbic acid and amino acids. Pharm Res. 2005;22:1642–8.
48. Desreumaux P, Ghosh S. Review article: Mode of action and delivery of 5-aminosalicylic acid – new evidence. Aliment Pharmacol Ther. 2006;24(Suppl. 1):2–8.
49. Luciani MG, Campregher C, Fortune JM et al. 5-ASA affects cell cycle progression in colorectal cells by reversibly activating a replication checkpoint. Gastroenterology. 2007;132:221–5.
50. D'Haens G, van Bodegraven AA. Mesalazine is safe for the treatment of IBD. Gut. 2004;53:155.
51. Ransford RAJ, Langman MJS. Sulphasalazine and mesalazine: serious adverse reactions re-evaluated on the basis of suspected adverse reaction reports to the Committee on Safety of Medicine. Gut. 2002;51:536–9.
52. Loftus Jr EV, Kane SV, Bjorkman D. Systematic review: short-term adverse effects of 5-aminosalicylic agents in the treatment of ulcerative colitis. Aliment Pharmacol Ther. 2004;19:179–89.
53. Herrlinger KR, Noftz MK, Fellermann K et al. Minimal renal dysfunction in inflammatory bowel disease is related to disease activity but not to 5-ASA use. Aliment Pharmacol Ther. 2003;15:369–9.

54. Fraser JS, Muller AF, Smith DJ, Newman DJ, Lamb EJ. Renal tubular inquiry is present in acute inflammatory bowel disease prior to the introduction of drug therapy. Aliment Pharmacol Ther. 2001;15:1131–7.
55. Mahmud N, O'Toole D, O'Hae N et al. Evaluation of renal function following treatment with 5-aminosalicylic acid derivatives in patients with ulcerative colitis. Aliment Pharmacol Ther. 2002;16:207–15.
56. Dehmer C, Greinwald R, Loffler J et al. No dose-dependent tubulotoxicity of 5-aminosalicylic acid: a prospective study in patients with inflammatory bowel diseases. Int J Colorectal Dis. 2003;18:406–12.
57. van Staa TP, Travis S, Leufkens HG, Logan RF. 5-Aminosalicylic acids and the risk of renal damage: a large British epidemiologic study. Gastroenterology. 2004;126:1733–9.
58. Müller AF, Stevens PE, McIntyre AS, Ellison H, Logan RF. Experience of 5-aminosalicylate nephrotoxicity in the United Kingdom. Aliment Pharmacol Ther. 2005;21:1217–24.
59. deJong DJ, Tielen J, Habraken CM, Wetzels JFM, Naber AHJ. 5-Aminosalicylates and effects on renal function in patients with Crohn's disease. Inflamm Bowel Dis. 2005;11:972–6.
60. Otake K, Uchida K, Inoue M et al. Successful treatment with topical 5-aminosalicylic acid ointment and spray of refractory oral and pharyngeal ulcerations in a child with Crohn disease. J Ped Gastroenterol Nutr. 2007;44:378–81.
61. Shale MJ, Riley SA. Studies of compliance with delayed-release mesalazine therapy in patients with inflammatory bowel disease. Aliment Pharmacol Ther. 2003;18:191–8.

16
Immunosuppressive drugs in inflammatory bowel disease: azathioprine

I. ATREYA and M. F. NEURATH

INTRODUCTION

Active flares of inflammatory bowel disease (IBD) are characterized by an overwhelming immune response of lamina propria T lymphocytes in the gut, resulting in severe mucosal inflammation and tissue damage. Aiming at the control and inhibition of this harmful pathogenetic T cell-mediated process, immunosuppressive therapy plays a major role in the treatment of IBD. Azathioprine and its metabolite 6-mercaptopurine represent the first-line immunosuppressive drugs for maintenance of remission in this field.

The initial fundament of azathioprine therapy was created in 1957 by Elion and Hitchings[1], who developed this immunosuppressive drug in order to improve the rate of inactivation of its parent drug 6-mercaptopurine and who were awarded the Nobel Prize in 1988 for this work. Only 5 years later, in 1962, the potential value of this new drug for IBD therapy was recognized by Robert Bean[2], who described for the first time the successful use of 6-mercaptopurine in the treatment of a patient with severe ulcerative colitis. Today, after 45 years of clinical experience with azathioprine as an immunosuppressive tool in IBD therapy, azathioprine is considered as classic immunosuppressive drug with a well-confirmed pattern of therapeutic effects in Crohns' disease (CD), as well as in ulcerative colitis (UC).

With regard to this strong and evidence-based clinical establishment of azathioprine in the therapy of IBD, it seems amazing that the exact molecular mechanism of action of azathioprine is still an object of ongoing research and a matter of debate. During recent years a number of new and unexpected data have been generated concerning the function of azathioprine at the cellular and molecular level.

On the one hand this chapter aims at describing the high clinical standing and position of azathioprine in current IBD therapy, and on the other hand at summarizing recent data, concerning the molecular mechanism of action of this classic immunosuppressive drug.

AZATHIOPRINE AS AN IMMUNOSUPPRESSIVE TOOL IN IBD THERAPY

Since 1962, when Robert Bean tested the efficiency of 6-mercaptopurine for the treatment of IBD for the first time[2], the immunosuppressive properties, the benefit in IBD therapy and the safety of azathioprine and 6-mercaptopurine have been confirmed in a large number of clinical trials[3–6] and meta-analyses[7–9]. Today it is broadly accepted that azathioprine represents the gold standard for maintenance of remission in CD[10,11], shows mentionable steroid-sparing effects[9] and even supports the induction of remission in combination with steroids in severe acute flares of CD[11]. In addition, recent studies suggest beneficial effects of azathioprine and 6-mercaptopurine for prevention of postoperative relapse in patients with CD. Including 131 CD patients, Hanauer et al.[12] showed in a placebo-controlled 2-year trial that postoperative treatment with 6-mercaptopurine was more effective than placebo in preventing clinical and endoscopic recurrence. Consistently, Myrelid et al.[13] described in a subset of CD patients with an aggressive disease course, that treatment with azathioprine after surgery prolonged the time to symptomatic relapse and reduced symptoms after surgery significantly. Referring to the European evidence-based consensus on the management of CD[14], it is recommended to use azathioprine or 6-mercaptopurine in CD patients who have severe relapse, who require two or more corticosteroid courses within one calendar year, whose disease relapses as the dose of corticosteroids is reduced below 15 mg or who relapse within 3 months of stopping corticosteroids or, in case of fistulizing or extensive disease, as a postoperative prophylaxis. In case of UC the use of azathioprine may be advisable for patients who have failed or cannot tolerate standard maintenance therapy with mesalazine or sulphalazine or who require repeated courses of corticosteroids to induce remission[15].

Regarding the long-term effectiveness of azathioprine in IBD, it is important to differentiate between CD and UC. A European multicentre study in 1176 patients provided strong evidence that azathioprine is effective in the long-term treatment of patients with UC, for at least 4 years, with clinical benefit in both disease activity and steroid requirement[10]. When azathioprine treatment of UC patients was discontinued after more than 4 years in the same study, there was a trend towards reactivation of disease activity, with an increase in the flare incidence and in steroid consumption. In contrast, discontinuation of azathioprine treatment in CD patients after 4 years did not result in any clinical deterioration, nor in increased flare incidence or steroid requirement. Therefore this study suggested that azathioprine treatment can be safely stopped after 3–4 years of therapy in those CD patients, who have been in complete remission without requiring steroids while on azathioprine therapy, but that it is recommendable to continue azathioprine treatment for more than 4 years in all other CD patients and in UC patients in general[10].

MOLECULAR MECHANISM OF ACTION OF AZATHIOPRINE – RECENT INSIGHTS

With regard to this central role of azathioprine in different therapeutic settings, it is understandable that there is a strong interest in the understanding of the exact molecular mechanism of this effective immunosuppressive drug. For a long period of time there had been only a vague hypothesis about this topic, which tried to explain the capacity of the purine-agonist azathioprine to mediate immunosuppressive effects. These early models were mainly based on the fact that azathioprine and its metabolites are able to interfere with biosynthesis and integrity of cellular DNA. Within the body, azathioprine is metabolized via 6-mercaptopurine towards biologically active 6-thioguanine nucleotides (6-TGN) and these 6-TGN nucleotides can be incorporated randomly into cellular DNA by DNA polymerase during DNA replication[16,17]. Subsequently, 6-TGN containing DNA is known to show an altered DNA structure and a decreased base pair stability of 6-thio-GTP/CTP base pairs, finally resulting in an increased sensitivity of this DNA to DNA processing enzymes, for example RNase H, Topo II and DNA ligase and therefore in a reduced DNA stability[18]. In addition to these azathioprine-mediated effects on DNA stability, therapy with purine-agonist azathioprine is known to result in inhibition of nucleic acid synthesis[19]. It could be shown by Dayton et al.[20] that especially the amount of adenosine nucleotides, but to a lesser extent also of guanosine nucleotides, is markedly reduced in azathioprine-treated T lymphocytes. Through this DNA-affecting quality of azathioprine it is possible to explain the capacity of this drug to mediate immunosuppressive effects in tissue with high rates of cell division and of DNA replication. For example, the successful use of azathioprine in the treatment of childhood leukaemia[21] could be explained as a result of 6-TGN-mediated effects on the DNA integrity of highly proliferating and dividing malignant leukaemic cells. But regarding the fact that lamina propria T lymphocytes in the colon of IBD patients show comparatively low rates of proliferation and cell division[22] and should therefore be less susceptible to faults within their DNA replication system, neither the random incorporation of 6-TGN into DNA nor a general inhibition of nucleic acid syntheses is able to explain sufficiently the specific suppressive effects of azathioprine on the mucosal immune system in the gut in case of IBD treatment. Another point, which conflicts with the assumption that the immunosuppressive effects of azathioprine could be mainly explained by random incorporation of 6-TGN into cellular DNA, is the very stable pattern of adverse side-effects of azathioprine[23]. One would imagine that a randomly distributed integration of 6-TGN into cellular DNA would more probably result in a mixed and variable pattern of side-effects, mainly affecting tissue with high rates of cell division, than in the constant pattern of side-effects which is recognized during azathioprine therapy and which is dominated by a rather frequently occurring leukopenia.

So obviously there must be alternative or complementary explanations for the immunosuppressive effects of azathioprine in IBD, which are mainly concentrated on the function of T lymphocytes. For this reason it was very

interesting to find in 2003 that azathioprine and 6-mercaptopurine (6-MP) are able to induce apoptosis in CD3/CD28 co-stimulated human $CD4^+$ T lymphocytes[24] (Figure 1). In line with this important finding it could be demonstrated that IBD patients, who responded well to therapy with azathioprine, showed an increased rate of apoptotic lamina propria mononuclear cells, while in non-responders there were only basal levels of

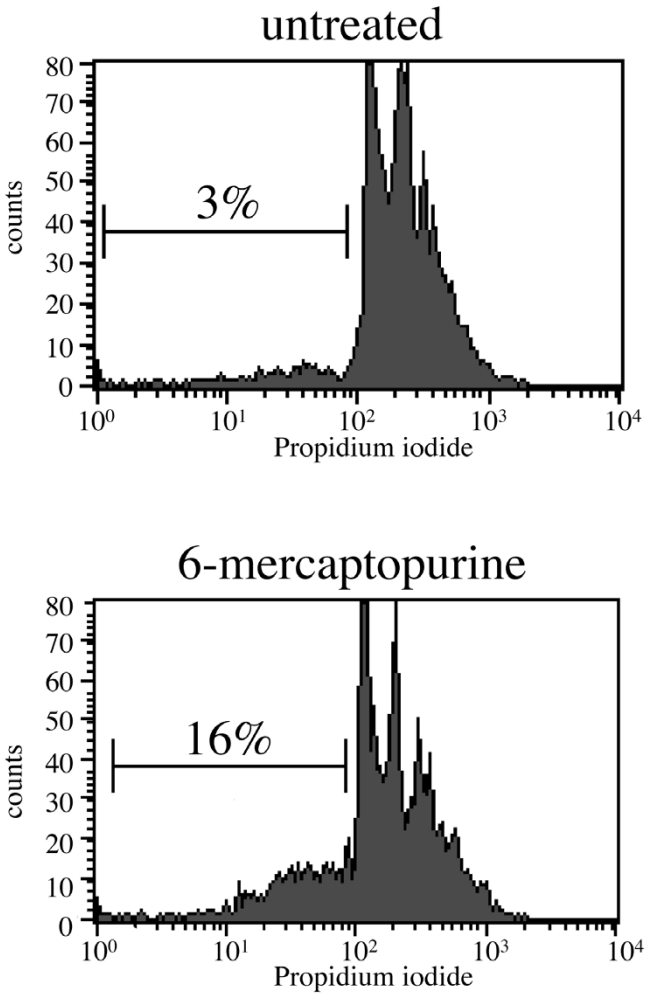

Figure 1 6-Mercaptopurine is able to induce apoptosis in co-stimulated human T lymphocytes. Human $CD4^+$ T cells were isolated from the blood of healthy donors, stimulated with anti-CD3/CD28 antibodies and interleukin 2 and treated with 6-mercaptopurine (5 μM) *in vitro* or left untreated. After 6 days the rate of apoptotic cells was analysed by propidium iodide-based Nicoletti staining

apoptotic cells detectable[24]. So obviously the capacity of azathioprine and 6-MP to induce apoptosis in co-stimulated human T cells is associated with therapeutic response to azathioprine treatment. Looking for a possible explanation for this apoptosis-inducing capacity of azathioprine, which was strongly dependent on a prior co-stimulation of T cells via CD28, the small GTPase Rac received much attention[24,25]. Rac represents a member of the Rho family of small GTPases, which are characterized by the ability to switch between inactive GDP-bound state and active GTP-bound state[26]. The GDP/GTP exchange on small GTPases is catalysed by guanosine nucleotide exchange factors (GEF). In case of Rac, Vav represents an important GEF, responsible for Rac activation[27,28]. Interestingly, the azathioprine metabolite 6-thio-GTP is able to bind to small GTPase Rac instead of GTP, to inhibit the GEF activity of Vav on Rac and thereby to reduce the amount of activated GTP-bound Rac within the cells[25]. Regarding the well-known involvement of activated Rac in the CD28-initiated intracellular signalling cascade in T lymphocytes[26–28] and the described ability of azathioprine metabolite 6-thio-GTP to inhibit Rac activation, the specific influence of azathioprine on CD28 co-stimulated T lymphocytes becomes understandable. In detail, the resulting model, describing the azathioprine-mediated effects in co-stimulated T lymphocytes, looks as follows[24,25]: physiologically, CD28-mediated co-stimulation of $CD4^+$ T lymphocytes is followed by a strong activation of Rac and subsequently by increased activation of transcription factors NF-κB and STAT-3[29,30], which in turn are able to induce their target genes. Due to STAT-3 and NF-κB-induced increased expression of anti-apoptotic protein bcl-x_L, co-stimulation via CD28 originally represents an anti-apoptocic signalling for human $CD4^+$ T cells[31]. In the case of azathioprine treatment there are increased intracellular concentrations of the azathioprine metabolite 6-thio-GTP. 6-thio-GTP is then able to compete with endogenous GTP for the binding to the small GTPase Rac, finally resulting in inhibited GEF activity of Vav on Rac and in decreased levels of activated GTP-bound Rac. Consequently, azathioprine treatment leads to decreased activation of transcription factors NF-κB and STAT-3 and to decreased expression of anti-apoptotic protein bcl-x_L. In this way the azathioprine metabolite 6-thio-GTP is able to convert the anti-apoptotic signalling of CD28-initiated co-stimulation into a pro-apoptotic signalling for human T lymphocytes (Figure 2).

Beside this important regulatory influence of activated Rac on T cell apoptosis, activated Rac also plays an essential role in the organization of cytoskeleton, and is thus involved in the formation of immunological synapse and conjugates between T lymphocytes and antigen-presenting cells (APC)[32]. In this way treatment of co-stimulated $CD4^+$ T lymphocytes with azathioprine not only results in increased apoptosis of stimulated T cells, but in addition shows inhibitory effects on the interaction between T lymphocytes and APC[25].

In summary, this more recent model, based on azathioprine-mediated inhibition of Rac activation and subsequent blockade of the CD28-mediated pathway, might be more relevant for an improved understanding of azathioprine-mediated effects in IBD treatment than models based on incorporation of 6-thio-GTP into DNA, as CD28-mediated co-stimulation is essential for survival of lamina propria T cells in the gut.

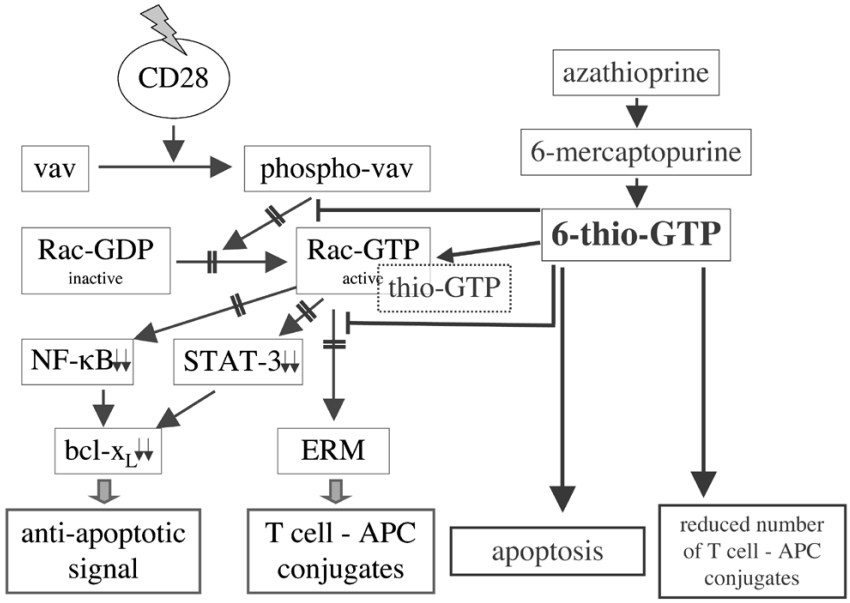

Figure 2 Azathioprine metabolite 6-thio-GTP is responsible for azathioprine-mediated immunosuppressive effects. Therapy with azathioprine results in the formation of the active metabolite 6-thio-GTP, which is able to bind to the small GTPase Rac instead of GTP and thereby interferes with the CD28-mediated intracellular signalling cascade. Physiologically, co-stimulation of T lymphocytes via CD28 results in an early activation of guanosine exchange factor Vav and subsequently in an increased formation of active GTP-bound Rac. Via NF-κB or STAT-3, activated Rac is able to increase the expression of anti-apoptotic protein bcl-x_L and thereby mediates an anti-apoptotic signalling in T cells. Additionally, activated Rac supports the formation of conjugates between lymphocytes and APC. Binding of 6-thio-GTP to Rac results in an inhibition of Vav-mediated Rac activation and subsequently in decreased expression of anti-apoptotic protein bcl-x_L. Therefore, the immunosuppressive effects of azathioprine, especially its capacity to induce apoptosis of stimulated T cells and to inhibit the formation of conjugates between T cells and APC, can be explained by 6-thio-GTP-mediated inhibition of Rac activation

NEW VISIONS – FROM BENCH TO BEDSIDE

Identification of 6-thio-GTP as the main mediator of azathioprine-related immunosuppressive effects in IBD allows the development of new strategies for the prediction of individual clinical success of azathioprine therapy, but also for the optimization of classic drug azathioprine.

Many different clinical studies have attempted to correlate the intracellular 6-TGN levels of IBD patients and the clinical benefit of azathioprine therapy, in order to predict response or non-response to azathioprine in a very early stage of the therapy[33–38]. The inconsistent and partly conflicting results of these attempts might be due to the fact that none of these trials analysed the different subtypes of 6-TGN separately. Actually, 6-TGN represents a heterogeneous mix of 6-thio-GMP, 6-thio-GDP and 6-thio-GTP. Based on the described

model of azathioprine-mediated inhibition of Rac activation and subsequent induction of T cell apoptosis, only 6-thio-GTP is responsible for the therapeutic effects of azathioprine in IBD, whereas 6-thio-GMP and 6-thio-GDP are functionally inactive. Therefore recently, in a pilot study, the concentrations of different 6-TGN, 6-thio-GMP, 6-thio-GDP and 6-thio-GTP, were analysed simultaneously in red blood cells of azathioprine-treated IBD patients by a newly developed HPLC-based method and compared separately with clinical data of included patients[16]. Interestingly, it could be shown that 6-TGN-based monitoring of azathioprine therapy could be markedly refined by regarding the 6-thio-GTP ratio, defined as the ratio of 6-thio-GTP concentration and the sum of 6-thio-GDP and 6-thio-GTP levels, in the group of patients with high total 6-TGN levels. Whereas simple measurement of total 6-TGN levels did not correlate significantly with the number of flares or the infliximab demand, additionally identified patients with low 6-thio-GTP ratio within the 6-TGN high group showed a significantly worse outcome with lower response rates, more flares and higher infliximab use than patients with high 6-thio-GTP ratio and high total 6-TGN levels[16]. Obviously, with regard to these early and very promising results, it is possible to predict a poor response to azathioprine therapy even in the presence of high 6-TGN levels by identifying patients with relatively high levels of functionally inactive 6-thio-GDP. This study is thus a very successful example of how to integrate recent experimentally gained insights into clinical practice; it will be very interesting to see whether these results can be confirmed or even extended in a subsequent multicentre prospective study.

Based on the same Rac-dependent model of azathioprine-mediated immunosuppressive effects, another very interesting approach could be the possible development of chemically modified derivatives of 6-thio-GTP, which might then be able to compete more effectively with GTP for the binding to Rac, or which might show an increased capacity to block the GEF activity of Vav on Rac. The idea of such an attempt would be that such a somehow stronger inhibition of Rac activation by newly developed 6-thio-GTP derivatives might help to reduce the well-known delayed onset of action of the classic drug azathioprine, and might thereby strengthen the role of thiopurines in the therapy of acute flares of IBD.

References

1. Elion GB. The George Hitchings and Gertrude Elion Lecture. The pharmacology of azathioprine. Ann NY Acad Sci. 1993;685:400–7.
2. Bean RHD. Treatment of ulcerative colitis with anti-metabolites. Br Med J. 1996;1:1081–4.
3. O'Donoghue DP, Dawson AM, Powell-Tuck J, Bown RL, Lennard-Jones JE. Double-blind withdrawal trial of azathioprine as maintenance treatment for Crohn's disease. Lancet. 1978;2:955–7.
4. Kirk AP, Lennard-Jones JE. Controlled trial of azathioprine in chronic ulcerative colitis. Br Med J (Clin Res Ed). 1982;284:1291–2.
5. Hawthorne AB, Logan RF, Hawkey CJ et al. Randomised controlled trial of azathioprine withdrawal in ulcerative colitis. Br Med J. 1992;305:20–2.
6. Ewe K, Press AG, Singe CC et al. Azathioprine combined with prednisolone or monotherapy with prednisolone in active Crohn's disease. Gastroenterology. 1993;105: 367–72.

7. Sandborn W, Sutherland L, Pearson D, May G, Modigliani R, Prantera C. Azathioprine or 6-mercaptopurine for inducing remission of Crohn's disease. Cochrane Database Syst Rev. 2000;2:CD000545.
8. Pearson DC, May GR, Fick GH, Sutherland LR. Azathioprine and 6-mercaptopurine in Crohn disease. A meta-analysis. Ann Intern Med. 1995;123:132–42.
9. Pearson DC, May GR, Fick G, Sutherland LR. Azathioprine for maintaining remission of Crohn's disease. Cochrane Database Syst Rev. 2000;2:CD000067.
10. Holtmann MH, Krummenauer F, Claas C et al. Long-term effectiveness of azathioprine in IBD beyond 4 years: a European multicenter study in 1176 patients. Dig Dis Sci. 2006;1: 1516–24.
11. Baumgart DC, Sandborn WJ. Inflammatory bowel disease: clinical aspects and established and evolving therapies. Lancet. 2007;369:1641–57.
12. Hanauer SB, Korelitz BI, Rutgeerts P et al. Postoperative maintenance of Crohn's disease remission with 6-mercaptopurine, mesalamine, or placebo: a 2-year trial. Gastroenterology. 2004;127:723–9.
13. Myrelid P, Svärm S, Andersson P, Almer S, Bodemar G, Olaison G. Azathioprine as a postoperative prophylaxis reduces symptoms in aggressive Crohn's disease. Scand J Gastroenterol. 2006;41:1190–5.
14. Travis SP, Stange EF, Lémann M et al.; European Crohn's and Colitis Organisation. European evidence based consensus on the diagnosis and management of Crohn's disease: current management. Gut. 2006;55(Suppl. 1):i16–35.
15. Timmer A, McDonald JW, Macdonald JK. Azathioprine and 6-mercaptopurine for maintenance of remission in ulcerative colitis. Cochrane Database Syst Rev. 2007;1: CD000478.
16. Neurath MF, Kiesslich R, Teichgräber U et al. 6-Thioguanosine diphosphate and triphosphate levels in red blood cells and response to azathioprine therapy in Crohn's disease. Clin Gastroenterol Hepatol. 2005;3:1007–14.
17. Ling YH, Nelson JA, Cheng YC et al. 2′-Deoxy-6-thioguanosine 5′-triphosphate as a substrate for purified human DNA polymerases and calf thymus terminal deoxynucleotidyltransferase *in vitro*. Mol Pharmacol. 1991;40:508–14.
18. Somerville L, Krynetski EY, Krynetskaia NF et al. Structure and dynamics of thioguanine-modified duplex DNA. J Biol Chem. 2003;278:1005–11.
19. Maltzman JS, Koretzky GA. Azathioprine: old drug, new actions. J Clin Invest. 2003;111: 1122–4.
20. Dayton JS, Turka LA, Thompsen CB et al. Comparison of the effects of mizoribine with those of azathioprine, 6-mercaptopurine, and mycophenolic acid on T lymphocyte proliferation and purine ribonucleotide metabolism. Mol Pharmacol. 1992;41:671–6.
21. Hyman CB, Brubaker C, Sturgeon P. 6-Mercaptopurine in childhood leukemia; comparison of large dose interrupted with small dose continuous therapy. Cancer Res. 1957;17:851–6.
22. Fuss IJ, Neurath M, Boirivant M et al. Disparate CD4+ lamina propria (LP) lymphokine secretion profiles in inflammatory bowel disease. Crohn's disease LP cells manifest increased secretion of IFN-gamma, whereas ulcerative colitis LP cells manifest increased secretion of IL-5. J Immunol. 1996;157:1261–70.
23. Sandborn WJ. Azathioprine: state of the art in inflammatory bowel disease. Scand J Gastroenterol Suppl. 1998;225:92–9.
24. Tiede I, Fritz G, Strand S et al. CD28-dependent Rac1 activation is the molecular target of azathioprine in primary human CD4+ T lymphocytes. J Clin Invest. 2003;111:1133–45.
25. Poppe D, Tiede I, Fritz G et al. Azathioprine suppresses ERM-dependent T cell-APC conjugation through inhibition of Vav guanosine exchange activity on Rac proteins. J Immunol. 2005;176:640–51.
26. Cantrell DA. GTPases and T cell activation. Immunol Rev. 2003;192:122–30.
27. Bustelo XR. Vav proteins, adaptors and cell signaling. Oncogene. 2001;20:6372–81.
28. Movilla N, Dosil M, Zheng Y, Bustelo XR. How Vav proteins discriminate the GTPases Rac1 and RhoA from Cdc42. Oncogene. 2001;20:8057–65.
29. Darnell JE. STATs and gene regulation. Science. 1997;277:1630–5.
30. Fanger GR, Johnson NL, Johnson GL. MEK kinases are regulated by EGF and selectively interact with Rac/Cdc42. EMBO J. 1997;16:4961–72.

31. Kirchhof, S, Muller WW, Li-Weber M, Krammer PH. Up-regulation of c-FLIPshort and reduction of activation-induced cell death in CD28-costimulated human T cells. Eur J Immunol. 2000;30:2765–74.
32. Faure S, Salazar-Fontana LI, Semichon M et al. ERM proteins regulate cytoskeleton relaxation promoting T cell-APC conjugation. Nature Immunol. 2004;5:272–9.
33. Cuffari C, Seidmann EG, Latour S et al. Quantification of 6-thio-guanine in peripheral blood leukocyte DNA in Crohn's disease patients on maintenance 6-mercaptopurine therapy. Can J Physiol Pharmacol. 1996;170:4986–95.
34. Dubinsky MC, Yang H, Hassard PV et al. 6-MP metabolite profiles provide a biochemical explanation for 6-MP resistancein patients with inflammatory bowel disease. Gastroenterology. 2002;122:904–15.
35. Dubinsky MC. Azathioprine, 6-mercaptopurine in inflammatory bowel disease: pharmacology, efficacy and safety. Clin Gastroenerol Hepatol. 2004;2:731–43.
36. Wright S, Sanders DS, Lobo AJ et al. Clinical significance of azathioprine active metabolite concentrations in inflammatory bowel disease. Gut. 2004;53:1123–8.
37. Lowry PW, Franklin CL, Weaver AL et al. Measurement of thiopurine methyltransferase activity and azathioprine metabolites in patients with inflammatory bowel disease. Gut. 2001;49:665–70.
38. Reuther LO, Sonne J, Larsen NF et al. Pharmacological monitoring of azathioprine therapy. Eur J Clin Pharmacol. 2003;38:972–7.

17
Antibiotics and probiotics

P. GIONCHETTI, F. RIZZELLO, C. MORSELLI, R. TAMBASCO and M. CAMPIERI

INTRODUCTION

The rationale for using antibiotics and probiotics in inflammatory bowel disease (IBD) is based on convincing evidence implicating intestinal bacteria in the pathogenesis of the disease[1].

The distal ileum and colon are the areas with the highest bacterial concentrations, and represent the sites of inflammation in IBD. In addition, pouchitis, the non-specific inflammation of the ileal reservoir after ileo-anal anastomosis, appears to be associated with bacterial overgrowth and dysbiosis. Furthermore, pouchitis does not occur prior to closure of the ileostomy.

Patients with Crohn's disease (CD) consistently respond to diversion of the faecal stream, with immediate recurrence of inflammation after restoration of intestinal continuity or infusion of luminal content into the bypassed ileum[2,3]. Moreover, the composition of the enteric flora is altered in patients with IBD, and enteric bacteria or their products have been found within the inflamed mucosa of patients with CD[4]. Increased numbers of aggressive bacteria, such as *Bacteroides*, adherent/invasive *Escherichia coli*, and enterococci; and decreased numbers of protective lactobacilli and bifidobacteria have been observed in IBD[5].

However, the most compelling evidence that intestinal bacteria play a role in IBD has been derived from animal models. Despite great diversity in genetic defects and immunopathology, a consistent feature of many transgenic and knockout mutant murine models of colitis is that the presence of normal enteric flora is required for full expression of inflammation[6]. Indeed, there is evidence of a loss of immunological tolerance to commensal bacteria in patients with IBD[7,8].

These findings have led to the proposal that manipulation of intestinal microbiota flora, either with antibiotics or probiotics, may be therapeutic in IBD. Some suggested mechanisms of action of antibiotics and probiotics are shown in Table 1.

There is a growing body of evidence from animal studies and clinical trials supporting a therapeutic role for antibiotics and probiotics in ulcerative colitis (UC), CD and pouchitis.

Table 1 Suggested mechanisms of action of antibiotics and probiotics

Antibiotics
Eradication of bacterial antigenic triggers
Elimination of bacterial overgrowth
Reduction of proinflammatory bacterial toxins
Potential immunosuppressive properties of antibiotics
Probiotics
Inhibition of pathogenic enteric bacteria by:
Decreasing luminal pH
Secretion of bactericidal proteins
Resisting colonization
Blocking epithelial binding
Improvement in epithelial and mucosal barrier function by:
Production of short-chain fatty acids
Enhancing mucus production
Increasing barrier integrity
Alteration of immunoregulation by:
Increasing interleukin-10 and transforming growth factor beta, and decreasing TNF levels
Increasing immunoglobulin A production

ANTIBIOTICS

Animal model studies

In several rodent models the use of broad-spectrum antibiotics can both prevent the onset of, and treat, experimental colitis, whereas metronidazole and ciprofloxacin can only prevent experimental colitis but not reverse established disease[9–13]. Broad-spectrum antibiotics are effective in almost all models of acute and chronic colitis[13–16]; however, they have only a transient efficacy in HLA-B27 transgenic rats[17]. Interestingly, ciprofloxacin and metronidazole were found to have selective efficacy in different colonic regions in interleukin-10 (IL-10) knockout mice, suggesting that different bacteria cause inflammation in different colonic segments[15]. These studies suggest that most clinical forms of IBD may respond to a specific combination of broad-spectrum antibiotics.

Ulcerative colitis

Only a few trials of antibacterial agents have been carried out in UC, and results are controversial. Most clinicians have used antibiotics as an adjuvant therapy in severe UC. Dickinson et al. have carried out a double-blind controlled trial on the use of oral vancomycin as an adjunct in acute exacerbations of idiopathic colitis. No significant difference was found between the two treatment groups, with only a trend towards a reduction in the need for surgery in patients treated with vancomycin[18].

Intravenous metronidazole, used in conjunction with corticosteroids, was similarly as effective as placebo in inducing remission in patients with severe UC[19].

In a double-blind, placebo-controlled trial in patients with acute relapse of UC, 84 patients were randomized to receive corticosteroids plus oral tobramycin or placebo. After 1 week of treatment, 74% of patients in the tobramycin treatment group vs 43% in the placebo group ($p < 0.003$) achieved complete symptomatic remission[20]. However, the combination of tobramycin and metronidazole did not have any beneficial effect when compared with a standard steroid treatment in severely acute UC[21].

Ciprofloxacin has been investigated in a randomized, placebo-controlled study; 70 patients with mild to moderate active UC were randomized to receive either ciprofloxacin 250 mg twice a day or placebo for 14 days. At the end of the study, 70.5% of patients in the ciprofloxacin group vs 72% in the placebo group had achieved remission[22]. Moreover, a short course of intravenous ciprofloxacin was not effective as an adjunctive treatment to corticosteroids in severe UC in a prospective, randomized, double-blind, placebo-controlled trial[23]. In contrast, some efficacy was observed in a more recent randomized placebo-controlled trial when ciprofloxacin was administered for 6 months to patients with active UC, poorly responding to conventional therapy with steroids and mesalazine. At the end of the study the treatment-failure rate was 21% in the ciprofloxacin-treated group and 44% in the placebo group ($p < 0.002$). This difference was detected using clinical criteria; while endoscopic and histological findings showed differences only at 3 months but not at 6 months[24].

The non-absorbable, broad-spectrum antibiotic, rifaximin, was investigated in a small controlled study, to evaluate its efficacy and systemic absorption in patients with moderate to severe active UC, refractory to steroid treatment. Twenty-eight patients were randomized to receive either rifaximin 400 mg twice daily or placebo for 10 days as an adjunct to standard steroid treatment. Although there was no significant difference in the clinical efficacy score between the two treatments, only rifaximin determined a significant improvement in stool frequency, rectal bleeding and sigmoidoscopic score[25]. Whilst rifaximin did not permanently alter the colonic microbiota, resistant *Bifidobacterium* species were found after three intermittent courses in patients with UC[26].

Crohn's disease

Broad-spectrum antibiotics are widely used to treat CD[27], but large controlled trials have not yet been performed.

Metronidazole has been the most investigated agent. In 1978 Blichfeldt et al. found no difference between metronidazole- and placebo-treated patients in a placebo-controlled, double-blind, crossover trial. However, a positive trend in favour of metronidazole was observed when only the colon was involved[28]. In the National Cooperative Swedish study, metronidazole was compared with sulphasalazine as a primary treatment for CD. Although no significant difference was found between the two groups, interestingly, in the crossover section of the study, metronidazole was effective in patients who did not respond to sulphasalazine[29]. In another study metronidazole was used either as a single therapy or in combination with cotrimoxazole, and compared to

cotrimoxazole alone and a double placebo, in patients with a symptomatic relapse of CD. After 4 weeks of treatment there was no difference in response among the three treatment groups[30]. In a Canadian randomized, placebo-controlled trial, Sutherland et al. demonstrated that treatment with metronidazole for 16 weeks significantly decreased the Crohn's Disease Activity Index (CDAI), but no difference was found in the rates of remission compared with placebo; benefit was dose-dependent with 20 mg/kg having a greater benefit than 10 mg/kg[31]. As in the Swedish study[29], the Canadian study found metronidazole to be effective for colonic and ileocolonic CD, but not for ileitis. Unfortunately, metronidazole has numerous side-effects including nausea, anorexia, dysgeusia, dyspepsia and peripheral neuropathy, which limit its use in approximately 20% of patients.

An antibiotic combination was used in an Italian randomized controlled study in which metronidazole 250 mg four times daily plus ciprofloxacin 500 mg twice daily were compared to a standard steroid treatment for 12 weeks[32]. No significant differences were reported in the rates of remission between treatments (46% with ciprofloxacin plus metronidazole vs 63% with methylprednisolone), suggesting that this antibiotic combination is a potential alternative to steroid treatment in the acute phase of CD[32]. In another trial this combination of metronidazole and ciprofloxacin was supplemented with budesonide 9 mg/day in active CD; no difference was registered compared to placebo but, surprisingly, the overall response in the two groups was lower than in previous studies using budesonide. This study also found antibiotic treatment to be more effective in colonic disease than in isolated small bowel disease[33].

In another trial ciprofloxacin 1 g/day was compared to mesalazine 4 g/day in a controlled study of mild to moderate active CD for 6 weeks. The results suggest that ciprofloxacin is as efficacious as mesalazine (remission observed in 56% and 55% of patients treated with ciprofloxacin and mesalazine respectively), thus offering a potential alternative treatment for active CD[34]. Furthermore, in a small study, ciprofloxacin was shown to be effective when combined with standard treatment in patients with resistant disease[35].

Other antibiotics have also been investigated. Shafran et al. carried out an open-label study on the efficacy and safety of rifaximin 600 mg/day for 16 weeks in the treatment of mild to moderate active CD. At the end of the study 59% of patients were in remission (CDAI <150), with a significant reduction of the mean CDAI score compared to baseline ($p<0.0001$)[36]. In another open-label trial Leiper et al. reported an impressive positive response to clarithromycin (64% patients improved or were in remission after 4 weeks) in a group of 25 patients with active CD, many of whom were unresponsive to other treatments[37].

Many studies have tried to evaluate the efficacy of antimycobacterial drugs in patients with CD, pursuing the possibility that a strain of *Mycobacterium* might be an aetiological agent in CD. Borgaonkar et al. performed a meta-analysis of all randomized controlled trials in which antimycobacterial therapy was compared with placebo[38]. It suggested that antimycobacterial therapy was efficacious only in the maintenance of remission after a combined treatment of corticosteroids and antimycobacterial agents. However, the investigator

emphasized that, because of the high incidence of side-effects and the small number of studies included in the meta-analysis, the data were inconclusive and should be interpreted with caution.

The same antibiotics used to treat luminal CD have been reported to be beneficial in the treatment of perianal CD, but no controlled trials have been performed[39]. Metronidazole 20 mg/kg has shown rates of fistula closure between 62% and 83%[40,41]. The combination of metronidazole and ciprofloxacin resulted in an improvement in 64% of patients with fistula closure in 21%[42]. Unfortunately, fistulas tend to recur in most patients following cessation of treatment. Although the results of these uncontrolled studies are inconclusive, metronidazole and ciprofloxacin, alone or in combination, are used by most clinicians as first-line treatments in patients with perianal disease, in conjunction with surgical drainage of abscesses.

The use of antibiotics in the prevention of postoperative disease recurrence has also been investigated. Rutgeerts et al. assessed the efficacy of metronidazole at 20 mg/kg per day in a placebo-controlled double-blind study[43]. Sixty patients were randomized to receive either metronidazole or placebo for 12 weeks. At the end of the treatment, endoscopic relapse was evaluated by Rutgeerts score. Metronidazole significantly decreased the incidence of severe endoscopic relapse (grade 3 or 4) in the neoterminal ileum 6 months after surgery and the clinical recurrence rates at 1 year, with a trend towards a protective effect after 3 years. More recently the similar antibiotic, ornidazole, used continuously for 1 year, was shown to be significantly more effective than placebo in the prevention of clinical and endoscopic recurrence in the neoterminal ileum at 12 months. Significantly more patients in the ornidazolo group dropped out because of side-effects[44].

Pouchitis

The awareness of the crucial importance that faecal stasis and bacterial overgrowth may have in the pathogenesis of acute pouchitis has led clinicians to treat patients with antibiotics.

Antibiotics have become the mainstay of treatment for pouchitis, despite an absence of controlled trials. Metronidazole is the first-line treatment, and most patients with acute pouchitis respond quickly to administration of 1–1.5 g/day [45,46]. A double-blind, randomized, placebo-controlled, crossover trial was carried out by Madden et al. to assess the efficacy of 400 mg of metronidazole three times daily per os for 2 weeks in 13 patients (11 completed both arms of the study) with chronic, unremitting pouchitis. Metronidazole was significantly more effective than placebo in reducing stool frequency (73% vs 9%), even without improvement in endoscopic appearance and histological grade of activity. However, a significant proportion of patients (55%) experienced side-effects while using metronidazole, including nausea, vomiting, abdominal discomfort, headache, skin rash and metallic taste[47].

Recently Shen et al. compared the efficacy and side-effects of ciprofloxacin and metronidazole in treating acute pouchitis in a randomized clinical trial. Seven patients received ciprofloxacin 1 g/day and nine patients received metronidazole 20 mg/kg per day for a period of 2 weeks. The results of this

study have shown that both ciprofloxacin and metronidazole are efficacious in the treatment of acute pouchitis: both reduced the total Pouchitis Disease Activity Index (PDAI) scores and led to a significant improvement in symptoms and endoscopic and histological scores. However, ciprofloxacin led to a greater reduction in PDAI scores, and improvement in symptoms and endoscopic scores. Furthermore ciprofloxacin was better tolerated than metronidazole (33% of metronidazole-treated patients reported adverse effects, compared with none in the ciprofloxacin group)[48].

Given the management difficulties posed by chronic refractory pouchitis, the use of combined antibiotic treatment has been explored. In an open trial, 18 patients with active pouchitis not responding to standard therapy (metronidazole or ciprofloxacin) for 4 weeks, were treated orally with rifaximin 2 g/day plus ciprofloxacin 1 g/day for 15 days. Symptom assessment, endoscopic and histological evaluations were performed at screening and after 15 days using PDAI scores. Sixteen out of 18 patients (88.8%) either improved ($n = 10$) or went into remission ($n = 6$); the median PDAI scores before and after therapy were 11 and 4 respectively ($p < 0.002$)[49].

More recently, 44 patients with refractory pouchitis received metronidazole from 800 mg to 1 g/day and ciprofloxacin 1 g/day for 28 days. Thirty-six patients (82%) went into remission; the median PDAI scores before and after therapy were 12 and 3 respectively ($p < 0.0001$), and the patients' quality of life significantly improved with the treatment (median IBD Questionnaire score increased from 96.5 to 175)[50].

PROBIOTICS

The first to propose the use of probiotics for the purpose of health maintenance and disease prevention was Elie Metchnikoff, the Russian Nobel prize winner, who at the turn of the last century suggested that a high concentration of lactobacilli in the intestinal flora was important for the health and longevity of humans[51]. Probiotics are defined as 'living organisms, which upon ingestion in certain numbers, exert health benefits beyond inherent basic nutrition'[52].

There are a number of bacteria associated with probiotic activity (Table 2). For clinical application, probiotic strains must possess certain characteristics: they need to be resistant to acid and bile; they must have the ability to be metabolically active within the luminal flora, where they should ideally survive, but not persist in the long term; they should be antagonistic to pathogenic bacteria; they must be safe for human use; and, finally, they should maintain their viability and beneficial properties during the manufacturing processes[53].

Animal model studies

Encouraging results have been obtained with probiotic therapy in experimental colitis. Administration of *Lactobacillus reuteri* was shown to significantly reduce inflammation in acetic acid- and methotrexate-induced colitis in rats[54,55]. More recently a *Lactobacillus* sp. was shown to be able to prevent the development of spontaneous colitis in interleukin 10 (IL-10)-deficient mice[56],

Table 2 Organisms associated with probiotic activity

Most commonly	*Other bacterial strains*	*Yeast*
Lactobacilli Bifidobacteria Streptococci	Enterococci Non-pathogenic *E. coli*	*Saccharomyces boulardii*

and continuous feeding with *L. plantarum* improved an established colitis in the same knockout model[57]. A strain of *L. salivarius* (subsp. *salivarius*) reduced the rate of progression from inflammation to dysplasia and colonic cancer in IL-10-deficient mice[58], and a strain of *Bifidobacterium infantis* and of *L. salivarius* were able to attenuate inflammation and reduce the ability to produce Th1-type cytokines in the IL-10 knockout model[59].

Rachmilewitz and colleagues investigated the efficacy of the probiotic preparation, VSL#3, in the treatment of iodoacetamide-induced colitis in rats[60]. VSL#3 is characterized by a very high bacterial concentration (each packet containing 450 billion viable bacteria) and the presence of a cocktail of eight different bacterial species. This product contains viable lyophilized bacteria of four strains of lactobacilli (*L. casei*, *L. plantarum*, *L. acidophilus*, *L. delbrueckii* subsp. *bulgaricus*), three strains of bifidobacteria (*B. longum*, *B. breve*, *B. infantis*) and one strain of *Streptococcus salivarius* subsp. *thermophilus*. Rachmilewitz and colleagues found that VSL#3 resulted in a significant attenuation of inflammation with a decrease of myeloperoxidase and nitric oxide synthase activity of the iodoacetamide-induced colitis[60]. In a similar study Madsen and colleagues reported a significant improvement in inflammation together with a reduction in mucosal levels of proinflammatory cytokines and a normalization of colonic barrier integrity in IL-10 knockout mice[61].

Ulcerative colitis

Promising results have been found with probiotic studies in the treatment of UC. In three recent trials involving the non-pathogenic strain of *Escherichia coli* Nissle 1917, similar efficacy was observed to that of mesalazine in the maintenance treatment of UC[62–64].

We carried out a pilot study using the probiotic cocktail, VSL#3, as maintenance treatment in patients with UC in remission, allergic or intolerant to sulphasalazine and mesalazine, to assess its impact on the faecal flora. Twenty patients received 6 g/day of VSL#3 (1800 billion bacteria) for 12 months and were assessed clinically and endoscopically at baseline, at 6 and 12 months, and in the event of a relapse. Stool culture and determination of faecal pH were also performed at different intervals[65].

Microbiological determination showed a significant increase in concentration of lactobacilli, bifidobacteria and *Streptococcus thermophilus*, evident after just 20 days, which persisted throughout the treatment period, and returned to basal levels within 15 days of ceasing treatment. Faecal

concentration of *Bacteroides*, enterococci, coliforms, *Clostridia* and total anaerobes and aerobes was not affected, but faecal pH was significantly reduced by the treatment. Fifteen of the 20 patients (75%) remained in remission throughout the treatment period[65].

Furthermore, in an uncontrolled pilot study, VSL#3 at very high dosage (3600 billion bacteria/day), induced remission in 53%, with a positive response in a further 23% of patients with active mild to moderate disease[66].

In addition, an open uncontrolled 4-week study found that the yeast *Saccharomyces boulardii* induced remission in 71% of patients with mild to moderate UC[67]. These studies highlight the wide range of organisms that may be beneficial as probiotic therapy for UC.

Crohn's disease

Campieri et al. performed a randomized trial to evaluate the efficacy of a combination of rifaximin and the probiotic preparation, VSL#3, in the prevention of postoperative recurrence of CD. Rifaximin 1.8 g/day for 3 months, followed by VSL#3 at 6 g daily for 9 months, was compared with mesalazine 4 g/day for 12 months, in 40 patients after curative resection for CD. After 3 months of treatment the antibiotic–probiotic combination resulted in a significantly lower incidence of severe endoscopic recurrence compared to mesalazine (2/20 (10%) vs 8/20 (40%)). This difference was maintained throughout the study period (4/20 (20%) vs 8/20 (40%))[68].

No such clinical effect was seen in a study by Prantera et al., who reported no benefit of the probiotic *Lactobacillus GG* in preventing postoperative disease recurrence in a 1-year double-blind, placebo-controlled trial[69]. Similar negative results were recently reported by the GETAID French group. In a randomized double-blind, placebo-controlled study *L. johnsonii* LA1 (4×10^9 cfu/day) was not superior to placebo in preventing endoscopic recurrence of CD[70].

In a small pilot study, treatment with capsules containing *E. coli* Nissle 1917 was compared to placebo in the maintenance of steroid-induced remission of colonic CD. Twelve patients were treated with *E. coli* Nissle and 11 with placebo; at the end of the 12-week treatment period relapse rates were 33% in the *E. coli* group and 63% in the placebo group, but unfortunately, because of the small number of patients treated, this difference did not reach statistical significance[71].

However, in a small comparative open study, the combination of *Saccharomyces boulardii* 1 g/day plus mesalazine 2 g/day was significantly superior to mesalazine 3 g/day in maintenance of remission in a 6-month trial[72], suggesting probiotic treatment in CD may be beneficial. More recently, in a double-blind trial *Lactobacillus GG* was shown not be superior to placebo in prolonging remission in children with CD when given as an adjunct to standard therapy[73].

Pouchitis

Although less widely used in clinical practice than antibiotics, probiotics may be just as efficacious in the prevention and treatment of pouchitis. In a double-

blind study we have compared the efficacy of VSL#3 with placebo in the maintenance and treatment of chronic pouchitis. Forty patients who obtained clinical and endoscopic remission after 1 month of combined antibiotic treatment (rifaximin 2 g/day + ciprofloxacin 1 g/day) were randomized to receive VSL#3 at 6 g daily (1800 billion bacteria/day) or a placebo of identical appearance for 9 months. Clinical assessment was carried out every month; endoscopic and histological assessments were performed at entry and subsequently every 2 months. Stool samples were cultured before and after antibiotic treatment and subsequently every month during maintenance treatment. Relapse was defined as an increase of at least two points in the clinical portion of the PDAI and confirmed endoscopically and histologically. Whilst all 20 patients treated with placebo had a relapse during the 9-month follow-up period, 17 of the 20 (85%) patients treated with VSL#3 remained in remission at this point. Interestingly, all of these 17 patients had a relapse within 4 months of ceasing the active treatment. Faecal concentration of lactobacilli, bifidobacteria and *Streptococcus salivarius* subsp. *thermophilus* were significantly increased within 1 month of starting VSL#3 treatment, and remained stable throughout the study. However, this increase did not affect the concentration of the other bacterial groups, suggesting that the beneficial effect of treatment was not mediated by suppression of endogenous luminal bacteria[74].

A recent study examining the maintenance of remission in patients with refractory or recurrent pouchitis has substantiated these results: remission was seen in 85% of patients treated with VSL#3 at 6 g/day (1800 billion bacteria/day) vs 6% in the placebo group after 1 year of treatment[75].

In addition, we found that continuous administration of VSL#3 resulted in a significant increase in IL-10 tissue levels, a significant decrease in tissue levels of the proinflammatory cytokines tumour necrosis factor alpha, IL-1 and interferon gamma, and a decrease in matrix metalloproteinase activity[76]. This may aid understanding of the mechanisms of action by which VSL#3 maintains remission in pouchitis. In contrast, *Lactobacillus GG* was ineffective in preventing relapse in patients with chronic pouchitis in a placebo-controlled trial[77].

We have also carried out a double-blind, placebo-controlled trial to evaluate the efficacy of VSL#3 in the prevention of pouchitis onset in patients, following ileal–anal anastomosis for UC. Forty patients were randomized to receive VSL#3 at 3 g per day (900 billion bacteria/day) or an identical placebo for 12 months, within 1 week of ileostomy closure. Patients were assessed clinically, endoscopically and histologically at 1, 3, 6, 9 and 12 months, according to the PDAI[76]. Patients treated with VSL#3 had a significantly lower incidence of acute pouchitis compared with those treated with placebo during the first year of ileostomy closure (10% vs 40%; $p < 0.05$). Moreover, the IBD Questionnaire score was significantly improved in the group treated with VSL#3, and the median stool frequency in patients not developing pouchitis, was significantly less in the VSL#3 group compared with the placebo group[78].

CONCLUSIONS

There is strong evidence that enteric commensal bacteria are involved in the pathogenesis of IBD. Therefore, it is sensible to propose that modification of the gut bacterial flora by antibiotics and probiotics may be effective in treating UC, CD and pouchitis.

Antibiotics are a well-established, efficacious treatment option for various manifestations of IBD. They have an essential role in treating the septic complications of CD, including intra-abdominal and perianal abscesses and perianal fistulas, although their use as primary therapy in CD is poorly documented. There is good evidence that ciprofloxacin, metronidazole or their combination are effective in Crohn's colitis and ileocolitis, though not in isolated ileal disease, and large controlled trials are needed to define optimal antibiotic regimens. In addition, their use in UC is not supported by the available studies and large trials with broad-spectrum agents are required. Although proper controlled trials have not yet been conducted, the use of antibiotics in pouchitis is largely justified.

Probiotics provide an attractive alternative to antibiotics in the treatment of IBD as trials to date have shown them to be safe and without side-effects. Promising results have been obtained from studies using probiotics, in both the prevention of relapse and the treatment of mild to moderate attacks of UC. Studies using probiotics in the treatment of CD are less clear, due to conflicting and limited data. There is also considerable evidence supporting the efficacy of the highly concentrated cocktail of probiotics, VSL#3, in the prevention of pouchitis onset and relapse.

Studies have highlighted the importance of selecting a well-characterized probiotic preparation for treatment. Viability and survival of bacteria in many available preparations are as yet unproven. It should be remembered that the beneficial effect of one probiotic preparation does not imply efficacy of other preparations containing different bacterial strains, because each individual probiotic strain has unique biological properties.

There is a need to improve our understanding of the composition of the enteric flora and the relationship between intestinal physiology and the luminal ecosystem. Only then can we truly optimize the bacteria-modifying treatments now available, in our quest to effectively treat IBD.

References

1. Sartor RB. Enteric microflora in IBD: pathogens or commensal? Inflamm Bowel Dis. 1997;3:230–5.
2. Rutgeerts P, Geboes K, Peeters M et al. Effect of faecal stream diversion on recurrence of Crohn's disease in the neoterminal ileum. Lancet. 1991;338:771–4.
3. D'Haens GR, Goboes K, Peeters M, Penninckx F, Rutgeerts P. Early lesions of recurrent Crohn's disease caused by infusion of intestinal contents in excluded ileum. Gastroenterology. 1998;114:262–7.
4. Guarner F, Casellas F, Borruel N et al. Role of microecology in chronic inflammatory bowel diseases. Eur J Clin Nutr. 2002;56(Suppl. 4):S34–8.
5. Neut C, Bulois P, Desreumaux P et al. Changes in bacterial flora of the neoterminal ileum after ileocolonic resection for Crohn's disease. Am J Gastroenterol. 2002;97:939–46.
6. Sartor RB, Rath HC, Sellon RK. Microbial factors in chronic intestinal inflammation. Curr Opin Gastoenterol. 1996;12:327–33.

7. Duchman R, Kaiser I, Hermann E, Mayet W, Ewe K, Meyer Zum Buschenfelde KH. Tolerance exists towards resident intestinal flora but is broken in active inflammatory bowel disease (IBD). Clin Exp Immunol. 1995;102:448–55.
8. Duchmann R, May E, Heike M, Knolle P, Neurath M, Meyer zum Buschenfelde KH. T cell specificity and cross reactivity towards enterobacteria, *Bacteroides*, *Bifidobacterium* and antigens from resident intestinal flora in humans. Gut. 1999;44:812–18.
9. Rath HC, Schultz M, Freitag R et al. Different subsets of enteric bacteria induce and perpetuate experimental colitis in rats and mice. Infect Immun. 2001;69:2277–85.
10. Madsen KL, Doyle JS, Tavernini MM, Jewell LD, Rennie RP, Fedorak RN. Antibiotic therapy attenuates colitis in interleukin 10 gene-deficient mice. Gastroenterology. 2000;118:1094–105.
11. Hoentjen F, Harmsen HJ, Braat H et al. Antibiotics with a selective aerobic or anaerobic spectrum have different therapeutic activities in various regions of the colon in interleukin-10 gene deficient mice. Gut. 2003;52:1721–7.
12. Fiorucci S, Distrutti E, Mencarelli A, Barbanti M, Palazzini E, Morelli A. Inhibition of intestinal bacterial translocation with rifaximin modulates lamina propria monocytic cells reactivity and protects against inflammation in a rodent model of colitis. Digestion. 2002;66:246–56.
13. Bamias G, Marini M, Moskaluk CA et al. Down-regulation of intestinal lymphocyte activation and Th1 cytokine production by antibiotic therapy in a murine model of Crohn's disease. J Immunol. 2002;169:5308–14.
14. Yamada T, Deitch E, Specian RD, Perry MA, Sartor RB, Grisham MB. Mechanisms of acute and chronic intestinal inflammation induced by indomethacin. Inflammation. 1993;17:641–62.
15. Onderdonk AB, Hermos JA, Dzink JL, Bartlett JG. Protective effect of metronidazole in experimental ulcerative colitis. Gastroenterology. 1978;74:521–6.
16. Videla S, Villaseca J, Guarner F et al. Role of intestinal microflora in chronic inflammation and ulceration of the rat colon. Gut. 1994;35:1090–7.
17. Dieleman LA, Goerres M, Arends A et al. *Lactobacillus GG* prevents recurrence of colitis in HLA-B27 transgenic rats after antibiotic treatment. Gut. 2003;52:370–6.
18. Dickinson RJ, O'Connor HJ, Pinder I, Hamilton I, Johnston D, Axon AT. Double-blind controlled trial of oral vancomycin as adjunctive treatment in acute exacerbations of idiopathic colitis. Gut. 1985;26:1380–4.
19. Chapman RW, Selby WS, Jewell DP. Controlled trial of intravenous metronidazole as adjunct to corticosteroids in severe ulcerative colitis. Gut. 1986;27:1210–12.
20. Burke DA, Axon ATR, Clayden SA, Dixon MF, Johnston D, Lacey RW. The efficacy of tobramycin in the treatment of ulcerative colitis. Aliment Pharmacol Ther. 1990;4:123–9.
21. Mantzaris GJ, Hatzis A, Kontogiannis P, Triadaphyllou G. Intravenous tobramycin and metronidazole as an adjunct to corticosteroids in acute, severe ulcerative colitis. Am J Gastroenterol. 1994;89:43–6.
22. Mantzaris GJ, Archavlis E, Christoforidis P et al. A prospective randomised controlled trial of oral ciprofloxacin in acute ulcerative colitis. Am J Gastroenterol. 1997;92:454–6.
23. Mantzaris GJ, Petraki K, Archavlis E et al. A prospective randomised controlled trial of intravenous ciprofloxacin as an adjunct to corticosteroids in acute, severe ulcerative colitis. Scand J Gastroenterol. 2001;36:971–4.
24. Turunen UM, Farkkila MA, Hakala K et al. Long-term treatment of ulcerative colitis with ciprofloxacin: a prospective, double-blind, placebo-controlled study. Gastroenterology. 1998;115:1072–8.
25. Gionchetti P, Rizzello F, Ferrieri A et al. Rifaximin in patients with moderate or severe ulcerative colitis refractory to steroid-treatment: a double-blind, placebo-controlled trial. Dig Dis Sci. 1999;44:1220–1.
26. Brigidi P, Swennen E, Rizzello F, Bozzolasco M, Matteuzzi D. Effects of rifaximin administration on the intestinal microbiota in patients with ulcerative colitis. J Chemother. 2002;14:290–5.
27. Present DH. How to do without steroids in inflammatory bowel disease. Inflamm Bowel Dis. 2000;6:48–57.
28. Blichfeldt P, Blomhoff JP, Myhre E, Gjone E. Metronidazole in Crohn's disease. A double-blind cross-over clinical trial. Scand J Gastroenterol. 1978;13:123–7.

29. Ursing B, Alm T, Barany F et al. A comparative study of metronidazole and sulphasalazine for active Crohn's disease: the cooperative Crohn's disease study in Sweden. II results. Gastroenterology. 1982;83:550–62.
30. Ambrose NS, Allan RN, Keighley MR, Burdon DW, Youngs D, Lennard-Jones JE. Antibiotic therapy for treatment in relapse of intestinal Crohn's disease. A prospective randomised study. Dis Colon Rectum. 1985;28:81–5.
31. Sutherland LR, Singleton J, Sessions J et al. Double-blind, placebo controlled trial of metronidazole in Crohn's disease. Gut. 1991;32:1071–5.
32. Prantera C, Zannoni F, Scribano ML et al. An antibiotic regimen for the treatment of active Crohn's disease: a randomised controlled clinical trial of metronidazole plus ciprofloxacin. Am J Gastroenterol. 1996;91:328–32.
33. Steinhart AH, Feagan BG, Wong CJ et al. Combined budesonide and antibiotic therapy for active Crohn's disease: a randomised controlled trial. Gastroenterology. 2002;123:33–40.
34. Colombel JF, Lemann M, Cassagnou M et al. A controlled trial comparing ciprofloxacin with mesalazine for the treatment of active Crohn's disease. Am J Gastroenterol. 1999;94:674–8.
35. Arnold GL, Beaves MR, Prydun VO, Mook WJ. Preliminary study of ciprofloxacin in active Crohn's disease. Inflamm Bowel Dis. 2002;8:10–15.
36. Shafran I, Dondelinger PJ, Johnson LK. Efficacy and tolerability of rifaximin, a nonabsorbed, gut-selective, oral antibiotic in the treatment of active Crohn's disease: results of an open-label study. Am J Gastroenterol. 2003;98:S250.
37. Leiper K, Morris AI, Rhodes JM. Open label trial of oral clarithromycin in active Crohn's disease. Aliment Pharmacol Ther. 2000;14:801–6.
38. Borgaonkar MR, MacIntosh DG, Fardy JM. A meta-analysis of antimycobacterial therapy for Crohn's disease. Am J Gastroenterol. 2000;95:725–9.
39. Schwartz DA, Pemberton JH, Sandborn WJ. Diagnosis and treatment of perianal fistulas in Crohn's disease. Ann Intern Med. 2001;135:906–18.
40. Bernstein LH, Frank MS, Brandt LJ, Boley SJ. Healing of perianal Crohn's disease with metronidazole. Gastroenterology. 1980;79:357–65.
41. Brandt LJ, Bernstein LH, Boley SJ. Metronidazole therapy for perianal Crohn's disease: a follow-up study. Gastroenterology. 1982;83:383–7.
42. Solomon MR, McLeod R. Combination ciprofloxacina and metronidazole in severe perianal Crohn's disease. Can J Gastroenterol. 1993;7:571–3.
43. Rutgeerts P, Hiele M, Geboes K et al. Controlled trial of metronidazole treatment for prevention of Crohn's recurrence after ileal resection. Gastroenterology. 1995;108:1617–21.
44. Rutgeerts P, Van Assche G, Vermeire S et al. Ornidazol for prophilaxis of postoperative Crohn's disease recurrence: a randomised, double-blind, placebo-controlled trial. Gastroenterology. 2005;128:856–61.
45. Sandborn WJ, Mc Leod, Jewell DP. Medical therapy for induction and maintenance of remission in pouchitis. A systematic review. Inflamm Bowel Dis. 1999;5:33–9.
46. Hurst RD, Molinari M, Chung P, Rubin M, Michelassi F. Prospective study of the incidence, timing and treatment of pouchitis in 104 consecutive patients after restorative proctocolectomy. Arch Surg. 1996;131:497–502.
47. Madden M, McIntyre A, Nicholls RJ. Double-blind cross-over trial of metronidazole versus placebo in chronic unremitting pouchitis. Dig Dis Sci. 1994;39:1193–6.
48. Shen B, Achkar JP, Lashner BA et al. A randomised clinical trial of ciprofloxacin and metronidazole to treat acute pouchitis. Inflamm Bowel Dis. 2001;7:301–5.
49. Gionchetti P, Rizzello F, Venturi A et al. Antibiotic combination therapy in patients with chronic, treatment-resistant pouchitis. Aliment Pharmacol Ther. 1999;13:713–18.
50. Mimura T, Rizzello F, Helwig U et al. Four week open-label trial of metronidazole and ciprofloxacin for the treatment of recurrent or refractory pouchitis. Aliment Pharmacol Ther. 2002;16:909–17.
51. Metchnikoff E. The Prolongation of Life: Optimistic Studies. Mitchell C ed. London: William Heinemann, 1907:161–83.
52. Schaafsma G. State of the art concerning probiotic strains in milk products. IDF Nutr Newsl. 1996;5:23–4.
53. Lee YK, Salminen S. The coming age of probiotics. Trends Food Sci Technol. 1995;6:241–5.

54. Fabia R, Ar'rajab A, Johansson M-L et al. The effect of exogenous administration of *Lactobacillus reuteri* R2LC and oat fiber on acetic acid-induced colitis in the rat. Scand J Gastroenterol. 1993;28:155–62.
55. Mao Y, Nobaek S, Kasravi B et al. The effects of *Lactobacillus* strains and oat fiber on methotrexate-induced enterocolitis in rats. Gastroenterology. 1996;111:334–44.
56. Madsen KL, Tavernini MM, Doyle JSG, Fedorak RN. *Lactobacillus* sp. prevents development of enterocolitis in interleukin-10 gene-deficient mice. Gastroenterology. 1999;116:1107–14.
57. Schultz M, Veltkamp C, Dieleman LA et al. *Lactobacillus plantarum* 299V in the treatment and prevention of spontaneous colitis in interleukin-10 deficient mice. Inflamm Bowel Dis. 2002;8:71–80.
58. O'Mahony L, Feenex M, O'Halloran S et al. Probiotic impact on microbial flora, inflammation and tumour development in IL-10 knockout mice. Aliment Pharmacol Ther. 2001;15:1219–25.
59. McCarthy J, O'Mahony L, O'Callaghan L et al. Double-blind, placebo-controlled trial of two probiotic strains in interleukin 10 knockout mice and mechanistic link with cytokine balance. Gut. 2003;52:975–80.
60. Shibolet O, Karmeli F, Eliakim R et al. Variable response to probiotics in two models of experimental colitis in rats. Inflamm Bowel Dis. 2002;8:399–408.
61. Madsen K, Cornish A, Soper P et al. Probiotic bacteria enhance murine and human intestinal epithelial barrier function. Gastroenterology. 2001;121:580–91.
62. Kruis W, Schuts E, Fric P, Fixa B, Judmaier G, Stolte M. Double-blind comparison of an oral *Escherichia coli* preparation and mesalazine in maintaining remission of ulcerative colitis. Aliment Pharmacol Ther. 1997;11:853–8.
63. Rembacken BJ, Snelling AM, Hawkey P, Chalmers DM, Axon TR. Non pathogenic *Escherichia coli* vs mesalazine for the treatment of ulcerative colitis: a randomised trial. Lancet. 1999;354:635–9.
64. Kruis W, Fric P, Potrotnieks J et al. Maintaining remission of ulcerative colitis with *Escherichia coli* Nissle 1917 is as effective as with standard mesalazine. Gut. 2004;53:1617–23.
65. Venturi A, Gionchetti P, Rizzello F et al. Impact on the faecal flora composition of a new probiotic preparation. Preliminary data on maintenance treatment of patients with ulcerative colitis (UC) intolerant or allergic to 5-aminosalicylic acid (5 ASA). Aliment Pharmacol Ther. 1999;13:1103–8.
66. Bibiloni R, Fedorak RN, Tannock GW et al. VSL#3 probiotic mixture induces remission in patients with active ulcerative colitis. Am J Gastroenterol. 2005;100:1539–46.
67. Guslandi M, Giollo P, Testoni PA. A pilot trial of *Saccharomyces boulardii* in ulcerative colitis. Eur J Gastroenterol Hepatol. 2003;15:697–8.
68. Campieri M, Rizzello F, Venturi A et al. Combination of antibiotic and probiotic treatment is efficacious in prophylaxis of post-operative recurrence of Crohn's disease: a randomised controlled study vs. mesalazine. Gastroenterology. 2000;118:A781.
69. Prantera C, Scribano ML, Falasco G Andreoli A, Luzi C. Ineffectiveness of probiotics in preventing recurrence after curative resection for Crohn's disease: a randomised controlled trial with *Lactobacillus GG*. Gut. 2002;51;405–9.
70. Marteau P, Lemann M, Seksik P et al. Ineffectiveness of *Lactobacillus johnsonii* LA1 for prophylaxis of post-operative recurrence in Crohn's disease: a randomized, double-blind, placebo-controlled GETAID trial. Gut. 2006;55:842–7.
71. Malchow HA. Crohn's disease and *Escherichia coli*. A new approach in therapy to maintain remission of colonic Crohn's disease? J Clin Gastroenterol. 1997;25:653–8.
72. Guslandi M, Mezzi G, Sorghi M Testoni PA. *Saccharomyces boulardii* in maintenance treatment of Crohn's disease. Dig Dis Sci. 2000;45:1462–4.
73. Bousvaros A, Guandalini S, Baldassano RN et al. A randomized, double-blind trial of *Lactobacillus GG* versus placebo in addition to standard maintenance therapy for children with Crohn's disease. Inflamm Bowel Dis. 2005;11:833–9.
74. Gionchetti P, Rizzello F, Venturi A et al. Oral bacteriotherapy as maintenance treatment in patients with chronic pouchitis: a double-blind, placebo-controlled trial. Gastroenterology. 2000;119:305–9.
75. Mimura T, Rizzello F, Helwig U et al. Once-daily high-dose probiotic therapy for maintaining remission in recurrent or refractory pouchitis. Gut. 2004;53:108–14.

76. Ulisse S, Gionchetti P, D'Alò S et al. Increased expression of cytokines, inducible nitric oxide synthase and matrix metalloproteinases in pouchitis: effects of probiotic treatment (VSL#3). Gastroenterology. 2001;120(Suppl.):A281.
77. Kuisma J, Mentula S, Jarvinen H, Kahri A, Saxelin M, Farkkila M. Effect of *Lactobacillus rhamnosus GG* on ileal pouch inflammation and microbial flora. Aliment Pharmacol Ther. 2003;17:509–15.
78. Gionchetti P, Rizzello F, Helvig U et al. Prophylaxis of pouchitis onset with probiotic therapy: a double-blind placebo controlled trial. Gastroenterology. 2003;124:1202–9.

18
Features of drugs used in inflammatory bowel disease

A. GANGL

ALTERNATIVE MEDICATIONS

Alternative medications are not well defined. In general this term denotes medications used in complementary and alternative medicine (CAM). Although great progress has been made in the development and use of effective drugs for inflammatory bowel diseases (IBD), about 30–50% of patients with IBD in Europe and North America are using unconventional therapies. Such therapies are mostly used complementary to conventional drugs and frequently patients do not inform their treating physicians about the use of CAM. Mostly used are homeopathy and herbal medicines, making up about 50% of CAM in IBD. Several studies suggest that long-lasting use of drugs (for instance cortisone) with limited beneficial effects, serious side-effects of conventional therapies and poor quality of the relationship between the patient and his treating physician determine the use of CAM. Despite its widespread use the evaluation of the efficacy of CAM in IBD is difficult for many reasons, including chronicity and heterogeneity of IBD, different unpredictable and highly variable courses of the disease as well as weak definitions for treatment indications and for therapeutic endpoints (review in refs 1 and 2; viewpoint in ref. 3).

HOMEOPATHY IN IBD

Homeopathy, established in about 1810 by Christian Friedrich Samuel Hahnemann in Saxony, now part of Germany, has not so far been studied specifically in IBD. A literature search in May 2007 (Pubmed, Google, Medline search) revealed no clinical study assessing homeopathic preparations and their efficacy in IBD. However, a recent meta-analysis, comparing placebo-controlled trials of homeopathy and allopathy including 110 homeopathy trials and 110 matched conventional-medicine trials, led the authors to conclude that the clinical effects of homeopathy are placebo effects[4]. There is no reason to assume that homeopathy should work better in patients with IBD.

Table 1 Herbal drugs tested in IBD

Plantago ovata seeds
Boswellia serrata gum resin and extracts
Aloe vera gel
Wheat grass juice
Germinated barley foodstuff
Sophora flavescens enemas
Curcuma longa

HERBAL DRUGS TESTED IN IBD

Contrary to homeopathy several herbal drugs used in CAM have been tested prospectively in IBD (Table 1).

Plantago ovata

Plantago ovata belongs to the plant family Plantaginaceae; it is an annual plant, originally derived from India, Iran and Pakistan, and distributed worldwide. It is also known as Indian wheat, and the seeds of this plant are named *Ispaghula* seeds. *P. ovata* seed is degraded by colonic bacteria to short-chain fatty acids, including butyrate and acetate. Various species of *P. ovata* flowers and roots have long been used in Chinese and Ayurvedic medicine, as well as in Western herbal medicine.

Fernandez-Banares et al. conducted an open-label, randomized, controlled trial of *P. ovata* seeds as compared with mesalamine in maintaining remission in ulcerative colitis[5]. The aim was to assess efficacy and safety of *P. ovata* seeds over a period of 12 months, assuming that colonic fermentation of *P. ovata* yields butyrate which could indeed be demonstrated in faecal samples of seven patients under study. A total of 105 patients with ulcerative colitis in remission for more than 3 months were recruited at 11 medical centres in Spain and randomized into three groups.

Three patients dropped out and the final analysis could be done on 35 patients, who received twice-daily 10 g *P. ovata* seeds only, 37 patients who received three times daily 500 g of mesalamine and 30 patients receiving 20 g *P. ovata* plus 1.5 g mesalamine per day. Upon completion of the study the remission curves by intention-to-treat analysis for the three groups showed no differences in the probability of maintained remission over 1 year[5].

As the authors state, the hypothesis of this study was to show that dietary fibre was better than mesalamine. The results, however, failed to show a difference between therapies. Although these results suggest that *P. ovata* seeds might be as effective as mesalamine in preventing a relapse of the disease over a 12-month period, the authors themselves claim that, to test the equivalence hypothesis between the two therapies, a larger sample with a sample size of 217 patients per study arm would have been required[5]. Nevertheless this is the first study to show that *P. ovata* seeds may be effective in maintaining remission in ulcerative colitis.

Boswellia serrata

Boswellia serrata also has a long-standing tradition in Ayurveda, where it is believed that incense acts by harmonizing in the human energy circuit. Incense is the oleogum resin produced in the bark of *B. serrata* and other *Boswellia* tree species. All ancient civilizations used incense, and francincense was one of the precious gifts of the three Magi, besides gold and myrrh. More than 200 different compounds were identified in the oleogum resin and some boswellic acids are probably responsible for anti-inflammatory effects of *B. serrata* medicines[6]. Boswellic acids inhibit the activity of 5-lipoxygenase and thus interfere with leukotriene biosynthesis[7]. As the most potent anti-inflammatory component of the gum resin of *B. serrata* a semi-synthetic form of acetyl-11-keto-beta-boswellic acid (sAKBA) was shown to confer protection in experimental murine colitis induced by dextran sodium sulphate[8].

Gerhardt et al.[9] performed the so far only randomized, double-blind, verum-controlled parallel group comparison for 8 weeks to compare efficacy and safety of the *B. serrata* extract H15 with mesalazine for the treatment of active Crohn's disease. A total of 102 patients with a Crohn's disease activity index between 450 and 150 were randomized and finally 83 patients could be treated and evaluated per protocol. The verum group of 44 patients was treated with three times three tablets of *Boswellia* extract H15 with a daily total dose of 3.6 g for 8 weeks. A control group of 39 patients was treated with three times daily three tablets mesalazine 0.5 g, amounting to a total daily dose of 4.5 g. The Crohn's disease activity index between the status of enrolment and end of therapy after treatment with H15 was reduced by 90, and after therapy with mesalazine by 53 scores in the mean. In the authors' conclusion the study confirms that therapy with *B. serrata* extract H15 is not inferior to mesalazine, and in a separate benefit/risk evaluation even superior to mesalazine[9]. The German IBD Competence Net has recently started a prospective controlled trial to evaluate a *B. serrata* preparation in the maintenance treatment of patients with Crohn's disease in remission. The study intends to enrol 266 patients for a treatment period of 52 weeks with 3-month follow up (Holtmeier, Frankfurt, oral communication at the 18th Interdisciplinary Symposium Chronic Inflammatory Bowel Diseases, 13 April, 2007, Wiesbaden).

Aloe vera

Aloe vera belongs to the plant family Asphodelaceae, is native to the Canary Islands, and has a long history of cultivation throughout the drier tropical and subtropical regions of the world. The plant is stemless, grows to about 100 cm tall, with lanceolate thick and fleshy leaves, containing a yellowish sap with bitter taste and a watery gel which is alleged to be useful as a conservative for fresh fruit. *Aloe* gel has been used for several thousand years by Indian, Chinese and European ancient cultures in various traditional forms of medicine. *A. vera* gel, the mucilaginous aqueous extract of the leaf pulp of *A. barbadensis* Miller, has been promoted for the treatment of inflammatory diseases of skin and of the digestive tract. Only recently *A. vera* gel has been studied for the treatment of mild to moderately active ulcerative colitis[10].

Between March 1999 and July 2003, 49 patients with active ulcerative colitis were enrolled in a randomized, double-blind, placebo-controlled trial at hospitals in London and in Oxford, Great Britain. Three patients were lost immediately from further review and two patients did not fulfill inclusion criteria, thus five patients were excluded. From 44 evaluable patients 30 were given 100 ml *A. vera* gel twice daily for 4 weeks and 14 patients received placebo.

After 4 weeks of treatment 24 patients having received *A. vera* and 11 patients having received placebo could be evaluated, and 30% in the *A. vera* group were in clinical remission whereas only 7% reached clinical remission in the placebo group. Sigmoidoscopic scores, however, were not different between the groups, and adverse events were only minor and were similar in both groups. The authors concluded that their results are encouraging although not conclusive. They emphasize that any possible clinical benefits suggested by this trial are modest[10]. These results, however, indicate the need for further, larger controlled trials of *A. vera* gel, not only in moderately active ulcerative colitis but also in the maintenance of remission in ulcerative colitis and in Crohn's disease.

The anti-inflammatory effects of *A. vera* gel were further investigated by Langmead et al. in human colorectal mucosa *in vitro*[11]. *A. vera* gel exerted a dose-dependent inhibitory effect on reactive oxygen metabolite production and inhibited the production of prostaglandin E2, thus supporting the proposal that *A. vera* gel may have a therapeutic effect in IBD[11].

Wheat grass juice

Wheat grass juice is an extract from mature sprouts of wheat seeds (*Triticum aestivum*), which is believed to possess therapeutic qualities when it is consumed immediately after extraction on an empty stomach. Ben-Arie et al. report that this kind of treatment was brought to their attention by several patients with ulcerative colitis in Israel, who regularly used such an extract and experienced clinical improvement[12]. Based on these reports Ben-Arie et al. designed and performed a randomized, double-blind, placebo-controlled trial to examine the effects of wheat grass juice in the treatment of active distal ulcerative colitis[12].

Out of a group of more than 600 telephone-contacted patients about one-third reported a clinical history suggestive of IBD, but finally only 24 patients with active ulcerative colitis fulfilled all inclusive criteria and entered the trial. Following baseline clinical and endoscopic assessment patients were randomized into a group of 12 who received wheat grass juice and a group of 12 patients who received a placebo juice. Wheat grass juice was especially produced from organically grown wheat seeds using the wheat grass harvested at a height of 20 cm. The dose of wheat grass and of placebo juice was gradually increased from 20 ml on the first day to 100 ml by the fifth day, and continued at this dose daily for 1 month. Within 3 days following the termination of treatment sigmoidoscopies were repeated on each participant. A disease activity index created by combining stool frequency, rectal bleeding, sigmoidoscopic score and physician's assessment of disease activity was

documented, and statistical analyses were performed. The disease activity index improved in 10 wheat grass juice patients versus in only five placebo patients, and rectal bleeding improved in nine wheat grass juice patients versus seven placebo patients. Furthermore several clinical symptoms tended to improve. The authors concluded that wheat grass juice is safe and may prove to be beneficial. They also realize, critically, that they may have investigated a very small superselected group of patients, and that much more work needs to be done in order to prove that wheat grass juice indeed possesses anti-inflammatory properties, and to establish the active components[12].

Germinated barley foodstuff

Germinated barley foodstuff (GBF) is obtained by milling and sieving the residual brewer's spent grain and contains glutamine-rich protein and hemicellulose rich dietary fibre[13]. It has recently been used by a group of Japanese researchers as prebiotic treatment of experimental colitis in comparison with probiotic or antibiotic treatment[14]. Encouraged by the finding of a significant reduction of colonic inflammation in a dextran sodium sulphate colitis model in rats, and based on the finding that in this model GBF increased the luminal butyrate concentration in the caecum[14], a clinical investigation of the efficacy of GBF as a maintenance therapy in patients with ulcerative colitis while in remission was performed[15] in Japan. In this trial 59 patients with ulcerative colitis in remission were randomized to receive either conventional treatment with 5-aminosalicylic acid (5-ASA) and/or steroids or conventional treatment plus daily 20 g of GBF for 12 months. The response to treatment was assessed by monitoring the Rachmilewitz clinical activity index and an endoscopic score at 3, 6 and 12 months. After 3 months a significant decrease in steroid dose was observed in both groups compared with entry levels; the dose of steroid in the GBF group was significantly lower compared with the control group. But most importantly the cumulated recurrence rate in the GBF group was significantly lower than in the control group[15].

The authors reported no side-effects related to GBF and concluded that GBF appears to be effective and safe as a maintenance therapy in patients with ulcerative colitis[15]. As this study was a non-randomized, open-label trial the results must be considered with caution, and the authors realize the need for double-blind and well-controlled clinical trials with long-term follow-up to fully evaluate the clinical efficacy of GBF in preventing relapse of ulcerative colitis[15].

Sophora flavescens enemas

Sophora flavescens belongs to the family of Leguminosae and grows as evergreen shrub in Asia. It is an important medicinal herb in China, where roots and flowers are used for a wide variety of indications including ulcerative colitis. In 1994 Chen et al. reported on a clinical study in treating intractable ulcerative colitis with traditional Chinese medicine[16], in which the Chinese phytopharmacon Jian Pi Ling tablets, together with retention enemas of radix sophorae flavescentis and flos sophora decoction per night, was applied. This

article is written in Chinese and unfortunately only a translation of the abstract in English is available[16]. In this clinical double-blind study 153 patients with intractable ulcerative colitis were treated with traditional Chinese medicine in comparison with salicylazosulphapyridine (SASP) and retention enema of dexamethasone. Patients were randomized into three treatment groups: Group 1 was treated with Jian Pi Ling tablets with retention enema of radix sophorae flavescentis and flos sophora decoction per night, Group 2 was treated with SASP and rentention enema of dexamethasone, and Group 3 with placebo with rentention enema as in Group 1.

After 90 days fibre endoscopy, and checks for pathological and immunological parameters were performed, and a group comparison was also performed. The results are reported as curative rates in Groups 1, 2 and 3 with 53%, 28% and 19%, respectively. Furthermore a total effective rate is reported for Group 1 with 86%, Group 2 with 60% and Group 3 with 45%. In Group 1 B lymphocytes decreased markedly. The authors conclude that Group 1 (Jian Pi Ling tablets with retention enema of radix sophorae flavescentis and flos sophorae decoction per night) seems to be the best therapeutic programme. Unfortunately no details for randomization, content of Jian Pi Ling tablets, quality of sophora flavescentis preparations nor detailed laboratory data and definition of intractable ulcerative colitis, curative rates and total effective rates are given in the abstract. Therefore the true therapeutic effect cannot be judged reliably. The Guidelines of the German Society of Gastroenterology (DDG) and the German Morbus Crohn/Colitis Ulcerosa Vereinigung (DCCV) recommend that Jian Pi Ling tablets should not be used outside traditional Chinese medicine, as in Europe little experience is available[17].

Curcuma longa

Curcuma longa is a member of the Zingiberaceae (ginger) family, a perennial herb commonly named turmeric. *C. longa* grows to a height of about 1 m and is cultivated extensively in Asia, India, China and other countries with a tropical climate.

Dried *C. longa* is the source of the spice turmeric, the ingredient that gives curry powder its characteristic yellow colour. The rhizome is the portion of the plant used medicinally. Turmeric has a long tradition of use in the Chinese and Ayurvedic systems of medicine, particularly as anti-inflammatory agent[18]. Recently it was shown that curcumin, a component of turmeric, is able to attenuate colitis in the dinitrobenzene sulphonic acid (DNB)-induced murine model of colitis[19].

Based on this observation Holt et al. administered a pure curcumin preparation in an open-label study to five patients with ulcerative proctitis, and five with Crohn's disease. Within 12 weeks of treatment nine of 10 patients improved clinically, had lowered Crohn's disease activity index scores and sedimentation rates. Based on this encouraging pilot study the authors postulated the need for double-blind, placebo-controlled, follow-up studies[20].

Table 2 Other CAM modalities tested in IBD

Bovine colostrum enema
Seal oil via nasuduodenal feeding tube
Acupoint catgut embedding
Acupuncture and moxibustion

Non-herbal CAM treatments tested in IBD

In addition to herbal therapies a variety of other CAM modalities have been tested for efficacy in IBD (Table 2).

Bovine colostrum enema

Colostrum is the first diet of mammalian neonates and is rich in immunoglobulins, antimicrobial peptides, growth factors and valuable nutritional components. Recently colostrum enemas have been suggested for the treatment of inflammatory bowel diseases[21] and Khan et al. have reported a pilot study utilizing bovine colostrum enemas in a small group of 14 patients with distal ulcerative colitis[22]. Fourteen patients with distal colitis of mild to moderate disease activity were randomized to receive either twice-daily colostrum enemas (100 ml of 10% solution) or placebo (albumin solution) for 4 weeks. Both groups also received a daily dose of 1.6 g mesalazine or, if already taking it, had a dose increment of 1.6 g per day. Disease activity was documented at 0, 2 and 4 weeks and all patients underwent fibreoptic colonic examination on the initial visit and a rigid sigmoidoscopy with biopsies at subsequent visits. After 4 weeks the eight patients in the colostrum group showed a reduction in symptom score while the six patients in the placebo group experienced no improvement. The histological score improved in five of eight patients in the colostrum group but in only two of six patients in the placebo group. The authors conclude that bovine colostrum enema showed potential as a novel therapy for left-sided colitis with additional benefits over using mesalazine alone[22]. Further clinical trials of the use of colostrum enemas for the treatment of active colitis are awaited with interest.

Seal oil

A recent pilot study by the National Institute of Nutrition and Seafood Research, Bergen, Norway, tested the effect of n-3-rich seal oil in comparison with n-6-fatty acid rich soy oil on joint pain in IBD. Nineteen patients with IBD and joint pain as extraintestinal manifestation of IBD (nine patients with Crohn's disease and 10 patients with ulcerative colitis) were treated through a nasoduodenal feeding tube with three-times daily 10 ml seal oil or 10 ml soy oil for 10 days. At the end of the 10-day treatment period the authors reported a significant reduction in the duration of morning stiffness, number of tender joints, intensity of pain and the doctor's scoring of rheumatic disease activity in patients receiving seal oil[23]. Furthermore, over a 6-month post-treatment

observation period these beneficial effects of seal oil administration were maintained, whereas soy oil tended to aggravate the condition. Surprisingly this short-time administration of seal oil reduced in IBD patients the initially elevated ratios of n-6 to n-3 fatty acids and arachidonic acid to eicosapentaenoic acid, both in serum[23] and in rectal mucosa[24], to the level in controls. These observations warrant further investigations.

Acupoint catgut embedding

A Chinese group of authors recently reported clinical observations on acupoint catgut embedding therapy for treatment of ulcerative colitis compared with oral administration of salicylazosulphapyridine, 4–6 g per day. The article is written in Chinese and only a short English abstract is available[25].

A total of 116 patients with ulcerative colitis were randomly divided into a treatment group of 56 patients, who were treated with catgut embedding at several traditional acupoints and a control group treated with oral administration of 4–6 g of salicylazosulphapyridine per day. The authors report an improvement of clinical symptoms and of endoscopy findings at 4 and 6–8 weeks after treatment in both groups, concluding with the statement that acupoint catgut embedding has a better therapeutic effect on ulcerative colitis with less adverse reactions[25]. As this report neither states the severity of the disease at the onset of the trial nor gives any detailed information on clinical symptom scoring, endoscopy scoring or other clinical or biochemical parameters, nor any evidence on statistical calculations, the validity of these observations cannot be judged.

Acupuncture and moxibustion

Acupuncture and moxibustion are established treatment modalities in traditional Chinese medicine. Recently a group of Chinese researchers in Shanghai[26] reported a detailed investigation on the mechanisms of acupuncture and moxibustion treatment for ulcerative colitis in a locally devised animal model involving SD rats. In this rat model for ulcerative colitis effects of acupuncture and moxibustion on gene expression of cytokines were investigated, and some data suggested that acupuncture and moxibustion can greatly inhibit the expression of interleukins IL-1β and IL-6 mRNA in this colitis model[26]. Moxibustion cones contained mugwort, which is botanically *Artemisia vulgaris*, in Europe also known as St John's Plant (Cingulum Sancti Johannis).

In Germany the Research Group for Alternative Medicine at F.A. University of Erlangen-Nuremberg established two prospective randomized controlled trials to evaluate the effect of acupuncture and moxibustion in the treatment of active Crohn's disease[27] and in the treatment of ulcerative colitis[28].

A total of 51 patients with mild to moderately active Crohn's disease were treated in a single centre for complementary medicine by three trained acupuncturists and randomly assigned to receive either traditional acupuncture (TCM group, $n = 27$) or control treatment at non-acupuncture points (control group, $n = 24$). Patients were treated in 10 sessions over a period

of 4 weeks and followed up for 12 weeks. Outcome measures were the change in Crohn's disease activity index, in quality of life and general well-being, and in serum markers of inflammation. In the TCM group the CDAI decreased by 56 points as compared with a mean decrease of 46 points in the control group. In both groups these changes were associated with improvements in general well-being and quality of life. With regard to general well-being, traditional acupuncture was superior to control treatment. The authors conclude that, apart from a marked placebo effect, traditional acupuncture offers an additional therapeutic benefit in patients with mild to moderately active Crohn's disease[27].

Joos et al. designed and performed another prospective randomized, controlled, clinical trial involving 29 patients with mild to moderately active ulcerative colitis to investigate the efficacy of acupuncture and moxibustion in the treatment of active ulcerative colitis[28]; 15 patients were randomized to the verum group, treated in 10 sessions over a period of 5 weeks by traditional acupuncture and moxibustion, 14 patients served as control group, treated with sham acupuncture in the same schedule. Outcome parameters were changes in colitis activity index, quality of life, well-being and several markers of inflammation. The authors describe decreases in colitis activity index scores in both groups, significant improvements in general well-being and quality of life; however, they showed no differences between treatment and control groups. In their conclusion the authors state that both traditional and sham acupuncture seem to offer some therapeutic benefit in patients with mild to moderately active ulcerative colitis[28].

SUMMARY AND CONCLUSIONS

Complementary and alternative medications (CAM) obviously are frequently used by patients with IBD. Despite its widespread use homeopathy so far has not been tested and evaluated in IBD. A recent meta-analysis comparing placebo-controlled trials of homeopathy and allopathy concluded that the clinical effects of homeopathy are placebo effects[4]. In Great Britain, where alternative therapies are now a degree subject (Bachelor of Science, BSc) at some British universities, the debate rages over whether homeopathy should be given scientific status in the British education system, as many scientists and advocates of evidence-based medicine feel that homeopathy is unscientific[29].

By contrast, an increasing number of herbal drugs have been tested both in ulcerative colitis and in Crohn's disease. Most of the studies with herbal and other complementary and alternative therapies have enrolled small numbers of patients and confirmatory studies by a different group of authors are lacking.

Presently *P. ovata* seeds may be considered as effective in maintaining remission in ulcerative colitis, *B. serrata* extract H15 may have a positive effect to induce remission of mild Crohn' disease, within 8 weeks. Jian Pi Ling tablets have been reported to be effective in 'intractable ulcerative colitis'[16]. Confirmatory studies are lacking, however, and there is little experience with Jian Pi Ling tablets in Western medicine. German guidelines advise that Jian Pi Ling tablets should not be used outside traditional Chinese medicine[17].

Studies with *A. vera* gel, wheat grass juice, germinated barley foodstuff, curcumine, colostrum enemas, seal oil, acupuncture and acupuncture with moxibustion presently cannot yet be considered as established effective treatments of IBD. Published preliminary results in the study author's conclusions look promising, and warrant further investigations. Time and future trials will show whether some of these CAM can establish themselves as evidence-based in well-defined clinical conditions of patients with IBD, as happened with probiotics[17].

Since CAM treatments compete with EBM treatments, and the development of new effective drugs is expensive and strictly regulated, the same regulations should be mandatory also for CAM treatments, especially with respect to purity, admixture of potentially harmful impurities, toxicity and stability of herbal drugs. Although CAM treatments in general are considered safe by their users, adverse reactions, especially with herbal medicines, have been reported. Drug compatibility with conventional medicines and potentially harmful drug interactions should therefore be followed with care.

References

1. Langmead L, Rampton DS. Review article: Complementary and alternative therapies for inflammatory bowel disease. Aliment Pharmacol Ther. 2006;23:341–9.
2. Joos St, Rosemann T, Szecsenyi J, Hahn EG, Willich StN, Brinkhaus B. Use of complementary and alternative medicine in Germany – a survey of patients with inflammatory bowel disease. Complement Altern Med. 2006;6:19.
3. Gangl A. Alternative and complementary therapies for inflammatory bowel disease. Nature Clin Prac Gastroenterol Hepatol. 2006;3:180–1.
4. Shang A, Huwiler-Müntener K, Nartey L et al. Are the clinical effects of homeopathy placebo effects? Comparative study of placebo controlled trials of homeopathy and allopathy. Lancet. 2005;366:726–32.
5. Fernández-Bañares F, Hinojosa J, Sánchez-Lombraña JL et al. Randomized clinical trial of *Plantago ovata* seeds (dietary fiber) as compared with mesalamine in maintaining remission in ulcerative colitis. Am J Gastroenterol. 1999;94:427–33.
6. Ammon HPT. Boswellic acids in chronic inflammatory diseases. Planta Med. 2006;72:1100–16.
7. Ammon HPT. Boswelliasäuren (Inhaltsstoffe des Weihrauchs) als wirksame Prinzipien zur Behandlung chronisch entzündlicher Erkrankungen. Wien Med Wochenschr. 2002;152:373–8.
8. Anthoni C, Laukoetter MG, Rijcken E et al. Mechanisms underlying the anti-inflammatory actions of boswellic acid derivatives in experimental colitis. Am J Physiol Gastrointest Liver Physiol. 2006;290:G1131–17.
9. Gerhardt H, Seifert F, Buvari P, Vogelsang H, Repges R. Therapie des aktiven Morbus Crohn mit dem Boswellia-serrata Extrakt H15. Z Gastroenterol. 2001;39:11–17.
10. Langmead L, Feakins RM, Goldthorpe S et al. Randomized, double-blind, placebo-controlled trial of oral *Aloe vera* gel for active ulcerative colitis. Aliment Pharmacol Ther. 2004;19:739–47.
11. Langmead L, Makins RJ, Rampton DS. Anti-inflammatory effects of *Aloe vera* gel in human colorectal mucosa *in vitro*. Aliment Pharmacol Ther. 2004;19:521—7.
12. Ben-Arie E, Goldin E, Wengrower D, Stamper A, Kohn R, Berry E. Wheat grass juice in the treatment of active distal ulcerative colitis. Scand J Gastroenterol. 2002;37:444–9.
13. Bamba T, Kanauchi O, Andoh A, Fujiyama Y. A new prebiotic from germinated barley for nutraceutical treatment of ulcerative colitis. J Gastroenterol Hepatol. 2002;17:818–24.
14. Fukuda M, Kanauchi O, Araki Y et al. Prebiotic treatment of experimental colitis with germinated barley food stuff: a comparison with probiotic or antibiotic treatment. Int J Mol Med. 2002;9:65–70.

15. Hanai H, Kanauchi O, Mitsuyama K et al. Germinated barley foodstuff prolongs remission in patients with ulcerative colitis. Int J Mol Med. 2004;13:643–7.
16. Chen ZS, Nie ZW, Sun OL. Clinical study in treating intractable ulcerative colitis with traditional Chinese medicine. Zhongguo Zhong Xi Yi Jie He Za Zhi. 1994;14:400–2.
17. Leitlinie der Deutschen Gesellschaft für Verdauungs- und Stoffwechselkrankheiten (DDG) und der Deutschen Morbus Crohn/Colitis ulcerosa Vereinigung (DCCV). Diagnostik und Therapie der Colitis ulcerosa. AWMF online Nr. 021/009p; www.uni.duesseldorf.de.
18. *Curcuma longa* (turmeric). Monograph. Altern Med Rev. 2001;6(Suppl):S62–6.
19. Salh B, Assi K, Templeman V et al. Curcumin attenuates DNB-induced murine colitis. Am J Physiol Gastrointest Liver Physiol. 2003;285:G235–43.
20. Holt PR, Katz S, Kirshoff R. Curcumin therapy in inflammatory bowel disease: a pilot study. Dig Dis Sci. 2005;50:2191–3.
21. Playford RJ, Macdonald ChE, Johnson WS. Colostrum and milk derived peptide growth factors for the treatment of gastrointestinal disorders. Am J Clin Nutr. 2000;72:5–14.
22. Khan Z, Macdonald C, Wicks AC et al. Use of the nutriceutical, bovine colostrum, for the treatment of distal colitis: results from an initial study. Aliment Pharmacol Ther. 2002;16:1917–22.
23. Bjorkkjaer T, Brunborg LA, Arslan G et al. Reduced joint pain after short-term duodenal administration of seal oil in patients with inflammatory bowel disease: comparison with soy oil. Scand J Gastroenterol. 2004;39:1088–94.
24. Bjorkkjaer T, Brun JG, Valen M et al. Short-term duodenal seal oil administration normalised n-6 to n-3 fatty acid ratio in rectal mucosa and ameliorated bodily pain in patients with inflammatory bowel disease. Lipids Health Dis. 2006;5:6.
25. Li HJ, Li GP, Li HY. Clinical observation on acupoint catgut embedding therapy for treatment of ulcerative colitis. Zhongguo Zhen Jiu. 2006;26:261–3.
26. Wu HG, Zhou LB, Pan YY et al. Study of the mechanisms of acupuncture and moxibustion treatment for ulcerative colitis rats in view of the gene expression of cytokines. World J Gastroenterol. 1999;6:515–17.
27. Joos S, Brinkhaus B, Maluche C et al. Acupuncture and moxibustion in the treatment of active Crohn's disease: a randomized controlled study. Digestion. 2004;69:131–9.
28. Joos S, Wildau N, Kohnen R et al. Acupuncture and moxibustion in the treatment of ulcerative colitis: a randomized controlled study. Scand J Gastroenterol. 2006;41:1056–63.
29. Giles J. Degrees in homeopathy slated as unscientific. Nature. Published online: 21 March 2007:doi:10.1038/446352a.

Section VII
Treatment algorithms: “The standard patient”

Chair: IE KOUTROUBAKIS and A TROMM

19
Mild-to-moderate left-sided ulcerative colitis and proctitis

G. J. MANTZARIS

INTRODUCTION AND DEFINITIONS

Ulcerative colitis (UC) is a chronic disease characterized by varying degrees of inflammation of the colonic mucosa. The disease almost always affects the rectum, but variably extends proximally to involve other parts of or the entire colon and in some cases the far distal part of the terminal ileum.

Based on the extent of colonic involvement the Montreal classification has recently defined three subgroups of UC: ulcerative proctitis (UP), when the disease is limited to the rectum; left-sided colitis (LS-UC), when the disease extends up to but distal to the splenic flexure; and extensive colitis, when the disease extends proximally to the splenic flexure[1]. Previously, the terms LS-UC and distal colitis were used to define disease extending proximal to the sigmoid colon but distal to the splenic flexure or disease confined to the rectosigmoid area, respectively[2]. It is unlikely that different factors determine the extent of colitis, and the area of colonic involvement is not stable over time since extension but also regression and skip lesions have been described[3]. Furthermore, although most clinicians are inclined to use endoscopic criteria[2], there is no consensus as to whether the proximal area of colonic involvement should be defined by endoscopic or histological criteria[4,5]. However, defining the extent of disease in general, and during a given flare in particular, has several important therapeutic implications, albeit the ability of rectally administered therapy (enemas, foams, gels, or suppositories) to reach the proximal end of colonic inflammation.

In this chapter therapeutic strategies for the induction of remission, as well as for remission maintenance, of patients with active mild-to-moderate LC-UC and UP will be reviewed.

INDUCTION OF REMISSION

There are several strategies to treat patients with active mild-to-moderate attacks of LS-UC and UP, such as topical therapies with preparations of

aminosalicylates or corticosteroids, oral formulations of aminosalicylates or corticosteroids, or a combination of the two[2,6,7]. The decision depends on various patient- and disease-related parameters, such as patient preferences and compliance with treatment, disease extent, disease behaviour, latest treatment regimen employed for inducing remission, local availability of drug preparations, cost of treatment, etc.

Topical therapy

General rules of treatment

Corticosteroids and aminosalicylates have been used for the treatment of LS-UC and UP. Corticosteroids were the first agents to be used, almost 60 years ago[8–11]. Various aminosalicylate formulations (foam, gel, or liquid enemas, and suppositories) have also been commercially available for the treatment of LS-UC and UP. Hydrocortisone, prednisone, prednisolone, betamethasone, beclometasone, but also locally acting corticosteroids with low systemic availability have also been formulated for rectal administration. However, the availability of these formulations varies between countries.

Corticosteroid and aminosalicylate preparations for topical therapy differ in chemical properties, dose of active drug, retention time by the inflamed mucosa, and area of colonic distribution after rectal administration. Topical treatment should be directed according to the extent of disease and the patient's preferences. Thus, suppositories should be used for the treatment of UP and distal sigmoiditis because they may spread up to the distal sigmoid colon, as confirmed by scientigraphic studies[12,13]. In contrast, gels, foams, and especially high-volume liquid enemas may spread more proximally and are suitable for the treatment of proximal sigmoiditis and LS-UC[14]. Although higher-volume liquid enemas may achieve a more proximal distribution reaching the splenic flexure they are more difficult to tolerate; in contrast, gel enemas and especially foam enemas are more comfortable, easier to retain, and probably coat more homogeneously the area of inflamed mucosa, and this may be true especially for rectal preparations of aminosalicylates[15,16]. Studies have shown that, although equally effective, patients prefer mesalazine gel to mesalazine foam, and the latter over mesalazine enemas for the treatment of active LS-UC[15–17]. Furthermore, mesalazine suppositories are preferred over hydrocortisone foams for the treatment of active proctitis[18].

Rectally administered corticosteroids

Controlled trials and a meta-analysis have demonstrated that rectally administered foam/liquid enemas of hydrocortisone, betamethasone, and prednisolone are superior to placebo in achieving clinical, endoscopic, and histological response and remission in patients with acute LS-UC and UP[8–10,19]. Patients treated with corticosteroids were four or five times more likely to achieve symptomatic and endoscopic improvement compared with those treated with placebo[19].

The comparative effect of various preparations of corticosteroids for topical therapy has also been studied. In a 4-week controlled trial, Campieri et al. randomized 157 patients with ulcerative proctosigmoiditis to 3 mg/60 ml beclometasone dipropionate enemas or 30 mg/60 ml prednisolone sodium phosphate enemas. At the end of the trial there were no significant differences in the overall clinical and endoscopic remission rates between the two treatment regimens (29% vs 25%, respectively)[20]. Similar results were obtained in the study of Ruddell et al., who compared the efficacy of hydrocortisone enemas and rectal hydrocortisone foam in the treatment of relapses of ulcerative proctosigmoiditis[21].

Unwanted systemic effects of classical steroids prompted the development of glucocorticoids with potent local activity and low systemic bioavailability for the treatment of inflammatory bowel disease, but only budesonide has been marketed in formulations for rectal (liquid and foam enema) and oral [controlled ileal release (Entocort®) or pH-modified release (Budenofalk®)] treatment. Budesonide is rapidly absorbed by the inflamed colonic mucosa but undergoes rapid extensive first-pass metabolism in the liver, resulting in very few systemic corticosteroid effects. The levels of cortisol did not change, and suppression of ACTH was not detected after 4–6 weeks treatment with 2 mg/day budesonide liquid or foam enemas[22,23]. In a series of randomized controlled trials budesonide was more effective than placebo in inducing remission of active LS-UC and UP[22]. In a controlled trial Danielsson et al. randomized 41 patients with active ulcerative proctosigmoiditis to a 4-week treatment with budesonide liquid enemas (2 mg/100 ml) or placebo. Patients not responding after 2 weeks of treatment were switched to open budesonide treatment. After 4 weeks significantly more patients had achieved sigmoidoscopic and histological improvement on budesonide ($p<0.01$) than on placebo. In addition, significantly more patients in the placebo arm switched over to open budesonide treatment ($p<0.001$)[24]. In a large multicentre, double-blind, randomized, controlled North American study, 233 patients were randomized to a 6-week treatment with three different doses of budesonide enema (0.5, 2.0, or 8.0 mg/100 ml) or placebo. The lowest effective dose of budesonide was 2.0 mg/100 ml, and this achieved significantly higher clinical (21% vs 4%, $p<0.05$), endoscopic and histological remission scores compared with placebo without affecting the ACTH-stimulated cortisol response[25]. More recently, Lindgren et al. randomized 149 patients with active distal colitis to budesonide enemas (2 mg/100 ml) twice daily or placebo enema in the morning and budesonide enema in the evening[26]. Patients were followed for 8 weeks or until full remission of disease. Subsequently, patients in remission were randomized to either budesonide enema or placebo enema twice weekly for 24 weeks or until relapse. There were no significant differences with either doses at 4 and 8 weeks (33% vs 41%, and 51% vs 54%, for once-daily or twice-daily enemas, respectively), but the twice-daily dose led to increased frequency of impaired adrenal function. In contrast, twice-weekly budesonide enemas were insufficient to maintain remission of disease. Budesonide foam (2 mg/50 ml) was as effective as enemas in the treatment of active ulcerative proctitis or proctosigmoiditis; foam was preferred by most patients[27].

Studies comparing 2–8 weeks treatment with 2 mg or 4 mg/100 ml budesonide foam or liquid enemas with various formulations of classical corticosteroids (hydrocortisone acetate foam, prednisolone enemas, or beclometasone dipropionate enemas) have shown that the former are equally effective to classical corticosteroids in inducing clinical, endoscopic and histological remission and improving quality of life of patients with UP and proctosigmoiditis, but budesonide enemas produced less endogenous cortisol suppression[22,28–34]. This has also been confirmed by meta-analysis data[19]. Based on this evidence budesonide may substitute for classical corticosteroids in the treatment of LS-UC. However, one should keep in mind that the majority of the aforementioned studies have included patients with UC extending up to the descending colon ('distal' UC) rather than to the splenic flexure. Therefore, these data should be interpreted with caution when treating patients with true LS-UC.

Rectally administered aminosalicylates

The available evidence based on results of randomized controlled trials and meta-analyses data has confirmed that rectally administered aminosalicylates at dose ranges of 1–4 g/day are superior to placebo and rectally administered corticosteroid formulations in the induction of clinical, endoscopic, and histological remission of LS-UC and UP[2,6,7,35–44]. The effect of topical aminosalicylates in LS-UC appears to be time- but not dose-dependent, and they act more quickly than oral mesalazine. Patients tolerate easier gel enemas or foam enemas than liquid enemas of mesalazine, and prefer mesalazine suppositories to steroid suppositories for the treatment of UP[15–18,45,46].

In the most recent meta-analysis. by Cohen et al., 67 studies for active LS-UC were identified (17 placebo-controlled with 10 of them passing quality assessment)[42]. Mesalazine enemas achieved remission in a duration but not dose response. For all studies assessed the remission rates were higher than corticosteroid enemas, and clinical improvement rates were superior to oral therapies. For active UP, 18 studies were identified, nine placebo-controlled with three passing quality assessment. Again, mesalazine suppositories achieved clinical improvement and remission in a duration but also dose response, with higher rates of remission than topical corticosteroids. In the meta-analyses of Marshal and Irvine[19,41,43] the pooled odds ratio for clinical, endoscopic and histological response or remission of LS-UC treated with rectal mesalazine versus placebo was 7.71, 6.55, and 6.91, respectively. Furthermore, rectal mesalazine was significantly better than rectally administered classical corticosteroids for inducing symptomatic, endoscopic, and histological remission with a pooled odds ratio of 2.42 (95% CI 1.72–3.41), 1.89 (1.29–2.76), and 2.03 (1.28–3.20), respectively. The adverse event profile was minor, and a cost analysis showed mesalazine enemas to be less expensive than rectal corticosteroids.

Mesalazine enemas appear to be more effective or at least equally effective, compared with budesonide enemas in inducing remission of LS-UC and UP. In the multicentre single-blind group comparative 4-week trial by Leman et al.[47] 97 patients with active distal UC and UP were randomized to budesonide enema (2 mg/100 ml) or 5-aminosalicylic acid (5-ASA) enema (mesalazine 1 g/

100 ml). Budesonide and 5-ASA enemas led to a significant improvement in symptom, endoscopy and histopathology scores, although differences between the two groups were insignificant. However, the clinical remission rate at 4 weeks was significantly higher for 5-ASA enema than for budesonide enema (60% vs 38%, respectively, $p = 0.03$). The adverse event profile was similar. In the open 4-week study by Rufle et al.[33] patients with LS-UC or proctosigmoiditis were randomized to budesonide foam (1 mg/50 ml) twice daily or mesalazine enemas (4 g/60 ml) once daily[48]. Clinical, endoscopic and histological improvements were not significantly different between the two groups (67% and 71% for budesonide and mesalazine enemas, respectively). Similar results were obtained when mesalazine enemas were compared to beclometasone dipropionate enemas[49,50]. In the study by Mulder et al. 2 g/day mesalazine enemas were compared to 3 mg/day beclometasone dipropionate enemas or the combination of both topical treatments in patients with active UP. The combination treatment was superior to either monotherapy, but there were no significant differences between mesalazine and beclometasone enemas[49]. In an Italian study comparing 1 g/day 5-ASA to 3 mg/day beclometasone dipropionate enemas in an open, 6-week trial in patients with active ulcerative proctosigmoiditis, both treatments resulted in a significant decrease in disease activity scores but there were no significant differences in efficacy or safety between the topical therapies[50].

Oral therapy

Oral sulphasalazine and aminosalicylates

Sulphasalazine was the first oral drug to be used in the treatment of UC. Sulphasalazine is a pro-drug consisting of one moiety of 5-ASA bound to sulphapyridine, an antibiotic, via an azo-bond. Bacterial reductases break the azo-bond and 5-ASA is released in the colon. Sulphapyridine may also exert beneficial effects in patients with UC but it is also responsible for most of the adverse events attributed to sulphasalazine; this renders approximately 10–15% of patients intolerant of sulphasalazine. This has prompted the production of newer 5-ASA preparations with a much better safety and tolerance profile than sulphasalazine. 5-ASA preparations release the active 5-ASA moiety in the colon after cleavage of an azo-bond linking either two moieties of 5-ASA (olsalazine) or 5-ASA to a carrier molecule (balsalazide); alternatively, 5-ASA is released in the small intestine in a pH-dependent (mesalazine) or sustained-release manner and the active moiety is delivered in the colon. Finally, mixed delayed- and sustained-release formulations of 5-ASA (mesalazine pellets and mesalazine with MMX technology) aim at gradually releasing 5-ASA, beginning in the terminal ileum and continuing throughout the colon to the rectum.

Sulphasalazine induces remission in approximately 70–75% of patients with acute mild-to-moderate UC and it is also effective in patients with LC-UC[51]. In the latest Cochrane meta-analysis newer release formulations of 5-ASA tended towards therapeutic benefit over sulphasalazine with a pooled Peto odds ratio of 0.83 (95% CI 0.60–1.13) for the failure to induce global/clinical improvement or remission, and 0.66 (95% CI 0.42–1.04) for the failure to

induce endoscopic improvement in acute mild-to-moderate UC. However, these differences were not statistically significant[52]. The effect of sulphasalazine is dose-dependent but doses higher than 4 g/day are usually intolerable. Oral 5-ASA preparations may also induce remission in approximately 50–75% of patients with mild-to-moderate UC[52,53]. In the previously mentioned meta-analysis the pooled Peto odds ratio for the failure to induce global/clinical improvement or remission over placebo was 0.40 (95% CI 0.30–0.53); in other words, 5-ASA was at least twice as effective as placebo for inducing improvement or remission of mild-to-moderate UC. A dose–response trend was observed for oral 5-ASA. In the ASCEND II trial, mesalazine induced a dose–response effect in clinical response but no clinically relevant differences in remission rates were seen. There were also no significant differences between various extents of LS-UC in patients receiving equal doses of mesalazine[54].

Although there are very few studies directly comparing the effect of topical mesalazine to oral mesalazine or sulphasalazine, it seems that topical mesalazine is superior in the treatment of acute LS-UC and UP[55,56]. In a 6-week, double-blind, double-dummy, parallel-group, multicentre study in patients with active mild-to-moderate distal UC, 4 g/day 5-ASA suspension enema was as effective as 4 g/day oral sulphasalazine in the treatment of active disease. However, topical therapy resulted in earlier clinical improvement and had fewer and milder adverse events compared with oral sulphasalazine[55]. Topical therapy is much more effective for UP because oral mesalazine is unlikely to reach the rectum in amounts sufficient to induce remission because of proximal colonic stasis, increased absorption, and rapid transit through the inflamed colonic mucosa[56,57]. Another advantage is that local treatment acts more rapidly, resulting in a faster resolution of symptoms, especially bleeding and tenesmus.

Oral corticosteroids

Oral corticosteroids have been the mainstay of therapy for moderate-to-severe attacks of UC for more than 50 years after the original studies of Truelove. However, their therapeutic efficacy has not been specifically tested in controlled trials in patients with mild-to-moderate LS-UC, although in routine clinical practice many of these patients will inevitably be treated with corticosteroids if 5-ASA fails to control the disease. With the advent of orally administered topically acting corticosteroids which lack remarkable systemic steroid activity few randomized, placebo-controlled trials have included patients with acute LS-UC. In a 9-week randomized, double-blind, controlled trial in patients with acute extensive or LS-UC, Lofberg et al. compared the effect of 10 mg oral budesonide to that of 40 mg prednisolone on the clinical and endoscopic remission rates. Thirty-four patients received budesonide, and 38 patients received prednisolone[58]. The endoscopic scores of UC improved significantly and were not different between the two groups, but histological scores were significantly reduced only in the prednisolone group. Similar numbers of patients were withdrawn from the study as non-responders and, as expected, the morning plasma levels of cortisol were less affected by budesonide.

Budesonide was also effective in patients with LS-UC. However, the power of this study does not allow any firm and strong conclusions. Another randomized controlled trial failed to show any superiority of oral fluticasone propionate (5 mg q.i.d.) over placebo in reducing the clinical, endoscopic, or histological activity of mild-to-moderate active LS-UC after 4 weeks of treatment[59]. In another study Rizzello et al. evaluated the additive effect of oral beclometasone dipropionate (5 mg/day) to oral 5-ASA (3.2 g/day) in a 4-week, placebo-controlled, double-blind study in patients with mild-to-moderate active extensive or LS-UC. Fifty-eight patients received oral 5-ASA and beclometasone dipropionate and 61 received oral 5-ASA and placebo. Although patients in both groups improved clinically, endoscopically and histologically, the effect of combined treatment was even more enhanced irrespective of the extent of UC[60,61]. Subsequently, another Italian multicentre study evaluated the comparative effect of 5 mg/day beclometasone dipropionate to that of 2.4 g/day 5-ASA in a 4-week randomized, parallel-group, single-blind study in patients with active mild-to-moderate extensive and LS-UC. Ninety patients were administered beclometasone dipropionate and 87 patients were administered 5-ASA. The clinical remission rates were significantly reduced and were not different between the two study groups, but the Disease Activity Index was more significantly reduced in the beclometasone arm of the study only in patients with extensive disease[62].

Combined oral and rectal therapy

There is growing evidence that combined oral and rectal preparations of 5-ASA may further increase the rates of clinical response and remission of extensive UC compared with either orally or rectally administered monotherapy[2,6,7,19,40,63]. In the study by Marteau et al., combined oral treatment with 4 g of a slow-release formulation of 5-ASA with either 1 g 5-ASA enema or placebo enema achieved clinical response and remission rates after 4 and 8 weeks respectively, which were among the highest ever reported for a 5-ASA treatment[63]. It is reasonable to assume that this effect may be even more augmented in LS-UC, probably as a result of higher mucosal concentrations of active 5-ASA in the inflamed colon[57,64]. However, robust evidence is lacking since very few studies have addressed the issue of combined therapy for distal disease in view of the high efficacy of rectal 5-ASA monotherapy. In a small randomized, double-blind, placebo-controlled study in patients with active mild-to-moderate distal UC, Safdi et al. randomized 18 patients to 4 g/day mesalazine rectal suspension enema, 22 patients to 2.4 g/day oral mesalazine, and 20 patients to combined therapy with oral and rectal mesalazine. Although differences in clinical remission rates were insignificant, combination therapy at weeks 3 and 6 produced a greater improvement in total Disease Activity Index scores than did oral or rectal monotherapy. Improvement was mirrored both by physician and patient independent rating of disease status. Furthermore, the combination therapy had a faster effect on complete relief of rectal bleeding[40]. Combination therapy may be especially useful for the more refractory patients before recommending oral corticosteroids[2,6,7,36].

Patients who are refractory to treatment with high doses of combined oral and rectal aminosalicylates may benefit by the addition of topical corticosteroids[49].

TREATMENT OF ACTIVE MILD-TO-MODERATE LS-UC AND UP

Treatment of active LS-UC

Topical mesalazine should be considered as first-line treatment for patients with active LS-UC (Figure 1). Overall, clinical response or remission may be achieved in up to 80% of patients after 4–6 weeks with topical mesalazine[11]. As mentioned previously, a variety of mesalazine formulations (gel, enemas, foams) have been produced for treating LS-UC, but enemas are more widely available worldwide. A variety of mesalazine doses have been tested clinically, ranging from 1 to 4 g daily, but the effect of topical mesalazine is not dose- but rather time-dependent[6,7,11,42]. Therefore, prolongation of treatment may increase the proportion of patients achieving remission of disease.

Topical treatment with classical corticosteroids, such as 100 mg hydrocortisone enemas, 10% hydrocortisone acetate foams, prednisolone enemas, betamethasone enemas, beclometasone dipropionate enemas, etc., are an effective second-line treatment for patients intolerable of or unresponsive to topical mesalazine for 2 weeks. Budesonide enemas dosed at 2 mg offer an excellent alternative by being at least as effective as topical corticosteroids but with fewer systemic side-effects.

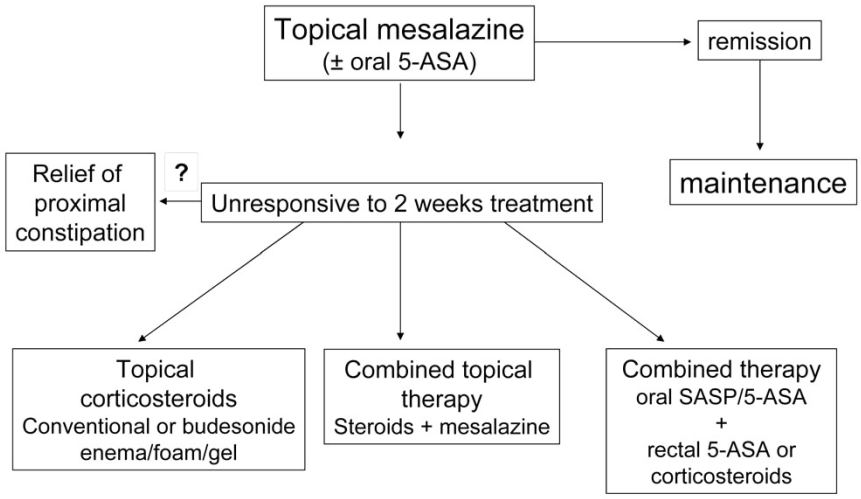

Figure 1 Induction of remission of left-sided colitis (and proctosigmoiditis)

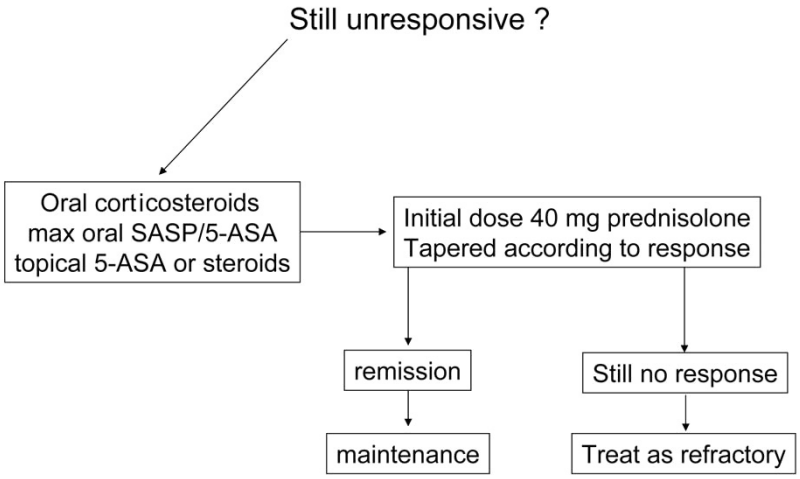

Figure 2 Induction of remission of left-sided colitis (and proctosigmoiditis)

Patients without adequate rapid response to topical mesalazine or corticosteroids, as well as patients with moderate disease activity, may still be benefit from combined therapy with topical mesalazine and corticosteroids. Alternatively, these patients should be treated with a combination of oral sulphasalazine or any other 5-ASA formulation (mesalazine, olsalazine, balsalazide) and rectal mesalazine or oral 5-ASA and rectal corticosteroids. Effective doses for oral sulphasalazine range between 4 and 6 g/day in four divided doses, for oral mesalazine 2.4–4.8 g/day (Eudragit-S coated) or 3–4.5 g/day (Eudragit-L coated), for oral olsalazine 1.5–3 g/day, for oral balsalazide 6.5 g/day in three divided doses, for mesalamize pellets 3 g/day as a single daily dose, and for MMX mesalazine 2.4–4.8 g/day[2,65,66]. A dose–response is seen with Eudragit-S coated mesalazine and balsalazide but not with MMX mesalazine. Mucosal healing has been documented with sulphasalazine, slow-release formulations of 5-ASA and MMX mesalazine.

It should be remembered that topical therapy is not easily tolerated and compliance is compromised with prolongation of treatment because of local irritation, discomfort, and inconvenience. Furthermore, it should be emphasized that prolonging inadequate treatment should be avoided because it leads to refractory disease or even to extensive UC. Therefore, patients with inadequate response to the combination of oral 5-ASA and topical mesalazine or corticosteroids should be treated aggressively with oral corticosteroids (Figure 2). An initial dose of 40 mg prednisolone or equivalent should be administered until the patient shows a satisfactory clinical response. Then the dose should be tapered progressively according to disease severity and rapidity of clinical response[2,6]. However, some experts recommend relief of proximal constipation by smooth laxatives before stepping up therapy in patients with LS-UC or UP unresponsive to combined oral and topical therapy.

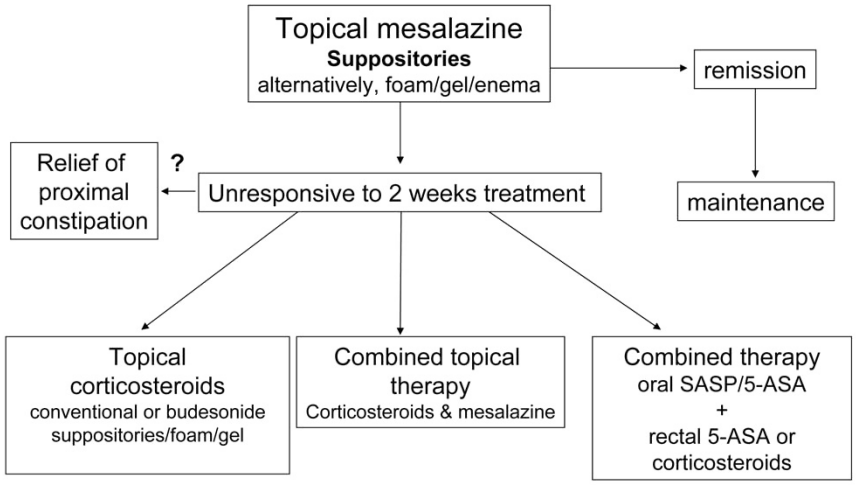

Figure 3 Induction of remission of ulcerative proctitis

Treatment of active UP

Endoscopically confirmed UP should be treated initially with mesalazine suppositories (500 mg twice daily or 1 g at night) for at least 1 month (Figure 3). Treatment is well tolerated and produces very few local or systemic side-effects. If suppositories are not tolerated, mesalazine foams or gels, or even liquid enemas, in a dose of 1–4 g/day are an effective alternative.

Patients who are intolerant of, or do not respond rapidly to, topical mesalazine should be treated with rectally administered corticosteroid suppositories, foams, or enemas, or with a combination of topical mesalazine and corticosteroids. The formulation to be chosen is totally dependent upon patient preferences and tolerance and local availability, as has been previously discussed. Oral 5-ASA (such as mesalazine 2.4–4.8 g/day or 3–4.5 g/day) may also be added. For the occasional patient who is still unresponsive to all these regimens relief of proximal constipation and re-evaluation of the extent of disease is recommended before UP is considered as refractory to 5-ASA and treated with oral steroids.

For patients who are refractory to treatment with a combination of oral and topical 5-ASA or topical corticosteroids, oral corticosteroids should be added (Figure 4). The initial dose of corticosteroids should be tailored by the severity and activity of disease. The rate of tapering should be guided by the relief of clinical symptoms and endoscopic healing.

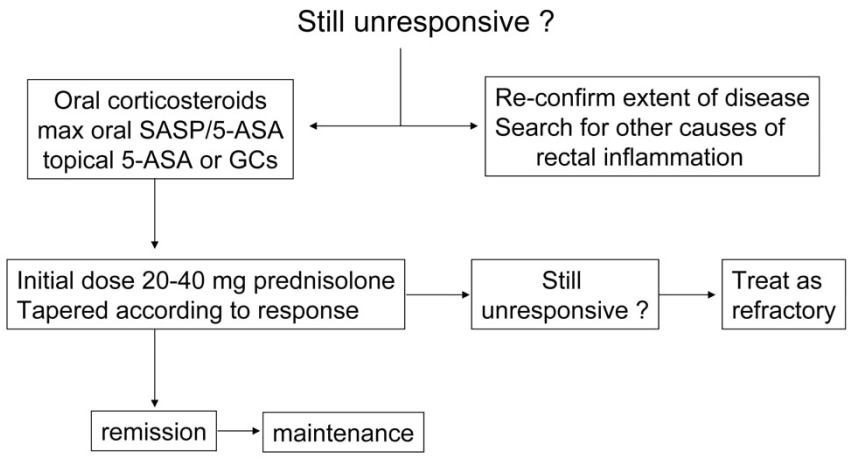

Figure 4 Induction of remission of ulcerative colitis

MAINTENANCE OF REMISSION

Patients with UC achieving remission with any of the aforementioned treatment regimens are at increased risk for disease relapse if they stop treatment. The goals of maintenance treatment are to maintain complete relief of symptoms of active UC, heal the inflamed mucosa and, ideally, eliminate the infiltration of the colonic mucosa by acute inflammatory cells, because all these ensure true quiescent disease and guarantee an excellent quality of life. A plethora of adequately powered randomized controlled trials have documented beyond any doubt the efficacy of oral sulphasalazine and oral 5-ASA formulations (mesalazine, balsalazide, olsalazine) to reduce the incidence of relapse in ulcerative colitis and maintain effective remission of disease (Figure 5)[2,6,7,51,67–74]. However, it has become increasingly evident over the past 20 years that topical aminosalicylates may also play an important role in the maintenance of remission, at least in patients with LC-UC and UP[67–69,71,75–79].

Oral and rectal corticosteroids

Oral and topically administered classical corticosteroids cannot maintain remission of disease and their long-term use is associated with potentially serious and occasionally life-threatening systemic adverse events[42,80]. In addition, repeated therapy with corticosteroids leads inevitably on to steroid-dependency and/or steroid-refractoriness. These patients should be treated with corticosteroid-sparing agents, such as azathioprine, 6-mercaptopurine, or even infliximab. Topical therapy with the locally acting corticosteroid budesonide has also proven ineffective to maintain remission of LS-UC and UP[26].

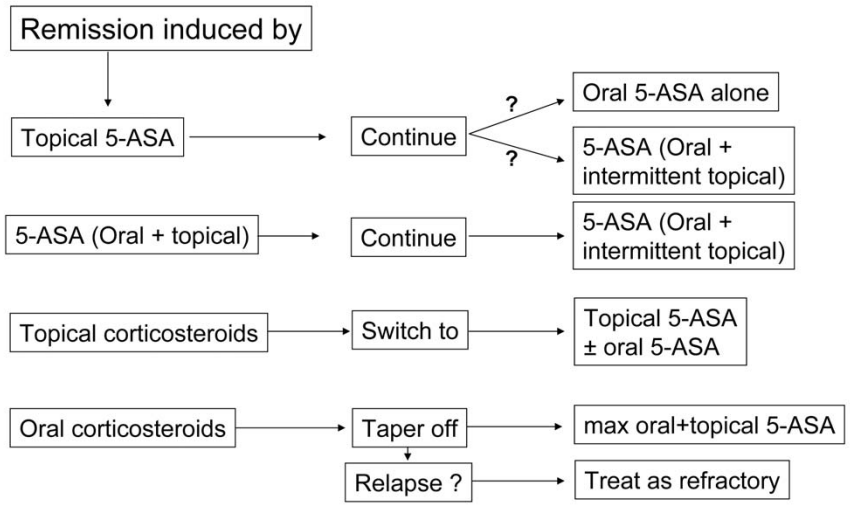

Figure 5 Maintenance of remission of left-sided colitis (and proctosigmoiditis)

Oral and rectal aminosalicylates

Sulphasalazine is effective in maintaining remission of LS-UC; the lowest effective maintenance dose was 2 g/day[2]. Furthermore, in randomized-controlled trials the newer oral formulations of 5-ASA, mesalazine (Eudragit-S coated 2.4–3.2 g/day or Eudragit L-coated 1.5–3 g/day), olsalazine (1 g/day), balsalazide (3–6 g/day) have proven efficacy for maintaining remission of LS-UC and proctitis[2,6,7,72–74]. Mesalazine pellets have not yet been evaluated for remission maintenance but MMX mesalazine (1.2 g b.i.d.) maintains remission of extensive and LS-UC (S. Hanauer, personal communication).

Rectally administered aminosalicylates are also effective as monotherapy in maintaining remission of LS-UC and UP when administered even intermittently, such as twice or three times a week. D'Albasio et al. randomized 60 patients with ulcerative proctosigmoiditis in remission to oral sulphasalazine (2 g/day) or intermittent therapy with mesalazine enemas (4 g for the first 7 days each month). After 2 years, 43% of patients in the former group versus 37% of patients in the latter group were in remission; these differences were not significant[75]. In another study of more or less similar design, Mantzaris et al. randomized 38 patients with quiescent distal UC to rectal mesalazine (4 g enemas every third night) or oral Eudragit-L coated mesalazine (1.5 g/day). After 2 years, 74% and 32% ($p < 0.001$) of patients maintained on enemas and oral 5-ASA, respectively, were still in remission[76].

Combination therapy with oral and rectally administered 5-ASA may further increase the remission rates of LS-UC and UP. D'Albasio et al. randomized 72 patients with quiescent ulcerative proctosigmoiditis to combination therapy

with Eudragit-S coated tablets (1.6 g/day) and mesalazine enemas (4 g b.i.w.) or oral therapy (as previously) and twice-weekly placebo enemas, and showed a significantly lower annual relapse rate in favour of the active treatment (39% versus 69%, $p = 0.036$)[69]. In an open investigator-blind trial, Mantzaris et al. treated a group of patients with frequent relapses of ulcerative proctosigmoiditis and proctitis with a combination of oral Eudragit-L coated mesalazine (1.5 g/day) and rectal mesalazine (4 g enemas three times a week) and showed that most of the patients were able to maintain remission of disease without any corticosteroids or steroid-sparing agents[77].

Topical therapy is even more effective for maintaining remission of UP, and the effect appears to be dose-dependent. In an Italian study, 111 patients with UP in remission were randomized to 1 year treatment with 500 mg mesalazine suppositories twice a day (36 patients), 500 mg mesalazine once daily (40 patients), or placebo. A dose–response relationship was seen with a cumulative annual relapse rate of 10% (95% CI 0–21), 32% (95% CI 16–49), and 47% (95% CI 29–65) in the mesalazine twice-daily, once-daily, and placebo groups, respectively[71]. In another study Hanauer et al. administered 1 g mesalazine suppositories in divided daily doses or placebo to a group of 65 patients with UP and showed significantly fewer relapses and prolongation of the remission period with active treatment[79]. Finally, Marteau et al. randomized 95 patients to intermittent mesalazine suppositories (1 g three times a week) or placebo. Patients who relapsed received 1 g of mesalazine suppositories daily. At the end of 1-year follow-up, 39% of patients on active treatment per protocol versus 72% on placebo ($p < 0.001$) had relapsed[80].

Thus, rectal aminosalicylates are effective for maintaining remission of LS-UC and UP: suppositories are suitable for UP, gels or foams for proctosigmoiditis, whereas enemas are convenient for more proximal disease. In contrast to acute treatment where efficacy is time-dependent rather than dose-dependent, a dose–response relationship appears to exist for suppositories in maintaining remission of UP. Enemas may be still effective for maintaining remission of LS-UC and proctosigmoiditis when dosed even every 3 days. Sulphasalazine and the newer oral 5-ASA formulations are also effective for maintaining remission of LS-UC, but their efficacy as monotherapy is poorer for patients with UP. The choice of oral or topical therapy is also dependent on patient preferences and compliance. Oral 5-ASA may also prevent proximal extension in subsequent flares of UC[67]. Finally, treatment of more frequently relapsing patients may be optimized by combining daily rectal and oral 5-ASA formulations since combination therapy is more effective than either oral or topical monotherapy for maintaining remission of LS-UC and UP. In contrast, corticosteroids are ineffective for maintaining remission of LS-UC and UP.

MAINTENANCE OF REMISSION OF LS-UC AND UP

Maintenance treatment of LS-UC

The choice of maintenance treatment regimen is dependent on various patient and disease parameters, such as disease extent, disease behaviour, latest treatment regimen employed for inducing remission, patient preferences and

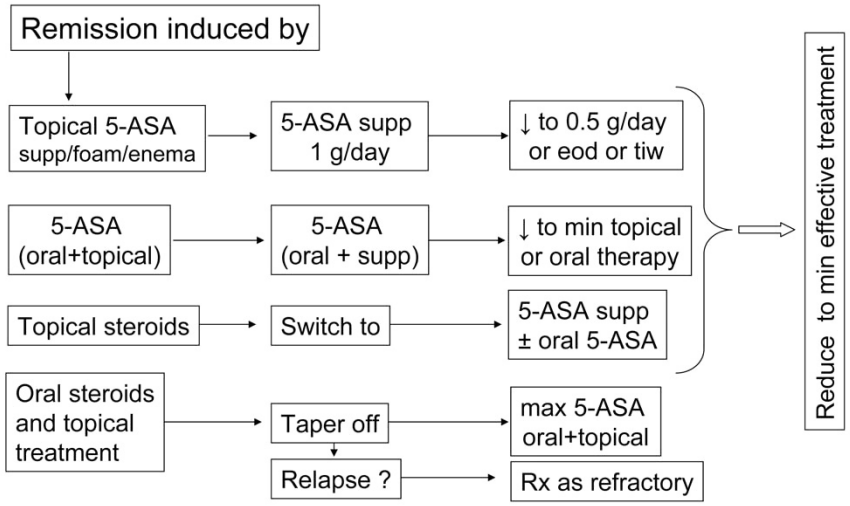

Figure 6 Maintenance of remission of ulcerative proctitis

compliance with treatment, etc. Therefore, patients who have achieved remission on rectal 5-ASA enemas, gels, or foams should continue to use these indefinitely. If intolerable, oral therapy may substitute effectively for topical therapy; the latter may be administered intermittently (every other or every third day) if the disease tends to relapse more frequently (Figure 5).

Patients who have achieved remission of disease on oral and rectal mesalazine should continue the same regimen for at least 1–2 months. Subsequently, the topical therapy may be reduced to every other day or every third day administration of liquid enemas (gels or foams for ulcerative proctosigmoiditis). In the unlucky occasion of a frequently relapsing disease the patient should be maintained on combination therapy indefinitely before stepping up to immunomodulators.

Patients who have achieved remission on topical corticosteroids may switch to 5-ASA preparations for remission maintenance (with or without oral 5-ASA according to experience from the frequency of previous relapses).

Patients who have achieved clinical remission on oral corticosteroids but are in the process of tapering steroids should receive a maximal dose of oral 5-ASA and mesalazine or corticosteroid enemas. The decision to add azathioprine or 6-MP is dependent upon prior experience with similar flares or clinical judgement. Patients who have tapered off corticosteroids without becoming steroid-dependent should be maintained on long-term oral and topical 5-ASA preparations. In contrast, patients who relapse upon or soon after withdrawal of corticosteroids should be treated as steroid-dependent using immunomodulators or infliximab.

Patients who are in long-term remission on a given combination 5-ASA regimen may attempt to reduce or even taper off either oral or (more commonly) rectal 5-ASA. This may also stand true for patients in remission on oral or rectal monotherapy. Oral therapy may be reduced to the lowest dose of 2 g/day sulphasalazine, 1 g/day olsalazine, 1.2 g/day or 1.5 g/day mesalazine, or 3 g/day balsalazide. For topical therapy, enemas may be administered as infrequently as between three times a week and once weekly. In any case, renal function should be monitored frequently to pick up on rare cases of aminosalicylate-induced nephropathy.

Maintenance treatment of UP

Patients who have achieved remission on rectal suppositories, gels, foams, or even enemas should maintain remission on 5-ASA suppositories 1 g/day (as a single or divided dose) (Figure 6). This dose can be effectively tapered to 500 mg–1 g every other day or three times a week. Additional oral 5-ASA therapy may be required for maintaining remission in patients who have achieved remission on rectal corticosteroids or combination therapy with oral corticosteroids and rectal corticosteroids or 5-ASA preparations.

Patients unable to maintain remission on topical 5-ASA preparations and who require continuous treatment with corticosteroids to relieve rectal bleeding and tenesmus should probably be treated with immunomodulators.

References

1. Satsangi J, Silverberg MS, Vermeire S, Colombel J-F. The Montreal classification of inflammatory bowel disease: controversies, consensus, and implications. Gut. 2006;55: 749–53.
2. Kornbluth A, Sachar DB. Ulcerative colitis practice guidelines in adults (Update): American College of Gastroenterology, Practice Parameters Committee. Am J Gastroenterol. 2004;99:1371–85.
3. Silvernerg MS, Satsangi J, Ahmad T et al. Toward an integrated clinical, molecular and serological subclassification of inflammatory bowel disease: report of a Working Party of the 2005 Montreal World Congress of Gastroenterology. Can J Gastroenterol. 2005;19 (Suppl. A):5–36.
4. Modigliani R. Endoscopic management of inflammatory bowel disease. Am J Gastroenterol Supplement. 1994;8:53–65.
5. Waye JD. Endoscopy in inflammatory bowel disease. In: Kirsner JD, Shorter RG, editors. Inflammatory Bowel Disease, 3rd edn. Philadelphia: Lea & Fabiger, 1988:353–76.
6. Rugeiro M, Loftus EVJr, Steinhart H, Cohen RD. Clinical guidelines for the medical management of left-sided ulcerative colitis and ulcerative proctitis: Summary statement. Inflamm Bowel Dis. 2006;12:972–8.
7. Rugeiro M, Loftus EVJr, Steinhart H, Cohen RD. Medical management of left-sided ulcerative colitis and ulcerative proctitis: clinical evaluation of therapeutic trials. Inflamm Bowel Dis. 2006;12:979–94.
8. Truelove SC. Treatment of ulcerative colitis with local hydrocortisone hemisuccinate sodium: a report on a controlled therapeutic trial. Br Med J. 1958;2:1072–7.
9. Watkinson G. Treatment of ulcerative colitis with topical hydrocortisone hemisuccinate sodium: a controlled trial employing restricted sequential analysis. Br Med J. 1958;2:1077–82.
10. Lennard-Jones JE, Baron JH, Connell AM et al. A double blind controlled trial of prednisolone21-phosphate suppositories in the treatment of idiopathic proctitis. Gut. 1962;3:207–10.

11. Vecchi M, Saibeni S, Devani M, Rondonotti E, De Franchis R. Review article: Diagnosis, monitoring and treatment of distal colitis. Aliment Pharmacol Ther. 2003;17(Suppl. 2):2–6.
12. Jay M, Beihn RM, Digenis GA et al. Disposition of radiolabelled suppositories in humans. J Pharmacol. 1985;37:266–8.
13. Williams CN, Haber G, Aquino JA. Double-blind placebo-controlled evaluation of 5-ASA suppositories in active proctitis and measurement of extent of spread using 99m-Tc labeled 5-ASA suppositories. Dig Dis Sci. 1987;32:71–5S.
14. Campieri M, Corbelli C, Gionchetti P et al. Spread and distribution of 5-ASA colonic foam and 5-ASA enemas in patients with ulcerative colitis. Dig Dis Sci. 1992;37:1890–7.
15. Pokrotnieks J, Marlicz K, Paradowski L, Margus B, Zaborowski P, Greinwald R. Efficacy and tolerability of mesalazine foam enema (Salofalk foam) for distal ulcerative colitis: a double-blind, randomized, placebo-controlled study. Aliment Pharmacol Ther. 2000;14: 1191–8.
16. Ardizzone S, Doldo P, Ranzi T et al. Mesalazine foam (Salofalk foam) in the treatment of active distal ulcerative colitis. A comparative trial vs Salofalk enema. The SAF-3 study group. Ital J Gastroenterol Hepatol. 1999;31:685–7.
17. Gionchetti P, Ardizzone S, Benvenuti ME et al. A new mesalazine gel enema in the treatment of left-sided ulcerative colitis: a randomized controlled multicentre trial. Aliment Pharmacol Ther. 1999;13:381–8.
18. Farup PG, Hovde O, Halvorsen FA, Raknerud N, Brodin U. Mesalazine suppositories versus hydrocortisone foam in patients with distal ulcerative colitis. A comparison of the efficacy and practicality of two topical treatment regimens. Scand J Gastroenterol. 1995;30: 164–70.
19. Marshall JK, Irvine EJ. Rectal corticosteroids versus alternative treatments in ulcerative colitis: a meta-analysis. Gut. 1997;40:775–81.
20. Campieri M, Cottone M, Miglio F et al. Beclomethasone dipropionate enemas versus prednisolone sodium phosphate enemas in the treatment of distal colitis. Aliment Pharmacol Ther. 1998;12:361–6.
21. Ruddell WS, Dickinson RJ, Dixon MF, Axon AT. Treatment of distal ulcerative colitis (proctosigmoiditis) in relapse: comparison of hydrocortisone enemas and rectal hydrocortisone foam. Gut. 1980;21:885–9.
22. Marin-Jimenez I, Pena AS. Budesonide for ulcerative colitis. Rev Esp Enferm Dig. 2006;98: 362–73.
23. Ostergaard-Thomsen O, Andersen T, Langholz E et al. Lack of adrenal gland suppression with budesonide enemas in active distal ulcerative colitis. Eur J Gastroenterol Hepatol. 1994;6:507–11.
24. Danielsson A, Lofberg R, Persson T et al. A steroid enema, budesonide, lacking systemic effects for the treatment of distal ulcerative colitis or proctitis. Scand J Gastroenterol. 1992; 27:9–12.
25. Hanauer SB, Robinson M, Pruitt R et al. on behalf of the US Budesonide Enema Group. Budesonide enema for the treatment of active, distal ulcerative colitis and proctitis: a dose-ranging study. Gastroenterology. 1998;115:525-32.
26. Lindgren S, Lofberg R, Bergholm L et al. Effect of budesonide enema on remission and relapse rate in distal ulcerative colitis and proctitis. Scand J Gastroenterol. 2002;37:705–10.
27. Gross V, Bar-Meir S, Lavy A et al. on behalf of the International Budesonide Foam Study Group. Budesonide foam versus budesonide enema in active ulcerative proctitis and proctosigmoiditis. Aliment Pharmacol Ther. 2006;23:303–12.
28. Danielsson A, Hellers G, Lyrenas E et al. A controlled randomized trial of budesonide versus prednisolone retention enemas in active distal ulcerative colitis. Scand J Gastroenterol. 1987;22:987–92.
29. Group DBS. Budesonide enema in active distal ulcerative colitis. A randomized dose–response trial with prednisolone enema as positive control. Danish Budesonide Study Group. Scand J Gastroenterol. 1991;26:1225–30.
30. Bianchi-Poro G, Prantera C, Campieri M et al. Comparative trial of methylprednisolone and budesonide enemas in active distal ulcerative colitis. Eur J Gastroenterol Hepatol. 1994;6:125–30.
31. Lofberg R, Ostergaard Thomsen O, Langholz E et al. Budesonide versus prednisolone retention enemas in active distal ulcerative colitis. Aliment Pharmacol Ther. 1994;8:623–9.

32. Tarpila S, Turunen U, Seppala K et al. Budesonide enema in active haemorrhagic proctitis – a controlled trial against hydrocortisone foam enema. Aliment Pharmacol Ther. 1994;8: 591–5.
33. Bar-Meir S, Fidder HH, Faszczyk M et al. for the International Budesonide Study Group. Budesonide foam versus hydrocortisone acetate foam in the treatment of active ulcerative proctosigmoiditis. Dis Colon Rectum. 2003;46:929–36.
34. Hammond A, Andus T, Gierend M, Ecker KW, Scholmerich J, Herfarth H on behalf of the German Budesonide Foam Study Group. Controlled, open, randomized multicenter trial comparing the effects of treatment on quality of life, safety and efficacy of budesonide foam and betamethasone enemas in patients with active distal ulcerative colitis. Hepatogastroenterology. 2004;51:1345–9.
35. Gionchetti P, Rizzello F, Morselli C, Tambasco R, Campieri M. Review article: Aminosalicylates for distal colitis. Aliment Parmacol Ther. 2006(Suppl. 3):41–4.
36. Travis SPL. Review article: Induction therapy for patients with active ulcerative colitis. Aliment Pharmacol Ther. 2006;24(Suppl. 1):10–16.
37. Hanauer SB. Review article: High-dose aminosalicylates to induce and maintain remission in ulcerative colitis. Aliment Pharmacol Ther. 2006;24(Suppl. 3):37–40.
38. Sutherland LR, Martin F, Greer S et al. 5-Aminosalicylic acid enema in the treatment of distal ulcerative colitis, proctosigmoiditis, and proctitis. Gastroenterology. 1987;92:1894–8.
39. Hanauer SB. Dose-ranging study of mesalamine (PENTASA) enemas in the treatment of acute ulcerative proctosigmoiditis: results of a multicentered placebo-controlled trial. The US Pentasa Enema Study Group. Inflamm Bowel Dis. 1998;4:79–83.
40. Safdi M, DeMicco M, Sninsky C et al. A double-blind comparison of oral versus rectal mesalamine versus combination therapy in the treatment of distal ulcerative colitis. Am J Gastroenterol. 1997;92:1867–71.
41. Marshall JK, Irvine EJ. Rectal aminosalicylate therapy for distal ulcerative colitis: a meta-analysis. Aliment Pharmacol Ther. 1995;9:293–300.
42. Cohen RD, Woseth DM, Thisted RA, Hanauer SB. A meta-analysis and overview of the literature on treatment options for left-sided ulcerative colitis and ulcerative proctitis. Am J Gastroenterol. 2000;95:1263–76.
43. Marshall JK, Irvine EJ. Putting rectal 5-aminosalicylic acid in its place: the role in distal ulcerative colitis. Am J Gastroenterol. 2000;95:1628–36.
44. Lee FI, Jewell DP, Mani V et al. A randomised trial comparing mesalazine and prednisolone foam enemas in patients with acute distal ulcerative colitis. Gut. 1996;38: 229–33.
45. Campieri M, Paoluzi P, D'Albasio G, Brunetti G, Pera A, Barbara L. Better quality of therapy with 5-ASA colonic foam in active ulcerative colitis. A multicenter comparative trial with 5-ASA enema. Dig Dis Sci. 1993;38:1843–50.
46. Malchow H, Gertz B; CLAFOAM Study Group. A new mesalazine foam enema (Claversal foam) compared with a standard liquid enema in patients with active distal ulcerative colitis. Aliment Pharmacol Ther. 2002;16:415–23.
47. Lemann M, Galian A, Rutgeerts P et al. Comparison of budesonide and 5-aminosalicylic acid enemas in active distal ulcerative colitis. Aliment Pharmacol Ther 1995;9:557–62.
48. Rufle W, Fruhmorgen P, Huber W, Kimmig JM. Budesonide foam as a new therapeutic principle in distal ulcerative colitis in comparison with mesalazine enema. An open, controlled, randomized and prospective multicenter pilot study. Z Gastroenterol. 2000;38:287–93.
49. Mulder CJ, Fockens P, Meijer JW, van der Heide H, Wiltink EH, Tytgat GN. Beclomethasone dipropionate (3 mg) versus 5-aminosalicylic acid (2 g) versus the combination of both (3 mg/2 g) as retention enemas in active ulcerative proctitis. Eur J Gastroenterol Hepatol. 1996;8:549–53
50. Gionchetti P, D'Arienzo A, Rizzello F et al. Topical treatment of distal active ulcerative colitis with beclomethasone diproprionate or mesalamine: a single-blind randomized controlled trial. J Clin Gastroenterol. 2005;39:291–7.
51. Sutherland LR, May GR, Shaffer EA. Sulfasalazine Revisited: A meta-analysis of 5-aminosalicylic acid in the treatment of ulcerative colitis. Ann Intern Med. 1993;118:540–9.
52. Sutherland L, Macdonald JK. Oral 5-aminosalicylic acid for induction of remission in ulcerative colitis. Cochrane Database Syst Rev. 2006;(2):CD000543.

53. Sutherland L, Roth D, Beck P. Alternatives to sulfasalazine: a meta-analysis of 5-ASA in the treatment of ulcerative colitis. Inflamm Bowel Dis. 1997;3:665–78.
54. Hanauer SB, Sandborn WJ, Kornbluth A et al. Delayed-release oral mesalamine at 4.8 g/day (800 mg tablet) for the treatment of moderately active ulcerative colitis: the ASCEND II trial. Am J Gastroenterol. 2005;100:2478–85.
55. Kam L, Cohen H, Dooley C, Rubin P, Orchard J. A comparison of mesalamine suspension enema and oral sulfasalazine for treatment of active distal ulcerative colitis in adults. Am J Gastroenterol. 1996;91:1338–42.
56. Gionchetti P, Rizzello F, Venturi A et al. Comparison of oral with rectal mesalazine in the treatment of ulcerative proctitis. Dis Colon Rectum. 1998;41:93–7.
57. Hebden JM, Blaskshaw PE, Perkins AC et al. Limited exposure of the healthy distal colon to orally-dose formulation is further exaggerated in active left-sided ulcerative colitis. Aliment Pharmacol Ther. 2000;14:155–61.
58. Lofberg R, Danielsson A, Suhr O et al. Oral budesonide versus prednisolone in patients with active extensive and left-sided ulcerative colitis. Gastroenterology. 1996;110:1713–18.
59. Angus P, Snook JA, Reid M et al. Oral fluticasone propionate in active distal ulcerative colitis. Gut. 1992;33:711–14.
60. Rizzello F, Gionchetti P, Galeazzi R et al. Oral beclomethasone dipropionate in patients with mild to moderate ulcerative colitis: a dose-finding study. Adv Ther. 2001;18:261–71.
61. Rizzello F, Gionchetti P, D' Arienzo A et al. Oral beclometasone dipropionate in the treatment of active ulcerative colitis: a double-blind placebo-controlled study. Aliment Pharmacol Ther. 2002;16:1109–16.
62. Campieri M, Adamo S, Valpiani D et al. Oral beclometasone dipropionate in the treatment of extensive and left-sided active ulcerative colitis: a multicentre randomised study. Aliment Pharmacol Ther. 2003;17:1471–80.
63. Marteau P, Probert CS, Lindgren S et al. Combined oral and enema treatment with Pentasa (mesalazine) is superior to oral therapy alone in patients with extensive mild/moderate active ulcerative colitis: a randomised, double blind, placebo controlled study. Gut. 2005;54:960–5.
64. Frieri G, Pimpo MT, Palombo GC et al. Rectal and colonic mesalazine concentration in ulcerative colitis: oral versus oral plus topical therapy. Aliment Pharmacol Ther. 1999;13:1413–17.
65. Kamm MA, Sandborn WJ, Gassull M et al. Once-daily high concentration MMX mesalamine in active ulcerative colitis. Gastroenterology. 2007;132:66–75.
66. Lichtenstein GR, Kamm MA, Boddu P et al. Effect of once- or twice-daily MMX mesalamine (SPD476) for the induction of remission of mild to moderately active ulcerative colitis. Clin Gastroenterol Hepatol. 2007;5:95–102.
67. Pica R, Paoluzi OA, Iacopini F et al. Oral mesalazine (5-ASA) treatment may protect from proximal extension of mucosal inflammation in ulcerative proctitis. Inflamm Bowel Dis. 2004;10:731–6.
68. D'Albasio G, Trallori G, Gavazzi O et al. Combined therapy with 5-aminosalicylic tablets and enemas for maintaining remission in ulcerative colitis. Ital J Gastroenterol. 1991;23:12–14.
69. D'Albasio G, Pacini F, Gamari E et al. Combined therapy with 5-aminosalicylic acid tablets and enemas for maintaining remission in ulcerative colitis: a randomized double-blind study. Am J Gastroenterol. 1997;92:121143–7.
70. Miner P, Hanauer SB, Robinson M et al. Safety and efficacy of controlled release mesalamine for maintenance of remission in ulcerative colitis. Dig Dis Sci. 1995;40:296–304.
71. D'Albasio G, Paoluzi P, Campieri M et al. Maintenance treatment of ulcerative proctitis with mesalazine suppositories: a double-blind placebo-controlled trial. Italian IBD Study Group. Am J Gastroenterol. 1998;93:799–803.
72. Green JRB, Gibson JA, Kerr GD et al. Maintenance of remission of ulcerative colitis: a comparison between balsalazide 3 g daily and mesalazine 1.2 g daily over 12 months. Aliment Pharmacol Ther. 1998;12:1207–16.
73. Kruis W, Schreiber S, Theuer D et al. Low dose balsalazide (1.5 g twice daily) and mesalazine (0.5 g three times daily) maintained remission of ulcerative colitis but high dose balsalazide (3.0 g twice daily) was superior in preventing relapses. Gut. 2001;49:783–9.

74. Sutherland L, Macdonald JK. Oral 5-aminosalicylic acid for maintenance of remission in ulcerative colitis. Cochrane Database Syst Rev. 2006;(2):CD000544.
75. D'Albasio G, Trallori G, Ghetti A et al. Intermittent therapy with high dose 5-aminosalicylic acid enemas for maintaining remission in ulcerative proctosigmoiditis. Dis Colon Rectum. 1990;44:394–7.
76. Mantzaris GJ, Hatzis A, Petraki K, Spiliadi Ch, Triantaphyllou G. Intermittent therapy with high-dose 5-aminosalicylic acid enemas maintains remission in ulcerative proctitis and proctosigmoiditis. Dis Colon Rectum. 1994;37:58–62.
77. Mantzaris GJ, Kontogiannis Ph, Georgiou G et al. Combined oral and rectal administration of 5-aminosalicylic acid maintains remission of frequently relapsing ulcerative proctosigmoiditis and proctitis. Hel J Gastroenterol. 1994;7:212–16.
78. Marteau P, Grand J, Foucault M et al. Use of mesalazine slow-release suppositories 1 g three times per week to maintain remission in ulcerative proctitis: a randomized double blind placebo controlled multicentre study. Gut. 1998;42:195–9.
79. Hanauer SB, Good LI, Goodman MW et al. Long-term use of mesalamine (Rowasa) suppositories in remission maintenance of ulcerative proctitis. Am J Gastroenterol. 2000; 95:1749–54.
80. Lennard-Jones JE, Misiewicz JJ, Connell AM et al. Prednisone as maintenance treatment for ulcerative colitis in remission. Lancet. 1965;191:188–9.

20
Treatment of mild-to-moderate episodes of extensive ulcerative colitis

P. MARTEAU and X. DRAY

INTRODUCTION

The treatment of ulcerative colitis (UC) depends on the severity of the episode and on the extent of the lesions[1]. A mild-to-moderate episode of active disease is defined by the presence of mild-to-moderate symptoms and mucosal lesions. The most frequent symptoms are bloody stools, diarrhoea, tenesmus, and mild abdominal pain. Severe episodes of UC defined as the presence of bloody stool frequency $\geqslant 6$/day associated with either tachycardia (>90 bpm), or temperature $>37.8^{\circ}$C, or anaemia (haemoglobin <10.5 g/dl), or an elevated ESR (>30 mm/h) are not discussed in this chapter. Lesions are said to be 'extensive' when they spread above the splenic flexure of the colon (often even the whole colon); they cannot be fully reached by a topically acting therapeutic enema and therefore require that at least part of the treatment should be given orally or using a systemic route.

The four main drugs which can be used either alone or in combination are aminosalicylates, steroids, calcineurin inhibitors and infliximab (IFX). Thiopurines (such as azathioprine or 6-mercaptopurine) can also (and often must) be used but they are too slow-acting to be considered as single therapies in this situation. Other drugs (including antibiotics and probiotics) or treatments (for example leucocytapheresis) are currently studied but the evidence for their efficacy is presently still too low to recommend their use in the common practice. Finally, surgery (i.e. subtotal colectomy or proctocolectomy), may be needed.

Choosing the optimal treatment strategy requires confirmation of active UC by endoscopy before starting treatment, or in case of treatment failure, and search for alternative causes for inflammation, especially infection by stool and mucosal biopsy examination. One must also consider the balance between drug potency and side-effects and the previous response to treatment. A consensus of international experts of the ECCO Group has been elaborated and the statements in Table 1 reproduce the main conclusions of these present treatment recommendations which should be published in 2007.

Table 1 Statement of the ECCO consensus group for the treatment of extensive mild-to-moderate UC

Extensive UC of mild–moderate severity should initially be treated with mesalazine $\geqslant 2$ g/day, combined with topical mesalazine.
Systemic corticosteroids are appropriate if symptoms of active colitis do not respond rapidly to mesalazine, or in those who are already taking appropriate maintenance therapy.
The response to intravenous steroids is best assessed objectively (by stool frequency, CRP and abdominal radiography) on or about the third day. Surgical options should be considered and discussed at this stage or earlier. Second-line therapy with either cyclosporin or infliximab or tacrolimus will often be appropriate. If there is clinical deterioration colectomy is recommended. If there is no improvement within a further 4–7 days, colectomy should usually be recommended.
Third-line therapy may be considered at a specialist centre

AMINOSALICYLATES

Oral aminosalicylates are effective in extensive UC; they have a dose–response effect and the the dose of 4 g/day is recommended[2]. Mesalazine is shown to be as effective as sulphasalazine and is better tolerated[3].

Combination with topical mesalazine is better, and this 'combined treatment' has been proposed in the ECCO consensus as the first-line strategy[4] (ECCO consensus to be published in 2007). This is based on the results of our randomized controlled trial (RCT) in which oral mesalazine (Pentasa®) 4 g/day with a 1 g mesalazine enema was shown to be more effective than oral mesalazine alone[4]. As a majority of clinical symptoms in extensive UC probably relate to the disease activity in the distal part of the colon, we speculated that combining an oral and an enema therapy would decrease the distal symptoms (bleeding, frequency of bowel movement) more effectively than oral therapy alone. This double-blind RCT was performed in 127 ambulatory patients in six European countries. All patients received 4 g/day oral mesalazine for 8 weeks. During the initial 4 weeks they additionally received an enema at bedtime containing 1 g mesalazine or placebo. Disease activity was assessed using the UCDAI with clinical and endoscopic signs at 4 and 8 weeks. Remission was obtained in 64% of the mesalazine enema group vs 43% of the placebo enema group at 8 weeks ($p = 0.03$). Improvement of the symptoms was also significantly better in this group at 4 and 8 weeks[4].

Tolerance to mesalazine (and of the oral + rectal combined treatment) is usually excellent but intolerance occurs in up to 15%. Diarrhoea (3%), headache, nausea, rash, pancreatitis, hepatitis and thrombocytopenia are reported. Acute intolerance in 3% may resemble a flare of colitis since it includes bloody diarrhoea. Renal impairment (including interstitial nephritis and nephrotic syndrome) is rare and idiosyncratic. A population-based study found the risk (OR 1.60, CI 1.14–2.26 compared to normal) to be associated with disease severity rather than the dose or type of mesalazine[5,6].

Failure to respond to mesalazine (combination strategy) is an indication to consider oral steroids.

STEROIDS

The efficacy of the treatment combining oral and rectal corticosteroids has been established in two studies. In the initial trial, by Truelove et al., oral prednisolone (40 mg daily) combined with steroid enemas induced remission within 2 weeks in 77% of 118 patients with mild to moderate UC of any extent, compared to 48% treated with 8 g/day sulphasalazine and steroid enemas[7]. In a second trial, Lennard-Jones et al.[8] reported that the combination of oral and rectal steroids was better than either alone. Steroid dependence is often observed (30%) and it is then recommended to start immunomodulators (usually thiopurines) as early as possible (as in Crohn's disease) in order to limit the adverse effects of long-term use[9].

Oral steroids with low systemic bioavailability are available or being developed, but their efficacy to properly treat the whole colon is not yet very well established (ECCO consensus to be published in 2007).

CALCINEURIN INHIBITORS

The placebo-controlled trial performed by Lichtiger et al. in 1994 identified cyclosporin A (CsA) as a rescue therapy for intravenous steroid-resistant UC[10]. Nine of 11 patients failing steroids improved on intravenous CsA whilst all nine on placebo failed to improve. First trials used a daily dose of 4 mg/kg but van Assche et al. clearly showed that 2 mg/kg was equivalent (83% and 82%, respectively)[11], and this dose is now recommended (ECCO consensus to be published in 2007). The efficacy of CsA is around 60–80% on the short-term basis[10–13]; however, this drug has many potential risks including 1–3% mortality[10–14], and the risk for colectomy in the next year because of long-term treatment failure is high. Finding predictors for efficacy or failure to these drugs may certainly help the clinician to take treatment decisions. The metabolism of CsA has a high inter-individual variability[14]. It is a substrate of P-glycoprotein (P-gp), a transporter expressed at the surface of epithelial and lymphoid cells which is encoded by the multidrug resistance gene-1 (MDR1). Single-nucleotide polymorphisms (SNP) have been found in various regions of the MDR1 gene. In a series of 154 patients treated with CsA for steroid-resistant UC (the majority had extensive UC), we observed a significant association between the SNP G2677T/A (exon 21) polymorphism distribution and the risk of resistance to CsA ($p = 0.0001$)[14].

Tacrolimus, another calcineurin inhibitor, when given at a dose that achieves a trough concentration of 10–15 ng/ml is also probably effective in patients with UC[15]. It carries many of the risks (including nephrotoxicity) of CsA.

INFLIXIMAB

Efficacy of IFX has now been clearly demonstrated. The best evidence is coming from the ACT 1 and 2 RCT, which both included 364 patients with moderately active UC refractory to oral steroids and/or thiopurines[16]. Patients

were given either placebo or IFX 5 mg/kg, or 10 mg/kg, at 0, 2 and 6 weeks, then every 8 weeks for a year. The primary endpoint, i.e. response at week 8 ($>30\%$ and a three-point decrease in the Mayo activity index), was observed in 61% and 69% of patients who received 5 mg of infliximab, vs 37 % and 29% of those who received placebo ($p<0.001$). This efficacy has been confirmed in daily practice[17], and other anti-TNF agents are currently being studied[18].

TAKING THE RIGHT DECISION BETWEEN MEDICAL OPTIONS AND SURGERY (FIGURE 1)

The first-line treatment is clearly combination treatment with oral and rectal aminosalicylates (ECCO consensus to be published in 2007). Steroid use should be limited and steroid resistance should rapidly lead to either colectomy or the prescription of CsA or IFX. Colectomy must always be considered in patients with severe UC but attempts to save the colon in patients with mild-to-moderate disease are logical. A recommendation on the best choice between calcineurin inhibitor and IFX is not possible until there has been a comparative RCT. This trial has just begun (CYSIF trial by the GETAID). Validation of the value of the MDR1 polymorphism to predict CsA resistance would be helpful[14].

Before prescribing CsA or IFX for a patient with steroid-resistant extensive UC, other causes of persistent symptoms, including coexistent cytomegalovirus or cancer, should be considered. Sepsis and other contraindications to

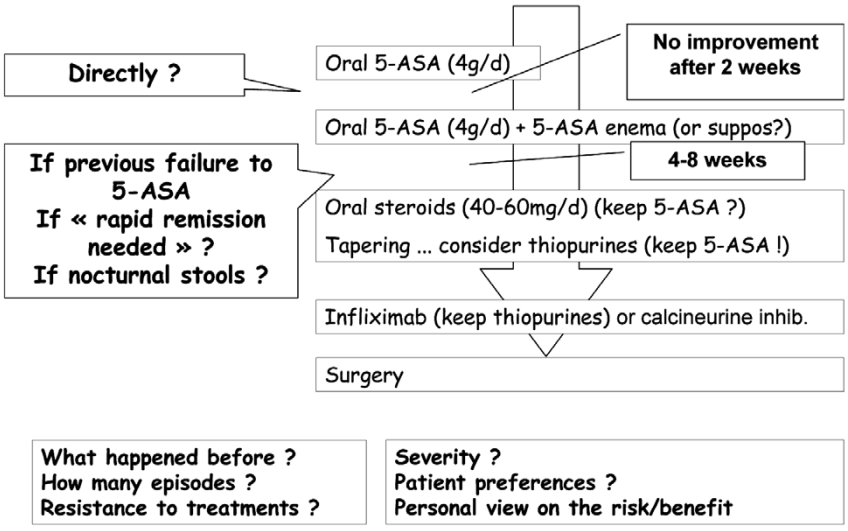

Figure 1 Taking the right decision between medical options and surgery

immunomodulators should be excluded. At the present time it seems that, for patients with UC who are not in a very severe situation but require long-term treatment, IFX would offer a higher chance of success than CsA (as IFX would be administered on a long-term basis).

References

1. D'Haens G, Sandborn WJ, Feagan BG et al. Clinical trials of medical therapy in adults with ulcerative colitis. Gastroenterology. 2007;132:763–86.
2. Kane SV, Bjorkman DJ. The efficacy of oral 5-ASAs in the treatment of active ulcerative colitis: a systematic review. Rev Gastroenterol Disord. 2003;3:210–18
3. Sutherland L, MacDonald JK. Oral 5-aminosalicylic acid for induction of remission in ulcerative colitis. Cochrane Database Syst Rev 2006; 2:CD000543.
4. Marteau P, Probert CS, Lindgren S et al. Combined oral and enema treatment with Pentasa (mesalazine) is superior to oral therapy alone in patients with extensive mild/moderate active ulcerative colitis: a randomised, double blind, placebo controlled study. Gut. 2005; 54:960–5.
5. Loftus EV Jr, Kane SV, Bjorkman D. Systematic review: short-term adverse effects of 5-aminosalicylic acid agents in the treatment of ulcerative colitis. Aliment Pharmacol Ther. 2004;19:179–89.
6. Van Staa TP, Travis S, Leufkens HG, Logan RF. 5-Aminosalicylic acids and the risk of renal disease: a large British epidemiologic study. Gastroenterology. 2004;126:1733–9.
7. Truelove SC, Watkinson G, Draper G. Comparison of corticosteroid and SASP therapy in ulcerative colitis. Br Med J. 1962;2:1708–11.
8. Lennard-Jones JE, Longmore AJ, Newell AC, Wilson CWE, Avery Jones F. An assessment of prednisone, Salazopyrin and topical hydrocortisone hemisuccinate used as outpatient treatment for ulcerative colitis. Gut. 1960;1:217–22.
9. Travis SP, Stange EF, Lémann M et al. European evidence based consensus on the diagnosis and management of Crohn's disease: current management. Gut. 2006;55(Suppl. 1):i16–35.
10. Lichtiger S, Present DH, Kornbluth A, Gelernt I. Cyclosporine in severe ulcerative colitis refractory to steroid therapy. N Engl J Med. 1994;330:1841–5.
11. Van Assche G, D'Haens G, Noman M et al. Randomized, double-blind comparison of 4 mg/kg versus 2 mg/kg intravenous cyclosporine in severe ulcerative colitis. Gastroenterology. 2003;125:1025–31.
12. Cohen RD, Stein R, Hanauer SB. Intravenous cyclosporine in ulcerative colitis: a five year experience. Am J Gastroenterol. 1999;94:1587–92.
13. Campbell S, Travis SPL, Jewell DP. Ciclosporin use in acute ulcerative colitis: a long-term experience. Eur J Gastroenterol Hepatol. 2005;17:79–84.
14. Daniel F, Loriot MA, Seksik P et al. Multidrug resistance gene-1 polymorphisms and resistance to cyclosporine A in patients with steroid resistant ulcerative colitis. Inflamm Bowel Dis. 2007;13:19–23.
15. Ogata H, Matsui T, Nakamura M et al. A randomised dose finding study of oral tacrolimus (FK506) therapy in refractory ulcerative colitis. Gut. 2006;55:1255–62.
16. Rutgeerts P, Sandborn WJ, Feagan B et al. Infliximab for induction and maintenance therapy for ulcerative colitis. N Engl J Med. 2005;233:2462–73.
17. Jakobovits SL, Jewell DP, Travis SP. Infliximab for the treatment of ulcerative colitis: outcomes in Oxford from 2000 to 2006. Aliment Pharmacol Ther. 2007;25:1055–60.
18. Peyrin-Biroulet L, Laclotte C, Roblin X, Bigard MA. Adalimumab induction therapy for ulcerative colitis with intolerance or lost response to infliximab: an open-label study. World J Gastroenterol. 2007;13:2328–32.

21
Mild to moderately active ileocaecal Crohn's disease

V. GROSS

INTRODUCTION

Various guidelines of different countries, e.g. those from the UK[1], Germany[2], France or from the ECCO[3], exist for the management of Crohn's disease (CD). The recent European evidence-based consensus on the diagnosis and management of CD[3] defines mildly active CD by a CDAI of 150–220 points and moderately active CD by a CDAI of 220–450 points. For the treatment of mildly active ileocaecal CD the consensus recommended budesonide 9 mg/day as the preferred treatment. The consensus stated that the benefit of mesalazine is limited. Antibiotics are not recommended. No treatment is an option for some patients with mild symptoms. Moderately active ileocaecal CD should preferably be treated with budesonide 9 mg/day or with systemic corticosteroids. Antibiotics should be added if septic complications are suspected.

WHAT IS THE EVIDENCE FOR THESE RECOMMENDATIONS?

Budesonide in active CD

A number of randomized controlled trials used oral budesonide in patients with active CD. Budesonide is a topically active glucocorticoid with a high glucocorticoid receptor affinity and anti-inflammatory potency. Due to its rapid first-pass metabolism and inactivation in the liver, the systemic availability of budesonide is less than 10% after oral dosing. For the treatment of CD budesonide has to be administered as an oral retarded preparation in order to avoid its absorption in the upper gastrointestinal tract. Two types of oral budesonide preparations are currently available: a pH-modified release formulation and a controlled ileal-release formulation. These budesonide preparations have been evaluated for the treatment of patients with active CD.

Two trials[4,5] compared budesonide to placebo for the treatment of active CD. Five trials[6–10] compared budesonide to conventional systemic glucocorticoids;

one trial[11,12] compared budesonide to mesalazine; one trial[13] combined budesonide and antibiotic therapy; and one trial[14] studied different budesonide doses. A meta-analysis assessing the effectiveness and safety of oral budesonide in comparison to placebo, oral systemic glucocorticoids, and mesalazine has been published[15]. The two studies comparing oral budesonide to placebo found that budesonide induces remission of CD more frequently than placebo (RR = 1.85, 95% CI 1.31–2.61). In the study of Greenberg et al.[4] oral doses of 3 mg, 9 mg, and 15 mg of budesonide were compared to placebo in 228 patients with active CD. After 8 weeks of treatment, remission was obtained in 51% of the patients in the 9 mg budesonide group, in 43% of the patients in the 15 mg group, and in 33% of the patients in the 3 mg group. The remission rate in the placebo group was 20%. Budesonide caused a dose-related reduction in basal and corticotropin-stimulated plasma cortisol concentrations, but was not associated with clinically important steroid-related symptoms or other toxic effects. The authors concluded from their study that budesonide at on optimal daily dose of 9 mg is well tolerated and effective against active CD in the ileum and proximal colon.

Tremaine et al.[5] treated 200 patients with mild to moderate CD either with 9 mg of budesonide once daily, 4.5 mg twice daily, or placebo for 8 weeks. Remission was achieved in 48%, 53%, and 33% with 9 mg budesonide once daily, 4.5 mg twice daily, or placebo, respectively. Differences between the groups were not statistically significant.

In both studies[4,5] total corticosteroid-associated adverse events were not significantly different for patients receiving budesonide compared with patients receiving placebo (50% versus 40%, RR = 1.06, 95% CI 0.92–1.23) (Table 1).

In none of the five trials comparing budesonide to conventional glucocorticoids[6–10] was there a statistically significant difference between the remission rates induced by budesonide or conventional glucocorticoids. However, the meta-analysis[15] showed that conventional glucocorticoids induced remission of CD more frequently than budesonide (RR = 0.87, 95% CI 0.76–0.995). In a subgroup analysis of patients with low disease activity, however, budesonide and conventional glucocorticoids induced remission at similar rates (RR = 0.91, 95% CI 0.77–1.07).

Table 1 Summary of trials comparing budesonide with placebo for the induction of remission in active ileocaecal Crohn's disease[4,5] (remission rates/side-effects)*

		Budesonide		
Study	*Placebo*	*3 mg*	*9 mg*	*15 mg*
Greenberg et al. (1994)[4]	20%/26%	33%/15%	51%/26%	43%/38%
Tremaine et al. (2002)[5]	33%		48%*; 53%**	

RR = 1.85 (95% CI 1.31–2.61).

*9 mg budesonide once daily, **4.5 mg budesonide twice daily.

Table 2 Meta-analysis of trials comparing budesonide with conventional sytemic glucocorticoids for the induction of remission in active Crohn's disease[15]

	RR	*95% CI*
Remission rate all patients	0.87	0.76–0.995
Remission rate low disease activity	0.91	0.77–1.07
Steroid side-effects	0.65	0.53–0.80

The meta-analysis of the studies comparing budesonide to conventional glucocorticoids showed that total glucocorticoid-associated adverse events were significantly less frequent for patients who received budesonide than for patients who received conventional glucocorticoids (RR = 0.65, 95% CI 0.533–0.80)[14]. This means a relative risk reduction of 35% (Table 2).

Rutgeerts et al.[6] compared budesonide with prednisolone in 176 patients with active ileal or ileocaecal CD (88 patients in each group). Budesonide was administered at a dose of 9 mg/day for 8 weeks and then at a dose of 6 mg/day for 2 weeks. Prednisolone was administered at a dose of 40 mg/day for 2 weeks and thereafter was gradually reduced to 5 mg/day. At 10 weeks 53% of the patients in the budesonide group were in remission as compared with 66% of those treated with prednisolone. The mean Crohn's Disease Activity Index (CDAI) decreased from 275 to 175 in the budesonide group and from 279 to 136 in the prednisolone group. Glucocorticoid-associated side-effects were significantly less in the budesonide group (29 versus 48 patients, $p = 0.003$).

Gross et al.[7] compared 3×3 mg budesonide per day with 6-methylprednisolone in patients with active ileal or ileocolonic CD. Budesonide induced remission in 55.9% of the patients, 6-methylprednisolone in 72.7%. This difference was statistically not significant. There was a similar CDAI decrease in both groups. Patients in the budesonide group suffered significantly less frequently from steroid-related side-effects than patients in the 6-methylprednisolone group (28.6% versus 69.7%, $p = 0.0015$).

Campieri et al.[8] compared two regimens of budesonide (1×9 mg and 2×4.5 mg/day) with prednisolone (40 mg starting dose with conventional tapering): 9 mg of budesonide was equally effective as prednisolone, with 60% remission at 8 weeks. The divided dose was somewhat less effective (42%). The suppression of plasma cortisol was much more pronounced in the prednisolone group and was more often severe in the once-daily budesonide group as compared to the divided-dose group.

Bar-Meir et al.[9] compared 3×3 mg budesonide per day with prednisone (starting dose 40 mg/day) in 201 patients with ileocolonic CD. Remission rates after 8 weeks were comparably high in the budesonide group (51%) and in the prednisone group (52.5%). However, in the budesonide group twice as many patients reached remission without steroid side-effects (30%) than in the prednisone group (14%).

The study comparing budesonide to mesalazine[11] showed that budesonide induced remission of CD more frequently than mesalazine (62% versus 36%), respectively. This produced a relative risk of 1.73 (95% CI 1.26–2.39). Further

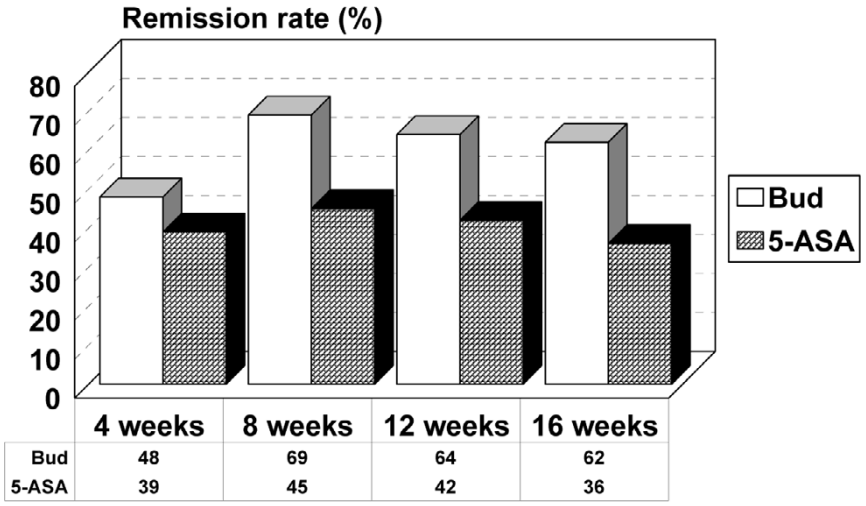

Figure 1 Trial comparing budesonide with mesalazine for the induction of remission in active ileocaecal Crohn's disease[11] (RR = 1.73; 95% CI 1.26–2.39)

analysis of this study showed that budesonide improved health-related quality of life to a greater extent than mesalazine in patients with mild to moderate CD[12] (Figure 1).

In a recent double-blind multicentre study of patients with active CD of the ileum, right colon, or both, patients were randomized to receive 9 mg oral budesonide once daily and in addition oral ciprofloxacin and metronidazole, both 500 mg twice daily, or placebo for 8 weeks[13]. Sixty-six patients received placebo, 64 received antibiotics. At week 8, 21 patients (33%) of the antibiotic group achieved remission as compared with 25 patients (38%) of the placebo group (n.s.). Among patients with involvement of the right colon 9/17 (52%) were in remission on the treatment with antibiotics compared with 4/16 (25%) of those who received placebo ($p = 0.15$). Thus, the addition of ciprofloxacin and metronidazole to budesonide was ineffective in patients with active CD, but there were some indications that the antibiotic combination may improve the outcome when the colon is involved.

In a further trial[14] oral budesonide was tested in a randomized double-blind dose-finding study. Patients with active Crohn's ileocolitis without steroid pretreatment received either 3×2 mg, 3×3 mg or 3×6 mg budesonide. Remission rates after 6 weeks were 36% (3×2 mg budesonide), 55% (3×3 mg budesonide), and 66% (3×6 mg budesonide), respectively. Only the remission rates of 3×6 mg versus 3×2 mg were significantly different ($p = 0.017$). Patients with high disease activity (CDAI $\geqslant 300$) responded better to the highest budesonide dose (remission rates 0%, 25%, and 75% for 3×2 mg, 3×3 mg, and 3×6 mg budesonide). Steroid-typical side-effects were observed

Table 3 Dose-escalation study with oral pH-modified release budesonide in patients with active ileocolonic Crohn's disease[15] (percentages)

	Budesonide		
Remission rate	*3 × 2 mg (n = 39)*	*3 × 3 mg (n = 33)*	*3 × 6 mg (n = 32)*
Total	36	55	66
CDAI <300	47	71	64
CDAI $\geqslant 300$	0	25	75
Ileum/ascending colon	30	60	59
Ileum/left colon	31	46	73

*$p<0.05$ 3×6 mg vs 3.2 mg.

in seven patients (3×2 mg), one patient (3×3 mg), and eight patients (3×6 mg) after 6 weeks of treatment. This study demonstrates that oral pH-modified release budesonide shows a dose-dependent effectiveness in patients with active ileocolonic CD. In the majority of patients 9 mg budesonide per day is sufficient; in patients with highly active disease higher doses can increase the therapeutic response (Table 3).

MESALAZINE

The utility of mesalazine in the treatment of mild to moderately active ileocaecal CD is controversial. There are few randomized controlled trials which used mesalazine for the induction of remission of CD. Danish investigators reported that Pentasa (1.5 g/day) was not better than placebo, but suggested that trials using larger doses should be performed[16].

Singleton et al.[17] performed a dose-ranking study with Pentasa in patients with active CD. While remission rates of 18%, 23%, and 24% were obtained with placebo, 1 g 5-aminosalicylic acid (5-ASA)/day or 2 g 5-ASA/day, respectively, 4 g 5-ASA/day induced remission in 43% of patients ($p<0.005$)[17]. However, a combined analysis of three studies using Pentasa in active CD, including the above-mentioned study[18], showed a significant but only modest benefit from the Pentasa treatment. While in the ITT analysis group ($n = 615$) the mean CDAI decrease in Pentasa-treated patients was 63 ± 6 points, it was 45 ± 6 points in placebo-treated patients (difference 18 ± 9 points, $p = 0.004$). The per-protocol analysis showed a slightly higher difference in the CDAI decrease (25 ± 11 points, $p = 0.002$); the clinical benefit of this effect is, however, questionable.

There are two studies[19,20] which compared Salofalk with conventional glucocorticoids in patients with active CD. Martin et al.[19] compared Salofalk 3 g/day with prednisone in 44 patients with CD. Although prednisone treated patients had a prompter decline in the CDAI, by the end of the study both groups had an equivalent response. Gross et al.[20] compared Salofalk 4.5 g/day with 6-methylprednisolone (48 mg/day starting dose) in patients with active

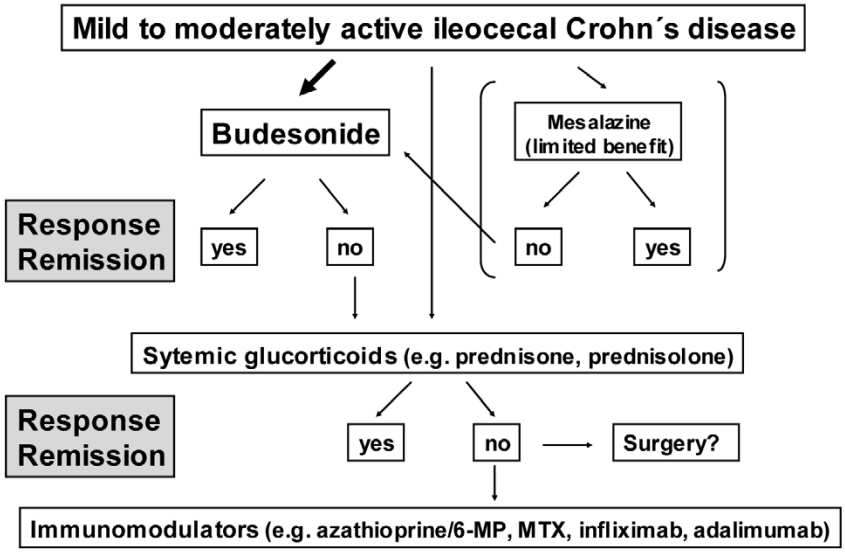

Figure 2 Treatment algorithm for patients with mild to moderately active ileocaecal Crohn's disease

Crohn's ileocolitis. Duration of treatment was 8 weeks. The mean CDAI decrease in the mesalazine group was 85 points, in the 6-methylprednisolone group it was 122 points. Remission rates (intention-to-treat) after 8 weeks were 40.0% in the mesalazine group and 56.3% in the 6-methylprednisolone group. The size of the study, however, was too small to calculate a statistically significant difference between the two treatment groups.

The most favourite data for mesalazine were published by Prantera et al.[21]. They performed a randomized study including patients with mild to moderately active Crohn's ileitis (CDAI 180–360). The patients were treated for 12 weeks with either mesalazine tablets (4 g/day), mesalazine microgranules (4 g/day) or 6-methylprednisolone (40 mg/day starting dose). The median CDAI decrease was 113.5 points in the mesalazine tablet group, 123 points in the mesalazine microgranule group and 154 points in the 6-methylprednisolone group. The corresponding remission rates were 61% for mesalazine tablets, 79% for mesalazine microgranules, and 60% for 6-methylprednisolone.

CONCLUSIONS

For the treatment of patients with mild to moderately active ileocaecal CD budesonide 9 mg/day is the preferred treatment. Budesonide is superior to placebo (RR 1.85, 95% CI 1.31–2.61) and superior to mesalazine 4 g/day (RR 1.73, 95% CI 1.26–2.39). Budesonide achieves remission in 51–60% over 8–10

weeks. Budesonide is preferred to prednisolone for patients with mild to moderately ileocaecal CD, since it is associated with fewer glucocorticoid side-effects (RR 0.65, 95% CI 0.53–0.80), although a systematic review has shown that budesonide is somewhat less effective than conventional glucocorticoids (RR 0.87, 95% CI 0.76–0.995). In patients with low disease activity, however, budesonide was not significantly less effective than conventional glucocorticoids (RR 0.91, 95% CI 0.77–1.07). If budesonide fails, oral systemic glucocorticoids, e.g. prednisone or prednisolone, are indicated. If systemic glucocorticoids fail, which is rarely the case in mild to moderately active ileocaecal CD, immunomodulators or surgery are indicated.

References

1. Carter MJ, Lobo AJ, Travis SP et al. Guidelines for the management of inflammatory bowel disease in adults. Gut. 2004;53(Suppl. 5):V1–16.
2. Stange EF, Schreiber S, Fölsch UR et al. Diagnostik und Therapie des M. Crohn – Ergebnisse einer evidenzbasierten Konsensuskonferenz der Deutschen Gesellschaft für Verdauungs- und Stoffwechselkrankheiten. Z Gastroenterol. 2003; 41:19–48.
3. Stange EF, Travis SPL, Vermeire S et al. for the European Crohn's and Colitis Organisation (ECCO). European evidence based consensus on the diagnosis and management of Crohn's disease: definitions and diagnosis. Gut. 2006;55(Suppl. I): I1–15.
4. Greenberg GR, Feagan BG, Martin F et al. and the Canadian Inflammatory Bowel Disease Study Group. Oral budesonide for active Crohn's disease. N Engl J Med. 1994;331:836–841.
5. Tremaine WJ, Hanauer SB, Katz S et al. and the Budesonide CIR United States Study Group. Budesonide CIR capsules (once or twice daily divided-dose) in active Crohn's disease: a randomized placebo-controlled study in the United States. Am J Gastroenterol. 2002;97:1748–54.
6. Rutgeerts P, Löfberg R, Malchow H et al. A comparison of budesonide with prednisolone for active Crohn's dsiease. N Engl J Med. 1994;331:842–5.
7. Gross A, Andus T, Caesar I et al. and the German/Austrian Budesonide Study Group. Oral pH-modified release budesonide versus 6-methylprednisolone in active Crohn's disease. Eur J Gastroenterol Hepatol. 1996; 8: 905–9.
8. Campieri M, Ferguson A, Doe W et al. and the Global Budesonide Study Group. Oral budesonide is as effective as oral prednisolone in active Crohn's disease. Gut. 1997;41:209–14.
9. Bar-Meir S, Chowers Y, Lavy A et al. and the Israeli Budesonide Study Group. Budesonide versus prednisolone in the treatment of active Crohn's disease. Gastroenterology. 1998;115:835–40.
10. D'Haens G, Verstraete A, Cheyns K et al. Bone turnover during short-term therapy with methylprednisolone or budesonide in Crohn's disease. Aliment Pharmacol Ther. 1998;12:419–24.
11. Thomson OØ, Cortot A, Jewell D et al. and the International Budesonide–Mesalamine Study Group. A comparison of budesonide and mesalamine for active Crohn's disease. N Engl J Med. 1998;339:370–4.
12. Thomson OØ, Cortot A, Jewell D et al. for the International Budesonide–Mesalamine Study Group. Budesonide and mesalazine in active Crohn's dsiease: a comparison of the effects on quality of life. Am J Gastroenterol. 2002;97:649–53.
13. Steinhart AH, Feagan BG, Wong CJ et al. for the Crohn's and Colitis Foundation of Canada Inflammatory Bowel Disease Network Investigators. Combined budesonide and antibiotic therapy for active Crohn's disease: a randomized controlled trial. Gastroenterology. 2002;123:33–40.
14. Herfarth H, Gross V, Andus T et al. and the German/Austrian Budesonide Study Group. Analysis of the therapeutic efficacy of different doses of budesonide in patients with active Crohn's ileocolitis depending on disease activity and localization. Int J Colorectal Dis. 2004;19:147–52.

15. Kane SV, Schoenfeld P, Sandborn WJ, Tremaine W, Hofer T, Feagan BG. Systematic review: the effectiveness of budesonide therapy for Crohn's disease. Aliment Pharmacol Ther. 2002;16:1509–17.
16. Rasmussen SN, Lauritsen K, Tage-Jensen U et al. 5-Aminosalicylic acid in the treatment of Crohn's disease. A 16 week double-blind, placebo-controlled, multicenter study with Pentasa. Scand J Gastroenterol. 1987;22:877–83.
17. Singleton JW, Hanauer SB, Gitnick GL et al., Pentasa Crohn's disease Study Group. Mesalamine capsules for the treatment of active Crohn's disease: results of a 16-week trial. Gastroenterology. 1993;104:1293–301.
18. Hanauer SB, Strömberg U. Oral Pentasa in the treatment of active Crohn's disease: a meta-analysis of double-blind, placebo-controlled trials. Clin Gastroenterol Hepatol. 2004;2:379–88.
19. Martin F, Sutherland LR, Beck IT et al. Oral 5-ASA versus prednisone in short term treatment of Crohn's disease: a multicenter controlled trial. Can J Gastroenterol. 1990;4:452–7.
20. Gross V, Andus T, Fischbach W et al. and the German 5-ASA Study Group. Comparison between high dose 5-aminosalicylic acid and 6-methylprednisolone in active Crohn's ileocolitis. A multicenter randomized double-blind study. German 5-ASA Study Group. Z Gastroenterol. 1995;33:581–4.
21. Prantera C, Cottone M, Pallone F et al. and a Cooperative Study Group. Mesalamine in the treatment of mild to moderate active Crohn's ileitis: results of a randomized, multicenter trial. Gastroenterology. 1999;116:521–6.

22
Therapy of mild to moderate colonic Crohn's disease

H. A. AKPINAR

INTRODUCTION

Crohn's disease (CD) is a chronic inflammatory bowel disease characterized by a relapsing inflammatory process. It can affect any part of the gastrointestinal (GI) tract and is associated with transmural inflammation of the gut wall[1].

Management targets of CD are to induce and maintain remission and to keep or improve quality of life of patients. The treatment plan for CD depends on the disease behaviour pattern, severity and location, and should be individualized according to the patient's response and tolerability to medical intervention. The main factors influencing therapeutic decisions in CD are shown in Table 1[2].

The activity of CD can be assessed clinically, endoscopically or by some indices. According to the European Crohn's and Colitis Organization (ECCO), Crohn's Disease Activity Index (CDAI) values between 150 and 220 are accepted as 'mild disease'. These ambulatory patients are able to eat and drink without manifestations of obstruction, fever, dehydration, abdominal mass or tenderness and $>10\%$ weight loss. C-reactive protein (CRP) levels are usually

Table 1 Main factors influencing therapeutic decisions in Crohn's disease[2]

Pattern of disease behaviour Penetrating, stenosing, inflammatory
Disease activity Mild, moderate, severe
Disease location Ileal, ileocolonic, colonic
Patient's age
Lack of response to other drugs in the same flare-up
Drug intolerance and/or contraindication

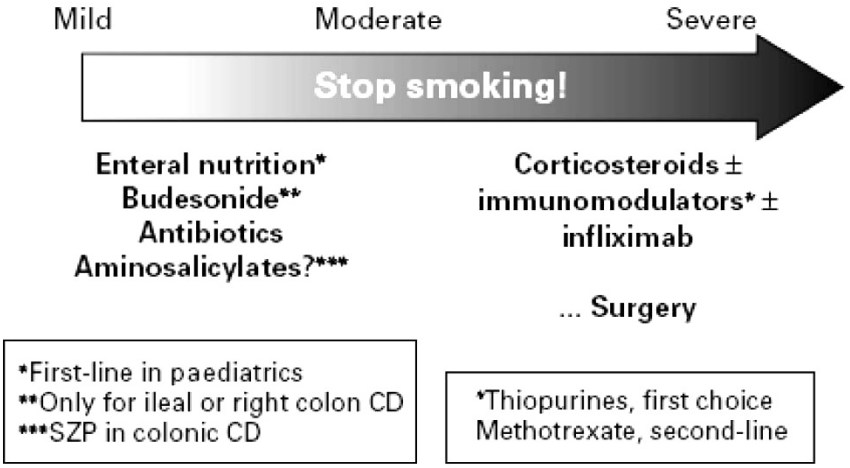

Figure 1 Medical therapeutic approach in 'inflammatory' active Crohn's disease[2] (CD: Crohn's disease, SZP: salazopyrine)

above the upper limit of normal. CDAI values between 220 and 450 are accepted as 'moderate' disease. These patients have failed to respond to treatment for mild disease and have prominent symptoms such as intermittent vomiting (without obstruction), $>10\%$ weight loss and tender mass. CRP levels are raised above the upper limit of normal[3].

There are many drugs which can be successfully used for inducing remission. The patients should be educated about the side-effects of drugs used and the importance of adherence to treatment[4]. There is only one therapeutic measure that should be advised in all CD patients: quit smoking. Treatments currently used for mild to moderate colonic CD are aminosalicylates, antibiotics and budesonide, as shown in Figure 1[2]. Treatments for moderate to severe CD are corticosteroids, infliximab and immunomodulators[2,5].

ENTERAL NUTRITION

Enteral nutrition, given orally or via tube feeding, can be an alternative treatment. Because of the absence of important side-effects and the presence of beneficial effects on nutritional status, the enteral nutrition can be preferred as an alternative treatment, especially in paediatric patients and adult patients with malnutrition[6].

According to ECCO consensus comment, nutritional therapy may be less effective in colonic than in small bowel disease in adults, but a meta-analysis was unable to confirm this, because numbers from controlled trials are too few[4]. After the ECCO consensus on CD, a meta-analysis which was done by Zachos et al., evaluating the effectiveness of enteral nutrition as a primary therapy to induce remission in CD and to examine the importance of formula

composition on effectiveness, was published in 2007[7]. It was concluded that corticosteroids are more effective than enteral nutrition for treating active CD. There was no difference in the effectiveness for treating active CD when one form of enteral nutrition was compared with another form, and there was a trend favouring low-fat formulations[7].

AMINOSALICYLATES

Aminosalicylates have been used for over three decades to treat mild to moderate CD[8]. Sulphasalazine (SASP) and 5-aminosalicylic acid (mesalazine or mesalamine) are the commonly used aminosalicylates. SASP contains a sulphonamide antibiotic (sulphapyridine) linked by an azo band to an anti-inflammatory salicylate (mesalamine)[4]; it requires colonic bacteria to release the active moiety of the drug. Since sulphapyridine is responsible for most of the side-effects, mesalamine formulations were developed.

SASP and mesalamine inhibit tumour necrosis factor alpha and interleukin 1, arachidonate metabolites, transcription factor NF-κB and leukocyte chemotaxis, and induce apoptosis, activate PPAR-γ and scavenge free reactive oxygen metabolites (Figure 2)[9].

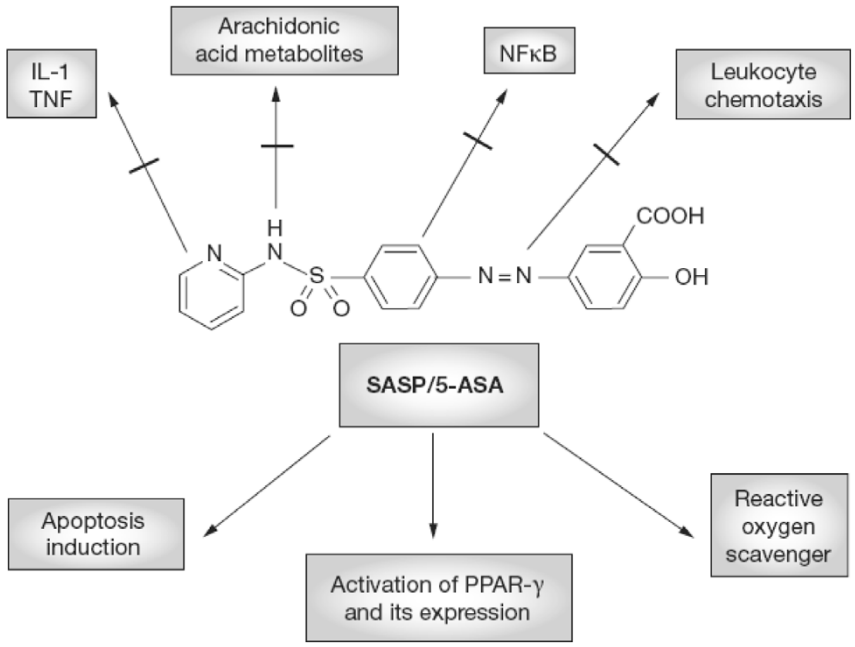

Figure 2 Inflammatory mediators and other factors in the diseased intestine that are affected by SASP or mesalamine[9]

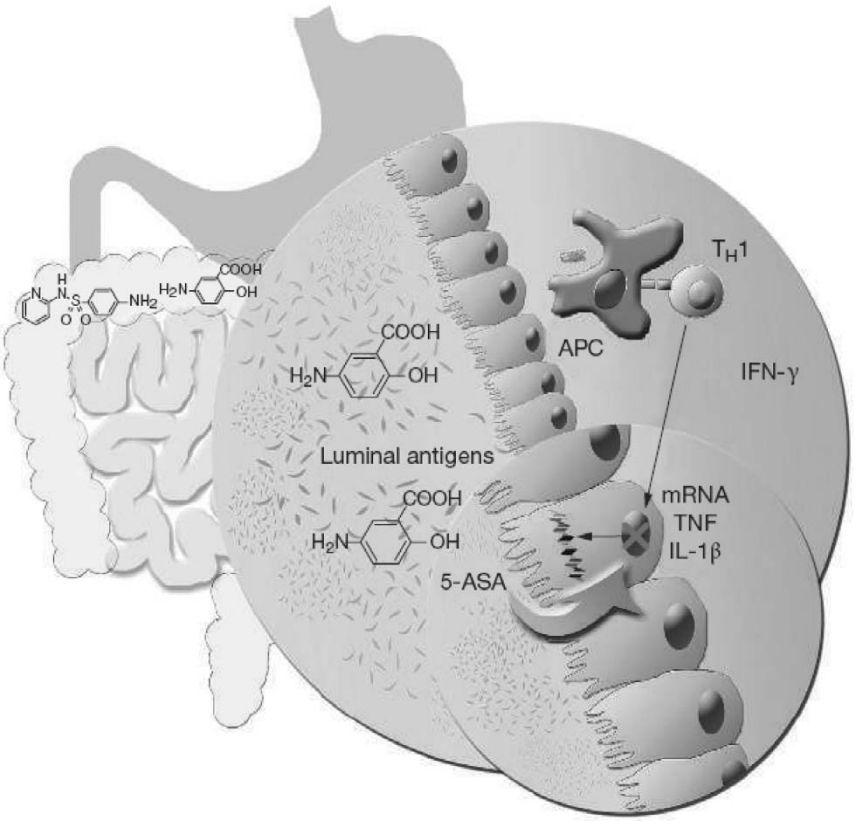

Figure 3 The mechanism of action of mesalamine in the colon[9]. (APC: antigen-presenting cell, T_H1: T helper 1 cell, IFN-γ: interferon gamma: TNF: tumour necrosis factor, IL-1β: interleukin 1β

The mechanism of action of mesalamine in the colon can be seen in Figure 3. Antigen-presenting cells are able to recognize luminal antigens penetrating the colonic wall and, through interaction with Th1 cells, induce the production of interferon gamma (IFN-γ). This activates epithelial cells. Mesalamine is able to block transcription of inflammatory cytokines in intestinal epithelial cells[9].

Sulphasalazine

Two large studies and several smaller ones, have shown that SASP is modestly effective in inducing remission in CD, especially in colonic CD[10]. The efficacy of SASP has been compared to placebo or to corticosteroids in several controlled trials. Before the development of CDAI, Anthonisen et al. randomized 31 patients to a 4-month crossover study with SASP 3 g/day or placebo. SASP was superior to placebo in improving the overall clinical

condition in 17 patients[11]. In another small study, 27 patients with active small bowel and/or colonic CD were randomly allocated to receive SASP 6 g/day or placebo for 26 weeks. A decrease in CDAI $\geqslant$25% was achieved in 61.5% of patients randomized to SASP compared with only 7.7% of patients in the placebo group[12]. The American National Cooperative Crohn's Disease multicentre study randomly allocated 295 active patients (CDAI $\sim$250) to receive placebo (n = 77), prednisone 0.25–0.75 mg/kg (n = 85), azathioprine 2.5 mg/kg (n = 59), or SASP (1 g/15 kg of body weight) (n = 74). Remission (CDAI $\leqslant$150 points) was obtained and maintained by 26% of patients on placebo (20/77) compared with 38% on SASP (28/74) for the 17-week follow-up period, a statistically significant difference[13]. Patients with involvement limited to the colon responded to SASP better than to placebo[14]. The European Cooperative Crohn's Disease Study also evaluated the effectiveness of SASP and 6-methylprednisolone, alone or in combination, compared with placebo in 455 patients with CD. Two hundred and fifteen patients had active disease at entry and were treated with SASP (3 g/day) (n = 54), 6-methylprednisolone (48 mg/day weekly, tapered to 12 mg/day weekly) (n = 47), combination therapy (n = 56) and placebo (n = 58). The proportion of patients achieving remission (CDAI $<$150) before the end of 18 weeks was 50% with SASP compared with 37.9% in the placebo group[15]. The lower efficacy of SASP observed in this study is probably secondary to a lower dose of SASP compared with the American National Cooperative Crohn's Disease Study, where the mean dose was 4 g/day[16].

Mesalamine

Mesalamine has been accepted as first-line treatment of choice for mild to moderately active CD by the majority of gastroenterologists. Both Rasmussen et al. and Mahida and Jewell found no difference in remission rates at 16 and 6 weeks respectively using the relatively low-dose mesalamine (1.5 g/day) vs placebo[17,18]. Tremaine et al. randomized 38 patients to either placebo or Asacol 3.2 g/day. There was a statistically significant difference at the combination of partial remission (fall in CDAI more than 70 points) and clinical remission[19].

The series of studies of mesalamine in mild to moderately active CD were those carried out by Singleton et al. In the initial study 310 CD patients were randomized to placebo, 1 g, 2 g or 4 g Pentasa daily. A significant difference after 8 weeks was observed only in the 4 g/day group. Quality of life also improved in the 4 g/day group[19]. Although this initial result was promising, the same group undertook a second trial with identical entry criteria randomizing to placebo 2 g/day and 4 g/day mesalamine; this study has been reported only as a short letter, but this time the findings were negative for a benefit of the 4 g/day dose over placebo. A third study focusing on the 4 g dose alone was undertaken, but has never been reported. The meta-analysis of these three studies showed a mean reduction in CDAI, 63 points in 304 subjects who received Pentasa 4 g/day vs 45 points for the 311 who received placebo. Although this 18-point difference in CDAI achieved statistical significance, it is not clinically significant[20].

According to ECCO consensus on CD, active colonic CD may be treated with sulphasalazine if only mildly active by the evidence level (EL) 1b, and the recommendation grade (RG) A (statement 5D)[4]. According to ECCO consensus comment, SASP 4 g daily is effective for active colonic disease, but cannot be recommended as first-line therapy in view of a high incidence of side-effects. It may be appropriate in selected patients, such as those with an associated arthropathy. Opinion varies about the value of topical 5-ASA as adjunctive therapy in left-sided colonic disease. There has been no controlled trial of topical therapy of CD, so there is no evidence base. Distal colonic CD presents an occasional therapeutic dilemma. The consensus believes it should be considered in these circumstances, but a similar proportion advise or recommend it as do not use it[4].

ANTIBIOTICS

Many experimental and clinical observations suggest that intestinal microflora plays a potential role in the pathogenesis of inflammatory bowel disease (IBD). Manipulation of luminal contents using antibiotics is a potentially effective therapeutic option. Although antibiotics are used in the septic complications of CD, their use as a primary therapy in CD is more controversial[21]. Metronidazole and ciprofloxacin are the most commonly used antibiotics in the treatment of CD. Suggested mechanisms of action of antibiotics in IBD can be seen in Table 2[21,22].

Metronidazole

Blichfeldt et al. examined the effectiveness of metronidazole 1 g/day for the duration of 16 weeks as an adjunctive therapy to SASP in CD. Although there was no overall benefit with metronidazole, significant improvement of symptoms and laboratory parameters was found in six of the 22 patients with isolated colonic CD[23]. In another smaller placebo-controlled study, metronidazole showed no benefit over placebo[24]. In a Swedish Cooperative Crohn's Disease Study the efficacy of metronidazole 800 mg daily compared with SASP was evaluated in a randomized, double-blind 4-month crossover study. CDAI and plasma orosomucoid levels were the evaluation criteria. Those patients who responded during the first study period remained stable during the second study period regardless of the treatment. However, plasma orosomucoid levels increased significantly in the SASP-treated group. In the patients with active disease during the first phase, a significant fall in CDAI

Table 2 Antibiotics in IBD: suggested mechanisms of action[21]

Eradication of bacterial antigenic trigger
Elimination of bacterial overgrowth
Reduction of proinflammatory bacterial toxins
Potential immunosuppressive properties

was observed in those patients who were switched to the metronidazole group; not for those who were switched to SASP treatment[25]. Sutherland and co-workers performed a double-blind study which compared the efficacy of metronidazole in two doses (20 mg/kg and 10 mg/kg) with placebo in patients with CD. One hundred and five patients participated but only 56 completed the study period of 16 weeks. Although patients receiving metronidazole 20 mg/kg per day had a greater improvement in disease activity than those receiving 10 mg/kg per day, the difference did not reach statistical significance, probably due to the small sample size. Preliminary analysis suggested that metronidazole was more effective in patients with disease confined to the large intestine or affecting both small and large bowel than in those with small bowel disease only[26].

Ciprofloxacin

Prantera et al. conducted a study which aimed to investigate the efficacy and safety of a combination of metronidazole and ciprofloxacin, compared with methylprednisolone, in treating 41 consecutive patients with active CD. Thirty-one patients were randomly allocated to receive, for 12 weeks, ciprofloxacin 500 mg twice daily plus metronidazole 250 mg four times daily or methylprednisolone 0.7–1 mg/kg per day, followed by tapering of 4 mg weekly. Ten of the 22 antibiotic patients (45.5%) and 12 of the 19 steroid patients (63%) obtained clinical remission (CDAI $\leqslant$150) at the end of the 12-week study (p = n.s.)[27]. Colombel et al. performed a 6-week study which comprised 40 patients and compared the efficacy of ciprofloxacin 1 g/day to mesalamine 4 g/day. They did not observe any difference between treatment groups (56% remission with ciprofloxacin vs 55% remission with mesalamine)[28].

Greenbloom et al. examined the efficacy and safety of combination ciprofloxacin and metronidazole for patients with active CD of the ileum and/or colon. Seventy-two patients with active CD of the ileum (n = 27), ileocolon (n = 22) or colon (n = 23) were treated with ciprofloxacin 500 mg b.i.d. and metronidazole 250 mg t.i.d. for a mean of 10 weeks. Clinical remission was observed in 49 patients (68%), and 55 patients (76%) showed a clinical response. A clinical response also occurred in a greater proportion of patients with colonic disease, with or without ileal involvement (84%), compared with patients with ileal disease alone (64%), and in patients without resection (86%) compared with those with previous resection (61%)[29].

Arnold et al. studied the efficacy of adding the antibiotic ciprofloxacin to the treatment of moderately active, but resistant, cases of CD. Forty-seven adults with moderately active CD were randomly assigned treatment with ciprofloxacin 500 mg twice daily vs placebo twice daily for 6 months. Mean CDAI scores at the completion of the study were 112 for the ciprofloxacin group (n = 25) and 205 for the placebo group (n = 12) ($p < 0.001$). They observed modest clinical benefit in the patients receiving ciprofloxacin[30]. In contrast, in a larger placebo-controlled trial reported by Steinhart et al., the addition of both ciprofloxacin and metronidazole to budesonide did not result in any theurapeutic gain compared with budesonide therapy[31].

An important issue when considering the use of antibiotics in IBD is the requirement of a long treatment period; thus tolerability becomes a problem. Metronidazole is associated with some untoward side-effects such as GI intolerance, metallic taste and peripheral neuropathy, which are reported to be 50% or more. Ciprofloxacin is better tolerated; its side-effects are nausea, diarrhoea and skin rashes. The concomitant use of antibiotics significantly increases the incidence of side-effects and the number of drug withdrawals[32]. Taken together these data suggest that neither metronidazole nor ciprofloxacin alone is effective in the management of CD. The combination of these antibiotics may have some beneficial effects[33].

According to ECCO consensus comment, metronidazole 10–20 mg/kg induces a response for colonic disease, but not remission. It is consequently not recommended as first-line therapy, and has a high incidence of side-effects, but has a role in selected patients with colonic disease who wish to avoid corticosteroids[4].

BUDESONIDE

Although the efficacy of classic steroids is well known for induction of remission of mild to moderate CD, their side-effects are also well documented. Furthermore, one-third of CD patients treated with systemic glucocorticoids become steroid-dependent[34]. Budesonide is a useful alternative to systemic steroids when steroid treatment is needed[35]. There are two formulations of budesonide used for treatment of CD; controlled ileal release (Entocort, Astra Zeneca) and pH-dependent release (Budenofalk, Dr Falk Pharma)[36].

It is clear from the studies reviewed that budesonide is superior to placebo (number needed to treat, NNT: 5) and to mesalamine (NNT: 4) for induction of remission of CD. However, the efficacy of budesonide is inferior to conventional steroids (NNT: 12)[36]. There are insufficient data showing the effect of disease location on efficacy of budesonide. There are only two studies evaluating pH-dependent release budesonide in those patients with involvement anywhere in colon except with ileal or ascending colon involvement. The first one is a prospective multicentre, randomized, double-blind study which assessed the effectiveness and safety of oral pH-modified release budesonide in patients with active CD in comparison with 6-methylprednisolone (Metpred). A total of 67 patients with active CD (CDAI >150) were included. Patients were treated with 3×3 mg budesonide ($n = 34$) or Metpred ($n = 33$) according to a weekly tapering schedule (48–32–24–20–16–12–8 mg). Baseline demographics, disease activity and localization of CD in the small bowel and the colon were similar in both treatment groups. On an intention-to-treat basis 19/34 patients in the budesonide group (55.9%) and 24/33 patients in the Metpred group (72.7%) were in remission after 8 weeks ($p = 0.237$)[37].

The second study, performed by Bar-Meir et al., compared the efficacy and safety of budesonide and prednisone in the treatment of active CD involving the terminal ileum and/or the colon. In a randomized, double-blind, controlled trial, patients received either 9 mg budesonide once daily for 8 weeks ($n = 100$)

or 40 mg prednisone once daily for the first 2 weeks tapered gradually to 5 mg/day by the end of the study ($n = 101$). By intention-to-treat analysis, treatment efficacy defined as CDAI <150 at completion was 51% and 52.5% for the budesonide and prednisone groups, respectively[38].

According to ECCO consensus on CD; active colonic CD may be treated with sulphasalazine if only mildly active (EL 1b, RG A) or with systemic corticosteroids (Statement 5D) by the EL 1a and RG A. According to ECCO consensus comment, in its current formulation, oral budesonide has no role in therapy of colonic disease, unless it primarily affects the proximal colon (with or without ileal involvement)[4].

CONCLUSIONS

According to ECCO consensus on CD:

- Active colonic CD may be treated with sulphasalazine if only mildly active (EL 1b, RG A) or with systemic corticosteroids.
- For those who have relapsed, azathioprine/mercaptopurine should be added (EL 1a, RG B) or, if intolerant, methotrexate should be added (EL 1a, RG B).
- Infliximab should be considered in addition for corticosteroid or immunomodulator refractory disease or intolerance (EL 1b, RG B), although surgical options should be considered and discussed.
- Topical treatment should be considered for distal disease (EL 5, RG D)[4].

References

1. Gionchetti P. Conventional therapy of Crohn's disease. World J Gastroenterol. 2006;12: 4794–806.
2. Domenech E. Inflammatory bowel disease: current therapeutic options. Digestion. 2006;73 (Suppl. 1):67–76.
3. Stange EF, Travis SPL, Vermiere S et al. for the European Crohn's and Colitis Organisation (ECCO). European evidence based consensus on the diagnosis and management of Crohn's disease: definitions and diagnosis. Gut. 2006;55(Suppl. I): i1–15.
4. Travis SPL, Stange EF, Lemann M et al. for the European Crohn's and Colitis Organisation (ECCO). European evidence based consensus on the diagnosis and management of Crohn's disease: current management. Gut. 2006;55(Suppl. I):i16–35.
5. Lichtenstein GR, Hanauer SB, Kane SV, Present DH. Crohn's is not a 6-week disease. A life long management of mild to moderate Crohn's disease. Inflamm Bowel Dis. 2004:10: S2–10.
6. Gassull MA. The role of nutrition in the treatment of inflammatory bowel disease. Aliment Pharmacol Ther. 2004;20(Suppl. 4):79–83.
7. Zachos M, Tondeur M, Griffiths AM. Enteral nutrition therapy for induction of remission in Crohn's disease (review). Cochrane Database of Systematic Reviews 2007; Issue 1.
8. Hanauer SB. Review article: Aminosalicylates in inflammatory bowel disease. Aliment Pharmacol Ther. 2004;20(Suppl. 4):60–5.
9. Nielsen OH, Munck LK. Drug insight: aminosalicylates for the treatment of IBD. Nat Clin Pract Gastroenterol Hepatol. 2007;4:160–70.

10. Michetti P, Juillerat P, Mottet C et al. Therapy of mild to moderate luminal Crohn's disease. Digestion. 2005;71:13–18.
11. Anthonisen P, Barany F, Folkenberg O et al. The clinical effect of salazosulphapyridine (Salazopyrin r) in Crohn's disease. A controlled double-blind study. Scand J Gastroenterol. 1974;9:549–54.
12. Van Hees PA, Van Lier HJ, Van Elteren PH et al. Effect of sulphasalazine in patients with active Crohn's disease: a controlled double-blind study. Gut. 1981;22:404–9.
13. Summers RW, Switz DM, Sessions JT Jr et al. National Cooperative Crohn's Disease Study: Results of drug treatment. Gastroenterology. 1979;77:847–69.
14. Sandborn WJ, Feagan BG. Review article. Mild to moderate Crohn's disease – defining the basis for a new treatment algorithm. Aliment Pharmacol Ther. 2003;18:263–77.
15. Malchow H, Ewe K, Brandes JW et al. European Cooperative Crohn's Disease Study (ECCDS): Results of drug treatment. Gastroenterology. 1984;86:249–66.
16. Hanauer SB. The case for using 5-aminosalicylates in Crohn's disease. Pro. Inflamm Bowel Dis. 2005;11:609–12.
17. Rasmussen SN, Lauritsen K, Tage-Jensen U et al. 5-Aminosalicylic acid in the treatment of Crohn's disease. A 16-week double-blind, placebo-controlled, multicentre study with Pentasa. Scand J Gastroenterol. 1987;22:877–83.
18. Mahida YR, Jewell DP. Slow release of 5-aminosalicylic acid (Pentasa) for the treatment of active Crohn's disease. Digestion. 1990:45:88–92.
19. Tremaine WJ, Schroeder KW, Harrison JM, Zinsmeister AR. A randomized, double-blind, placebo-controlled trial of the oral mesalamine (5-ASA) preparation, Asacol, in the treatment of symptomatic Crohn's colitis and ileocolitis. J Clin Gastroenterol. 1994;19: 278–82.
20. Bergman R, Parkes M. Systematic review: The use of mesalazine in inflammatory bowel disease. Aliment Pharmacol Ther. 2006;23:841–55.
21. Gionchetti P, Rizzello F, Lammers KM et al. Antibiotics and probiotics in treatment of inflammatory bowel disease. World J Gastroenterol. 2006;12:3306–13.
22. Gionchetti P, Rizzello F, Lammers KM, Morselli C, Tambasco R, Campieri M. Antimicrobials in the management of inflammatory bowel disease. Digestion. 2006;73 (Suppl. 1):77–85.
23. Blichfeldt P, Blomhoff JP, Myhre E, Gjone E. Metronidazole in Crohn's disease: a double blind cross-over clinical trial. Scand J Gastroenterol. 1978;13:123–7.
24. Ambrose NS, Allan RN, Keighley MR et al. Antibiotic therapy for treatment in relapse of intestinal Crohn's disease. A prospective randomized study. Dis Colon Rectum. 1985;28: 81–5.
25. Ursing B, Alm T, Barany F et al. A comparative study of metronidazole and sulfasalazine for active Crohn's disease: the cooperative Crohn's disease study in Sweden. II. Result. Gastroenterology. 1982;83:550–62.
26. Sutherland L, Singleton J, Sessions J et al. Double blind, placebo controlled trial of metronidazole in Crohn's disease. Gut. 1991;32:1071–5.
27. Prantera C, Zannoni F, Scribano ML et al. An antibiotic regimen for the treatment of active Crohn's disease: a randomized, controlled clinical trial of metronidazole plus ciprofloxacin. Am J Gastroenterol. 1996;91:328–32.
28. Colombel JF, Lemann M, Cassagnou M et al. A controlled trial comparing ciprofloxacin with mesalazine for the treatment of active Crohn's disease. Groupe d'Etudes Therapeutiques des Affections Inflammatoires Digestives (GETAID). Am J Gastroenterol. 1999;94:674–8.
29. Greenbloom SL, Steinhart AH, Greenberg GR. Combination ciprofloxacin and metronidazole for active Crohn's disease. Can J Gastroenterol. 1998;12:53–6.
30. Arnold GL, Beaves MR, Pryjdun VO, Mook VJ. Preliminary study of ciprofloxacin in active Crohn's disease. Inflamm Bowel Dis. 2002;8:10–15.
31. Steinhart AH, Feagan BG, Wong CJ et al. Combined budesonide and antibiotic therapy for active Crohn's disease: a randomized controlled trial. Gastroenterology. 2002;123:33–40.
32. Guslandi M. Antibiotics for inflammatory bowel disease: do they work? Eur J Gastroenterol Hepatol. 2005;17:145–7.
33. Wild GE. The role of antibiotics in the management of Crohn's disease. Inflamm Bowel Dis. 2004;10:321–3.

34. Chopra A, Darrell SP, Loftus EV et al. Budesonide in the treatment of inflammatory bowel disease: the first year of experience in clinical practice. Inflamm Bowel Dis. 2006;12:29–32.
35. Schölmerich J. Review article. Systemic and topical steroids in inflammatory bowel disease. Aliment Pharmacol Ther. 2004;20(Suppl. 4):66–74.
36. Otley A, Steinhart AH. Budesonide for induction of remission in Crohn's disease. Cochrane Database of Systematic Reviews 2005; Issue 4.
37. Gross V, Andus T, Caesar I et al. Oral pH-modified release budesonide versus 6-methylprednisolone in active Crohn's disease. German/Austrian Budesonide Study Group. Eur J Gastroenterol Hepatol. 1996;8:905–9.
38. Bar-Meir S, Chowers Y, Lavy A et al. Budesonide versus prednisone in the treatment of active Crohn's disease. Israeli Budesonide Study Group. Gastroenterology. 1998;115:835–40.

23
State-of-the-Art Lecture: Future therapies in inflammatory bowel diseases

J. SCHÖLMERICH

INTRODUCTION

There is obviously a need for new therapies in inflammatory bowel disease (IBD). Among the goals of treatment, induction and maintenance of clinical remission without steroids is currently the most important practical aim and can be achieved in the majority of patients. However, in the future the preservation of the main functions of the intestinal tract for many decades of life in these young patients seems to be a more attractive goal. Those functions comprise the absorption of nutrients, the retention of water and electrolytes, and the controlled release of faeces. This implies the avoidance of surgery and/or structural damage due to continuous inflammation and scarring. The ultimate goal is obviously the healing of the disease, which means elimination or neutralization of susceptibility factors which are mostly not yet even known.

EPIDEMIOLOGY AS BASIS OF TREATMENT

It is obvious that IBD comprise a number of entities which currently are named 'Crohn's disease', 'ulcerative colitis' and 'colitis undetermined' (formerly 'indeterminate colitis'). However, looking at symptoms of these disorders it is obvious that there is a huge overlap between the main entities (Figure 1). This is also apparent when looking at the clinical picture of IBD, which ranges from oral aphthae to perianal fistulas and many even extraintestinal problems such as eye, joint and skin lesions. Therefore it does not seem to be appropriate in the long term to discuss treatments of Crohn's disease (CD) and ulcerative colitis (UC) separately, but to discuss treatments for subgroups of the 'inflammatory bowel syndrome' according to the genotypes and phenotypes.

The course of disease in CD, as well as in UC, seems to be more benign, as discussed and presented in most of the referal centre data[1,2]. For example the disease course in CD over 10 years as demonstrated from the IBSEN study in Oslo shows that almost half of the patients have a benign course with an initial

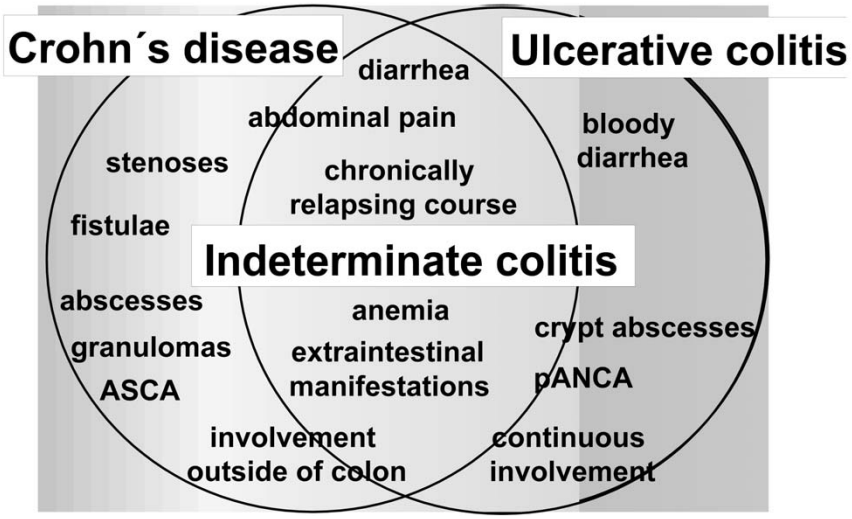

Figure 1 Inflammatory bowel diseaases

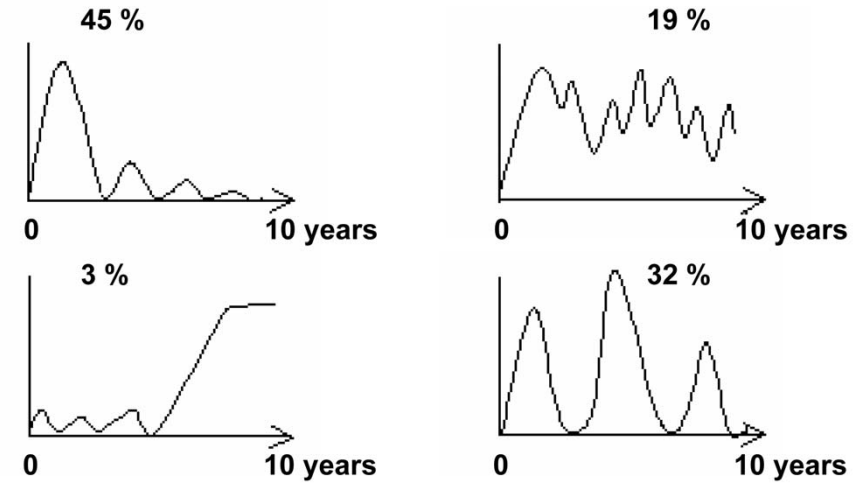

Figure 2 Disease course in Crohn's disease over 10 years (Ibsen study[2])

flare and then very low activities over the next 10 years; 32% have relapsing activity without clear-cut aggravation; 3% have an initial mild course and then change to a severe form, and 20% have continuous activity and those patients are probably the ones who really need new treatment approaches (Figure 2)[2].

Currently the principles of pathophysiology are based on the immunology paradigm which describes the disturbed functions of gut lymphocytes for whatever reason reacting in an uncontrolled fashion to luminal components and producing a whole orchestra of chemokines and cytokines, among many other tumour necrosis factors (TNF). These mediators have many effects including the attraction of new immune cells such as neutrophils and macrophages. Those in turn again release, after activation, another orchestra of cytokines and more mediators such as leucotrienes, platelet-activating factor, reactive oxidant species and many more which finally lead to destruction of the target cell, namely the epithelial cell in the gut. Based on this paradigm many treatments have been aimed at immune suppression, i.e. inhibiting lymphocyte activity or even inducing lymphocyte destruction.

APOPTOSIS AS A TARGET

The example of *Caenorhabditis elegans* demonstrates how important apoptosis is for normal function of an organism. In addition it has been shown in many examples that apoptosis is a basic principle of dysfunction of many organs. Consequently it has been shown that inhibition and induction of apoptosis can be useful in a number of different diseases (Table 1). Some of these treatments are already registered by the FDA, some are still in clinical testing. It appears from current knowledge that modern immune suppressants such as azathioprine and methotrexate, but also biologicals such as infliximab and others, induce apoptosis of activated lymphocytes. This has been beautifully shown by recent work from the Netherlands[3].

Table 1 Manipulation of apoptosis as therapeutic approach (examples)

Substance	*Phase*	*Disease*
Inhibition		
Rasagiline	FDA⊕	Parkinson
Amifostine	FDA⊕	Chemotherapy effects
IDN 6556	II	Hepatitis C, alcoholic hepatitis
VX-740	II	Rheumatoid arthritis
Induction		
Survivin	I	Cancer
Advexin	III	Cancer
Onyx-015	III	Colon–pancreas cancer
Cetuximab	FDA⊕	Colon cancer
Gefitinib	III	Pancreas cancer
Imatinib mesylate	FDA⊕	GIST
Erlotinib	III	Pancreas cancer

⊕ = approved; GIST = gastrointestinal stroma tumour

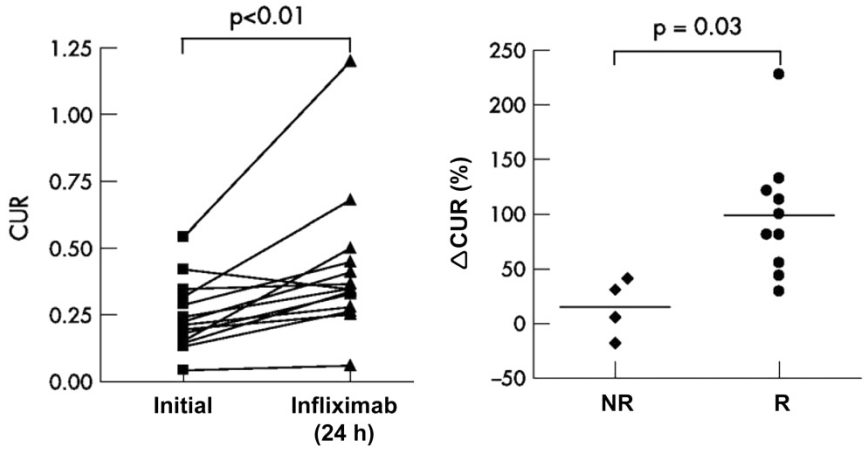

Figure 3 Annexin V SPECT before and after infliximab[3]. CUR = colonic uptake ratio, R = responder, NR = non-responder

There it was shown in humans, using a nuclear medicine technology, that there is apoptosis in the gut in patients treated with the TNF-antibody infliximab (Figure 3). Actually the amount of apoptosis induction determined patient response to this treatment. It is not clear, however, if all of the new anti-TNF agents really induce apoptosis, but it seems likely to be a major mechanism of immunosuppression.

However, unfortunately long-term results of all currently used biologics, when all patients initially treated are calculated, show a 6-month remission percentage between 20% and 30%, which is obviously unsatisfactory. Recently it has been shown that better patient selection, and restricting treatment to patients who are truly refractory and have definitive inflammation in the gut, increases remission rates significantly towards the range of 80% in CD but fails to do so in patients with UC, where it only reaches 44% (Table 2)[4].

In addition to those (at least at first glance) disappointing results, it needs to be mentioned that all types of immunosuppression actually increase the infection risk in patients treated. This is the more obvious the more combined immunosuppressants patients are using, which includes steroids, azathioprine, methotrexate, cyclophosphamide and obviously all the modern biologics. A recent publication demonstrated that quite a significant number of patients treated with conventional immunosuppression fell below a level of 250 T-helper cells per microlitre, and that a majority of those presented clinically with opportunistic infections[5]. Recent data from other series and papers demonstrate clearly that this is also the case with anti-TNF therapy, regardless of the indication, i.e. in rheumatoid arthritis as well as in CD[6,7].

It is obvious from these data that the current wave of biologics is probably not the final solution for the goals discussed at the beginning of this chapter.

Table 2 Patient selection increases response to infliximab in IBD[4]

	Remission (%)	*Response (%)*
Fistulizing Crohn's disease	50.0	77.2
Luminal Crohn's disease	80.4	91.3
Isolated colonic Crohn's disease	92.9	–
Isolated small bowel Crohn's disease	71.9	–
Ulcerative colitis/indeterminate colitis	43.8	75.0

Selection: truly refractory patients, ascertainment of inflammation; 70 patients.

DO WE HAVE ALTERNATIVES?

There are quite a number of possibilities of progress in medicine; this reaches from the understanding of basic mechanisms leading to the solution of the problem, through the trial-and-error system which actually succeeds by the sheer number of attempts made, to the 'belief and accident system' which actually hits the target by luck. All of these are rather unlikely in a situation where the disease or the syndrome has many causes, influential factors and modifiers making it very unlikely that a single cause will ever be detected, and therefore be treated.

THE DEFECTIVE BARRIER AS A TARGET

When taking the human organism as a simple dual entity as shown in Figure 4 it becomes very understandable that the symbiosis between our genome and the coded proteins and cells on one side and the ten times more frequent bacteria with their hundred times more genes and unknown number of cell wall and cytoplasmic products on the other side of the intestinal barrier is the basis of the homeostasis in our gut, and is the reason why most of us have a very quiet intestinal system. It has been known for quite a while that the innate immune system is responsible for this homeostasis and that disturbances of the system can lead to autoimmune disorders and other diseases[8]. This can be nicely shown when looking at the first gene where a mutation leads to manifestation of CD in a significant number of patients, namely with the NOD2/CARD15 gene. This gene codes for an intracellular receptor for bacterial products, namely peptidoglycans, and seems to be rather important for the maintenance of this intestinal homeostasis. This is nicely illustrated by our recent study showing that recipients of stem cell transplantation for leukaemia, being heterozygote for this gene, have a significantly increased mortality after stem cell transplantation. Homozygotes or compound heterozygotes for this mutation actually reached a transplantation-related mortality of 80%. This problem could be almost abolished when patients have been conditioned with an antibiotic system eliminating Gram-positive and Gram-negative bacteria. This study demonstrates that these receptors of the innate immune system play a very significant role related to our intestinal homeostasis (Figure 5)[9,10].

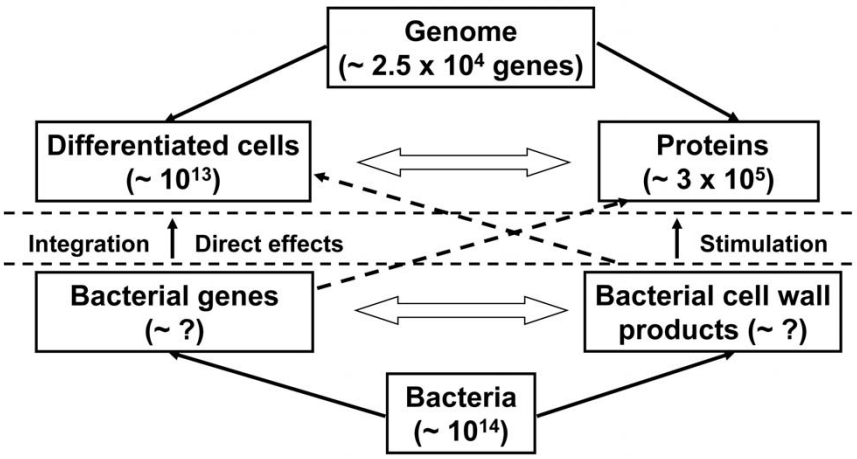

Figure 4 Components of the human organism

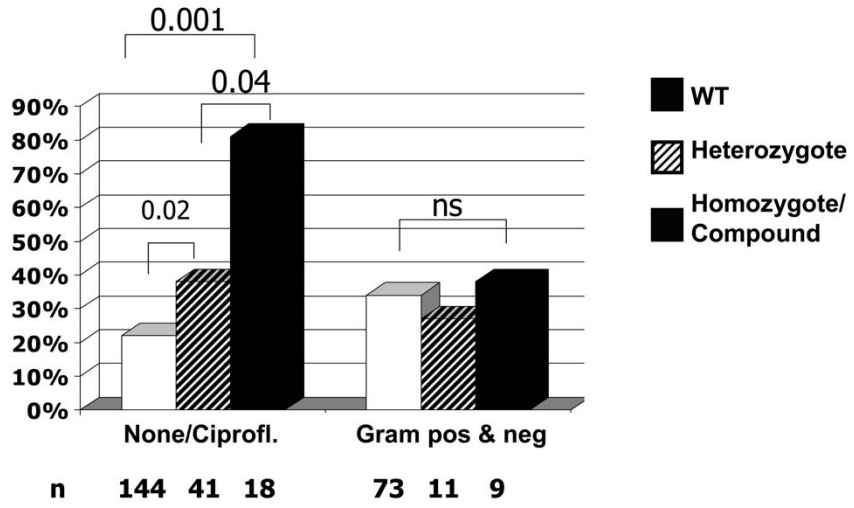

Figure 5 Role of decontamination for transplant related mortality in relation to NOD2/CARD15 genotype. WT = wild type

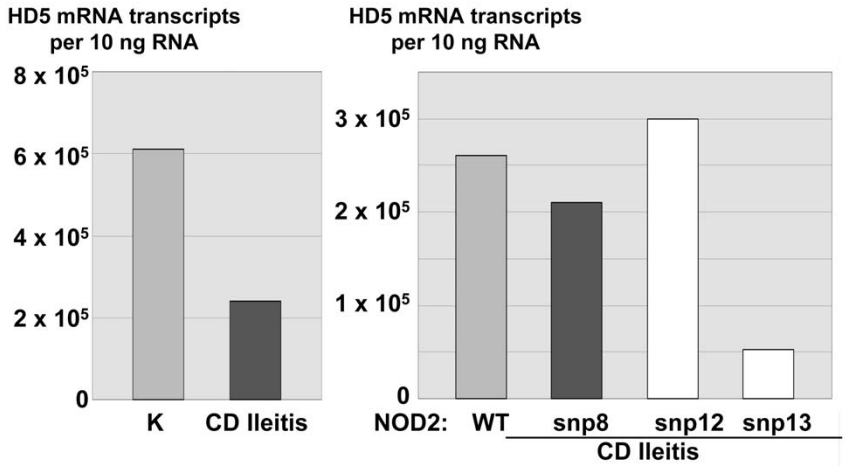

Figure 6 Expression of defensin 5 RNA in the ileum – effect of NOD2 mutations[11]

Another nice example is the finding that intestinal antibacterial proteins such as defensins and others seem to be deficient (in addition related to mutations such as the NOD2 mutation) in patients with the ileal CD type of IBD, and are not produced by the Paneth cells which are normally responsible for their release (Figure 6)[11].

It may be concluded from these and many recent findings that the intestinal symbiosis of host and flora may be one of the keys to IBD and its future treatment. Many interactions and dysfunctions of our immune system depend at least in part on the 'correct' intestinal colonization. This cannot be shown in detail in this chapter. Most known genetic defects, and probably in addition particular exogenous alterations, disturb the homeostasis and are the cause of very different disorders, in our case IBD, but also asthma and sarcoidosis.

Attempts to modify the luminal side of this symbiosis have been successful. A recent study with rifaximin, a non-absorbal antibiotic, showed significant effects in CD which are more pronounced in patients with proven inflammation (Table 3)[12]. These data confirm many other earlier data on the positive effects of antibiotics in the Crohn syndrome. Similarly in pouchitis, and also in UC, probiotics have been found to be effective, mostly in maintaining remission, but also in the induction of remission in more recent studies[13]. The application of *Trichuris suis* ova, the eggs of a helminth which cannot replicate in the human gut, has been found to be effective in UC in a placebo-controlled trial and in CD in a pilot study (Table 4)[14]. These helminths may induce regulatory T cells which seem to be effective in experimental colitis in several models of IBD[15]. The application of modified bacteria which produce anti-inflammatory cytokines such as interleukin-10 has shown promising effects in human CD, and this needs to be replicated in further clinical trials (Figure 7)[16]. Thus far defensins or other endogenous anti-infectious peptides have not been tested in humans, but this is under way.

Table 3 Rifaximin in active Crohn's disease[12]

	Placebo (*n* = 27)	*Rifaximin* 1 × 800 mg (*n* = 25)	*Rifaximin* 2 × 800 mg (*n* = 27)
All patients			
Remission (%)	33	32	52
Patients with CRP ↑			
No.	14	16	16
Remission (%)	21	25	63*

*Significant.

Table 4 *Trichuris suis* ova (TSO) in active ulcerative colitis[14]

	TSO (*n* = 30)	*Placebo* (*n* = 24)
Improvement (%)**	43.3	16.7*
Cross-over positive (%)	56.3	13.3*
All phases (%)	47.8	15.4*

*Significant; **Δ CAI $\geqslant$ 4.

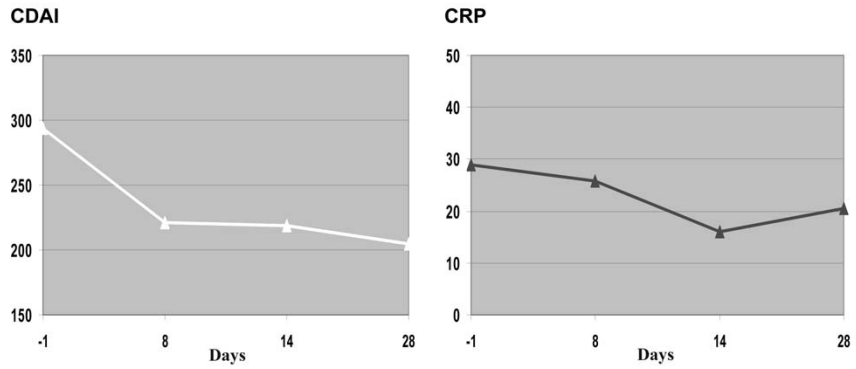

Figure 7 *Lactococcus lactis* (IL-10 expression) in active Crohn's disease – phase I study[16]

Table 5 Phosphatydylcholine orally for active ulcerative colitis[19]

	3 months	
	Phosphatidylcholine (*6 g/day*)	*Placebo*
Remission (%)	53	10*
Response (%)	90	10*
Δ Histology score > 50% (%)	44	13*

*Significant.

Table 6 Lack of intestinal bile and bacterial translocation in the rat[20]

	MLN *(%)*	*Spleen* *(%)*	*All organs* *(%)*
Controls ($n = 20$)	5	5	3
Sham operation ($n = 22$)	14	0	5
Bile duct ligation ($n = 25$)	48	24	32
Bile diversion ($n = 23$)	65	35	41

MLN = mesenterial lymph nodes.

Table 7 UDCA improves experimental ileitis and colitis

	Macroscopic score	
	Vehicle	*UDCA*
Indomethacin ileitis	6.2 ± 1.4	3.0 ± 2.5*
TNBS colitis	3.6 ± 1.4	1.8 ± 1.6*

*$p < 0.05$.

Microscopic score, myeloperoxidase, gut weight were also improved.

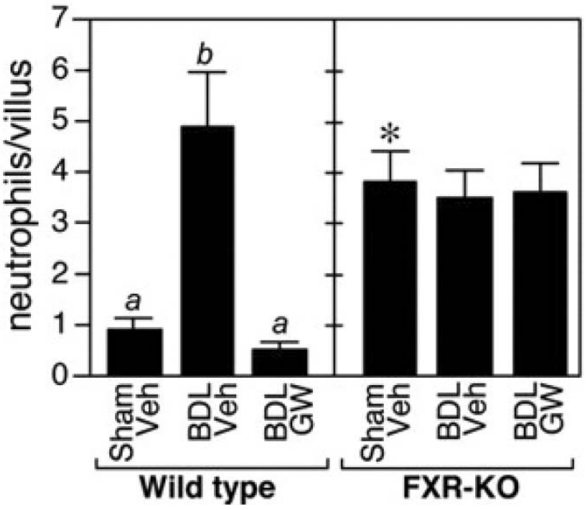

Figure 8 FXR activation blocks mucosal injury due to bile duct ligation[23]. BDL = bile duct ligation, Sham = sham operation, Veh = vehicle, GW = FXR ligand, FXR-KO = knockout for FXR (nuclear bile acid receptor)

NUTRIENTS, LIPIDS AND ABDOMINAL FAT AS A TARGET

More recently it has been shown that mesenteric fat and its products seem to be very much involved in the inflammation in the gut in IBD[17,18]. This is in particular obvious from the 'creeping fat' and other macroscopic phenomena observed by surgeons many years ago. The modulation of adipocyte products such as adiponectin, resistin and others has not yet been tested, but is currently being examined in animal experiments. The recent clinical trial using a simple fat such as phosphatidylcholine given orally in active UC has shown surprising remission rates (Table 5)[19], and has been reproduced in another trial using patients with steroid-refractory colitis. These exciting data need to be reproduced in an independent study, and look very interesting and promising.

Another approach may be even more related to endogenous compounds. It has long been known that the lack of intestinal bile increases bacterial translocation in different animals when different models are used (Table 6)[20]. Accordingly it was shown several years ago that one bile acid, namely ursodeoxycholic acid, improves experimental ileitis and colitis when given orally in animals. This is true for indomethacin ileitis and TNBS colitis regarding macroscopic and microscopic scores, neutrophil infiltration and other signs of inflammation (Table 7)[21,22]. Very recently this has been nicely explained by the fact that the activation of the nuclear receptor for bile acids, FXR, can attenuate mucosal injury and even mucosal fibrogenesis very effectively. Therefore bile acids or analogues binding to the FX receptor, given in the right concentration and delivered to the right place, may be a very interesting new approach to IBD (Figure 8)[23].

There are many such novel approaches which are all related to the homeostasis of the bacterial–human interaction in the gut. Most of these approaches do not need to be expensive, and probably will not be of immunosuppressive nature. It remains to be seen if any of those approaches are as good as our current standard treatment. It will not be too difficult to reach the effectiveness level of the current wave of very expensive and aggressive biological immunosuppression.

References

1. Henriksen M, Jahnsen J, Lygren I et al.; IBSEN Study Group. Ulcerative colitis and clinical course: results of a 5-year population-based follow-up study (the IBSEN study). Inflamm Bowel Dis. 2006;12:543–50.
2. Henriksen M, Jahnsen J, Lygren I et al.; the IBSN Study Group. Clinical course in Crohn's disease: results of a five-year population-based follow-up study (the IBSEN study). Scand J Gastroenterol. 2007;42:602–10.
3. Van den Brande JM, Koehler TC, Zelinkova Z et al. Prediction of antitumour necrosis factor clinical efficacy by realtime visualization of apoptosis in patients with Crohn's disease. Gut. 2007;56:509–17.
4. Pearce CB, Lawrance IC. Careful patient selection may improve response rates to infliximab in inflammatory bowel disease. J Gastroenterol Hepatol. 2007;22:1671–7.
5. Glück T, Kiefmann B, Grohmann M, Falk W, Straub RH, Schölmerich J. Immune status and risk for infection in patients receiving chronic immunosuppressive therapy. J Rheumatol. 2005;32:1473–80.
6. Bongartz T, Sutton AJ, Sweeting MJ, Buchan I, Matteson EL, Montori V. Anti-TNF antibody therapy in rheumatoid arthritis and the risk of serious infections and

malignancies: systemic review and meta-analysis of rare harmful effects in randomized controlled trials. J Am Med Assoc. 2006;295:2275–85.
7. Siegel CA, Hur C, Korzenik JR, Gazelle GS, Sands BE. Risks and benefits of infliximab for the treatment of Crohn's disease. Clin Gastroenterol Hepatol. 2006;8:1017–24.
8. Gregersen PK. Modern genetics, ancient defenses, and potential therapies. N Engl J Med. 2007;356:1263–6.
9. Holler E, Rogler G, Herfarth H et al. Both donor and recipient NOD2/CARD15 mutations associate with transplant-related mortality and gastrointestinal GvHD following allogeneic stem cell transplantation. Blood. 2004;104:889–94.
10. Holler E, Rogler G, Brenmoehl J et al. Prognostic significance of NOD2/CARD15 variants in HLA-identical sibling transplantation: effect on long term outcome is confirmed in 2 independent cohorts and may be modulated by the type of gastrointestinal decontamination. Blood. 2006;107:4189–93.
11. Wehkamp J, Fellermann K, Herrlinger KR, Bevins CL, Stange EF. Mechanisms of disease: defensins in gastrointestinal diseases. Nat Clin Pract Gastroenterol Hepatol. 2005;2:406–15.
12. Prantera C, Lochs H, Campieri M et al. Antibiotic treatment of Crohn's disease: results of a multicentre, double blind, randomized, placebo-controlled trial with rifaximin. Aliment Pharmacol Ther. 2006;23:1117–25.
13. Biblione R, Fedorak RN, Tannock GW et al. VSL#3 probiotic-mixture induces remissions in patients with active ulcerative colitis. Am J Gastroenterol. 2005;100:1539–46.
14. Summers RW, Elliott DE, Urban JF, Thompson RA, Weinstock JV. *Trichuris suis* therapy for active ulcerative colitis: a randomized controlled trial. Gastroenterology. 2005;128:825–32.
15. Braat H, McGuirk P, Ten Kate FJ et al. Prevention of experimental colitis by parenteral administration of a pathogen-derived immunomodulatory molecule. Gut. 2007;56:351–7.
16. Braat H, Rotties P, Hommes DW et al. A phase I trial transgenic bacteria expressing interleukin-10 in Crohn's disease. Clin Gastroenterol Hepatol. 2006;4:754–9.
17. Peyrin-Biroulet L, Chamaillard M, Gonzales F et al. Mesenteric fat in Crohn's disease: a pathogenetic hallmark or an innocent bystander? Gut. 2007;56:577–83.
18. Schäffler A, Schölmerich J, Buechler C. Mechanisms of disease: adipocytokines and visceral adipose tissue: emerging role in intestinal and mesenteric diseases. Nature Clin Pract Gastroenterol Hepatol. 2005;2:103–11.
19. Stremmel W, Merle U, Zahn A, Autschbach F, Hinz U, Ehehalt R. Retarded release phosphatidylcholine benefits patients with chronic active ulcerative colitis. Gut. 2005;54:966–71.
20. Slocum MM, Sittig KM, Specian RD, Deitch EA. Absence of intestinal bile promotes bacterial translocation. Am Surg. 1992;58:305–10.
21. Kullmann F, Gross V, Rüschoff J et al. Effect of ursodeoxycholic acid on the inflammatory activity of indomethacin-induced intestinal inflammation in rats. Z Gastroenterol. 1997;35:171–8.
22. Kullmann F, Arndt H, Gross V, Rüschoff J, Schölmerich J. Beneficial effect of ursodeoxycholic acid on mucosal damage in trinitrobenzene sulphonic acid-induced colitis. Eur J Gastroenterol Hepatol. 1997;9:1205–12.
23. Inagaki T, Moschetta A, Lee YK et al. Regulation of antibacterial defense in the small intestine by the nuclear bile acid receptor. Proc Natl Acad Sci USA. 2006;103:3920–5.

Section VIII
Cases and controversies

Chair: D KARAMANOLIS and C GASCHE

24
Management of fistulizing Crohn's disease: the conservative approach

H. Ö. HAMZAOĞLU

INTRODUCTION

Fistulas with Crohn's disease (CD) are reported to occur in between 20% and 40% in different studies[1–4]. The development of fistulas may precede or be coincident with the diagnosis of CD. Patients with colonic diseases have a higher incidence of perianal fistulas. The effective management of fistulas in patients with CD presents an extremely challenging problem. A population-based study has shown that 82% of fistulizing patients will eventually need surgery. Most of these surgical procedures (83%) are minor ones such as abscess drainage or seton replacement, but 23% resulted in bowel resection with proctectomy[5].

The first step should be to define the exact anatomical feature of the fistula and rule out infectious complications such as an abscess. The only treatment for abscess is surgical drainage. Good collaboration between the gastroenterologist and the surgeon is essential in management. Endoscopy and either magnetic resonance imaging (MRI) or endoscopic ultrasound (EUS) must be performed in all patients before starting treatment. Examination under anaesthesia (EUA) is also very helpful in determining the extent and type of fistula. Abscess drainage and/or seton placement may also be performed at the same time. It is possible to reach 100% diagnostic accuracy by using the combination of MRI or EUS with EUA[6].

CLASSIFICATION

Several fistula classification systems have been proposed. The most anatomically precise one is Parks' classification, which uses the external sphincter as a central point of reference. This has five subtypes: inter-sphincteric, trans-sphincteric, supra-sphincteric, extra-sphincteric, and superficial fistula[7]; however, it is not easy to use in everyday clinical practice. A simpler classification that we use in our clinical practice is based on the American Gastroenterological Association medical position statement paper. In this classification the fistula is either simple or complex. A simple fistula is

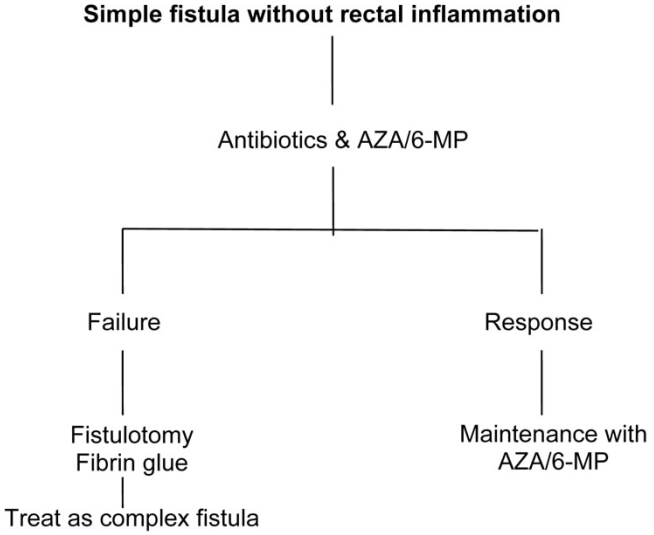

Figure 1 Management algorithm of simple fistula without rectal inflammation

defined as a superficial, inter-sphincteric or low trans-sphincteric fistula with only one opening, and it is not associated with an abscess or connected to an adjacent structure. In contrast, a complex fistula is one that involves more of the anal sphincters, has multiple openings, is associated with a perianal abscess and/or connects to an adjacent structure, such as the vagina or bladder[8]. The choice of treatment modality could be based on this simple classification system, but it is important to keep in mind that every patient is unique and must be treated individually.

MEDICAL THERAPY IN FISTULIZING CD

Fistulizing CD can be very difficult to treat. A combination of medical therapies is almost always necessary. Despite the lack of sufficient evidence, metronidazole and/or ciprofloxacin is the first choice in simple fistulizing CD without colonic inflammation. Antibiotics can be a bridge until azathioprine–6-mercaptopurine (AZA/6-MP) start to work. If colonic inflammation with simple fistula or complex fistula is the case it would be wiser to start with antibiotics and AZA/6-MP together as first-line treatment. AZA/6-MP still seems to be the mainstay of long-term treatment of fistulas. Infliximab has clearly proved its efficacy in the short-term treatment of fistulizing CD. Complete healing of fistula was reported in 55% of patients compared to 13% on placebo in the first trial[9]. Infliximab is generally well tolerated and safe, but serious side-effects such as infection, demyelinating disorders, congestive heart failure, autoimmunity, and malignancy might occur, and careful follow-up is mandatory[10–12].

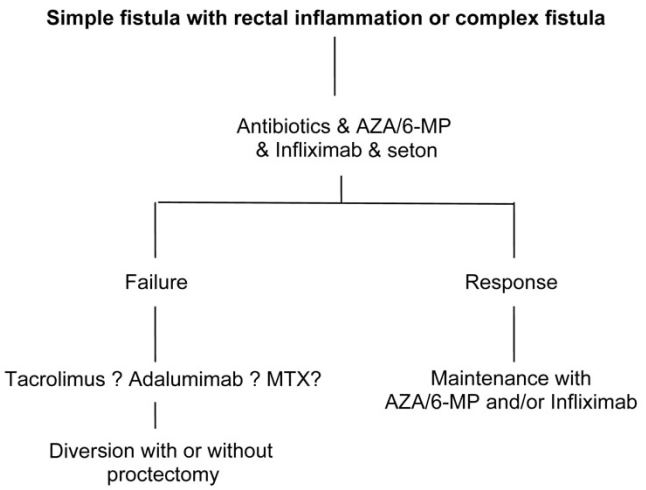

Figure 2 Management algorithm of simple fistula with rectal inflammation or complex fistula

The ACCENT II maintenance trial showed that, at 54 weeks, 46% of patients receiving infliximab continued to respond to treatment, compared with 23% in the placebo group[13]. Patients receiving infliximab therapy may develop antibodies to infliximab and the presence of these antibodies has been associated with the development of infusion reactions and a reduction in therapeutic efficacy over time[14]. Concomitant use of immunomodulators with infliximab seems to decrease antibody formation.

Adalimumab is a fully human IgG1 monoclonal antibody to tumour necrosis factor alpha. Experience with fistulizing CD is still limited. In CHARM-I (Crohn's trial of the fully human antibody adalimumab for remission maintenance-I) study, the effect of adalumimab on CD was evaluated. In a subgroup of patients with fistula, maintenance adalimumab therapy resulted in complete fistula closure at the last two visits in 14% of placebo-treated patients, in 36% of patients receiving adalimumab 40 mg every other week, and in 46% of patients receiving adalimumab 40 mg weekly[15]. The GAIN (gauging adalimumab efficacy in infliximab non-responders) study evaluated the efficacy and safety of adalimumab maintenance therapy in CD patients who had previously responded to infliximab and then lost response or became intolerant. This study demonstrated that adalimumab therapy was also effective in this group of patients. Adalumimab seems to be an alternative for future use in fistulizing CD management[16].

Methotraxate is another immunosuppressant drug shown to be superior to placebo both in remission induction and maintenance therapy in CD, but the data on fistulizing CD are very limited. A 25% complete fistula response rate and a 31% partial response rate were reported in a small study[17]. The toxicity and teratogenicity with chronic use of methotrexate is well known. Cyclosporin and tacrolimus could be tried as induction therapy in refractory cases, but they have no role in maintenance[18,19].

AZA/6-MP still seems to be the mainstay of long-term treatment of fistulas, but current evidence suggest that infliximab has proven its efficacy in controlled studies. Duration, intervals and dosage of infusions still need to be determined for longer periods, since CD is a lifelong disorder. Hopefully time will provide us with more and accurate data, not just for infliximab but also for developing other new biological therapies.

References

1. Farmer RG, Hawk WA, Turnbull RB Jr. Clinical patterns in Crohn's disease: a statistical study of 615 cases. Gastroenterology. 1975;68:627–35.
2. Rankin GB, Watts HD, Melnyk CS, Kelley ML Jr. National Cooperative Crohn's Disease Study: extraintestinal manifestations and perianal complications. Gastroenterology. 1979; 77:914–20.
3. Hellers G, Bergstrand O, Ewerth S, Holmstrom B. Occurrence and outcome after primary treatment of anal fistulae in Crohn's disease. Gut. 1980;21:525–7.
4. Tang LY, Rawsthorne P, Bern CN. Are perineal and luminal fistulas associated in Crohn's disease? A population-based study. Clin Gastroenterol Hepatol. 2006;4:1130–4.
5. Schwartz DA, Loftus EV, Tremaine WJ et al. The natural history of fistulizing Crohn's disease in Olmsted County, Minnesota. Gastroenterology. 2002;122:875–80.
6. Wise P, Schwartz DA. Management of perianal Crohn's disease. Clin Gastroenterol Hepatol. 2006;4:426–30.
7. Parks AG, Gordon PH, Hardcastle JD. A classification of fistula-in-ano. Br J Surg. 1976; 63:1–12.
8. American Gastroenterological Association. American Gastroenterological Association medical position statement: perianal Crohn's disease. Gastroenterology. 2003;125:1503–7.
9. Present DH, Rutgeerts P, Targan S et al. Infliximab for the treatment of fistulas in patients with Crohn's disease. N Engl J Med. 1999;340:1398–405.
10. Blonski W, Lichtenstein GR. Complications of biological therapy for inflammatory bowel diseases. Curr Opin Gastroenterol. 2006;22:30–43.
11. Desai SB, Furst DE. Problems encountered during anti-tumour necrosis factor therapy. Best Pract Res Clin Rheumatol. 2006;20:757–90.
12. Reddy JG, Loftus EV. Safety of infliximab and other biologic agents in the inflammatory bowel diseases. Gastroenterol Clin N Am. 2006;35:837–55.
13. Sands BE, Anderson FH, Bernstein CN et al. A randomized controlled trial of infliximab maintenance therapy for fistulizing Crohn's disease (ACCENT II). N Engl J Med. 2004; 350:876–85.
14. Baert F, Noman M, Vermeire S et al. Influence of immunogenicity on the long-term efficacy of infliximab in Crohn's disease. N Engl J Med. 2003;348:601–8.
15. Colombel J, Sandborn WJ, Rutgeerts P et al. Adalimumab induces and maintains clinical response and remission in patients with active Crohn's disease: results of the CHARM trial. Gastroenterology. 2006;130 (Abstract 686d).
16. Sandborn WJ, Rutgeerts P, Enns RA et al. Adalimumab rapidly induces clinical remission and response in patients with moderate to severe Crohn's disease who had secondary failure to infliximab therapy: results of the GAIN study. Am J Gastroenterol. 2006;101:S448 (Abstract).
17. Mahadevan U, Marion JF, Present DH. Fistula response to methotrexate in Crohn's disease: a case series. Aliment Pharmacol Ther. 2003;18:1003–8.
18. Present DH, Lichtiger S. Efficacy of cyclosporine in treatment of fistula of Crohn's disease. Dig Dis Sci. 1994;39:374–80.
19. Sandborn WJ, Isaacs KL. Tacrolimus (FK506) for the treatment of perianal and enterocutaneous fistulas in patients with Crohn's disease: a randomized, double blind, placebo-controlled trial. Gastroenterology. 2002;122:A81.

25
Fistulizing Crohn's disease: the aggressive approach

B. E. SANDS

Fistulizing Crohn's disease (CD) presents unique challenges for both patient and physician. Fistulas are a frequent complication of CD and reflect the transmural nature of the inflammatory process. These may arise at any time in the course of the disease, and 46% of all fistulas in CD have been noted either before or at the time of diagnosis[1,2]. The cumulative incidence of fistulization in CD has been estimated to range between 20% and 40%[1,2]. It is also estimated that surgery is needed for approximately 83% of fistula episodes[1,2]. Enterocutaneous, perianal, and rectovaginal fistulas have important and detrimental impacts upon the patient's quality of life. In addition, patients with perineal fistulas are at increased risk for surgery, and in the most severe cases this may include proctectomy and permanent stoma. Other surgical approaches to fistula, such as mucosal flaps or gracilis muscle interposition, have a high rate of failure. Therefore, a diagnosis of fistulizing CD calls for aggressive treatment, regardless of the activity of the luminal disease.

The majority of fistulas occur in a perianal location[1,2]. Approximately one-quarter of fistulas are enteroenteric and may be of little consequence in the quality of life of the patient[3]. These may be asymptomatic, if short segments of bowel have been bypassed; however, these may also be a focus of abscess, particularly if obstruction occurs distal to the fistula. Nine per cent of all fistulas are rectovaginal; these may present as complaints of dyspareunia or chronic vaginal drainage. Other fistula locations may include enterovesicular fistulas[4], enterocutaneous fistulas to the abdomen or groin, or very rarely cologastric[5] or fistulas to the urinary collecting system or ureter[6]. Fistulas of this sort are highly refractory to medical therapy, and most will require surgical approaches in order to be corrected.

The gastroenterologist plays a key role in managing fistulizing CD. The first task is to control the overall disease activity. Regardless of the effect on the fistula itself, controlling diarrhoea will serve to decrease the output through fistulas to the skin and decrease symptoms related to this. The gastroenterologist's next task is to determine the course and complexity of the fistula. This is a key point in choosing appropriate combinations of medical and surgical approaches. An obvious goal is to induce closure of the fistula and to

use medical therapy to maintain closure over time. The gastroenterologist should also serve to limit the scope of surgical intervention, which in many cases will result in poor or undesirable outcomes. Ultimately the goal is to improve the patient's quality of life.

Careful assessment is required before embarking on a course of medical or surgical therapy for fistula. Perianal fistulas, in particular, may be simple and superficial, or may be deep, traversing the pelvic floor musculature, and complex. Other complicating factors that need to be defined include the presence of anal stricture or abscess. Proper evaluation of perianal abscess starts with physical examination to note the location and number of draining abscesses. Most importantly, the presence of tenderness, and possibly fluctuance, should be a clue to underlying inadequately treated abscess. The Park's classification system has been used to describe the course of perianal fistulas[2]. Superficial fistulas are those which typically arise in the anorectal glands or an anal crypt and pass superficially to the external and internal anal sphincter muscles. Intersphincteric fistulas are those which pass superficially to the external anal sphincter muscle but then course through the internal anal sphincter muscle group. Trans-sphincteric fistulas are those which pass through both the external and internal anal sphincter muscle groups. Suprasphincteric fistulas are those which pass around the external sphincter muscles, but penetrate the internal sphincter muscle group, while extrasphincteric fistulas are those which completely bypass both the external and internal anal sphincters, proceeding over both muscle groups in a very deep fashion.

While this classification system is useful for colorectal surgeons in their daily practice, for the purposes of the gastroenterologist it is simply important to differentiate between the superficial fistulas and all other types. Superficial fistulas may sometimes respond to antibiotic therapy alone; however, these are most often cured by fistulotomy and healing by secondary intention. Careful validation studies have demonstrated that the most accurate diagnosis of course and complexity of perianal fistulas will include two of the following three modalities: examination under anaesthesia, endoscopic ultrasound, or magnetic resonance imaging (MRI)[7]. In practice it is usually preferable for examination under anaesthesia (EUA) to be one of the diagnostic procedures, because this also offers the opportunity for drainage of previously unsuspected abscess and placement of a non-cutting seton. Setons are non-absorbable ligatures that are threaded by the surgeon through the internal os and then out of the external os of the fistula, with the internal os end brought through the anal canal, and the two ends then being secured to make a loop. This permits constant drainage of the fistula and prevention of recurrent perianal sepsis, which has a negative impact on healing of fistulas by medical therapy.

Medical therapies thought to have utility in the treatment of fistulizing CD include antibiotics, the thiopurine analogues mercaptopurine and azathioprine, the calcineurin inhibitors cyclosporin and tacrolimus, and the anti-tumour necrosis factor alpha (TNF-α) antibodies infliximab and adalimumab. Among these agents the largest evidence base exists for the anti-TNF antibodies.

Limited data exist in support of antibiotics for healing perianal fistulas. The data in support of metronidazole for fistulizing CD include case series each ranging from eight to 34 patients studied[8–10]. One case series reported complete

healing of fistulas in approximately half of all patients who receive metronidazole either alone or in combination with other agents. However, for most patients this effect was not durable, as somewhat less than 30% of patients treated with metronidazole were successfully able to discontinue the antibiotic. A single controlled trial of 52 patients demonstrated complete closure of fistulas in 40% of patients treated with metronidazole alone[10]. In addition, two uncontrolled trials suggest that ciprofloxacin may be beneficial in perineal CD[11–13]. One study of eight patients showed improvement in physician and global patient assessments with treatment consisting of ciprofloxacin 1000–1500 mg/day over 3–12 months. Another very small study consisting of five patients with perineal CD reported that four of the five patients had resolution of perineal pain with 4 days to 5 weeks of treatment[14]. Despite this relative paucity of data for antibiotics as a primary therapy for fistulizing CD, these agents are widely used, and it is unlikely that randomized controlled trials for this indication will be completed.

Reports from cases studied in clinical trials of azathioprine and mercaptopurine have also suggested the ability of these agents to treat not only luminal disease but also fistulas[15–19]. Meta-analyses of results from randomized controlled trials suggest that 54% of patients treated with these agents will respond with complete healing or decrease in discharge from their fistulas, compared to 21% for placebo treatment[19]. It should be noted, however, that the total number of patients in the studies was small, with only 70 patients included in this analysis. Nevertheless, the results support an odds ratio in favour of azathioprine and mercaptopurine with regard to fistula healing at 4.44 (CI 1.50–13.20) compared to placebo. Of note, these agents are relatively slow in onset and are of limited utility in rapid induction of fistula closure.

In the 1980s and 1990s the possibility of using the potent immunosuppressive agent cyclosporin for CD was greeted with great enthusiasm. A number of case series were reported, in which 64 patients received intravenous cyclosporin, usually at a dose of 4 mg/kg per day[20–23]. The onset of effect was rapid, often less than 1 week. Although the initial response rate was very high, approaching 83%, it was reported that a high rate of relapse occurred upon switching to oral treatment. A similar lack of maintenance effect at lower doses has also been observed with regard to purely luminal CD[20]. Given the potent immunosuppressive effects of cyclosporin as well as the potential for renal and other toxicities, which may be severe, this agent has largely been abandoned as a treatment for CD in general and specifically for fistulizing disease.

However, the related calcineurin inhibitor tacrolimus, which has excellent oral bioavailability, has also been studied[24–27]. In 2003 the results of a randomized double-blind placebo-controlled trial were reported for tacrolimus in healing of enterocutaneous (primarily perianal) fistulas[24]. Patients were randomized to placebo or oral tacrolimus at a dose of 0.2 mg/kg per day divided twice daily. The primary endpoint was a 50% or greater reduction from baseline in the number of draining fistulas for at least 4 weeks. A total of 48 patients were treated over 10 weeks, and were randomized 1:1 to active treatment versus placebo. Overall, 43% of patients randomized to tacrolimus responded, compared to 8% of patients treated with placebo (p = 0.004). However, the short-term response came at the cost of a variety of dose-

limiting side-effects including hypertension, tremor, and renal dysfunction. It should be noted that this study did not investigate long-term closure of fistulas, and this treatment is unlikely to have a durable effect. It is expected that the toxicity of tacrolimus would be similar to that of cyclosporin and prohibitive of long-term use. Therefore, this agent is best considered as an inductive, rather than a maintenance, agent.

By far the greatest evidence base for medical therapy of fistulizing CD exists in the area of anti-TNF antibodies, in particular the chimeric anti-TNF antibody infliximab. Anecdotal reports of closure of perianal fistulas with infliximab supported the first randomized controlled trial of this agent for fistulizing CD. In 1999 Present et al. reported the first double-blind, randomized, placebo-controlled trial of infliximab for patients with fistulas visible on the skin surface, primarily perianal fistulas[28]. The primary endpoint was a fistula response, defined as lack of drainage despite gentle compression around the fistula orifices in at least 50% or more of the fistula openings. Patients were given a three-dose induction regimen of placebo or infliximab 5 or 10 mg/kg intravenously at weeks 0, 2, and 6. Of greatest interest was the secondary endpoint of closure of all fistulas, or what was called fistula remission. Overall, 55% of patients given infliximab 5 mg/kg achieved complete fistula closure, compared to 38%, giving infliximab 10 mg/kg and only 13% for patients infused with placebo. The study also demonstrated that fistula closure was not durable, with a mean duration of fistula closure of 12.3 weeks.

Accordingly, the ACCENT 2 study was undertaken to answer the following question: among patients with fistulizing CD who respond to three-dose induction therapy, is maintenance dosing with infliximab 5 mg/kg every 8 weeks superior to placebo maintenance dosing over a 1-year period of follow up[28]? This study was the largest blinded, randomized placebo-controlled study yet undertaken in fistulizing CD. A total of 195 patients, comprising 69% of those treated with open-label induction therapy of infliximab 5 mg/kg given at weeks 0, 2 and 6 intravenously, or randomized in a blinded fashion to receive infliximab 5 mg/kg intravenously every 8 weeks or placebo. Patients were followed until week 54. The primary endpoint was time to loss of response. Loss of response was defined as loss of fistula response as had been defined in the previous study, or flare of luminal disease as defined by Crohn's Disease Activity Index (CDAI) criteria, or need for rescue medication or surgery. The primary analysis demonstrated that infliximab maintenance therapy was significantly superior to placebo maintenance therapy in maintaining remission over a 54-week period. Median time to loss of response was more than 40 weeks for patients randomized to infliximab maintenance compared to 14 weeks for those randomized to placebo maintenance. A major secondary endpoint demonstrated that, among responders, complete fistula response was maintained through week 54 in 36% maintained on infliximab compared to only 19% maintained on placebo ($p = 0.009$). These results demonstrate the possibility of durable closure and clinical improvement for a subset of patients with fistulizing CD.

A variety of other important observations came from the ACCENT 2 study. These included the observation that patients who did not respond to induction

therapy were unlikely to respond to continued infliximab 5 mg/kg given every 8 weeks (18% versus 10% among non-responders randomized to receive placebo infusion every 8 weeks)[29]. Patients who initially responded, and who were maintained on infliximab 5 mg/kg every 8 weeks, but who later lost their response, were also demonstrated to regain response more than half the time (57%) when the dose was increased to 10 mg/kg[29]. It was found that perianal fistulas, which comprised the majority of fistulas studied, were most likely to close (97.2% of perianal fistulas closed at any time during the study) compared to abdominal wall fistulas (79.5% closure at any time) or rectovaginal fistulas (64.0% closure at any time)[30]. Finally, an additional analysis demonstrated that infliximab maintenance therapy was associated with reduced rates of hospitalization and surgery[31]. This may lead to improved long-term outcomes and reduced costs[32].

Subsequent studies have demonstrated the benefit of achieving control of perianal sepsis before attempting treatment with anti-TNF antibodies. Patients who have an examination under anaesthesia and seton placement have a higher initial response rate (100%), a lower recurrence rate (44%), and a longer time to recurrence of fistula (13.5 months) compared to patients who did not have examination under anaesthesia prior to beginning infliximab (82.6%, 79%, and 3.6 months; $p = 0.014$, 0.001, 0.0001, respectively)[33]. Another study compared rate of closure of perianal fistulas in patients receiving inflixlimab 5 mg/kg at weeks 0, 2 and 6 who were randomized to receive placebo or ciprofloxacin as concomitant therapy[34]. At week 12, fistula closure rate in the ciprofloxacin group was 91%, compared to 62% in the placebo group. By week 16 the fistula closure rate remained superior in the ciprofloxacin group: 73% versus 39%. These studies suggest that response to anti-TNF antibodies is more likely, as well as more durable, with control of the septic complications of fistulas.

Additional data suggest that other anti-TNF antibodies may be successful in healing fistulas. Data from the CHARM study, a blinded, randomized, placebo-controlled study of adalimumab maintenance therapy, suggests that this human anti-TNF antibody is effective in reducing draining fistulas[35]. In a pre-specified secondary analysis of 123 patients enrolled with draining fistulas, 30% of patients randomized to adalimumab maintenance therapy (33% randomized to 40 mg subcutaneously every other week (EOW) and 28% randomized to 40 mg subcutaneously every week (EW)) had complete healing at week 26, compared to 13% maintained on placebo ($p = 0.04$ for adalimumab versus placebo). At week 56, complete healing rates were similar: 33% for both adalimumab groups combined (37% for 40 mg EOW, 30% for EW), compared to 13% for placebo ($p = 0.02$ for adalimumab versus placebo). Although this represents a subgroup analysis from a larger study and not a dedicated randomized controlled trial for fistulizing CD, these outcomes are similar to those observed with infliximab. At present, fistula response rates have not been reported for a third anti-TNF antibody, certolizumab pegol.

Other medical therapies have a relatively small body of evidence in fistulizing CD, but may be useful should the previously mentioned agents fail. These include methotrexate[36], mycophenolate mofetil[37], sargramostim (granulocyte–monocyte colony-stimulating factor, or GM-CSF)[38], octreotide[39], thalidomide[40], and hyperbaric oxygen[41]. With the relative paucity of data in

support of the efficacy of these agents in healing fistulas, these should be used infrequently if at all.

An evidence-based algorithm for the treatment of perianal fistulizing CD can be described[2]. A diagnostic evaluation should be undertaken in order to understand the course and complexity of the fistulas, and to exclude the presence of abscess. Appropriate modalities may include endoscopic ultrasound, MRI, or examination under anaesthesia. All foci of abscess should be adequately drained before immune-suppressing therapies are begun. A seton should generally be placed by the surgeon in complex fistulas. Simple fistulas should be treated with antibiotics and, if this is not successful, fistulotomy should be done. Complex fistulas will probably require the addition of an immune-suppressing agent. Azathioprine may be tried first, if sufficient time is available to wait for response to this slow-acting agent. However, many patients will not have healing of their fistula until infliximab is added to the regimen. Induction therapy, with a dose of 5 mg/kg intravenously given at weeks 0, 2 and 6, should induce a fistula response in about two-thirds of patients over the first 2 months of treatment. Successful response should be followed by maintenance dosing every 8 weeks. Currently available data would support the use of adalimumab either as an alternative to infliximab, or to be used in the event of loss of response to infliximab. Should the patient fail to respond to anti-TNF antibody, treatment with tacrolimus may be considered; however, successful short-term induction is unlikely to be maintained with tacrolimus alone, which has a poor safety profile at high doses.

In summary, the treatment of fistulizing CD remains challenging. Optimal approaches incorporate careful evaluation, and an aggressive approach that judiciously includes effective elements of medical and surgical therapy timed appropriately and targeted to patients with correct indications.

References

1. Schwartz DA, Loftus EF Jr, Tremaine WJ et al. The natural history of fistulizing Crohn's disease in Olmsted County, Minnesota. Gastroenterology. 2002;122:875–80.
2. Schwartz DA, Pemberton JH, Sandborn WJ. Diagnosis and treatment of perianal fistulas in Crohn disease. Ann Intern Med. 2001;135:906–18.
3. Glass RE, Ritchie JK, Lennard-Jones JE, Hawley PR, Todd IP. Internal fistulas in Crohn's disease. Dis Col Rectum. 1985;28:557–61.
4. Margolin M, Korelitz B. Management of bladder fistula in Crohn's disease. J Clin Gastroenterol. 1989;11:399–402.
5. Greenstein A, Present D, Sachar D et al. Gastric fistulas in Crohn's disease. Report of cases. Dis Colon Rectum. 1989;32:888–92.
6. Solem CA, Loftus EV Jr, Tremaine WJ, Pemberton JH, Wolff BG, Sandborn WJ. Fistulas to the urinary system in Crohn's disease: clinical features and outcomes. Am J Gastroenterol. 2002;97:2300–5.
7. Schwartz DA, Wiersema MJ, Dudiak KM et al. A comparison of endoscopic ultrasound, magnetic resonance imaging, and exam under anesthesia for evaluation of Crohn's perianal fistulas. Gastroenterology. 2001;121:1064–72.
8. Bernstein LH, Frank MS, Brandt LJ, Boley SH. Healing of perineal Crohn's disease with metronidazole. Gastroenterology. 1980;79:357–65.
9. Brandt LJ, Bernstein LH, Boley SJ, Frank MS. Metronidazole therapy for perineal Crohn's disease: a follow-up study. Gastroenterology. 1982;83:383–7.
10. Jakobovits J, Schuster MM. Metronidazole therapy for Crohn's disease and associated fistulae. Am J Gastroenterol. 1984;79:533–40.

11. Turunen U, Farkkila M, Seppala K. Long-term treatment of peri-anal or fistulous Crohn's disease with ciprofloxacin. Scand J Gastroenterol. 1989;24(Suppl. 48):144.
12. Wolf J. Ciprofloxacin may be useful in Crohn's disease. Gastroenterology. 1990;98:A212 (Abstract).
13. Solomon M, McLeod R, O'Connor B, Steinhart A, Greenberg G, Cohen Z. Combination ciprofloxacin and metronidazole in severe perineal Crohn's disease. Can J Gastroenterol. 1993;7:571–3.
14. Dejaco C, Harrer M, Waldhoer T, Miehsler W, Vogelsang H, Reinisch W. Antibiotics and azathioprine for the treatment of perianal fistulas in Crohn's disease. Aliment Pharmacol Ther. 2003;18:1113–20.
15. Present DH, Korelitz BI, Wisch N, Glass JL, Sachar DB, Pasternack BS. Treatment of Crohn's disease with 6-mercaptopurine: a long-term randomized double blind study. N Engl J Med. 1980;302:981–7.
16. Jeshion WC, Larsen KL, Jawad AF et al. Azathioprine and 6-mercaptopurine for the treatment of perianal Crohn's disease in children. J Clin Gastroenterol. 2000;30:294–8.
17. Korelitz BI, Present DH. Favorable effect of 6-mercaptopurine on fistulae of Crohn's disease. Dig Dis Sci. 1985;30:58–64.
18. O'Brien J, Bayless T, Bayless J. Use of azatioprine in the treatment of Crohn's disease. Gastroenterology. 1991;101:39–46.
19. Pearson DC, May GR, Fick GH, Sutherland LR. Azathioprine and 6-mercaptopurine in Crohn disease. A meta-analysis. Ann Intern Med. 1995;123:132–42.
20. McDonald JW, Feagan BG, Jewell D, Brynskov J, Stange EF, Macdonal JK. Cyclosporine for induction of remission in Crohn's disease. Cochrane Database Syst Rev. 2005;18: CD000297.
21. Hanauer SB, Smith MB. Rapid closure of Crohn's disease fistulas with continuous intravenous cyclosporine A. Am J Gastroenterol. 1993;88:627–30.
22. Present DH, Lichtiger S. Efficacy of cyclosporine in treatment of fistula of Crohn's disease. Dig Dis Sci. 1994;39:374–80.
23. Egan U, Sandborn WJ, Tremaine WJ. Clinical outcome following treatment of refractory inflammatory and fistulizing Crohn's disease with intravenous cyclosporine. Am J Gastroenterol. 1998;93:442–8.
24. Sandborn WJ, Present DH, Isaacs KL et al. Tacrolimus for the treatment of fistulas in patients with Crohn's disease: a randomized, placebo-controlled trial. Gastroenterology. 2003;125:380–8.
25. Sandborn WJ. Preliminary report on the use of oral tacrolimus (FK506) in the treatment of complicated small bowel and fistulizing Crohn's disease. Am J Gastroenterol. 1997;92:876–9.
26. Lowry PW, Weaver AL, Tremaine WJ, Sandborn WJ. Combination therapy with oral tacrolimus (FK506) and azathioprine or 6-mercaptopurine for treatment-refractory Crohn's disease perianal fistulzae. Inflamm Bowel Dis. 1999;5:239–45.
27. Ierardi E, Principi M, Rendina M et al. Oral tacrolimus (FK506) in Crohn's disease complicated by fistulae of the perineum. J Clin Gastroenterol. 2000;30:200–2.
28. Present DH, Rutgeerts P, Targan S et al. Infliximab for the treatment of fistulae in patients with Crohn's disease. N Engl J Med. 1999;340:1398–405.
29. Sands BE, Anderson FH, Bernstein CN et al. Infliximab maintenance therapy for fistulizing Crohn's disease. N Engl J Med. 2004;350:876–85.
30. Sands BE, Blank MA, Patel K, van Deventer SJH. Long-term treatment of rectovaginal fistulas in Crohn's disease: response to infliximab in the ACCENT II study. Clin Gastroenterol Hepatol. 2004;2:912–20.
31. Lichtenstein GR, Yan S, Bala M, Blank M, Sands B. Infliximab maintenance treatment reduces hospitalizations, surgeries, and procedures in fistulizing Crohn's disease. Gastroenterology. 2005;128:862–9.
32. Arseneau, KO, Cohn, SM, Cominelli, F, Connors, AF. Cost-utility of initial medical management for Crohn's disease perianal fistulae. Gastroenterology. 2001;120:1640–56.
33. Regueiro M. Mardini H. Treatment of perianal fistulizing Crohn's disease with infliximab alone or as an adjunct to exam under anesthesia with seton placement. Inflamm Bowel Dis. 2003;9:98–103.

34. West RL, van der Woude CJ, Hansen BE et al. Clinical and endosonographic effect of ciprofloxacin on the treatment of perianal fistulae in Crohn's disease with infliximab: a double-blind placebo-controlled study. Aliment Pharmacol Ther. 2004;20:1329–36.
35. Schwartz D, Rutgeerts P, Colombel JF et al. Induction, maintenance, and sustainability of the healing of draining fistulas in patients with Crohn's disease treated with adalimumab: results of the CHARM Study. Am J Gastroenterol. 2006;101(Suppl. 2):S458.
36. Mahadevan U, Marion JF, Present DH. Fistula response to methotrexate in Crohn's disease: a case series. Aliment Pharmacol Ther. 2003;18:1003–8.
37. Wenzl HH, Hinterleitner TA, Aichbichler BW, Fickert P, Petritsch W. Mycophenolate mofetil for Crohn's disease: short-term efficacy and long-term outcome. Aliment Pharmacol Ther 2004;19:427–34.
38. Korzenik JR, Dieckgraefe BK, Valentine JF, Hausman DF, Gilbert MJ. Sargramostim for active Crohn's disease. N Engl J Med. 2005;352:2193–201.
39. Lavy A, Yasin K. Octreotide for enterocutaneous fistulas of Crohn's disease. Can J Gastroenterol. 2003;17:555–8.
40. Vasiliauskas EA, Kam LY, Abreu-Martin MT et al. An open-label pilot study of low-dose thalidomide in chronically active, steroid-dependent Crohn's disease. Gastroenterology. 1999;117:1278–87.
41. Noyer CM, Brandt LJ. Hyperbaric oxygen therapy for perineal Crohn's disease. Am J Gastroenterol. 1999;94:318–21.

26
Refractory ulcerative colitis: the conservative approach

S. ODES

INTRODUCTION

In considering the conservative approach to refractory ulcerative colitis (UC) the following topics will be addressed: prognostic factors, corticosteroids, companion treatments for patients on corticosteroids (5-aminosalicylates, thiopurine analogues, methotrexate), antibiotics and hyperalimentation. Cyclosporin, biologics and surgery will be discussed in other chapters.

PROGNOSTIC FACTORS IN SEVERE UC

A severe attack of UC remains a potential threat to life. Furthermore, of the 50% of UC cases expected to relapse once a year, some will be serious enough to require admission to hospital and intravenous medications, usually corticosteroids. Information about prognostic variables, available at admission, will identify patients who are likely to respond to intravenous corticosteroids, or hasten the introduction of non-corticosteroid medical therapies or surgery in those individuals who are unlikely to remit when treated with corticosteroids. Failure of intravenous corticosteroid therapy can be defined nowadays as death, surgery within 30 days, or introduction of cyclosporin or infliximab.

Before proceeding with the treatment options in severe UC, it is worth reviewing what we actually know about the factors that prognosticate a bad outcome in these unfortunate patients. Truelove and Witts in 1955 defined severe UC by the following criteria which have stood the test of time: >6 bloody stools per day, fever $>37.8^{\circ}$C, pulse rate >90/min, haemoglobin <10.5 g/dl, ESR >30 mm/h[1]. In 10% of cases the first attack of UC will fulfil the criteria of severe UC[2]. A colonic diameter >55 mm (or 90 mm for the caecum) or mucosal islands by abdominal X-ray on any day of admission is a poor prognostic factor. Continuing tachycardia and fever after 24 h on treatment with corticosteroids signified a high failure rate with this therapy[3]. More recently, in a prospective study, a stool frequency of $\geqslant 8$ or a C-reactive

protein (CRP) of >45 on day 3 of admission was shown to foretell an 85% likelihood of colectomy occurring during that admission[4]. A short duration of illness prior to admission, as well as prior corticosteroid use, forecast a poor response to medical therapy[5]. It is logical to presume that the degree of colonic involvement and the grade of inflammation will impact on the outcome of therapy. Sigmoidoscopy assists in assessing the severity of colonic inflammation, and colonoscopy (often partial) has been used successfully in the acute setting. Patients with severe colitis at colonoscopy demonstrated deep extensive ulcerations in 97%, large mucosal abrasions and mucosal detachment; the estimated time to colectomy in these patients was significantly shorter than in those without such lesions[6].

UC is a multifaceted disease entity which has been classified recently into clinical phenotypes based on disease extent (E1, E2, E3) and severity (S0, S1, S2, S3), this being the Montreal Classification of Ulcerative Colitis[7]. Patients with E3 (extensive disease, proximal to splenic flexure, or pancolitis) are more likely to require corticosteroids and have a higher actuarial risk for colectomy than those with E2 (left-sided, distal to the splenic flexure) and E1 (proctitis). Furthermore, the disease severity classification reliably predicts the clinical course in the short term, where S0 implies remission, S1 is mild, S2 moderate, and S3 severe disease. Note that the severity classification follows closely the Truelove–Witts scale[1] and the latest guidelines of the American College of Gastroenterology[8]. Present knowledge of the age phenotype in UC is insufficient to allow its use as a prognostic variable in the Montreal Classification[7].

ROLE OF CORTICOSTEROIDS

It was Dr Sidney Truelove who pioneered the use of corticosteroids in UC. Corticosteroids have rapid and potent anti-inflammatory activity. In 1955 he showed with Witts that oral corticosteroids dramatically improved the prognosis of acute severe UC, with reduction of mortality from 24% to 7% in a controlled trial[1]. His paper with Jewell in 1974 showed conclusively that intravenous corticosteroids were highly effective in severe UC[9]. In 1978 Truelove published the results of a series of 100 treatments with intravenous corticosteroids in 87 severe UC patients[10]. Sixty-five per cent of the cases were symptom-free by day 5, 15% had partial improvement, and 25% did not respond and were sent for immediate surgery; there were no deaths. It may well be asked whether we are doing that much better today. To quote from Truelove in a 1981 state-of-the-art paper[11]: 'The importance of treating the acute episode early and efficiently is emphasized, and a differentiated therapeutic regimen according to disease activity is outlined.'

Even today, the inexpensive intravenous corticosteroid preparations remain the mainstay of therapy in acute severe UC, and are given as hydrocortisone 100 mg four times daily or methylprednisone 60 mg daily (0.4 mg/kg twice daily) for 5 days[6,12]. Lower doses of corticosteroids were not found to be effective[13]. Moreover, extending intravenous corticosteroid therapy beyond 7 days was unlikely to be successful and could result in late introduction of other

therapies or emergency surgery. The response rate to intravenous corticosteroids in UC was 60–70%[4,14]. It was likely (in the pre-infliximab era) that 80% of the corticosteroid failures would require colectomy within a decade[15]. Of note, failure to achieve full remission before tapering intravenous corticosteroids is a prescription for early relapse. In children and teenagers corticosteroid resistance appeared to be commoner than in adults[16]. This is speculated to be the result of more extensive distribution of disease in the colon and a greater genetic component in the young age group.

A cohort of 85 French patients with severe UC was admitted, subjected to colonoscopy, and treated according to a very strict protocol of intravenous methylprednisone for 9 ± 5 days, bowel rest for 7 ± 4 days, intravenous fluids and electrolytes, and parenteral nutrition[6]. In addition antibiotics, steroid enemas and low molecular weight heparin were used in up to half the patients. The outcome was 34 complete remissions, seven incomplete remissions, and 44 failures. Thirteen patients were treated with intravenous cyclosporin and 37 required emergency colectomy (including seven who had received cyclosporin) at median 12 days, range 3–37 days. By multivariate analysis the factors predicting increased risk of failure of intravenous corticosteroid therapy included the duration of attack >6 weeks (relative risk 2.1), finding of severe colonoscopic lesions (RR 2.3) and presence of Truelove–Witts criteria (RR 2.3). Patients having two or three of these variables had a 75–86% risk of failure of treatment.

An extensive review of 29 studies (time period 1974–2005, total of 1948 adult patients) of intravenous corticosteroid therapy in severe UC was published in 2007[17]. The mean dose of intravenous corticosteroids expressed as methylprednisolone equivalent, while presuming an adult weight of 70 kg, was 68 ± 13 mg/kg; in a meta-regression controlled for disease severity at baseline the dose did not correlate with the colectomy rate. The mean response rate to intravenous corticosteroid therapy was 67% (95% CI 65–69); the surgical rate was 27% (95% CI 26–29) and death rate 1% (95% CI 0.7–1.6). Notably, the colectomy rates in patients requiring intravenous corticosteroids, controlled for dose, disease severity at baseline and time to evaluation for colectomy, were unchanged over the 30-year study interval. Similarly the overall surgical rate in Danish UC patients has not diminished over 1962–2005[18]. By comparison the European Collaborative Study Group of Inflammatory Bowel Disease has recently reported a low 8.7% 10-year colectomy rate in a prospectively recruited community-based multinational UC cohort with 619 patients[19].

COMPANION TREATMENTS FOR PATIENTS ON CORTICOSTEROIDS

These therapies include the 5-aminosalicylates (5-ASA, mesalazine, mesalamine), azathioprine and 6-mercaptopurine (thiopurine analogues), and methotrexate. 5-ASA and the immunomodulators are steroid-sparing in maintenance therapy. Such adjuvant treatment may reduce the considerable morbidity of prolonged administration of corticosteroid therapy[20].

In a recent meta-analysis of nine clinical trials, 5-ASA $\geqslant 3$ g/day was twice as effective as placebo in improving acute UC, 65% versus 35%[21]. The number to

treat to achieve improvement was three (95% CI 3–5), and to achieve remission was eight (95% CI 5–20). Lower dosages were less effective. The efficacy of 5-ASA in the maintenance of remission in UC patients was found to be 56% in five placebo-controlled trials; this was significantly higher than placebo. The dose of 5-ASA varied from 1–4 g/day and the follow-up period was 6–12 months. Side-effects of 5-ASA may require stopping the drug in up to 15% of patients, but these are usually mild. Nephrotoxicity is rare, being partly idiosyncratic and partly dose-related.

Azathioprine 1.5–2.5 mg/kg per day and 6-mercaptopurine 0.75–1.5 mg/kg per day are administered for maintenance of remission in UC. Dose optimality and avoidance of serious bone marrow and hepatic toxic effects can be achieved, at least in theory, by pretreatment measurement of TPMT (thiopurine methyltransferase enzyme) activity or genotype, or the 6-thioguanine level[22]. A high 6-thioguanine level carries a serious risk of leucopenia. The TPMT level correlates inversely with the drug response. Co-administration of 5-ASA may raise 6-thioguanine levels. The addition of allopurinol to thiopurine analogue non-responders appeared to optimize 6-thioguanine production and reduce disease activity in six UC patients, with a trend towards reducing the need for corticosteroids[23]. In general azathioprine or 6-mercaptopurine is introduced during corticosteroid therapy and continued for 3–4 years in responders. The 20% of patients who relapse when the thiopurine immunomodulator is stopped can be retreated with the same medication. Surprisingly, the use of these agents in UC has been little studied. Azathioprine and 6-mercaptopurine were categorized using the RAND method as inappropriate therapy (score 5/9) for induction of remission in severe UC extending >60 cm from the anus[24]. However, azathioprine and 6-mercaptopurine were appropriate companion treatments (score 9/9) in steroid-dependent and -resistant UC extending >60 cm from the anus. Azathioprine and 6-mercaptopurine were deemed appropriate in the maintenance of remission in UC.

The problem with thiopurine analogue intolerance occurring in 10–20% of patients shifted attention to the possible use of methotrexate in the maintenance treatment of UC. An initial controlled trial of oral methotrexate in active UC gave a disappointing result[25]. A recent retrospective analysis was reported of the use of methotrexate 20 mg weekly (range 10–40 mg) in 50 UC patients in the UK, resistant to or dependent on corticosteroids, and resistant to or intolerant of azathioprine[26]. Remission was achieved in 21 patients and improvement in nine patients, with a marked drop of the CRP level; five colectomies were performed. These data intimated a possible role for methotrexate in active UC patients intolerant of thiopurines. By application of the RAND method, methotrexate was judged to be appropriate in the maintenance of remission in UC where azathioprine therapy had failed[24].

ANTIBIOTICS FOR PATIENTS ON CORTICOSTEROIDS

Since UC does not develop in germfree murine models, it is postulated that bacteria have a role in the pathogenesis of human UC; the corollary to this is

that broad-spectrum antibiotics should be given as primary or adjunctive therapy in each flare of UC. Nonetheless the benefit of antibiotics in this situation has not been substantiated in several controlled studies, so that the role of antibiotics as concomitant therapy in UC patients requiring intravenous corticosteroids remains unproven. Randomized controlled trials have shown no short-term clinical benefit of intravenous metronidazole (500 mg thrice daily) or ciprofloxacin (400 mg twice daily) in addition to standard intravenous and local corticosteroid therapy for severe UC[27,28]. The non-absorbable oral antibiotic rifaximin given as 800 mg/day for 10 days decreased the number of episodes of diarrhoea and rectal bleeding compared with placebo in severe steroid-refractory UC but offered no advantage in altering the clinical outcome of disease[29]. Trials of tobramycin have generally yielded poor results. Recent guidelines of the British Society of Gastroenterology have restricted the use of intravenous antibiotics in active severe UC to those patients in whom infection is suspected prior to surgery[12]. Such a situation may arise with fulminant UC and toxic megacolon with risk of perforation. Additionally, in areas where colonic amoebiasis is rampant it may be prudent to give metronidazole at the onset of corticosteroid therapy while the results of the stool examination are awaited. In giving antibiotics to hospitalized UC patients the potentially serious complication of superimposed *Clostridium difficile* colitis should be remembered. *C. difficile* superinfection in UC is largely community-acquired, is difficult and expensive to diagnose by toxin assay, has an atypical endoscopic appearance, and may be resistant to fluoroquinolones. It is commoner in patients with colonic disease, chronic co-morbidities, and those on immunomodulator therapy[30]. The use of probiotics in severe UC has not been studied.

HYPERALIMENTATION FOR UC PATIENTS ON CORTICOSTEROIDS

The importance of maintaining a normal blood chemistry, albumin and haemoglobin in very ill UC patients cannot be overemphasized. Whether treatment should include routine total parenteral nutrition and full bowel rest is a moot point. The effect of enforced peripheral parenteral nutrition in a recent study cannot be assessed since this parameter was not subjected to univariate analysis[6]. There is still no evidence that total parenteral nutrition benefits the acutely ill UC patient except as supplementary therapy or prior to surgery in a moribund individual, and the procedure carries a high risk of sepsis.

CONCLUSIONS

Intravenous corticosteroids are the mainstay of conventional treatment of acute severe UC but a considerable proportion of patients will be found to be steroid-resistant. Companion treatments have limited value in altering the clinical course in such patients, indicating a real need for giving cyclosporin and particularly biologics to these very ill patients.

References

1. Truelove SC, Witts LJ. Cortisone in ulcerative colitis: final report on a therapeutic trial. Br Med J. 1955;2:1041–8.
2. Cima RR, Penberton JH. Surgical management of ulcerative colitis. In: Sartor RB, Sandborn WJ, editors. Kirsner's Inflammatory Bowel Diseases, 6th edn. London: Saunders, 2004;603–13.
3. Lennard-Jones JE, Ritchie JK, Hilder W et al. Assessment of severity in colitis: a preliminary study. Gut. 1975;16:579–84.
4. Travers SPL, Farrant JM, Ricketts C et al. Predicting outcome in severe ulcerative colitis. Gut. 1996;38:905–10.
5. Lindgren SC, Flood LM, Kilander AF et al. Early predictors of glucocorticosteroid treatment failure in severe and moderately severe attacks of ulcerative colitis. Eur J Gastroenterol Hepatol. 1998;10:831–5.
6. Carbonnel F, Gargouri D, Lemann M et al. Predictive factors of outcome of intensive intravenous treatment for attacks of ulcerative colitis. Aliment Pharmacol Ther. 2000;14: 273–9.
7. Silverberg MS, Satsangi J, Ahmad T et al. Toward an integrated clinical, molecular and serological classification of inflammatory bowel disease. Report of a Working Party of the 2005 Montreal World Congress of Gastroenterology. Can J Gastroenterol. 2005;19(Suppl. A):5–36.
8. Kornbluth A, Sachar DB. Ulcerative colitis practice guidelines in adults (update): American College of Gastroenterology, Practice Parameters Committee. Am J Gastroenterol. 2004;99:1371–85.
9. Truelove SC, Jewell DP. Intensive intravenous regimen for severe attacks of ulcerative colitis. Lancet. 1974;1:1067–70.
10. Truelove SC, Willoughby CP, Lee EG, Kettlewell MG. Further experience in the treatment of severe attacks of ulcerative colitis. Lancet. 1978;2:1086–8.
11. Truelove SC. The treatment of ulcerative colitis. Schweiz Med Wochenschr. 1981;111:1342–6.
12. Carter MJ, Lobo AJ, Travis SLP. Guidelines for the management of inflammatory bowel disease. Gut. 2004;53(Suppl. V):V1–16.
13. Rosenberg W, Ireland A, Jewell DP. High-dose methylprednisolone in the treatment of active ulcerative colitis. J Clin Gastroenterol. 1990;12:40–1.
14. Faubion WA, Loftus EV, Hannsen WS, Zinsmeister AR, Sandborn WJ. The natural history of corticosteroid therapy for inflammatory bowel disease; a population-based study. Gastroenterology. 2001;121:255–60.
15. Bojic D, Al-Ali M, Jewel DP et al. Pattern and outcome of severe ulcerative colitis. Gut. 2005;54(Suppl. VIII):A155.
16. Tung J, Loftus EV, Freese D et al. A population-based study of the frequency of corticosteroid resistance and dependence in pediatric patients with Crohn's disease and ulcerative colitis. Inflamm Bowel Dis. 2006;12:1093–100.
17. Turner D, Walsh CM, Steinhart AH, Griffiths AM. Response to corticosteroids in severe ulcerative colitis: a systematic review of the literature and a meta-regression. Clin Gastroenterol Hepatol. 2007;5:103–10.
18. Jess T, Riis L, Vind I et al. Changes in clinical characteristics, course, and prognosis of inflammatory bowel disease during the last 5 decades: a population-based study from Copenhagen, Denmark. Inflamm Bowel Dis. 2007;13:481–9.
19. Hoie O, Wolters FL, Riis L et al. Low colectomy rates in ulcerative colitis in an unselected European cohort followed for 10 years. Gastroenterology. 2007;132:507–15.
20. Stein RB, Hanauer SB. Comparative tolerability of treatments for inflammatory bowel disease. Drug Saf. 2000;23:429–48.
21. Bebb JR, Scott BB. Systematic review: how effective are the usual treatments for ulcerative colitis? Aliment Pharmacol Ther. 2004;20:143–9.
22. Aberra FN, Lichtenstein GR. Review article: Monitoring of immunomodulators in inflammatory bowel disease. Aliment Pharmacol Ther. 2005;21:307–19.
23. Sparrow MP, Hande SA,Friedman S, Cao D, Hanauer SB. Effect of allopurinol on clinical outcomes in inflammatory bowel disease nonresponders to azathioprine or 6-mercaptopurine. Clin Gastroenterol Hepatol. 2007;5:209–14.

24. Caprilli R, Angelucci E, Cocco A et al. Appropriateness of immunosuppressive drugs in inflammatory bowel diseases assessed by the RAND method: Italian group for IBD (IG-IBD) position statement. Dig Liver Dis. 2005;37:407–17.
25. Oren R, Arber N, Odes S et al. Methotrexate in chronic active ulcerative colitis: a double-blind, randomized, Israeli multicenter trial. Gastroenterology. 1996;110:1416–21.
26. Cummings JRF, Herrlinger KR, Travis SPL, Gorard DA, McIntyre AS, Jewell DP. Oral methotrexate in ulcerative colitis. Aliment Pharmacol Ther. 2005;21:385–9.
27. Chapman RW, Selby WS, Jewell DP. Controlled trial of intravenous metronidazole as an adjunct in severe ulcerative colitis. Gut. 1986;27:1210–12.
28. Mantzaris GJ, Pettraki K, Archavlis E et al. A prospective randomized controlled trial of intravenous ciprofloxacin in severe ulcerative colitis. Scand J Gastroenterol. 2001;36:971–4.
29. Gionchetti P, Rizzello F, Ferrieri A. Rifaximin in patients with moderate or severe ulcerative colitis refractory to steroid treatment: a double-blind, placebo-controlled trial. Dig Dis Sci. 1999;44:1220–1.
30. Tremaine WJ. Inflammatory bowel disease and *Clostridium difficile*-associated diarrhea: a growing problem. Clin Gastroenterol Hepatol. 2007;5:310–11.

27
Refractory ulcerative colitis: the aggressive approach

W. J. SANDBORN

INTRODUCTION

Patients with refractory ulcerative colitis (UC) can be broadly classified into two categories: (1) outpatients; or (2) in-hospital patients. Outpatients with refractory UC would typically be defined as patients failing oral corticosteroids and azathioprine or 6-mercaptopurine. In-hospital patients with refractory UC would typically be defined as patients failing intravenous corticosteroids. This chapter will review the aggressive medical treatment options for patients with refractory UC.

TREATMENT OF OUTPATIENTS WITH REFRACTORY UC

The requirements for a medication to treat outpatients with refractory UC include: (1) the need to act reasonably quickly (probably within 1–2 weeks); (2) efficacy for both induction and maintenance of response and remission; and (3) an acceptable safety profile. Cyclosporin and tacrolimus do not meet these criteria because of their unfavourable long-term safety profile. Infliximab (chimeric anti-tumour necrosis factor (TNF) antibody) does meet all of these criteria. The investigational visilizimab (humanized anti-CD3 antibody) does not meet these criteria because maintenance therapy with this agent would result in prolonged T-cell depression which presumably would lead to toxicity from opportunistic infection, malignancy, etc.

Infliximab

Two large clinical trials, the ACT I and ACT II trials, have been conducted to evaluate infliximab for both induction and maintenance of response and remission in outpatients with refractory UC[1]. Patients received induction therapy with infliximab 5 mg/kg or placebo at weeks 0, 2, and 6. At week 8 the clinical response rates in the ACT I trial were 69% for infliximab 5 mg/kg versus 37% for placebo, and in the ACT II trial they were 65% for infliximab 5 mg/kg versus 29% for placebo. Similarly, the remission rates at week 8 in the

ACT I trial were 39% for infliximab 5 mg/kg versus 15% for placebo, and in the ACT II trial they were 34% for infliximab 5 mg/kg versus 6% for placebo. Patients were then subsequently treated with maintenance therapy with infliximab 5 mg/kg or placebo every 8 weeks for 54 weeks in the ACT I trial and 30 weeks in the ACT II trial. The week 54 clinical response rates in the ACT I trial were 46% for infliximab 5 mg/kg versus 20% for placebo and the week 30 clinical response rates in the ACT II trial were 47% for infliximab 5 mg/kg versus 26% for placebo. The clinical remission rates at week 54 in the ACT I trial were 35% for infliximab 5 mg/kg versus 17% for placebo, in the ACT II trial they were 26% for infliximab 5 mg/kg versus 11% for placebo. Mucosal healing (endoscopic healing) occurred at week 54 in the ACT I trial in 46% of patients treated with infliximab 5 mg/kg versus 18% of patients treated with placebo, and at week 30 in the ACT II trial in 46% of patients treated with infliximab 5 mg/kg versus 30% of patients treated with placebo. The median C-reactive protein concentration in patients who received infliximab dropped very rapidly with a maximal effect observed at the first measured time point – week 2. Similarly, in the ACT I trial, the clinical components of the Mayo score (partial Mayo score composed of stool frequency, rectal bleeding, and physician global assessment (score ranges from 0 to 9)) decreased from a median score of six points at week 0 to a median score of three points at week 2 and a median score of two points at week 8. Remission and complete steroid discontinuation occurred at week 30 in the ACT I trial in 24% of patients treated with infliximab 5 mg/kg versus 10% of patients treated with placebo, and in the ACT II trial in 18% of patients treated with infliximab versus 3% of patients treated with placebo. In the ACT I trial, the rates of formation of antibodies to infliximab (ATI) were somewhat greater in patients who received monotherapy with infliximab 5 mg/kg as compared to patients who received combination therapy with infliximab plus immunomodulator therapy with azathioprine or 6-mercaptopurine (20% versus 0%)[2]. In the ACT II trial, 15% of patients who received monotherapy with infliximab 5 mg/kg developed ATI compared to 2% of patients who received combination therapy with infliximab plus an immunomodulator[2]. However, these modest differences in the rates of ATI formation did not impact clinical efficacy. In the ACT I trial, at week 54 the clinical response rate was 45% for both patients who received monotherapy with infliximab 5 or 10 mg/kg and patients who received combination therapy with infliximab plus an immunomodulator[3]. Similarly, in the ACT II trial, at week 30 the clinical response 53% in patients who received montherapy with infliximab 5 or 10 mg/kg and 55% in patients who received combination therapy with infliximab plus an immunomodulator[3].

A variety of important toxicities can occur in patients treated with infliximab, including: acute infusion reactions, delayed hypersensitivity reactions, formation of autoantibodies (such as antinuclear antibodies, anti-double-stranded DNA antibodies, and antihistone antibodies), drug-induced lupus (rare), non-Hodgkin's lymphoma (including hepatosplenic T cell lymphoma in children receiving infliximab in combination with azathioprine), possibly skin cancer, serious infections and opportunistic infections (including tuberculosis, histoplasmosis, coccidiomycosis) and demyelinating diseases such as multiple sclerosis and optic neuritis[4,5].

NOVEL THERAPIES FOR REFRACTORY UC

A variety of novel treatments are currently being evaluated for patients with refractory UC. These can be categorized as follows. Anti-tumour necrosis factor (TNF) agents including: adalimumab, certolizumab pegol, and golimumab. Anti-selective adhesion molecules including the anti-intergrin antibodies (anti-α4 integrin (natalizumab), anti-α4β7 integrin (MLN-002), anti-β7 integrin, anti-MAdCAM-1, and anti-αEβ7 integrin) and anti-sense to ICAM-1 (alicaforsen)[6–10]. Anti-interleukin-2 receptor (anti-CD25) antibodies including basiliximab and daclizumab (failed)[11–13]. The anti-CTLA-4 fusion protein abatacept is currently in clinical trials. A phase III study with apheresis has been completed; the results are pending. The humanized anti-CD3 antibody visilizumab is currently in clinical trials[14,15].

TREATMENT OF IN-HOSPITAL PATIENTS WITH REFRACTORY UC

The requirements for a medication to treat in-hospital patients with moderate to severely active UC refractory to intravenous corticosteroids include: (1) the need to act very quickly (probably within 4–7 days); (2) efficacy for induction of response and remission; (3) an acceptable safety profile. It would be ideal also to have a maintenance effect, but this is not mandatory since a rapidly acting agent could be used as a bridge to maintenance therapy with a second agent. Cyclosporin, tacrolimus, and infliximab do meet all of these criteria. The investigational agent visilizumab (humanized anti CD3 antibody) also appears to have a rapid onset of action and to induce response and remission. The safety profile of visilizumab is still being characterized.

Cyclosporin

The initial placebo-controlled trial with intravenous cyclosporin 4 mg/kg administered as a continuous intravenous infusion was performed in 20 patients[16]. At week 2 a clinical response occurred in 82% of patients treated with intravenous cyclosporin compared to 0% of patients treated with placebo[16]. Remission rates were not reported. The disease activity index which was used to measure disease activity was the Lichtiger score (ranges from 0 to 21)[16]. In the patients who received cyclosporin the Lichtiger score decreased from a median score of 13 points at week 0 to a median score of 6 points at week 2 as compared to patients who received placebo, in whom no significant change occurred (median Lichtiger score decreased from 14 points at week 0 to 13 points at week 2)[16]. Subsequently another trial was performed in 30 patients who were being hospitalized for severe UC[17]. Patients were randomized to receive initial therapy with continuous infusion intravenous cyclosporin 4 mg/kg as a monotherapy or intravenous corticosteroids as a monotherapy[17]. After 7 days of treatment a clinical response occurred in 64% of patients treated with cyclosporin and 53% of patients treated with placebo[17]. Remission rates were not reported. A third study evaluated the dose response of intravenous cyclosporin in patients who were being hospitalized for severe UC[18]. Patients

were randomized to treatment with continuous infusion intravenous cyclosporin 4 mg/kg or 2 mg/kg as a monotherapy. The response rate at day 8 was 84% in the cyclosporin 4 mg/kg group and 86% in the cyclosporin 2 mg/kg group.

Despite the impressive results reported with cyclosporin in the short term, in the long term most patients relapse after cyclosporin is discontinued[19,20]. At the University of Leuven 142 patients with severe acute UC have received intravenous cyclosporin, of whom 118 (83%) responded[19]. Forty-four of these patients (31%) were already receiving maintenance azathioprine at the time of the relapse that required treatment with cyclosporin whereas 74 patients (52%) were started on azathioprine for the first time[19]. At 1 year, 52% of patients who were already receiving azathioprine, and failing it at the time cyclosporin was initiated, required colectomy compared to 16% of patients who were starting azathioprine for the first time[19]. Considering the entire patient population of 142 patients, 5 years after treatment of cyclosporin only 31% of patients had avoided colectomy and only 12% of patients were still in clinical remission[19]. A similar experience was seen at Oxford University where 90% of patients had relapsed by 42 months after receiving cyclosporin and over a 7-year period 42% of patients required colectomy[20].

A variety of toxicities have been associated with cyclosporin, including headache, tremor, paraesthesias, seizures (predominantly with intravenous cyclosporin), hypertrichosis, gingival hyperplasia, renal insufficiency, hypertension, serious and opportunistic infections, hepatotoxicity, nausea and vomiting, and anaphylaxis[21,22]. At the University of Leuven, among 142 patients' treatment with intravenous cyclosporin, four patients died (2.8% mortality rate) from causes probably associated with cyclosporin, including one patient who died from *Pneumocystis carinii* pneumonia, two patients who died from systematic aspergillosis, and one patient who died from chronic mylegenous leukaemia[19].

Tacrolimus

One controlled trial has been performed with tacrolimus in patients with severely active UC[23]. Sixty-three patients were hospitalized for moderate to severely active UC that was steroid-dependent or steroid-refractory. Stable doses of 5-aminosalicylates were continued. Patients were randomized to receive oral tacrolimus at a high dose (target trough concentration of 10–15 ng/ml) or a low dose (target trough concentration of 5–10 mg/ml) or placebo for 14 days. Disease activity was measured with the Mayo score. The primary endpoint for the study was the percentage of patients who experienced improvement defined as a complete response (Mayo score = 0) or partial response (decrease from baseline in the Mayo score $\geqslant$4 points) at day 14. Improvement occurred at day 43 in 68% of patients treated with high-dose tacrolimus, 38% of patients received low-dose tacrolimus, and 10% of patients treated with placebo[23]. Secondary endpoints included clinical remission (absolute Mayo score $\leqslant$2 points) at day 14 and mucosal healing (decrease in the Mayo score endoscopy sub-score from 2–3 points at baseline to 0–1 points at day 14). Clinical remission occurred in 20% of patients who received high-

dose tacrolimus, 11% of patients who received low-dose tacrolimus, and 6% of patients who received placebo[23]. Mucosal healing occurred in 79% of patients who received high-dose tacrolimus, 44% of patients who received low-dose tacrolimus, and 13% of patients who received placebo[23]. Nephrotoxicity was reported in 5% of patients in the high-dose tacrolimus group, 5% of patients of patients in the low-dose tacrolimus group and none in the patients treated with the placebo[23].

Infliximab

Both placebo-controlled and corticosteroid-positive controlled trials have been performed with infliximab for the treatment of severe UC in the in-hospital setting. Sands and colleagues randomized 11 patients with severe steroid-refractory UC to infliximab or placebo and reported a 50% clinical response to infliximab at week 2 compared to 0% of patients treated with placebo[24]. Jarnerot et al. reported that 71% of patients with severe, steroid-refractory UC treated with infliximab were able to avoid colectomy by day 90 compared to only 33% of placebo-treated patients[25]. Ochsenkuhn and colleagues compared intravenous infliximab monotherapy to intravenous steroid monotherapy in patients with severe UC who were initially being hospitalized[26]. At week 3, and again at week 13, 83% of patients responded to infliximab and 86% of patients responded to corticosteroids[26]. Finally, Armuzzi et al. compared intravenous infliximab monotherapy to intravenous steroid monotherapy in patients with severe UC who were initially being hospitalized[27]. They reported 100% response in both the infliximab- and steroid-treated patients at week 2[27].

Visilizumab

Visilizumab is a humanized anti-CD3 antibody. The prototype antibody in this class is the murine anti-CD3 antibody OKT3. OKT3 is commonly associated with cytokine release syndrome mediated both by inhibition of the CD3 T-cell receptor and by the interaction between the Fc receptor on antigen-presenting cells and the Fc portion of the antibody. The Fc receptor-mediated T-cell activation contributes to the toxicity of OKT3 but is not required for its immunosuppressive activities. Visilizumab also targets the CD3, but has been engineered so that it does not bind the Fc receptor. *In vitro*, visilizumab causes apoptosis of activated but not resting T cells. Plevy and colleagues reported a phase I study of visilizumab in 32 patients with severe UC who were refractory to at least 5 days of intravenous corticosteroids[14]. Patients received visilizumab at a dose of either 10 μg/kg or 15 μg/kg administered as an intravenous bolus on two consecutive days. Response was defined as a decrease from baseline in the modified Truelove and Witts Severity Index (Lichtiger score) of $\geqslant 3$ points to an absolute score of <10 points. Remission was defined as an absolute modified Truelove and Witts Severity Index score of <4 points. At day 60, 60–80% of patients had experienced a clinical response and 20–35% had experienced a clinical remission[14]. Significant decreases from week 0 in the total Mayo score and the endoscopy subscore were also observed. When these patients were followed over time, the median time to a requirement for salvage

therapy was greater than 1 year. These preliminary phase I data suggest that visilizumab may be a rapidly acting induction agent with efficacy in this clinical setting.

A second phase I/II trial was performed in 76 patients with severe UC again refractory to 5 days of intravenous corticosteroids[15]. Patients were stratified according to whether or not they had detectable Epstein–Barr virus DNA copies in the blood. Patients with undetectable Epstein–Barr virus DNA were randomized to visilizumab 5, 7.5, 10, or 12.5 μg/kg administered as an intravenous and bolus on two consecutive days[15]. Patients with Epstein–Barr virus present at $\leqslant$5000 copies/ml were treated in a dose escalation regimen with visilizumab 5, 7.5, 10, or 12.5 μg/kg[15]. Response was defined as a modified Truelove and Witts Severity Index (Lichtiger) score of <10 points and a reduction from baseline $\geqslant 3$ points[15]. Remission was defined as a modified Truelove and Witts Severity Index (Lichtiger) score of <4 points[15]. High proportions of clinical response and clinical remission were observed with all four visilizumab doses, with the best numeric results being seen in the lowest dose group, 5 μg/kg. Significant reductions in the median Mayo score from week 0 to week 30 were seen with all four dose groups[15]. Similarly, approximately 15–50% of patients in the various dose groups were in endoscopic remission at day 30, with the best results being seen in patients who received the lowest visilizumab dose of 5 μg/kg[15].

Adverse events associated with visilizumab consist primarily of cytokine release syndrome symptoms. These include chills, nausea, headache, fever, arthralgias, vomiting and myalgias[14,15].

CONCLUSIONS

For outpatients who are failing corticosteroids and azathioprine or 6-mercaptopurine, at the present time, the only agent available that works quickly and is effective for both induction and maintenance is infliximab. For in-hospital patients who are failing intravenous corticosteroids, cyclosporin, tacrolimus, and infliximab all have a rapid onset of action and are effective for induction of clinical response. Only infliximab is effective for maintenance of response and remission. The anti-CD3 antibody visilizumab has substantial promise as a rapid induction agent but is still in clinical trials. A large number of biotechnology agents are likely to become available for patients with refractory UC in the future.

References

1. Rutgeerts P, Sandborn WJ, Feagan BG et al. Infliximab induction and maintenance therapy for ulcerative colitis. N Engl J Med. 2005;353:2462–76.
2. Sandborn WJ, Wagner CL, Fasanmade AA et al. Effects of immunomodulators on pharmacokinetics and immunogenicity of infliximab administered as 3-dose induction followed by systematic maintenance therapy in IBD. Gastroenterology. 2007;132(Suppl. 2):A504–5.
3. Lichtenstein GR, Diamond RH, Wagner C et al. Infliximab administered as 3-dose induction followed by scheduled maintenance therapy in IBD: comparable clinical

outcomes with or without concomitant immunomodulators. Gastroenterology. 2007;132 (Suppl. 2):A146.
4. Lichtenstein GR, Feagan BG, Cohen RD et al. Serious infections and mortality in association with therapies for Crohn's disease: TREAT registry. Clin Gastroenterol Hepatol. 2006;4:621–30.
5. Prescribing information for Remicade (infliximab). Package Insert 2006.
6. Gordon FH, Hamilton MI, Donoghue S et al. A pilot study of treatment of active ulcerative colitis with natalizumab, a humanized monoclonal antibody to alpha-4 integrin. Aliment Pharmacol Ther. 2002;16:699–705.
7. Feagan BG, Greenberg GR, Wild G et al. Treatment of ulcerative colitis with a humanized antibody to the α4β7 integrin. N Engl J Med. 2005;352:2499–507.
8. van Deventer SJ, Tami JA, Wedel MK. A randomised, controlled, double blind, escalating dose study of alicaforsen enema in active ulcerative colitis. Gut. 2004;53:1646–51.
9. Van Deventer SJH, Wedel MK, Baker BF, Xia S, Chuang E, Miner PB. A phase II dose ranging, double-blind, placebo-controlled study of alicaforsen enema in subjects with acute exacerbation of mild to moderate left-sided ulcerative colitis. Aliment Pharmacol Ther. 2006;23:1415–25.
10. Miner PB, Wedel MK, Xia S, Baker BF. Safety and efficacy of two dose formulations of alicaforsen enema compared with mesalazine enema for treatment of mild to moderate left-sided ulcerative colitis: a randomized, double-blind, active-controlled trial. Aliment Pharmacol Ther. 2006;23:1403–13.
11. Creed TJ, Norman MR, Probert CS et al. Basiliximab (anti-CD25) in combination with steroids may be an effective new treatment for steroid-resistant ulcerative colitis. Aliment Pharmacol Ther. 2003;18:65–75.
12. Creed TJ, Probert CS, Norman MN et al. Basiliximab for the treatment of steroid-resistant ulcerative colitis: further experience in moderate and severe disease. Aliment Pharmacol Ther. 2006;23:1435–42.
13. Van Assche G, Sandborn WJ, Feagan BG et al. Daclizumab, a humanized monoclonal antibody to the interleukin-2 receptor (CD25), for the treatment of moderately to severely active ulcerative colitis: a randomized, double-blind, placebo-controlled, dose-ranging trial. Gut. 2006;55:1568–74.
14. Plevy S, Salzberg B, van Assche G et al. A humanized anti-CD3 monoclonal antibody, visilizumab, for treatment of severe steroid-refractory ulcerative colitis: results of a phase I study. Gastroenterology. 2004;126:A75.
15. Baumgart DC, Hommes DW, Reinisch W et al. The phase I/II visilizumab study. A report of safety and efficacy of treament and retreatment in ulcerative colitis patients refractory to treatment with i.v. steroids (IVSR-UC). Gut. 2006;54:A57.
16. Lichtiger S, Present DH, Kornbluth A et al. Cyclosporine in severe ulcerative colitis refractory to steroid therapy. N Engl J Med. 1994;330:1841–5.
17. D'Haens G, Lemmens L, Geboes K et al. Intravenous cyclosporine versus intravenous corticosteroids as single therapy for severe attacks of ulcerative colitis. Gastroenterology. 2001;120:1323–9.
18. Van Assche G, D'Haens G, Noman M et al. Randomized, double-blind comparison of 4 mg/kg versus 2 mg/kg intravenous cyclosporine in severe ulcerative colitis [see comment]. Gastroenterology. 2003;125:1025–31.
19. Moskovitz DN, Van Assche G, Maenhout B et al. Incidence of colectomy during long-term follow-up after cyclosporine-induced remission of severe ulcerative colitis. Clin Gastroenterol Hepatol. 2006;4:760–5.
20. Campbell S, Travis S, Jewell D. Cyclosporin use in acute ulcerative colitis: a long-term experience. Eur J Gastroenterol Hepatol. 2005;17:79–84.
21. Sandborn WJ. A critical review of cyclosporine therapy in inflammatory bowel disease. Inflamm Bowel Dis. 1995;1:48–63.
22. Loftus CG, Egan LJ, Sandborn WJ. Cyclosporine, tacrolimus, and mycophenolate mofetil in the treatment of inflammatory bowel disease. Gastroenterol Clin N Am. 2004;33:141–69.
23. Ogata H, Matsui T, Nakamura M et al. A randomised dose finding study of oral tacrolimus (FK506) therapy in refractory ulcerative colitis [see comment]. Gut. 2006;55:1255–62.
24. Sands BE, Tremaine WJ, Sandborn WJ et al. Infliximab in the treatment of severe, steroid-refractory ulcerative colitis: a pilot study. Inflamm Bowel Dis. 2001;7:83–8.

25. Jarnerot G, Hertervig E, Friis-Liby I et al. Infliximab as rescue therapy in severe to moderately severe ulcerative colitis: a randomized, placebo-controlled study. Gastroenterology. 2005;128:1805–11.
26. Ochsenkuhn T, Sackmann M, Goke B. Infliximab for acute, not steroid-refractory ulcerative colitis: a randomized pilot study. Eur J Gastroenterol Hepatol. 2004;16:1167–71.
27. Armuzzi A, De Pascalis B, Lupascu A et al. Infliximab in the treatment of steroid-dependent ulcerative colitis. Eur Rev Med Pharmacol Sci. 2004;8:231–3.

28
The surgical approach for refractory ulcerative colitis

S. BARATSIS

Ulcerative colitis (UC) is characterized by chronic colonic mucosal inflammation of unknown aetiology. UC is pathologically limited to the rectum and colon, facilitating definitive surgical therapy. Whereas the role of surgery in the other major form of inflammatory bowel disease (IBD), Crohn's disease, is primarily to treat complications of the disease process, surgery in UC is curative for the intestinal manifestations of the disease and almost eliminates the risk of future malignancy.

There exist three major indications for surgical intervention in UC:

- Treatment of acute, medically unresponsive flares;
- Treatment of poorly controlled symptomatic disease or addressing intolerable treatment side-effects; and
- Prevention or treatment of malignancy after long-standing symptomatic or even asymptomatic disease.

Within a lifetime approximately 15% of UC patients will have a severe relapse necessitating admission to hospital. Fulminant UC is a potentially life-threatening disorder that must be expertly managed if optimal outcomes are to be achieved. This condition was once associated with a very high mortality, but medical and surgical treatments have improved dramatically, to the point where the mortality associated with fulminant UC is now lower than 3%. Optimal management depends on close coordination between medical and surgical therapy, and multidisciplinary strategies are essential. Despite intravenous steroid treatment, approximately 25% will require either surgery or cyclosporin-A (CsA) rescue therapy. Care must be exercised, to exclude concomitant infections, especially *Cytomegalovirus* infection, and not to overtreat patients with fulminant UC.

Patients with perforation or severe gastrointestinal bleeding require urgent surgical treatment. The debilitation resulting from the disease, coupled with the immunosuppression resulting from intensive medical therapy, can mask the signs and symptoms of sepsis and peritonitis associated with perforation.

Perioperative mortality in cases of fulminant colitis is as much as 10 times higher when perforation occurs than when it does not. For this reason, patients with high fever, marked leukocytosis, and persistent tachycardia should be referred for early surgery, regardless of whether other indications of perforation or peritonitis are noted.

Toxic megacolon, though an uncommon complication of severe UC, is important in that it is associated with impending colonic perforation and therefore must be watched for and aggressively managed if present. Two specific conditions must be satisfied to establish the diagnosis of toxic megacolon: first, there must be colonic dilation; second, the patient must be in a toxic state. Patients admitted to the hospital with fulminant UC will, by definition, exhibit some degree of toxicity. Colonic dilation in these patients is a very worrisome phenomenon, in that it completes the picture of toxic megacolon. Accordingly, in all patients with fulminant UC, an abdominal X-ray should be obtained to look for colonic dilation. Those in whom abdominal distension develops, or who experience a sudden decrease in the number of bowel movements without signs of significant clinical improvement, should also be assessed for colonic dilation and toxic megacolon. In patients with toxic megacolon who are otherwise stable, conservative management, consisting of elimination of narcotics and anticholinergic agents, may be briefly tried. Patients who do not rapidly respond to conservative management, and those who show signs of peritonitis or are otherwise unstable, require urgent surgical treatment.

Patients who do not show significant improvement in response to intravenous (i.v.) steroid therapy within 5–7 days should be started on i.v. CsA therapy or referred for operative treatment. The introduction of CsA for use in patients with severe UC has provided an alternative to patients previously facing only surgical options. Those who do not respond to cyclosporin therapy within 4 days, or in whom remission of major symptoms is not achieved within 2 weeks, should be treated surgically. Patients whose symptoms progress during the course of i.v. therapy, or who show no sign of improvement at all, should be considered for early surgery. Eighty-five per cent of patients with more than eight bowel movements daily, or three to eight bowel movements combined with CRP >45, will undergo surgery unrelavent with the treatment with steroids or CsA. Persistent bowel dilation indicates no or partial response to conservative treatment and a high risk for toxic megacolon. Patients known to have deep longitudinal ulcerations are less likely to respond to i.v. medical therapy and thus may also be referred for early surgery. The decision regarding when to abandon medical therapy for fulminant UC in favour of surgical therapy is difficult, and requires considerable experience and special expertise.

The immunosuppressive effects of high-dose corticosteroids and i.v. cyclosporin, along with the debilitation induced by prolonged severe disease, can place patients at high risk for perioperative complications.

Although initial response rates to CsA have been encouraging, remission rates have been disappointing. If patients achieved initial remission with CsA, after 1 year 65% had relapsed and after 3 years 90% had relapsed. After 7 years 58% had come to colectomy. In large series so far reported the precise assessment of the occurrence of adverse events was difficult because the trials

described different adverse reactions, which reversed after discontinuation of cyclosporin. The major side-effects of cyclosporin treatment are renal insufficiency, opportunistic infections, and seizures. The risk of seizures appears to be highest in patients with hypocholesterolaemia. Consequently, cyclosporin should not be given to patients with significant hypocholesterolaemia (serum cholesterol concentration <100 mg/dl). Hypomagnesaemia is commonly seen in patients with fulminant UC who undergo CsA treatment; accordingly, serum magnesium levels should be closely followed. As most of the side-effects of cyclosporin therapy are dose-dependent, monitoring of whole-blood levels by high-power liquid chromatography or radioimmunoassay is needed.

As many as 60% of patients experience recurrence of disease after initial CsA-induced remission. Recurrence rates can be substantially lowered by means of maintenance therapy with 6-mercaptopurine (6-MP) or azathioprine, and with appropriate maintenance therapy the rate of early recurrence of symptoms after successful i.v. cyclosporin treatment may be reduced to levels as low as 22%. Azathioprine, acting as a prodrug, is converted intracellularly to mercaptopurine. Both are purine antimetabolite agents; however, the exact mechanism of their action is not known. During therapy it is important to monitor the patient's haematological profile for evidence of significant bone marrow suppression, and liver function tests for evidence of hepatotoxicity, which can occur infrequently.

These agents, similar to the aminosalicylates, have allergic and non-allergic adverse effects that can limit their use. The most common allergic reactions include pancreatitis, fevers, skin reactions, and gastrointestinal tract disturbances. Approximately 5–10% of patients will experience some type of allergic reaction that prevents continued use. Often, if a patient has a reaction to one of the two drugs, he or she will have a similar reaction to the other. The non-allergic adverse effects are primarily related to bone marrow suppression. The leukopenia and thrombocytopenia can be profound, leading to opportunistic infections or bleeding. Although there are reports of an association with long-term use of these agents and an increased risk for malignancies, particularly lymphoma, this has not been demonstrated in larger reviews.

In a surgical perspective, even if the disease does recur, the initial success of cyclosporin or other immunosuppressive therapy in aborting the acute phase of the UC allows patients to recover from the acute illness, so that they are in better condition to undergo elective surgical treatment at a later date if such treatment ultimately proved to be necessary. This is a major benefit, in that operative management of UC carries a much higher risk of complications when carried out on an urgent basis than when carried out in an elective setting.

As the indications for surgery are clear, both early in the course of the disease and during the chronic disease phase, which may be asymptomatic, early curative surgical intervention is a reasonable alternative to prolonged medical management. Currently, surgical intervention is accomplished safely, with good functional results, and with a high degree of patient satisfaction.

Since the early 1980s surgical therapy has evolved to removal of the entire colon and rectum followed by construction of an ileal pouch that is

anastomosed to the anal canal. This procedure is known as a proctocolectomy and ileal pouch anal anastomosis (IPAA). IPAA avoids the need for a permanent ostomy and maintains the normal route of defecation, albeit with altered frequency. The operation is usually performed in one or two stages: a preliminary colectomy with rectal preservation, which adds a third stage to the procedure, is reversed for severely ill patients. The severity and frequency of complications related to surgery have decreased significantly and the functional results are durable. The most common early complications are pelvic sepsis from a pouch leak (3%), abdominal wound infections (3%), and early small bowel obstructions (15%). Equally important as good functional results are the patients' perceptions of their life after the procedure. Numerous studies using validated quality of life (QoL) measures indicate that there is markedly improved QoL after restorative surgery. Most patients report 'perfect health' at 12 months after surgery. Even UC patients with well-controlled disease preoperatively continue to perceive themselves as having a lower QoL due to living with a chronic illness and score similarly to patients with diabetes. What is clear from these and other studies is that patients who undergo surgery for UC have an overall and health-related QoL comparable with the healthy general population, despite alterations in their bowel habits.

Although a large number of different surgeons and institutions have reported their experience with IPAA, the functional results are quite similar. Most patients report good to excellent function with their ileal pouch. The markers of function that are most often recorded are the number of bowel movements during the day and the night, episodes of soiling, and use of medications to control bowel activity. Normal function is five to seven bowel movements per day and one to two at night. Most patients report complete daytime continence. In those patients who have had their ileal pouch for longer than 10 years, stool frequency and continence are remarkably stable over time. However, episodes of incontinence, particularly nocturnal incontinence, increase slightly over time. Even with the slight decline in function over time, most patients report a high degree of satisfaction with their ileal pouch function and QoL.

Although IPAA has become the procedure of choice for most patients with chronic UC, some authors have shown that QoL improves no matter what procedure is performed, and is probably due mostly to improvement in the patients' general health after eradication of the disease. These findings suggest that existing QoL measurement tools may not completely address all of the important variables influenced by these procedures.

While fulminant disease, failure of medical therapy, or intractability of disease symptoms are clear indications for surgical intervention, protracted disease, even when asymptomatic, is an equally important indication for colectomy due to the increased risk of malignancy. While there are reports of no increased risk of malignancy in the setting of chronic UC, the majority of the literature supports the contention that chronic UC patients represent a population at high risk for developing colorectal cancer.

It is generally accepted that the risk of colorectal cancer in the setting of UC increases with duration of disease activity. One estimate puts that risk at 25–30% after 25 years of disease activity. In those UC patients managed chronically with medications, it is recommended that they undergo

colonoscopic surveillance. Yet the optimum time course and frequency for surveillance is currently unclear, and recent evidence suggests that the presence of any dysplasia, even low-grade, occurring in the background of chronic UC may be an indication for colectomy. The knowledge of dysplasia-associated risk by gastroenterologists leads to inconsistency of management, which can harm patients. Only 53% of surveyed gastroenterologists recommended colectomy in the setting of high-grade dysplasia while 16% were unaware of the significance of a dysplasia-associated lesion or mass. Also important to this discussion is whether asymptomatic patients who have the IPAA procedure for dysplasia are any less satisfied with the result. Many of these patients may have near-normal preoperative bowel function. However, in QoL measurements among patients who had the IPAA for active colitis compared with those with dysplasia, there were no differences in QoL; both groups reported a high degree of satisfaction with their postoperative condition. The inconsistency of practice by gastroenterologists, the risks and costs of surveillance, and the known cancer risk, make it unclear if there is any benefit to deferring surgery in those patients with chronic disease, even if they are asymptomatic.

The cost of UC to the individual and society is significant, and productivity loss due to illness and the impact on patient lifestyle related to symptoms or treatment side-effects must be considered. With increased use of immunomodulator therapy for UC, there might be an increasing tendency to defer surgical treatment. However, aggressive medical therapy in the long term might be more costly to the individual and society due to a delay in definitive surgical therapy that a large proportion of patients will eventually require. In a cost analysis of patients treated with aggressive medical therapy, Sher et al.[11] reported that the mean total hospital cost for medically treated patients admitted with a severe UC flare was $28 477 per individual for that hospitalization. The one-time cost for a three-stage restorative procedure was $33 041 per patient. If patients had undergone surgery earlier they might have avoided the three-stage procedure that is associated with increased morbidity and cost.

Surgical therapy for UC provides definitive treatment for the intestinal manifestations of the disease. When performed by experienced surgeons the complication rate is low, and the functional results are predictably good. Most patients report a QoL comparable with that of normal healthy patients. While many patients with symptoms can be managed effectively on medical therapy, these same patients will eventually be referred for surgery due to the duration of the disease and the increased risk of malignancy. Given the annual financial and personal cost of prolonged medical therapy, the risk of failed medical therapy and possible emergent surgery, and the need for surveillance procedures due to the risk of malignancy, early planned surgical therapy should be advocated. Definitive surgical therapy in the form of IPAA allows UC patients to resume normal healthy lives with an excellent QoL.

Bibliography

1. Campell S, Travis S, Jewell D. Ciclosporin use in acute ulcerative colitis: a long-term experience. Eur J Gastroenterol Hepatol. 2005;17:79–84.
2. Shibolet O, Regushevskaya E, Brezis M, Soares-Weiser K. Cyclosporine A for induction or remission in severe ulcerative colitis. Cochrane Database Syst Rev 2005;1:CD004277.
3. Cima R, Pemberton J. Medical and surgical management of ulcerative colitis. Arch Surg. 2005;140:300–10.
4. Carter M, Lobo A, Travis SP. Guidelines for the management of inflammatory bowel disease in adults. Gut. 2004;53:1–16.
5. Pikarsky A, Zmora O, Wexner S. Immunosuppressants and operation in ulcerative colitis. J Am Coll Surg. 2002;195:251–60.
6. Marteau P. Inflammatory bowel disease. Endoscopy. 2002;34:63–8.
7. Hyman N, Cataldo P, Osler T. Urgent subtotal colectomy for severe inflammatory bowel disease. Dis Colon Rectum. 2005;48:70–3.
8. Fazio V, Tekkis P, Remzi F et al. Quantification of risk for pouch failure after ileal pouch anal anastomosis surgery. Ann Surg. 2003;238:605–17.
9. Wong D, Rothenberger D, Goldberg S, Madoff R. Colorectal cancer in inflammatory bowel disease. Dis Colon Rectum. 1999;42:343–7.
10. Moskovitz D, Van Assche G, Maenhout B et al. Incidence of colectomy during long-term follow-up after cyclosporine-induced remission of severe ulcerative colitis. Clin Gastroenterol Hepatol. 2006;4:760–5.
11. Sher ME, Weiss EG, Norgueras JJ et al. Morbidity of medical therapy for ulcerative colitis: what are we really saving? Int J Colorectal Dis. 1996;11:287–93.

Index

Falk Symposium Series

43. Reutter W, Popper H, Arias IM, Heinrich PC, Keppler D, Landmann L, eds.: *Modulation of Liver Cell Expression*. Falk Symposium No. 43. 1987 ISBN: 0-85200-677-2*
44. Boyer JL, Bianchi L, eds.: *Liver Cirrhosis*. Falk Symposium No. 44. 1987 ISBN: 0-85200-993-3*
45. Paumgartner G, Stiehl A, Gerok W, eds.: *Bile Acids and the Liver*. Falk Symposium No. 45. 1987 ISBN: 0-85200-675-6*
46. Goebell H, Peskar BM, Malchow H, eds.: *Inflammatory Bowel Diseases – Basic Research & Clinical Implications*. Falk Symposium No. 46. 1988 ISBN: 0-7462-0067-6*
47. Bianchi L, Holt P, James OFW, Butler RN, eds.: *Aging in Liver and Gastrointestinal Tract*. Falk Symposium No. 47. 1988 ISBN: 0-7462-0066-8*
48. Heilmann C, ed.: *Calcium-Dependent Processes in the Liver*. Falk Symposium No. 48. 1988 ISBN: 0-7462-0075-7*
50. Singer MV, Goebell H, eds.: *Nerves and the Gastrointestinal Tract*. Falk Symposium No. 50. 1989 ISBN: 0-7462-0114-1
51. Bannasch P, Keppler D, Weber G, eds.: *Liver Cell Carcinoma*. Falk Symposium No. 51. 1989 ISBN: 0-7462-0111-7
52. Paumgartner G, Stiehl A, Gerok W, eds.: *Trends in Bile Acid Research*. Falk Symposium No. 52. 1989 ISBN: 0-7462-0112-5
53. Paumgartner G, Stiehl A, Barbara L, Roda E, eds.: *Strategies for the Treatment of Hepatobiliary Diseases*. Falk Symposium No. 53. 1990 ISBN: 0-7923-8903-4
54. Bianchi L, Gerok W, Maier K-P, Deinhardt F, eds.: *Infectious Diseases of the Liver*. Falk Symposium No. 54. 1990 ISBN: 0-7923-8902-6
55. Falk Symposium No. 55 not published

55B. Hadziselimovic F, Herzog B, Bürgin-Wolff A, eds.: *Inflammatory Bowel Disease and Coeliac Disease in Children.* International Falk Symposium. 1990 ISBN 0-7462-0125-7

56. Williams CN, eds.: *Trends in Inflammatory Bowel Disease Therapy*. Falk Symposium No. 56. 1990 ISBN: 0-7923-8952-2
57. Bock KW, Gerok W, Matern S, Schmid R, eds.: *Hepatic Metabolism and Disposition of Endo- and Xenobiotics*. Falk Symposium No. 57. 1991 ISBN: 0-7923-8953-0
58. Paumgartner G, Stiehl A, Gerok W, eds.: *Bile Acids as Therapeutic Agents: From Basic Science to Clinical Practice.* Falk Symposium No. 58. 1991 ISBN: 0-7923-8954-9
59. Halter F, Garner A, Tytgat GNJ, eds.: *Mechanisms of Peptic Ulcer Healing*. Falk Symposium No. 59. 1991 ISBN: 0-7923-8955-7
60. Goebell H, Ewe K, Malchow H, Koelbel Ch, eds.: *Inflammatory Bowel Diseases – Progress in Basic Research and Clinical Implications*. Falk Symposium No. 60. 1991 ISBN: 0-7923-8956-5
61. Falk Symposium No. 61 not published
62. Dowling RH, Folsch UR, Löser Ch, eds.: *Polyamines in the Gastrointestinal Tract*. Falk Symposium No. 62. 1992 ISBN: 0-7923-8976-X
63. Lentze MJ, Reichen J, eds.: *Paediatric Cholestasis: Novel Approaches to Treatment*. Falk Symposium No. 63. 1992 ISBN: 0-7923-8977-8
64. Demling L, Frühmorgen P, eds.: *Non-Neoplastic Diseases of the Anorectum*. Falk Symposium No. 64. 1992 ISBN: 0-7923-8979-4

64B. Gressner AM, Ramadori G, eds.: *Molecular and Cell Biology of Liver Fibrogenesis*. International Falk Symposium. 1992 ISBN: 0-7923-8980-8

*These titles were published under the MTP Press imprint.

Falk Symposium Series

65. Hadziselimovic F, Herzog B, eds.: *Inflammatory Bowel Diseases and Morbus Hirschprung*. Falk Symposium No. 65. 1992 ISBN: 0-7923-8995-6
66. Martin F, McLeod RS, Sutherland LR, Williams CN, eds.: *Trends in Inflammatory Bowel Disease Therapy.* Falk Symposium No. 66. 1993 ISBN: 0-7923-8827-5
67. Schölmerich J, Kruis W, Goebell H, Hohenberger W, Gross V, eds.: *Inflammatory Bowel Diseases – Pathophysiology as Basis of Treatment*. Falk Symposium No. 67. 1993 ISBN: 0-7923-8996-4
68. Paumgartner G, Stiehl A, Gerok W, eds.: *Bile Acids and The Hepatobiliary System: From Basic Science to Clinical Practice*. Falk Symposium No. 68. 1993 ISBN: 0-7923-8829-1
69. Schmid R, Bianchi L, Gerok W, Maier K-P, eds.: *Extrahepatic Manifestations in Liver Diseases*. Falk Symposium No. 69. 1993 ISBN: 0-7923-8821-6
70. Meyer zum Büschenfelde K-H, Hoofnagle J, Manns M, eds.: *Immunology and Liver*. Falk Symposium No. 70. 1993 ISBN: 0-7923-8830-5
71. Surrenti C, Casini A, Milani S, Pinzani M , eds.: *Fat-Storing Cells and Liver Fibrosis*. Falk Symposium No. 71. 1994 ISBN: 0-7923-8842-9
72. Rachmilewitz D, ed.: *Inflammatory Bowel Diseases – 1994*. Falk Symposium No. 72. 1994 ISBN: 0-7923-8845-3
73. Binder HJ, Cummings J, Soergel KH, eds.: *Short Chain Fatty Acids.* Falk Symposium No. 73. 1994 ISBN: 0-7923-8849-6

73B. Möllmann HW, May B, eds.: *Glucocorticoid Therapy in Chronic Inflammatory Bowel Disease: from basic principles to rational therapy.* International Falk Workshop. 1996 ISBN 0-7923-8708-2

74. Keppler D, Jungermann K, eds.: *Transport in the Liver.* Falk Symposium No. 74. 1994 ISBN: 0-7923-8858-5

74B. Stange EF, ed.: *Chronic Inflammatory Bowel Disease*. Falk Symposium. 1995 ISBN: 0-7923-8876-3

75. van Berge Henegouwen GP, van Hoek B, De Groote J, Matern S, Stockbrügger RW, eds.: *Cholestatic Liver Diseases: New Strategies for Prevention and Treatment of Hepatobiliary and Cholestatic Liver Diseases.* Falk Symposium 75. 1994. ISBN: 0-7923-8867-4
76. Monteiro E, Tavarela Veloso F, eds.: *Inflammatory Bowel Diseases: New Insights into Mechanisms of Inflammation and Challenges in Diagnosis and Treatment.* Falk Symposium 76. 1995. ISBN 0-7923-8884-4
77. Singer MV, Ziegler R, Rohr G, eds.: *Gastrointestinal Tract and Endocrine System.* Falk Symposium 77. 1995. ISBN 0-7923-8877-1
78. Decker K, Gerok W, Andus T, Gross V, eds.: *Cytokines and the Liver.* Falk Symposium 78. 1995. ISBN 0-7923-8878-X
79. Holstege A, Schölmerich J, Hahn EG, eds.: *Portal Hypertension.* Falk Symposium 79. 1995. ISBN 0-7923-8879-8
80. Hofmann AF, Paumgartner G, Stiehl A, eds.: *Bile Acids in Gastroenterology: Basic and Clinical Aspects.* Falk Symposium 80. 1995 ISBN 0-7923-8880-1
81. Riecken EO, Stallmach A, Zeitz M, Heise W, eds.: *Malignancy and Chronic Inflammation in the Gastrointestinal Tract – New Concepts.* Falk Symposium 81. 1995 ISBN 0-7923-8889-5
82. Fleig WE, ed.: *Inflammatory Bowel Diseases: New Developments and Standards.* Falk Symposium 82. 1995 ISBN 0-7923-8890-6

Falk Symposium Series

82B. Paumgartner G, Beuers U, eds.: *Bile Acids in Liver Diseases.* International Falk Workshop. 1995 ISBN 0-7923-8891-7

83. Dobrilla G, Felder M, de Pretis G, eds.: *Advances in Hepatobiliary and Pancreatic Diseases: Special Clinical Topics.* Falk Symposium 83. 1995. ISBN 0-7923-8892-5

84. Fromm H, Leuschner U, eds.: *Bile Acids – Cholestasis – Gallstones: Advances in Basic and Clinical Bile Acid Research.* Falk Symposium 84. 1995 ISBN 0-7923-8893-3

85. Tytgat GNJ, Bartelsman JFWM, van Deventer SJH, eds.: *Inflammatory Bowel Diseases.* Falk Symposium 85. 1995 ISBN 0-7923-8894-1

86. Berg PA, Leuschner U, eds.: *Bile Acids and Immunology.* Falk Symposium 86. 1996 ISBN 0-7923-8700-7

87. Schmid R, Bianchi L, Blum HE, Gerok W, Maier KP, Stalder GA, eds.: *Acute and Chronic Liver Diseases: Molecular Biology and Clinics*. Falk Symposium 87. 1996 ISBN 0-7923-8701-5

88. Blum HE, Wu GY, Wu CH, eds.: *Molecular Diagnosis and Gene Therapy.* Falk Symposium 88. 1996 ISBN 0-7923-8702-3

88B. Poupon RE, Reichen J, eds.: *Surrogate Markers to Assess Efficacy of TReatment in Chronic Liver Diseases.* International Falk Workshop. 1996 ISBN 0-7923-8705-8

89. Reyes HB, Leuschner U, Arias IM, eds.: *Pregnancy, Sex Hormones and the Liver.* Falk Symposium 89. 1996 ISBN 0-7923-8704-X

89B. Broelsch CE, Burdelski M, Rogiers X, eds.: *Cholestatic Liver Diseases in Children and Adults.* International Falk Workshop. 1996 ISBN 0-7923-8710-4

90. Lam S-K, Paumgartner P, Wang B, eds.: *Update on Hepatobiliary Diseases 1996.* Falk Symposium 90. 1996 ISBN 0-7923-8715-5

91. Hadziselimovic F, Herzog B, eds.: *Inflammatory Bowel Diseases and Chronic Recurrent Abdominal Pain*. Falk Symposium 91. 1996 ISBN 0-7923-8722-8

91B. Alvaro D, Benedetti A, Strazzabosco M, eds.: *Vanishing Bile Duct Syndrome – Pathophysiology and Treatment*. International Falk Workshop. 1996 ISBN 0-7923-8721-X

92. Gerok W, Loginov AS, Pokrowskij VI, eds.: *New Trends in Hepatology 1996*. Falk Symposium 92. 1997 ISBN 0-7923-8723-6

93. Paumgartner G, Stiehl A, Gerok W, eds.: *Bile Acids in Hepatobiliary Diseases – Basic Research and Clinical Application.* Falk Symposium 93. 1997 ISBN 0-7923-8725-2

94. Halter F, Winton D, Wright NA, eds.: *The Gut as a Model in Cell and Molecular Biology.* Falk Symposium 94. 1997 ISBN 0-7923-8726-0

94B. Kruse-Jarres JD, Schölmerich J, eds.: *Zinc and Diseases of the Digestive Tract.* International Falk Workshop. 1997 ISBN 0-7923-8724-4

95. Ewe K, Eckardt VF, Enck P, eds.: *Constipation and Anorectal Insufficiency.* Falk Symposium 95. 1997 ISBN 0-7923-8727-9

96. Andus T, Goebell H, Layer P, Schölmerich J, eds.: *Inflammatory Bowel Disease – from Bench to Bedside.* Falk Symposium 96. 1997 ISBN 0-7923-8728-7

97. Campieri M, Bianchi-Porro G, Fiocchi C, Schölmerich J, eds. *Clinical Challenges in Inflammatory Bowel Diseases: Diagnosis, Prognosis and Treatment.* Falk Symposium 97. 1998 ISBN 0-7923-8733-3

98. Lembcke B, Kruis W, Sartor RB, eds. *Systemic Manifestations of IBD: The Pending Challenge for Subtle Diagnosis and Treatment.* Falk Symposium 98. 1998 ISBN 0-7923-8734-1

99. Goebell H, Holtmann G, Talley NJ, eds. *Functional Dyspepsia and Irritable Bowel Syndrome: Concepts and Controversies.* Falk Symposium 99. 1998 ISBN 0-7923-8735-X

Falk Symposium Series

100. Blum HE, Bode Ch, Bode JCh, Sartor RB, eds. *Gut and the Liver.* Falk Symposium 100. 1998 ISBN 0-7923-8736-8
101. Rachmilewitz D, ed. *V International Symposium on Inflammatory Bowel Diseases.* Falk Symposium 101. 1998 ISBN 0-7923-8743-0
102. Manns MP, Boyer JL, Jansen PLM, Reichen J, eds. *Cholestatic Liver Diseases.* Falk Symposium 102. 1998 ISBN 0-7923-8746-5

102B. Manns MP, Chapman RW, Stiehl A, Wiesner R, eds. *Primary Sclerosing Cholangitis.* International Falk Workshop. 1998. ISBN 0-7923-8745-7

103. Häussinger D, Jungermann K, eds. *Liver and Nervous System.* Falk Symposium 102. 1998 ISBN 0-7924-8742-2

103B. Häussinger D, Heinrich PC, eds. *Signalling in the Liver*. International Falk Workshop. 1998 ISBN 0-7923-8744-9

103C. Fleig W, ed. *Normal and Malignant Liver Cell Growth*. International Falk Workshop. 1998 ISBN 0-7923-8748-1

104. Stallmach A, Zeitz M, Strober W, MacDonald TT, Lochs H, eds. *Induction and Modulation of Gastrointestinal Inflammation.* Falk Symposium 104. 1998 ISBN 0-7923-8747-3
105. Emmrich J, Liebe S, Stange EF, eds. *Innovative Concepts in Inflammatory Bowel Diseases.* Falk Symposium 105. 1999 ISBN 0-7923-8749-X
106. Rutgeerts P, Colombel J-F, Hanauer SB, Schölmerich J, Tytgat GNJ, van Gossum A, eds. *Advances in Inflammatory Bowel Diseases.* Falk Symposium 106. 1999 ISBN 0-7923-8750-3
107. Špičák J, Boyer J, Gilat T, Kotrlik K, Mareček Z, Paumgartner G, eds. *Diseases of the Liver and the Bile Ducts – New Aspects and Clinical Implications.* Falk Symposium 107. 1999 ISBN 0-7923-8751-1
108. Paumgartner G, Stiehl A, Gerok W, Keppler D, Leuschner U, eds. *Bile Acids and Cholestasis.* Falk Symposium 108. 1999 ISBN 0-7923-8752-X
109. Schmiegel W, Schölmerich J, eds. *Colorectal Cancer – Molecular Mechanisms, Premalignant State and its Prevention.* Falk Symposium 109. 1999 ISBN 0-7923-8753-8
110. Domschke W, Stoll R, Brasitus TA, Kagnoff MF, eds. *Intestinal Mucosa and its Diseases – Pathophysiology and Clinics.* Falk Symposium 110. 1999 ISBN 0-7923-8754-6

110B. Northfield TC, Ahmed HA, Jazwari RP, Zentler-Munro PL, eds. *Bile Acids in Hepatobiliary Disease*. Falk Workshop. 2000 ISBN 0-7923-8755-4

111. Rogler G, Kullmann F, Rutgeerts P, Sartor RB, Schölmerich J, eds. *IBD at the End of its First Century.* Falk Symposium 111. 2000 ISBN 0-7923-8756-2
112. Krammer HJ, Singer MV, eds. *Neurogastroenterology: From the Basics to the Clinics.* Falk Symposium 112. 2000 ISBN 0-7923-8757-0
113. Andus T, Rogler G, Schlottmann K, Frick E, Adler G, Schmiegel W, Zeitz M, Schölmerich J, eds. *Cytokines and Cell Homeostasis in the Gastrointestinal Tract.* Falk Symposium 113. 2000 ISBN 0-7923-8758-9
114. Manns MP, Paumgartner G, Leuschner U, eds. *Immunology and Liver.* Falk Symposium 114. 2000 ISBN 0-7923-8759-7
115. Boyer JL, Blum HE, Maier K-P, Sauerbruch T, Stalder GA, eds. *Liver Cirrhosis and its Development*. Falk Symposium 115. 2000 ISBN 0-7923-8760-0
116. Riemann JF, Neuhaus H, eds. *Interventional Endoscopy in Hepatology.* Falk Symposium 116. 2000 ISBN 0-7923-8761-9

Falk Symposium Series

116A. Dienes HP, Schirmacher P, Brechot C, Okuda K, eds. *Chronic Hepatitis: New Concepts of Pathogenesis, Diagnosis and Treatment*. Falk Workshop. 2000 ISBN 0-7923-8763-5

117. Gerbes AL, Beuers U, Jüngst D, Pape GR, Sackmann M, Sauerbruch T, eds. *Hepatology 2000 – Symposium in Honour of Gustav Paumgartner*. Falk Symposium 117. 2000 ISBN 0-7923-8765-1

117A. Acalovschi M, Paumgartner G, eds. *Hepatobiliary Diseases: Cholestasis and Gallstones.* Falk Workshop. 2000 ISBN 0-7923-8770-8

118. Frühmorgen P, Bruch H-P, eds. *Non-Neoplastic Diseases of the Anorectum*. Falk Symposium 118. 2001 ISBN 0-7923-8766-X

119. Fellermann K, Jewell DP, Sandborn WJ, Schölmerich J, Stange EF, eds. *Immunosuppression in Inflammatory Bowel Diseases – Standards, New Developments, Future Trends.* Falk Symposium 119. 2001 ISBN 0-7923-8767-8

120. van Berge Henegouwen GP, Keppler D, Leuschner U, Paumgartner G, Stiehl A, eds. *Biology of Bile Acids in Health and Disease.* Falk Symposium 120. 2001 ISBN 0-7923-8768-6

121. Leuschner U, James OFW, Dancygier H, eds. *Steatohepatitis (NASH and ASH).* Falk Symposium 121. 2001 ISBN 0-7923-8769-4

121A. Matern S, Boyer JL, Keppler D, Meier-Abt PJ, eds. *Hepatobiliary Transport: From Bench to Bedside.* Falk Workshop. 2001 ISBN 0-7923-8771-6

122. Campieri M, Fiocchi C, Hanauer SB, Jewell DP, Rachmilewitz R, Schölmerich J, eds. *Inflammatory Bowel Disease – A Clinical Case Approach to Pathophysiology, Diagnosis, and Treatment*. Falk Symposium 122. 2002 ISBN 0-7923-8772-4

123. Rachmilewitz D, Modigliani R, Podolsky DK, Sachar DB, Tozun N, eds. *VI International Symposium on Inflammatory Bowel Diseases*. Falk Symposium 123. 2002 ISBN 0-7923-8773-2

124. Hagenmüller F, Manns MP, Musmann H-G, Riemann JF, eds. *Medical Imaging in Gastroenterology and Hepatology.* Falk Symposium 124. 2002 ISBN 0-7923-8774-0

125. Gressner AM, Heinrich PC, Matern S, eds. *Cytokines in Liver Injury and Repair.* Falk Symposium 125. 2002 ISBN 0-7923-8775-9

126. Gupta S, Jansen PLM, Klempnauer J, Manns MP, eds. *Hepatocyte Transplantation.* Falk Symposium 126. 2002 ISBN 0-7923-8776-7

127. Hadziselimovic F, ed. *Autoimmune Diseases in Paediatric Gastroenterology.* Falk Symposium 127. 2002 ISBN 0-7923-8778-3

127A. Berr F, Bruix J, Hauss J, Wands J, Wittekind Ch, eds. *Malignant Liver Tumours: Basic Concepts and Clinical Management.* Falk Workshop. 2002 ISBN 0-7923-8779-1

128. Scheppach W, Scheurlen M, eds. *Exogenous Factors in Colonic Carcinogenesis.* Falk Symposium 128. 2002 ISBN 0-7923-8780-5

129. Paumgartner G, Keppler D, Leuschner U, Stiehl A, eds. *Bile Acids: From Genomics to Disease and Therapy.* Falk Symposium 129. 2002 ISBN 0-7923-8781-3

129A. Leuschner U, Berg PA, Holtmeier J, eds. *Bile Acids and Pregnancy.* Falk Workshop. 2002 ISBN 0-7923-8782-1

130. Holtmann G, Talley NJ, eds. *Gastrointestinal Inflammation and Disturbed Gut Function: The Challenge of New Concepts*. Falk Symposium 130. 2003 ISBN 0-7923-8783-X

131. Herfarth H, Feagan BJ, Folsch UR, Schölmerich J, Vatn MH, Zeitz M, eds. *Targets of Treatment in Chronic Inflammatory Bowel Diseases*. Falk Symposium 131. 2003 ISBN 0-7923-8784-8

Falk Symposium Series

132. Galle PR, Gerken G, Schmidt WE, Wiedenmann B, eds. *Disease Progression and Carcinogenesis in the Gastrointestinal Tract*. Falk Symposium 132. 2003 ISBN 0-7923-8785-6

132A. Staritz M, Adler G, Knuth A, Schmiegel W, Schmoll H-J, eds. *Side-effects of Chemotherapy on the Gastrointestinal Tract*. Falk Workshop. 2003 ISBN 0-7923-8791-0

132B. Reutter W, Schuppan D, Tauber R, Zeitz M, eds. *Cell Adhesion Molecules in Health and Disease.* Falk Workshop. 2003 ISBN 0-7923-8786-4

133. Duchmann R, Blumberg R, Neurath M, Schölmerich J, Strober W, Zeitz M. *Mechanisms of Intestinal Inflammation: Implications for Therapeutic Intervention in IBD.* Falk Symposium 133. 2004 ISBN 0-7923-8787-2

134. Dignass A, Lochs H, Stange E. *Trends and Controversies in IBD – Evidence-Based Approach or Individual Management?* Falk Symposium 134. 2004 ISBN 0-7923-8788-0

134A. Dignass A, Gross HJ, Buhr V, James OFW. *Topical Steroids in Gastroenterology and Hepatology.* Falk Workshop. 2004 ISBN 0-7923-8789-9

135. Lukáš M, Manns MP, Špičák J, Stange EF, eds. *Immunological Diseases of Liver and Gut.* Falk Symposium 135. 2004 ISBN 0-7923-8792-9

136. Leuschner U, Broomé U, Stiehl A, eds. *Cholestatic Liver Diseases: Therapeutic Options and Perspectives.* Falk Symposium 136. 2004 ISBN 0-7923-8793-7

137. Blum HE, Maier KP, Rodés J, Sauerbruch T, eds. *Liver Diseases: Advances in Treatment and Prevention*. Falk Symposium 137. 2004 ISBN 0-7923-8794-5

138. Blum HE, Manns MP, eds. *State of the Art of Hepatology: Molecular and Cell Biology.* Falk Symposium 138. 2004 ISBN 0-7923-8795-3

138A. Hayashi N, Manns MP, eds. *Prevention of Progression in Chronic Liver Disease: An Update on SNMC (Stronger Neo-Minophagen C).* Falk Workshop. 2004 ISBN 0-7923-8796-1

139. Adler G, Blum HE, Fuchs M, Stange EF, eds. *Gallstones: Pathogenesis and Treatment*. Falk Symposium 139. 2004 ISBN 0-7923-8798-8

140. Colombel J-F, Gasché C, Schölmerich J, Vucelic C, eds. *Inflammatory Bowel Disease: Translation from Basic Research to Clinical Practice.* Falk Symposium 140. 2005. ISBN 1-4020-2847-4

141. Paumgartner G, Keppler D, Leuschner U, Stiehl A, eds. *Bile Acid Biology and its Therapeutic Implications.* Falk Symposium 141. 2005 ISBN 1-4020-2893-8

142. Dienes H-P, Leuschner U, Lohse AW, Manns MP, eds. *Autoimmune Liver Disease.* Falk Symposium 142. 2005 ISBN 1-4020-2894-6

143. Ammann RW, Büchler MW, Adler G, DiMagno EP, Sarner M, eds. *Pancreatitis: Advances in Pathobiology, Diagnosis and Treatment*. Falk Symposium 143. 2005 ISBN 1-4020-2895-4

144. Adler G, Blum AL, Blum HE, Leuschner U, Manns MP, Mössner J, Sartor RB, Schölmerich J, eds. *Gastroenterology Yesterday – Today – Tomorrow: A Review and Preview.* Falk Symposium 144. 2005 ISBN 1-4020-2896-2

145. Henne-Bruns D, Buttenschön K, Fuchs M, Lohse AW, eds. *Artificial Liver Support.* Falk Symposium 145. 2005 ISBN 1-4020-3239-0

146. Blumberg RS, Gangl A, Manns MP, Tilg H, Zeitz M, eds. *Gut–Liver Interactions: Basic and Clinical Concepts.* Falk Symposium 146. 2005 ISBN 1-4020-4143-8

147. Jewell DP, Colombel JF, Peña AS, Tromm A, Warren BS, eds. *Colitis: Diagnosis and Therapeutic Strategies.* Falk Symposium 147. 2006 ISBN 1-4020-4315-5

Falk Symposium Series

148. Kruis W, Forbes A, Jauch K-W, Kreis ME, Wexner SD, eds. *Diverticular Disease: Emerging Evidence in a Common Condition*. Falk Symposium 148. 2006 ISBN 1-4020- 4317-1
149. van Cutsem E, Rustgi AK, Schmiegel W, Zeitz M, eds. *Highlights in Gastrointestinal Oncology*. Falk Symposium 149. 2006. ISBN 1-4020-5108-5
150. Galle PR, Gerken G, Schmidt WE, Wiedenmann B, eds. *Disease Progression and Disease Prevention in Hepatology and Gastroenterology*. Falk Symposium 150. 2006 ISBN 1-4020-5109-3
151. Fraser A, Gibson PR, Hibi T, Qian J-M, Schölmerich, eds. *Emerging Issues in Inflammatory Bowel Disease*. Falk Symposium 151. 2006 ISBN-13 978-1-4020-5701-4
152. Not published.
153. Dignass A, Rachmilewitz D, Stange E-F, Weinstock JV, eds. *Immunoregulation in Inflammatory Bowel Diseases – Current Understanding and Innovation*. Falk Symposium 153. 2007 ISBN-13 978-1-4020-5888-2
154. Adler G, Fiocchi C, Lazebnik LB, Vorobiev GI, eds. *Inflammatory Bowel Disease – Diagnostic and Therapeutic Strategies*. Falk Symposium 154. 2007 ISBN-13 978-1-4020-6115-8
155. Keppler D, Beuers U, Leuschner U, Stiehl A, Trauner M, Paumgartner G, eds. *Bile Acids: Biological Actions and Clinical Relevance.* Falk Symposium 155. 2007 ISBN 978-1-4020-6251-3
156. Blum HE, Cox DW, Häussinger D, Jansen PLM, Kullak-Ublick GA, eds. *Genetics in Liver Diseases.* Falk Symposium 156. 2007 ISBN 978-14020-6393-0
157. Diehl AM, Hayashi N, Manns MP, Sauerbruch T, eds. *Chronic Hepatitis: Metabolic, Cholestatic, Viral and Autoimmune.* Falk Symposium 157. 2007 ISBN 978-1-4020-6522-4
158. Gasche G, Herrerías Gutiérrez JM, Gassull M, Monterio E, eds. *Intestinal Inflammation and Colorectal Cancer.* Falk Symposium 158. 2007 ISBN 978-1-4020-6825-6
159. Tözün N, Mantzaris G, Dağlı Ü, Schölmerich J, eds. *IBD 2007 – Achievements in Research and Clinical Practice*. Falk Symposium 159. 2008 ISBN 978-1-4020-6986-4