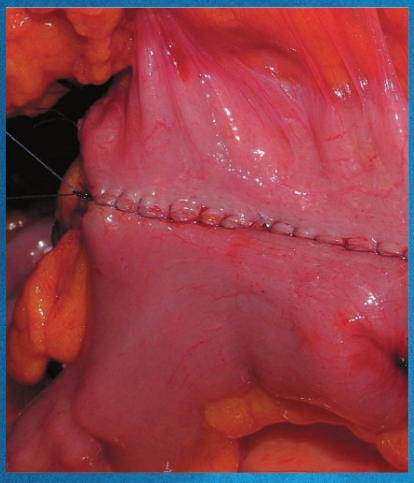
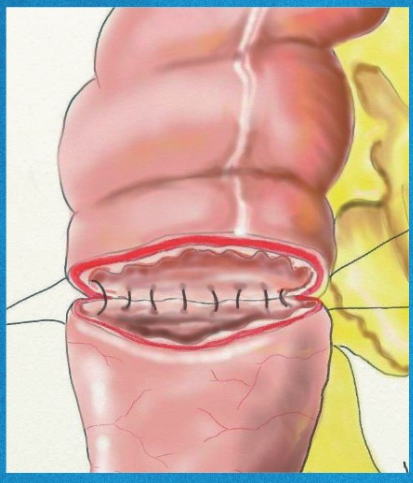
UPDATES in SURGERY

Roberto Tersigni
Cosimo Prantera

Crohn's Disease

A Multidisciplinary Approach





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Roberto Tersigni · Cosimo Prantera (Eds.)

Crohn's Disease

A Multidisciplinary Approach

Foreword by
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Springer

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Foreword

In 1761, J.B. Morgagni described the clinical case of a young man who died from an ileal perforation. The patient's clinical presentation included ileal ulceration and inflammation of the mesenteric lymphonodes with splenomegaly. This was the first description of Crohn's disease (CD).

In 1904, a Polish physician, Lesmowski, wrote a case report for a local medical journal, describing a patient with symptoms similar to those of CD. In 1913, an English author, C. Combe, and a Scottish colleague, T.K. Daziel, reported several cases of "chronic interstitial enteritis" with the features of CD. However, these descriptions were completely ignored by the academic world.

The pathology became known only after the classic description of Crohn, Ginzburg, and Oppenheimer in 1932. They described a "chronic granulomatous inflammation of the ileal ansa." Nevertheless, it was only in 1960, after the well-known publication of Lockhart-Mummery and Morson, that the existence of the disease entity, by then known as CD, was accepted.

While CD can affect any part of the digestive tract, without distinction, from the mouth to the anus, it shows very different localizations. The etiopathology of CD remains controversial; the disease has a variable clinical presentation and thus a number of therapeutic options, both medical and surgical. However, treatment of the frequent precocious and late complications of CD remains problematic.

More recently, research has focused on the immunological origins of the disease, specifically, on the humoral and cell-mediated responses of the immune system to sensitizing agents. Furthermore, environmental factors acting in concert with a genetic predisposition have been considered and may explain the increased incidence of CD in certain geographical areas over the past few decades.

The Italian Society of Surgery has recognized the need for an authoritative update on the subject and has entrusted this task to Roberto Tersigni, in recognition of his extensive experience in the field of CD and the inflammatory bowel diseases, and taking advantage of his numerous case files. The result is an exceedingly interesting scientific work, rich in details and in critical analyses of all the important issues related to CD. In addition, the book presents a wealth of new information that will

certainly be of interest to physicians and researchers seeking to broaden their knowledge of this disease.

As President of the Society and as someone who is passionately involved in this field of surgery, my compliments to Roberto and his co-workers for this excellent work. I am convinced it is destined to attain a prestigious place for itself in the world of international surgical knowledge.

Rome, October 2009

Enrico De Antoni
President of Italian Society of Surgery

Preface

Several prominent members of our institution, San Camillo-Forlanini Hospital of Rome, and of the University Faculty of Medicine of Rome, La Sapienza of Rome, joined together to present a book intended to provide a comprehensive discussion of Crohn's disease. The result, "*Crohn's Disease: A Multidisciplinary Approach*", attempts to systematically review all medical and surgical aspect of the disease. The first group of chapters deals with problems regarding the etiology, pathogenesis, and clinical manifesta-tions of Crohn's disease, while the second group concentrates on diagnostic techniques and clinical practice. This is followed by chapters addressing current concepts of medical and surgical treatment. The selected topics are those wherein recent developments have altered previous practices. The goals of this book are to offer general practitioners basic knowledge of Crohn's disease, to provide the specialist with more detailed information, and to stimulate students, residents, and fellows in their search for knowledge of this challenging but satisfying field.

The Editors express their thanks to all of the book's authors and to the Italian Society of Surgery.

Rome, October 2009

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Preface

This is not a typical book on Crohn's disease (CD); rather, it is a synthesis of a long effort that began in the 1970s at the "Regina Margherita" Hospital of Rome, when the Gastroenterology Unit began its study of inflammatory bowel diseases (IBD).

Dalziel was the first to describe CD, in a 1913 report published in a Scottish medical journal. Unfortunately, the journal was quite unknown to the medical community and, consequently, this pathological entity was re-discovered in 1932, by Burrill Crohn, a surgeon at Mount Sinai Hospital, in NY, who named this "new" entity after himself. In the 1960s, Basil Morson, a pathologist at St. Mark's Hospital, in London, described CD in the colon, thus distinguishing it from ulcerative colitis.

In the last 40 years, there have been many discoveries related to IBD and many new therapeutic approaches have been developed. However, the causes and mechanism of CD remain elusive. Today, CD is thought to arise from the interaction of genetic and environmental factors. Bacteria and food antigens, among the environmental causes, and the expression of certain, no doubt several genes account for the phenotypic aspects of the disease.

This intricate relationship is likely the source of differences in the location, pathological behavior, and, most importantly, therapeutic response or resistance that characterize CD. The newly developed biologicals have led to a reduction in the number of surgical operations, even if surgery remains an important strategy of CD treatment. The appropriate choice of a medical vs. a surgical approach is one of the major challenges that currently confront gastroenterologists.

In this book authors with a great deal of expertise in different disciplines and working in the same hospital have written chapters concerning the many aspects of CD. Neither the authors nor the editors have attempted to resolve what might appear to be conflicting information, as different points of view can increase the sum of our knowledge.

Rome, October 2009

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Etiology

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M. Marrollo, A. Armuzzi, F. Zannoni

Introduction

Although the etiology of Crohn's disease (CD) remains largely unexplained, there have been major advances in our understanding of the pathogenic mechanisms underlying intestinal inflammation. There is general agreement with regard to the multifactorial character of the disease, in which a genetic predisposition, the external environment, intestinal microbial flora, and the immune system are all involved. Together, they result in the generation and maintenance of the chronic intestinal inflammatory reaction [1].

Environmental Factors

Environmental factors seem to play an important role in the manifestation, course and prognosis of CD. Epidemiological studies suggest that the prevalence of CD is higher in industrialized countries. Moreover it seems that, at least in Western countries [2], but also in other parts of the world, there is currently a change in the type of the most common diseases affecting humans, with a switch from infectious to chronic inflammatory, including IBD, and neoplastic disease. This has been explained by the "hygiene hypothesis," i.e., the lack of exposure to microbial antigens early in life due to changing sanitation conditions [3,4]. A variety of environmental factors are considered to be risk factors for CD, among them, smoking, drugs, diet, and the composition of the enteric flora [5]. Smoking is a recognized risk factor for CD; patients who smoke, or have smoked in the past, are at greater risk of

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Crohn's Disease. Roberto Tersigni, Cosimo Prantera (Eds.)

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developing CD than non-smokers. Patients with CD who never smoked have a higher incidence of early-onset (< 20 years) disease and perhaps even a stronger genetic component, whereas current and former smokers seem to be diagnosed at a later age [6]. A history of smoking also has deleterious effects on the course of the CD, including greater recurrence after surgical and endoscopic procedures, more operations and complications, and a reduced quality of life [7,8]; and the discontinuation of smoking improves the disease course [9]. It has been proposed that passive smoking in children is associated with an increased risk of developing CD; however, a recent meta-analysis did not find a positive relationship between childhood passive smoke exposure and CD [10]. The exact mechanism of the effect of smoking on CD remains poorly understood but several mechanisms have been suggested. Macrophages from smokers express a selective functional deficiency in their ability to kill intracellular bacteria [11]. Nicotine or smoking could alter gut motility, reduce smooth muscle tone and contractility (modulated by nitric oxide) [12], decrease permeability [13], and alter the microcirculation [14]. Smoking, through the associated increased carbon monoxide concentration, may amplify impairment of the vasodilatation capacity in chronically inflamed microvessels, resulting in ischemia and perpetuating ulceration and fibrosis [15]. Smoking also increases the thrombotic potential associated with vascular damage.

Other environmental agents associated with CD are oral contraceptives (OC), although the association between OC and CD remains controversial. While a direct causal relationship has not been found, women taking OC have twice the risk of developing CD than those not taking contraceptives [16]. The pathogenic mechanism is thought to be the formation of microthrombi and multifocal gastrointestinal infarction [17]. The lack of breastfeeding during infancy was found to be positively associated with the risk of CD [18]. Beside providing immunological protection to the newborn, breastfeeding might reduce the stimulatory effect of bacteria or endotoxins on the immune system, either by direct reduction of exposure or by passive transfer of immune responses from the mother, possibly leading to immature immune response mechanisms in the infant [19].

Immunopathogenesis of Crohn's Disease

The epithelial barrier acts as first-line of defense to limit antigen penetration to the mucosal immune system. The intestinal epithelium, a polarized single layer covered by mucus in which commensal bacteria are embedded, is a critical mediator involved in the generation of immune tolerance and controlled inflammation to intestinal antigens, and of host defense responses against pathogens [20]. The importance of the epithelial barrier in disease predisposition is supported by the finding of abnormal intestinal permeability in patients with CD as well as their first-degree relatives. Moreover, defects in mucus production have been reported in patients with the disease [21]. Proinflammatory cytokines (TNF-α, IFN-γ, IL-17) released during intestinal inflammation can induce an increase in epithelial permeability through a

dynamic regulation of tight junctions. Finally, the bidirectional epithelial transport of immunoglobulins, which neutralizes antigens in the lumen and provides information on the contents of the lumen to the mucosal immune system, appears to be disrupted under the inflammatory conditions of CD [20]. Intestinal epithelial cells express various receptors (Toll-like, NOD1 and 2, chemokine receptors, antibody-specific Fc) that mediate communication between the luminal flora and the underlying network of innate and adaptive immune cells. In particular, epithelial NF-kB modulation (activation/suppression) seems to be a key point in the inhibition or stimulation of immune responses in CD [1]. In vitro and animal models support the role of bacteria in minimizing epithelial cell NF-kB activation and inhibiting IKB-α degradation, thus dampening intestinal inflammation. Moreover, spontaneous colitis develops and the expression of antimicrobial peptides is significantly attenuated in mice with deletion of IKBKG in intestinal epithelial cells [22]. Conversely, deletion of IKK-β in mice intestinal epithelial cells does not trigger spontaneous colitis, as in the case of NOD2 knockout mice. Together, these findings underline the role of the NF-kB signaling pathway within the intestinal epithelium in sustaining mucosal homeostasis, in mediating pathogen-specific responses, and in protecting or enhancing the inflammatory cascade in case of specific disease processes. Besides columnar epithelial cells, the small intestine also contains other specialized epithelial cells. Paneth cells play an important role in innate intestinal defense at the base of the crypts, where they produce and secrete various antimicrobial peptides, including α-defensins. It has been reported that a reduction in α-defensin expression by Paneth cells may contribute to the pathogenesis of CD of the terminal ileum in patients bearing NOD2 mutations [23]. Goblet cells produce trefoil peptides, which are important for mucosal defense and repair. In animal models, a goblet-cell-specific protein (RELMβ) is induced upon bacterial colonization, and disruption of the RELMβ gene reduces the severity of colitis [24]. Moreover, a goblet-cell-derived secretory mucin, MUC2, is differentially expressed in human inflammatory bowel disease (IBD), and MUC2 knockout mice develop spontaneous colitis [25]. The two main types of IBD, CD and ulcerative colitis (UC), represent clearly distinct forms of intestinal inflammation and profound alteration in mucosal immunity have been demonstrated in these patients. CD is characterized by a CD4 T-helper cell type 1 (Th1-phenotype) response dominated by the production of IFN-γ [26], while UC represents an atypical CD4 T-helper cell type 2 (Th2-phenotype) response characterized by an increased production of IL-13 [27]. This simplistic interpretation of the adaptive immune response is probably fundamentally correct, although recent studies suggest a more complex overlap between the two forms of IBD. In particular, the dysfunction of regulatory cells (originally “suppressor cells”) seems to contribute to mucosal immune abnormalities and the recently described Th17 cells appear to be involved in the intestinal inflammatory response of both CD and UC. Moreover, defects of the innate immune response seem to be a prerequisite for the excessive activation of adaptive immunity [1]. Innate immunity is a defense system with limited specificity that acts within minutes or hours and is mediated by several different cell types (epithelial cells, neutrophils, monocytes, macrophages, dendritic cells, NK cells). This immune response is primarily directed at the recognition of microbial antigens, through pattern

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recognition receptors on the cell surface (Toll-like receptors) and in the cytoplasm (NOD proteins). The genetic association of CD with polymorphisms in the NOD2 gene and autophagy-related genes (ATG16L1 and IRGM) suggests that alterations in the recognition and intracellular processing of bacterial components have a role in disease pathogenesis [28]. The exact mechanisms by which NOD2 dysregulation contributes to CD development remains incompletely understood. Transfection studies in primary human mononuclear cells showed that CD-associated NOD2 mutations induce a defective ability to activate NF-kB in response to muramyl dipeptide stimulation. This finding contrasts with the increased activation of NF-kB observed in the lamina propria of patients with CD. It has been hypothesized that loss-of-function NOD2 mutations results in impaired host-microbial interaction through different mechanisms (altered tolerance to bacterial stimulation, decreased clearance of oral pathogens, and abnormal colonization of the mucosal layer resulting from decreased production of α-defensins by epithelial cells). Under normal conditions, the constant exposure to luminal bacteria leads to chronic activation of the NOD2 signaling network and a hyporesponsiveness of cells to stimulation with NOD2 and TLR ligands. This tolerizing effect is impaired in individuals with disease-associated NOD2 variants [29]. Moreover, NOD2 knockout mice show increased *Listeria monocytogenes* spleen and liver colonization following oral exposure to this pathogen, and intestinal epithelial cells expressing a NOD2 frameshift variant have an impaired capacity to control the growth of *Salmonella typhimurium*. Although it has been reported that the level of some defensin-related sequences are decreased in NOD2 knockout mice and that the expression of human defensins 5 and 6 are decreased in the affected ileum of patients with CD, recent data have shown that decreased α-defensins expression is not related to NOD2 variants, but probably reflects a loss of surface epithelial and Paneth cells secondary to inflammation [28]. Finally, the recently identified genetic associations between autophagy genes and CD point out the importance of impaired intracellular processing of bacterial components as a pathogenic step leading to disease. It has been shown that ATG16L1 expression by intestinal epithelial cells is essential for autophagy of *Salmonella typhimurium* [30]. Moreover, mice deficient in the IRGM autophagy gene have an impaired capacity to eliminate intracellular microrganisms (*Toxoplasma gondii* and *Listeria monocytogenes*). In this context, it seems that alterations in the intracellular processing of bacteria, with a subsequent compensatory increase in the activity of Th1 cells, could represent a key feature in the pathogenesis of CD. Adaptive immunity is a defense system that acts within a few up to several days and depends on the type and number of various cells. In this context, responses are evoked through the combination of resident and recruited cell populations (mucosal B cells, T cells of Th1, Th17 or Th2 phenotype, regulatory T/B cells). In particular, Th1 development is triggered by microbial antigens that stimulate the production of IFN-γ and IL-12 subunit p40, while STAT6 enhances Th2 development and GATA3 induces the typical cytokine pattern (IL-4,IL-5, IL-13) [31]. Recent reports have highlighted the role of T regulatory cells and Th17 cells in the pathogenesis of CD. T regulatory cells exist in different subtypes: Tr1 cells (CD4+CD25-Tcells) exert their suppressor function through the secretion of IL-10; Tregs (CD4+CD25+FOXP3+Tcells) require cytokines and cell contact for their func-

tion; NKT cells act through cytokine secretion and cytotoxicity; and a variety of CD8+ cells use different mechanisms [32]. Studies focusing on the role of regulatory T cells in the pathogenesis of IBD have been extensively performed in animal models and have shown that normal CD4+ T-regulatory cell responses and commensal bacteria are required for the maintenance of gut immune homeostasis and that the administration of regulatory T cells may cure experimental colitis. Human studies have provided evidence that the relative proportion of Tregs is significantly increased in patients with active IBD and directly related to disease activity [33]. The recent discovery of a third Th lineage (Th17 cells), both developmentally and functionally distinct from Th1 and Th2 cells, has challenged the paradigm according to which CD4+ effector T helper cells are divided in two main separate subpopulations (Th1 and Th2). Th17 cells require IL-23, Il-6, and TGF-β1 for their differentiation and growth, and they produce IL-17, IL-6, and TNF-α. These cells mediate inflammation, immunity, and tissue damage in various conditions (autoimmunity, infections, inflammatory processes) [34]. As far as the pathogenesis of CD is concerned, Th17 cells and a subset of T cells sharing features of both Th1 and Th17 cells (i.e., producing both IFN-γ and IL-17) have been detected in the inflamed mucosa of patients. Cells sharing features of both Th1 and Th17 can arise from the modulation of Th17 cells by IL-12 and are partially inhibited by IL-23. These cells raise intriguing questions about the functional relationships and the role of Th17 and Th1 in mediating inflammatory processes in CD [35]. The role of Th17 cells in the pathogenesis of IBD is currently under intense investigation. In particular, Th17 cells express on their surface the IL-23 receptor. It has been recently reported that variations in the gene encoding the IL-23 receptor subunit are associated with CD [28]. IL-23, a cytokine needed for the production of IL-17, shares the common subunit p40 with IL-12 (a key cytokine for IFN-γ production in the Th1 response). In this context, anti-p40 antibodies, originally thought to block IL-12 and thus of interest for the treatment of CD, also down-regulate IL-23 and, downstream, IL-17. Thus, the beneficial effects of these antibodies, which were originally ascribed to inhibition of the Th1 response (IL-12 and IFN-γ), may indeed be determined by inhibition of the Th17 response (IL-23 and Th17 cells) [36].

Microbial Factors

Commensal bacteria are involved in the pathogenesis of human IBD and in experimental colitis. In at least 11 different animal models, colitis and immune activation fail to develop in the absence of commensal bacteria [37], while multiple animal models of colitis respond to antibiotics and probiotics. *Klebsiella* monoassociation induces moderate pancolitis [37] and *Bifidobacterium animalis* monoassociation causes distal colonic and duodenal inflammation [38]. These results demonstrate that even a traditionally probiotic bacterial species can induce inflammation in a susceptible host, raising concern over the safety of probiotic therapy in some patients. IBD patients can respond favorably to antibiotic and probiotic treatment [39]. Antibiotics

are effective for Crohn's colitis but not isolated ileitis, except in the postoperative state in which loss of the ileocecal valve probably changes the luminal microenvironment. The dominant bacterial stimuli are different in ileal and colonic CD, such that commensal bacteria probably have a more important role in CD and pouchitis than in UC; thus, subsets of patients will respond selectively to individual treatments. There is reasonable experimental support for only two pathogens in the development of CD. The granulomatous inflammation of CD resembles that of intestinal tuberculosis or the spontaneous enteritis caused by *Mycobacterium avium* subspecies *paratuberculosis* (MAP) in ruminants, which is known as Johne's disease [40]. Noncontrolled studies have reported that up to 84% of patients respond to treatment with combinations of antibiotics effective against MAP [40,41]. It is more likely that this relatively common environmental agent selectively colonizes or lodges in the ulcerated mucosa of CD patients but does not cause disease. Darfeuille-Michaud and Colombel recovered a virulent strain of *Escherichia coli* from 22% of mucosal biopsies from CD patients. These results are consistent with reports of immunohistochemical evidence of increased *E. coli* mucosal adherence, invasion of ulcers and fistulae, and their presence within lamina propria macrophages in CD patients [42,43], and with the concept of defective clearance of intracellular infections by patients with *CARD15* polymorphisms [44]. An altered balance of beneficial versus aggressive microbial species could lead to a proinflammatory luminal milieu that drives chronic intestinal inflammation in a susceptible host. Numerous studies have implicated several commensal organisms, such as *E. coli* as well as *Bacteroides*, *Enterococcus,* and *Klebsiella* species, in the pathogenesis of experimental intestinal inflammation and human IBD [37]. By contrast, various *Lactobacillus* and *Bifidobacterium* species have predominantly protective effects and have been used therapeutically as probiotics [37]. Dietary components can alter the composition and virulence of enteric commensal bacteria, providing a potential explanation for the marked increase in the incidence of IBD in Western countries in the second half of the twentieth century, and more recently in Eastern countries, as they adopt Western dietary practices [45]. A defect in mucosal barrier integrity could lead to increased uptake of luminal antigens and/or adjuvants that overwhelm the net suppressive tone of the mucosal immune system. Alternatively, a defect in epithelial repair could potentiate damage by environmental triggers, such as NSAIDs or infections, that cause only transient damage in normal hosts. The association of *CARD15* with CD and the therapeutic activity of granulocyte-macrophage colony-stimulating factor (GM-CSF) support the novel hypothesis that CD is the result of defective bacterial killing.

References

1. Xavier RJ, Podolsky DK (2007) Unravelling the pathogenesis of inflammatory bowel disease. Nature 448:427-433
2. Cohen ML (2000) Changing patterns of infectious disease Nature 406:762-767
3. Gent AE, Hellier MD, Grace RH et al (1994) Inflammatory bowel disease and domestic hygiene in infancy. Lancet 343:766-767

4. Bach JF (2002) The effect of infections on susceptibility to autoimmune and allergic disease. N Engl J Med 347:911-920
5. Danese S, Fiocchi C (2006) Etiopathogenesis of inflammatory bowel disease. World J Gastroenterol 12(30):4807-4812
6. Mahid SS, Minor KS, Stromberg AJ, Galandiuk S (2007) Active and passive smoking in childhood is related to the development of inflammatory disease. Inflamm Bowel Dis 13:431-438
7. Lindberg E, Jarnerot G, Huitfeldt B (1992) Smoking in Crohn's disease: effect on localization and clinical course. Gut 33:779-782
8. Russel M, Volovics A, Schoon E et al (1988) Inflammatory bowel disease: is there any relationship between smoking status and disease presentation? European Collaborative IBD Study Group. Inflammatory Bowel Dis 4:182- 186
9. Cottone M, Rosselli M, Orlando A et al (1994) Smoking habits and recurrence in Crohn's disease. Gastroenterology 106:643-648
10. Jones DT, Osterman MT, Bewtra M, Lewis JD (2008) Passive smoking and inflammatory bowel disease: a meta-analysis. Am J Gastroenterol 103(9):2382-2393
11. King TE Jr, Savici D, Campbell PA (1988) Phagocytosis and killing of Listeria monocytogenes by alveolar macrophages: smokers versus nonsmokers. J Infect Dis 158:1309-1316
12. Green JT, Richardson C, Marshall RW et al (2000) Nitric oxide mediates a therapeutic effect of nicotine in ulcerative colitis. Aliment Pharmacol Ther 14:1429-1434
13. Suenaert P, Bulteel V, Den Hond E et al (2000) The effects of smoking and indomethacin on small intestinal permeability. Aliment Pharmacol Ther 14:819-822
14. Danese S (2007) Inflammation and the mucosal microcirculation in inflammatory bowel disease: the ebb and flow. Curr Opin Gastroenterol 23:384-389
15. Hatoum OA, Binion DG, Otterson MF et al (2003) Acquired microvascular dysfunction in inflammatory bowel disease: loss of nitric oxide-mediated vasodilatation. Gastroenterology 125:58-69
16. Timmer A, Sutherland LR, Martin F (1998) 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 114:1143-1150
17. Wakefield AJ, Sawyerr AM, Hudson M et al (1991) Smoking, the oral contraceptive pill, and Crohn's disease. Dig Dis Sci 36:1147-1150
18. Corrao G, Tragnone A, Caprilli R et al (1998) Risk of inflammatory bowel disease attributable to smoking, oral contraception and breastfeeding in Italy: a nationwide case-control study. Int J Epidemiol 27:397-404
19. Sears MR, Greene JM, Willan AR et al (2002) Long-term relation between breastfeeding and development of atopy and asthma in children and young adults: a longitudinal study. Lancet 360:901-907
20. Kucharzik T, Maaser C, Lugering A et al (2006) Recent understanding of IBD pathogenesis: implications for future therapies. Inflamm Bowel Dis 12:1068-1083
21. Baumgart DC, Carding SR (2007) Inflammatory bowel disease: cause and immunobiology. Lancet 369:1627-1640
22. Nenci A, Becker C, Wullaert A (2007) Epithelial NEMO links innate immunity to chronic intestinal inflammation. Nature 446:557-561
23. Wehkamp J, Salzman NH, Porter E (2005) Reduced Paneth cell α-defensins in ileal Crohn's disease. Proc Natl Acad Sci 102:18129-18134
24. McVay LD, Keilbaugh SA, Wong TM (2006) Absence of bacterially induced RELMβ reduces injury in the dextran sodium sulphate model of colitis. J Clin Invest 116:2914-2923
25. Van der Sluis M, De Koning BA, De Bruijn AC (2006) Muc2-deficient mice spontaneously develop colitis, indicating that MUC2 is critical for colonic protection. Gastroenterology 131:117-129
26. Cobrin GM, Abreu MT (2005) Defects of mucosal immunity leading to Crohn's disease. Im-

munol Rev 206:277-295
27. Targan SR, Karp LC (2005) Defects of mucosal immunity leading to ulcerative colitis. Immunol Rev 206:296-305
28. Cho JH (2008) The genetics and immunopathogenesis of inflammatory bowel disease. Nature Rev Immunol 8:458-465
29. Hedl M, Li J, Cho JH, Abraham C (2007) Chronic stimulation of Nod2 mediates tolerance to bacterial products. Proc Natl Acad Sci 104:19440-19445
30. Rioux JD, Xavier RJ, Taylor KD (2007) Genome-wide association study identifies new susceptibility loci for Crohn disease and implicates autophagy in disease pathogenesis. Nat Genet 39:596-604
31. Weaver CT, Hatton RD, ManganPR, Harrington LE (2007) IL-17 family cytokines and the expanding diversity of effector T cell lineages. Annu Rev Immunol 25:851-852
32. Jiang H, Chess L (2004) An integrated view of suppressor T cell subsets in immunoregulation. J Clin Invest 114:1198-1208
33. Himmel ME, Hardenberg G, Piccirillo CA et al (2008) The role of T-regulatory cells and Toll-like receptors in the pathogenesis of human inflammatory bowel disease. Immunology 125:145-153
34. Steinman L (2007) A brief history of Th17, the first major revision in the Th1/Th2 hypothesis of T cell–mediated tissue damage. Nat Med 13:139-145
35. Annunziato F, Cosmi L, Santarlasci V et al (2007) Phenotypic and functional features of human Th17 cells. J Exp Med 204:1849-1861
36. Fuss IJ, Becker C, Yang Z (2006) Both IL-12p70 and IL-23 are synthesized during active Crohn's disease and are down-regulated by treatment with anti-IL-12 p40 monoclonal antibody. Inflamm Bowel Dis 12:9-15
37. Sartor RB (2004) Microbial influences in inflammatory bowel disease: role in pathogenesis and clinical implications. In: Sartor RB and Sandborn WJ (eds) Kirsner's inflammatory bowel diseases. Elsevier, Philadelphia, pp 138–162
38. Moran JP et al (2006) Bifidobacterium animalis causes mild inflammatory bowel disease in interleukin-10 knockout mice [abstract]. Gastroenterology 130:A6
39. Sartor RB (2004) Therapeutic manipulation of the enteric microflora in inflammatory bowel diseases: antibiotics, probiotics and prebiotics. Gastroenterology 26:1620–1633
40. Sartor RB (2005) Does Mycobacterium avium subspecies paratuberculosis cause Crohn's disease? Gut 54:896–898
41. Shafran I, Kugler L, El-Zaatari FA et al (2002) Open clinical trial of rifabutin and clarithromycin therapy in Crohn's disease. Dig Liver Dis 34:22–28
42. Swidsinski , Ladhoff A, Pernthaler A et al (2002) Mucosal flora in inflammatory bowel disease. Gastroenterology 122:44–54
43. Liu Y Liu Y, van Kruiningen HJ, West AB et al (1995) Immunocytochemical evidence of Listeria, Escherichia coli, and Streptococcus antigens in Crohn's disease. Gastroenterology 108:1396–1404
44. Hisamatsu T, Suzuki M, Reinecker HC et al (2003) CARD15/NOD2 functions as an anti-bacterial factor in human intestinal epithelial cells. Gastroenterology 124:993–1000
45. Loftus EV Jr (2004) Clinical epidemiology of inflammatory bowel disease: incidence, prevalence, and environmental influences. Gastroenterology 126:1504–1517

Epidemiology

2

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Introduction

Epidemiology studies the frequency of illness and in some cases can suggest causes and mechanisms for chronic idiopathic diseases, including Crohn's disease (CD). One well-received current hypothesis for the etiology of inflammatory bowel diseases (IBD) is a genetic predisposition to dysregulation of the gastrointestinal immune system. This possibility is supported by studies of monozygotic twins, in which the concordance of CD is 50–60%. However, the remaining 50% are discordant for CD, suggesting that environmental factors also play an important role in disease development.

In epidemiology, both the incidence and the prevalence of a disease are important parameters. The frequency of newly diagnosed cases of a disease over a definite time interval is defined as the incidence and it is expressed as a rate. In CD literature, the number of cases is conventionally reported per 100,000 person-years. Prevalence measures the proportion of individuals of a population that, at any given time, have the disease. It is expressed as cases per 100,000 persons.

Geographical and Genetic Factors

There are several reports concerning the incidence of CD in many regions of the world [1]. Some parts of the world are historically associated with CD. Specifically, Northern Europe [2,3], the UK [4,5], and North America [6–10], which have the highest incidence and prevalence of the disease. More recently, however, the inci-

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dence and prevalence of CD have been increasing in other areas of the world, i.e., Southern and Central Europe [11–13], Asia [14], Africa, and South America [15]. In North America, incidence rates range from 3.6 (South California) to 14.6 (Manitoba, Canada) cases per 100,000 person-years and prevalence from 26 to 199 cases per 100,000 persons. In Europe, incidence rates are between 0.7 (Croatia) and 9.8 (Scotland) cases per 100,000 person-years while prevalence ranges from 8.3 (Croatia) to 214 (UK).Thus, a "north-south gradient" is often cited because the rates seem to be highest in northern countries [7–9], but this difference has lessened [2] as developing countries "gain affluence" [16]. Typical examples of this trend are Eastern Europe countries [17]. Historically, CD was rare in Middle and South America, Asia, and Africa, with the exceptions of Israel and South Africa. Recently, the incidence of CD has grown in many countries of these regions, including in Japan, South Korea, Singapore [14], India, South America while all of which were previously characterized by a very low incidence of CD [15]. Data from Japan and Korea suggest that urbanization and industrialization, and ultimately a more "Western" lifestyle as responsible for the change in the incidence [18]. Nonetheless, it must be underlined that in most of these areas CD remains a rare disease. Also noteworthy is the fact that the incidence of CD is stable in many high-incidence areas, such as Scandinavia and Minnesota. In Olmsted County, Minnesota, the prevalence of CD is rising while its incidence is stable, perhaps because of the increase in the average life-span of CD patients, the earlier age of disease onset or earlier diagnosis, and the growing number of immigrants [19].

An analysis of the temporal and geographic tendencies in CD incidence and of the time course of the disease shows that the incidence appears to be low in developing countries. This observation may reflect the limited availability of diagnostics and in general the low diagnostic capacities. Moreover, in these areas there is a frequent diagnostic overlap with infectious causes of diarrhea.

Aside from environmental factors, are there differences in the incidence of CD that involve genetics? According to literature reports, the incidence of CD among African-Americans is approaching that of white Americans. Regarding the course of the disease, hospitalization rates for CD are superimposable among whites and blacks in southern California, and the prevalence of CD among whites is about one-third higher than among blacks [6]. It seems that Asian-Americans, Americans of Hispanic background, and aboriginal North Americans are less likely to develop CD [20], although available data are limited. In a British prospective study, the risk of CD among African-Caribbean and white children was similar [21]. The incidence of CD in Derby, UK, evaluated in a retrospective study, was likewise found to be similar among whites and African-Caribbean adults [22].

Hormonal Factors

As a rule, in CD there is a small female predominance, although in some low-incidence areas the disease is more prevalent in males. The latter might be explained by

the reluctance of women in those countries to undergo a physical examination and, in general, by the decreased attention to women's health in under-developed regions of the world.

The female predominance, especially among women in late adolescence and early adulthood, has led to suggestions that hormonal factors are involved in the expression of CD. This link is strengthened by the increased risk of CD in women who take oral contraceptives, reported in some case-control and cohort studies from the United States [23] and the UK [24]. The risk of CD was reported to be three-fold higher than in women not taking contraceptives; however, after the results were adjusted to take cigarette smoking into account, they were not statistically significant. A dose-response effect, with increased risk related to the prolonged [25] and high-dose [26] intake of estrogen has been reported. Data about the role of contraceptives in the course of existing CD are contradictory. Timmer et al. showed that oral contraceptive use is a predictor of relapse [27], while Cosnes et al. did not find an association with recurrence [28]. Ultimately, a weak association between oral contraceptive use and CD seems likely, perhaps mediated by the thrombogenic properties of oral contraceptives and by alterations of the intestinal microcirculation.

In the light of what we know about the higher rates of venous thromboembolism in CD patients compared to non-IBD controls (OR 1.48, 95% CI 1.35–1.62) [29] and about the thrombogenic properties of estrogen [30], it seems reasonable to recommend that CD patients with additional risk factors for thromboembolic disease should avoid oral contraceptives and other forms of exogenous estrogen.

Age

Crohn's disease is mostly diagnosed in young people, with the mean age at diagnosis ranging from 33.4 to 45 years according to the results of a systematic review of population-based cohorts of CD from North America [31]. Another study reported a median age at diagnosis of 29.5 years [7]. A bimodal age distribution has been proposed, with an incidence peak around 20–30 years and a second, smaller peak in later decades. However, this has not been unequivocally confirmed by more recent epidemiological studies. In particular it remains unclear whether the differences in age distribution are real or represent variations in the diagnostic criteria and/or classification of CD.

Cigarette Smoking

Several studies have shown that cigarette smoking is a risk factor for CD and that smokers have a two-fold higher risk of developing CD than non-smokers [32]. Nevertheless, this association between smoking and CD is not seen in some ethnic groups or geographic regions, for example in the Israeli Jewish population [33].

Moreover, the clinical course and expression of CD may be influenced by smoking. In smokers, CD more frequently involves the ileum [34] and the disease is purely inflammatory [35]. If CD patients continue their smoking habit after surgical resection, the risk of recurrent disease is increased [36,37] and the need for second surgery is more than four times higher than in non-smokers [36]. Also, CD patients who smoke have a greater need of immunosuppressants [37]. The discontinuation of smoking reduces the number and the severity of disease flares and thus spares patients the need for corticosteroid and immunosuppressant therapy [38]. In children, the exposure to passive cigarette smoke seems to increase the risk of developing CD, according to a Swedish study [39].

Appendectomy

Several studies have examined the relationship between appendectomy and the risk of these patients to develop CD [40]. Most of these analyses showed a trend but not a statistically significant relationship between these two events. Moreover, we have to consider that symptoms of CD and appendicitis can be similar, and many diagnoses of appendicitis can mask an undiagnosed CD. Assuming that appendectomy indeed increases the risk of CD, we can propose that removal of the appendix causes an imbalance of the gut immune system, which may be responsible or co-responsible for initiating the disease [40]. A similar explanation for the inverse correlation between ulcerative colitis and absence of previous appendectomy has been suggested.

Dietary Factors

Diet might be involved in the development of CD, and a plausible role for diet is based on the fact that luminal antigens are mainly represented by dietary and bacterial antigens. Moreover, the differences in CD risk across different countries and the increases in CD incidence in migrant populations can be explained by differences in diet habit. In the studies available thus far, the only consistent association was between increased sugar intake and CD, identified by Martini et al. [41] and confirmed in several case-control studies [42].

Childhood Factors and Infection

Events in early childhood have been suggested to shape and modify the risk of CD and disease expression. While several factors have been studied for an association with CD, including type of feeding (in particular breastfeeding), domestic hygiene, and perinatal infections, a significant association has not been found. The only data

of interest in this respect are a four-fold increased risk for CD in patients with a recorded perinatal health event, such as infection or illness in mother or child, and a three-fold increased risk for CD in infants from families of low socioeconomic status, reported by Ekbom et al. [43].

The analogies between tuberculosis and CD have resulted in controversy for over 20 years [44]. *Mycobacterium avium* subspecies *paratuberculosis* (MAP) causes chronic diarrhea in ruminants and other species, with histological evidence of a granulomatous inflammation of the intestine. In the 1980s, MAP was found in the intestinal specimens of patients with CD [45]. Initially, higher rates of MAP antibody in CD patients were measured in seroprevalence studies, but subsequent investigations could not confirm this result [46]. Moreover, many open-label studies reported a clinical improvement of CD in patients administered drugs with antimycobacterial activity [46–48], but, again, randomized clinical trials could not confirm these findings.

Conclusions

The role of epidemiology is not only to describe the impact of diseases in a population and to study associations between variables and disease, but also to provide a useful key to understanding the causes and mechanisms underlying disease development.

In CD, genetic factors appear to be important in the pathogenesis of the disease, but environmental factors can significantly modify its expression. Factors so far identified that can alter the risk and expression of CD include a family history of IBD, cigarette smoking, and appendectomy.

It is essential to continue research to identity the risk factors of CD and their ability to influence the disease expression in the hope of finding the root causes of CD and start looking for means to remove them.

References

1. Loftus EV Jr, Sandborn WJ (2002) Epidemiology of inflammatory bowel disease. Gastroenterol Clin North Am 31:1–20
2. Shivananda S, Lennard-Jones J, Logan R et al (1996) 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 39:690–697
3. Bjornsson S, Johannsson JH (2000) Inflammatory bowel disease in Iceland, 1990–1994: a prospective, nationwide, epidemiological study. Eur J Gastroenterol Hepatol 12:31–33
4. Yapp TR, Stenson R, Thomas GAO et al (2000) Crohn's disease incidence in Cardiff from 1930: an update for 1991–1995. Eur J Gastroenterol Hepatol 12:907–911
5. Rubin GP, Hungin APS, Kelly PJ, Ling J (2000) Inflammatory bowel disease: epidemiology and management in an English general practice population. Aliment Pharmacol Ther 14:1553–1559
6. Kurata JH, Kantor–Fish S, Frankl H et al (1992) Crohn's disease among ethnic groups in a

large health maintenance organization. Gastroenterology 102:1940–8194
7. Loftus EV Jr, Silverstein MD, Sandborn WJ et al (1998) Crohn's disease in Olmsted County, Minnesota, 1940–1993: incidence, prevalence, and survival [published erratum appears in Gastroenterology (1999) 116:1507. Gastroenterology 114:1161–1168
8. Bernstein CN, Blanchard JF, Rawsthorne P, Wajda A (1999) Epidemiology of Crohn's disease and ulcerative colitis in a central Canadian province: a population–based study. Am J Epidemiol 149:916–924
9. Loftus EV Jr, Silverstein MD, Sandborn WJ et al (2000) Ulcerative colitis in Olmsted County, Minnesota, 1940–1993: incidence, prevalence, and survival. Gut 46:336–343
10. Loftus CG, Loftus EV, Sandborn WJ et al (2003) Update on incidence and prevalence of Crohn's disease and ulcerative colitis in Olmsted County, Minnesota (abstr) Gastroenterology 124:A36
11. Manousos ON, Koutroubakis I, Potamianos S et al (1996) A prospective epidemiologic study of Crohn's disease in Heraklion, Crete. Incidence over a 5–year period. Scand J Gastroenterol 31: 599–603
12. Trallori G, Palli D, Saieva C et al (1996) A population–based study of inflammatory bowel disease in Florence over 15 years (1978–92). Scand J Gastroenterol 31:892–899
13. Tragnone A, Corrao G, Miglio F et al (1996) Incidence of inflammatory bowel disease in Italy: a nationwide population–based study. Intl J Epidemiol 25:1044–1052
14. Lee YM, Fock KM, See SJ et al (2000) Racial differences in the prevalence of ulcerative colitis and Crohn's disease in Singapore. J Gastroenterol Hepatol 15:622– 625
15. Linares de la Cal JA, Canton C, Hermida C et al (1999) Estimated incidence of inflammatory bowel disease in Argentina and Panama (1987–1993). Rev Esp Enferm Dig 91:277–286
16. Ekborn A (2003) The changing faces of Crohn's disease and ulcerative colitis. In: Targan SR, Shanahan F, Karp LC et al (eds) Inflammatory bowel disease, from bench to bedside. Dordrecht: Kluwer Academic, pp.:5–21
17. Lakatos L, Lakatos PL (2006) Is the incidence and prevalence of inflammatory bowel disease increasing in Western Europe? Postgrad Med J 82:332–337
18. Yang SK, Loftus EV, Sandborn WJ (2001) Epidemiology of Inflammatory Bowel Disease in Asia. Inflammatory Bowel Diseases 7(3):260–270
19. Loftus CG, Loftus EV, Harmsen WS et al (2007) Update on the incidence and prevalence of Crohn's disease and ulcerative colitis in Olmsted County, Minnesota, 1940–2000. Inflamm Bowel Dis 14:254–261
20. Blanchard JF, Bernstein CN, Wajda A, Rawsthorne P (2001) Small–area variations and sociodemographic correlates for the incidence of Crohn's disease and ulcerative colitis. Am J Epidemiol 154:328–335
21. Sawczenko A, Sandhu BK, Logan RFA et al (2001) Prospective survey of childhood inflammatory bowel disease in the British Isles. Lancet 357:1093–1094
22. Fellows IW, Freeman JG, Holmes GK (1990) Crohn's disease in the city of Derby, 1951–85. Gut 31:1262–1265
23. Ramcharan S (1981) The Walnut Creek Contraceptive Drug Study: a prospective study of the side effects of oral contraceptives. DHEW Publication No. 74–562. US Government Printing Office, Washington DC
24. Vessey M, Jewell D, Smith A et al (1986) Chronic inflammatory bowel disease, cigarette smoking, and use of oral contraceptives: findings in a large cohort study of women of childbearing age. BMJ 292:1101–1103
25. Godet PG, May GR, Sutherland LR (1995) Meta–analysis of the role of oral contraceptive agents in inflammatory bowel disease. Gut 37:668–673
26. Boyko EJ, Theis MK, Vaughan TL, Nicol-Blades B (1994) Increased risk of inflammatory bowel disease associated with oral contraceptive use. Am J Epidemiol 140:268–278
27. Timmer A, Sutherland LR, Martin F (1998) Oral contraceptive use and smoking are risk factors for relapse in Crohn's disease. Gastroenterology 114:1143–115

28. Cosnes J, Carbonnel F, Carrat F et al (1999) Oral contraceptive use and the clinical course of Crohn's disease: a prospective cohort study. Gut 45:218–222
29. www.ebgh.it/ebgh/areetematiche/ibd/40-ibd/291-prevalenza-e-mortalità-per-tromboembolismo-nelle-ibd.html
30. Lacut K, Mottier D, Pottier P (2008) Drugs and venous thromboembolism. Rev Pneumol Clin 64(6):290–297
31. Loftus EV Jr, Schoenfeld P, Sandborn WJ (2002) The epidemiology and natural history of Crohn's disease in population–based patient cohorts from North America: a systematic review. Aliment Pharmacol Ther 16:51–60
32. Calkins BM (1989) A meta-analysis of the role of smoking in inflammatory bowel disease. Dig Dis Sci 34:1841–1854
33. Reif S, Lavy A, Keter D et al (2000) Lack of association between smoking and Crohn's disease but the usual association with ulcerative colitis in Jewish patients in Israel: a multicenter study. Am J Gastroenterol 95:474–478
34. Russel MG, Volovics A, Schoon EJ et al (1998) Inflammatory bowel disease: is there any relation between smoking status and disease presentation? Inflamm Bowel Dis 4:182–186
35. Picco MF, Bayless TM (2003) Tobacco consumption and disease duration are associated with fistulizing and stricturing behaviors in the first 8 years of Crohn's disease. Am J Gastroenterol 98:363–368
36. Sutherland LR, Ramcharan S, Bryant H, Fick G (1990) Effect of cigarette smoking on recurrence of Crohn's disease. Gastroenterology 98:1123–1128
37. Cosnes J, Carbonnel F, Carrat F et al (1999) Effects of current and former cigarette smoking on the clinical course of Crohn's disease. Aliment Pharmacol Ther 13:1403–1411
38. Cosnes J, Beaugerie L, Carbonnel F, Gendre JP (2001) Smoking cessation and the course of Crohn's disease: an intervention study. Gastroenterology 120:1093–1099
39. Lindberg E, Jarnerot G, Huitfeldt B (1992) Smoking in Crohn's disease: effect on localisation and clinical course. Gut 33:779–782
40. Radford-Smith GL (2003) The role of the appendix and appendectomy in patients with IBD. IBD Monitor 4:120–128
41. Martini GA, Brandes JW (1976) Increased consumption of refined carbohydrates in patients with Crohn's disease. Klin Wochen 54:367–371
42. Riordan AM, Ruxton CH, Hunter JO (1998) A review of associations between Crohn's disease and consumption of sugars. Eur J Clin Nutr 52:229–338
43. Ekbom A, Adami HO, Helmick CG et al (1990) Perinatal risk factors for inflammatory bowel disease: a case–control study. Am J Epidemiol 132:1111–1119
44. Greenstein RJ (2003) Is Crohn's disease caused by a mycobacterium? Comparisons with leprosy, tuberculosis, and Johne's disease. Lancet Infect Dis 3:507–514
45. Chiodini RJ, Van Kruiningen HJ, Merkal RS et al (1984) Characteristics of an unclassified Mycobacterium species isolated from patients with Crohn's disease. J Clin Microbiol 20:966–971
46. Van Kruiningen HJ (1999) Lack of support for a common etiology in Johne's disease of animals and Crohn's disease in humans. Inflamm Bowel Dis 5:183–191
47. Shafran I, Kugler L, El–Zaatari FA et al (2002) Open clinical trial of rifabutin and clarithromycin therapy in Crohn's disease. Dig Liver Dis 34:22–28
48. Borody TJ, Leis S, Warren EF, Surace R (2002) Treatment of severe Crohn's disease using antimycobacterial triple therapy – approaching a cure? Dig Liver Dis 34:29–38

Genetics

3

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Introduction

In the last few years, the application of molecular studies in medicine has dramatically increased our knowledge of the biological basis of human diseases. This is evident not only for rare single-gene disorders, but also for many common disorders, including Crohn's disease (CD).

The genetics of CD parallels those of ulcerative colitis, because most genetic studies have been performed and/or replicated in both disorders with similar results [1]. These two conditions are autoimmune disorders of the gastrointestinal tract, grouped under the umbrella terminology of inflammatory bowel disease (IBD). This grouping was originally guided by well-consolidated clinical and pathogenic similarities. More recently, an increasing number of genetic studies have demonstrated that this phenotypic overlap is mirrored by a relatively common genetic background. From a molecular perspective, these findings suggest that CD and ulcerative colitis most probably represent two ends of a continuous clinical spectrum that is partly explained by the same etiopathogenic mechanism. The evidence that at least 25–33% of IBD patients with chronic inflammation of the gastrointestinal mucosa cannot be definitively diagnosed with either CD or ulcerative colitis and are therefore classified as having "indeterminate colitis" further illustrates this assumption [2]. Additional support of this hypothesis is provided by evidence that, among familial cases of IBD, more than 20% of affected family members are discordant for the IBD phenotype [3].

This chapter summarizes current knowledge of the genetic/molecular basis of CD, defines the applications and limits of genetic counseling in this complex disorder, and discusses future perspectives in the growing field of genetics.

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Crohn's Disease. Roberto Tersigni, Cosimo Prantera (Eds.)

3

Genetic Susceptibility in Crohn's Disease

A first clue to the possible role of a genetic factor in the pathogenesis of CD was the observed discrepancy in disease incidence among different populations. CD (as well as ulcerative colitis) is a relatively common disorder in North America and Europe, and an accurate estimation of disease frequency in the various European countries is now available [4,5]. By contrast, CD is rarer in other regions of the world, including Japan, India, Latin America, and Singapore (for further reading, see the chapter on "Epidemiology and Pathophysiology") [6]. However, these differences may also reflect lifestyle and environmental factors. Accordingly, recent studies have demonstrated an increasing trend in incidence in several Asian countries, in which CD was previously considered rare [7]. Possible non-genetic factors include modification of dietary habits, improved sanitation, and industrialization. Moreover, migration studies indicate that disease incidence rises when populations move from low-risk to higher-risk geographic areas [8]. Thus, taken together, this evidence offers only weak support for a major role of genetic predisposition in the pathogenesis of CD.

A stronger indication of the existence of a genetic susceptibility in CD is offered by family and twin studies. It is well known that 5–20% of CD patients have a family history of IBD [9,10]. However, in most familial cases the disease clearly does not segregate as a Mendelian (i.e., monogenic) trait. Robust data collected by multiple studies show that monozygotic twin concordance is higher than dizygotic twin concordance (20–50% vs. 0–7%, respectively) [11–14]. Moreover, the lack of complete concordance among monozygotic twins further supports a role for one or more environmental factors in disease pathogenesis. Together, these data indicate that no single gene mutation is sufficient and/or necessary to cause CD, which, accordingly, can be considered a multifactorial disorder. In contrast to monogenic conditions, in multifactorial (or complex) disorders genetic variants do not cause the disease, but rather merely modify individual susceptibility to it. Genetic variations (polymorphisms) involved in multifactorial disorders are frequently (>1%) encountered in the general population. Therefore, in any given individual, the presence/absence of specific polymorphisms increases or decreases her/his relative risk of developing the associated disorder. The disease is therefore ultimately caused by a variable mixture of genetic, environmental and, probably, stochastic factors that cooperate in its etiopathogenesis (Fig. 3.1).

Unraveling the pathogenic sequence variants implicated in disease development is dramatically more difficult in complex disorders than in monogenic diseases. In the second half of the 1990s, a multitude of studies tried to identify inherited susceptibility markers for CD. The vast majority of these works consisted of case-control association and linkage studies. These methods tested the likelihood of an association between a known polymorphism or chromosomal segment (locus) and CD. The power of these associations was evaluated by statistical analyses and the results usually expressed as an odds ratio and LOD (logarithm of odds) score. Nonetheless, despite the association strength that may be established in a single study, a major limit of this type of work is the need for further evidence in order for the findings to

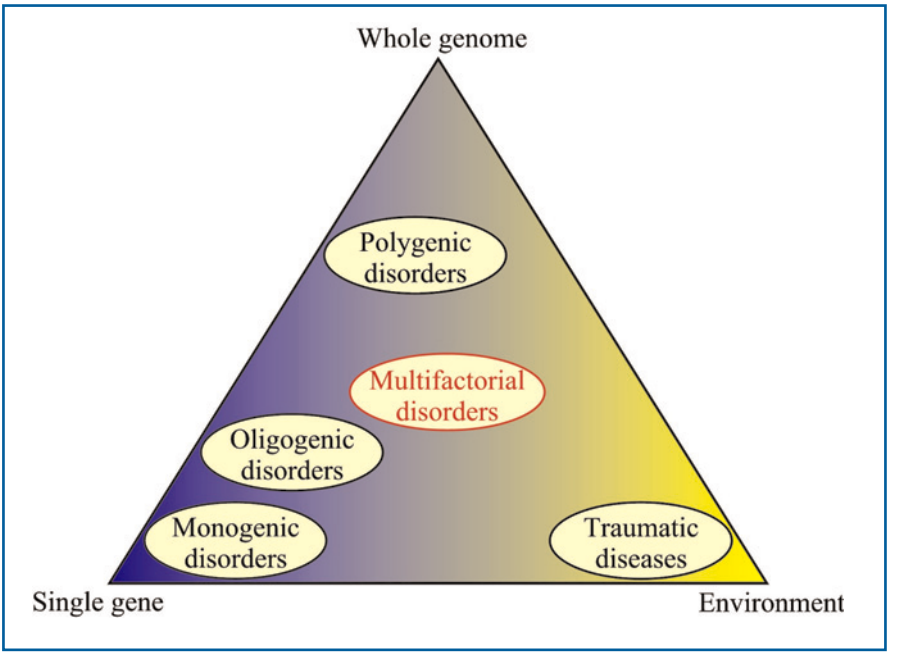

Fig. 3.1 Ideogram illustrating the relationships between monogenic, oligogenic, polygenic, multifactorial, and environmental diseases. In monogenic disorders (*left*), the clinical features are caused by mutations in single genes. However, lack of penetrance and variable expressivity may reflect minor influences of modifier genes, environmental factors, and stochastic events. Oligogenic and polygenic disorders are caused by mutations in 2–3 genes and >3 genes, respectively. Therefore, the disease is not determined by single mutations but results from the concurrence of several or many pathogenic variants in multiple loci. Multifactorial disorders (*middle*) are caused by the interaction between complex genetic susceptibility and non-genetic factors, which are equally essential for disease development. Finally, there are environmental disorders (*right*), including traumatic diseases, in which genetic predisposition plays very little role in disease pathogenesis

be considered scientifically relevant. In fact, among the astonishingly high number of genes/loci claimed to be associated with CD, only a minority of them has been identified in other populations and/or supported by functional studies.

Since 2007, research in the field of CD genetics has moved from studies testing one or more genetic variants to analyses that screen the entire genome simultaneously in hundreds of patients (genome-wide association studies). In these studies, genotyping platforms assess several hundred thousand single-nucleotide polymorphisms and compare allele frequencies in individuals with the disease vs. healthy controls. With rigorous sample selection, genome-wide association studies have been able to confirm previous associations and to identify new loci/polymorphisms linked to CD (for a review, see [1]). To date, more than 40 loci have been associated with CD (Table 3.1); however, most of these associations are inconsistent or, as yet, preliminary, demonstrated in single populations and/or using a relatively small sample size.

Table 3.1 Identified susceptibility loci/genes in Crohn's disease

Chromosome region	Locus name	Gene(s)	Reference(s)
1p31.1	IBD17	IL23R	[15,16]
1p36	IBD7	PIK3CD	[17]
1q31-q32	IBD23	IL10	[18]
1q44	–	NLRP3	[19]
2q14	–	IL1B	[20]
2q14.2	–	IL1RA	[20]
2q37.1	IBD10	ATG16L1	[21]
3p21.3	IBD12	MST1	[22,23]
3p21.3	–	TLR9	[24]
3p26	IBD9	–	[25]
4p14	–	TLR1	[26]
4p14	–	TLR6	[26]
4q23-q24	–	NFKB1	[27]
4q32	–	TLR2	[26]
5p13.1	IBD18	PTGER4	[28,29]
5q31-q33	IBD5	SLC22A4, SLC22A5	[30,31]
5q31.1-q33.1	–	IL12B	[16]
5q33.1	IBD19	IRGM	[32]
6p21.3	IBD3	HLA (class II),	[33–35]
	–	MICA, TNFA	
7p15-p14		NOD1/CARD4	[36]
7q21	–	IL6	[37]
7q21.1	IBD13	ABCB1/MDR1	[38]
7q22	IBD11	MUC3A	[39]
7q32	IBD14	IRF5	[40]
9q32	IBD16	TNFS15	[41]
9q32-q33	–	TLR4	[42]
10q21	IBD15	–	[43]
10q23-q24	IBD20	DLG5	[23,44]
10q24.2	–	NKX2.3	[32,33]
11q22.2-q22.3	–	IL18	[45]
12p13.2-q24.1	IBD2	VDR, STAT6,	[46]
		INFG, MMP18	
14q11-q12	IBD4	IL25, TCR	[47]
15q26.1	–	IL16	[48]
16p	IBD8	–	[49]
16q12	IBD1	NOD2/CARD15	[50,51]
17q21	IBD22	STAT3	[23]
18p11	IBD21	PTPN2	[33]
19p13	IBD6	ICAM1	[52]
19p13.1	–	MYO9B	[53]
19p13.3	–	TUCAN/CARD8	[54]

Specific Loci Associated with Crohn's Disease

The recent CD immunopathogenesis paradigm suggests that a defective immune response impairs the clearance of luminal antigens and/or pathogens and leads to the development of chronic intestinal inflammation. Factors that very probably contribute to this process include alterations in: (i) barrier function of the intestinal epithelium, (ii) innate immune cells, (iii) lymphocyte function in the gut lamina propria and mesenteric lymph nodes, and (iv) lymphocyte- and stromal-cell-derived factors [1]. Accordingly, many of the CD-associated genes encode proteins expressed in one or more of these cell lines and involved in the immune response.

In CD, the strongest known association is with the *NOD2* gene [51,55]. *NOD2* is located in the long arm of chromosome 16 (16q12) and consists of 12 exons, encoding a protein of 1040 amino acids. NOD2 is a pattern recognition receptor, which, acting as an intracellular sensor activated by peptidoglycan-muramyl dipeptide, is directly involved in the innate immune response. Peptidoglycan-muramyl dipeptide is a barrier constituent in both gram-positive and gram-negative bacteria. Contact with NOD2 stimulates nuclear factor-κB and mitogen-activated protein kinase signaling pathways in paneth cells and macrophages [56,57]. The *NOD2* polymorphisms Arg702Trp, Gly908Arg, and 3020insC are the most common genetic variants associated with CD. In particular, approximately 33% of CD patients have one of these polymorphisms in one or both alleles compared with 10–15% of the normal population of European ancestry [50,58]. However, *NOD2* polymorphisms are absent in Japan, China, and Korea [59–61], and are rare in African Americans [62]. The odds ratio for *NOD2* heterozygotes is 2.4 while for homozygotes it is 17.1 [63]. This strongly indicates that the molecular pathogenesis of CD is different among populations and that *NOD2* is neither necessary nor sufficient for disease development. Additional studies have investigated the role of *NOD2* in the development of specific disease features, such as increased risk for extraintestinal manifestations, ileal localization, early-onset, fistula formation, and fibrostenosis, but the available data are currently too preliminary to be considered useful for clinical practice [63–65]. Details regarding the applications and limits of *NOD2* genetic testing are discussed in the following section.

ATG16L1 is another gene involved in the innate immune response and strongly associated with CD. It is located in 2q37.1 and consists of 17 exons. ATG16L1 has been implicated in autophagy and is expressed by intestinal epithelial cells, antigen-presenting cells, and $CD4^+$, $CD8^+$, $CD19^+$ primary human T cells. The *ATG16L1* single-nucleotide polymorphism Ala281Thr is associated with CD, with the less-common threonine allele conferring protection from disease development [21]. Other genes implicated in the innate immune response have been associated with CD, including *IRGM*, *NLRP3*, *NOD1*, *TLR1*, *TLR2*, *TLR4*, *TLR6*, and *TLR9* [19,24,26,32,36,42]. Interestingly, mutations in one of these genes, *NLRP3*, also have been implicated in rare Mendelian autoinflammatory syndromes, such as chronic infantile neurological, cutaneous, and articular (CINCA) syndrome and Muckle-Wells syndrome. This evidence indicates a pivotal role of *NLRP3* dysregulation in the

development of an abnormal immune response, resulting in autoimmune disorders.

The adaptive immune system also plays a relevant role in CD immunopathogenesis. Recent evidence suggests that the *IL23R* pathway is a key factor in disease development. In fact, IL32R is expressed by natural killer (NK) cells, $CD4^+$ T cells, and $CD8^+$ T cells. Its ligand, IL23, consists of the p19 and p40 subunits, with p19 overexpression resulting in multiorgan inflammation, including in the intestine, in mice [66]. IL23 is also necessary for the maximal induction of inflammation in innate-immune-cell-mediated intestinal inflammation [67]. More specifically, the Arg381Gln polymorphism of *IL23R* is significantly underrepresented in patients with CD compared with controls. The glutamine allele confers an approximately threefold increase (odds ratio = 0.26) in protection against the development of CD [15]. The role of IL23R signaling in CD pathogenesis is further supported by the identification of an association between CD and other genes, namely, *IL12B*, *STAT3*, and *JAK2*, involved in this pathway [16,23]. Finally, earlier studies identified an association between IBD and specific HLA class I and II alleles [34,35]. However, it remains to be further elucidated whether or not these polymorphisms are truly associated with the disease as opposed to being neutral variants in linkage disequilibrium with functional polymorphisms located in neighboring genes.

Genetic Counseling in Crohn's Disease

At the present time, genetic counseling is not a part of the standard multidisciplinary evaluation of patients with CD. In fact, despite recent advances in the field of genetics, the large amount of accumulated information about CD has had little or no role in the differential diagnosis, therapeutic planning, and establishment of prognosis for most affected individuals. However, genetic counseling could support the families of patients with CD for a number of reasons, as summarized in Table 3.2.

According to the theory of multifactorial disorders, unaffected family members of CD patients have a higher chance of developing IBD in their life compared to the general population. This risk does not follow Mendelian laws but is calculated empirically. In particular, the absolute risk of first-degree relatives (siblings, parents, and children) is 3–10%, while the relative risk of siblings (sibling risk compared to the

Table 3.2 Possible indications for genetic counseling in inflammatory bowel disease

1. Consideration of molecular testing in affected individuals and unaffected relatives[a]
2. Co-operating with first-line caregivers in family planning
3. Interpretation of molecular tests performed elsewhere
4. Supporting patients in coping with chronic and heritable disorders

[a]The setting for molecular test(s) in IBD is illustrated in Figure 3.2

risk in the general population) is 30–40 [9,68]. When both parents are affected, the absolute risk for their children rises to 30–50% [9]. For higher-degree relatives, our capacity of estimating risk is largely incomplete, as data on empirical risks are not yet available. Since communicating these risks and the limits of this information could be difficult in specific cases, the involvement of personnel with specific training in genetic counseling is desirable. Genetic counselors are trained in managing not only risk calculation but also inter-individual variability in risk perception and the spectrum of possible emotional responses by those seeking counseling.

A further point that should be stressed in the setting of genetic counseling is prevention. Although, at this time, the prevention strategies that would be effective in individuals at increased risk of developing CD are unknown, most experts recommend that cigarette smoking be avoided [69–72]. This information should be communicated to unaffected relatives together with information on empirical risks. Finally, genetic counselors are able to support patients in coping with the fear of transmitting a chronic and unpredictable disease to their children. In fact, sometimes, parental concerns are exaggerated regarding their child's health status. In these cases, the consequences of parental overprotection could be of greater consequence than the relatively small risk of the child developing the disease [73].

With the recent expansion of the molecular diagnosis of IBD, the number of patients who request predictive testing is increasing. However, the currently available assays are able to detect only the three most common *NOD2* polymorphisms (Arg702Trp, Gly908Arg, and 3020insC), while the number of potentially associated genetic variants is much higher. Therefore, in any given individual, a negative result cannot exclude the presence of other susceptibility markers that may equally influence the final individual risk. In addition, in predictive medicine, the clinical utility of a genetic test primarily depends on effective access to appropriate interventions [74]. Therefore, in the absence of a specific preventive strategy in CD, application of *NOD2* molecular screening is extremely limited. Thus, for now, *NOD2* testing should be performed only in affected subjects. Moreover, it is not indicated in unaffected individuals when an affected relative has a negative screening test. However, in case of a positive result in a CD patient, the same test could be considered in unaffected relatives in order to modify (in terms of increase and decrease) their absolute risk of developing IBD (Fig. 3.2). It should be emphasized that molecular screening for IBD should always be discussed in the setting of genetic counseling, where its applications and limits can be properly discussed.

Future Perspectives

Based on the enormous amount of accumulated data concerning the genetic basis of CD, it has been estimated that a well-established association with this disease accounts for only 20% of the genetic variance observed among patients [23]. Therefore, it is expected that, in the future, the number of associated loci will increase dramatically. This, in turn, will allow better delineation of the genetic fac-

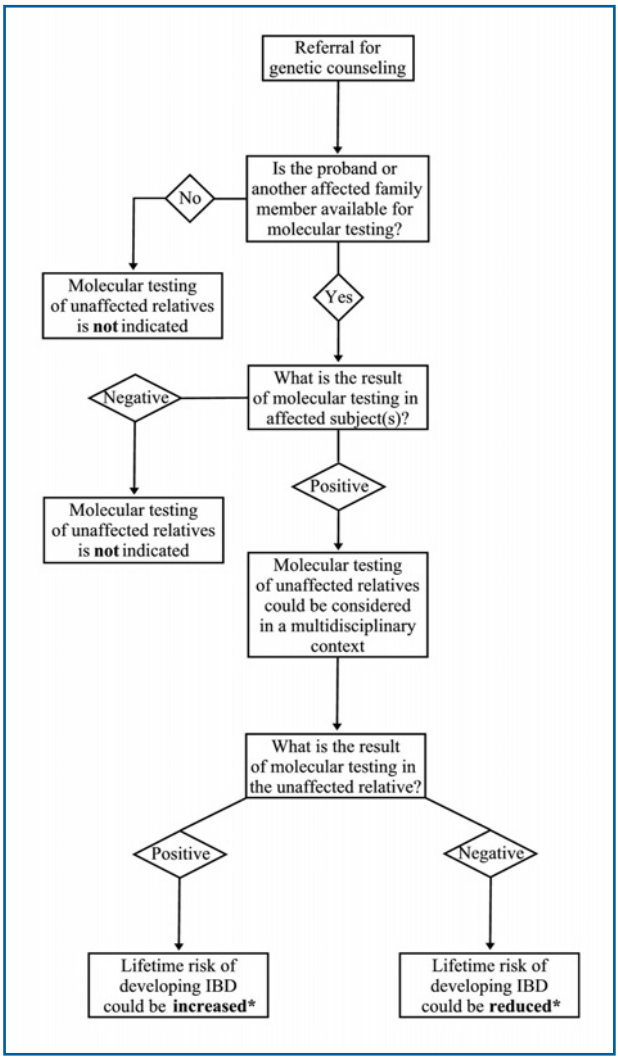

Fig. 3.2 Flow-chart for molecular testing in inflammatory bowel disease. *An interpretation of molecular test results is feasible in first-degree relatives, for whom absolute risks have been estimated, while it is less clear in second- and third-degree relatives because the empirical risks are still unknown

tors underlying the pathogenesis of CD and, potentially, a patient's prognosis. At the present time, the applications of molecular testing for CD in the establishment of the relative risk in unaffected relatives are limited. In the future, more stringent genotype-phenotype correlations between specific polymorphisms and distinct clinical features could predict the course of disease shortly after the diagnosis is made.

The multiplication of molecular studies in CD will certainly increase our understanding of its pathogenesis, with possible relevant implications in identifying environmental factors contributing to development of the disease. This information will

have a major impact on prevention and, possibly, therapeutic planning. In the last few years, a growing field of research into complex diseases has been established. Pharmacogenetics is the study of the association between variability in drug response and drug toxicity and genetic polymorphisms, two aspects that are essential for individualized therapy. Moreover, it enables the exclusion of any given patient determined to be susceptible to a drug's more severe side effects and, conversely, the selection of those drugs that may be better tolerated and more effective. Currently, for CD, the most robust data are those available for thiopurine S-methyltransferase (TPMT) polymorphisms and their influence on azathioprine and 6-mercaptopurine treatment [75]. In particular, based on a combination of three intragenic polymorphisms it is possible to establish a specific risk percentage for hematopoietic toxicity. A number of studies are now testing the effectiveness of searching for specific polymorphisms in other genes in order to guide the use of many drugs, including classic agents as well as the more recent immune-modulators and monoclonal antibodies. However, with the exception of TPMT testing, none of these studies has yet found application in clinical practice.

In the future, it is expected that molecular testing in CD will help families and caregivers not only in disease prevention in healthy subjects but also in a more precise evaluation of patient status, with accurate prognostic indicators and a wide repertoire of tests for establishing individualized therapy. This novel approach will certainly require a wider range of specialists, including molecular and clinical geneticists as well as genetic counselors who will support first-line practitioners in managing the daily problems of these patients.

References

1. Cho JH (2008) The genetics and immunopathogenesis of inflammatory bowel disease. Nat Rev Immunol 8:458–66
2. Geboes K, Colombel JF, Greenstein A et al (2008) Pathology Task Force of the International Organization of Inflammatory Bowel Diseases. Indeterminate colitis: a review of the concept—what's in a name? Inflamm Bowel Dis 14:850–857
3. Cho JH, Weaver CT (2007) The genetics of inflammatory bowel disease. Gastroenterology133:1327–1339
4. Shivananda S, Lennard-Jones J, Logan R et al (1996) 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 39:690–697
5. Frangos CC, Frangos CC (2007) Inflammatory bowel disease: reviewing an old study under a new perspective. Gut 56:1638–1639
6. Yang H, Taylor KD, Rotter JI (2001) Inflammatory bowel disease. I. Genetic epidemiology. Mol Genet Metab 74:1–21
7. Thia KT, Loftus EV Jr, Sandborn WJ, Yang SK (2008) An update on the epidemiology of inflammatory bowel disease in Asia. Am J Gastroenterol 103:3167–3182
8. Andres PG, Friedman LS (1999) Epidemiology and the natural course of inflammatory bowel disease. Gastroenterol Clin North Am 28:255–281
9. Satsangi J, Jewell DP, Bell JI (1997) The genetics of inflammatory bowel disease. Gut 40:572–574

10. Binder V (1998) Genetic epidemiology in inflammatory bowel disease. Dig Dis 16:351–355
11. Tysk C, Lindberg E, Järnerot G et al (1988) Ulcerative colitis and Crohn's disease in an unselected population of monozygotic and dizygotic twins. A study of heritability and the influence of smoking. Gut 29:990–996
12. Thompson NP, Driscoll R, Pounder RE, Wakefield AJ (1996) Genetics versus environment in inflammatory bowel disease: results of a British twin study. BMJ 312:95–96
13. Orholm M, Binder V, Sørensen TI et al (2000) Concordance of inflammatory bowel disease among Danish twins. Results of a nationwide study. Scand J Gastroenterol 35:1075–1081
14. Halfvarson J, Bodin L, Tysk C et al (2003) Inflammatory bowel disease in a Swedish twin cohort: a long-term follow-up of concordance and clinical characteristics. Gastroenterology 124:1767–1773
15. Duerr RH, Taylor KD, Brant SR et al (2006) A genome-wide association study identifies IL23R as an inflammatory bowel disease gene. Science 314:1461–1463
16. Fisher SA, Tremelling M, Anderson CA et al (2008) Genetic determinants of ulcerative colitis include the ECM1 locus and five loci implicated in Crohn's disease. Nat Genet 40:710–712
17. Cho JH, Nicolae DL, Gold LH et al (1998) Identification of novel susceptibility loci for inflammatory bowel disease on chromosomes 1p, 3q, and 4q: evidence for epistasis between 1p and IBD1. Proc Natl Acad Sci U S A 95:7502–7507
18. Fowler EV, Eri R, Hume G et al (2005) TNFalpha and IL10 SNPs act together to predict disease behaviour in Crohn's disease. J Med Genet 42:523–528
19. Villani AC, Lemire M, Fortin G et al (2009) Common variants in the NLRP3 region contribute to Crohn's disease susceptibility. Nat Genet 41:71–76
20. Nemetz A, Köpe A, Molnár T et al (1999) Significant differences in the interleukin-1beta and interleukin-1 receptor antagonist gene polymorphisms in a Hungarian population with inflammatory bowel disease. Scand J Gastroenterol 34:175–179
21. Hampe J, Franke A, Rosenstiel P et al (2007) A genome-wide association scan of nonsynonymous SNPs identifies a susceptibility variant for Crohn disease in ATG16L1. Nat Genet 39:207–211
22. Paavola P, Heliö T, Kiuru M et al (2001) Genetic analysis in Finnish families with inflammatory bowel disease supports linkage to chromosome 3p21. Eur J Hum Genet 9:328–334
23. Barrett JC, Hansoul S, Nicolae DL et al (2008) Genome-wide association defines more than 30 distinct susceptibility loci for Crohn's disease. Nat Genet 40:955–962
24. Török HP, Glas J, Tonenchi L et al (2004) Crohn's disease is associated with a toll-like receptor-9 polymorphism. Gastroenterology 127:365–366
25. Duerr RH, Barmada MM, Zhang L et al (2002) Evidence for an inflammatory bowel disease locus on chromosome 3p26: linkage, transmission/disequilibrium and partitioning of linkage. Hum Mol Genet 11:2599–2606
26. Pierik M, Joossens S, Van Steen K et al (2006) Toll-like receptor-1, -2, and -6 polymorphisms influence disease extension in inflammatory bowel diseases. Inflamm Bowel Dis 12:1–8
27. Karban AS, Okazaki T, Panhuysen CI et al (2004) Functional annotation of a novel NFKB1 promoter polymorphism that increases risk for ulcerative colitis. Hum Mol Genet 13:35–45
28. Libioulle C, Louis E, Hansoul S et al (2007) Novel Crohn disease locus identified by genome-wide association maps to a gene desert on 5p13. 1 and modulates expression of PTGER4. PLoS Genet 3:e58
29. Franke A, Balschun T, Karlsen TH et al (2008) Replication of signals from recent studies of Crohn's disease identifies previously unknown disease loci for ulcerative colitis. Nat Genet 40:713–715
30. Peltekova VD, Wintle RF, Rubin LA et al (2004) Functional variants of OCTN cation transporter genes are associated with Crohn disease. Nat Genet 36:471–475
31. Silverberg MS, Duerr RH, Brant SR et al; NIDDK IBD Genetics Consortium (2007) Refined

genomic localization and ethnic differences observed for the IBD5 association with Crohn's disease. Eur J Hum Genet 15:328–335
32. Parkes M, Barrett JC, Prescott NJ et al (2007) Sequence variants in the autophagy gene IRGM and multiple other replicating loci contribute to Crohn's disease susceptibility. Nat Genet 39:830–832
33. Bouma G, Xia B, Crusius JB et al (1996) Distribution of four polymorphisms in the tumour necrosis factor (TNF) genes in patients with inflammatory bowel disease (IBD). Clin Exp Immunol 103:391–396
34. Stokkers PC, Reitsma PH, Tytgat GN, van Deventer SJ (1999) HLA-DR and -DQ phenotypes in inflammatory bowel disease: a meta-analysis. Gut 45:395–401
35. Orchard TR, Dhar A, Simmons JD et al (2001) MHC class I chain-like gene A (MICA) and its associations with inflammatory bowel disease and peripheral arthropathy. Clin Exp Immunol 126:437–440
36. McGovern DP, Hysi P, Ahmad T et al (2005) Association between a complex insertion/deletion polymorphism in NOD1 (CARD4) and susceptibility to inflammatory bowel disease. Hum Mol Genet 14:1245–1250
37. Cantor MJ, Nickerson P, Bernstein CN (2005) The role of cytokine gene polymorphisms in determining disease susceptibility and phenotype in inflammatory bowel disease. Am J Gastroenterol 100:1134–1142
38. Brant SR, Panhuysen CI, Nicolae D et al (2003) MDR1 Ala893 polymorphism is associated with inflammatory bowel disease. Am J Hum Genet 73:1282–1292
39. Kyo K, Muto T, Nagawa H et al (2001) Associations of distinct variants of the intestinal mucin gene MUC3A with ulcerative colitis and Crohn's disease. J Hum Genet 46:5–20
40. Dideberg V, Kristjansdottir G, Milani L et al (2007) An insertion-deletion polymorphism in the interferon regulatory Factor 5 (IRF5) gene confers risk of inflammatory bowel diseases. Hum Mol Genet 16:3008–3016
41. Yamazaki K, McGovern D, Ragoussis J et al (2005) Single nucleotide polymorphisms in TNFSF15 confer susceptibility to Crohn's disease. Hum Mol Genet 14:3499–3506
42. Franchimont D, Vermeire S, El Housni H et al (2004) Deficient host-bacteria interactions in inflammatory bowel disease? The toll-like receptor (TLR)-4 Asp299gly polymorphism is associated with Crohn's disease and ulcerative colitis. Gut 53:987–992
43. Rioux JD, Xavier RJ, Taylor KD et al (2007) Genome-wide association study identifies new susceptibility loci for Crohn disease and implicates autophagy in disease pathogenesis. Nat Genet 39:596–604
44. Stoll M, Corneliussen B, Costello CM et al (2004) Genetic variation in DLG5 is associated with inflammatory bowel disease. Nat Genet 36:476–80
45. Tamura K, Fukuda Y, Sashio H et al (2002) IL18 polymorphism is associated with an increased risk of Crohn's disease. J Gastroenterol 37 (Suppl 14):111–116
46. Yang H, Ohmen JD, Ma Y et al (1999) Additional evidence of linkage between Crohn's disease and a putative locus on chromosome 12. Genet Med 1:194–198
47. Duerr RH, Barmada MM, Zhang L et al (2000) High-density genome scan in Crohn disease shows confirmed linkage to chromosome 14q11-12. Am J Hum Genet 66:1857–1862
48. Glas J, Török HP, Unterhuber H et al (2003) The -295T-to-C promoter polymorphism of the IL-16 gene is associated with Crohn's disease. Clin Immunol 106:197–200
49. Hampe J, Frenzel H, Mirza MM et al (2002) Evidence for a NOD2-independent susceptibility locus for inflammatory bowel disease on chromosome 16p. Proc Natl Acad Sci U S A 99:321–326
50. Hampe J, Cuthbert A, Croucher PJ et al (2001) Association between insertion mutation in NOD2 gene and Crohn's disease in German and British populations. Lancet 357:1925–1928
51. Hugot JP, Chamaillard M, Zouali H et al (2001) Association of NOD2 leucine-rich repeat vari-

ants with susceptibility to Crohn's disease. Nature 411:599–603
52. Rioux JD, Silverberg MS, Daly MJ et al (2000) Genomewide search in Canadian families with inflammatory bowel disease reveals two novel susceptibility loci. Am J Hum Genet 66:1863–1870
53. Latiano A, Palmieri O, Valvano MR et al (2008) The association of MYO9B gene in Italian patients with inflammatory bowel diseases. Aliment Pharmacol Ther 27:241–248
54. McGovern DP, Butler H, Ahmad T et al (2006) TUCAN (CARD8) genetic variants and inflammatory bowel disease. Gastroenterology 131:1190–1196
55. Ogura Y, Bonen DK, Inohara N et al (2001) A frameshift mutation in NOD2 associated with susceptibility to Crohn's disease. Nature 411:603–606
56. Inohara N, Ogura Y, Fontalba A et al (2003) Host recognition of bacterial muramyl dipeptide mediated through NOD2. Implications for Crohn's disease. J Biol Chem 278:5509–5512
57. Kobayashi KS, Chamaillard M, Ogura Y et al (2005) Nod2-dependent regulation of innate and adaptive immunity in the intestinal tract. Science 307:731–734
58. Cavanaugh JA, Adams KE, Quak EJ et al (2003) CARD15/NOD2 risk alleles in the development of Crohn's disease in the Australian population. Ann Hum Genet 67:35–41
59. Yamazaki K, Takazoe M, Tanaka T et al (2002) Absence of mutation in the NOD2/CARD15 gene among 483 Japanese patients with Crohn's disease. J Hum Genet 47:469–472
60. Croucher PJ, Mascheretti S, Hampe J et al (2003) Haplotype structure and association to Crohn's disease of CARD15 mutations in two ethnically divergent populations. Eur J Hum Genet 11:6–16
61. Leong RW, Armuzzi A, Ahmad T et al (2003) NOD2/CARD15 gene polymorphisms and Crohn's disease in the Chinese population. Aliment Pharmacol Ther 17:1465–1470
62. Kugathasan S, Loizides A, Babusukumar U et al (2005) Comparative phenotypic and CARD15 mutational analysis among African American, Hispanic, and White children with Crohn's disease. Inflamm Bowel Dis 11:631–638
63. Economou M, Trikalinos TA, Loizou KT et al (2004) Differential effects of NOD2 variants on Crohn's disease risk and phenotype in diverse populations: a metaanalysis. Am J Gastroenterol 99:2393–2404
64. Annese V, Lombardi G, Perri F et al (2005) Variants of CARD15 are associated with an aggressive clinical course of Crohn's disease – an IG-IBD study. Am J Gastroenterol 100:84–92
65. Crawford NP, Colliver DW, Eichenberger MR et al (2007) CARD15 genotype-phenotype relationships in a small inflammatory bowel disease population with severe disease affection status. Dig Dis Sci 52:2716–2724
66. Wiekowski MT, Leach MW, Evans EW et al (2001) Ubiquitous transgenic expression of the IL-23 subunit p19 induces multiorgan inflammation, runting, infertility, and premature death. J Immunol 166:7563–7570
67. Yen D, Cheung J, Scheerens H et al (2006) IL-23 is essential for T cell-mediated colitis and promotes inflammation via IL-17 and IL-6. J Clin Invest 116:1310–1316
68. Cummings SA, Rubin DT (2006) The complexity and challenges of genetic counseling and testing for inflammatory bowel disease. J Genet Couns 15:465–476
69. Lindberg E, Tysk C, Andersson K, Järnerot G (1988) Smoking and inflammatory bowel disease. A case control study. Gut 29:352–357
70. Calkins BM (1989) A meta-analysis of the role of smoking in inflammatory bowel disease. Dig Dis Sci 34:1841–1854
71. Sandler RS, Wurzelmann JI, Lyles CM (1992) Oral contraceptive use and the risk of inflammatory bowel disease. Epidemiology 3:374–378
72. Brant SR, Picco MF, Achkar JP et al (2003) Defining complex contributions of NOD2/CARD15 gene mutations, age at onset, and tobacco use on Crohn's disease phenotypes. Inflamm Bowel Dis 9:281–289

73. Gleason TR, Evans ME (2004) Perceived vulnerability: a comparison of parents and children. J Child Health Care 8:279–287
74. Grosse SD, Khoury MJ (2006) What is the clinical utility of genetic testing? Genet Med 8:448–450
75. Pierik M, Rutgeerts P, Vlietinck R, Vermeire S (2006) Pharmacogenetics in inflammatory bowel disease. World J Gastroenterol 12:3657–3667

The Role of the Pathologist in the Management of Patients with Crohn's Disease

4

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Gastrointestinal histopathology represents approximately half of the current workflow in our laboratory, and six to ten of the reports signed out daily concern patients studied for Crohn's disease (CD). This chapter summarizes the role of the pathologist in the management of patients with CD, starting from a glossary of the terms used in the information exchange between the pathologist, radiologist, gastroenterologist, and surgeon. This multidisciplinary team approach is crucial for the correct management of CD patients. The gastrointestinal tract has limited patterns of tissue response to triggers of inflammation and the diagnosis therefore relies upon the various combinations of these patterns with the clinical picture. The study of endoscopic specimens is based not only on a histopathological scheme but also on the presentation of the patient, which is of equal importance. Due to the limits in the information that can be extracted by studying the morphology of inflammation, molecular biology is expected to play an increasingly important role in providing the clinician with insight into the disease process in a single CD patient and thus in allowing the appropriate treatment strategy to be tailored accordingly.

A Practice-Based Glossary

Anal Cancer and Fistula

Patients with CD are at risk for anal cancer. A particular diagnostic difficulty is distinguishing between an adenocarcinoma in fistula and an adenocarcinoma presenting as a fistula. Biopsies taken from fistulas can show glands with minor atypias compatible with a regenerating process. Therefore, in these instances, to achieve a

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final diagnosis of adenocarcinoma it is often necessary to obtain a more extensive in-depth biopsy. Adenocarcinoma with minimal atypia has been described as low-grade tubuloglandular adenocarcinoma [1] and is particularly frequent in patients with inflammatory bowel disease (IBD). In our experience, vascular invasion was the diagnostic difference between benign and malignant disease in one patient and deep infiltration to the sphincter in another, because of the minimal amounts of atypia observed on the initial biopsy. By contrast, minor diagnostic difficulties are encountered with squamous cell carcinoma.

Architecture

Mucosal architecture plays a major role in the pathological interpretation of IBD. Changes in architecture are the results of damage to the crypts, superficial epithelium, and lamina propria. Moreover, the intestinal architecture may revert to normal in IBD in remission or in a quiescent phase. Patients referred to qualified centers after a first acute episode often have a normal histology at the second biopsy. It is therefore useful to review the slides prepared from biopsies taken during the first acute episode. Asymptomatic patients may have abnormalities in mucosal architecture as a result of major damage, leading to defective healing with crypt loss, branching, or metaplastic changes of the Paneth cell or pseudopyloric type. These cases, marked by alterations limited to the mucosal architecture without active inflammation, are often referred to as unclassified IBD, with further definition depending on the modality of inflammation during the active phase of disease. The appropriate timing of the biopsy is crucial in lesion detection. At disease onset, basal lymphoplasmacytosis in the lamina propria occurs over a period of 2 weeks but is, in fact, better observed during the third week.

Biases and Mimics

Patients treated with enemas (5-ASA, butyrate) were reported to have a statistically higher percentage of normal biopsies (36%) per patient than patients in the corresponding placebo-treated group (12%) [2]. In up to 60% of patients with ulcerative colitis (UC), previously inflamed segments of colon convert to normal following treatment with oral anti-inflammatory agents, such as sulfasalazine or steroids. Thus, evaluation of disease “continuity” by mucosal biopsies is not useful to distinguish UC from CD in previously treated IBD patients. Some patients develop a form of UC characterized by complete endoscopic and histologic sparing of the transverse colon in the presence of left-sided proctosigmoiditis in association with either cecal or ascending colon inflammation. Patients with fulminant UC and extensive mucosal ulceration may show CD-like histological features, such as relative or complete rectal sparing, patchy ulceration, fissuring ulceration, and transmural lymphoid inflammation.

Several inflammatory disorders can mimic CD: segmental inflammation, granu-

lomatous inflammation, ileal inflammation in UC before and after ileal pouch anal anastomosis (IPAA), and aphthoid ulcers with skip interval mucosa as in ischemic and NSAID colitis [2]. In addition, infections with *Entamoeba histolytica, Salmonella*, and *Yersinia*, which typically cause a right-side-predominant or segmental colitis, may mimic CD endoscopically. Superimposed infections caused by *Clostridium difficile* or cytomegalovirus should be considered in CD patients receiving steroids or immunosuppressive therapy, with unexpected recurrences, or with refractory illness [3,4].

Dysplasia

Patients with IBD are at risk of developing cancer of the small bowel in CD and colorectal cancer (CRC) in both UC and CD. The special risk for IBD patients is associated with active, extended, and long-duration disease.

Dysplasia is an intraepithelial neoplasia with a low or high grade of atypia and a flat, polypoid, or mass-forming (DALM) pattern of growth. Referral centers for IBD gather cohorts of thousands of patients and must therefore distinguish between IBD patients affected by a sporadic case of dysplasia and those with disease-driven dysplasia. IBD patients are treated either conservatively (polypectomy/mucosectomy) or by colectomy, with the latter reserved for high-grade neoplasias, infiltrating adenocarcinomas, and low-grade dyplasia in multiple sites of the large bowel. The decision-making process is often clinically and pathologically driven in the first round and follows clinician-patient discussions in the second round. In both cases, there are failures of good judgment and appropriate choices.

The usual course of assessment for any new patient with IBD includes open discussion and repeated diagnostic procedures. Conservative treatments are undertaken in patients with sporadic non-IBD exhibiting high-grade adenomas, intramucosal adenocarcinomas, and multiple low-grade dysplasias. Even in patients with transmural infiltrating carcinomas, a partial colectomy is usually performed.

Why perform colectomy in the IBD patient? The answer to this question requires a basic understanding of the problem. After 20–30 years of pancolic and active disease, a considerable number of patients have cancer, consisting of high-grade and low-grade dysplasia and flat or mass-forming lesions. The colectomy specimens of these patients may well be a jumble of these features. Interestingly, up to one-fifth of the colons removed for dyplasia can reveal an infiltrating adenocarcinoma missed at endoscopy. In this setting, colectomy seems a good choice, as it treats the tumor and the whole source of cancer risk excess.

What about the occurrence of dysplasia early in the disease history or in young patients? What about focal dysplasia in an extensively studied and biopsied colon? Recent data [5–6] reported a delay in the diagnosis of CRC in 17–28% of the patients eligible for colonoscopic surveillance according to the guidelines of the American and British Associations of Gastroenterology, which recommends an entry surveillance time from the first diagnosis of 8–10 years for CD and UC pancolitis, and 15–20 years for left-sided colitis. Only 17% of the CRC cases were detected under

surveillance; 41% because the patients were symptomatic, and 23% in non-surveillance colonoscopies. In 13% of the cases, CRC was an incidental finding in proctocolic resection for other causes, including refractory disease. As a consequence, a well-known risk group of subjects already present at the time of CRC diagnosis with lymph node (33%) or distant (18.7%) metastasis, and have a median age of 49 years at the time of diagnosis. Surprisingly, the life span of IBD patients is not as affected as their lifestyle, compared to the general population.

These considerations suggest the following:

1. these patients merit the same preventive facilities aimed at the general population, i.e., colonoscopy for prevention after age 45 in asymptomatic subjects;
2. the entry time of surveillance should be anticipated before a disease history of 8 years, possibly taking into account the duration of symptoms and not the time of first diagnosis;
3. the interval between colonoscopies should be shorter than 3 years;
4. biopsies should be taken at 10-cm intervals, labeling each site in order to locate the dysplasia; a total of 33 biopsy specimens are required to diagnose dysplasia with 90% confidence [6].

Granulomas

Granulomas have been traditionally considered a feature of CD; however, the emphasis given to this pattern of inflammation may be misleading. The more the histopathology is dominated by granulomas, the less reliable is the diagnosis of CD. Granulomas are formed around broken crypts, whatever the cause, be it as a reaction to the discharge of mucin deep in the intestinal wall or to penetrating intestinal luminal contents consequent to any fissuring process, including infection, UC, perfusion disorders such as vasculitis or heart surgery, etc. Due to the local environment of CD, infection is often if not always the second component of disease activity. Up to 80% of the cases of active CD respond to antibiotic therapy [7].

Immunity

CD has been linked to autoimmunity and autoinflammation, considered as a continuum rather than as separate entities [8]. The actual impact of the autoinflammation concept on clinical practice is not yet clear, but the overlap between the patterns of local intestinal expression of systemic disease and CD are noteworthy. Paradoxically, autoimmune disorders, such as as Behçet disease and immunological deficiency, along with common variable immunodeficiencies are characterized by an inflammatory process in the gastrointestinal tract that closely mimics CD. Moreover, CD patients who undergo radical native immune system suppression, as in bone marrow transplantation for hematological disorders or for acquired HIV-driven immunosuppression, experience remission of their CD and/or long disease-free intervals. This exceedingly complicated matter is mentioned here in order to point out that in CD

patients undergoing surgery as the first treatment an underlying systemic disease should be suspected, rather than simply classifying the disease in these patients as "usual CD" on the basis of a surgical emergency for a terminal-ileum disease.

Slide Reviewing

In referral centers for IBD, slide reviewing for a second opinion is frequently requested by gastroenterologists. The basic questions to the pathologist are: is it IBD? Is the differential between CD and UC feasible?

Frequently, the approach to the patient changes following the pathologist's opinion. The diagnosis of IBD is affected by problems in inter-observer reproducibility and by misunderstandings in communication. The most frequent source of the latter is the term "inflammation," regardless of whether it is qualified with the additional terms "chronic," "acute," or "nonspecific." If associated with symptoms of long duration, the automatic translation into chronic colitis or ileitis is common. In addition, two major normal functions are erroneously interpreted as pathological findings: inflammatory-cell trafficking inside the mucosa and reactive lymphoid hyperplasia in the lymphatic tissue (Peyer's patches) of the ileum or involving lymphoepithelial complexes in the large bowel. As in all equilibrium settings, the features of both vary within the range of normal. Another bias comes from the general definition of chronic inflammation as a response sustained by lymphocytes and plasma cells, and acute inflammation by granulocytes. In the chronic inflammatory process of IBD, granulocytes are a constant component of disease activity. Lymphocytes and plasma cells are normally present in the gastrointestinal tract mucosa and are increased in number in acute inflammation.

Surgical Margins and Surgical Specimens

Occasionally, the pathologist is requested to evaluate the surgical margins in order to limit the amount of intestinal resection. Frozen-section examination often leads to over-treatment because inflammation restricted to the mucosal level is not associated with anastomotic failure or relapse. Ulcers in the surgical margins or transmural inflammation, both of which are related to relapse, are better evaluated by the naked eye. In the studied and classified patient undergoing surgery for complicated unresponsive CD, the surgical specimen report outlines lesions related to stenosis, such as fibromuscular obliteration of the submucosa, or substitution of the tunica propria. Several attempts have been made to identify unequivocal histological markers predicting recurrence in the surgical intestinal specimens of CD patients [9,10]. In two different series, at the Leuven and St. Mark's hospitals, myenteric plexitis at the proximal resection margin was statistically associated with recurrence and second surgery.

4

The Biopsy-Based Histopathology of Crohn's Disease: A Stepwise Approach

While CD can affect different segments of the gastrointestinal tract, the microscopic features are similar at all sites. The diagnosis of CD is based on the presence of aphthoid ulcers, fissural ulcers, sinus tracts, granulomas, transmural lymphoid hyperplasia, fibromuscular obliterations, and vascular and neural changes. However, the majority of these findings cannot be detected on endoscopic biopsy samples. Furthermore, the mucosal lesions can be very mild. CD may demonstrate wide overlap with UC on mucosal biopsy examination. In addition, several types of chronic colitis may mimic the features of UC and CD. The analysis of multiple biopsies allows a correct diagnosis of IBD in 66–75% of newly diagnosed patients. Additional endoscopic and clinical data allow a final diagnosis in more than 90%. In practice, the first step in evaluating mucosal biopsies is to establish a diagnosis of colitis based on the presence of epithelial damage and inflammation. Subsequently, a possible etiology should be included in order to differentiate between IBD and other types of chronic colitis. The last step is to solve the differential diagnosis between CD and UC, which can be done by the location of the disease, the distribution of the lesions, and the presence of ileal lesions [11,12]. The diagnosis of IBD on endoscopic biopsies relies therefore on the detection of reproducible microscopic criteria that consist of changes in the architecture, a cellular inflammatory response, and epithelial cell alterations.

Abnormalities of the mucosal architecture include an irregular/pseudovillous mucosal surface, architectural distortion of the crypts (non-parallel, variable diameter, or cystically dilated), crypt branching and shortening, and decreased crypt density. Crypt distortion in the colon, one of the major features in the diagnosis of IBD, is usually not present in the early phase and generally takes two months to develop. The architectural alterations may be focal, discontinuous, or diffuse. The cellularity of the lamina propria changes in intensity, composition, and distribution pattern and may be diffuse or discontinuous (focal/patchy). In the diffuse pattern there is an increase in cellularity throughout the lamina propria, with basal plasmacytosis. The latter occurs in 38% of patients within two weeks following the onset of colitis and has a high predictive diagnostic value. Patchy inflammation is diagnosed when the mucosal background shows inflammation of varying intensity, while focal inflammation is defined as a small collection of inflammatory cells in an otherwise normal mucosa [13]. Sarcoid-type granulomas are traditionally considered the hallmark of CD, even if present in only 50% of the cases of small-bowel CD. In rectal biopsies of patients with CD, the overall incidence of granulomas is >20%. They are more common in young patients (42%) with short disease duration. Infectious diseases such as tuberculosis and yersiniosis should be considered when granulomas are seen. Pericryptal granulomatous inflammation is not a specific feature of CD and also can be seen in UC. A well-formed single granuloma, non-necrotizing, basally oriented, and unrelated to ruptured crypts is highly suggestive of CD and remains one of the most useful markers of the disease [14]. Epithelial alterations, including mucin

depletion, Paneth cell metaplasia, and flattening of the superficial epithelial cells, are indicative of damage and the ensuing repair process. Disease activity in IBD is usually assessed based on infiltration by polymorphonuclear neutrophils of the surface epithelium, crypt epithelium (cryptitis), or crypt lumina (crypt abscesses). Crypt destruction, erosions, and ulcerations are other markers of disease activity.

The presence of ileal lesions is one of the major features discriminating between CD and UC. In chronic ileitis, structural changes consist of abnormalities in villi shape and size (blunting, enlargement, broadening at the top, atrophy, or diffuse shortening) and in the crypts, which can become branching, shortened, atrophic or lost, and arranged in groups. The epithelial lining can be normal or show increased mucin production. Changes in the crypt epithelium include ulcer-associated cell lineage (pseudopyloric gland metaplasia) and an increase in the number of Paneth cells displaced higher up in the crypts [15]. Aphthoid erosion is defined as a mucosal erosion occurring over lymphoid aggregates such as Peyer's patches; it is present in > 50% of CD patients but it is not specific.

Features that favor a diagnosis of CD are chronic active inflammatory changes in the terminal ileum, less severe disease in the distal than in the proximal colon, aphthoid ulcers, epithelioid granulomas unassociated with crypt rupture, segmental architectural changes with patchy inflammation and focal cryptitis in the colonic mucosa, involvement of the upper gastrointestinal tract, and the presence of normal and inflamed samples from the same areas (skip areas). In 10–20% of all CD patients, the disease involves the colorectal mucosa with a diffuse pattern of inflammation without skip lesions and closely mimicking the endoscopic and biopsy features of UC [2–16].

A variety of circumstances can modify the usual histological patterns such that the separation of CD and UC becomes very difficult. Rectal sparing and patchy inflammation associated with focal minimal architectural alterations have been documented in biopsies from children presenting with new-onset UC, in adult patients under treatment, in patients with primary sclerosing cholangitis and IBD, and in patients presenting with severe fulminant disease. Therefore, in these conditions, the diagnosis of IBD unclassified (IBDU) has been proposed [11].

Molecular Mechanisms of Crohn's Disease

Main Molecular Mechanisms

It is well known that CD results from a dysregulated interaction between the three basic components of the host response to the intestinal microbiota: the intestinal epithelium, innate immunity, and the adaptive immune response. A thorough understanding of the basic molecular mechanisms that govern these interactions is of paramount importance to identify novel molecular targets that enrich the clinician's therapeutic armamentarium. The intestinal epithelium represents a solid and very selective barrier that, due to an individual's genetic predisposition and chronic exposure to

yet unidentified intraluminal antigens of bacterial origin, can eventually fail. The result is abnormal intestinal permeability, activation of the innate immune system, and intestinal inflammation. To understand the molecular mechanisms associated with this very complex pathological interaction, we need to become familiar with the diverse molecular players involved in the process. The identification of nucleotide-binding oligomerization domain-2 (*NOD2*) as a CD susceptibility gene has provided important clues into the dysregulated host immune response to luminal bacteria. The NOD2 protein functions as an intracellular receptor that plays a role in the immune response by recognizing muramyl dipeptide derived from bacterial lipopolysaccharides. In addition, NOD2 is expressed in intestinal epithelial cells, Paneth cells, macrophages, and dendritic cells (DC) and is thought to control the expression of specific antimicrobial agents known as defensins. Adaptive immunity relies on resident mucosal B cells that produce immunoglobulins and a mixture of T cells, predominantly represented by T-helper (Th) cells with a 1, 2, or 17 phenotype. The Th-1 lineage is induced by the expression of interleukin (IL)-12p40 and interferon (IFN)-γ and in turns activates the production of IL-1, IL-6 and TNF-α, while, the more recently discovered Th-17 phenotype is controlled by TGF-β, IL-6, and IL-23. Additional players have been identified that are involved in the process of leukocyte migration to sites of inflammation, most notably, integrins, cell adhesion molecules, matrix metalloproteinases (MMPs) and tissue inhibitors of MMPs (TIMPs). The identification of specific key molecules involved in the pathogenesis of CD has led to the development of novel biological therapies aimed at blocking their function as well as a great deal of new drug therapies that include cytokine and anti-cytokine therapies (anti-TNF drugs) and anti-adhesion molecules to block T-cell recruitment [17].

Molecular Mechanisms "Under the Microscope"

The pathologist relies extensively on immunohistochemistry (IHC) in studying how, when, and where a protein is expressed in a pathological tissue. Despite the great advances in IHC technology that have been exploited in other fields of study, such as oncology, very little has been done, until recently, with respect to the in situ molecular characterization of IBD, which, instead, mostly still relies on classic histopathological parameters. In the last 2–3 years, however, we have witnessed a rapidly growing interest in the immunohistochemical approach to IBD. Therefore, in the following, we report several important examples of solid initial and novel contributions to the field.

Patients with CD show a decrease in the production of defensins, probably leading to ineffective microbial elimination. Several studies have proposed a role in NOD2 regulation of the α-defensins production by the inflamed mucosa. Three recent independent studies have identified TCF-4, a Wnt signaling pathway transcription factor, in patients with ileal CD; specifically, that in Paneth cells there is a parallel decrease in mRNA expression of TCF-4 and human defensins 5 and 6 (HD-5 and HD-6). This phenomenon was linked to a specific single nucleotide polymorphism (rs 3814570) in the TCF-4 gene. In addition, a comparison between HD-5 immunohistochemical staining in serial biopsies from patients with either NOD-2 wild-type or NOD-2 mutant

proteins demonstrated that HD-5 reduction was independent of NOD2 status [18]. Animal studies have shown mucosal immune regulation by DC located in the subepithelial dome. Cells positive for the DC marker DC-sign(+) in Crohn's tissue were found to express toll-like receptor 4 and TNF-α, suggesting that DC cells are important for the onset and perpetuation of mucosal inflammation in CD [19]. Chronic inflammation also affects the local neuronal plexuses of the enteric nervous system. Plasminogen activator inhibitor-1(PAI-1) and urokinase receptor (uPAR) have been linked to nerve regeneration. In a study of 28 patients with CD and 17 patients with chronic UC, PAI-1 was found in a subset of neurons primarely located in the submucosal plexus of the small and large intestine in 24 of 28 the CD patients but in none of the 17 with UC. In addition, PAI-1 expression was restricted to the perikarya of neurons while the nerves were negative. The data imply that PAI-1 is a potential marker to discriminate CD from UC [20]. The balance between ileal T-effector cells vs. T-regulatory cells was studied in active and inactive CD, using RT-PCR and IHC with a large panel of antibodies to several cytokines. The numbers of cells positive for IL-4, IL-17, and IL-23 in the lamina propria were higher in patients with CD, both active and inactive, than in controls. In addition, Il-23 mRNA expression was increased in active vs. inactive disease. These findings indicate that activation of the IL-23/IL-17 axis is strictly connected to the etiology of CD [21].

IHC can also be helpful to study the therapeutic effect of novel drugs adopted for the pharmacological treatment of CD. The CD40/CD40L pathway is important in disease pathogenesis. In the intestine, CD40 is overexpressed in the microvasculature and CD40L in platelets and T cells. Mucosal biopsies from 18 patients with CD were evaluated before and after infliximab therapy. Infliximab treatment was found to abolish CD40 and VCAM-1 expression in mucosal microvessels. Moreover, in the same study, in vitro evaluation of infliximab therapy in human intestinal microvascular endothelial cells (HIMEC) demonstrated that this biological agent was able to prevent TNF-α-induced CD40 and VCAM-1 expression by HIMEC and to decrease T-cell-induced VCAM-1 expression in HIMEC by down-regulating CD40L in T cells and promoting T-cell apoptosis. A novel mechanism of action of infliximab, implying the disruption of CD40/CD40L-dependent cognate interaction between intestinal microvessels and T cells, was therefore discovered [22].

An absolutely novel and exciting highway of research has been opened by the discovery of microRNAs, which are single-stranded RNA molecules partially complementary to one or more mRNAs. The main function of microRNAs is to down-regulate gene expression with a tissue-specific pattern. Their role in the different pathologies is intriguing because they can be used as diagnostic markers of disease and to monitor disease progression. Furthermore, they are appealing molecular targets for novel therapeutic strategies. From the pathologist's point of view, microRNAs can be studied by in situ hybridization analysis. In a recent work, differential expression of 11 microRNAs was identified in tissue biopsies from patients with active UC. In situ hybridization analysis indicated that miR-192, whose expression in active UC was decreased, was mainly localized to colonic epithelial cells. Furthermore, macrophage inflammatory peptide (MIP)-2α, a chemokine expressed by epithelial cells, was identified as a target of miR-192 [23]. As far as CD is concerned, to the best of our

knowledge, no results on miRNAs have yet been published, but we can certainly foresee that the excitement for this new field of research will shortly produce important clues to new diagnostic and therapeutic approaches in the management of CD.

References

1. Levi GS, Harpaz N (2006) Intestinal low-grade tubuloglandular adenocarcinoma in inflammatory bowel disease. Am J Surg Pathol 30(8):1022–1029
2. Yantiss RK, Odze RD (2007) Pitfalls in the interpretation of non neoplastic mucosal biopsies in inflammatory bowel disease. Am J Gastroenterol 102:890–904
3. Garcia–Osogobio S, Takahashi T, Gamboa–Dominguez A et al (2000) Toxic pseudomembranous colitis in a patient with ulcerative colitis. Inflamm Bowel Dis 6:188–190
4. Papadakis KA, Tung JK, Binder SW (2001) Outcome of cytomegalovirus infections in patients with inflammatory bowel disease. Am J Gastroenterol 96:2137–2142
5. Lutgens MW, Vleggaar FP, Schipper ME et al (2008) High frequency of early colorectal cancer in inflammatory bowel disease. Gut 57(9):1246–1251
6. Stange EF, Travis SP, Vermeire S et al (2006) European Crohn's and Colitis Organisation. European evidence based consensus on the diagnosis and management of Crohn's disease: definitions and diagnosis. Gut 55(Suppl)1:i1-i15
7. Prantera C, Lochs H ,Campieri M et al (2006) Antibiotic treatment of Crohn's disease: results of a multicentre, double blind, randomized, placebo controlled trial with rifaximin. Aliment Pharmacol Ther 23:1117–1125
8. Touitou I, Notarnicola C, Grandemange S (2004) Identifying mutations in autoinflammatory diseases: towards novel genetic tests and therapies? Am J Pharmacogenomics 4(2):109–118
9. Ferrante M, de Hertogh G, Hlavaty T et al (2006) The value of myenteric plexitis to predict early postoperative Crohn's disease recurrence. Gastroenterology 130(6):1595–1606
10. Ng SC, Lied GA, Kamm MA et al (2009) Predictive value and clinical significance of myenteric plexitis in Crohn's disease. Inflamm Bowel Dis. Mar 31. (DOI)10.1002/ibd.20932
11. Geboes K, Van Eyken P (2009) Inflammatory bowel disease unclassified and indeterminate colitis: the role of the pathologist. J Clin Pathol 62: 201–205
12. Tsang P and Rotterdam H (1999) Biopsy diagnosis of colitis. Possibilities and pitfalls. Am J Surg Pathol 23:423–430
13. Geboes K (2001) Pathology of inflammatory bowel disease (IBD): variability with time and treatment. Colorectal Dis 3:2–12
14. Shepherd NA (2002) Granulomas in the diagnosis of intestinal Crohn's disease: a myth exploded? Histopathology 41:166–168
15. Cuvelier C, Demetter P, Mielants H et al (2001) Interpretation of ileal biopsies: morphological features in normal and diseased mucosa. Histopathology 38:1–12
16. Odze R (2003) Diagnostic problems and advances in inflammatory bowel disease. Mod Pathol 16:347–358
17. Peyrin–Biroulet L, Desreumaux P, Sandborn VJ et al (2008) Crohn's disease: beyond antagonists of tumour necrosis factor. Lancet 372:67–81
18. Simms LA, Doecke JD, Walsh MD et al (2008) Reduced alpha-defensin expression is associated with inflammation and not NOD2 mutation status in ileal Crohn's disease. Gut 57:903–910
19. Salim SY, Silva MA, Keita AV et al (2009) CD83+CCR7-dendritic cells accumulate in the subepithelial dome and internalize translocated Escherichia coli HB 101 in the Peyer's patches of ileal Crohn's disease. Am J Pathol 174:82–90

20. Laerum OD, Illemann N, Skarstein A et al (2008) Crohn's disease but not chronic ulcerative colitis induces the expression of PAI-1 in enteric neurons.Am J Gastroenterol 103:2350–2358
21. Holta V, Klemetti P, Sipponen T et al (2008) IL23/IL17 immunity as a hallmark of Crohn's disease. Inflamm Bowel Dis 14:1175–1184
22. Danese S, Sans M, Scaldaferri F et al (2006) TNF–alpha blockade down–regulates the CD40/CD40L pathway in the mucosal microcirculation: a novel anti–inflammatory mechanism of infliximab in Crohn's disease. J Immunol 176:2617–2624
23. Wu F, Zikusoka M, Trindade A et al (2008) MicroRNAs are differenttiallyexpressed in ulcerative colitis and alter expression of macrophage inflammatory peptide-2 alpha. Gastroenterology 135:1624–1635

Clinical Presentation

5

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Crohn's disease (CD) involves the entire alimentary tract, starting from the mouth, and is characterized by focal exacerbations, with intermittent activity throughout the patient's life. The initial manifestations are often insidious and vague. Systemic symptoms include unexplained fever, weight loss, and extraintestinal symptoms such as arthralgias and perianal abscess. The retrospective description of this initial disease course may be difficult and ill-defined, especially in the presence of intestinal symptoms that are rather nonspecific. Delays by the patient in seeking medical help and by the physician to identify the disease contribute to the long interval from onset to diagnosis, which tends to become even longer in patients with mild symptoms. More recently, however, owing to an improved awareness of the disease, this time interval has decreased.

The disease can be localized throughout the gastrointestinal system and histological features of inflammation can be evident in endoscopically uninvolved areas. Nonetheless, the large majority of new cases of grossly detectable CD are located in three sites: the small intestine only, the colon only, and in both the small and the large bowel. The presenting clinical features depend on the anatomical location, the extent of the disease, the occurrence of complications, and extraintestinal involvement, which, taken together, make CD a heterogeneous disease with extreme variability in its clinical features. These may be further explained, at least in part, by patients' different genetic backgrounds.

Clinical Symptoms

Symptoms of CD commonly include diarrhea for more than 6 weeks, abdominal pain, and/or weight loss (Fig. 5.1). Chronic diarrhea, defined as a decrease in fecal

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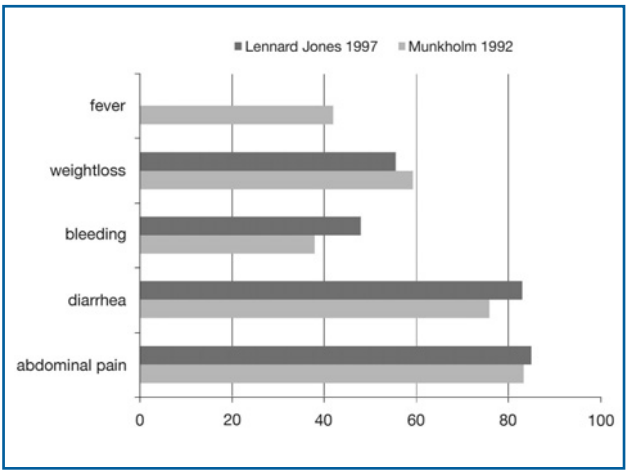

Fig. 5.1 Frequency of clinical symptoms: data from population-based studies. (From [27,32])

consistency for more than 6 weeks, and abdominal pain are present in more than 70% of all patients at diagnosis; blood and/or mucus in the stool may be seen in up to 50% of patients, and perianal lesions in 10% at the time of diagnosis [1]. Articular involvement is the most common extraintestinal manifestation of CD and may precede intestinal symptoms by months or years [2].

Although present at all localizations, a significant correlation has been found between ileal disease and abdominal pain, and between diarrhea or bloody stools and colonic disease. The risk of developing Crohn's perianal fistulas increases when the disease involves the distal bowel; only 12% of patients with isolated ileal disease develop perianal fistulas compared with 92% of patients with rectal involvement [3]. Hypoproteinemia due to intestinal protein loss, clubbing, and occult blood loss with anemia is frequent in Crohn's jejunoileitis [4]. Dysphagia, vomiting, and epigastric pain, related to upper gastrointestinal lesions, are less typical presentations and represent rare initial manifestations of the disease. The occurrence of systemic symptoms, such as malaise, anorexia, weight loss, and fever, are common and independent of disease location.

Anatomical Location

The single most affected segment is the terminal ileum, which is clinically inflamed in two-thirds of patients with CD. However, detailed knowledge about initial disease location is limited by the fact that most of the data originate from retrospective studies using different methodologies to define disease involvement (i.e., aphthous lesion or ulcer excluding erythema and edema) and by the lack of information regarding the condition of the entire gastrointestinal tract at the time of diagnosis. These limitations and the diverse definition of disease location account for the variability of the

disease location frequency reported by different studies. In an attempt to define the different localizations of CD, in 1998, at the World Gastroenterology meeting in Vienna, a clinical classification based on four distinct locations was proposed:

- Terminal ileum, with or without spillover into the cecum
- Colon, any colonic location between cecum and rectum
- Ileocolon, disease of the terminal ileum and any location of the colon
- Upper gastrointestinal tract, any location proximal to the terminal ileum irrespective of the other locations of the disease

The distribution of patients at diagnosis within each of these categories of disease location is shown in Table 5.1; the data derive from population-based studies following the Vienna classification.

Table 5.1 Localization of Crohn's disease at diagnosis in population-based studies

	Terminal ileum (%)	Colon (%)	Ileocolon (%)	Upper GI (%)
Vind et al 2006 [6] >18 years	32	39	22	7
<18 years	17	17	41	25
Louis et al 2003 [7]	45	28	22	5
Louis et al 2001 [5]	38	22	33	6
Witte et al 2000 [33]	29	38	31	?
Tarrant et al 2008 [12]	32	49	19	0.6

Only a minority of patients has involvement of the duodenum or jejunum, mainly associated with ileitis, and only a very small number of patients have isolated jejunal disease. The evolution of endoscopy, e.g., single- and double-balloon enteroscopy and capsule endoscopy, as well as the development of routine endoscopy in recent years, has revealed a higher frequency of CD in the upper gastrointestinal tract (5–7%) [5,6]. Approximately 1–4% of patients suffer from atypical manifestations, with oral lesions, pharyngeal and esophageal disease, and gastric involvement, which seldom occur without additional involvement of the ileum and/or of the large bowel. The location of the disease remain relatively stable over its course; while the proportion of patients with a change in disease location becomes significant after 5 years, over 10 years only 16% of patients typically experience a change in location [5].

Behavior of the Disease

The chronic transmural inflammatory process of CD may lead to stricture of the intestinal lumen by the formation of fibrosis within the bowel wall and to penetration throughout the bowel wall, with fistulae connecting to the skin or to nearby organs. This behavior of CD is defined as stricturing or penetrating.

The behavior of CD varies throughout its course; over 10 years, nearly 50% of patients have a change in disease behavior [5]. The majority of patients start with non-stricturing and non-penetrating disease at diagnosis, but after 25 years the majority progress to either a stricturing or a penetrating pattern. The development of a stricturing or penetrating behavior probably represents a multifactorial phenomenon, involving environmental and genetic factors. Environmental factors include disease treatments [7] and smoking [8–10]; the latter has been associated with more aggressive disease and with perianal penetrating disease. Genetic factors seem to influence the speed at which a penetrating or stricturing behavior develops rather than the ultimate behavior of the disease. The behavior of CD is clinically relevant and is associated with the development of complications and the need for surgery. The association between disease location and behavior and the potential influence of disease location on disease behavior evolution have been studied. A positive association was found between isolated small bowel location and stricturing disease, and between colonic disease and penetrating behavior [5,11]. This association was present at diagnosis and was more prominent after 10 years.

Recently, perianal disease was found to be associated with an increased likelihood and rate of progression to more complicated CD [12].

Extraintestinal Manifestations

CD is associated with a variety of systemic manifestations that influence its course, the therapeutic response, and the patient's quality of life. The reported frequency of extraintestinal manifestations (EIMs) varies from 16 up to 25% [13,14]. Several factors may be responsible for extraintestinal organ involvement in CD and it can be difficult to distinguish true EIMs from extraintestinal complications due to bowel dysfunction or to treatment side effects. While some EIMs, e.g., those associated with autoimmune dysregulation, may not correlate with disease activity, they generally follow the clinical course of the intestinal disease (Table 5.2) [15]. The identified pathogenetic autoimmune mechanisms include genetic susceptibility, antigenic display of autoantigen, aberrant self-recognition, and immunopathogenic autoantibodies against organ-specific cellular antigens shared by the colon and extracolonic organs [16]. Shared epitopes between bacterial and viral proteins and host self-antigens could initiate an autoimmune response that continuous after the initial antigenic signal remits. Several autoimmune phenomena are identifiable in CD and include antibodies to joint constituents (anti-collagen antibodies) and organ-based tissues (anti-pancreatic antibodies) [17]. It has been suggested that self-antigens located at different sites can evoke an autoimmune response; a 40-kDa protein resembling tropomyosin or its isoform and located on the surface of colonic epithelial cells, as well as on the ciliary body of the eye, joint chondrocytes, and bile duct epithelial cells has been identified. Some HLA haplotypes (HLA B27, A2, DR1, and DQW5) are more commonly associated with the development of EIMs but a rich intestinal bacterial flora is required for the full expression of EIMs in patients with colonic inflammatory bowel disease [15].

Table 5.2 Extraintestinal manifestations (EIMs) of Crohn's disease

EIMs associated with disease activity	EIMs not associated with disease activity
Muscoloskeletal Arthritis Peripheral arthropathies	**Muscoloskeletal** Sacroileitis Ankylosing spondylitis
Dermatologic Erythema nodosum Apthous stomatitis	**Dermatologic** Pyoderma gangrenosum
Ocular Scleritis Episcleritis	**Ocular** Uveitis
	Hepatobiliary disease Primary sclerosing cholangitis
	Vascular Thromboembolic events
	Renal Nephrolithiasis

An increase in endothelial cell permeability in the inflamed intestine and the expression of specific adhesion molecules permit bi-directional movement of cytokines, bacterial proteins, and inflammatory cells. A key to the full development of other EIMS may be the change in the migration path for homing lymphocytes. In inflammatory bowel disease, lymphocytes circulate to sites as far from the intestine as the peripheral joints and the eyes. These sites lose their normal immunological tolerance and, behaving as anomalous lymphoid organs, develop an inflammatory response clinically recognized as an extraintestinal complication [17].

Clinical Course

Inflammatory CD is a naturally remittent and recurring disease. The different disease patterns include chronically active disease, chronic intermittent disease with remission periods longer than 1 month without steroids, and disease with remission periods over many years without the need for steroids**.** Although the individual courses are unpredictable, statistical analyses in cohorts of patients have provided some information about the long-term course of CD. A study of disease activity carried out in Copenhagen County reported that in the first year after diagnosis 80% of patients had high disease activity, 15% had low activity, and 5% were in remission. Over the first 7 years, 20% of patients had active disease every year, 13% had a relapse-free course, and 67% had disease that fluctuated between relapse and remission [18]. A subsequent study from Olmsted County reported that over time a

decreasing proportion of patients were in clinical remission, while an increasing proportion were in a post-surgical remission state. The proportions of patients with mild disease (12%) severe (drug-responsive, drug-dependent, or drug-refractory) disease (7%) were relatively constant [19]. Conventional corticosteroids were required by 43–56% of CD patients at some point in their disease history, and roughly 10% required corticosteroids in any given year [20,21]. In addition, 57–60% of all CD patients require at least one surgical resection [20]. The reported cumulative probability of surgery 10 and 15 years after the diagnosis of CD is 43–70% [21]. Patients with colonic disease that includes rectal involvement have a similar cumulative probability of surgery: half will undergo surgical resection within the first 10 years and slightly more than half of those will ultimately receive an ileostomy [22]. Data from endoscopic follow-up of patients after resection of ileocecal disease have shown that, in the absence of treatment, the postoperative recurrence rate is 65–90% within 1 year and 80–100% within 3 years of surgery [23]. Clinical recurrence without therapy is seen in 20–25% per year [23]. The postoperative clinical course of CD is best predicted by the severity of the endoscopically detected lesions. Typically, symptoms appear only when severe lesions are present, although it is not uncommon to see patients with advanced recurrent lesions at endoscopy who remain asymptomatic [24].

Survival

Long-term follow up studies of population-based cohorts of patients have shown a slight increase in the mortality rate of patients with CD late in the disease course, with an overall standardized mortality ratio (SMR) of 1.3–1.5 [25,26]. An unexpectedly high mortality among women with CD was reported, being most pronounced among women younger than 50 years at diagnosis [25,27]; more than one-third of the deaths among women seemed to be connected to CD vs. 20% of the deaths among men. A large Italian population-based series comprising patients with inflammatory bowel disease found a four-fold increase in mortality risk for lung cancer in CD patients [28].

Complications

Fistulation and abscess formation: Long-standing inflammation may lead to local penetration. With transmural involvement of the bowel, inflammation of the serosal surface incites an inflammatory exudate that attaches the bowel to adjacent structures, such as other bowel loops, muscle, or extraintestinal structures such as the bladder or vagina. Advancement of the fissure into these other structures creates a fistula, with an eventually associated abscess. Fistulizing CD at diagnosis has been reported in 14–20% of patients [1,29]; the most frequent fistula sites [1] are perianal (54%), enteroenteric (24%), rectovaginal (9%), enterocutaneous (6%), and enterovesicular (3%).

Cancer: As in ulcerative colitis, CD carries an increased risk of colorectal and small bowel cancers. Estimates of the magnitude of these risks have been derived from referral-based and population-based studies, but they have varied. Regional differences measured at different centers and populations around the world have been noted.

The reasons for the variation in risk assessment have been proposed to be related to study design, population characteristics, and the different types of therapy. A recent meta-analysis of intestinal cancer risk in CD, according to population-based studies only, revealed an overall increased risk of colorectal and small bowel cancers among patients with CD, with an overall standardized incidence ratio (SIR) for colorectal cancer of 1.9 (95% CI 1.4–2.5) and an overall pooled standardized incidence of small bowel cancer of 27.1 (95% CI 14.9–49.2) [30,31].

Different Patterns of Crohn's Disease: Clinical Disease Classification

Crohn's disease is a multifactorial polygenic disease with probable genetic heterogeneity. Several disease phenotypes that may be genetically determined have been suggested. Classifications based on disease behavior have been proposed in an attempt to provide information to patients regarding prognosis as well as to assist in decision-making for treatment strategies and in identifying the causes of CD. The most widely used classification systems are reported in Table 5.3.

Table 5.3 Proposed classification of patient subgroups in Crohn's disease

	Rome 1991	Vienna 1998	Montreal 2003
Age at diagnosis		A1 <40 years A2 >40 years	A1 <16 years A2 16–40 years A3 >40 years
Location	1. Stomach/duodenum 2. Jejunum 3. Ileum 4. Colon 5. Rectum 6. Anal-perianal	L1 Terminal ileum L2 Colon L3 Ileocolon L4 Upper gastrointestinal	L1 Terminal ileum L2 Colon L3 Ileocolon L4 Upper gastrointestinal
Extent	Localized (<100 cm) Diffuse		
Behavior		B1 Non-stricturing/ non-penetrating B2 Stricturing B3 Penetrating	B1 Non-stricturing/ non-penetrating B2 Stricturing B3 Penetrating P Perianal disease
Operative history	Primary Recurrent		

5

References

1. Schwartz DA, Loftus EV Jr, Tremaine WJ et al (2002) The natural history of fistulizing Crohn's disease in Olmsted County, Minnesota. Gastroenterology 122:875–880
2. Orchard TR, Wordsworth BP, Jewell JP (1998) Peripheral arthropathies in inflammatory bowel disease: their articular distribution and natural history. Gut 42:387
3. Hellers G, Bergstrand O, Ewerth S, Holmstrom B (1980) Occurrence and outcome after primary treatment of anal fistulae in Crohn's disease. Gut 21: 525–527
4. Chrispin AR, Tempany E (1967) Crohn's disease of the jejunum in children. Arch Dis Child 42:631-635
5. Louis E, Collard A, Oger AF et al (2001) Behaviour of Crohn's disease according to the Vienna classification: changing pattern over the course of the disease. Gut 49:777–782
6. Vind I, Riis L, Jess T et al (2006) Increasing incidence of inflammatory bowel disease and decresing surgery rates in Copenhagen city and county, 2003–2005: a population –based study from the danish crohn Colitis Database. Am J Gastroenterol 10:1274–1282
7. Louis E, Michel V, Hugot JP et al (2003) Early development of stricturing or penetrating pattern in Crohn's disease is influenced by disease location, number of flares, and smoking but not by NOD2/CARD15 genotype. Gut 52 (4):552–557
8. Cottone M, Rosselli M, Orlando A et al (1994) Smoking habitus and recurrence in Crohn's disease. Gastroenterology 106:643–648
9. Breuer-Katschusky BD, Hollander N et al (1996) Effect of cigarette smoking on the course of Crohn's disease. Eur J Gastroenterol Hepatol 8:225–228
10. Sutherland LR, Ramcharan S, Bryant H et al (1994) Effect of cigarette smoking on recurrence of Crohn's disease. Gastroenterology 98:1123–1128
11. Gasche C, Scholmerich J, Brynskov J et al (2000) A simple classification of Crohn's disease: report of the working party of the world congresses of gastroenterology, Vienna 1998. Inflamm Bowel Dis 6:8–15
12. Tarrant KM, Biomed B, Barclay ML et al (2008) Perianal disease predicts changes in Crohn's disease phenotype – results of a population-based study of inflammatory bowel disease phenotype. Am J Gastroenterol 103:3082–3093
13. Tavarela Veloso F, Carvalho J, Magro F (1996) Immune-related systemic manifestations of inflammatory bowel disease: a prospective study of 792 patients. J Clin Gastroenterol 23(1):29–33
14. Barreiro de Acosta JM, Dominguez-Munoz JE, Nunez-Pardo de Vera MC et al (2007) Relationship between clinical features of Crohn's disease and the risk of developing extraintestinal manifestations Eur J Gastroenterology Hepatology 19:73–78
15. Rothfuss KS, Stange EF, Herrlinger KR (2006) Extraintestinal manifestations and complications in inflammatory bowel disease. J Gastroenterol 12(30):4819–4831
16. Ardizzone S, Sarzi Puttini P, Cassinotti A, Bianchi Porro G (2008) Extraintestinal manifestations of inflammatory bowel disease Dig Liver Dis 40S:253–259
17. Levine B (2003) Extraintestinal manifestations of inflammatory bowel disease. In: Kirsner (ed) Inflammatory bowel disease. Saunders, Philadelphia, pp 397–409
18. Munkholm P, Langholz E, Davidsen M, Binder V (1995) Disease activity courses in a regional cohort of Crohn's disease patients. Scand J Gastroenterol 30:699–706
19. Silverstein MD, Loftus EV, Sandborn WJ et al (1999) Clinical course and costs of care for Crohn's disease: Markov model analysis of a population-based cohort. Gastroenterology 117:49–57
20. Loftus EV, Schoenfeld P, Sandborn WJ (2002) The epidemiology and natural history of Crohn's disease in population-based patient cohorts from North America: a systematic review. Aliment Pharmacol Ther 16: 51–60

21. Munkholm P, Langholz E, Davidsen M, Binder V (1994) Frequency of glucocorticoid resistance and dependency in Crohn's disease. Gut 35:360–362
22. Lapidus A (2006) Crohn's disease in Stockholm County during 1990–2001: an epidemiological update. World J Gastroenterol 12(1):75–81
23. Rutgeerts P, Geboes K, Vantrappen G et al (1990) Predictability of the postoperative course of Crohn's disease. Gastroenterology 99:956–963
24. Borley NR, Mortensen NJ, Jewell DP (1997) Preventing postoperative recurrence of Crohn's disease. Br J Surg 84:1493–1502
25. Jess T, Winther KV, Munkholm P et al (2002) Mortality and causes of death in Crohn's disease: follow-up of a population-based cohort in Copenhagen County, Denmark. Gastroenterology 122:1808–1814
26. Persson PG, Bernell O, Leijonmarck CE et al (1996) Survival and cause-specific mortality in inflammatory bowel disease: a population-based cohort study. Gastroenterology 110:1339–1345
27. Weterman IT, Biemond I, Pena AS (2002) Mortality and causes of death in Crohn's disease: follow up of a population–based cohort in Copenhagen County Denmark. Gastroenterology 122:1808–1814
28. Palli D, Trallori G, Saieva C et al (1998) General and cancer specific mortality of a population based cohort of patients with inflammatory bowel disease: the Florence Study. Gut 42 (2):175–179
29. Munkholm P, Langholz E, Nielsen OH et al (1992) Incidence and prevalence of Crohn's Disease in the County of Copenhagen 1962-1987: a sixfold increase in incidence. Scand J Gastroenterol 27:609–614
30. Jess T, Gamborg M, Matzen P et al (2005) Increased risk of intestinal cancer in Crohn's disease: a meta-analysis of populatioan-based cohort studies. Am J Gastroenterol 100:2724–2729
31. Masal G, Bagnoli S, Ceroti M et al (2004) Divergent patterns of total and cancer mortality in ulcerative colitis and Crohn's disease patients: the Florence IBD study 1978–2001. Gut 53:1309–1313
32. Lennard-Jones JE, Shivananda S (1997) Clinical uniformity of inflammatory bowel disease a presentation and during the first year of disease in the north and south of Europe. EC-IBD Study Group. Eur J Gastroenterol Hepatol 9 (4):353–359
33. Witte J, Shivananda S, Lennard-Jones JE et al (2000) Disease outcome in inflammatory bowel disease: mortality, morbidity and therapeutic management of a 796-person inception cohort in the European Collaborative Study on Inflammatory Bowel Disease (EC-IBD) Scand J Gastroenterol 35(12):1272–1277

Diagnostic and Therapeutic Role of Endoscopy

6

A. Andreoli, P. Meddi, P. Pantanella, A. Tesi

Over the past few decades, endoscopy has become an essential tool for the gastroenterologist treating patients with Crohn's disease (CD). Indications for endoscopy in CD patients include a correct diagnosis (both endoscopic and histological), assessment of disease localization and activity, diagnosis of postoperative recurrence, evaluation of perianal disease, dilation of strictures, and surveillance in patients with long-standing colonic disease.

Diagnosis

Colonoscopy with intubation of the terminal ileum should be always performed during the initial evaluation of patients with clinical symptoms suggestive of inflammatory bowel disease, because it allows direct visualization and biopsy of the mucosa and is useful to distinguish CD from ulcerative colitis (UC) and to rule out other causes of colitis, such as bacterial infections, ischemia, non-steroidal anti-inflammatory drug (NSAID) use, or mucosal changes due to bowel-cleansing preparations [1,2].

In order to differentiate CD from UC patients, it is very important to perform index endoscopy before therapy is started, because treatment might obscure discriminating features useful for a correct diagnosis, such as segmental colitis or rectal sparing [3,4]. Even if some endoscopic features (aphthous ulcers, serpiginous ulcers, and cobble-stoning of the mucosa; Fig. 6.1a–c) are suggestive for CD, none of them is specific. The most useful endoscopic features used to differentiate CD from UC are segmental involvement of the colon, rectal sparing, involvement of the distal ileum, and perianal disease. The findings of mild inflammatory mucosal lesions around the periappendiceal orifice in the setting of UC with an otherwise normal

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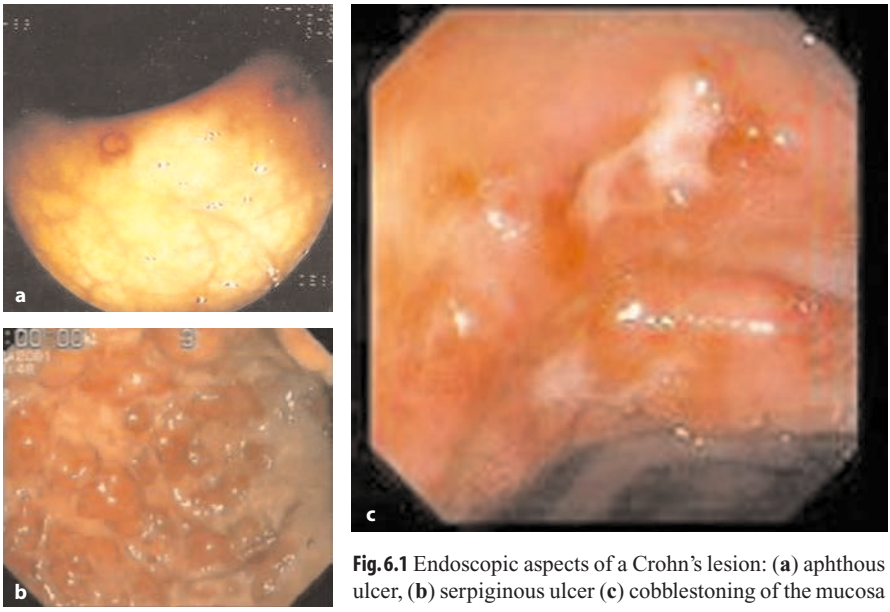

Fig. 6.1 Endoscopic aspects of a Crohn's lesion: (**a**) aphthous ulcer, (**b**) serpiginous ulcer (**c**) cobblestoning of the mucosa

right colon should not be confused with CD [5,6].

As reported in prospective studies, the experienced endoscopist is able to endoscopically distinguish CD from UC in about 90% of patients. In the 10% of patients in whom endoscopic features are specific for neither CD nor UC, the diagnosis of indeterminate colitis should be made [7,8].

Biopsies taken during endoscopy are important for distinguishing CD, UC, and other less frequent forms of chronic colitis, such as lymphocytic or collagenous colitis. Unluckily, although the histological criteria for distinguishing different forms of inflammatory bowel disease are well-established, there is important interobserver variability (65–76%) [9], and a final diagnosis should never be made based on histopathologic examination alone, but always also on clinical, radiological, and endoscopic criteria. During the course of the disease, new peculiarities may arise that allow a more definite diagnosis.

Endoscopic evaluation of the upper gastrointestinal tract may be important either for adequate assessment of disease localization, which can be useful for a correct therapy, or in the diagnosis of CD in a patient with indeterminate colitis [10]. Upper gastrointestinal tract involvement (proximal to the ligament of Treitz) occurs in up to 13% of patients with CD [8] and may include the esophagus [11], stomach, and duodenum [12]. It can be detected with a simple esophagogastroduodenoscopy (EGD).

It is interesting to note that granulomas (40–60%) are more often detected in duodenal biopsy specimens than in colon biopsy specimens [13,14]. Upper gastrointestinal lesions distal to the ligament of Treitz can be endoscopically visualized with wireless capsule endoscopy (WCE) and/or enteroscopy. The former technology allows for

direct visualization of the small bowel. The capsule is a small camera, about the size of a vitamin pill, that takes two pictures per second as it traverses the gastrointestinal tract. It is easily swallowed and transmits the images for 8 h to a recording device allocated around the abdomen. The images are downloaded to a computer and then reviewed by a gastroenterologist. The more frequent lesions visualized with WCE include mucous erosions, aphthae, lineal and serpiginous mucous lesions, ulcers, and fissures, but obviously these lesions are not specific for CD and it is possible to encounter them in patients taking NSAIDs and even in healthy subjects. A meta-analysis compared WCE with small bowel barium radiography: the yield of WCE was 64% vs. 24% for radiography, with an increasing yield of 40% and a number needed to treat (NNT) of 3 [15]. Four trials compared WCE to ileo-colonoscopy: WCE had a 61% yield compared to 46% for ileo-colonoscopy, with an increasing yield of 15% and a NNT of 7. Three studies compared WCE with CT enterography or enteroclysis: WCE had a 69% yield vs. 30% for CT scan, with an increasing yield of 39%. These results show an improvement in yield with WCE over all other modalities when used to evaluate small bowel involvement in patients with established CD, but there is only a trend toward significance in patients with suspected small bowel CD [15].

In CD studies, WCE-related adverse events occurred on average in 2% of patients, due to capsule battery expiration or capsule retention. Since 10% of CD patients suffer from intestinal strictures, they are not eligible for WCE. However, CD patients can be pre-tested for small bowel patency using a dissolving capsule: patients in whom the capsule is excreted intact and who do not experience any adverse event can be safely examined with standard WCE [16].

WCE is considered a less invasive, painfulness procedure that is well tolerated and accepted by patients; however, to date standardized criteria for the diagnosis of CD with WCE are still lacking. Therefore, instead, a push and pull enteroscopy (single or double balloon) is needed to provide pathologic confirmation of visualized lesions. Once the diagnosis of CD is made, the indications to perform repeat endoscopy apply only to particular aspects of the disease. In CD, the gold standard to monitor the presence of disease activity is the Crohn's disease activity index (CDAI), a clinical score that does not include endoscopic assessment. Accordingly, repeat colonoscopies have not been advocated to control therapeutic efficacy because most of the drugs prescribed to induce clinical remission are not useful in inducing mucosal healing.

There is recent evidence that with immunosuppression and infliximab treatment healing of the bowel can be achieved and that this affects the outcome of CD in terms of a decreased risk for disease-related procedures and hospital admissions [17]. For this reason, endoscopic assessment of mucosal healing may come to represent an independent therapeutic aim in evaluating new medical therapies for CD.

Diagnosis of Postoperative Recurrence

Recurrence after surgery is one of the main problems of patients with CD. In a majority of patients, disease recurs within the first year after surgery [18]. This recurrence

is often limited to the ileocolic anastomosis, with aphthous or linear ulcers extending only a few centimeters into the adjacent colon wall and neoterminal ileum. The severity of the endoscopic recurrence is graded using the Rutgeerts score (Table 6.1). It seems that an earlier and more severe endoscopically confirmed recurrence after surgery predicts a worse clinical course of the disease, suggesting the need for a more aggressive therapeutic approach.

Ileo-colonoscopy is considered the gold standard in detecting post-surgical recurrence, with a sensitivity of 90% and a specificity of 100%. WCE was proposed as an alternative tool for the diagnosis of recurrence but its sensitivity is low, 50–79%, with a specificity of 90%. Some studies showed that there is an underestimation of the Rutgeerts score in lesions detected by WCE compared with those identified by ileoscopy. In the absence of ileal recurrence, no proximal lesion was detected at WCE. In contrast, WCE could be useful in patients with established ileal recurrences because 50% of them have concurrent lesions in the proximal ileum or in the jejunum [19].

Table 6.1 Endoscopic scoring system for postoperative recurrence (Rutgeert's score)

Grade	Endoscopic findings
0	No lesions in the distal ileum
1	<5 aphthous lesions
2	>5 aphthous lesions with normal mucosa between the lesions, or skip lesions confined to ileocolonic anastomosis (i.e. <1 cm in length)
3	Diffuse aphthous ileitis with diffusely inflamed mucosa
4	Diffuse inflammation with already larger ulcers, nodules, and/or narrowing

Perianal Disease

During the last 10 years, the combination of endoscopy with ultrasound (EUS) has been shown to be a useful tool in the evaluation of colorectal, anal, and pelvic disorders, due to the high resolution of this technique, with its clearly distinguishable tissue-dependent echo signals.

To date, the role of EUS in inflammatory disease is limited to evaluating perianal disease in CD patients and the perirectal complications of CD. Some authors compared EUS, MRI, and examination under anesthesia (EUA) in patients with suspected Crohn's perianal fistulas. The accuracy of the three modalities for determining fistula anatomy in patients with perianal CD was comparable: EUS 91% (CI 75–98%), MRI 87% (CI 69–96%), and EUA 91% (CI 75–98%) [20].

A further study assessed whether the use of EUS as a guide to combination medical and surgical therapy during fistula healing led to a higher rate of resolution. The

conclusion of this report was that EUS may identify a subset of patients who can discontinue infliximab without recurrence of fistula drainage [21]. EUS allows the differentiation of simple from complex fistulae and evaluation of the fistula tracts in relation to the sphincter muscle. It is a highly sensitive procedure for detecting perianal abscesses and can be considered a valid alternative to MRI and to EUA for the assessment of perianal disease.

Dilation of Strictures

The course of CD is often complicated by gastrointestinal fibrosis and strictures, potentially leading to bowel obstruction and necessitating surgery in 20% of patients. Unfortunately, the disease commonly recurs after resection, sometimes leading to repeated surgery. For strictures accessible to endoscopy, alternative approaches have been proposed, including through-endoscope balloon dilation (Fig. 6.2a–c). Several studies [22] have reported the results of balloon dilation, showing a high technical success rate and good clinical efficacy (Table 6.2). Most of the dilations were performed in anastomotic strictures with a stricture length of generally <5 cm. The rate

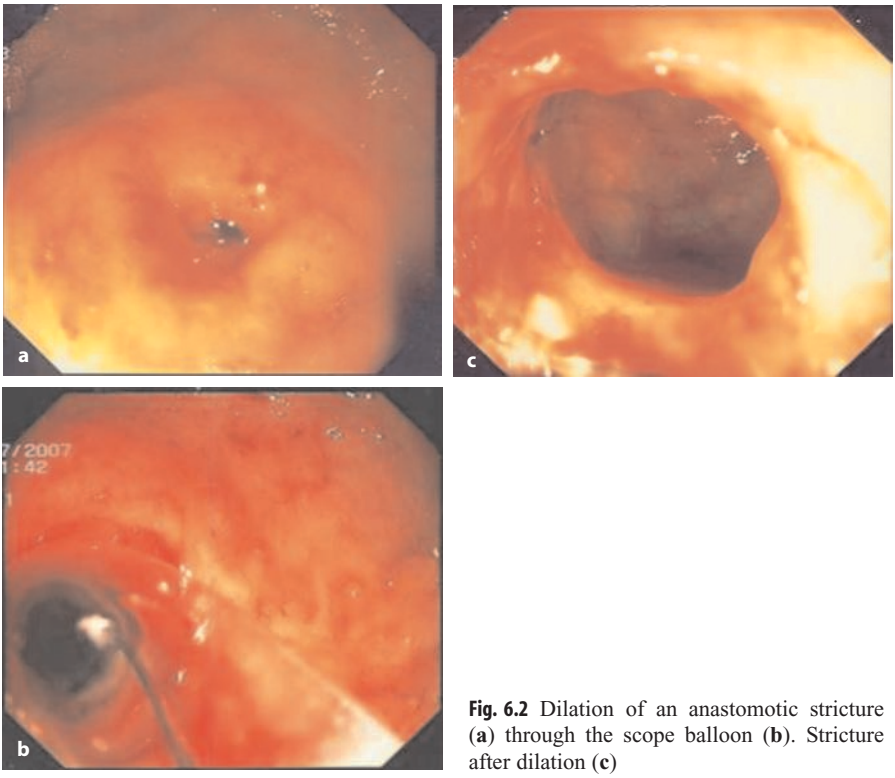

Fig. 6.2 Dilation of an anastomotic stricture (**a**) through the scope balloon (**b**). Stricture after dilation (**c**)

Table 6.2 Endoscopic dilation of strictures in Crohn's disease

Author	Patients) (n)	Postsurgical strictures (%)	Scope passing (%)	Technical success (%)	Clinical efficacy (%)	Complications (%)
Dear	22	91	45	100	73	0
Sabatè	38	65	56	84	53	3
Ferlitsch	46	62	–	85	57	4
Brooker	14	79	86	100	79	0
Gibson	59	73	74	73	41	3
Ramboer	13	71	77	100	100	0
Couckuyt	55	67	73	85	60	11
Morini	43	67	70	79	42	0
Blomberg	27	100	100	100	67	0
Singh	17	74	66	86	5	2

See [22] for more details

of major complications, including bowel perforation, was 2%, low enough to consider the procedure as relatively safe. Technical success, i.e., pneumatic dilation of the stenosis, was achieved in nearly 90% of patients even if in some cases the endoscopist was unable to pass the scope through the stricture, usually because of bowel angulations. Most of the patients who underwent dilatation had very good short-term clinical success, although only 30% of them maintained long-term clinical success with only one dilation. For this reason, subsequent scheduled dilations after an index dilation should be performed, even in the absence of obstructive symptoms, as this strategy could guarantee a longer surgery-free period. This approach supports the use of endoscopic dilatation to avoid or postpone the need for surgery in selected patients with CD strictures [23].

Surveillance Colonoscopy in Chronic Crohn's Colitis

The importance of surveillance colonoscopy in Crohn's colitis became particularly evident after a report by the group of Present, conducted in patients with long-standing extensive Crohn's colitis. In that longitudinal series of 259 patients, new low- or high-grade dysplasia (LGD, HGD) or cancer was found in 32 (12%) of the examined patients. After a negative screening result, the probability of finding neoplasia by the

fourth surveillance examination was 22%. Recently, the same group updated the results of that previous study: all patients had at least 7 years of Crohn's colitis affecting at least one-third of the colon; the patients were followed-up every 1–2 years. The cumulative risk of detecting an initial finding of any definite dysplasia or cancer after a negative screening colonoscopy was 25% by the tenth surveillance examination. The cumulative risk of detecting an initial finding of flat HGD or cancer after a negative screening colonoscopy was 7% by the ninth surveillance examination [24]. Therefore, periodic surveillance colonoscopy should be part of the routine management of patients with chronic extensive Crohn's colitis.

Chromoendoscopy with indigo-carmine or methylene blue dye in conjunction with magnifying endoscopes (Fig. 6.3a–c) is useful to detect subtle mucosal changes and has been shown to increase the yield of surveillance endoscopy in patients with inflammatory bowel disease [25]. Although these data need to be confirmed, chromoendoscopy is likely to become the procedure of choice for this indication.

Narrow band imaging (NBI) using a zoom scope presumably increases the sensitivity of the technique for the detection of dysplasia to a similar level as achieved with chromoendoscopy, but without the use of dyes. The accuracy of this technique for dysplasia screening is currently under investigation.

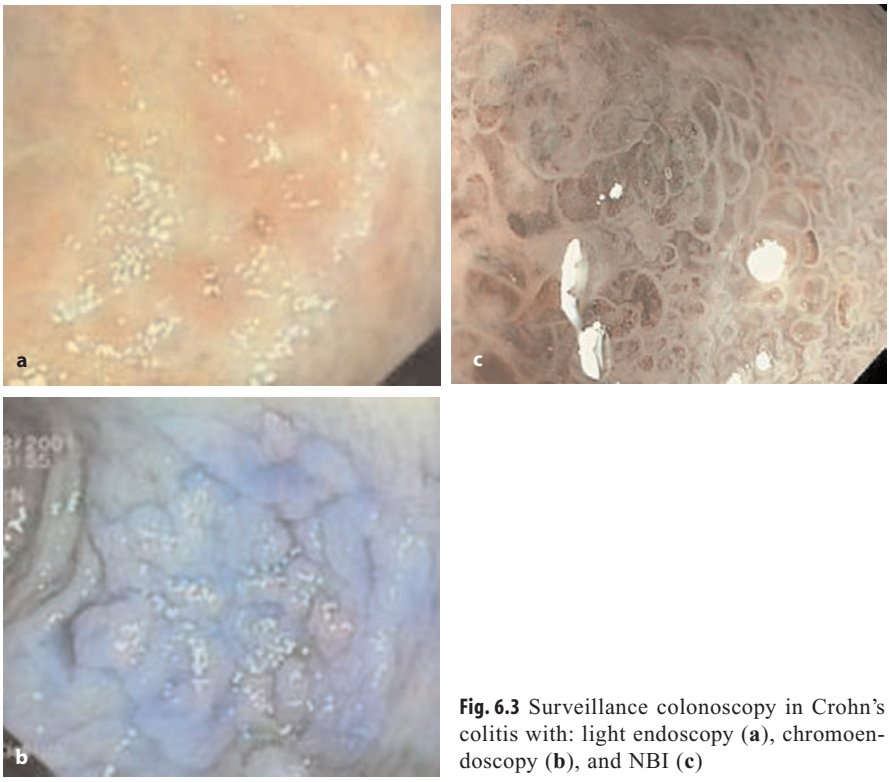

Fig. 6.3 Surveillance colonoscopy in Crohn's colitis with: light endoscopy (**a**), chromoendoscopy (**b**), and NBI (**c**)

References

1. Zwas FR, Cirillo NW et al (1996) Colonic mucosa abnormalities associated with oral sodium phosphate solution. Gastrointest Endosc 43:463–466
2. Rejchrt S, Bures J et al (2004) A prospective, observational study of colonic mucosal abnormalities associated with orally administered sodium phosphate for colon cleansing before colonoscopy. Gastrointest Endosc May 59(6):651–654
3. Kim B, Barnett JL et al (1999) Endoscopic and histological patchiness in treated ulcerative colitis. Am J Gastroenterol 94:3258–3262
4. Bernstein CN, Shanahan F et al (1995) Patchiness of mucosal inflammation in treated ulcerative colitis: a prospective study. Gastrointest Endosc 42:132–237
5. Okawa K, Aoki T et al (1998) Ulcerative colitis with skip lesions at the mouth of the appendix: a clinical study. Am J Gastroenterol 93:2405–2410
6. Byeon J-S, Yan S et al (2005) Clinical course of distal ulcerative colitis in relation to appendiceal orifice inflammation status. Inflamm Bowel Dis 11:366–371
7. Pera A, Bellando P et al (1987) Colonoscopy in inflammatory bowel disease. Diagnostic accuracy and proposal of an endoscopic score. Gastroenterology 92:181–185
8. Witte AM, Veenendaal RA et al (1998) Crohn's disease of the upper gastrointestinal tract: the value of endoscopic examination. Scand J Gastroenterol Suppl 225:100–105
9. Theodossi A, Spiegelhalter DJ et al (1994) Observer variation and discriminatory value of biopsy features in inflammatory bowel disease. Gut 35:961–968
10. Kundhal PS, Stormon MO et al (2003) Gastric antral biopsy in the differentiation of pediatric colitides. Am J Gastroenterol 98:557–561
11. Decker GA, Loftus EV Jr et al (2001) Crohn's disease of the esophagus: clinical features and outcomes. Inflamm Bowel Dis 7:113–119
12. Rutgeerts P, Onette E et al (1980) Crohn's disease of the stomach and duodenum: a clinical study with emphasis on the value of endoscopy and endoscopic biopsies. Endoscopy 12:288–294
13. Nugent FW, Roy MA (1989) Duodenal Crohn's disease: an analysis of 89 cases. Am J Gastroenterol 84:249–254
14. Tobin JM, Sinha B et al (2001) Upper gastrointestinal mucosal disease in pediatric Crohn's disease and ulcerative colitis: a blinded, controlled study. J Ped Gastroenterol Nutr 32:443–448
15. Triester S, Leighton et al (2006) A meta-analysis of the yield of CE compared to other diagnostic modalities in patients with non-stricturing small bowel Crohn disease. Am J Gastroenterol 101:954–964
16. Spada C, Riccioni ME et al (2007) Patients with known small bowel stricture or with symptoms of small bowel obstruction secondary to Crohn's disease should not perform video capsule endoscopy without being previously tested for small bowel patency. Am J Gastroenterol 102(7):1542–1543
17. Rutgeerts P, Vermeire S et al (2007) Mucosal healing in inflammatory bowel disease: impossible ideal or therapeutic target? Gut 56:453–455
18. Tytgat GNJ, Mulder GJJ et al (1988) Endoscopic lesions in Crohn's disease early after ileocecal resection. Endoscopy 20:260–262
19. A Bourreille, M Jarry et al (2006) Wireless capsule endoscopy versus ileocolonoscopy for the diagnosis of postoperative recurrence of Crohn's disease: a prospective study. Gut 55 (7):978–983
20. Schwartz DA, Wiersema MJ et al (2001) A comparison of endoscopic ultrasound, magnetic resonance imaging, and exam under anesthesia for evaluation of Crohn's perianal fistulas. Gastroenterology 121(5):1064–1072

21. Spradlin NM, Wise PE et al (2008) Use of endoscopic ultrasound to guide combination medical and surgical therapy for patients with Crohn's perianal fistulas. Am J Gastroenterol 103(10):2527–2535
22. Hassan C, Zullo A (2007) A systematic review: endoscopic dilatation in Crohn's disease. Aliment Pharmacol Ther 26:1457–1464
23. Andreoli A, Cosintino R et al (2008) Endoscopic dilation of Crohn's disease strictures: results of scheduled re-treatment. Gut 57(Suppl):P0690
24. Friedman S, Rubin PH et al (2008) Screening and surveillance colonoscopy in chronic Crohn's colitis: results of a surveillance program spanning 25 years. Clin Gastroenterol Hepatol 6(9):993–998
25. Kiesslich R, Jung M (2002) Magnification endoscopy: does it improve mucosal surface analysis for the diagnosis of gastrointestinal neoplasias? Endoscopy 34:819–822

Laboratory Tests

7

G. Famularo, G. Minisola

Introduction

It is unusual for patients with inflammatory bowel disease (IBD) to present with almost specific or pathognomonic clinical symptoms and signs. This implies that diagnosing Crohn's disease (CD) or ulcerative colitis (UC) is substantially dependent on the combination of a wide range of clinical, radiological, endoscopic, and histological findings that should be correctly interpreted by physicians in the appropriate setting. One additional important point is linked to recent advances in the field that have led to the widespread use of biological therapies in the management of patients with IBD, either CD or UC. This suggests that we urgently need reliable tools to stratify patients with different disease subtypes in terms of the risk of complications and a poor outcome as well as the probability to clinically respond to one treatment strategy, including biological therapies, rather than to others. Reliable tools are also required to appropriately follow-up patients during the course of their disease, especially methods that allow inflammatory activity and treatment effects to be monitored.

Several laboratory biomarkers are potentially helpful in the management of IBD patients, with C-reactive protein (CRP) being the most widely used in clinical practice. Fecal levels of calpoprotectin and lactoferrin are promising markers to be used in the near future in this field but data from evidence-based literature are relatively scarce and further properly designed investigations are required.

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C-Reactive Protein

Blood levels of CRP, the first acute-phase reactant to be described, are strongly increased in a wide range of inflammatory disorders [1]. CRP is elevated early after the onset of the inflammatory process and, due to the protein's short half-life, rapidly decreases after its resolution, making CRP an attractive marker of disease activity. Moreover, CRP testing is inexpensive and easy to perform and its levels are not affected by medications, except those such as corticosteroids that directly affect the underlying inflammatory disorder. Other advantages of measuring CRP are that there is no requirement for overnight fasting before blood sampling and the lack of any circadian cycle in CRP synthesis and release that might affect the reliability of measuring blood concentrations.

Important differences among IBD patients with regard to CRP blood concentrations have been identified: nonetheless, mechanisms responsible for those differences are still incompletely understood [2]. For example, CRP strongly increases during inflammatory flares of CD whereas, in contrast, UC patients usually have mildly increased or normal CRP concentrations even during the most severe inflammatory flares [2]. This suggests that measurement of blood CRP should be regarded as of little or no value to estimate and follow-up the inflammatory activity in UC patients, even though some investigators have suggested a relationship of CRP blood levels with both activity and extension of the disease [2]. However, the finding of significantly more elevated CRP concentrations among CD patients than in UC patients allows patients with active CD to be easily differentiated from those with inactive disease, but this remains an unreliable objective among UC patients.

The reasons for this discrepancy between CD and UC are as yet unknown. Moreover, blood and tissue levels of pro-inflammatory cytokines, such as interleukin (IL)-6, IL-1β and tumor necrosis factor (TNF)-α, are strongly increased in both conditions [2,3]. There is some evidence of a greater increase in peripheral blood IL-6 in CD patients than in UC patients and this might, at least in part, explain why blood CRP concentrations are more elevated in the former [4]. It is also worth noting that inflammation is limited to the colonic mucosa in UC whereas in CD there is transmural involvement with granulomatous inflammation, which indirectly suggests that a systemic inflammatory reaction with more elevated CRP blood levels is more likely to occur in CD than in UC [2,3].

Measurement of CRP has been shown to have sensibility and specificity as high as 80–85% among patients presenting with clinical features consistent with IBD, either CD or UC [2,3]. However, sensibility and specificity rise to near 100% if only CD patients are enrolled whereas they are 55–81% if only UC patients are enrolled [2,3]. One important point, however, relates to the crucial observation that CRP measurement does not add any further information in the work-up to the differential diagnosis between CD and UC.

In contrast, CRP blood levels have been demonstrated to strongly correlate with the inflammatory activity of CD, as measured by use of the Crohn's disease activity index (CDAI) [5]. Furthermore, CRP blood concentrations appear to closely corre-

late with several others inflammatory markers of disease activity, such as IL-6 and the fecal excretion of radiolabeled leukocytes [6]. In addition, because of CRP's short half-life (about 19 h), levels of the protein in CD patients are considered to reflect variations in inflammatory activity much more reliably than other acute-phase reactants with longer half-lives, such as fibrinogen. The close relationship linking blood concentrations of CRP with disease activity in CD patients has been demonstrated based on clinical parameters but also on the findings of endoscopic and histological investigations [7]. However, there is also evidence that about 10% of patients with active CD, according to clinical criteria, have persistently low to normal CRP blood levels [8]. The reasons for this discrepancy are unclear; nonetheless, it appears that those patients usually have ileal involvement with a stenosing phenotype along with a greater risk of intestinal resection rather than the fistulizing phenotype of the disease [8]. CRP stands as the best marker of clinical activity in CD patients compared with other biomarkers such as erythrocyte sedimentation rate (ESR), but ESR better correlates with endoscopic activity criteria of the disease [9].

There is no complete agreement about the clinical utility of measuring CRP in order to establish both the presence and activity of CD because of the large overlap in the CRP levels of patients with mild, moderate, or severe disease activity [2,3]. Consequently, CRP is a useful bio-marker to longitudinally monitor clinical disease activity in each single patient rather than to compare clinical activity between different subjects [2,3].

One important point is that the probability of experiencing a CD relapse appears to be significantly greater in patients with quiescent CD who have elevated CRP blood levels than in those with low to normal CRP [10]. However, several studies have clearly demonstrated that at least one-third of patients with disease relapse had low to normal CRP before the onset of a new flare-up [11]. Furthermore, there is also evidence that a non-negligible proportion of patients with inactive CD has elevated CRP blood levels but do not experience any disease relapse [11]. Therefore, the predictive value of CRP to estimate the relapse risk among patients with inactive CD remains to be firmly established and more studies are needed.

A relevant advantage of using CRP as a marker of CD inflammatory activity is that blood levels of the protein are not directly affected by the use of anti-inflammatory or immuno-modulating drugs. Rather, changes in blood CRP levels seem to reflect the impact of treatment on disease course and inflammatory activity [2,3]. This suggests that, when CRP decreases after a course of anti-inflammatory treatment, it reflects a positive effect of therapy upon intestinal inflammation even when there has been no clear-cut improvement in clinical parameters of disease activity. In contrast, if CRP levels remain persistently elevated despite treatment, it is reasonable to conclude that the underlying intestinal inflammation is not efficiently down-modulated by treatment. Despite the fact that CRP is not the gold-standard for the follow-up of UC patients, some studies have consistently suggested that when blood CRP remains constantly elevated above the cut-off level of 45 mg/l this predicts the need for colectomy in these patients despite treatment with intravenous steroids or cyclosporine [12].

The use of infliximab or other TNF antagonists has been a major advance in the

treatment of IBD. In this setting, there are data in favor of the view that patients with CRP levels >5 mg/l have the greatest probability to respond to infliximab or other anti-TNF agents, such as adalimumab [13,14].

Fecal Calprotectin

The advantage of fecal markers such as calprotectin is the greater specificity for the diagnosis and follow-up of intestinal disorders, including CD and UC. Several intestinal disorders, among them IBD, are characterized by the abundant fecal excretion of leukocytes. Calprotectin represents about the 60% of leukocyte cytosol proteins. Thus, the amount of fecal calprotectin mirrors the amount of leukocytes excreted in the feces and thus both the severity and the extent of intestinal inflammation. Several studies have indeed demonstrated a close relationship between fecal calprotectin and leukocytes labeled with indium-111 [15]. Furthermore, calprotectin remains stable in the feces for as long as one week and no more than 5 grams of feces are needed to consistently measure calprotectin concentrations [16]. An important disadvantage is represented by the fact that fecal levels of calprotectin may be substantially affected by the use of non-steroidal anti-inflammatory drugs and proton-pump inhibitors [16]. In addition, calprotectin levels may also change with advancing age, irrespective of the presence of an intestinal disorder, with diet, and also with physical activity [16].

Fecal calprotectin does not appear to be the gold-standard for the diagnosis and follow-up of CD and UC patients since many other gut disorders, ranging from cancer to infections, could result in a strong increase in the fecal excretion of calprotectin [17]. For example, no significant difference in fecal calprotectin levels has been demonstrated between IBD patients and those with colorectal cancer [18]. However, despite these caveats, it should be kept in mind that fecal concentrations of calprotectin are low to normal in patients with functional disorders such as irritable bowel syndrome; thus, this marker could be a useful tool to effectively differentiate irritable bowel syndrome from IBD or colorectal cancer [17]. It is otherwise clear that the finding of elevated fecal calprotectin should prompt an endoscopy to confirm suspicion of a potentially severe and life-threatening intestinal disorder. Among patients presenting with clinical features consistent with CD or UC, measurement of fecal calprotectin has been estimated to have a sensitivity and specificity for the correct diagnosis of around 90% [17,18]. Fecal calprotectin seems to be more elevated among CD patients than among those with UC, with sensitivity and specificity for the diagnosis of CD being >95% [19]. Since fecal calprotectin is a strong negative predictor for inflammatory and neoplastic disorders, it has been also proposed that the finding of low to normal fecal concentrations is an indication to not perform endoscopic investigations, which in these cases are deemed unnecessary [17,18].

Some studies have reported the interesting finding of abnormally elevated fecal calprotectin among healthy first-degree relatives of CD patients [20]. Whether this represents a marker for identifying healthy subjects at greater risk of developing CD or UC is still unproven and needs to be explored in further studies. It is also unclear whether

a close relationship can be established between fecal levels of calprotectin and clinical, endoscopic, and histological parameters of either CD or UC. There is some evidence that fecal calprotectin more reliably reflects inflammatory activity as evaluated based on histological findings rather than on endoscopic findings [21]. This indirectly suggests that fecal calprotectin is a more sensitive marker than endoscopy to estimate inflammatory activity in CD and UC patients. Furthermore, the severity rather than the extent of UC correlates with fecal calprotectin concentrations [22].

One promising advantage of using fecal calprotectin is the potential to predict the risk of IBD relapse. Elevated fecal concentrations of calprotectin among both CD and UC patients are closely associated with a clinically significant risk of disease relapse [23]. It has been estimated that the sensitivity and specificity of fecal calprotectin for predicting relapse are around 90% and 83%, respectively [24]. In addition, more elevated fecal calprotectin levels appear to predict an early relapse among patients with quiescent IBD [23]. It is also worth noting that the predictive value of CRP or ESR with respect to disease relapse remains to be firmly established.

Finally, since the normalization of fecal calprotectin during treatment for IBD closely mirrors the normalization of endoscopic findings, this result would provide a very reliable marker of treatment response and recovery probability [25].

Fecal Lactoferrin

Lactoferrin is an iron-binding glycoprotein of activated neutrophils that is much more stable in feces than other leukocyte proteins, such lysozyme, elastase, and myeloperoxidase. Fecal concentrations of lactoferrin are strongly elevated in IBD patients but also in patients with infectious enteritis and colitis, either viral or bacterial [16]. Therefore, the differential diagnosis of CD or UC and infectious enteritis or colitis cannot be established based upon fecal lactoferrin levels alone. It is unclear whether the elevated levels of fecal lactoferrin in IBD patients are triggered by a previous *Clostridium difficile* infection [26]. In patients with elevated fecal lactoferrin, it is mandatory to perform endoscopy in order to not overlook a cancer or IBD; however, low to normal fecal concentrations suggest that a functional disorder such as irritable bowel syndrome is a reasonable diagnosis. Among patients presenting with clinical features consistent with CD or UC, fecal lactoferrin has a sensitivity and specificity for the diagnosis of IBD of 82% and 93%, respectively [16]. However, measuring fecal lactoferrin seems to be less effective than measuring calprotectin for the differential diagnosis between IBD and irritable bowel syndrome [27,28].

No study has definitely demonstrated that fecal concentrations of lactoferrin closely mirror inflammatory activity in either CD or UC patients, mainly because of a large overlap in the lactoferrin levels of those with inactive disease and those with active disease [27].

Only one study has so far evaluated the effectiveness of using fecal lactoferrin as a marker to assay the clinical response to treatment among IBD patients. Buderus and colleagues indeed observed a close relationship linking the decrease in lactoferrin

concentrations with reduced CD inflammatory activity in the response to treatment with infliximab [29]. Even though these results are promising, the study has several limitations, including the small number of enrolled patients and the lack of an appropriately designed control group.

Autoantibodies

Perinuclear antineutrophilic cytoplasmic antibodies (pANCAs) may be found in 60–70% of UC patients and 5–10% of CD patients [30]. The antigens to which these autoantibodies are directed have not yet been identified but there is evidence that they are distinct from those associated with systemic vasculitides and likely to represent antibodies that cross-react with a wide range of bacteria of the indigenous gut flora. pANCA positivity is usually associated with pancolitis, early surgery, pouchitis, and primary sclerosing cholangitis in UC patients whereas ANCA-positive CD patients often have colonic disease closely resembling UC [30].

In contrast, anti-*Saccharomyces cerevisiase* antibodies (ASCAs) are detected in about 70% of CD patients and in no more than 15% of UC patients [31]. ASCA positivity is associated with an increased rate of early CD complications and seems to predict the risk that patients will require small bowel surgery [31].

References

1. Pepys MB, Hirschfield GM (2003) C-reactive protein: a critical update. J Clin Invest 111:1805–1812
2. Vermeire S, Van Assche G, Rutgeerts P (2004) C-reactive protein as a marker for inflammatory bowel disease. Inflamm Bowel Dis 10:661–665
3. Vermeire S, Van Assche G, Rutgeerts P (2005) The role of C-reactive protein as an inflammatory marker in gastrointestinal diseases. Nat Clin Pract Gastroenterol Hepatol 2:580–586
4. Gross V, Andus T, Caesar I et al (1992) Evidence for continuous stimulation of interleukin-6 production in Crohn's disease. Gastroenterology 102:514–519
5. Tromm A, Tromm CD, Huppe D et al (1992) Evaluation of different laboratory tests and activity indices reflecting the inflammatory activity of Crohn's disease. Scand J Gastroenterol 27:774–778
6. Nielsen OH, Vainer B, Madsen SM et al (2000) Established and emerging biological activity markers of inflammatory bowel disease. Am J Gastroenterol 95:359–367
7. Solem CA, Loftus EV Jr, Tremaine WJ et al (2005) Correlation of C-reactive protein with clinical, endoscopic, histologic, and radiographic activity in inflammatory bowel disease. Inflamm Bowel Dis 11:707–712
8. Florin TH, Paterson EW, Fowler EV et al (2006) Clinically active Crohn's disease in the presence of a low C-reactive protein. Scand J Gastroenterol 41:306–311
9. Cellier C, Sahmoud T, Froguel E et al (1994) Correlations between clinical activity, endoscopic severity, and biological parameters in colonic or ileocolonic Crohn's disease. A prospective multicentre study of 121cases. The Groupe d'Etudes Therapeutiques des Affections Inflammatoires Digestives. Gut 35:231–235

10. Lemann M, Mary JY, Colombel JF et al (2005) A randomized, double-blind, controlled withdrawal trial in Crohn's disease patients in long-term remission on azathioprine. Gastroenterology128:1812-1818
11. Boirivant M, Leoni M, Tariciotti D, Fais S, Squarcia O, Pallone F (1988) The clinical significance of serum C reactive protein levels in Crohn's disease. Results of a prospective longitudinal study. J Clin Gastroenterol 10:401–405
12. Travis SP, Farrant JM, Ricketts C et al (1996) Predicting outcome in severe ulcerative colitis. Gut 38:905–910
13. Louis E, Vermeire S, Rutgeerts P et al (2002) A positive response to infliximab in Crohn's disease: association with a higher systemic inflammation before treatment but not with -308 TNF gene polymorphism. Scand J Gastroenterol 37:818–824
14. Beaven SW, Abreu MT (2004) Biomarkers in inflammatory bowel disease. Curr Opin Gastroenterol 20:318–327
15. Tibble J, Teahon K, Thjodleifsson B et al (2000) A simple method for assessing intestinal inflammation in Crohn's disease. Gut 47:506–513
16. Poullis A, Foster R, Northfield TC, Mendall MA (2002) Review article: faecal markers in the assessment of activity in inflammatory bowel disease. Aliment Pharmacol Ther 16:675–681
17. Aadland E, Fagerhol MK (2002) Faecal calprotectin: a marker of inflammation throughout the intestinal tract. Eur J Gastroenterol Hepatol 14:823–825
18. Summerton CB, Longlands MG, Wiener K, Shreeve DR (2002) Faecal calprotectin: a marker of inflammation throughout the intestinal tract. Eur J Gastroenterol Hepatol 14:841–845
19. Costa F, Mumolo MG, Bellini M et al (2003) Role of faecal calprotectin as non-invasive marker of intestinal inflammation. Dig Liver Dis 35:642–647
20. Thjodleifsson B, Sigthorsson G, Cariglia N et al (2003) Subclinical intestinal inflammation: an inherited abnormality in Crohn's disease relatives? Gastroenterology 124:1728–1737
21. Bunn SK, Bisset WM, Main MJ et al (2001) Fecal calprotectin: validation as a noninvasive measure of bowel inflammation in childhood inflammatory bowel disease. J Pediatr Gastroenterol Nutr 33:14–22
22. Roseth AG, Aadland E, Jahnsen J, Raknerud N (1997) Assessment of disease activity in ulcerative colitis by faecal calprotectin, a novel granulocyte marker protein. Digestion 58:176–180
23. Costa F, Mumolo MG, Ceccarelli L et al (2005) Calprotectin is a stronger predictive marker of relapse in ulcerative colitis than in Crohn's disease. Gut 54:364–368
24. Tibble JA, Sigthorsson G, Bridger S et al (2000) Surrogate markers of intestinal inflammation are predictive of relapse in patients with inflammatory bowel disease. Gastroenterology 119:15–22
25. Roseth AG, Aadland E, Grzyb K (2004) Normalization of faecal calprotectin: a predictor of mucosal healing in patients with inflammatory bowel disease. Scand J Gastroenterol 39:1017–1020
26. Vaishnavi C, Kochhar R, Bhasin D et al (2003) Simultaneous assays for Clostridium difficile and faecal lactoferrin in ulcerative colitis. Trop Gastroenterol 24:13–16
27. Kane SV, Sandborn WJ, Rufo PA et al (2003) Fecal lactoferrin is a sensitive and specific marker in identifying intestinal inflammation. Am J Gastroenterol 98:1309–1314
28. Silberer H, Kuppers B, Mickisch O et al (2005) Fecal leukocyte proteins in inflammatory bowel disease and irritable bowel syndrome. Clin Lab 51:117–126
29. Buderus S, Boone J, Lyerly D, Lentze MJ (2004) Fecal lactoferrin: a new parameter to monitor infliximab therapy. Dig Dis Sci 49:1036–1039
30. Ferrante M, Henckaerts L, Joossens M et al (2007) New serological markers in in inflammatory bowel disease are associated with complicated disease behaviour. Gut 56:1394–1403
31. Schoepfer AM, Schaffer T, Mueller S et al (2009) Phenotypic associations of Crohn's disease with antibodies to flagellins A4-Fla2 and Fla-X, ASCA, p-ANCA, PAB, and NOD2 mutations in a swiss cohort. Inflamm Bowel Dis (in press)

Imaging

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Imaging modalities play an important role in the diagnosis and management of patients with Crohn's disease (CD). Imaging is used for diagnostic purposes as well as in the evaluation of the nature, gravity, and extraintestinal complications of the disease. It also provides information on the degree of inflammation and useful feedback after clinical or surgical therapy. To this end, endoscopy and conventional radiological and cross-sectional imaging methods (ultrasound, CT, and MR) are mainly used. Radiological and cross-sectional imaging tests are particularly significant in the evaluation of the small intestine, given the fact that its endoscopic investigation is incomplete or almost impossible.

Conventional Radiology

When dealing with CD, the clinician should no longer regard traditional radiological studies with barium as the first-line screening strategy; however, these studies may still have a complementary role, e.g., in more specifically targeting the alterations diagnosed with CT studies. Although barium radiology studies were the only means to investigate virtually every bowel disease for more than 50 years, the use of this method in clinical practice has progressively declined in the last 20 years [1,2]. The main reason is the long time that is required for the barium to opacify all of the intestinal loops, as expected when considering the remarkable length of the small bowel, the effect of an eventual delay in gastric emptying, and the physical obstacles due to Crohn's-disease-mediated alterations. Secondly, due to precipitation and deterioration of the contrast agent—inevitable during examinations of long duration–detailed imaging of the bowel loops cannot be achieved, severely hampering the accuracy of

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the study in its ability to detect early or small lesions and to estimate the potential extent of the Crohn's alterations.

Several techniques have been proposed to improve the efficacy of radiological studies with per os barium. These have been aimed at accelerating progression of the contrast agent through the bowel loops, resulting in shorter examination times and a reduction in artifacts due to barium degradation.

There is a wide agreement that distension of the bowel loops is necessary in order to appropriately visualize alterations in the bowel wall. This is usually achieved by positioning a nasojejunal catheter at the level of the duodenojejunal flexure of Treitz (enteroclysis). The technique may be performed using diluted barium or a mix of barium with either methyl-cellulose or air, after full-bowel preparation [3,4]. Double-contrast enteroclysis allows simultaneous imaging of the entire small bowel while providing optimal distention of the intestinal loops. It also allows visualization of the mucosal surface at different angles and exploration of the different bowel loops by transparency studies (Fig. 8.1).

When applied to CD, enteroclysis provides accurate assessment of the small bowel, which is extremely valuable in the differential diagnosis and in the grading and staging of CD. In particular, it allows a careful estimate of disease extent, the identification of discrete lesions, the characterization of strictures, and measurement of the length of the unaffected bowel. In addition, enteroclysis also provides accurate depiction of very early CD lesions, so that new findings, such as lymphoid nodular hyperplasia and apthoid ulcers, can be identified. Consequently, the specificity of the technique is very high, 96.9–98.3%, with an overall diagnostic accuracy of 93–99.3% in different series [5,6].

The first step in the pathogenesis of the swelling of the valvulae conniventes is edema of the lamina propria and the submucosal layer. Subsequently, with inflammatory hypertrophy of the lymphatic structures, there is irregular bulging of the mucosa, giving it a nodular appearance. Subsequent disruptions of the mucosal epithelium result in the onset of apthoid ulcer, which is the earliest mucosal lesion detectable by means of radiology (Fig. 8.2) [7]. In imaging studies, the lesion appears as small collection of barium surrounded by a radiotransparent halo. It is due to a discrete inflammatory process that interrupts the normal mucosa.

Fig. 8.1 Double-contrast enteroclysis with barium and methyl-cellulose. Normal findings

Fig. 8.2 Crohn's disease. The apthoid ulcer appears as a central lesion with a peripheral radiotransparent halo

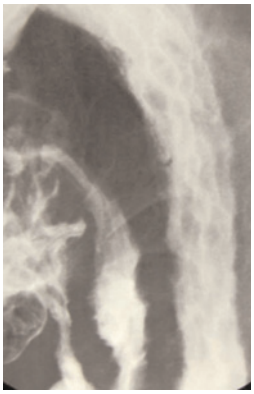

Fig. 8.3 Cobblestone appearance in a patient with Crohn's disease. The finding is due to the presence of deep linear ulcers separated by mucosal areas swollen because of the inflammatory process

During the progression of CD, the apthoid ulcers enlarge and merge to form linear ulcers. The mucosa entrapped between the ulcers thickens, because of swelling and inflammation, resulting in the characteristic cobblestone picture of CD (Fig. 8.3). As the disease continues to evolve, usually at the same location as the earlier lesions, two different radiological pictures develop that may be present separately or simultaneously. The first consists of concentric thickening of the bowel wall, lending a typical string-like aspect to the involved segment, usually the distal ileum. At this stage, obstruction is more frequently due to a spastic component rather than to a fibrotic process. Consequently, it is possible to detect partial resolution of the stricture and the reappearance of peristaltic activity during the same examination. The fibrotic evolution of the stricture results in the loss of its tubular aspect, because of the stiffness of the bowel wall (Fig. 8.4). The disease tends to be asymmetric, mainly involving and shortening the mesenteric side of the loops, so that the strictures are irregular and characterized by sparse pseudodiverticula on the anti-mesenteric side of the bowel wall.

The typical aspect of CD also involves extra-luminal extensions of the lesions, with the formation of entero-enteric or entero-colic fistulas. A more direct visualiza-

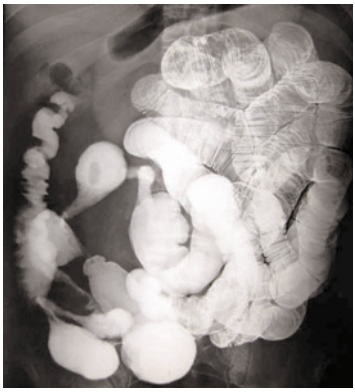

Fig. 8.4 Crohn's disease. Strictures alternate with dilated segments

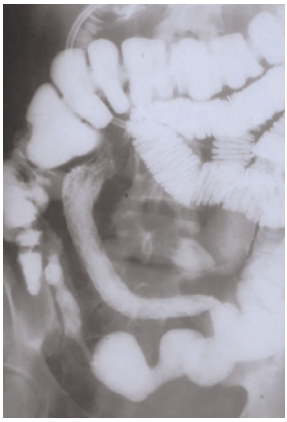

Fig. 8.5 In this patient with Crohn's disease, fibro-fatty mesenteric proliferation separates the various loops because of a mass effect

tion of the mesenteric alterations may be achieved with modern imaging techniques such as CT and MRI. However, these are inferior to enteroclysis in describing the mucosal alterations of CD (Fig. 8.5). Moreover, the development of double-contrast enteroclysis has allowed, for the first time, non-surgical evaluation of the entire length of the small bowel. This ability is extremely important in predicting the risk of short bowel syndrome, especially in Crohn's patients who need to undergo extensive surgical resections. Therefore, when performed before surgery, enteroclysis may result in more conservative and better targeted surgical approaches [8].

Cross-sectional Imaging

Ultrasound

Traditional ultrasound provides an accurate diagnosis of patients with CD and represents an initial screening procedure for patients with symptoms such as abdominal pain

and diarrhea. It is only slightly invasive and can be repeated during the follow-up phase to assess the response to therapy and potential disease complications. Ultrasound also highlights any recurrence of the disease in patients surgically treated for CD and in patients with relative or absolute contraindications, for example, children, pregnant women, and the disabled, to the usual radiological and endoscopic diagnostic investigations [9]. The sensitivity of ultrasound examinations in the study of CD is 60–95%, with a specificity of 80–100% [10]. In patients suspected of having the disease, the sensitivity increases to 74–92%. The sensitivity of ultrasound is also high in patients with active CD and in the detection of complications, due to the transmural nature of the inflammatory process [9]. Nonetheless, ultrasound has a number of shortcomings as it only provides access to the colon and lower portions of the small intestines, unlike CT and MR scans and other recognized radiological investigations. In addition, several factors prevent an accurate ultrasound examination, such as meteorism, peristalsis, and obesity. For the radiologist to make an accurate ultrasound diagnosis, the patient should abstain from food for at least 4 h in order to reduce intestinal peristalsis and endoluminal gas, but neither a special diet nor intestinal cleansing is normally necessary [11].

Conventional ultrasound based on linear or convex probes with a frequency range of 3.5–7.5 MHZ are used to examine the intestinal loops. The test is done with the patient in supine decubitus and begins with an exploration of the right lower abdominal quadrant to assess the last ileal loop and the ileocecal valve; the remaining abdominal quadrants are subsequently examined to evaluate the small intestine and colon loops. Color power Doppler imaging modalities provide additional information on the activity index of the disease based on the speed of blood flow through the vessels in the intestinal walls, spectral analysis and an assessment of the resistance index (RI), and flows in both the intraparietal area and in the superior and inferior mesenteric arteries (SMA-IMA) [9].

The examination is initially done without any contrast material; however, to more accurately evaluate the disease activity index, contrast agents can be subsequently administered, either orally or by intravenous injection. Recent studies showed that the use of endoluminal contrast agents increases sensitivity in the diagnosis of inflammatory bowel diseases (IBDs) involving the jejunum or colon [11]. If contrast agents based on sulfur fluoride microbubbles are used, it is necessary to obtain informed consent and an anamnesis to determine any allergy to the contrast agent or a history of recent myocardial infarction or ischemic heart diseases, both of which are contraindications to its use.

The ultrasound diagnosis of IBDs is carried out on the basis of several parameters referring to the parietal thickness, the features of the intestinal loops, and extraparietal abnormalities, such as mesenteric thickening, regional or localized lymphadenomegaly, abscess collections, enteroenteric, enterocutaneous, and enterovesical fistulas, and inflammation of adjacent areas such as the bladder and reproductive organs. Endocavitary ultrasound is necessary in patients with peri-rectal abscess and fistula [9,12].

Five layers with different ultrasound imaging features are differentiated in the wall of the digestive tract. From the endoluminal side these are: (1) the hypere-

chogenic layer, which corresponds to the mucosal surface, (2) the hypoechogenic layer of the tunica mucosa, (3) the hypoechogenic layer of the tunica submucosa, (4) the hypoechogenic layer of the tunica submuscularis propria, and (5) the hyperechogenic layer representing the serosa or the interface between the wall and the perivesicular spaces (Fig. 8.6). The imaging of each segment of the digestive tract is, among other factors, determined by the individual's functional status, which is influenced by peristaltic activity, the degree of repletion, and the nature of the items contained in the segment analyzed. A hollow viscus may appear collapsed, its distension caused by liquid, partially fluid contents, or gas [9].

The most common criterion used in the diagnosis of CD is parietal thickness and echo-structure (Fig. 8.7). Parietal thickening, measured as a wall thickness > 3 mm, is the typical sign although it is not specific for the disease. On longitudinal and transversal scans, it produces the typical layered or targeted pattern [9–11]. The degree of wall thickening is influenced by several factors; some of them are related, e.g., edema and cellular infiltration, whereas others are not, such as fibrosis and distension. In patients with CD, an increase in the hyperechogenic layer of the submucosa is the main cause of parietal thickening. Other factors are, on the one hand, the irregularity and segmented appearance of the central hyperchogenic line with respect to endoluminal gas, due to mucosal ulcers, and on the other, the interruption of the hypoechogenic tunica mucosa, due to the formation of microabscesses and intramucosal fistulas. In general, the altered loop is hypoperistaltic and fixed; a dilated loop with marked peristalsis is also frequently seen on the upper side of the stenotic tract (Fig. 8.8). It is sometimes possible to ultrasonographically differentiate between an intestinal loop regarded as normal based on its parietal thickness and echo-structure and an altered loop characterized by a loss of layering. Large polyps of the terminal ileum and of the ascending colon can be seen in patients with active disease [13]. Among the recognized IBDs, unaltered wall layering is visible in patients with ulcerative colitis, which is characterized by a more superficial inflammatory process. This criterion can be helpful in distinguishing ulcerative proctocolitis from CD [11].

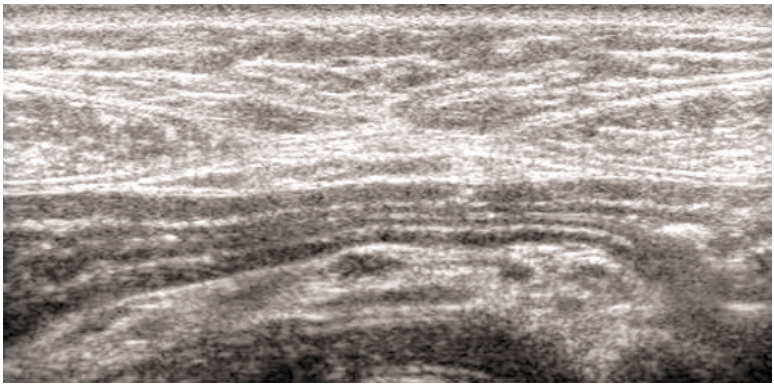

Fig. 8.6 Mural stratification of the bowel wall: ultrasound anatomy

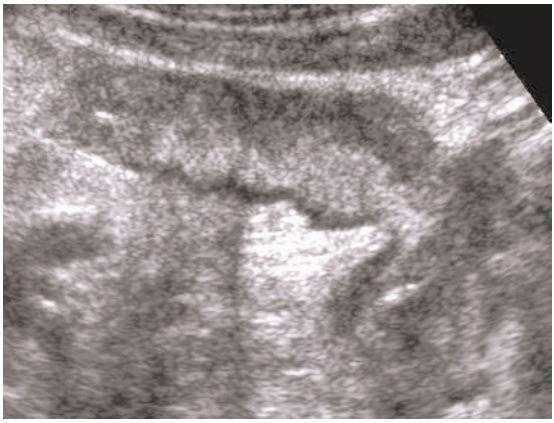

Fig. 8.7 Thickened wall of the ileum, with modified bowel wall stratification and fibro-fatty proliferation

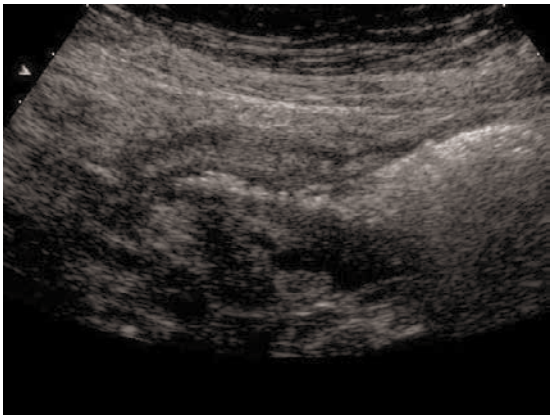

Fig. 8.8 Enlarged ileum in the presence of a stiff and thickened small bowel loop

Color power Doppler imaging is used to evaluate the neovascularization of the altered loop. Thus, where parietal thickening is associated with active inflammation, intraparietal flow signals can be visualized whereas they do not appear in individuals with fibrosis (Fig. 8.9). The relevance of spectral investigations (RI measurement) of the SMA is controversial [9,11]. However, in the presence of complications, color Doppler can often distinguish phlegmons, i.e., mesenteric vascularized masses with internal flow signals, from abscesses, which are seen as fluid collections with peripheral vascularization [10].

As noted above, contrast agents can be helpful in both the diagnosis of CD and the evaluation of the disease activity index. Orally administered contrast agents allow distension of the small bowel loops. Polyethylene glycol (PEG), a non-absorbable, non-fermenting, and non-digestible hydrophilic molecule, is typically provided at a dose of 200–600 ml (400 ml on average) and allows visualization of the intestinal walls [14].

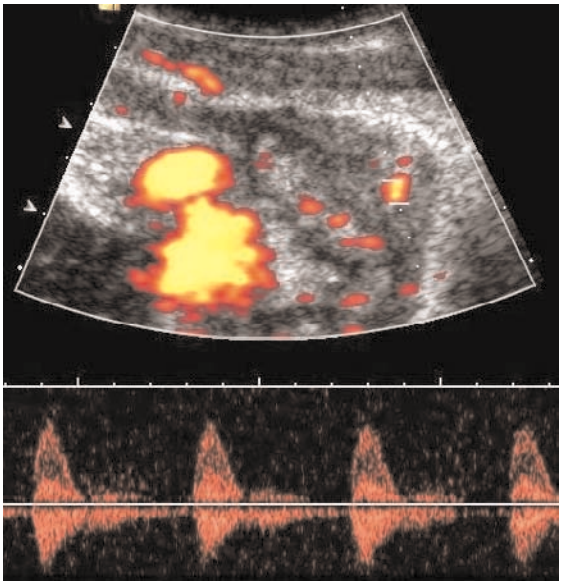

Fig. 8.9 Color Doppler ultrasound: increased vascularity of the thickened bowel wall

This approach results in more accurate observation of the affected area and of disease progression compared to traditional echographies. It also provides a better evaluation of stenosis [10]. Second-generation intravenous contrast agents together with the low mechanical index acquisitions performed by dedicated software have facilitated studies of the microcirculation of the parenchymatous organs and of neoangiogenesis of tumoral tissues. This technique has been used to achieve parietal enhancement in patients with CD and has led to the identification of four distinct patterns:

- In pattern I, or the monolayered pattern, there is an intense impregnation that affects the entire intestinal wall. This is observed in patients with thickened loops and consists of hypoechogenicity with loss of parietal layering and color Doppler parietal signals.
- In pattern II, or the double-layered pattern, the mucosa and submucosa, rather than the external muscular layer, are affected by impregnation. This is observed in patients with thickened loops, partially altered layering, and fewer parietal vessels.
- In pattern III, or the three-layered pattern, there is no impregnation of the mucosa and muscular tunicae, whereas impregnation occurs in the submucosa. This is observed in patients with thickened and layered loops, thickened submucosa, and few parietal signals.
- In pattern IV, there is an absence of parietal impregnation. This is observed in patients with stenosis in the radiologically documented loop and with a slightly thickened, layered, and fibrotic loop with parietal signals.

A statistically significant correlation between pattern I and pattern II and clinical disease activity has been established [10] (Fig. 8.10a–c).

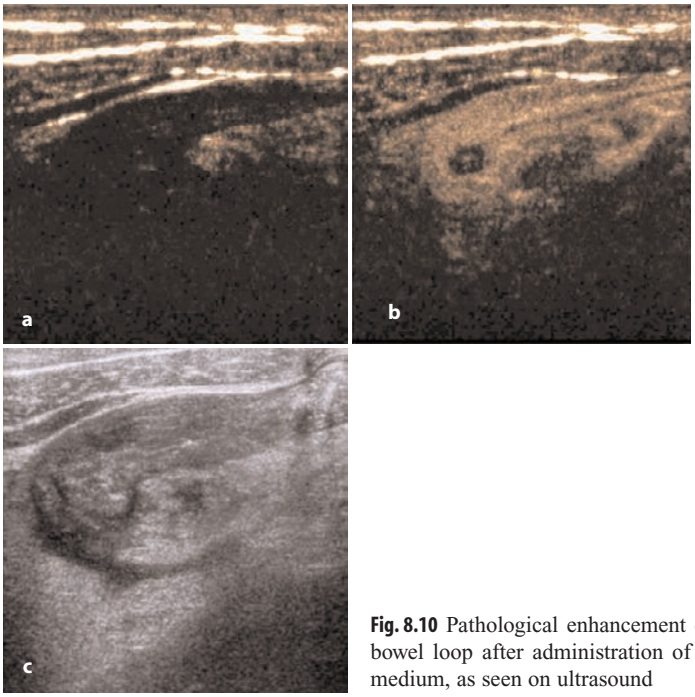

Fig. 8.10 Pathological enhancement of a thickened small bowel loop after administration of intravenous contrast medium, as seen on ultrasound

Computed Tomography

The success of CT in the diagnosis of CD or its complications relies on a number of procedures, such as a proper patient preparation, the use of multi-detector instruments (MDCT), the administration of intravenous or oral contrast agent, standardized technical acquisition parameters, and elaboration techniques for the post-processing of images through MPR, MIP, and VR reconstruction systems [15]. In CT scans done for screening purposes, patients must abstain from food for at least 6 h and laxatives are administered for proper intestinal cleansing. CT is also used in specific urgent/emergency cases for the evaluation of pre- and post-surgical complications.

Patients undergoing screening are administered an endoluminal contrast agent either orally (CT enterography) or through a tube inserted through the nose (CT enteroclysis) with the tip positioned at the duodenojejunal flexure of Treitz [16,17]. The contrast agent can be positive, for example 1500 ml of a 2% barium suspension or a suspension of hydrosoluble iodate contrast medium, or negative, for example, if water is used, a PEG or 5% methylcellulose diluted solution [16,17]. The administration of contrast agents in CT enterography is seriated and monitored to obtain complete and optimal lumen distension as far as the ileal valve. In CT enteroclysis, a nasojejunal tube is positioned, under radioscopic guidance, at the angle of Treitz. An injection pump programmed to calculate the quantity of contrast agent and its flow speed may also be used (1550–2000 ml at 100–250 ml/min) [16]. Both procedures require that the patient is administered a hypotonic drug prior to the scan and an

intravenous non-ionic iodate contrast agent during the examination. The acquisitions obtained in these two modalities substantially overlap and their technical parameters largely depend on the available equipment [16,17]. The scan must provide at least three acquisitions, one before and two following contrast agent infusion. Post-processing elaboration is based on the retro-reconstruction of images that are acquired in increasingly thinner slices (0.6–1.25 mm at equal or less frequent overlapping reconstruction intervals) to obtain the maximal number of images, thus producing a study on all three planes that includes an analysis of the vascular map (MIP) and a concise 3D representation of the intestinal loops (VR). Dedicated software provides curved or longitudinal representations and measures the exact size of the diseased area [16–18]. In specific urgent/emergency cases, similar technical acquisition parameters are used. An intravenous non-ionic iodate contrast agent is administered and routine post-processing procedures are followed. Oral contrast agent administration is not common.

MDCT is also used (CT colography) in patients with CD with colonic localization and provides an alternative to colonoscopy, which is often incomplete because of the stenotic complications of this disease [19–21]. The patient follows a low-fiber diet for 2–3 days prior to the scan and laxatives are taken the day before for proper intestinal cleansing. During the examination, a Foley catheter is inserted through the rectum and hypotonic drugs are administered. CO_2 or tolerable amounts of air, depending on the patient, are then introduced manually or automatically to inflate the colon. Thin-layered volume acquisition is done with the patient in prone decubitus, at low milliampere and kilovolt settings, and in supine decubitus after an intravenous contrast agent is administered [19–21]. Images are subsequently elaborated in 2D and 3D reconstructions using virtual colonoscopy and virtual dissection software (Fig. 8.11). This technique enables the identification of neoplastic or pseudo-inflammatory polypoid formations and is helpful in assessing the degree and progression of stenotic segments [20,21].

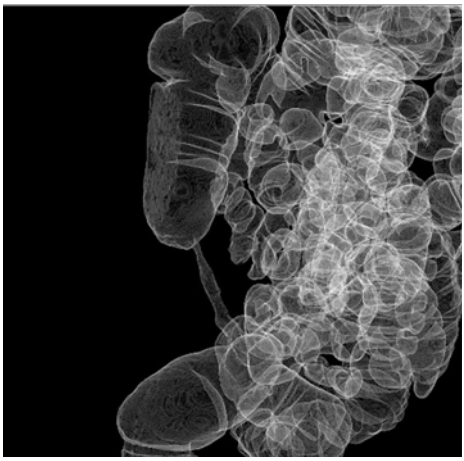

Fig. 8.11 Stenotic lesion evaluated by 3D virtual multi-detector CT (MDCT) colonoscopy

The main imaging criteria for the diagnosis of CD are parietal thickening and stratification, either a layered or targeted pattern with or without wall enhancement (Fig. 8.12), sinus tracts, edema and fibro-adipose proliferation of the mesenteric fat, occlusion of the vasa recta or the comb sign (Fig. 8.13), fibro-adipose infiltration of the submucosa, and mesenteric adenopathy [16–19,22]. Although several studies have shown that parietal thickening of 2.5–10 mm is a clear indication of the disease, values > 3 mm are regarded as being particularly meaningful in this context [18,22]. Parietal enhancement is considered the principal sign of transmural inflammation whereas low or no enhancement suggests a fibrotic evolution (Fig. 8.14). Superficial alterations of the mucosa, such as aphtoid lesions or lymphoid follicle hypertrophy, are not easily identifiable. Hence, CT is less helpful in evaluating the disease in its initial phase [11,22]. The sensitivity of CT depends on the disease phase but ranges between 70% in the initial phase and 98% in transmural and extramural localizations [15,17,18,22].

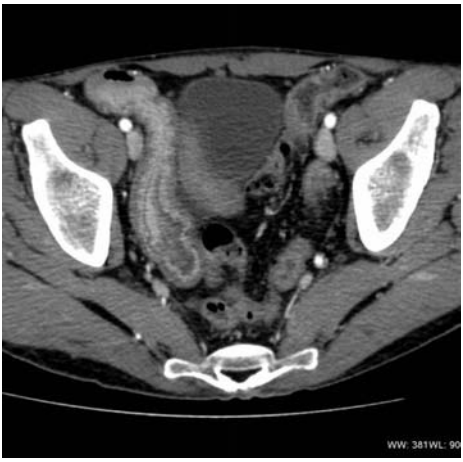

Fig. 8.12 MDCT axial data after the administration of i.v. contrast medium: target sign

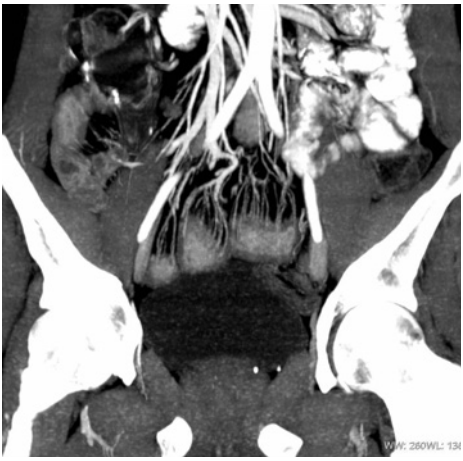

Fig. 8.13 MDCT coronal thin reformatted image after the administration of i.v. contrast medium: comb sign

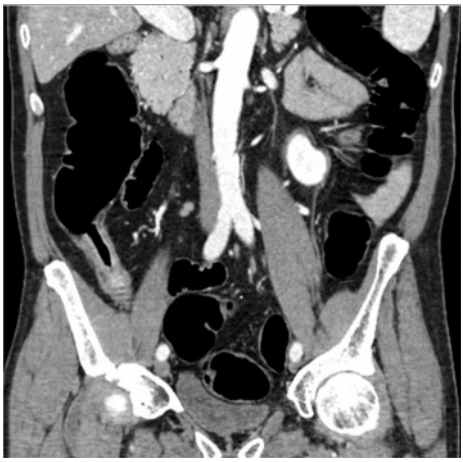

Fig. 8.14 MDCT coronal thin reformatted image after the administration of i.v. contrast medium: stenotic lesion without parietal contrast enhancement in the presence of fibrotic evolution

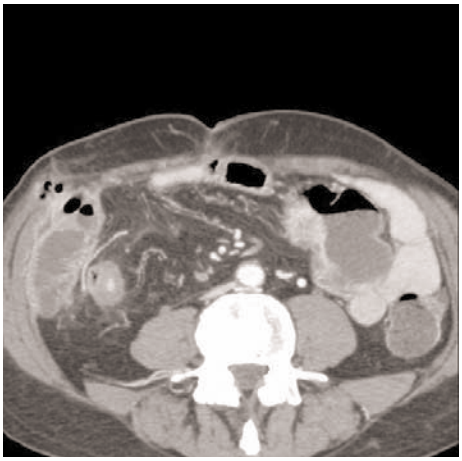

Fig. 8.15 MDCT axial data after the administration of i.v. contrast medium: abscess fluid collection

Among the most frequent complications of CD that are likely to arise, CT offers a panoramic visualization of fistulous tracts, abscesses (Fig. 8.15), and stenotic tracts [18]. Fistulous tracts (enteroenteric, enterocolic, enterovesicular or vaginal and enterocutaneous, duodeno-pancreatic, gastrocolic) may be either hyperdense, in the presence of positive endoluminal contrast agents, or hypodense, with reduced enhancement with or without the use of negative contrast agents [11,18,22]. Abscesses are easily identified, including their mesenteric and retroperitoneal extension, and can be subsequently monitored for their response to therapy.

MDCT evaluates a number of signals and parietal attenuation patterns that emerge in the post-contrast graphic phase and provides information on the disease activity index. Moreover, because of its spatial resolution, MDCT is particularly important in the diagnosis of disease activity and progression, as well as in the detection of extraintestinal complications and the determination of the best therapeutic approach [18,22].

This modality is therefore recommended because it is rapid, readily available, and less invasive than other techniques. However, it does have a few limitations, such as reduced sensitivity to superficial lesions compared to small bowel enema, exposure of the patient to a high dose of ionizing radiations, possible reactions to the iodate contrast agent, and a lower contrast resolution than obtained with MRI [11,15].

Magnetic Resonance Imaging

The advantage of MRI is that it is a non-invasive imaging technique that does not involve the use of ionizing radiation. It provides multi-planar images and thus information on many of the parameters needed to evaluate the parietal and extraparietal inflammation of CD as well any of the disease's complications [23,24]. Since the beginning of the 1990s, MRI has become an increasingly common technique in clinical practice due to technological progress and the introduction of phased array coils, fast and ultrafast gradient-echo and steady-state sequences, and the use of endocavitary and intravenous Gd-based paramagnetic contrast media. These advances have improved spatial and contrast resolution in images obtained during short periods of apnea, thereby addressing many of the limiting issues in the study of the abdomen, particularly those concerning respiratory movements, intestinal peristalsis, and the lack of adequate endoluminal contrast. Intestinal cleansing is necessary prior to the test and different procedures are prescribed to achieve a complete and optimal distension of the intestinal loops.

Contrast agents may be endoluminal or intravenous and include hypotonic drugs and the use of dedicated sequences. Positive, negative, and biphasic endoluminal contrast media are used in MRI to study the gastrointestinal tract [25–27]. Positive contrast media (gadolinium chelate, ferric ammonium citrate, manganese chloride) increase the signal intensity of intestinal structures in T1- and T2-weighted sequences. Negative contrast media such as superparamagnetic iron oxides result in low signal intensity at either frequency. Biphasic contrast media (water solutions of PEG, methyl-cellulose, and mannitol), which are currently favored, can act either as positive or negative agents depending on the sequence, thus generating a low-intensity signal in T1-weighted sequences and a high-intensity signal in T2-weighted sequences. As in MDCT or conventional radiology examinations, contrast agents can be administered in amounts of 1000–2500 ml via an infusion pump (with a speed of 100–250 ml/min), through a nasojejunal tube (MR enteroclysis), or orally (MR enterography) [28,29]. Intestinal transit and distension and possible stenosis with intestinal obstruction can be monitored during MR enteroclysis through MR fluoroscopy sequences [28,29]. Oral administration of the contrast agent is the most common and is undoubtedly better tolerated by patients. Recent studies have shown a similar diagnostic accuracy of MR enteroclysis vs. MR enterography when employed in the follow-up phase [30].

MRI investigations normally last 20–30 min and are carried out using instruments with 1.5T resonance and phased array coils. Most protocols include acquisitions in the axial and coronal planes, without intravenous contrast agent; atypical

T1/T2-weighted FIESTA or TRUE FISP sequences; and T2-weighted half-Fourier single-shot fast spin echo (ssFSE) or turbo spin echo (TSE) sequences, with and without suppression of the fat signal [31,32].

The first type of sequence, due to its speed of acquisition, provides a high contrast between the homogenously hyperintense lumen and the intestinal wall, without any movement artifact (Fig. 8.16). The only disadvantage is the black boundary artifact, which is caused by the chemical shift and is characterized by a thin hypointense external parietal line. This may prevent the visualization of a slight wall thickening, which is resolved only through sequences that suppress the fat signal. The second type of sequence, T2-weighted sequences, provides a high-contrast resolution between the hyperintense lumen and the intestinal wall and results in a better definition of parietal edema (Fig. 8.17) when fat is suppressed [28,29,32]. However, it is

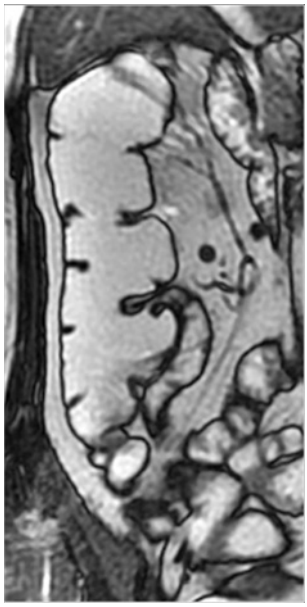

Fig. 8.16 Coronal T1/T2-weighted FIESTA sequence. Normal appearance of the ileocecal region

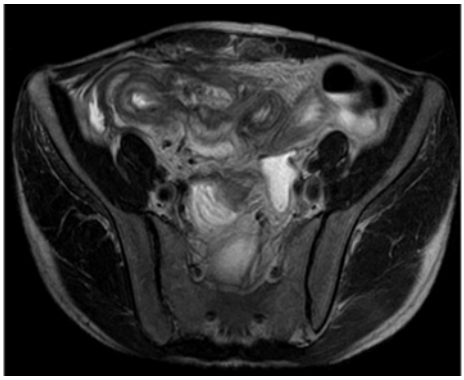

Fig. 8.17 Fat-saturated axial T2-weighted sequence: parietal intramural edema

very sensitive to endoluminal fluid flow, as these cause flow void, and hypotonic drugs must be prescribed prior to the examination. The third type of sequences, 3D fast spoiled gradient echo T1-weighted sequences with fat signal suppression [28,29,33], are produced before and during intravenous contrast agent administration (gadolinium chelate 0.1–0.2 mmol/kg intravenous followed by 10–20 ml of saline flush) in a multi-phasic technique (3 acquisitions at 30–70 and 120 s) and with the patient administered hypotonic drugs.

These sequences provide excellent visualization of the parietal enhancement and stratification generated by the contrast between the hypointense lumen signal and the surrounding mesenteric fat. In any type of sequence, coronal planes are critical to produce a panoramic evaluation of the intestinal loops. The multi-phasic technique may be relevant to identifying enhancement of the submucosa—a specific disease activity criterion–in arterial blood flow [34].

MRI scans also depict some of the superficial alterations of CD, such as the ulcers (although not the small aphthoid or linear ulcers), plical thickening, and flattening of the circular folds that mark the initial phase of the disease. However, it more accurately identifies the transmural and extramural alterations of CD [28,29,32] because of the reduced spacial resolution compared with that obtained following clysma of the small bowel.

Prassopoulos et al. [32] reported that blunting, flattening, thickening, distortion, and straightening of the valvulae conniventes and tiny aphthae were clearly shown on conventional enteroclysis but were not consistently depicted with MR enteroclysis. The latter procedure is as accurate as clysma of the small intestine in the depiction of parietal lesions. Deep ulcerative lesions are represented as lines of increased signal intensity moving transversally or longitudinally (Fig. 8.18a, b). In addition, a cobblestone pattern is seen.

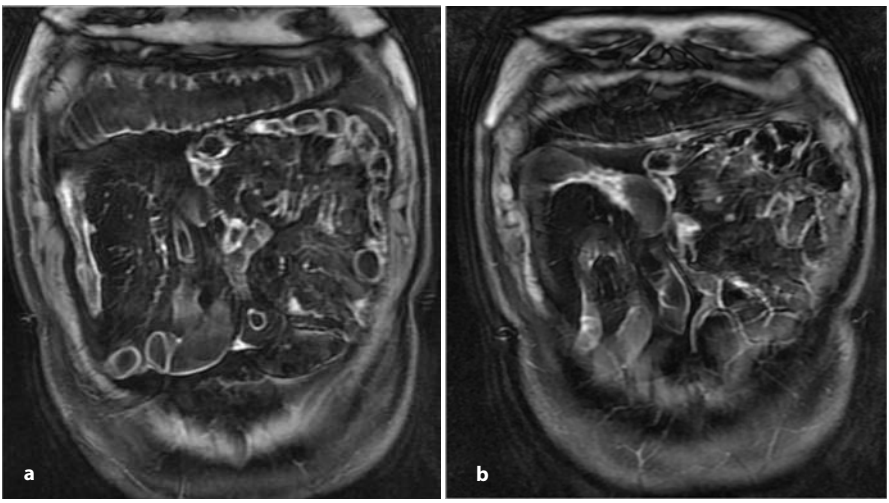

Fig. 8.18 Coronal 3D spoiled gradient-echo (SPGR) T1-weighted sequence after i.v. administration of a paramagnetic contrast agent: longitudinal and transversal ulcerative lesion findings

Inflammatory wall thickening, seen as the target pattern (Fig. 8.19), as well as stenosis and the extent of the diseased area can be accurately visualized, particularly through T1-weighted sequences and after an intravenous contrast agent is administered (Fig. 8.20). The main advantage over clysma of the small intestine is that extra-

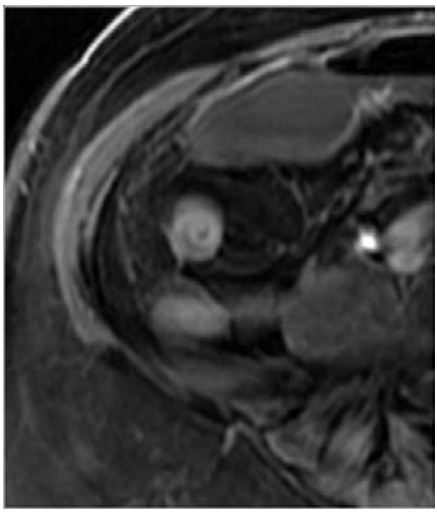

Fig. 8.19 Axial SPGR T1-weighted sequence after i.v. administration of a paramagnetic contrast agent: target sign

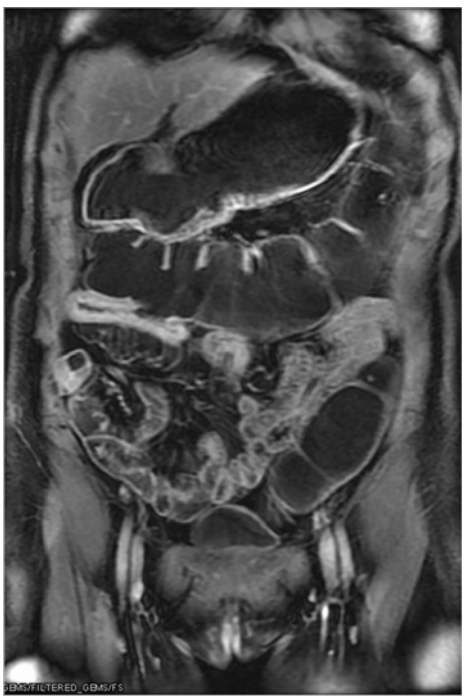

Fig. 8.20 Coronal SPGR T1-weighted sequence after i.v. administration of a paramagnetic contrast agent: optimal detection of the length of the stenotic lumen

parietal extent and disease complications, such as fibro-fatty proliferation (Fig. 8.21), mesenteric lymphadenopathies (Fig. 8.22), fistulous tracts (Fig. 8.23), phlegmons, and abscesses [23,28,29,32], can be identified.

Therefore, MRI can be regarded as an optimal procedure to correctly diagnose CD, based on the technique's multi-planar and multi-parametric features, higher contrast resolution compared to CT scans, and the absence of the need for ionizing radiation. The accuracy of MRI is > 85% [15,35]. Nonetheless, MRI has a few limitations. Its sensitivity to the initial alterations caused by the disease is relatively low because of the reduced spatial resolution. Also, MRI is expensive, time-consuming, and of limited availability, and it cannot be performed on claustrophobic patients or on individuals with metallic implants.

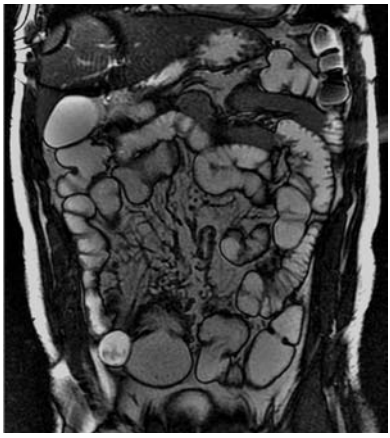

Fig. 8.21 Coronal T1/T2-weighted FIESTA sequence: fibro-fatty proliferation

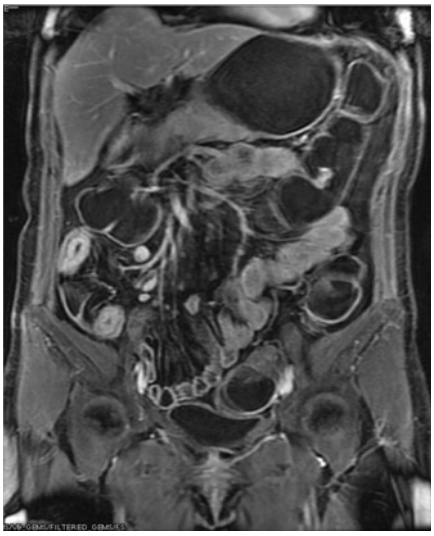

Fig. 8.22 Coronal SPGR T1-weighted sequence after i.v. administration of a paramagnetic contrast agent: mesenteric adenopathy, target sign, and comb sign

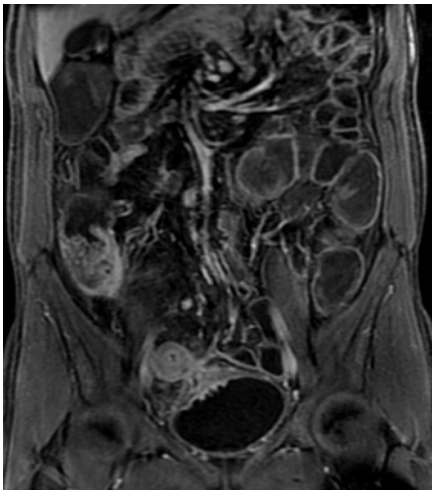

Fig. 8.23 Coronal SPGR T1-weighted sequence after i.v. administration of a paramagnetic contrast agent: enterovesicular fistula

References

1. Fanucci A (1994) L'imaging del canale alimentare: un approccio oroscopico. In: Cirillo R, Comino E (eds) Il tubo digerente. Atti del II Corso di aggiornamento in radiologia, Turin
2. Levine MS, Laufer I (1993) The upper gastrointestinal series at a crossroads. AJR Am J Roentgenol 161:1131–1137
3. Maglinte DDT, Herlinger H (1989) Single contrast and biphasic enteroclysis. In: Herlinger H, Maglinte DDT (eds) Clinical radiology of the small intestine. WB Saunders, Philadelphia, pp 107–119
4. Di Mizio R (2002) Morbo di Crohn del tenue. Atlante di Radiologia Verduci Editore, Rome
5. Maglinte DT, Chernish SM, Kelvin FM et al (1992) Crohn disease of the small intestine: accuracy and relevance of enteroclysis. Radiology 184:541–545
6. Dixon PM, Roulston ME, Nolan DJ (1993) The small bowel enema: a ten year review. Clin Radiol 47:46–48
7. Laufer J, Costopoulos L (1978) Early lesions in Crohn's diesease. AJR Am J Roentgenol 130:307–311
8. Fanucci A, Cerro P, Fraracci L, Ietto F (1984) Small bowel length measured by radiography. Gastrointest Radiol 9:349–351
9. Rossi S (2001) Ecografia addominale in Epato-Gastroenterologia. Testo Atlante Poletto Editore Torino
10. Rossi S, Callida F, Martegani A (2004) Mezzi di contrasto in ecografia. Testo Atlante. Poletto Editore, Turin
11. Horsthuis K, Stokkers P, Stoker J (2008) Detection of inflammatory bowel disease: diagnostic performance of cross-sectional imaging modalities. Abdom Imaging 33:407–416
12. Marc J, Van Outryve ET, Pelckmans PA et al (1991) Value of transrectal ultrasonography in Crohn's disease. Gastroenterol 101:117
13. Barros N, Cerri GG, Souza Rocha M, Goncalves MO (2000) Sonographic appearences of conglomerated polyps (giant polyposis) in patients with Crohn's disease. J Clin Ultrasound 28:199–205
14. Sarrazin J, Wilson SR (1996) Manifestations of Crohn disease at US. RadioGraphics 16:499–503

15. Saibeni S, Rondonotti E, Iozzelli A et al (2007) Imaging of the small bowel in Crohn's disease: a review of old and new techniques. World J Gastroenterol 24:3279–3287
16. Schmidt S, Felley C, Meuwly J et al (2006) CT enteroclysis: technique and clinical applications. Eur Radiol 16:648–660
17. Paulsen SR, Huprich JE, Fletcher JG et al (2006) CT enterography as a diagnostic tool in evaluating small bowel disorders: review of clinical experience with over 700 cases. RadioGraphics 26:641–666
18. Furukawa A, Saotome T, Yamasaki M et al (2004) Cross-sectional imaging in Crohn disease. RadioGraphics 24:689–702
19. Svensson MH, Svensson E, Lasson A, Hellström M (2002) Patient acceptance of CT colonography and conventional colonoscopy: prospective comparative study in patients with or suspected of having colorectal disease. Radiology 222:337–345
20. Silva AC, Vens EA, Hara AK et al (2006) Evaluation of benign and malignant rectal lesions with CT colonography and endoscopic correlation. RadioGraphics 26:1085–1099
21. Perry J, Pickhardt PJ (2004) Differential diagnosis of polypoid lesions seen at CT. RadioGraphics 24:1535–1556
22. Wittenberg J, Mukesh G, Harisinghani MG et al (2002) Algorithmic approach to CT diagnosis of the abnormal bowel wall. RadioGraphics 22:1093–1109
23. Umschaden HW,Szolar D, Gasser J et al (2000) Small bowel disease: comparison of MR enteroclysis images with conventional enteroclysis and surgical findings. Radiology 215:717–725
24. Laghi A, Passariello R (2003) Magnetic resonance in the study of the small bowel. Radiol Med (Torino) 106:1–15
25. Lauenstein TC, Schneemann H, Vogt FM et al (2003) Optimization of oral contrast agents for MR imaging of the small bowel. Radiology 228:279–283
26. Debatin JF, Patak MA (1999) MRI of the small and large bowel. Eur Radiol 9:1523–1534
27. Maccioni F, Viscido A, Marini M, Caprilli R (2002) MRI evaluation of Crohn's disease of the small and large bowel with the use of negative superparamagnetic oral contrast agents. Abdom Imaging 27:384–393
28. Umschaden HW, Gasser J (2003) MR enteroclysis. Radiol Clin N Am 231–248
29. Masselli G, Brizi MG, Menchini L et al (2005) Magnetic resonance enteroclysis imaging of Crohn's disease. Radiol Med 110:221–233
30. Schreyer AG, Geissler A, Albrich H et al (2004) Abdominal MRI after enteroclysis or with oral contrast inpatients with suspected or proven Crohn's disease. Clin Gastroenterol Hepatol 2:491–487
31. Lee JK, Marcos HB, Semelka RC (1998) MR imaging of the small bowel using the HASTE sequence. AJR Am J Roentgenol 170:1457–1463
32. Prassopoulos P, Papanikolaou N, Grammatikakis J et al (2001) MR enteroclysis imaging of Crohn's disease. Radiographics 21:S161-S172
33. Gourtsoyiannis N, Papanikolaou N, Grammatikakis J et al (2001) MR enteroclisys protocol optimization: comparison between 3D flash with fat saturation after intravenous gadolinium injection and true Fisp sequences. Eur Radiol 11:908–913
34. Koh DM, Miao Y, Chinn RJ et al (2001) MR imaging evaluation of the activity of Crohn's disease. AJR Am J Roentgenol 177:1325–1332
35. Rottgen R, Herzog H, Lopez Hanninen E et al (2005) Combination of dynamic MR enteroclysis (Sellink) and MR colonography in diagnose Crohn's disease. Rofo 177:1131–1138

Diagnosis of Anorectal Disease

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One-third of patients affected by chronic inflammatory bowel diseases (IBDs) also suffer from fistulas of different sizes and types. The majority of fistulous tracts are identified in the anal rectal area, with chronic inflammation, and in the surrounding perivisceral adipose tissues [1]. Diagnostic imaging aims to identify and depict the complex fistulous tracts that can appear at the top of the anal canal as well as the potential involvement of the anal sphincter and the extension of the fistula to the ischioanal fossa.

Over the years, a number of investigative techniques have been explored with different results. Fistulography requires cannulation of the external fistulous opening [2] and the administration of a radio-opaque contrast agent. The high spatial resolution of this modality fails to compensate for its inability to visualize soft tissues or to show the relationship between the fistulous tract and the sphincter apparatus. Tracts in which the external fistulous opening has been cannulated as well as the relevant active communications can be shown whereas hidden tracts and non-communicating abscesses are not visualized.

Ultrasound diagnostic techniques are currently still in use in a large number of clinics [3]. The examination can be carried out through the endocavitary route, using dedicated tubes, or via the perineal route. It is generally acknowledged that the widespread availability of ultrasound and its relatively low cost do not compensate for the difficult visualization of the external sphincter and the perivisceral areas [4]. As in other endocavitary methods, this is particularly complex for patients with acute perineal pain caused by active inflammation or patients in the post-operative phase. Neither the type of examination, which is operator-dependent, nor the information produced during the investigation is easily interpreted during the pre-operative phase.

Good representation of the pelvic pathology frequently associated with the disease is obtained with CT scans, although the poor contrast resolution of this technique makes it difficult to identify the sphincter organ [5].

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The most common method currently used to study perianal pathologies is MRI [6]. It addresses the bottlenecks presented by other modalities because of its high contrast resolution, which provides visualization of the soft tissues. The rectoanal muscular structures are therefore easily depicted, as is their relationship with the complex fistulous tracts typical of chronic inflammatory diseases. MRI with endocavitary coils was carried out in the past. The coils used for rectoanal studies were of a smaller caliber than those used for prostate examinations, and they worked in association with an external detection system [7]. These techniques, while common during the 1990s, have been almost completely abandoned today because of the high sensitivity of the external multichannel coils that are currently used. The earlier techniques are nowadays limited to dubious cases (rectovaginal fistulas) in patients able to tolerate the insertion of a rectal tube [8].

MRI provides information on a large number of parameters. Most image acquisitions of the pelvic plane are based on the high signal intensity detected in areas affected by the inflammatory disease. A fistulous tract comprises a canal filled with serous fluid and pus and often surrounded by a variably extended area of granulation tissue. This is associated with a large water content, resulting in an increase in resonant protons and thus of signal intensity [9].

Some acquisitions, namely, morphological ones, are highly detailed and thus well-depict both the anatomy and the associated pathology, facilitating disease diagnosis. These images are obtained through fat suppression techniques in which the signal from adipose tissue is suppressed, yielding high sensitivity in the representation of pelvic pathologies [10].

Images acquired before and after an intravenous contrast agent is administered are based on a different principle. Here, signal enhancement is produced by the edema and, ultimately, by the increased vascularization, which in turn reflects the degree of inflammation. In practice, intravenous contrast agent is hardly ever used to depict chronic inflammation, although it may be administered to aid in the diagnosis of complex localized pelvic abscesses and rectoanal cancerous lesions [11]. Similarly, endoanal insertion of contrast agent through cannulation of the external fistulous opening does not have any concrete application in MRI because of the high sensitivity of the superficial coils.

Many protocols, used in numerous studies, have been tested and optimized according to the type of patient in order to represent the pelvic area and to diagnose fistulous tracts. The anatomy and pathology of the concerned area are represented in 3D according to different weightings [12]. A basic examination that addresses most diagnostic issues normally lasts 30 min, including patient preparation and accommodation in the MRI room. Due to the strong magnetic fields produced by MR devices, this technique cannot be used to examine patients with ferromagnetic metallic prostheses (especially patients who underwent surgery before 1995), unknown external metallic components, pacemakers, and similar intracardiac devices.

Although clinical reality is undoubtedly more complex, it is possible to analyze the coronal and sagittal images produced by MRI according to Park's classification, whereas the St. James's University Hospital classification is used to assess disease activity index (Table 9.1) [13].

Table 9.1 Classification of perianal fistulas (St. James Hospital)

Grade 0	Normal
Grade 1	Intersphincteric fistula
Grade 2	Intersphincteric fistula with abscess
Grade 3	Trans-sphincteric fistula
Grade 4	Trans-sphinteric fistula with abscess
Grade 5	Extrasphinteric fistula

MRI in longitudinal planes is consistent with Park's classification [4], as it visualizes the internal anal sphincter (involuntary muscle), direct continuation of the circular smooth muscle of the rectum, and external anal sphincter, whose muscular extensions are attached to the puborectalis muscle and levator ani muscle (voluntary muscle). An ischioanal fossa with homogeneous adipose content appears external to the muscular plane [14]. In Park's article, published in 1976, some 400 patients were evaluated. It is likely that superficial fistulas were not included in the classification because of the complex type of patients treated at St. Mark's Hospital in London. The transversal axial plane is used as reference to identify the fistulous internal opening according to the hands of the clock (12 and 6 positions). Information produced by MRI often confirms Goodsall's rule: the anterior or posterior external opening of an anal fistula relates to tracts located anterior or posterior to the transverse line drawn across the anal verge.

According to the St. James's University Hospital classification (Table 9.1), a grade 1 pathology is a simple fistulous tract that forms longitudinally within the intersphincteric planes (Fig. 9.1), whereas abscesses are also present in grade 2 pathologies (Fig. 9.2). Neither one is associated with disorders in the ischioanal fossa (Fig. 9.3). The passage of fistulous tracts through the intersphincteric planes and the presence of alterations in the ischioanal fossa are seen in grade 3 pathology (Fig. 9.4), and localized abscesses in grade 4 (Figs. 9.5, 9.6). In grade 5, fistulous tracts originate from above the sphincter (Fig. 9.7). However, there are also extremely complex alterations involving different associated fistulous tracts and lacunae formed by communicating abscesses (Fig. 9.8). In post-operative examination, it is often possible to recognize the seton, depicted as a low-intensity signal line (Fig. 9.9). Subsequent examinations of such patients are often carried out to evaluate the relationship between the seton and the sphincter apparatus and to assess the efficiency of clinical therapy of the diseased areas.

MRI is therefore the preferred diagnostic imaging modality for evaluating perianal disease, as it is non-invasive, well-tolerated, and can ultimately be used to examine patients with chronic inflammatory diseases.

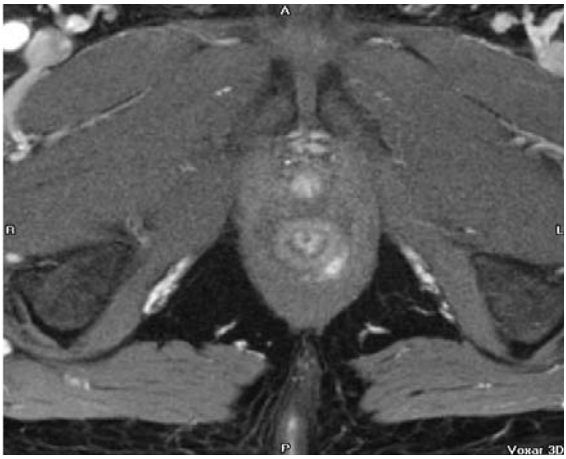

Fig. 9.1 Axial T1-weighted image with fat saturation. Grade 1 intersphinteric fistula

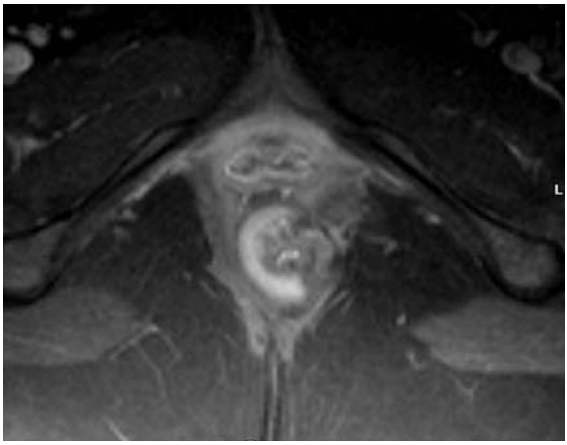

Fig. 9.2 Axial T2-weighted image with fat saturation. Grade 2 horseshoe perianal fistula

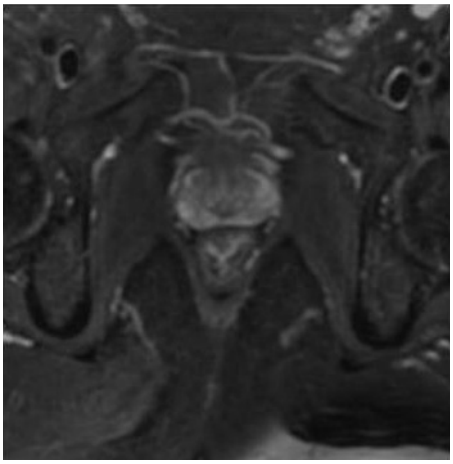

Fig. 9.3 Axial T2-weighted image with fat saturation. Intersphincteric fistula with normal appearance of the ischioanal space

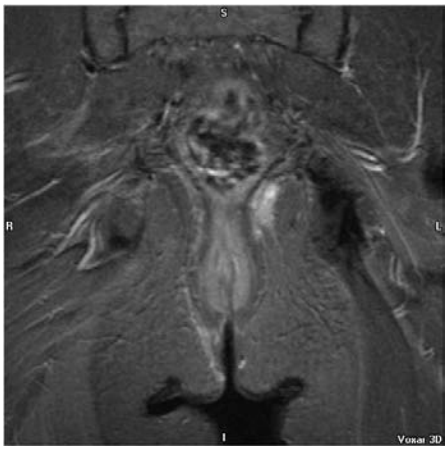

Fig. 9.4 Coronal T2-weighted image with fat saturation. Trans-sphincteric fistula with inflammatory change in the left ischiorectal fossa

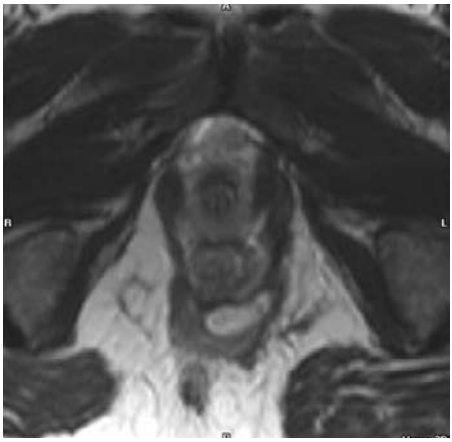

Fig. 9.5 Axial T1-weighted image. Perianal fistula with an ischiorectal fossal abscess

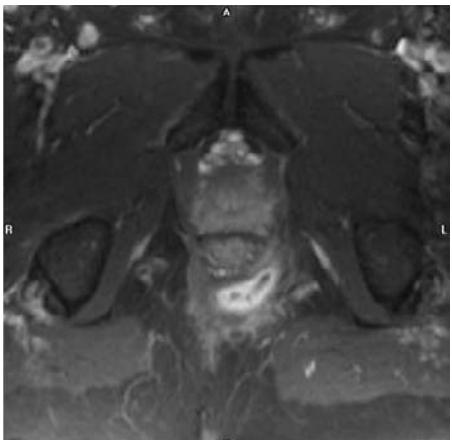

Fig. 9.6 Axial T2-weighted image with fat saturation. Grade 4 perianal fistula with an ischiorectal fossa abscess

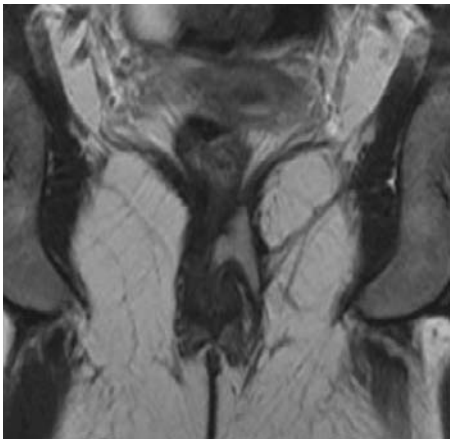

Fig. 9.7 Coronal T2-weighted image with fat saturation. Grade 5 trans-elevator fistula traversing the left ischirectal fossa

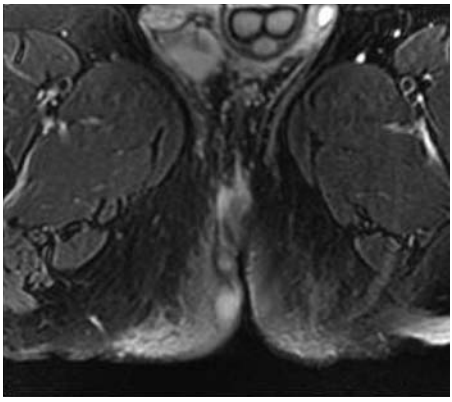

Fig. 9.8 Axial T2-weighted image with fat saturation. Complex fistula with abscesses in the ischioanal fossa

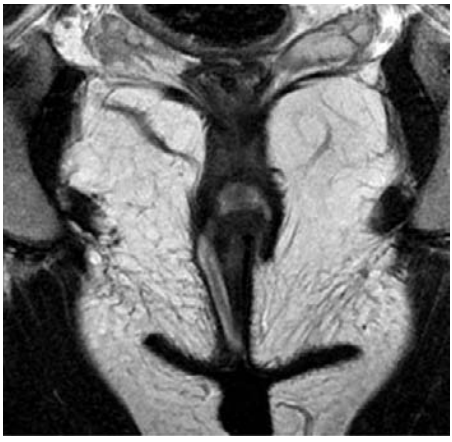

Fig. 9.9 Coronal T2-weighted image with fat saturation. Trans-sphincteric fistula after surgery (seton placement)

References

1. Schwartz DA, Loftus EV Jr, Romaine WJ et al (2002) The natural history of fistulizing Crohn's disease in Olmsted County, Minnesota. Gastroenterology 122(4):875–880
2. Kuijpers HC, Schulpen T (1985) Fistulography for fistula-in-ano: is it useful? Dis Colon Rectum 28:103–104
3. Choen S, Burnett S, Bartram CI, Nicholls RJ (1991) Comparison between anal endosonography and digital examination in the evaluation of anal fistulae. Br J Surg 78:445–447
4. Morris J, Spencer JA, Ambrose NS (2000) MR imaging classification of perianal fistulas and its implications for patient management. RadioGraphics 20:623–635
5. Yousem DM, Fishman EK, Jones B (1988) Crohn disease: perirectal and perianal findings at CT. Radiology 167:331–334
6. Spencer JA, Ward J, Beckingham IJ et al (1996) Dynamic contrast-enhanced MR imaging of perianal fistulas. AJR Am J Roentgenol 167:735–741
7. deSouza NM, Gilderdale DJ, Coutts GA et al (1998) MRI of fistula-in-ano: a comparison of endoanal coil with external phased array coil techniques. J Comput Assist Tomogr 22:357–363
8. Stoker J, Rociu E, Schouten WR, Lameris JS (2002) Anovaginal and rectovaginal fistulas: endoluminal sonography versus endoluminal MR imaging. AJR Am J Roentgenol 178:737–741
9. Maier AG, Funovics MA, Kreuzer SH et al (2001) Evaluation of perianal sepsis: comparison of anal endosonography and magnetic resonance imaging. J Magn Reson Imaging14:254–260
10. Halligan S, Bartram CI (1998) MR imaging of fistula in ano: are endoanal coils the gold standard? AJR Am J Roentgenol171:407–412
11. Spencer JA, Ward J, Beckingham IJ et al (1996) Dynamic contrast-enhanced MR imaging of perianal fistulas. AJR Am J Roentgenol 167(3):735–741
12. Stoker J, Jong Tjien Fa VE, Eijkemans MJ et al (1998) Endoanal MR imaging of perianal fistulas: the optimal imaging planes. Eur Radiol 8:1212–1216
13. Parks AG, Gordon PH, Hardcastle JD (1976) A classification of fistula-in-ano. Br J Surg 63:1–12
14. Hallingan S, Stoker J (2006) Imaging of fistula in ano. Radiology 239:18–33

Role of Interventional Radiology

10

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Introduction

Crohn's disease (CD) is a chronic condition in which patients often require multiple surgical procedures during their lifetime. Specifically, approximately 70–90% of patients with CD require surgery, and 33–82% undergo a second operation during the course of the disease [1]. Up to 33% may require more than two bowel resections. The results of percutaneous abscess drainage (PAD) must be viewed within this context, with the goal of minimizing the invasiveness of interventions equally as important as minimizing the number of interventions. Abdominal or pelvic abscesses occur in 10–28% of patients with CD during their lifetimes [2-6]. The mechanisms of abscess formation in these patients include peritoneal contamination at the time of previous surgery, remote hematologic seeding from diseased bowel, and direct extension from involved bowel. The latter is the most common mechanism, reflecting the transmural nature of the disease process, with deep, fissuring ulceration that may result in perforation of the bowel wall.

Treatment of abscesses, which traditionally has required clinical diagnosis and surgical intervention, has been revolutionized by high-resolution radiologic techniques such as computed tomography (CT) scanning (Fig. 10.1), magnetic resonance imaging (MRI), and ultrasound (US). These studies permit precise anatomic localization of abscess cavities and allow for non-surgical drainage of the infected cavity. Liver abscess is a rare complication of CD [7-8], probably due to Crohn's colitis, since there have been no documented cases of liver abscess associated with ulcerative colitis. The incidence of liver abscess in patients with CD is about 10–15 times that found in the general population [9]. Diagnosis of the abscess is often delayed because its clinical presentation can be similar to an exacerbation of CD or may pre-

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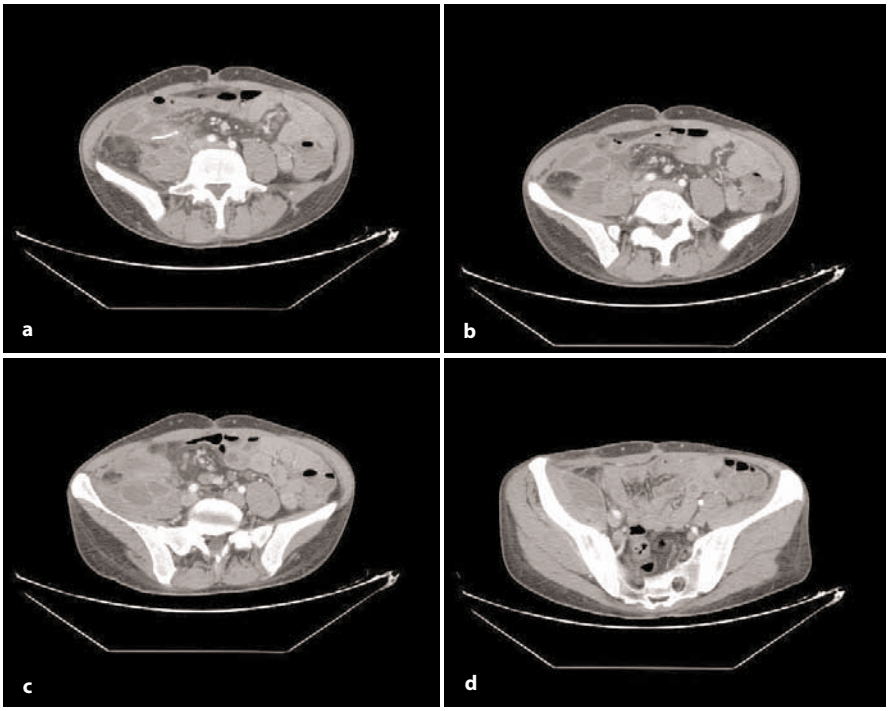

Fig. 10.1 Patient (A). Patient with CD, undergone surgery for ileo-colitis. Transverse CT images obtained at the level of ileo-psoas muscle, shows deep pelvic abscess

cede diagnosis of the disease. Liver abscesses usually appear as late sequelae of CD.

PAD may represent a safe and sometimes definitive therapy for abdominal and pelvic abscesses in the absence of indications for immediate surgery. The choice of PAD as an initial drainage modality takes into account patient age, immunomodulator use, and admission to a medical service. In the setting of CD complications, many variables must be considered: origin, location, and size of the abscess; volume of the initial aspirate; organisms identified in fluid culture; duration of catheter drainage; incidence of catheter-related pain and procedure-related complications; short- and long-term outcomes.

Drainage and Catheter Management

Abscess drainage is performed by using standard imaging-guided techniques [10,11]. Catheters can be placed using either the trocar technique tandem to a guiding needle or the Seldinger technique, at the discretion of the attending radiologist. Catheters are placed with US or CT guidance or with fluoroscopic guidance for wire manipulation following initial needle placement under CT or US guidance. Catheter size

ranges from 8 to 16 F; size and number of catheters are determined by the attending interventional radiologist, who performs the abscess drainage on the basis of the nature of the fluid obtained at needle aspiration and the extent of the abscess. A highly skilled team of interventional radiologists is crucial to affording patients the benefits of PAD. Initial abscess drainage is done under CT or sonographic guidance, with tandem placement of an 18-gauge sheath needle after location of the abscess cavity with a 22-gauge needle [12]. Subsequent guidewire placement, tract dilatation, and catheter positioning are done under fluoroscopic guidance (Fig. 10.2). After initial abscess drainage, if communication to the gastrointestinal tract is suspected by the nature of the drainage, or by persistently elevated catheter drainages, the abscessogram is scrutinized for signs of enteric communication, such as a pointed or angular contrast collection. If present, the collection is probed with angiographic catheters in an attempt to document the location and size of the communication. Attempts to document communication are routinely repeated after drainage if the communication is not immediately apparent (Fig. 10.3). Once found, the drainage catheter is repositioned adjacent to the entry site of the communication; or a second catheter is placed for this purpose, with the original catheter retained for continued abscess drainage. Catheter manipulation, if necessary, is considered part of the man-

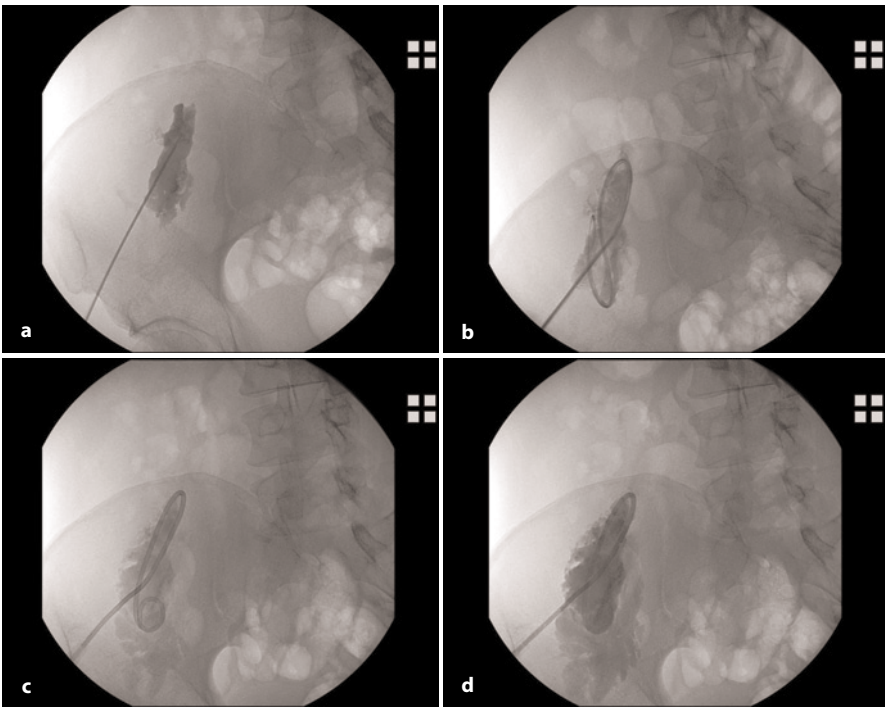

Fig. 10.2 Patient (A). Percutaneous abscess drainage (PAD) is placed in deep pelvic abscess with Seldinger Tecnique under ultrasonografic and Fluoroscopic guidance. After initial abscess drainage, abscessogram was performed to evaluate enteric comunication and no signs were found

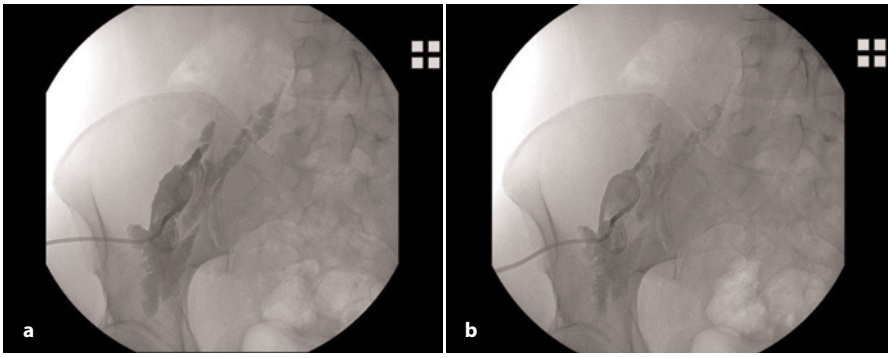

Fig. 10.3 Patient (A). **a** Abscessogram performed 7 days after PAD placement. **b** Abscessogram performed 20 days after PAD placement. Abscess appear decreased in size and no potential enteric comunication were found

agement of drainage catheters and not a complication or failure. Catheters are removed when drainage is diminished to < 10–20 ml/day. In patients with persistently high outputs (> 50 ml/day), injection of contrast material through a catheter, contrast-material-enhanced study of the small bowel, or enema examination with contrast material is performed to assess the presence and location of a fistula to the bowel.

Percutaneous Abdominal Abscesses Drainage

Patients undergoing percutaneous abdominal abscess drainage must be categorized on the basis of the nature of the underlying abscess. Spontaneous abscesses are those that arise in the setting of CD in patients who do not have a history of surgery in the preceding 30 days. Postoperative abscesses are those diagnosed within 30 days of CD-related surgery. Patients must be also evaluated for the location, size, and number of abscesses at presentation for initial drainage. Furthermore, abscesses can be categorized as initial versus recurrent. A recurrent abscess is defined as an abscess that occurs in the same location as a previously drained abscess after resolution of the initial abscess. In the setting of abscesses management, short- and long-term successes must be considered. Both are defined with respect to the need for surgery for active CD or complications of the disease following percutaneous abdominal abscess drainage. A short-term success is defined as a patient who does not undergo CD-related surgery within 60 days of PAD, and a short-term failure as a patient who requires CD-related surgery within 60 days. A long-term success is defined as a patient who does not require CD-related surgery beyond 60 days after PAD, and a long-term failure as a patient who undergoes surgery beyond 60 days after PAD. With respect to spontaneous versus postoperative abscesses, Sahai et al. [13] observed a trend in which postoperative abscesses were more likely to be short-term successes

than spontaneous abscesses. This trend toward better outcomes with postoperative abscesses may be due to the fact that the diseased segment is left in situ in the setting of PAD for spontaneous abscesses. Therefore, unless medical treatment substantially mitigates disease activity, the conditions that led to abscess formation remain, and the abscess can easily recur. In postoperative abscesses, the diseased segment or segments of bowel have presumably been removed. An abscess may form from a localized intraoperative contaminated site or anastomotic leak. Without persistent bowel inflammation or persistent leak, this abscess would be expected to resolve with PAD and the administration of antibiotics. Thus, success rates of percutaneous drainage are higher for postoperative abscesses than for spontaneous abscesses. Patients with recurrent and additional abscesses represent a sizable minority of patients; recurrent abscesses appear to be equally likely to undergo successful short-term drainage as initial abscesses. Accordingly, we are confident that there is no reason to approach a recurrent abscess differently from an initial abscess. New abscesses in different locations reflected noncontained and/or multifocal perforation, which may be difficult to treat percutaneously. However, abscesses that developed beyond 60 days, likely reflecting recurrent disease in a different location, do not appear to seriously limit the success of their treatment by PAD. After initial abscess drainage, patients with abscess-intestinal communication can be administered TPN (Total Parenteral Nutrition), which is important for healing and closure of the communicating tract and plays an ancillary role in percutaneous abscess management by decreasing intestinal luminal contents and peristalsis as well as intestinal flora. Indeed, temporary closure of the intestinal-abscess communication can be effected by TPN therapy alone [14,15]; however, long-term follow-up revealed recurrence of the abscesses in the same location once oral feedings were reinstituted [16]. This observation shows that the closure is transient, with no true healing of the bowel wall. Moreover, it underscores the need for long-term follow-up in the percutaneous management of abscesses with intestinal communication in CD before they can be considered "cured." It also suggests that TPN can have a considerable palliative effect that is independent of percutaneous abscess management. The technical success rate of PAD is important both for patients who undergo surgery and for those who do not undergo surgery in the short term. In patients who undergo subsequent surgery, PAD results in complete drainage of the abscess, and thus in a cleaner surgical field and possibly in a less complicated procedure and postoperative course. Casola et al. [17] advocated this approach of PAD followed by later bowel resection as opposed to the two-stage surgical approach, which surgeons sometimes use for patients who are too ill to undergo immediate definitive surgery. In this approach, surgical abscess drainage is performed first, followed by resection of the diseased bowel at surgery later. Indeed, PAD is less invasive than surgical drainage and has a lower rate of enterocutaneous fistula. It therefore provides an approach to abscess treatment that is associated with lower morbidity, even for patients who ultimately undergo bowel resection. Furthermore, patients who avoided surgery in the short term were found to be not significantly more likely to require surgery in the long term. This suggests that PAD is not simply helping to defer surgery, as a temporizing measure, but possibly helps to avoid surgery altogether. In our experience, the only major complication of

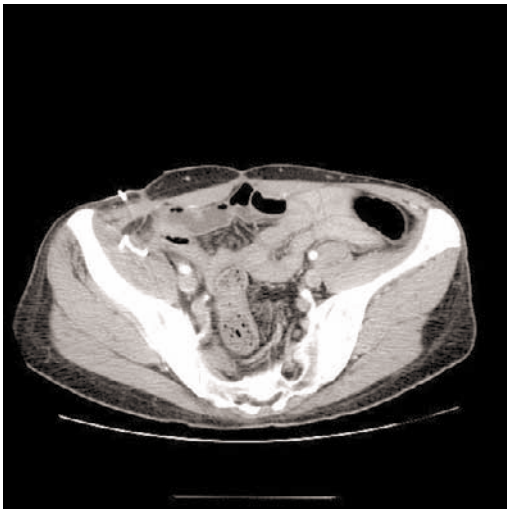

Fig. 10.4 Patient (A). Follow-up: transverse CT images performed after 2 months from the first CT shows decrease in size of the deep ileo-psoas abscess wich was completely drained out. PAD was removed after 2 days

abscess drainage is a single enterocutaneous fistula at the site of catheter placement, which occurred in 2% of the procedures; this rate is acceptably low and compares very favorably with the 21–85% rate reported following surgical abscess drainage [18]. Thus, given the benefits of PAD and its minimally invasive nature, this rate is acceptable and should not limit PAD in the setting of CD. In conclusion, in patients with CD and abdominal abscesses, catheter placement by interventional radiologists is successful in almost all cases in which a safe percutaneous path exists, and technical adequacy of abscess drainage is achieved in 96% of these patients (Fig. 10.4). For some patients, surgery can be avoided both in the short and long term. Patients with recurrent abscesses can be offered PAD, because they are not universally fated for surgery. The rate of surgery following PAD will likely vary according to local practice patterns, but patients can expect an approximately 50% chance of undergoing surgery in the short term following PAD, and success rates of percutaneous drainage are higher for post-operative abscesses than for spontaneous abscesses.

Percutaneous Pelvic Abscess Drainage

Percutaneous drainage is standard therapy for pelvic abscesses in the absence of indications for immediate surgery [19]. Abscesses deep in the pelvis pose a unique problem because numerous intervening structures create obstacles to safe percutaneous access. These include the pelvic bones, bowel, bladder, iliac vessels, and female reproductive organs. Among the techniques designed to overcome these obstacles to percutaneous drainage are the transgluteal approach through the greater sciatic foramen, the transvaginal approach, and the transrectal approach [20-23]. For drainage of pelvic collections of fluid near the vagina and rectum, excellent results have been

reported with the transvaginal and transrectal approaches, respectively. However, the transgluteal approach has advantages over these others. First, the transvaginal approach cannot be employed to drain presacral abscesses effectively and is obviously impossible in men. If long-term catheter drainage is indicated, catheter fixation is achieved much more easily with a transgluteal approach. Use of the transgluteal approach under CT guidance was first reported in 1986 by Butch et al [24]. However, it also has draw-backs, because patients must lie prone on the transgluteally placed catheter for proper gravity drainage, which may cause discomfort as well as catheter kinking. All transgluteal drainage procedures are performed with CT guidance and with intravenous conscious sedation of the patient to achieve adequate pain control for the procedure. Patients who tolerate the prone position can be placed prone, whereas those with recent surgical incision or respiratory distress must be placed in a prone oblique or lateral decubitus position. The administration of intravenous antibiotics to all patients immediately prior to the procedure is recommended. Self-retaining locking pigtail catheters with distal hydrophilic tips are used; these catheters range in size from 8 to 14 F and are placed using either a tandem trocar or Seldinger technique. The tandem trocar technique involves initial CT-guided placement of an 18–22-gauge spinal needle into the abscess. This needle provides an externally visible guide, enabling catheter insertion at the correct angle, perfectly parallel to the needle. The Seldinger technique involves the insertion of an 18-gauge sheathed needle into the abscess. The sheath allows the needle to be exchanged for a 0.035-inch Amplatz guidewire over which dilators are inserted and serial dilation is performed. The catheter is then inserted over the wire to a depth determined on the basis of the initial transverse CT scan. The preferred approach for transgluteal access is the infra piriformis approach, in which the catheter is inserted as close to the sacrum as possible, at the level of the sacrospinous ligament, below the piriformis muscle. At this level, the sciatic nerve is situated laterally and can be avoided easily. The location of the abscess or of the bowel, however, sometimes requires a transpiriformis approach with insertion of the catheter through the piriformis muscle. Pain limited to the site of needle or catheter insertion is common and is usually related to muscle transgression. If pain is severe, the patient often tenses the involved muscle (usually the gluteal muscle or the piriformis muscle). In turn, this muscular tension makes catheter placement more difficult, both in terms of traversing the muscles and in accurately directing the catheter to the target abscess. This muscle-localized pain is easily controlled, however, with intravenous analgesics administered prior to catheter insertion. Immediately after catheter placement, a syringe is inserted and the fluid is aspirated until flow ceases; after aspiration, the catheter is left in place to allow gravity drainage. Post-drainage images of the area of interest must be obtained to verify the adequacy of drainage. To maintain catheter patency, the catheters are flushed every 8 h with 5–20 ml of 0.9% saline solution. The success of percutaneous drainage is assessed postprocedurally with radiologic imaging and is verified by observation of improvement in the patient's general clinical condition, as indicated by defervescence, diminishing catheter output, and diminishing leukocytosis. The major complication of percutaneous pelvic abscesses drainage is hemorrhage, espe-

cially when the transpiriformis approach is performed [25]. Usually, hemorrhage occurs through the catheter tract after catheter removal. In this case arteriography must be done to visualize the source of bleeding; pseudoaneurysm of the inferior gluteal artery is the most common finding. After visualization, the pseudoaneurysms can be embolized with coils and a gelatin sponge. If arteriography shows no arterial injury, spontaneous resolution of the hematoma with conservative management is expected.

Percutaneous Liver Abscess Drainage

Liver abscess is a rare complication of CD [26]. The incidence is reported to be 114–297 per 100,000 cases of CD vs. 8–16 per 100,000 in the general population [27]. All of the case series and case reports support a male predominance, with a male: female ratio of 23:8 [28-32]; this is in contrast to the equal male-to-female ratio found in CD in general. A predisposing factor seems to be an inflammatory or perforating disease as opposed to fibrostenotic disease or the presence of fistulous disease and intra-abdominal abscesses. Several mechanisms have been proposed regarding the development of a liver abscess: direct extension of an intra-abdominal abscess into the liver via the portal vein from intra-abdominal infection, indirectly from complications of CD (i.e., biliary disease), and from sepsis occurring in a carcinoma that had metastasized to the liver. Because of the infrequency of this serious complication in clinical practice, controversy still exists as to the appropriate treatment modality. Attempts have been made to treat liver abscesses with antibiotics alone [33], but the results have been dismal. Percutaneous liver abscess drainage may be performed, usually with insertion of a pigtail catheter. A consensus as to type and duration of antibiotics has not been reached, and the duration of intravenous antibiotics ranges from 2 to 6 weeks. Patients have also been administered oral antibiotics. Regarding percutaneous vs. surgical drainage, many patients can be managed satisfactorily with percutaneous drainage and antibiotics. In those patients in whom percutaneous drainage was unsuccessful (i.e., multiple abscesses that were not connected) surgery is necessary, especially if there is additional intra-abdominal pathology that must also be addressed. Under these circumstances, the majority of authors advocate surgical drainage.

References

1. Krupnick AS, Morris JB (2000) The long-term results of resection and multiple resections in Crohn's disease. Semin Gastrointest Dis 11:41-51
2. Nagler SM, Poticha SM (1979) Intraabdominal abscess in regional enteritis. Am J Surg 137:350-354
3. Steinberg DM, Cooke WT, Alexander-Williams J (1973) Abscess and fistulae in Crohn's disease. Gut 14:865-869

4. Edwards H (1969) Crohn's disease. An inquiry into its nature and consequences. Ann R Coll Surg Engl 44:121-139
5. Greenstein AJ, Kark AE, Dreiling DA (1975)Crohn's disease of the colon. III. Toxic dilatation of the colon in Crohn's colitis. Am J Gastroenterol 63:117-128
6. Keighley MR, Eastwood D, Ambrose NS et al (1982) Incidence and microbiology of abdominal and pelvic abscess in Crohn's disease. Gastroenterology 83:1271-1275
7. Mir-Madjlessi SH, McHenry MC, Farmer RG (1986) Liver abscess in Crohn's disease. Report of four cases and review of the literature. Gastroenterology 91:987-993
8. Valero V, Senior J, Wantanakunakorn C (1985) Liver abscess complicating Crohn's disease presenting as thoracic empyema. Am J Med 79:659-662
9. Greenstein AJ, Sachar DB, Lowenthal D et al (1985) Pyogenic liver abscess in Crohn's disease. Quart J Med 56:505-518
10. vanSonnenberg E, Mueller PR, Ferrucci JT Jr (1984) Percutaneous drainage of 250 abdominal abscesses and fluid collections. Part I: Results, failures, and complications. Radiology 151:337-341
11. Mueller PR, vanSonnenberg E, Ferrucci JT Jr (1984) Percutaneous drainage of 250 abdominal abscesses and fluid collections. Part II: Current procedural concepts. Radiology 151:343-347
12. Haaga JR. Weinstein AJ (1980) CT-guided percutaneous aspiration and drainage of abscess. AJR Am J Roentgenol 135:1187-1194
13. Sahai A, Bèlair M, Gianfelice D et al (1997) Percutaneous drainage of intra-abdominal abscesses in Crohn's disease: short and long-term outcome. Am J Gastroenterol 92:275-278
14. Mùller JM, Keller HW, Erasmi H, Pichlmaier H (1983) Total parenteral nutrition as the sole therapy in Crohns disease: a prospective study. Br J Surg 70:40-43
15. Fischer JE, Foster GS, Abel RM et al (1973) Hyperalimentation as primary therapy for inflammatory bowel disease. Am J Surg 125:165-171
16. Elson CO, Layden TJ, Nemchausky BA et al (1980) An evaluation of total parenteral nutrition in the management of inflammatory bowel disease. Dig Dis Sci 25:42-48
17. Casola G, vanSonnenberg E, Neff CC et al (1987) Abscesses in Crohn disease: percutaneous drainage. Radiology 163:19-22
18. Safrit HD, Mauro MA, Jaques PF (1987) Percutaneous abscess drainage in Crohn's disease. AJR Am J Roentgenol 148 :859-862
19. Bennett JD, Kozak RI, Taylor BM, Jory TA (1992) Deep pelvic abscesses: transrectal drainage with radiologic guidance. Radiology 185:825-828
20. Alexander AA, Eschelman DJ, Nazarian LN, Bonn J (1994) Transrectal sonographically guided drainage of deep pelvic ab-scesses. AJR Am J Roentgenol 162: 1227-1230, discussion 1231-1222
21. Varghese JC, O'Neill MJ, Gervais DA et al (2001) Transvaginal catheter drainage of tuboovarian abscess using the trocar method: technique and literature review. AJR Am J Roentgenol 177:139-144
22. VanDerKolk HL (1991) Small, deep pelvic abscesses: definition and drainage guided with an endovaginal probe. Radiology 181:283-284
23. Yeung EY, Ho CS (1993) Percutaneous radiologic drainage of pelvic abscesses. Ann Acad Med Singapore 22:663-669
24. Butch RJ, Mueller PR, Ferrucci JT Jr et al (1986) Drainage of pelvic abscesses through the greater sciatic foramen. Radiology 158:487-491
25. Malden ES, Picus D (1992) Hemorrhagic complication of transgluteal pelvic abscess drainage: successful percutaneous treatment. J Vasc Interv Radiol 3:323-326, discussion, 327-328
26. Valero V, Senior J, Wantanakunakorn C (1985) Liver abscess complicating Crohn's disease presenting as thoracic empyema. Am J Med 79:659-662
27. Mir-Madjlessi SH, McHenry MC, Farmer RG (1986) Liver abscess in Crohn's disease. Re-

port of four cases and review of the liter-ature. Gastroenterology 91:987-993
28. Fagge GH (1870) Hepatic abscesses following ulceration of the large intestine. Trans Pathol Soc Lond 21:235-236
29. Teague M, Baddour LM, Wruble LD (1988) Liver abscess: a harbinger of Crohn's disease. Am J Gastroenterol 83:1412-1414
30. Kotanagi H, Sone S, Fukuoka T et al (1991) Liver abscess as the initial manifestation of colonic Crohn's disease: report of a case. Jpn J Surg 21:348-351
31. Czernichov B, Filippi de la Palavesa MM, Bergier JM (1993) Liver abscess revealing Crohn's disease. Gastroenterol Clin Bio 17:153-155
32. Andre M, Aumaitre O (1998) Disseminated aseptic abscesses associated with Crohn's disease. Digestive Dis 43:420-428
33. Greenstein AJ, Sachar DB, Lowenthal D et al (1985) Pyogenic liver abscess in Crohn's disease. Quart J Med 56(220):505-518

Standard Therapeutic Approach

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R. Mangiarotti

Introduction

Crohn's disease (CD) is characterized by patchy, transmural inflammation that may affect any part of the gastrointestinal tract. It may be defined by location (distal ileum, colonic, ileocolic, upper gastrointestinal) or by pattern of disease (inflammatory, fistulizing, or stricturing). Both these aspects have been combined in the Vienna classification. CD may cause intestinal obstruction due to strictures, fistulae (often perianal), or abscesses, and the clinical course of the disease is characterized by exacerbations and remission. Therapy for inflammatory bowel disease (IBD) is a rapidly evolving field, with the many new biological agents under investigation likely to change therapeutic strategies in the future. The general approach for treating active CD must consider the degree of activity, location, and behavior of the disease, including its course, response to previous medications, side effects of medication, and possible presence of extraintestinal manifestations). In this chapter, the principal drugs employed as a first-line treatment are discussed.

Aminosalicylates

Aminosalicylates (mesalazine or 5-aminosalicylic acid (5-ASA), "mesalamine" in the USA) [1] are available in different formulations that deliver millimolar concentrations to the gut lumen, including as oral tablets, sachets or suspension, liquid or foam enemas, or suppositories. They act on epithelial cells by a variety of mechanisms to moderate the release of lipid mediators, cytokines, and reactive oxygen species. Oral forms include:

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- pH-dependent release/resin coated (Asacol, Salofalk, Claversal)
- time-controlled release (Pentasa)
- delivery by carrier molecules, with release of 5-ASA after splitting by bacterial enzymes in the large intestine: sulfasalazine (Salazopyrin), olsalazine (Dipentum), balsalazide (Colazide)

Active Disease

Higher doses of 5-ASA (4 g/day) are more effective than placebo for inducing remission in mild CD [2]. In active Crohn's ileocolitis, a meta-analysis of the three placebo-controlled trials of Pentasa (4 g daily for 16 weeks in a 615 patients) showed a mean reduction of the CD activity index (CDAI) from baseline of –63 points, compared with –45 points for placebo ($p = 0.04$). The clinical significance of this marginal advantage is irrelevant. Subgroup analyses do not provide answers to whether one group of patients benefit more than another.

Maintaining Remission

5-ASA is less effective for maintaining remission in CD [3]. Mesalazine (>2 g/day) reduces relapse after surgery (NNT = 8), especially after small bowel resection (40% reduction at 18 months). It is ineffective after steroid-induced remission, except for those at high risk of relapse, at a dose of 4 g/day (relapse risk on placebo 2.0, CI 1.0–3.8).

Adverse Effects of Aminosalicylates

5-ASA intolerance occurs in up to 15% of patients taking the drug [4]. Diarrhea (3%), headache (2%), nausea (2%), rash (1%), and thrombocytopenia (<1%) are reported, but a systematic review has confirmed that all new 5-ASA agents are safe, with adverse events that are similar to placebo. Acute intolerance in 3% of patients may mimic a flare of colitis. Recurrence of symptoms on re-challenge confirms this side effect. Renal impairment (including interstitial nephritis and nephrotic syndrome) is rare and idiosyncratic. A population-based study found that the risk of this side effect (OR 1.60, CI 1.14–2.26 compared with normal) is associated with disease severity rather than the dose or type of 5-ASA.

Patients with pre-existing renal impairment who are taking other potentially nephrotoxic drugs, or with comorbid disease should have renal function monitored during 5-ASA therapy. Most clinicians believe that creatinine, urine analysis, and full blood count should be measured every 3–6 months during 5-ASA therapy, although there is no evidence favoring one monitoring regimen over another.

Corticosteroids

The many forms of corticosteroids include oral prednisolone, prednisone, budesonide, beclomethasone dipropionate (BDP), intravenous hydrocortisone, and methylprednisolone. Hydrocortisone, prednisolone metasulphobenzoate, betamethasone, and budesonide are available as topical suppositories, foam, or liquid enemas.

Many strategies attempt to maximize the topical effects while limiting the systemic side effects of steroids. Budesonide and BDP are poorly absorbed corticosteroids with limited bioavailability and thus have therapeutic benefits in ileocecal CD but with reduced systemic toxicity.

Choice and Mechanism

Corticosteroids are potent anti-inflammatory agents used for the treatment of moderate to severe flares of CD [5]. They have no role in maintenance therapy because of the large number of side effects. These compounds inhibit several inflammatory pathways: the suppression of interleukin transcription; the induction IB, which stabilizes the NFκB complex; the suppression of arachidonic acid metabolism; and the stimulation of apoptosis by lymphocytes within the lamina propria of the gut.

Efficacy for Active CD

Two major trials established that corticosteroids provided effectively induced remission of CD [6-7]. The National Cooperative Crohn's Disease Study randomized 162 patients, achieving 60% remission with 0.5–0.75 mg prednisone/kg/day (the higher dose for more severe disease) and tapering over 17 weeks, compared with 30% on placebo (NNT = 3). The comparable European Cooperative Crohn's Disease Study in 105 patients achieved 83% of remission on 1 mg prednisone/kg/day compared with 38% on placebo (NNT = 2) over 18 weeks. No formal dose–response trial has been performed, but a remission rate of 92% within 7 weeks was achieved in 142 patients with moderately active CD who received 1 mg prednisone/kg/day. Budesonide is slightly less effective than prednisolone, but is an appropriate alternative for patients with active ileo-ascending colonic disease.

Adverse Effects of Corticosteroids

Three broad groups of side effects can be identified, although 50% of patients report no adverse events. Early effects due to supraphysiological doses include cosmetic (acne, moon face, edema), sleep and mood disturbances, dyspepsia, and glucose intolerance. Effects associated with prolonged use (usually >12 weeks, but sometimes less) include posterior subcapsular cataracts, osteoporosis, osteonecrosis of the

femoral head, myopathy, and susceptibility to infection. Effects during withdrawal include acute adrenal insufficiency (from sudden cessation), a syndrome of myalgia, malaise, and arthralgia (similar to recrudesence of CD), or increased intracranial pressure. Complete steroid withdrawal is facilitated by the early introduction of azathioprine, adjuvant nutritional therapy, or timely surgery. Osteoprotective therapy is considered advisable if the duration of therapy is likely to be >12 weeks, although some clinicians advocate supplements of calcium and vitamin D for all patients.

Thiopurines

Purine antimetabolites inhibit ribonucleotide synthesis, but they induce T cell apoptosis by modulating cell (Rac1) signaling. Thiopurines such as azathioprine (AZA), which is is metabolized to mercaptopurine (6-MP) and then to 6-thioguanine nucleotides, and thioguanine have been used for the treatment of IBD, but caution is appropriate because of potential hepatotoxicity [8–10].

Efficacy

Thiopurines are effective both for the treatment of active disease and for maintaining remission in CD. A Cochrane review of the efficacy of AZA and MP for inducing remission in active CD showed a benefit for thiopurine therapy compared with placebo, with an odds ratio of 2.36 (95% CI 1.57–3.53). Thiopurines have effect as maintenance therapy for CD for up to 4 years. In a prospective trial, 83 patients with CD who had been in remission for 3.5 years on AZA were randomized to continue AZA or placebo and followed for 18 months. Relapse rates were 21% and 8% in the placebo and AZA groups, respectively ($p = 0.0195$). Patients with CD treated by AZA are recommended to continue treatment for 3–4 years and then determine whether AZA can be stopped. For the 20% who relapse, AZA can be restarted and continued. Direct comparisons of the efficacy of AZA and 6-MP in IBD are not available. Some patients who are intolerant of AZA may tolerate MP.

Indications for Thiopurines

The main role for thiopurines is to avoid the need for steroids; therefore thiopurines should be considered for patients who require two or more corticosteroid courses within a calendar year, and for steroid-dependent or steroid-refractory patients. Purines also should be employed in postoperative prophylaxis in case of severe (fistulating or extensive) CD.

Dosing

The maintenance dose of AZA should be 2–2.5 mg/kg/day and that of 6-MP 1–1.5 mg/kg/day in patients with ulcerative colitis (UC) or CD. Tailoring or optimization of thiopurine therapy can occur before or during treatment. The "maximum" dose will differ between individuals and it depends on at which level the patient develops leukopenia. The latter is a myelotoxic side effect of thiopurines and the metabolic phenotype of the individual can be defined by measuring thiopurine methyl transferase (TPMT) activity or determining the TPMT genotype.

Monitoring Thiopurine Therapy

Manufacturers suggest weekly full blood counts (FBCs) for the first 8 weeks of therapy followed by blood tests at least every 3 months. However, there is no evidence that this provides effective monitoring. Instead, less frequent monitoring (within 4 weeks of starting therapy and then every 6–12 weeks) may be sufficient. It is just as important to advise patients to promptly report a sore throat or other sign of infection.

Adverse Effects of Thiopurines

Flu-like symptoms (myalgia, headache, diarrhea) are the most common cause of intolerance (affecting up to 20%). They usually occur after two to three weeks and stop abruptly when the drug is withdrawn. Serious leukopenia can develop suddenly and unpredictably, in between blood tests, although it is rare (around 3%). Hepatotoxicity and pancreatitis are uncommon (<5%). Generally, if the drug is tolerated for 3 weeks, long-term tolerance and benefit can be expected. Thiopurines can reasonably be continued during pregnancy if CD has been refractory. In a study of 155 men and women with IBD who were parents of 347 pregnancies while taking 6-MP, there was no difference in miscarriage, congenital abnormality, or infection rates in the thiopurine group compared with a control group. The risk of malignancy related to thiopurine is low. Large audits of up to 755 patients have shown no increased risk of lymphoma or other cancers in IBD patients treated with AZA. Nevertheless, a meta-analysis of six studies evaluating thiopurines and lymphoma in IBD reported a pooled relative risk of 4.18 (95% CI 2.07–7.51; 11 observed cases, 2.63 expected). The approximate fourfold increased risk of lymphoma could be a result of the medications, the severity of the underlying disease, or a combination of the two. In general, the opinion of most experts is that the benefits of AZA outweigh any risk of lymphoma in IBD patients. Although this is best discussed with the patients, the meta-analysis was unable to show that the magnitude of risk was related to the duration of therapy. To put the data in perspective, the incidence of lymphoma rises with age. Consequently, the number needed to harm (NNH) to cause one lymphoma by

treating patients with thiopurines in their third decade (age 20–29) is 4357, while the NNH for treating patients in their sixth decade is 1126. Serious, systemic viral infections, including varicella zoster and cytomegalovirus, can complicate thiopurine therapy; in such cases, rapid treatment with antiviral agents under expert guidance is appropriate. There is some evidence that lymphomas occurring in patients taking AZA/MP are caused by Epstein-Barr virus infection.

Methotrexate

Methortrexate (MTX) [11-13] is administered orally or via subcutaneous, or intramuscular injection. Polyglutamated metabolites of MTX inhibit dihydrofolate reductase, but this cytotoxic effect does not explain its anti-inflammatory actions; rather, inhibition of cytokine and eicosanoid synthesis probably contributes.

Efficacy of Methotrexate

The indications for MTX are the same as for thiopurine therapy, but at present, the drug is generally reserved for treatment of active or relapsing CD in patients refractory to or intolerant of AZA or MP. Most in the consensus adopted this approach (86% always started AZA/6-MP before treatment with MTX, 14% used them interchangeably).

Dose, Delivery, and Duration

The standard doses of MTX in the treatment of CD are 25 mg/week. For practical reasons relating to the reconstitution of parenteral cytotoxic drugs, oral dosing is most convenient, although parenteral administration may be more effective. Subcutaneous administration is reserved for patients with CD of the small intestine, who are unable to absorb oral MTX. There are no conclusions on the duration of therapy. The 3-year remission rate for MTX in one series was 51%, which compares with data on azathioprine from the same center (69% 3-year remission rate for azathioprine).

Monitoring Therapy

Measurement of FBC and liver function tests are recommended before and within 4 weeks of starting MTX therapy, then monthly. The same caveats as for monitoring thiopurine therapy apply. Patient follow up must be performed by a specialist.

Side Effects

A high percentage of patients (10–18%) discontinue MTX because of its side effects. Early toxicity is primarily gastrointestinal (nausea, vomiting, diarrhea, and stomatitis) but may be limited by co-prescription of folic acid (5 mg at a 2- or 3-day interval from the MTX). Moreover, MTX causes hepatotoxicity and pneumonitis. A study of liver biopsies in IBD patients taking MTX showed mild histological abnormalities, despite cumulative doses of up to 5410 mg. Surveillance liver biopsy is not warranted, but if the AST doubles then it is sensible to withhold MTX until it returns to normal, before a re-challenge. The prevalence of pneumonitis has been estimated at two to three cases per 100 patient years of exposure, but large series have not reported any cases.

Other Immunomodulators

Among the immunomodulators [14-17] used in the treatment of CD are cyclosporin (CsA) and tacrolimus. However, these calcineurin inhibitors are of limited value in CD. Their mechanism of action is thought to result from inhibition of the nuclear translocation of the transcription factor NFAT (nuclear factor of activated T cells), thereby preventing the downstream start of T-cell cytokine transcription.

Efficacy and Selection

A single trial has shown some efficacy for treatment of CD with oral CsA whereas in three further placebo-controlled trials, no efficacy of oral CsA for treatment of CD was found. Three small, uncontrolled case series have, however, reported efficacy of intravenous CsA (4–5 mg/kg/day) for both inflammatory and fistulating CD, but randomized controlled studies of intravenous CsA are lacking. Therefore, the use of oral CsA for corticosteroid-refractory or corticosteroid-dependent CD cannot be recommended, but the use of short-term intravenous CsA to induce remission is still debated.

In contrast, oral tacrolimus for inflammatory CD has only been reported in uncontrolled studies and case reports; consequently, the experience with this drug is too limited to allow its recommendation for general use in the therapy of CD.

Antibiotics

The use of antibiotics in the treatment of CD [18] is based on the most widely accredited hypothesis on the etiology of IBD, in which the disease is ascribed to an excessively aggressive immune response to antigens in the gut of genetically susceptible individuals [19]. Intestinal bacteria are the most important antigens present in the gut

and their role in IBD is supported by extensive experimental and clinical data. Antibiotics are widely used in clinical practice for treating the septic complications of IBD. In the last few years, several groups have reported the presence of *Escherichia coli* in the ileal and colonic tissues of CD patients. *E. coli* with adherent-invasive properties (AIEC) have been found in these specimens and shown to be able to colonize the intestinal mucosa and to invade and replicate within macrophages, inducing the secretion of a large quantity of tumor necrosis factor (TNF).

Different randomized studies on antibiotic therapy, mainly in CD but also in UC and pouchitis, have yielded varied results. The antibiotics employed were those with antimycobacterial activity; metronidazole, active against anaerobic bacteria; ciprofloxacin, active against *E. coli*, and, most recently, the non-absorbable agent rifaximin.

Efficacy

Randomized studies have shown that metronidazole is no better than placebo with respect to remission, but the drop in CDAI was 67–97 points in the metronidazole group compared with one point in the placebo group ($p = 0.002$). Patients with isolated small-bowel disease showed no benefit, but only 56 of 105 patients completed the trial, with 17 withdrawing due to adverse events. In a 16-week crossover trial, the responses to metronidazole and sulfasalazine were similar (25% remission rates in each arm, no placebo), but there were more patients who failed sulfasalazine and then responded to metronidazole than vice versa.

Results on ciprofloxacin show that its efficacy is similar to that of 5-ASA in active CD, with a response rate of 40–50% after 6 weeks. The combination of ciprofloxacin and metronidazole has been compared with corticosteroids, showing 46% vs. 63% remission (NS) [20].

Rifaximin is active against anaerobic bacteria and *E. coli* and has been approved for the treatment of traveler's diarrhea. In 2008, two case reports described the response to rifaximin in 6 patients with CD. In one of these articles, the clinical remission obtained in three patients with newly diagnosed ileal CD was confirmed also by a capsule endoscopy, which showed mucosal improvement [21]. The doses employed varied from 600 to 800 mg/daily for a period of 3 weeks to over 6 months. A previous controlled trial had shown that 800 mg of rifaximin, given twice daily for 3 months to patients with elevated CRP, provided the best result.

Other antibiotics require additional testing. A meta-analysis of six trials of antimycobacterial treatment showed that only the two trials including corticosteroids for induction of remission influenced the disease. A subsequent randomized trial consisting of 216 patients that was conducted in Australia showed that triple therapy in conjunction with corticosteroids improved the response at 16 weeks, although the pattern of disease was unchanged over 3 years (Selby 2005, personal communication). At present, antibiotics are only considered specific for septic complications, symptoms attributable to bacterial overgrowth, or perineal disease. Antimycobacterial therapy cannot be recommended based on the evidence from controlled trials. The duration of antibiotic therapy is still debated.

Adverse Effects

Side effects of antibiotics remain a concern. Apart from short-term intolerance in around 50% (nausea, metallic taste, abreaction to alcohol), polyneuropathy secondary to metronidazole limits their long-term use. Ciprofloxacin is better tolerated in the short term, but is associated with tendonitis and Achilles tendon rupture, especially with concomitant corticosteroids.

It is our opinion that antibiotics are extremely useful for treating CD complications, especially when immunosuppressors may be counter-indicated because of abscesses and septic symptoms. In the active phases of disease, when the colon is involved, antibiotics are a possible first-step therapy; however, there are serious doubts about their efficacy in Crohn's ileitis. The combination of antibiotics with steroids needs to be reassessed.

Treatment According to Disease Site and Activity

Active Ileal/Ileocolonic/Colonic Disease

Initial treatment of active ileal or ileocolonic CD with high-dose mesalazine, corticosteroids, nutritional therapy, or surgery should be tailored to the severity of disease and take the views of the patient into account. The European Crohn's and Colitis Organisation (ECCO) published several recommendations on the treatment of CD [22]:

Recommendations

- In mild ileocolonic CD, high-dose mesalazine (4 g/daily) may be sufficient initial therapy.
- For patients with moderate to severe disease, or those with mild to moderate ileocolonic CD that has failed to respond to oral mesalazine, oral corticosteroids such as prednisolone (40–60 mg daily) is appropriate.
- Prednisolone should be reduced gradually according to severity and patient response, generally over 8 weeks. More rapid reduction is associated with early relapse.
- Budesonide (9 mg daily) is appropriate for patients with isolated ileocecal disease with moderate disease activity, but marginally less effective than prednisolone.
- Intravenous steroids (hydrocortisone 400 mg/day or methylprednisolone 60 mg/day) are appropriate for patients with severe disease. Concomitant intravenous metronidazole is often advisable because it may be difficult to distinguish between active disease and a septic complication.
- Sulfasalazine (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.
- Metronidazole (10–20 mg/kg/day), although effective, is not usually recommend-

ed as first-line therapy for CD in view of its potential side effects. It has a role in selected patients with colonic or treatment resistant disease, or those who wish to avoid steroids.
- Ciprofloxacin (500 mg bid) has shown similar efficacy to 5-ASA in active CD and is better-tolerated in the short term.
- Topical mesalazine may be effective in left-sided colonic CD of mild to moderate activity.
- Azathioprine (1.5–2.5 mg/kg/day) or mercaptopurine (0.75–1.5 mg/kg/day) may be used in active CD as adjunctive therapy and as a steroid-sparing agent. However, its slow onset of action precludes its use as a sole therapy.

Fistulating and Perianal Disease

Active CD is often complicated by perianal disease or fistulae elsewhere in the gastrointestinal tract. The initial aim should be to treat active disease and sepsis. For more complex, fistulating disease, the approach involves defining the anatomy, supporting nutrition, and potential surgery. Further treatment will be discussed in another chapter of this book.

Recommendations
- Metronidazole (400 mg tds) and/or ciprofloxacin (500 mg bid) are appropriate first-line treatments for simple perianal fistulae.
- Azathioprine (1.5–2.5 mg/kg/day) or mercaptopurine (0.75–1.5 mg/kg/day) is potentially effective for simple perianal fistulae or enterocutaneous fistulae when distal obstruction and abscess have been excluded.

Maintenance of Remission

The indication and choice of medications for prevention of relapse in patients with medically induced remission should take into account three main factors: the course of the disease (initial presentation, frequency, and severity of flares); the effectiveness and tolerance of treatments previously used for induction of remission or maintenance; and the extent of disease. Other factors, such as the presence of biological signs of inflammation and smoking status, should also be considered, as well as constraints (logistic, social, or financial) affecting the choice of treatment. Patients should be encouraged to participate in the decision-making process. After the first presentation if remission has been achieved medically, maintenance with mesalazine is a treatment option, although there is no consistent evidence for its efficacy. If remission has been achieved with systemic corticosteroids, azathioprine should be considered.

Recommendations
- All smokers should be strongly advised to stop, and to seek help (counseling, nicotine patches, or substitutes) in order to achieve this.
- Mesalazine has limited benefit and is ineffective at doses <2 g/day, or for those

who have needed steroids to induce remission.
- Immunomodulation with azathioprine, mercaptopurine, or MTX is usually appropriate if patients relapse more than once per year as steroids are withdrawn.
- Azathioprine (1.5–2.5 mg/kg/day) or 6-mercaptopurine (0.75–1.5 mg/kg) is effective, but reserved as second-line therapy because of potential toxicity.
- Methotrexate (15–25 mg IM weekly) is effective for patients whose active disease has responded to intramuscular MTX. It is appropriate for those intolerant of, or who have failed azathioprine/mercaptopurine therapy, once potential toxicity and other options, including surgery, have been discussed with the patient. Folic acid (5 mg once a week, taken 3 days after MTX) may reduce side effects. Subcutaneous or oral therapy may be effective.

Chronic-Active and Steroid-Dependent Disease

Long-term treatment with steroids, including budesonide, is undesirable. Patients who have a poor response to steroids are divided into steroid-refractory (active disease in spite of an adequate dose and duration of prednisolone): and steroid-dependent (a relapse when the steroid dose is reduced below 20 mg/day, or within 6 weeks of stopping steroids). Such patients should be considered for treatment with immunomodulators if surgery is not an immediate consideration.

Recommendations
- Azathioprine (1.5–2.5 mg/kg/day) and mercaptopurine (0.75–1.25 mg/kg/day) are the first-line agents of choice for steroid-dependent disease.
- Methotrexate (IM 25 mg weekly for up to 16 weeks followed by 15 mg weekly) is effective for chronic active disease. Oral dosing is effective for many patients.

Preventing Postoperative Recurrence

For patients who smoke, cessation significantly reduces postoperative relapse. Additional medical therapy should be considered for at least 18 months after surgery, especially if disease has frequently relapsed prior to surgery, or after surgery for fistulating disease, or after a second operation. Prophylactic treatment is recommended after resection of the small intestine. The drug of choice is mesalazine. Imidazole antibiotics have been shown to be effective after ileocolic resection. Other drugs, including azathioprine/6-mercaptopurine, should be considered as first-line therapy in high-risk patients. The start of prophylaxis should be within 2 weeks of surgery, on the basis of pathophysiological considerations, although an early start has not been demonstrated superior to later treatment. The duration of prophylaxis should be at least 2 years.

Recommendations
- Mesalazine (≥2 g/day) lowers postoperative recurrence in small bowel disease, but is ineffective after colonic resection.

- Azathioprine (1.5–2.5 mg/kg/day) or mercaptopurine (0.75–1.5 mg/kg/day) may be used for preventing postoperative recurrence and may be better than mesalazine.
- Metronidazole (20 mg/kg/day for 3 months) effectively delays recurrence after ileocolic resection for up to 18 months, but potential side effects include peripheral neuropathy.

References

1. Sandborn WJ, Hanauer SB (2003) Systematic review: the pharmacokinetic profiles of oral mesalazine formulations and mesalazine prodrugs used in the management of ulcerative colitis. Aliment Pharmacol Ther 17:29–42
2. Prantera C, Cottone M, Pallone F et al (1999) Mesalamine in the treatment of mild to moderate active Crohn's ileitis: results of a randomized multicenter trial. Gastroenterology 116:521–526
3. Lochs H, Mayer M, Fleig WE et al (2000) Prophylaxis of post-operative relapse in Crohn's disease with mesalamine: European Cooperative Crohn's Disease Study VI. Gastroenterology 118:264–273
4. van Staa T P, Travis S P L, Leufkens H J M et al (2004) 5-aminosalicylic acids and the risk of renal disease: a large British epidemiological study. Gastroenterology 126:1733–1739
5. Franchimont D, Kino T, Galon J et al (2003) Glucorticoids and inflammation revisited: the state of the art. Neuroimmunomodulation 10:247–260
6. Summers RW, Switz DM, Sessions JT et al (1979) National co-operative Crohn's disease study group: results of drug treatment. Gastroenterology 77:847–869
7. Steinhart AH, Ewe K, Griffiths AM et al (2001) Corticosteroids for maintaining remission of Crohn's disease. Cochrane Database Syst Rev (3) :CD000301
8. Sandborn W, Sutherland L, Pearson D et al (2000) Azathioprine or 6-mercaptopurine for inducing remission of Crohn's disease. Cochrane Database Syst Rev (2) :CD000545
9. Lemann M, Bouhnik Y, Colombel J et al (2002) Randomized, double-blind, placebo-controlled, multicentre, azathioprine withdrawal trial in Crohn's disease. Gastroenterology 122:A23
10. Colombel JF, Ferrari N, Debuysere H et al (2000) Genotypic analysis of thiopurine S-methyltransferase in patients with Crohn's disease and severe myelosuppression during azathioprine therapy. Gastroenterology 118:1025–1030
11. Fraser AG (2003) Methotrexate: first or second-line immunomodulator? Eur J Gastroenterol Hepatol 15:225–231
12. Alfadhli AA, McDonald JW, Feagan BG (2003) Methotrexate for induction of remission in refractory Crohn's disease (Cochrane Review). Cochrane Database Syst Rev (1):CD003459
13. Feagan BG, Fedorak RN, Irvine EJ et al (2000) A comparison of methotrexate with placebo for the maintenance of remission in Crohn's disease. North American Crohn's Study Group Investigators. N Engl J Med 342:1627–1632
14. Feagan BG, McDonald JW, Rochon J et al (1994) Low-dose cyclosporine for the treatment of Crohn's disease. The Canadian Crohn's Relapse Prevention Trial Investigators. N Engl J Med 330:1846–1851
15. Feagan BG (1995) Cyclosporin has no proven role as a therapy for Crohn's disease. Inflamm Bowel Dis 1:335–339
16. Egan LJ, Sandborn WJ, Tremaine WJ (1998) Clinical outcome following treatment of refractory inflammatory and fistulizing Crohn's disease with intravenous cyclosporine. Am J Gastroenterol 93:442–448

17. Sandborn WJ, Present DH, Isaacs K L et al (2003) Tacrolimus for the treatment of fistulas in patients with Crohn's disease. Gastroenterology 125:380–388
18. Sasaki M, Sitaraman SV, Babbin BA et al (2007) Invasive Escherichia coli are a feature of Crohn's disease. Lab Invest 87:1042–1054
19. Prantera C, Scribano ML (2009) Antibiotics and probiotics in inflammatory bowel disease why, when, and how. Curr Opin Gastroenterol 25:329–333
20. Prantera C, Zannoni F, Scribano M L et al (1996) An antibiotic regimen for the treatment of active Crohn's disease: a randomized, controlled clinical trial of metronidazole plus ciprofloxacin. Am J Gastroenterol 91:328–332
21. Prantera C, Lochs H, Campieri M et al (2006) Antibiotic treatment of Crohn's disease: results of a multicentre, double-blind, randomized, placebo-controlled trial with rifaximin. Aliment Pharmacol Ther 23:1117–1125
22. Stange EF, Travis SPL, S Vermeire S et al for the European Crohn's and Colitis Organisation (ECCO) (2006) European evidence based consensus on the diagnosis and management of Crohn's disease: definitions and diagnosis Gut 55(Supplement 1):i1–i15

New Therapies

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Introduction

The advent of biological therapies over the past decade has initiated a new therapeutic era for the treatment of Crohn's disease (CD), especially for patients with corticosteroid-dependent, corticosteroid-refractory or fistulizing disease. For years the primary goal of traditional therapies has been the control of disease-related symptoms. However, emerging evidence from inflammatory bowel disease (IBD) research suggests that the new biological agents not only control symptoms but may also alter the natural history of the disease. Accordingly, the therapeutic goals in the medical treatment of CD are changing and now include the rapid induction of clinical remission, the maintenance of steroid-free clinical remission, the healing of mucosal lesions, improvements in health-related quality of life, and a reduction in both the need for surgery and hospital stay.

The biological drugs include infliximab, adalimumab, and certolizumab pegol, which are directed against tumor necrosis factor-alpha (TNF-α), a pro-inflammatory cytokine that plays a central role in the pathogenesis of CD, and natalizumab, an anti-integrin antibody. These and other such agents have provided new and effective treatment options for patients with CD. The efficacy and safety of biological therapy, for induction and for the maintenance of remission, have been evaluated in multicenter, double-blind, placebo-controlled trials in patients with moderate to severe CD and an inadequate response to traditional drugs.

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Infliximab

Infliximab (Remicade), a chimeric mouse-human monoclonal antibody directed against TNF-α, was the first biological therapy to be approved in the US for the treatment of CD (1998). In Europe, infliximab is approved for the treatment of severe active or fistulizing active CD in patients who are intolerant or have not responded to a full and adequate course of conventional therapy. Infliximab also has been extensively used for the treatment of rheumatoid arthritis, ankylosing spondylitis, psoriasis, and psoriatic arthritis.

In the ACCENT I study, a large maintenance trial, patients with moderate to severe CD who responded to a single intravenous infusion of infliximab 5 mg/kg, administered at week 0, were randomized to receive infliximab 5 mg/kg or placebo at weeks 2 and 6 followed by a scheduled therapy every 8 weeks with infliximab (5 or 10 mg/kg) or placebo [1]. At week 14, patients who had responded but then worsened were eligible to crossover to episodic treatment with the active drug. At week 54, remission was observed in 28% of patients who received infliximab 5 mg/kg, in 38% of patients who received infliximab 10 mg/kg, and in 14% among those receiving placebo. Furthermore, at week 54 significantly more infliximab recipients than placebo recipients were able to discontinue steroid therapy while remaining in remission.

Data from both the ACCENT I trial and other studies have shown that episodic treatment leads to significantly higher rates of anti-infliximab antibody formation than scheduled administration [1,2]. The development of immunogenicity correlates with a decrease in the degree and duration of response. Comparisons between ACCENT I patients who received therapy as scheduled and those treated with episodic therapy confirmed that patients in the scheduled group had a better response, also with respect to the healing of mucosal lesions, as shown in the endoscopic substudy of ACCENT I [2,3]. Mucosal healing was associated with reduced hospitalization and need for surgery at 1 year [3]. Therefore, scheduled maintenance therapy (5 mg/kg every 8 weeks) is more appropriate for disease maintenance than an episodic treatment regimen.

In some patients, there is a gradual or complete loss of infliximab efficacy over time, in part due to the development of antibodies to the drug, necessitating increased dosing to 10 mg/kg, shortened dosing intervals, or the use of an alternative agent [4]. In the ACCENT I study, 88% of patients who had initially responded to infliximab but were no longer responsive during maintenance therapy regained response when the drug dose was increased to up to 10 mg/kg. Concomitant therapy with immunosuppressive drugs, such as 6-mercaptopurine, azathioprine [5,6], or methotrexate [7], reduces the likelihood of developing antibodies and is associated with an increased likelihood of response but may enhance the risk of side effects.

The efficacy of infliximab specifically in patients with fistulas has also been evaluated in randomized, placebo-controlled trials [8,9]. In the first study, 94 patients with draining abdominal or perianal fistulas of ≥ 3 months were randomized to receive infliximab (5 or 10 mg/kg) or placebo at 0, 2, and 6 weeks [8]. Complete ces-

sation of drainage from fistulas was observed in significantly more patients treated with infliximab 5 mg/kg and 10 mg/kg than in placebo patients. The fistulas remained closed for a median time of 3 months.

The subsequent ACCENT II trial evaluated the efficacy of infliximab in the maintenance of fistulizing CD and yielded similar results [9]. At week 54, among patients responding to the induction phase (infliximab 5 mg/kg at weeks 0, 2, and 6) and randomized to infliximab 5 mg/kg or placebo, significantly more patients receiving infliximab were able to maintain response (46%) compared to the placebo group (23%).

Adalimumab

Adalimumab (Humira), the second anti-TNF agent to be developed, is a fully human monoclonal anti-TNF antibody administered subcutaneously. The drug has been approved for use in the treatment of rheumatoid arthritis, ankylosing spondylitis, psoriatic arthritis, psoriasis. In November 2007, adalimumab was approved by the Italian Drug Agency (AIFA) for use in the treatment of patients with severe active CD who have had an inadequate response or intolerance to conventional therapy.

The first placebo-controlled trial demonstrating adalimumab efficacy was the CLASSIC I study, in which patients with active moderate to severe CD, naïve to anti-TNF therapy, were randomized to one of three different dose regimens (160/80, 80/40, or 40/20 mg) administered at weeks 0 and 2 [10]. The 160/80 mg dose regimen proved to be the most efficacious, with a remission rate at week 4 of 36% compared to 12% of the placebo group. The CLASSIC II trial showed the long-term efficacy of adalimumab in the maintenance of remission in patients who completed the CLASSIC I study [11].

In the large CHARM trial, patients who responded to open-label doses of adalimumab 80/40 mg at weeks 0 and 2 were randomized to either adalimumab 40 mg every week, adalimumab 40 mg every other week, or placebo [12]. Both dose regimens produced significantly greater remission rates at week 56 (41% and 36%, respectively) compared with placebo (12%). Complete fistula closure at week 56 was observed in 33% of adalimumab-treated patients versus 13% in the placebo group.

In contrast to infliximab, adalimumab has not been specifically studied in fistulizing CD.

The GAIN study showed that, among patients who had lost response to infliximab or were intolerant to the drug, those treated with adalimumab 160/80 mg achieved better clinical remission rates than those in the placebo group at week 4 (21.4 vs. 7.2%) [13]. Although statistically significant, this result was lower than the result observed in the CLASSIC I trial. A subanalysis of the CHARM study showed that response rates to adalimumab therapy in patients who were previously treated with infliximab were approximately 10% less than those for anti-TNF naïve patients. This is in agreement with the observation that patients who previously received anti-TNF therapy had a lower response when exposed to a second agent within this class. It is

unclear whether previous treatment with a biological drug implies more serious disease or reflects a loss of efficacy to the drug's mechanism of action; however, the development of antibodies to an anti-TNF drug does not correlate with the development of antibodies to another agent.

A loss of efficacy to adalimumab over time has also been noted. In patients who show a loss of response to 40 mg every other week, response can be re-established by decreasing the interval to 40 mg weekly.

Certolizumab Pegol

The latest anti-TNF agent to be approved by the Food and Drug Administration (FDA), certolizumab pegol (Cimzia), is a humanized anti-TNF pegylated Fab' fragment. Pegylation is thought to increase the half-life of the drug, allowing it to be administered subcutaneously once every 4 weeks. Differently from infliximab and adalimumab, this agent does not induce the apoptosis of T cells. However, in March 2008, the European Medicines Agency (EMEA) confirmed the initial negative opinion of certolizumab pegol, refusing marketing authorization because of insufficient evidence showing a benefit of the drug. Therefore, to date, certolizumab is approved and commercially available for the treatment of CD only in the US and Switzerland.

The efficacy of certolizumab as an induction and maintenance therapy in patients with moderate to severe CD was evaluated in the two large trials, PRECISE I and PRECISE 2 [14,15]. In the first study, certolizumab 400 mg at weeks 0, 2, and 4 and then every 4 weeks showed a modest advantage compared to placebo in inducing clinical response in patients with baseline C reactive protein (CRP) ≥ 10 mg/l [14]. The PRECISE 2 trial demonstrated a better efficacy of certolizumab 400 mg every 4 weeks through week 24 compared to placebo in maintaining clinical response in patients with high baseline CRP, following an open-label induction phase with certolizumab itself [15]. A lower response rate in patients previously exposed to infliximab confirmed the observation made in other anti-TNF trials.

In the recent open-label WELCOME study, patients with active CD who previously lost response or were intolerant to infliximab received certolizumab 400 mg at weeks 0, 2, and 4 in the induction phase [16]. At week 6, those who achieved remission or response (39 and 62%, respectively) were randomized to maintenance therapy with certolizumab 400 mg either every 2 or 4 weeks. The results of this double-blind, maintenance phase and of other studies are awaited.

Safety of Anti-TNF Therapy

The safety of anti-TNF drugs has been evaluated in clinical studies and in post-marketing surveillance, both in the rheumatology and the IBD setting. An increased risk of infections is the main safety concern. Upper respiratory and urinary tract infec-

tions occur commonly with TNF inhibitors but are easily treated. However, more serious infections, such as pneumonia, tuberculosis, sepsis, opportunistic fungal infections and viral infections, have been observed following their use. TNF antagonists should not be given to patients with an active infection and appropriate screening for latent tubercular infection is mandatory. Severe hepatic reactions have been reported, promoting recommendations to screen patients for hepatitis B and C before anti-TNF therapy is initiated.

Long-term safety data for infliximab are available through the TREAT registry [17]. An analysis of the registry's data suggested that the increased risk of serious infections is due to the severity of CD in these patients and to the concomitant use of corticosteroids and immunosuppressive drugs.

The risk of lymphoma and malignancy was not confirmed by the TREAT registry, and an Italian study showed that the risk of neoplasia in CD patients who had received infliximab was comparable to that in CD patients never treated with the drug [17,18]. However, an association between infliximab and adalimumab as well as with thiopurines and the development of a rare, aggressive form of non-Hodgkin lymphoma, classified as hepatosplenic T-cell lymphoma, has been reported in young patients with IBD [19].

The formation of antibodies to infliximab may lead to acute and delayed hypersensitivity reactions. The incidence of antibody development can be reduced by scheduled maintenance therapy, concomitant use of immunosuppressive drugs, and pre-medication with corticosteroids. Antibody formation has also been reported with adalimumab, but it was not associated with increased side effects.

Other adverse events associated with the use of anti-TNF agents are skin lesions, worsening of congestive heart failure, rare hematological events, and new onset or exacerbation of demyelinating disorders.

Natalizumab

Natalizumab (Tysabri) is a humanized monoclonal antibody against the α4 integrin subunit expressed on the surface of leukocytes. The drug therefore prevents leukocyte transmigration across the endothelium into areas of inflammation.

Subsequent to two pivotal placebo-controlled trials showing the efficacy of natalizumab in patients with moderate to severe CD [20,21], Sandborn et al. evaluated the drug in the induction and maintenance of clinical response. In the first of their two tandem large studies, ENACT-1 and ENACT-2 [22], 905 patients with moderate to severe CD were randomized to receive an infusion of 300 mg of natalizumab or placebo at weeks 0, 4, and 8. Similar rates of clinical response and remission between active drug and placebo were obtained at week 10. The ENACT-2 trial showed that natalizumab effectively maintains remission. In this study, patients who had responded to natalizumab in the first trial were randomized to receive natalizumab 300 mg or placebo every 4 weeks. Patients treated with active drug experienced significantly better response rates at 36 weeks than those receiving placebo (61 vs. 28%).

Furthermore, among patients previously treated with anti-TNF therapy, maintenance of response occurred in 54% of patients in the natalizumab group compared to 15% of placebo-treated patients.

In the ENCORE study, patients with active CD and elevated serum CRP levels were randomized to receive natalizumab 300 mg or placebo for a total of three infusions given at weeks 0, 4, and 8 [23]. Natalizumab was significantly superior to placebo with respect to both the induction of clinical response (48 vs. 32%) and remission (26 vs. 16%) at week 8, sustained through week 12. A Cochrane Systematic Review confirmed the efficacy of natalizumab for inducing clinical response and remission in patients with moderately to severely active CD [24].

However, natalizumab was withdrawn from the market in 2005 because of three reported cases of progressive multifocal leukoencephalopathy (PML), caused by the JC virus, in patients who concomitantly received immunosuppressive drugs [25]. After an investigation that found no additional cases [26], the FDA approved the reintroduction of natalizumab as monotherapy, initially for relapsing forms of multiple sclerosis under a careful risk management program (TOUCH Prescribing Program) and later, in 2007, for the treatment of moderate to severe active CD. Therefore, natalizumab may represent an alternative treatment option with a different mechanism of action for those patients who have failed to respond or who have lost response or are intolerant to anti-TNF therapy. These patients must be informed of the rare but mostly fatal risk of PML.

Other Biological Agents

Other biological therapies in development include agents that target pro-inflammatory and recombinant human anti-inflammatory cytokines, such as a fully human monoclonal antibody that targets the interleukin 12/23 shared p40 subunit (ustekinumab), monoclonal antibodies to interleukin-12 (ABT-874, CNTO 1275), interleukin- 6 receptors (tocilizumab), and interferon-γ (fontolizumab); monoclonal antibody to α_4 γ_7 integrins (MLN 0002); and antibodies to interleukin-2 receptor (basiliximab, daclizumab). Other interesting drugs include a fully human recombinant fusion protein, categorized as second-signal blocker of T-cell activation (abatacept), and granulocyte-macrophage colony–stimulating factor (sargramostim).

References

1. Hanauer SB, Feagan BC, Lichtenstein GR et al (2002) Maintenance infliximab for Crohn's disease: the ACCENT I randomised trial. Lancet 359:1541-1549
2. Rutgeerts P, Feagan BG, Lichtenstein GR et al (2004) Comparison of scheduled and episodic treatment strategies of infliximab in Crohn's disease. Gastroenterology 126; 402-413
3. Rutgeerts P, Diamond RH, Bala M et al (2006) Scheduled maintenance treatment with inflix-

imab is superior to episodic treatment for the healing of mucosal ulceration associated with Crohn's disease. Gastrointest Endosc 63:433-442
4. Gisbert JP, Panes J (2009) Loss of response and requirement of infliximab dose intensification in Crohn's disease: a review. Am J Gastroenterol 104:760-767
5. Lemann M, Mary JY, Duclos B et al (2006) Infliximab plus azathioprine for steroid-dependent Crohn's disease patients: a randomized, placebo-controlled trial. Gastroenterology 130:1054-1061
6. Sanborn W, Rutgeerts P, Reinisch et al (2008) SONIC: a randomized, double-blind, controlled trial comparing infliximab and infliximab plus azathioprine to azathioprine in patients with Crohn's disease naïve to immunomodulators and biologic therapy. Am J Gastroenterol 103:1117 (Abstract)
7. Schroder O, Blumenstein I, Stein J et al (2006) Combining infliximab with methotrexate for the induction and maintenance of remission in refractory Crohn's disease: a controlled pilot study. Eur J Gastroenterol Hepatol 18:11-16
8. Present DH, Rutgeerts P, Targan S et al (1999) Infliximab for the treatment of fistulas in patients with Crohn's disease. N Engl J Med 340:1398-1405
9. Sands BE, Anderson FH, Bernstein CN et al (2004) Infliximab maintenance therapy for fistulizing Crohn's disease. N Engl J Med 350:876-885
10. Hanauer SB, Sandborn WJ, Rutgeerts P et al (2006) Human anti-tumor necrosis factor monoclonal antibody (adalimumab) in Crohn's disease: the CLASSIC-I trial. Gastroenterology 130:323-333
11. Sandborn WJ, Hanauer SB, Rutgeerts P et al (2007) Adalimumab for maintenance treatment of Crohn's disease: results of the CLASSIC-II trial. Gut 56:1232-1239
12. Colombel JF, Sanborn WJ, Rutgeerts P et al (2007) Adalimumab for maintenance of clinical response and remission in patients with Crohn's disease: the CHARM trial. Gastroenterology 132:52-65
13. Sandborn WJ, Rutgeerts P, Enns R et al (2007) Adalimumab induction therapy for Crohn's disease previously treated with infliximab. Ann Intern Med 146:829-838
14. Sandborn WJ, Feagan BG, Stoinov S et al (2007) Certolizumab pegol for the treatment of Crohn's disease. N Engl J Med 357:228-238
15. Schreiber S, Khaliq-Kareemi M, Lawrence IC et al (2007) Maintenance therapy with certolizumab pegol for Crohn's disease. N Engl J Med 357:239-250
16. Vermeire S, Abreu MT, D'Haens G et al (2008) Efficacy and safety of certolizumab pegol in patients with active Crohn's disease who previously lost response or were intolerant to infliximab: open-label induction preliminary results of the Welcome study. Gastroenterology 134:A67
17. Lichtenstein GR, Feagan BG, Cohen RD et al (2006) Serious infections and mortality in association with therapies for Crohn's disease: TREAT registry. Clin Gastroenterol Hepatol 4:621-630
18. Biancone L, Orlando A, Kohn A et al (2006) Infliximab and newly diagnosed neoplasia in Crohn's disease: a multicentre, pair study. Gut 55:228-233
19. Mackey AC, Green L, Liang LC et al (2007) Hepatosplenic T cell lymphoma associated with infliximab use in young patients treated for inflammatory bowel disease. J Pediatr Gastroenterol Nutr 44:265-267
20. Gordon FH, Lai CW, Hamilton MI et al (2001) A randomized placebo-controlled trial of a humanized monoclonal antibody to alpha4 integrin in active Crohn's disease. Gastroenterology 121: 268-274
21. Ghosh S, Goldin E, Gordon FH et al (2003) Natalizumab for active Crohn's disease. N Engl J Med 348:24–32
22. Sandborn WJ, Colombel JF, Enns R et al (2005) Natalizumab induction and maintenance therapy for Crohn's disease. N Engl J Med 353:1912–1925

23. Targan SR, Feagan BG, Fedorak RN et al (2007) Natalizumab for the treatment of active Crohn's disease: results of the ENCORE trial. Gastroenterology 132:1672-1683
24. Macdonald JK, McDonald JW (2006) Natalizumab for induction of remission in Crohn's disease. Cochrane Database Syst Rev (3): CD006097
25. Van Assche G, Van Ranst M, Sciot R et al (2005) Progressive multifocal leukoencephalopathy after natalizumab therapy for Crohn's disease. N Engl J Med 353:362-368
26. Yousry TA, Major EO, Ryschkewitsch C et al (2006) Evaluation of patients treated with natalizumab for progressive multifocal leukoencephalopathy. N Engl J M 354:924–933

Cell and Stem-Cell Therapies of Crohn's Disease and Complications

13

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Introduction

Crohn's disease (CD) is an inflammatory acquired pathological process of the small intestine that occurs in adult men and women, with an annual incidence of about 7 new cases in a population of 100,000 [1,2]. This disease has a significant prevalence in adults between the second and fourth decades of life. It is three times more common in individuals of Jewish ancestry and least common in blacks. Up to 5% of those with CD have one or more affected relatives. Despite the partially documented familial pattern of this disease, a pattern of Mendelian inheritance has not been identified. A slightly higher prevalence of CD has been observed in women. The etiology of CD is unknown but recent advances in our knowledge of the cellular mechanisms that sustain gut mucosal inflammation indicate that these patients suffer from an abnormal immune response to intraluminal microbial flora and/or other potential immunogens present in the intestinal mucosa [1,3]. For several years, researchers have reported an increased activity of type 1 helper T cells (TH1), which secrete relevant amounts of interferon (IFN)-γ and interleukin (IL)-2, as a key starting point in both chronic immune activation and secondary, persistent, mucosal inflammation. Recent experimental evidence suggests that CD is also sustained by a defective innate immune response to luminal microbial species. Specifically, the NOD2/CARD15 pathway, in which muramyl dipeptides of the bacterial cell wall are recognized by leukocytes, with subsequent activation of the transcription factor NF-κB and pro-inflammatory cytokine secretion, could be impaired in CD [4]. In fact, some NOD2 mutations known to recur in individuals who develop CD are associated with decreased IL-8, tumor necrosis factor (TNF)-α, IL-6 and anti-inflammatory

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IL-10 secretion by monocytes and dendritic cells. Mutations of NOD2 are also associated with defective production of α-defensin by the intestinal mucosa, and thus in the diminished local anti-microbial defense response. Regulatory T cells (Treg), a T-cell subset implicated in the maintenance of immune tolerance, have been shown to contribute to the cure of experimental colitis [5], underpinning their potential pathogenetic and therapeutic role in inflammatory bowel disease (IBD) in humans.

In this pathogenetic context, stem cell and other cellular-based therapies aimed at the immune response represent current and promising therapeutic modalities for the treatment of resistant/persisting cases of CD.

High-Dose Immunosuppression and Hematopoietic Stem-Cell Transplantation

At least 15% of CD patients suffer from a chronic, unremittingly active course of the disease, and therefore from an impaired health-related quality of life that impacts not only their employment status but also their mental and physical functioning. The consequence of this chronic alteration of health is a life expectancy at 20 years that is < 80% whereas in the normal control population it is about 86%. For patients with chronic active CD, there is no effective cure. Patients who are steroid-refractory or steroid-intolerant may have a chance of better disease control by immune modulators and biological agents, such as infliximab or adalimumab, but the beneficial effects are tempered by the potential complications of these drugs, such as life-threatening infections or the development of malignancies. The risk of immunosuppression-related complications produced by maintenance treatments may be theoretically overcome by the induction of intense, high-dose, single-shot immunosuppression achieved with alkylating agents, total body irradiation (TBI), and anti-thymocyte globulin (ATG). The accompanying immuno-hematopoietic suppression could be rapidly rescued by autologous stem cell transplantation, using a treatment modality similar to that employed in lymphoproliferative disorders or leukemia. In the case of CD, as in other autoimmune disease such as scleroderma, multiple sclerosis, and systemic lupus erythematosus, immunosuppression by high-dose cyclophosphamide, TBI, and ATG produces a powerful abrogation of immune effector responses—including those clones conferring autoreactivity or immune dysregulation– as well as an immune space where regenerating T/B cells derived from re-infused autologous stem cells can reconstitute a renewed and controlled response to endogenous and foreign molecules [2,6]. In case of an occurrence of overt autoimmune disease, an oligoclonal repertoire of T-cell effectors is usually seen, as judged by T-cell receptor analysis. This immune alteration is one of the cellular mechanisms underlying the prevalent and persistent lymphocyte response to a limited number of endogenous molecules and, consequently, to the destruction of autologous cells or cell components. The accumulated experience on autologous stem cell transplantation after high-dose immunosuppression by chemotherapy and/or radiotherapy indicates that the regenerating immune system produces a sequential post-transplant response that starts with an increase of natural killer (NK) cell regeneration and proceeds with an

increase in CD4/CD8 cells and then B cells [7]. Subset analysis of regenerating T cells 3–4 weeks after transplant shows that nascent immune cells have a prevalent naive profile, with the reduced presence of memory cell elements. This may be the beginning of renewed modulation of the immune response through a new generation of T cells and, later, of B cells [6]. Usually, 6 months after autotransplantation, immune peripheral cell counts return to within normal values and normal immunoglobulin levels are observed in the peripheral blood of these patients. Reportedly, after immunosuppression and during brief- and long-term immune-regeneration, some autografted patients suffering from malignancy and concomitant autoimmune disease have experienced remission from their autoaggressive disease, with long-lasting control [8]. From a theoretical point of view, the use of an allograft after high-dose immunosuppression could provide a radical abrogation of the dysregulated immune status of patients with autoaggressive disease via immune replacement by donor immunity; however, this approach requires a HLA-matched sibling donor and produces graft versus host disease (GVHD) in 20–30% of grafted patients, translating into a high treatment-related mortality. The "autologous" approach, by contrast, carries the risk of re-infusing autoreactive clones with the graft, despite possible manipulation of the latter through clinical-scale T-cell depletion prior to re-infusion; nonetheless, this treatment modality has a transplant-related mortality consistently below 10% for most autografted diseases. On a cellular basis, allogeneic transplantation has the additional advantage of replacing both antigen-presenting cells and effector lymphocytic cells by donor cells instead of the sole regeneration of effector cells, as is the case with autografts from autologous re-infused progenitors. Hence, although less feasible and more toxic than autotransplantation, allotransplantation may represent a chance of cure for progressive, treatment-refractory autoimmune disorders affecting patients within the sixth decade of life. The current practice in allogeneic transplantation precludes the use of matched unrelated donors or cord blood in autoimmune disease due to the prevalent and appropriate use of these sources of stem cells in malignancies, in which allografts are the standard of cure. This fact translates into a no more than 20% chance to identify matched sibling donors in the relatives of patients with these acquired immune disorders. Collectively, the potential occurrence of fatal GVHD and the frequent unavailability of matched donors render the allograft approach as a "not recommended" treatment modality in these disorders, albeit of continued interest. In this context, there have been sporadic observations of clinical remission of CD in some patients with concomitant leukemia or other hematological malignancies who underwent allogeneic transplantation for the cure of their primary disease. This confirms the hypothesis that replacement of the host immune system by nascent donor immune effectors and myeloid cells can reconstitute immune tolerance to endogenous gut mucosal components, overcoming dysregulation and the loss of tolerance to self-antigens. Available data on the allogeneic approach confirm that there are no reported cases of allogeneic stem cell transplantation performed for the primary treatment of CD. However, there are some patients reported in the literature who had CD and underwent allogeneic transplantation for hematological disorders [9]. Ditschkowski et al. reported a case series in which 10 out of 11 patients experienced remission from IBD following

allogeneic transplantation for hematological malignancy, with a median follow-up time longer than 30 months [10]. Seven out of the 11 patients with IBD had CD. All patients received cyclophosphamide as part of different conditioning regimens, and all except two received TBI. There was one transplant-related death at 10 months, from opportunistic infection. Nonetheless, this study is clearly limited with respect to furthering our understanding of the effectiveness of allotransplantation to produce sustained CD control, since all patients evaluable at follow-up had received immunosuppressive therapy at that time. Conversely, Lopez-Cubero et al. reported a case series of six patients with CD who underwent allogeneic marrow transplantation for leukemia. Four of the patients were free of CD in the absence of immunosuppressive treatment at a follow-up of more than one year [11]. Collectively, of the 14 reported cases of allogeneic bone marrow or stem cell transplantation in patients with CD, 11 patients achieved remission during a median follow-up time of 7 years. Of the seven patients evaluated for CD after immunosuppressive drugs had been suspended, six remained in remission of CD. Among the 14 patients, two transplant-related deaths were reported.

At variance with allogeneic transplantation, autologous stem cell transplantation has been used as primary treatment for CD and for other autoimmune diseases, as noted above. Particularly, autoimmune disorders, including CD, may benefit from full immune re-setting of T/B cells by high-dose immunosuppression and autologous stem cell transplantation. In this case, the patients coincide with the donors of myeloid/immune precursors, which, in this autologous setting, obviously do not generate any adverse immune events such as GVHD or graft rejection. Immune-hematopoietic reconstitution by autologous cells is rapid and the absence of serious immune events reduces the mortality associated with this procedure to < 5%. Hence, the theoretical feasibility of this treatment modality is near 100% in patients less than 65 years of age and not benefiting from the availability of a fully matched sibling donor. A recent report on autologous transplantation in patients with systemic lupus erythematosus clearly and convincingly demonstrated that autologous transplantation following high-dose immunosuppression produces the depletion of autoreactive immunological memory, as reflected by the disappearance of pathogenic anti-double-stranded DNA antibodies as well as protective antibodies in serum, and a fundamental resetting of the adaptive immune system. The latter includes the reappearance of $CD31^+CD45RA^+CD4^+$ T cells (recent thymic emigrants) with a doubling in absolute numbers compared with age-matched healthy controls at 3-year follow-up, the regeneration of thymic-derived $FoxP3^+$ regulatory T cells, and the normalization of peripheral T-cell receptor repertoire usage. Moreover, patients exhibited normalization of previously disturbed B-cell homeostasis, with recovery of the naive B-cell compartment no later than 1 year after transplantation.

In the context of this biological scenario, a significant number of patients with refractory or progressive CD treated by high-dose immunosuppression and autologous stem cell transplantation have been reported in the recent literature, including 19 patients who had both CD and autologous transplantation. These patients had previously failed standard medical treatment including infliximab. The first case was reported in 1993 [12]. Although the follow-up period was short, the patient main-

tained remission from CD at 6 months. Three additional patients who underwent transplantation for concomitant malignancies also remained in clinical remission. Recently, Oyama and colleagues reported 12 patients with refractory CD who received autologous hematopoietic cell transplantation [13]. All of these patients had failed standard medical treatment, including infliximab, and had a minimal Crohn's disease activity index (CDAI) ≥ 250. Autologous stem cell mobilization was obtained by cyclophosphamide and filgrastim, conditioning was accomplished by cyclophosphamide and equine ATG, and peripheral blood stem cells were T-cell depleted. After a median follow-up of 18.5 months, 11 of 12 patients had continuous remission of their disease (CDAI ≤ 150). One patient experienced a relapse of CD at 15 months post-transplantation. All patients tolerated transplantation well in the absence of transplant-related deaths. Collectively, 18 out of 19 patients obtained clinical remission over a median follow-up time of 20 months. Only two patients were on any ongoing medications, again suggesting that autologous transplantation results in CD remission without any need for continued medications. To date, there has been no mortality linked to transplant-related complications.

T-Cell Regulation by Growth Factors and Immune-Regulatory Cell Therapies

Functionally and phenotypically heterogeneous Treg cell populations have been recently described in animals and in humans. $CD4^+CD25^+$ cells, also referred to as naturally occurring Treg cells, are the most thoroughly characterized Treg cell subset. Naturally occurring Treg cells express the FoxP3 transcription factor and suppress pathogenic T cells through a cell-contact-dependent mechanism [14]. The expression of surface membrane transforming growth factor (TGF)-β may contribute to the regulation of undesired T-cell responses by naturally occurring Treg cells. Very recently, a previously unappreciated ability of Treg cells to kill B lymphocytes was reported, suggesting that defects in the Treg compartment favor the expansion of antibody-producing B-cell populations and thus contribute to autoimmune phenomena [15].

Functionally specialized subsets of $CD4^+$ T cells regulate intestinal immune responses [16]. For instance, $CD4^+CD25^+$ Treg cells can prevent disease in the T-cell transfer model of colitis. The same population of $CD4^+CD25^+$ Treg cells can cure established colitis, suggesting that these cells can be therapeutically exploited in the treatment of human IBD. The human disease IPEX (immune dysregulation, polyendocrinopathy, enteropathy, X-linked syndrome) is characterized by severe enteropathy, underscoring the importance of $CD4^+CD25^+Foxp3^+$ Treg cells in the control of destructive intestinal immune responses. Type 1 Treg cells (Tr1 cells) have also been shown to protect from autoimmune and inflammatory diseases, including experimental colitis. At variance with $CD4^+CD25^+$ Treg cells, Tr1 cells lack unique surface markers and can be operationally defined by their ability to release IL-10 and to suppress pathogenic T cells through a cell-contact-independent mechanism.

$CD4^+CD25^+Foxp3^+$ Treg cells can be visualized in the colonic lamina propria of

normal mice, suggesting a role for these cells in the prevention of aberrant responses to harmless intestinal antigens. They also accumulate in the intestine during cure of experimental colitis, where they are in close contact with $CD11c^+$ dendritic cells (DC). These studies suggest that Treg cells require interaction with colonic DC for their activation or expansion, or that they act on colonic DC to modulate their expression of pro-inflammatory cytokines or costimulatory molecules. Surprisingly, $Foxp3^+$ Treg cells have been shown to be over-represented in the lamina propria of IBD patients compared with healthy controls. This indicates that chronic intestinal inflammation is a result of impaired Treg function or the resistance of pathogenic effector cells to Treg-mediated suppression, in accordance with previous findings in other autoimmune/inflammatory conditions such as rheumatoid arthritis.

Based on pre-clinical studies, several theoretical approaches with the aim to restore Treg number and/or function in human IBD can be envisaged, including vaccination with tolerogenic DC [17], infusion of ex-vivo-expanded Treg cells [5], and administration of individual growth factors with the ability to foster Treg differentiation in vivo [18] (Fig. 13.1). DC were thought to be mainly involved in the initiation of autoaggressive T-cell responses. More recently, a role for DC in mediating peripheral T-cell tolerance has emerged [19]. Specifically, growth factors such as G-CSF (granulocyte colony-stimulating factor) [20], HGF (hepatocyte growth factor)

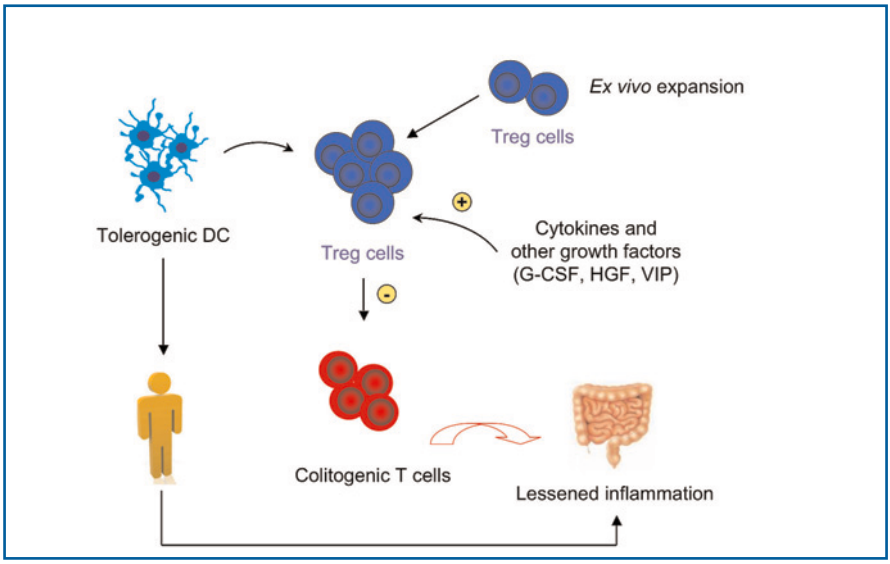

Fig. 13.1 Theoretical approaches to immune-based therapies in human inflammatory bowel disease (IBD). Dendritic cell (DC) preparations for tolerance induction have been generated in vitro with growth factors and cytokines, including G-CSF, HGF, and VIP. When administered to mice with experimental colonic inflammation, tolerogenic DC have shown therapeutic potential, inducing IL-10-secreting Treg cells with suppressive activity against colitogenic T cells [17]. Either freshly isolated or in-vitro-expanded Treg cells have been reported to ameliorate colitis [5], suggesting that the restoration of Treg numbers and/or function has therapeutic potential in human IBD

[21] and VIP (vasoactive intestinal peptide) [17] have been shown to differentiate tolerogenic DC preparations both in vitro and in vivo, suggesting that vaccination with tolerogenic, in-vitro-differentiated DC can be of therapeutic benefit in inflammatory/autoimmune conditions.

Several recombinant human growth factors have been reported to affect the number and/or function of Treg cells. For instance, G-CSF differentiates human Tr1 cells, with a remarkable ability to produce bioactive IL-10 [22]. This has been recently applied to the treatment of human IBD. A 28-day treatment with G-CSF skewed the immune response towards a regulatory profile in nine patients with CD [18]. During the G-CSF treatment period, six patients reported improvement in their CDAI score, achieving a clinical response (4 patients) or remission (2 patients). IL-10 production from memory T cells was significantly higher in the group of G-CSF-responders than in non-responders, suggesting in vivo polarization towards a Tr1-like phenotype. Also, plasmacytoid DC, defined as $CD123^+$ cells, were over-represented in G-CSF-responders compared with non-responders, both in the peripheral blood and in the lamina propria [18]. Finally, a non-significant increase in $CD25^+FoxP3^+$ cells in the lamina propria of responding patients at the end of G-CSF treatment was also documented. This interesting study provides convincing proof of principle in favor of the immune modulatory effects of human cytokines that could be exploited to achieve better cure rates for patients with IBD through their effect on the Treg cell compartment.

TNF-α antagonism has been reported to foster Treg cell number and function in pediatric IBD [23]. This potentially useful effect has been attributed to the reversal of inhibitory actions of TNF-α on FoxP3 expression by Treg cells [24]. TNF blockade with infliximab may restore Treg cell numbers also in adult patients with CD (Rutella S, Bonanno G, Guidi L; unpublished observations; 2009), pointing to TNF-α as a multifaceted target for molecularly based therapeutic approaches.

Tissue Engineering by Mesenchymal Stem Cells in Crohn's Disease Complications

Mesenchymal stem cells (MSC), as defined by The International Society for Cellular Therapy, are plastic-adherent cells with a specific surface phenotype that have the capacity to self-renew and to differentiate into cell types of mesodermal, ectodermal, and endodermal origin [25]. The ability of MSC to differentiate into these cell types results in a wide range of potential therapeutic applications. MSC were initially isolated from the bone marrow but other tissue sources have since been investigated, including adipose tissue and placenta. The biological characteristics of MSC are also exhibited by umbilical cord matrix stromal cells (UCMSC), which are derived from umbilical cord Wharton's jelly, the loose connective tissue surrounding the umbilical vessels. UCMSC can be easily collected from umbilical cords obtained from either vaginal delivery or caesarean section, and easily isolated and expanded in culture. These cells can be transplanted across major histocompatibility complex (MHC) bar-

riers without immune rejection, allowing a single production run to be used for multiple patients and numerous clinical sites. The capacity of UCMSC to maintain their expansion and differentiation potential even after cryopreservation permits their banking. Other readily accessible sources are human subcutaneous adipose tissue depots, which constitute a potential source of adipose-derived, multipotent MSC and hold promise for a range of therapeutic applications. Cells used for regenerative medicinal applications should meet the following criteria:

1. they must be found in abundant quantities (millions to billions of cells);
2. they must be harvested by a minimally invasive procedure;
3. they must differentiate along multiple cell lineage pathways in a regulatable and reproducible manner;
4. they must be safely and effectively transplanted to either an autologous or allogeneic host;
5. they must be manufactured in accordance with current Good Manufacturing Practice guidelines.

Management of fistulas in patients with CD continues to present an extremely challenging problem, because they often do not respond to currently available treatments. Fistulas and their recurrence are a distressing complication that significantly reduces the quality of life of affected patients. The failure of fistulas to heal is probably the result of the suboptimal quality of the tissues affected by CD. Administration of MSC has been reported to be of therapeutic value in patients with CD.

In a phase I clinical trial [26], the feasibility and safety of autologous adipose stromal stem cell transplantation for the treatment of unresponsive Crohn's fistulas were evaluated in four patients. From a phenotypical standpoint, MSC expressed vimentin, CD117, and CD90 but stained negatively for neuroectodermal antigens such as S100 protein and keratin. This is the first clinical trial to use autologous stem cells obtained from a lipoaspirate. The patients were selected according to the following inclusion criteria: older than age 18 years; diagnosis of CD at least 5 years before the trial; and presence of one or more complex Crohn's fistulas (enterocutaneous fistula, suprasphincteric fistula, and/or rectovaginal fistula) that had been unresponsive to medical treatment and unsuccessfully treated by classic surgery at least twice. Adipose-tissue-derived autologous MSC were obtained by liposuction under local anesthesia and general sedation. For clinical purposes, the cells were used after three or fewer in vitro passages, and for subsequent injections they were cryopreserved and thawed before implantation. The trial showed the complete histological healing of 75% of patients undergoing this treatment, indicating that these cells are safe for the treatment of fistulas in CD. Moreover, because the cells were autologous, no ethical conflicts were identified by the Ethics Committee.

A preparation for intravenous infusion of bone-marrow-derived MSC (Prochymal) has been recently developed at Osiris Therapeutics Inc (http://www.osiristx.com/products_prochymal.php) [27]. In a prospective, randomized, open-label phase II clinical trial, patients with moderate-to-severe CD who failed previous treatments with steroids, infliximab, and other immune-suppressive regimens received two intravenous injections of MSC. Clinical improvement occurred rapidly, with an average CDAI reduction of 62 points by day 7 of treatment.

MSC therapy was well tolerated and no severe adverse events were attributed to Prochymal. The FDA has recently allowed Prochymal to proceed to a phase III clinical trial. After the approval of Prochymal for GVHD, treatment of CD with Prochymal is the second indication to MSC therapy.

These results provide convincing proof of principle favoring the immune regulatory effects of MSC and strongly encourage the design of further clinical trials [28].

Concluding Remarks and Future Perspectives

Cell and stem-cell therapies offer new opportunities for therapeutic intervention in a wide series of human diseases, including cancer, autoimmune disease, and chronic inflammatory conditions. These approaches may be classified in two categories: (1) cell transfer of immune-active effectors or cells with regenerative capacity from autologous or allogeneic sources, and (2) modulation and/or reactivation of immune and/or regenerative functions by resident cell effectors.

As described in this chapter, patients with CD may benefit from both approaches through the reactivation of specific cell functions [2]. Specifically, hematopoietic cells and MSC may determine tolerance induction to autologous gut cell determinants or the regeneration of lost tissues, respectively, in progressive CD and in CD complications. Moreover, the induction of tolerance by T-cell regulation may occur through resident T-cell polarization following the systemic administration of growth factors or the infusion of clinical-grade preparations of MSC, which evoke a regulatory profile in relevant cell effectors. Further biological and clinical research in this field will hopefully define the effectiveness of transplant and non-transplant stem-cell and cell therapies in CD and other autoimmune diseases in the near future.

References

1. Yamamoto-Furusho JK, Korzenik JR (2006) Crohn's disease: innate immunodeficiency? World J Gastroenterol 12:6751-6755
2. Gersemann M, Wehkamp J, Fellermann K, Stange EF (2008) Crohn's disease—defect in innate defence. World J Gastroenterol 14:5499-5503
3. Shergill AK, Terdiman JP (2008) Controversies in the treatment of Crohn's disease: the case for an accelerated step-up treatment approach. World J Gastroenterol 14:2670-2677
4. Strober W, Kitani A, Fuss I, Asano N, Watanabe T (2008) The molecular basis of NOD2 susceptibility mutations in Crohn's disease. Mucosal Immunol 1 Suppl 1:S5-9
5. Mottet C, Uhlig HH, Powrie F (2003) Cutting edge: cure of colitis by CD4+CD25+ regulatory T cells. J Immunol 170:3939-3943
6. Muraro PA, Douek DC, Packer A et al (2005) Thymic output generates a new and diverse TCR repertoire after autologous stem cell transplantation in multiple sclerosis patients. J Exp Med 201:805-816
7. Rutella S, Rumi C, Laurenti L et al (2000) Immune reconstitution after transplantation of autologous peripheral CD34+ cells: analysis of predictive factors and comparison with unselected progenitor transplants. Br J Haematol 108:105-115

8. Alexander T, Thiel A, Rosen O et al (2009) Depletion of autoreactive immunologic memory followed by autologous hematopoietic stem cell transplantation in patients with refractory SLE induces long-term remission through de novo generation of a juvenile and tolerant immune system. Blood 113:214-223
9. Leung Y, Geddes M, Storek J, Panaccione R, Beck PL (2006) Hematopoietic cell transplantation for Crohn's disease; is it time? World J Gastroenterol 12:6665-6673
10. Ditschkowski M, Einsele H, Schwerdtfeger R et al (2003) Improvement of inflammatory bowel disease after allogeneic stem-cell transplantation. Transplantation 75:1745-1747
11. Lopez-Cubero SO, Sullivan KM, McDonald GB (1998) Course of Crohn's disease after allogeneic marrow transplantation. Gastroenterology 114:433-440
12. Burt RK, Traynor A, Oyama Y, Craig R (2003) High-dose immune suppression and autologous hematopoietic stem cell transplantation in refractory Crohn disease. Blood 101:2064-2066
13. Oyama Y, Craig RM, Traynor AE et al (2005) Autologous hematopoietic stem cell transplantation in patients with refractory Crohn's disease. Gastroenterology 128:552-563
14. Shevach EM (2002) CD4+CD25+ suppressor T cells: more questions than answers. Nat Rev Immunol 2:389-400
15. Lim HW, Hillsamer P, Banham AH, Kim CH (2005) Cutting edge: direct suppression of B cells by CD4+CD25+ regulatory T cells. J Immunol 175:4180-4183
16. Bluestone JA, Thomson AW, Shevach EM, Weiner HL (2007) What does the future hold for cell-based tolerogenic therapy? Nat Rev Immunol 7:650-654
17. Gonzalez-Rey E, Delgado M (2006) Therapeutic treatment of experimental colitis with regulatory dendritic cells generated with vasoactive intestinal peptide. Gastroenterology 131:1799-1811
18. Mannon PJ, Leon F, Fuss IJ et al (2009) Successful granulocyte-colony stimulating factor treatment of Crohn's disease is associated with the appearance of circulating interleukin-10-producing T cells and increased lamina propria plasmacytoid dendritic cells. Clin Exp Immunol 155:447-456
19. Rutella S, Danese S, Leone G (2006) Tolerogenic dendritic cells: cytokine modulation comes of age. Blood 108:1435-1440
20. Rutella S, Bonanno G, Pierelli L et al (2004) Granulocyte colony-stimulating factor promotes the generation of regulatory DC through induction of IL-10 and IFN-a. Eur J Immunol 34:1291-1302
21. Rutella S, Bonanno G, Procoli A et al (2006) Hepatocyte growth factor favors monocyte differentiation into regulatory interleukin (IL)-10++IL-12low/neg accessory cells with dendritic-cell features. Blood 108:218-227
22. Rutella S, Pierelli L, Bonanno G et al (2002) Role for granulocyte colony-stimulating factor in the generation of human T regulatory type 1 cells. Blood 100:2562-2571
23. Ricciardelli I, Lindley KJ, Londei M, Quaratino S (2008) Anti tumour necrosis-a therapy increases the number of FOXP3 regulatory T cells in children affected by Crohn's disease. Immunology 125:178-183
24. Valencia X, Stephens G, Goldbach-Mansky R et al (2006) TNF downmodulates the function of human CD4+CD25hi T-regulatory cells. Blood 108:253-261
25. Brooke G, Cook M, Blair C et al (2007) Therapeutic applications of mesenchymal stromal cells. Semin Cell Dev Biol 18:846-858
26. Garcia-Olmo D, Garcia-Arranz M, Herreros D et al (2005) A phase I clinical trial of the treatment of Crohn's fistula by adipose mesenchymal stem cell transplantation. Dis Colon Rectum 48:1416-1423
27. Taupin P (2006) OTI-010 Osiris Therapeutics/JCR Pharmaceuticals. Curr Opin Investig Drugs 7:473-481

28. Ascanelli S, de Tullio D, Gregorio C, Azzena G, Occhionorelli S (2007) Autologous fibroblasts transplant after infliximab administration: a new approach in Crohn's perianal fistulas? Brief clinical report. Int J Colorectal Dis 22:1135-1136

Surgical Treatment

14

R. Tersigni

Introduction

Surgery for Crohn's disease (CD) has changed considerably during the past 10 years as a result of the introduction of new models in medical therapy. Despite these advances, even conservatively minded physicians have needed to resort to surgical treatment for 70–90% of patients with CD. Since the disease is incurable, the indications for surgery remain relatively straightforward: failure of medical management and complications of the disease process. Surgery is considered by many gastroenterologists as a last resort and reserved essentially for patients in whom medical therapy has failed. However, this carries an increased risk for the patients, because the disease can become more complicated [1]. For this reason many surgeons opt for the elective surgical treatment of CD, advising early resection because of the fear that a delayed approach is associated with serious and frequent complications. It should be pointed out that early surgery prolongs clinical remission compared to late surgery, but the surgical recurrences and the natural history of the disease are not modified.

Small-Bowel Disease

Indications

The failure of medical management remains the most common indication for surgery in most series of patients with CD of the small bowel. Failure is defined by: (1)

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symptoms that cannot be controlled or which progress despite medical treatment, (2) increased number of side effects of the drugs, (3) inability of the patient to maintain compliance with a medical regimen. The extraintestinal manifestations of CD are another indication for surgery and may occur in as many as 25% of these patients [2]. Disorders of the skin, mouth, eye, and joints are common in CD and tend to parallel intestinal disease activity. As such, surgical resection of the diseased bowel tends to ameliorate these manifestations as well. Hepatic, vascular, hematologic, pulmonary, cardiac, and neurologic extraintestinal manifestations usually proceed independently from the intestinal disease. Biliary and urinary lithiases are complications due to altered intestinal absorption, and can be improved by surgical resection.

Intestinal obstruction represents a frequent complication of small-bowel CD. Acute obstruction can occur due to a primary stricture or series of strictures. Healthy, non-diseased bowel may be mechanically obstructed as part of an inflammatory mass or fistula. Acute obstructions are more likely to be the result of active inflammation and will often resolve with medical management. Conversely, chronic obstruction, which is usually the result of a fixed fibrostenotic lesion, tends to require surgical management. Surgery usually involves a resection of the diseased segment, but other options include intestinal bypass, ileostomy, and strictureplasty. Fistulas with associated abscess or stricture are another common complication of small-bowel CD that requires surgery. In a study of 1379 patients, Michelassi et al. found fistulas in 35% of surgically managed patients. However, fistula was the primary surgical indication in only 6.3% of these patients [3]. Enteroenteric fistulas are the most common type of fistula and may be relatively asymptomatic unless a large segment of intestine is bypassed or complications of obstruction arise. Broe et al. [4] published that 40% of patients with fistula, initially managed non-operatively, eventually required surgery within 1 year, usually secondary to medical intractability. The surgical management of enteroenteric fistulas generally involves resection of the primary site that has active disease, and simple debridement and primary closure of the secondary site, which is usually normal. Enteral fistulas to the vagina and urinary bladder often require surgery, with treatment following principles similar to those of primary site resection with repair of the secondary site.

Enterocutaneous fistulas deserve special mention as they will occasionally respond to medical therapy. The most promising trials to date have evaluated the use of infliximab [5,6]. Patients with short fistula tracts, exposed bowel mucosa, and high outputs will generally require operative intervention, but this should be delayed until the patient's health and nutritional status have been optimized.

The life-time risk for developing an abscess in CD is estimated to be around 25% [7]. Radiographic techniques can be used to drain abscesses percutaneously. However, some controversy exists as to whether abscesses must eventually be followed up with surgical resection of the associated diseased bowel. One study has shown that percutaneously drained abscesses recur more frequently than those that are surgically drained abscesses (56% vs. 12% respectively) [8]. However, Gutierrez and colleagues showed, in a study of 66 patients, that only one third who underwent percutaneous drainage of an abscess required surgery at 1-year follow-up [9]. Certainly, if an abscess contains enteric fluid, it is less likely to resolve without surgical resection.

Other less common indications for surgery in small-bowel CD include perforation, bleeding, and cancer. Free perforation is associated with a high mortality if not treated and surgical management should involve resection rather than repair [10]. Massive hemorrhage due to CD is a rare indication for surgery. Other, more common sources of bleeding, such as peptic ulcer disease or diverticulitis, must be actively ruled out. Adenocarcinoma of the small intestine, while rare, is increased 12- to 60-fold compared to the general population [11]. Prognosis is generally poor due to the often advanced stage of disease at the time of diagnosis. Mortality rates at 1 or 2 years have been reported to be 30–60%.

Surgical Strategy

Intestinal resection is the most commonly performed surgical procedure for small-bowel CD. In general, the nature and natural history of CD has resulted in a surgical philosophy of conservatism. Recurrence rates tend to increase with the passage of time and these patients may eventually require multiple resections, each increasing the risk of short-bowel syndrome and its associated metabolic morbidities. Of note, Glehen et al. reported that CD patients start out with a shorter bowel than the normal population [12]. The surgeon should consider the decision very carefully before advising further resection for patients who have already lost part of their small bowel. However, patients can survive in reasonably good health with only 1 m of upper small bowel when resection of the gut is done in stages and if the small bowel has been anastomosed to the right or transverse colon.

A number of studies have investigated whether certain technical factors, including resection margin and configuration of the anastomosis, influence the rate of recurrence. Two retrospective studies suggested that "radical" resection resulted in a much lower rate of resection and better quality of life. Krause and colleagues studied 186 patients with margins of uninvolved bowel of < 10 cm or > 10 cm (a radical resection). They reported a 31% recurrence rate and better quality of life with radical resection compared to an 83% recurrence rate in the other group [13]. Softley and colleagues used a 4-cm histological margin and found that impingement on this margin resulted in a ten-fold increase in recurrence [14]. However, the best available evidence that large resection margins do not decrease the rate of recurrence is based on a study performed by Fazio [15], in which 131 patients were randomized to resection with margins of either 2 cm of uninvolved bowel (75 patients) or 12 cm (56 patients). Although the rate of recurrence was lower in the group with more extensive resection (25 vs. 18%), this difference did not achieve statistical significance. In Fazio's study, grossly normal resection margins were used. Hamilton and colleagues studied the role of frozen-section examination of the bowel wall at the resection margins during surgery and found there was no difference in reoperation or recurrence rates in patients with disease-free margins that were detected histologically or grossly [16]. For CD confined to the ileocecum, limited resection is performed as a means of achieving remission for patients with obstructive symptoms [1]. There is good evidence that extensive resection is no longer necessary and potentially dangerous. The

actual trend is to leave microscopic diseased bowel behind. The risk of short-bowel syndrome is now lower than in the past, and the main cause is multiple operations within a short time span rather than multiple operations over several years for recurrent disease. After intestinal resection, there is a 50% chance that the operated patient will never need to undergo another surgical treatment.

In the presence of active disease with a concomitant abscess, there are three options: (1) percutaneous drainage and delayed resection in patients with obstructive symptoms, (2) percutaneous drainage and medical treatment in the absence of obstruction, and (3) medical treatment followed by resection in patients unfit for percutaneous drainage [1].

The type of anastomosis performed in small-bowel resections has also been speculated to affect recurrence rates. Since fibrostenotic disease is a clinical phenotype of CD, as described by the Vienna Classification [17], it was thought that the larger lumen of a side-to-side anastomosis would be less likely to obstruct and require reoperation [18]. Stapled anastomoses have been reported to be associated with lower morbidity and recurrence rates [19]. However, certain circumstances seem to clearly favor hand-sewn anastomosis, particularly when the thickened (but grossly disease-free) bowel that is joined exceeds the specifications of a bowel stapler [20].

Recently, Simillis et al. [21] carried out a meta-analysis of anastomotic configuration in CD as reported in two randomized and six non-randomized studies, with a total of 712 anastomoses, of which 53.8% were hand sewn end-to-end and 46.2% had other configurations (stapled side-to-side, end-to-side, side-to-end, and stapled end-to-end). The end-to-end anastomosis showed the higher rate of anastomotic leak. Side-to-side anastomoses may lead to fewer postoperative complications, shorter hospital stay, and fewer perianastomotic recurrences. Stapled or hand-sewn wide lumen (at least 5 cm) functional end-to-end anastomoses are actually the preferred technique, but further randomized, controlled trials should be performed.

Recurrence rates following resection remain high and although not all symptomatic recurrence requires surgery, it has been reported that surgical re-intervention occurs in 25–35% of patients at 5 years and 40–70% at 15 years [22]. However, several trials have been done to evaluate patient and disease factors as well as medication regimens, which may decrease recurrence. Yamamoto et al. recently published a comprehensive systematic review of factors affecting CD recurrence after surgery [23]. They concluded that the cessation of smoking seems to be the most consistent factor in reducing recurrence. While several other factors (including 5-ASA use, immunosuppressant drugs, wider anastomosis, and disease duration) may affect recurrence rates, further studies are still required. Post-surgical recurrence in CD is still largely unpredictable.

When preservation of intestinal length is an issue, strictureplasty represents an alternative to resection and can reduce the risk of short-bowel syndrome. Indications for strictureplasty include short fibrous strictures, diffuse involvement of the small bowel involving multiple strictures, a stricture in a patient with short-bowel syndrome or a history of multiple prior small-bowel resections, patients with a rapid recurrence of CD manifested as obstruction, or patients with duodenal involvement. Conventional strictureplasties should not be done in patients with multiple strictures

within a short bowel segment, a long (≥ 20 cm) stricture, or stricture close to a site of resection [3]. The largest strictureplasty experience published to date comes from the Cleveland Clinic, in which 698 strictureplasties were performed in 162 patients [24]. The authors of that study cited a recurrence rate at 5 years of 28%, which is similar to published rates of recurrence following resection. In addition to relieving obstruction, there have been reports of disease regression at sites of strictureplasty [25]. Indeed, in the Cleveland Clinic series, documented recurrences only occurred at the previous strictureplasty site in 5% of the patients [24]. Fearnhead and colleagues recently published a long-term follow-up of 479 strictureplasties in 100 patients [26]. Over a mean follow-up of 85 months, overall morbidity was 22.6%, with septic complications (leak, fistula or abscess) occurring in 11.3%. Obstruction occurred in 4.4% of patients while perioperative mortality was 0.6%. Although bleeding from strictureplasty sites was previously cited as a potentially serious problem due to the presence of a suture line in the diseased bowel, the incidence of gastrointestinal bleeding was only 3.8%.

In the ECCO statements [1], conventional strictureplasties (Heinecke-Mikulicz, Finney) are indicated when the length of the stricture is < 10–20 cm. For longer strictures, the alternative to resection, in selected patients, are the so-called unconventional strictureplasties, i.e., a side-to-side intestinal anastomosis without resection [27-30]. Michelassi noted that in patients with closed multiple strictures or long and rigid stenosis conventional strictureplasties may not be technically feasible [28]. Besides, Finney strictureplasty may be associated with a large lateral diverticulum with bacterial overgrowth. Michelassi also reported a side-to-side isoperistaltic strictureplasty in patients with extensive partial intestinal obstruction.

One disadvantage of strictureplasty is that a malignancy of the bowel can be missed. Several case reports have been published of adenocarcinoma arising from CD-related stricture sites. To avoid missing a cancer in a longstanding stricture, it has been suggested that full-thickness biopsies should be taken for frozen section to aid the surgeon in the decision to perform either strictureplasty or resection [31].

Two recent meta-analyses concluded that strictureplasty represents a safe and effective procedure for small-bowel CD, and it has the advantage of protecting against further small-bowel loss [25,32]. Patients with CD undergoing strictureplasty alone may have fewer postoperative complications than those undergoing a concomitant bowel resection; however, surgical recurrences may be higher following strictureplasty than following resection. In patients with multiple strictures in a short segment of the small bowel but who are not at risk for short-bowel syndrome, it is preferable to perform a resection. Phlegmon, abscess, fistulating disease, carcinoma, and active bleeding are contraindications for strictureplasty.

Other surgical options for the treatment of small-bowel CD include bypass operations or ileostomy. Bypass operations have largely been abandoned due to the risk of malignancy and continued disease activity. An ileostomy may be required when enteric anastomosis is unsafe (sepsis, unstable patient, severe malnutrition, chronic immunosuppression) or when small-bowel resection is done in conjunction with colonic or rectal resection.

14

Notes on Surgical Techniques

Strictureplasty

The presence of stenosis must be carefully assessed by observation and palpation. The intestinal lumen size may be evaluated using an 18-F catheter or by the fingers of an expert surgeon. The type of strictureplasty, described in the following, depends on the stricture's characteristics.

1. Heineke-Mikulicz strictureplasty. This technique is used for a short stenotic tract (< 10 cm). An antimesenteric incision is made in the anterior wall of the small bowel. Traction sutures are placed through the proximal and distal corners of the incision. All active bleeding points should be carefully controlled. The longitudinal incision is converted to transverse closure by 4-0 interrupted absorbable sutures (Fig. 14.1). Heineke-Mikulicz strictureplasty is successfully used in case of duodenal stenosis as an alternative to by-pass procedure or resection.

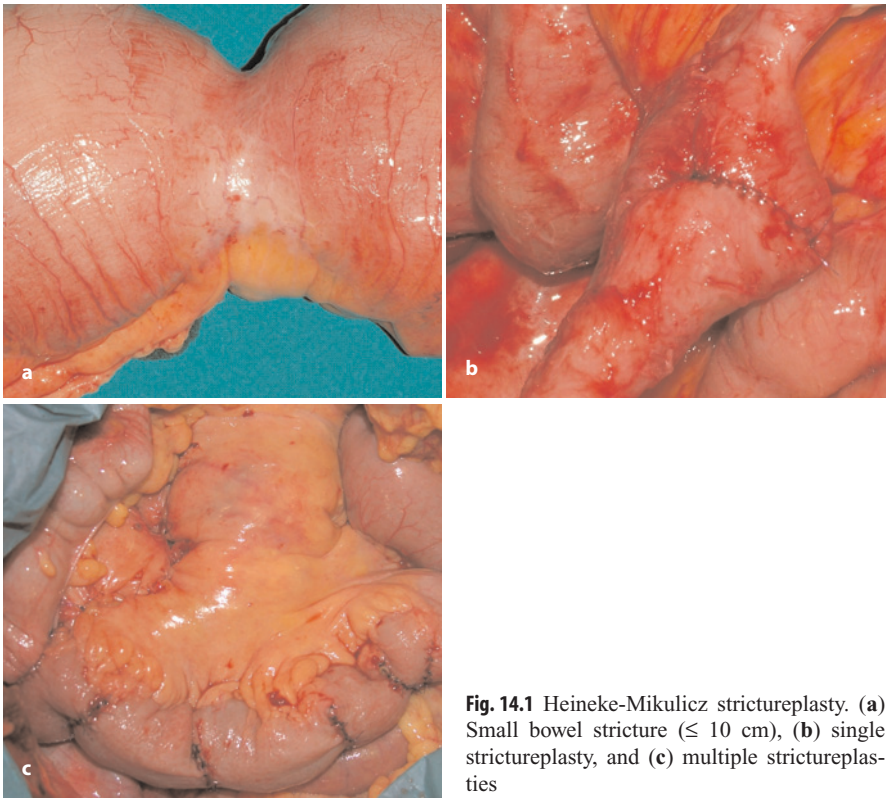

Fig. 14.1 Heineke-Mikulicz strictureplasty. (**a**) Small bowel stricture (≤ 10 cm), (**b**) single strictureplasty, and (**c**) multiple strictureplasties

2. Finney strictureplasty. This procedure is used for longer stenoses (10–20 cm). After an antimesenteric longitudinal incision, the opened bowel segment is bent into a U shape and posterior and anterior layers are closed with continuous 4-0 absorbable sutures (Fig. 14.2).
3. Fazio and Tijandra strictureplasty. This technique is used for two closed strictures. A long longitudinal antimesenteric incision is transversely closed with the Heineke-Mikulicz and the Finney techniques (Fig. 14.3).
4. Michelassi strictureplasty. Multiple strictures, up to 90 cm long, can be treated with this technique. After the intestinal transection and sliding of the proximal and distal intestinal loops, the loops are approximated in a side-to-side isoperistaltic fashion by a layer of non-absorbable stitches. A longitudinal enterotomy is performed and the intestinal ends are spatulated. A formal two-layer enteroenterostomy is then done with non-absorbable sutures (Fig. 14.4).

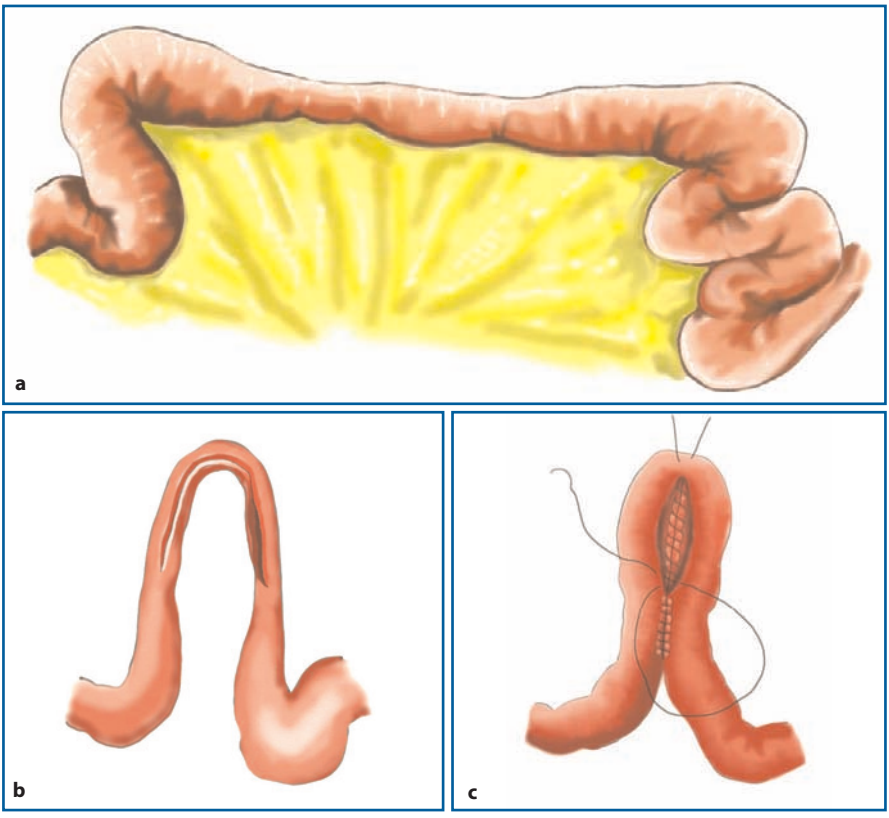

Fig. 14.2 Finney strictureplasty. (**a**) Small-bowel stricture (10-20 cm), (**b**) U-shaped long antimesenteric incision, (**c**) continuous 4-0 absorbable sutures

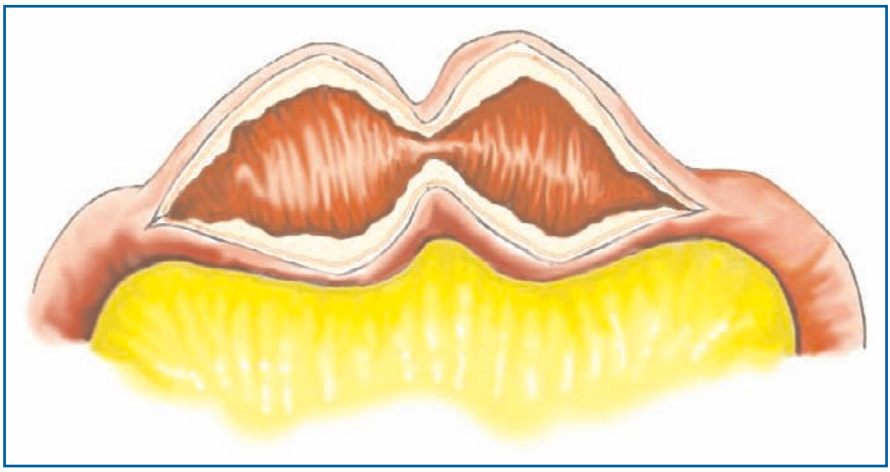

Fig. 14.3 The Fazio-Tjandra strictureplasty is a combination of the Heineke-Mikulicz and Finney procedures used in strictures up to 20 cm in size

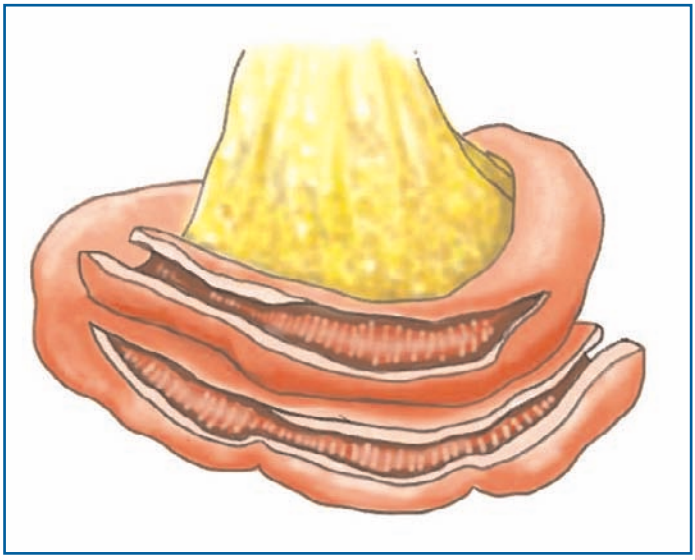

Fig. 14.4 Michelassi strictureplasty

5. Poggioli and Taschieri strictureplasty. This is a side-to-side, disease with disease-free anastomosis. Poggioli and Taschieri independently described a side-to-side enterocolic anastomosis in patients with disease involving the terminal ileum and or the ileocaecal valve (Fig. 14.5).
6. Jaboulay, Judd, Moskel-Walske, Neumayer, Sasaki strictureplasty. New models of strictureplasty have been published in the last years by many surgeons (Figs. 14.6–14.8).

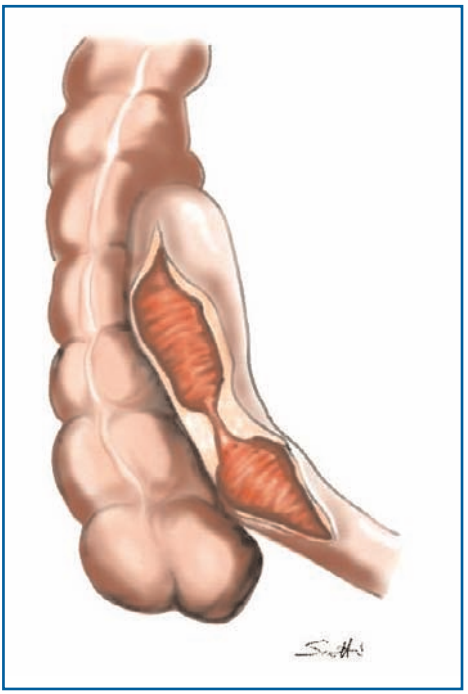

Fig. 14.5 Poggioli and Taschieri strictureplasty

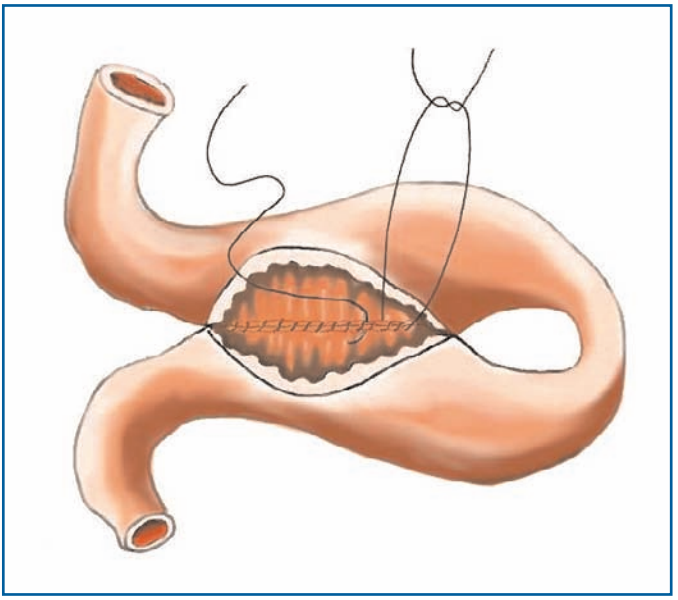

Fig. 14.6 Jaboulay strictureplasty. The loop of the stenotic intestine is positioned in a U shape. Two separate incisions are made in healthy facing sections

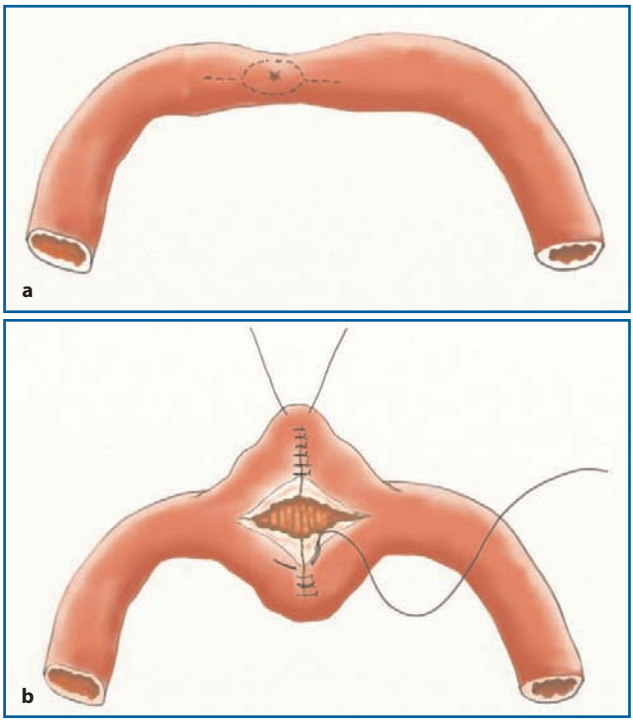

Fig. 14.7 Judd strictureplasty. This procedure is a modification of the Heineke-Mikulicz strictureplasty, used when a fistula is present in a short-segment stenosis

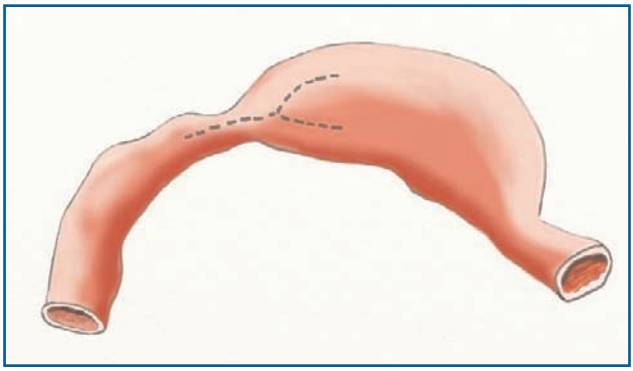

Fig. 14.8 Moskel-Walske-Neumayer strictureplasty. This technique is used in case of intestinal dilatation, when a Heineke-Mikulicz procedure can result in excessive suture-line-tension strictureplasty

Resection of the Small Intestine

A median incision is often used. The involved loop of intestine is mobilized carefully from the peritoneal cavity. The intestinal wall should be resected 5 cm beyond the grossly involved area between two applicants of the gastrointestinal anastomosing (GIA) instrument. Two double staggered staple lines seal the small bowel. The resec-

tion should not extend into the base of the mesentery and pulsating vessels are preserved to nourish the residual intestine. In case of an end-to-end anastomosis, a single layer of interrupted or continuous 4-0 absorbable sutures is made and the mesentery is approximated with continuous or interrupted 4-0 absorbable sutures. At the end, the patency of the stoma is tested manually, with the fingers. Based on the principle of triangulation, an end-to-end anastomosis can be created with three applications of the transverse anastomosis (TA) stapler. An alternative method of lateral anastomosis may be used. After division of the bowel, a row of continuous or interrupted 4-0 absorbable sutures is placed in the external layer. The bowel wall is incised close to the suture line. The posterior layer is closed with continuous or interrupted sutures. The anterior layer is closed with inverting stitches and the anterior serosal layer with interrupted or continuous sutures. A mechanical end-to-end functional anastomosis can be constructed using traction sutures and approximating the antimesenteric borders of the bowel. One fork of the GIA instrument is inserted into each lumen of the bowel segments after which the staples are released and an enteroenterostomy is created. The common opening of both ends of the small bowel is closed with the linear stapler.

Ileocolic Resection

A midline, or right paramedian, or right transverse, or Pfannenstiel incision provides excellent exposure (Fig. 14.9). The inflammatory lesion is inspected and palpated. The entire small bowel should be examined for skip lesions. In high-risk patients

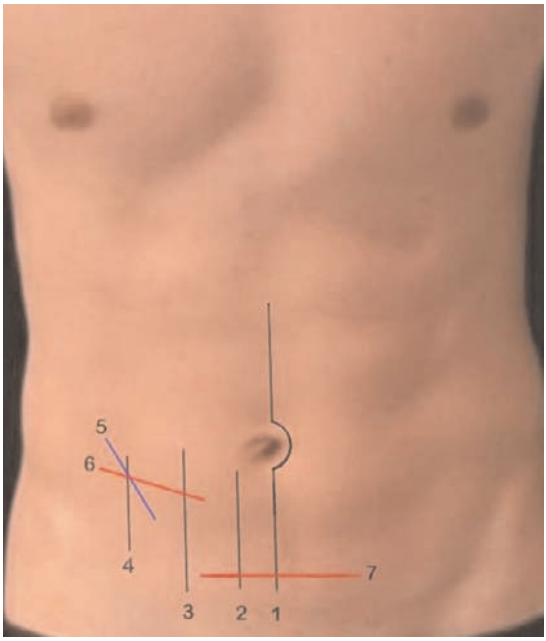

Fig. 14.9 Abdominal incision for ileocolic resection

with extensive involvement, it may be advisable to perform a bypass by constructing an ileocolostomy. Two tractions sutures approximate the antimesenteric border of the ileum and the transverse colon. A 1-cm stab wound is made. The lumen of the bowel and the two forks of the linear stapler are closed and the staples are released, taking care to overlap the ends of the previous staple lines. A bypass with unilateral exclusion with division of the small bowel proximal to the lesion and closure of the distal open end should also be considered. If an ileocolic resection is planned, with the lateral peritoneal attachment and the hepatocolic ligament divided, the large bowel is lifted medially. Care is taken to identify the right ureter and the third portion of the duodenum. The right colon and the terminal ileum are brought outside the peritoneal cavity. The mesenteric vessels are severed and tied; the colon and the diseased ileum prepared for resection 5 cm away from the macroscopic disease. The extent of the right colectomy should vary according to the degree of disease spread into the large bowel. If the disease terminates close to the ileocecal valve, it is perfectly satisfactory to take the distal line of the resection through the caecum. The stumps are closed with two applications of the linear stapler. If an end to side repair is used, the end of the colon is closed by continuous or interrupted suture or by a mechanical suture, reducing the possibility of contamination. If a side-to-end anastomosis is preferred, the closed stump of the small intestine is brought to the open end of the large bowel. In side-to-side anastomosis, the small intestine is brought up adjacent to the anterior taenia of the colon, a posterior serosal layer of continuous or interrupted sutures is placed; an opening is made into the large and the small intestine; posterior and anterior layers are started; and an anterior serosal suture completes the anastomosis (Fig. 14.10). A mechanical side-to-side enterocolostomy can be performed with the linear stapler. After the forks are inserted into the lumen of each bowel segment and the tissue edges aligned, the instrument is closed and the staples released. The ends of the previous staple lines must be overlapped. A direct end-to-end anastomosis can be selected. The discrepancy in the size of the terminal ileum and the colon can be safely overcome by enlarging the oblique division of the terminal ileum and by going slightly deeper into the colonic side (Fig. 14.11). If a functional end-to-end anastomosis is

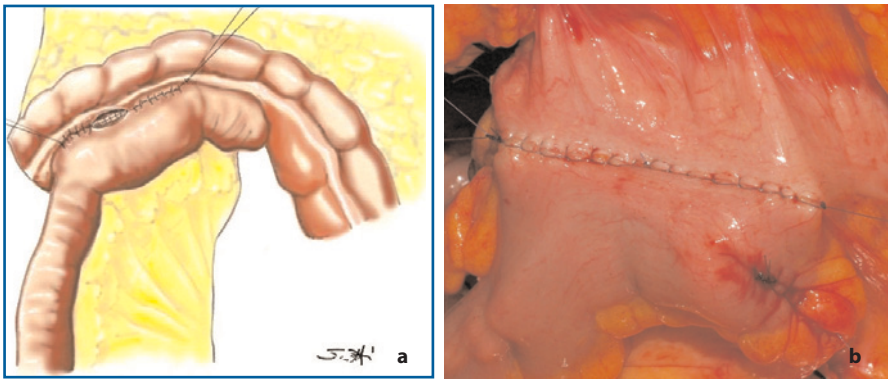

Fig. 14.10 Hand sewn side-to-side ileocolic anastomosis (**a**, **b**)

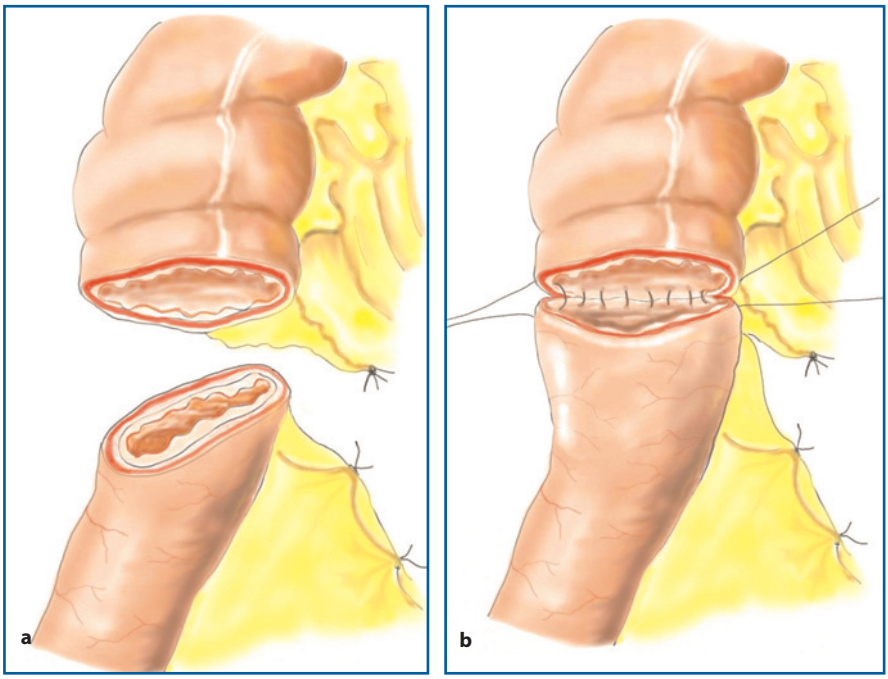

Fig. 14.11 Hand sewn end-to-end ileocolic anastomosis. (**a**) Oblique division of the ileum overcomes the discrepancy in the size of the terminal ileum and the colon; (**b**) direct one- or two-layer anastomosis

planned, a hand-sewn or mechanical anastomosis can be carried out. In case of a mechanical procedure, a 100-mm linear stapler is inserted into each lumen after approximation of the antimesenteric borders of the bowel. Two double-staggered staple lines join the small and the large bowel and the knife creates the stoma. It is important to inspect the anastomotic staple lines for hemostasis, prior to incorporating the common opening within the jaws of the linear instrument (Fig. 14.12). A two-stage resection can be performed in high-risk patients, the lateral anastomosis between the ileum and the colon having been performed as during the first stage.

Large Bowel

Indications

Crohn's colitis occurs in approximately one-quarter of patients although colonic disease is most frequently seen in conjunction with terminal ileal disease. As with small-bowel CD, the indications for surgery in colonic CD can be grouped into com-

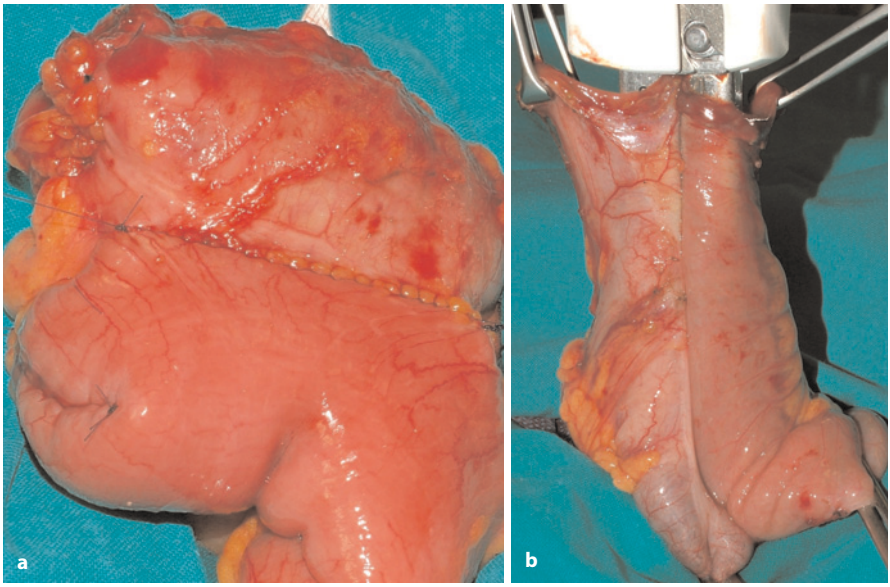

Fig. 14.12 End-to-end functional ileocolic anastomosis. (**a**) Hand sewn end-to-end functional anastomosis; (**b**) A 100-mm linear stapler is inserted into the ileal and colic lumen and the staples released

plications of the disease and failure of medical therapy. Indications specific to colonic disease include the development of dysplasia or colorectal cancer and toxic colitis. The treatment of obstruction and fistula of the colon may differ from that in small-bowel disease. Obstruction from colonic strictures may be present in as many as 17% of patients with Crohn's colitis.

All colonic strictures should be endoscopically biopsied, as 7–10% may contain malignancy. Despite earlier studies suggesting that the risk of colorectal cancer is lower than that seen in ulcerative colitis (UC), the current literature reports that the risk is equivalent. Maykel et al. published a retrospective analysis of 222 patients who underwent resection for colonic CD [33]. Five cases of dysplasia (2.3%) and six of adenocarcinoma (2.7%) were identified. Of note, in only three cases of dysplasia and one of adenocarcinoma was the abnormality identified preoperatively. Older age at diagnosis of CD, duration of disease (> 8 years) and extent of disease (pancolitis) are risk factors for the development of dysplasia or cancer. As with UC, in colonic CD surgery is indicated for a proven malignancy, high-grade dysplasia, or a dysplasia-associated lesion or mass (DALM), but the diagnosis and management of low-grade dysplasia is still controversial. A recent Cochrane review suggested that surveillance endoscopy does not necessarily improve survival despite the earlier detection of cancers [34]. Nonetheless, current recommendations for surveillance in Crohn's colitis mirror those for UC.

Fistulous disease can also occur with CD. However, it is important when assessing fistulas involving the colon to determine the primary site of the fistula. The colonic mucosa should be endoscopically evaluated. Enterocolonic fistulas are often

a result of primary small-bowel disease with the colon only secondarily involved. In these circumstances, it is generally preferable to debride the colonic side of the fistula and close the defect rather than perform a colonic resection. Despite the introduction of new medical therapies such as infliximab, 50–75% of patients with colonic fistulas will require surgery.

Toxic colitis in CD has a similar presentation to that in UC. Operative indications include perforation, lack of improvement with medical management, fulminant colitis, massive hemorrhage, and hemodynamic instability. As with UC, the most common procedure for toxic Crohn's colitis is colectomy and end ileostomy.

Surgical Strategy

The extent of resection in patients with only segmental colonic disease is a subject of some debate. A recent meta-analysis of six studies encompassing 488 patients suggested that there is no significant difference in overall recurrence rate, complications, or need for permanent stoma between segmental colectomy and total abdominal colectomy with ileorectal anastomosis, but the time to recurrence was longer in the total abdominal colectomy group by 4.4 years [35]. This is in contrast to previous studies suggesting that a more extensive procedure (total abdominal colectomy) was associated with a higher recurrence. Bernell published a review of 833 patients and found the 10-year recurrence rate of total abdominal colectomy and ileorectal anastomosis to be 58%, compared to 47% in the segmental colectomy group [36]. Another study found similar recurrence rates in both groups, but better functional outcomes in the segmental colectomy group [37]. In patients with diffuse colitis and proctitis, patients who underwent total proctocolectomy had less medication use 1 year after operation than patients who were treated by total abdominal colectomy or segmental colectomy [38]. For disease of multiple localization, the treatment of choice is colectomy but some authors have proposed separate segmental colic resections as an alternative [37]. Tekkis suggested that more extensive resection, such as total abdominal colectomy, may be more appropriate when two or more segments of colon are diseased [35]. Limited colonic CD treated by limited resection gives a higher rate of recurrence than a proctocolectomy. However, most agree that the avoidance of a permanent stoma usually outweighs the increased risk of recurrence.

According to the ECCO statement for surgical treatment [1], total proctocolectomy with permanent ileostomy is the treatment of choice in patients with severe disease unresponsive to medical therapy. If surgery is necessary for localized colonic disease, a segmental resection is preferable. Two segmental resections can be considered when macroscopic disease affects both ends of the colon, but total colectomy and ileorectal anastomosis may be preferable in the absence of rectal active disease.

Restorative proctocolectomy with ileal pouch-anal anastomosis (IPAA) for CD has most often occurred in the setting of a changed diagnosis following IPAA for UC or indeterminate colitis. The majority of studies have suggested that IPAA is associated with a significantly higher rate of morbidity (including pouch failure, incontinence, and pouchitis) in CD than in UC [39,40]. Although few authors have concluded

that IPAA may be appropriate in highly selected patients with CD, without small-bowel and perianal disease [41,42], the current literature certainly does not support its general use in the CD population. Retrospective analyses show that these patients with CD who underwent restorative proctocolectomy and IPAA are burdened with the most complications, with a reported failure rate of up to 50% [43,44]. However, Regimbeau et al. reported a very small increase in morbidity when restorative proctocolectomy was performed in patients affected by long-standing Crohn's colitis, without small-bowel or perianal disease, compared with patients with UC [45]. Due to the increased risk of cancer, strictureplasty for colonic stenosis in CD is feasible but not recommended [46].

Notes on Surgical Techniques

Loop Ileostomy

With the goal that acute colitis and proctitis will eventually resolve with defunctioning, a loop ileostomy, with a protruding afferent limb and a receding efferent limb, can be constructed. A 4-cm plastic rod through the mesentery supports the intestinal loop. The cut wall of the efferent limb is sutured to the skin. A semicircular row of 4-0 absorbable sutures is inserted between the skin and the everted wall of the afferent limb (Fig. 14.13).

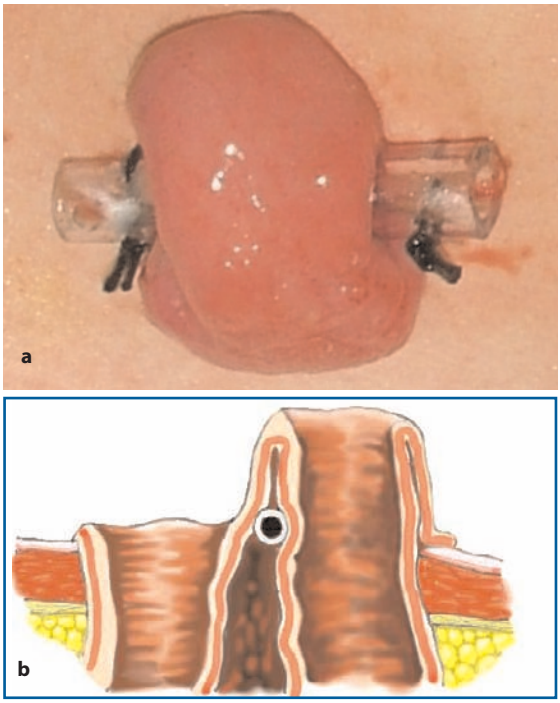

Fig. 14.13 Loop ileostomy. (**a**) A plastic rod supports the loop ileostomy; (**b**) the everted wall of the afferent limb is sutured to the skin

Turnbull Operation

There is no unanimity regarding the best approach in patients with sealed colic perforation and toxic megacolon. Turnbull recommended a loop ileostomy and decompressing blow-hole skin-level transversostomy and sigmoidostomy. The Turnbull operation is confined to very ill patients who have not responded to medical treatment and with frank intestinal perforation, with resulting flooding of the peritoneal cavity by colonic contents.

Colectomy and Proctocolectomy

Colectomy or proctocolectomy is in most cases the best operative solution. In the very high-risk patient, the operation should be carried out in two stages. Removal of the rectum is delayed until the patient's condition is improved. Preservation of the lower rectum by ileorectal anastomosis is controversial and is indicated in the absence of proven rectal disease. The patient is placed in the Lloyd-Davies position, with thighs widely extended. A midline incision provides easy exposure of the entire colon. The dissection is started in the region of the cecum. The right colon is retracted medially as the lateral peritoneum is incised. The terminal ileum and cecum are mobilized outside the peritoneal cavity. The right spermatic vessels and the right ureter are identified. Blunt dissection is used to sweep away the duodenum. The thickened greater omentum is separated from the stomach and the lesser sac entered from the right side. The splenocolic ligament is divided to avoid tearing of the splenic capsule. The splenic flexure and the left colon are freed down to the sigmoid colon. The left gonadal vessels and the right ureter are identified The mobilized colon is drawn outside and all colic vessels in the mesentery can be easily identified. Ileocolic branches, the right colic, middle colic, and inferior mesenteric vessels are ligated. The blood supply of the terminal ileum should be carefully preserved and a few centimeters of ileum denuded in preparation of an ileostomy. The terminal ileum is divided with a linear stapler. The mesentery is divided adjacent to the rectosigmoid, avoiding interference with the sympathetic nerve supply. If removal of the rectum is delayed, the inferior mesenteric artery is spared and only the sigmoid vessels are severed. Finally, the sigmoid colon and the rectum are easily divided with a mechanical instrument. In the presence of extensive pathological changes in the entire large bowel and rectum, a proctocolectomy with ileostomy is performed. A sharp and blunt dissection about the remaining rectum can be carried out, not necessarily as far laterally as in the presence of malignancy. The anus is excised as in the case of abdominoperineal resection for cancer. Usually it is not necessary to go wide on the elevator muscles, because a simple excision of the sphincter muscle can be carried out. Primary closure with drainage suction can be used in the absence of fistula-in-ano or gross contamination. Construction of the ileostomy is of major concern. The Brooke ileostomy is placed in the lower right quadrant. A button of skin is excised, and the rectus muscle fibers separated widely enough to admit two fingers. The ileum is exteriorized through the abdominal wall for at least 6–7 cm. The mesen-

tery is anchored to the peritoneum before the ileostomy is constructed, with a complete reperitonealization of the right paracolic gutter. The terminal ileum should be everted and the mucosa is anchored with interrupted fine absorbable sutures to the skin. The completed ileostomy should extend upward from the skin level at least 3–4 cm (Fig. 14.14). In case of an ileorectal side to end anastomosis (IRA), the purse-string instrument is applied around the rectum and the specimen resected. The circular stapler without anvil is introduced into the anus and advanced to the level of the purse-string suture or to the line of rectal transection. The anvil is passed through an opening in the jejunal wall and the purse-string suture tied. The circular stapler is closed and fired. As an obvious alternative to mechanical anastomosis, a one- or two-layer end to end or side-to-end anastomosis may be employed.

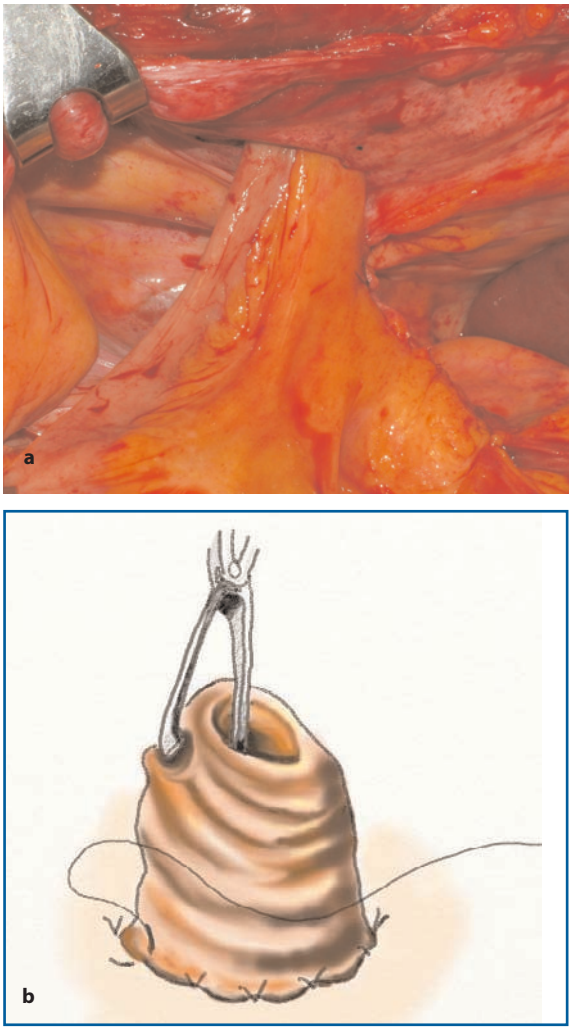

Fig. 14.14 Brooke end ileostomy. (**a**) Complete reperitonealization of the right paracolic gutter; (**b**) the everted wall of the terminal ileum

Restorative Proctocolectomy

As an alternative to proctolectomy with ileostomy, IPAA can be an appropriate procedure in a small selected group of patients. However, patients with CD have high complication and high failure rates after restorative proctocolectomy. Anorectal sepsis and pouchitis are the most common complications and the most important causes of failure.

Two-Stage Procedure

This surgical procedure can be life-saving for the patient during a severe attack of acute colitis. Subtotal or total colectomy, closure of the rectal stump, or sigmoid-rectal mucous fistula and ileostomy is the rule for fulminant Crohn's colitis. In fact, when the tissue of the bowel is so friable that it would be dangerous to try to suture it, the terminal sigmoid can be exteriorized as a suprapubic colostomy or fixed to the anterior abdominal wall by suture. An alternate procedure is a coloproctectomy with a retained rectal stump measuring 2 cm, the upper limit of the future mucosectomy being delimited by the transection line. Most surgeons avoid leaving any retained rectal cuff due to the associated incidence of pelvic sepsis.

Mucosectomy

Anal mucosectomy is always indicated in patients with coexisting dysplasia. The anus is dilated and exposed with Gelpi retractors; the mucosectomy starts a few millimeters above the dentate line and is conducted up to the transected rectal muscle. The length of a dissection ranges from 3 to 5 cm. An alternative procedure is a stapled IPAA 1–1.5 cm above the dentate line, which seems to be associated with an improvement in pouch function compared to the end-sewn anastomosis, with a lower rate of anastomotic complications (Fig. 14.15).

Construction of the Pouch

Most surgeons prefer the J pouch, but occasionally using the S pouch when the apex of a J pouch cannot be brought down to the dentate line. No reliable advantages are found using the bigger W pouch. The mesentery is severed up to the duodenum. It is sometimes necessary to divide some of the tethering secondary branches of the ileocolic artery. Occasionally the main ileocolic artery is tied to allow the pouch to reach the anus without tension. The distal 25–40 cm of ileum is looped, forming a J loop. Two ileal limbs, 12–20 cm in length are held side by side. A linear 100-mm stapler is introduced cephalad through a stab wound into the antimesenteric border and the staples released once or twice, until a large reservoir, 12–18 cm in length, is completed. Interrupted or continuous sutures may be required to reinforce the layers of stapled sutures. The anastomotic staple lines are inspected for hemostasis (Fig. 14.16).

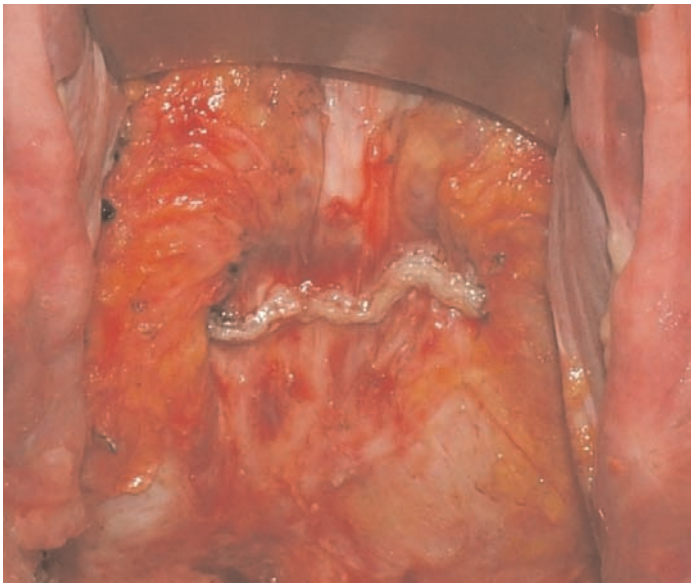

Fig. 14.15 Anorectal transection 2 cm above the dentate line

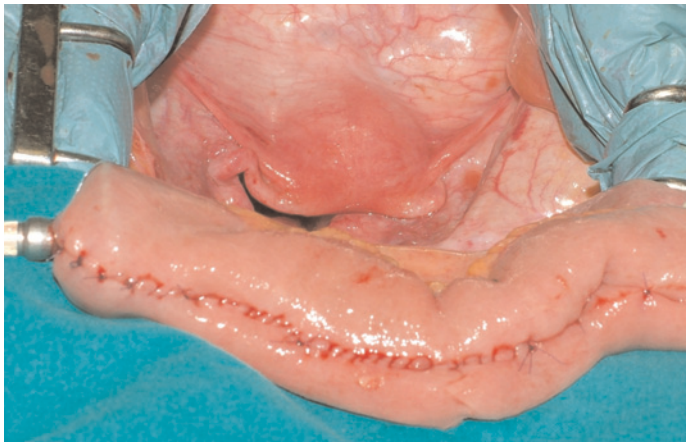

Fig. 14.16 Ileal J pouch

Hand-Sewn IPAA

A long Allis clamp passed through the rectal cuff delivers the apex of the pouch beyond the sphincters. Absorbable sutures are placed in the posterior half of the anastomosis, including the ileum, sphincters, and anoderm. An anterior suture line completes the anastomosis.

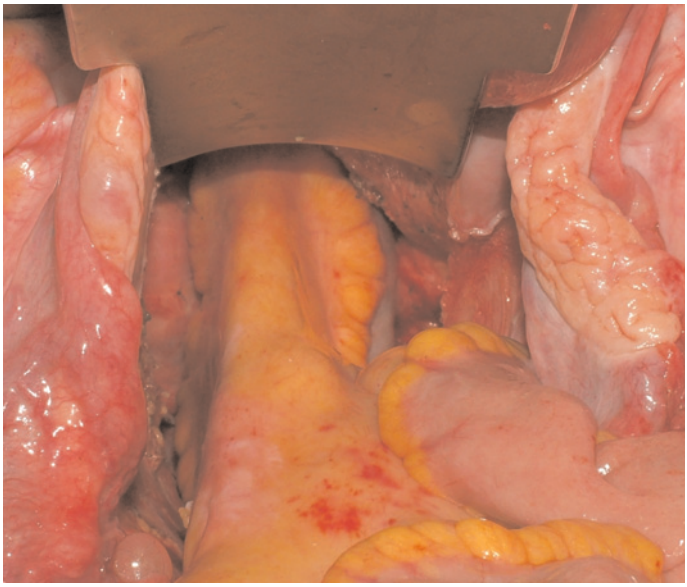

Fig. 14.17 Double-stapled restorative proctocolectomy with ileo pouch-anal anastomosis

Double-Stapled IPAA

This technique is associated with much less tension on the anastomosis than is the case with the hand-sewn technique, particularly in obese and tall patients. The perineum is pushed superiorly while at the same time the staples are released 1–2 cm above the dentate line. Leaving a long rectal cuff or transecting a portion of the internal and the external sphincters is not advisable. The anvil (diameter 28–29 mm) is inserted into the apex of the pouch and secured with a 3-0 suture. The stapling gun is placed into the anus, the trocar tip is fully advanced through the linear staple line at the midpoint of the closure, and the anvil shaft engaged with the stapler shaft. The instrument is closed and the staples released (Fig. 14.17).

Ileostomy

A temporary diverting-loop ileostomy is not always necessary, but is preferable in patients with pelvic complications and in toxic or malnourished patients. A loop of ileum, approximately 20–25 cm proximal to the pouch, is withdrawn through the abdominal wall, a small transverse enterotomy is made in the distal limb, and the proximal limb everted and sutured to the skin, so that an adequate nipple can be constructed (Fig. 14.13). About 1–3 months after IPAA, the stoma is removed with resection and an end-to-end anastomosis is performed. Alternatively, the mucosa can be unfolded and the stoma closed without resection.

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Laparoscopic Procedures for Crohn's Disease

The safety and feasibility of laparoscopic resections for the treatment of CD has been extensively studied. In a recent meta-analysis on 14 comparative studies vs. laparotomy (2 prospective controlled randomized trials, 2 prospective case-control studies, and 10 retrospective studies), 881 patients were collected [47]. Laparoscopy was associated with a longer operative time than laparotomy but had fewer postoperative complications, earlier restoration of intestinal functions, less postoperative pain, and shorter postoperative length of hospital stay. The rate of conversion to laparoscopy was 11.2% (range 4.8–29.2%). Robust data are available for laparoscopic resection in CD although not in UC. Two randomized, controlled studies have been published suggesting that laparoscopic ileocolic resection is associated with decreased morbidity and length of hospital stay [20, 48]. Maartense et al. further suggested that the overall costs of laparoscopic resection were lower [48]. However, while operative times have decreased with increased experience, most studies have still found significantly longer operative times than is the case in open procedures. The better cosmesis and body image in laparoscopic surgery are important because of the young age of the patients. Furthermore, at 4 weeks after surgery, the quality of life is similar in patients who underwent laparoscopic vs. open procedures [48]. Surgical relapses were similar in the two groups [49]. A surgical relapse after laparoscopic resection can be managed with a new laparoscopic approach.

Currently, a laparoscopic assisted procedure may be considered as the preferred operative approach for primary resections. For patients with complicated disease, laparoscopy is also feasible but the surgeon must be aware of the high rate of conversion. A laparoscopic assisted procedure also may be an option for those patients with acute colitis but who are usually excluded due to perforated acute colitis or toxic megacolon.

Notes on Surgical Techniques

Laparoscopic Ileocolic Resection

Under general anesthesia, the patient is placed in a supine position with both arms tucked at his or her sides. Three 5/10/12 mm trocars, a 30° angled laparoscope, and an intra-abdominal insufflation of 12 mmHg are used. The pneumoperitoneum is established following open Hasson trocar placement through an infraumbilical incision. Under laparoscopic vision, two trocars are placed in the patient's left flank, lateral to the rectus sheath. After a careful exploration of the abdomen to assess the site and extent of disease, the right colon, terminal ileum, and the inflammatory mass are fully mobilized, mainly with blunt dissection from the lateral to medial direction. The hepatic flexure is mobilized in most cases. No attempt should be made to transect the thick mesentery at this time. The right colon and the terminal ileum are extracted through a small (5–7 cm) right lower quadrant incision, using a

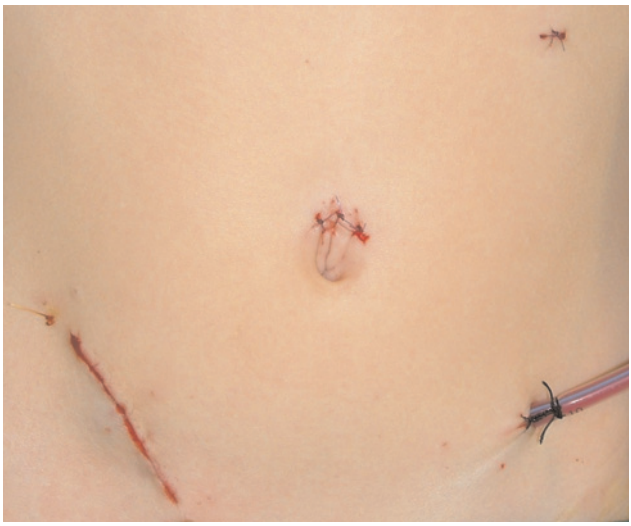

Fig. 14.18 Laparoscopic ileocolic resection. Three trocars are typically used, with extraction of the inflammatory mass through a lower right quadrant incision (5–6 cm)

Pfannenstiel or a midline incision. The terminal ileum and the cecum are transected with a linear stapler. A mechanical or hand sewn side-to side or end-to-end functional anastomosis is performed extracorporeally. In very simple cases, the ileocolic vascular pedicle is transected first with a 30-mm vascular stapler; transection of the bowel and formation of the ileocolic anastomosis are then performed intracorporeally (Fig. 14.18).

Laparoscopic Colectomy and Proctocolectomy

Five 5/10/12 mm trocars and a 30° angled laparoscope are used for colon mobilization and devascularization, followed by extraction through a right lower quadrant incision made where the diverting ileostomy is eventually planned. The terminal ileum is transected with a linear stapler near the valve. The entire colon is mobilized with extreme care. The spermatic vessels and ureters are accurately identified. Special attention is taken in dividing the gastrocolic ligament close to the stomach. The main ileocolic vessels are preserved if a future pouch construction is planned. When a rectum-preserving total colectomy is intended, the ileocolic branches, the right and middle colic vessels, and the sigmoid branches are isolated and tied. The main inferior mesenteric artery and the superior rectal artery are preserved. The colon is transected with an articulate linear stapler at the sacral promontory level, avoiding any dissection of the pelvic peritoneum. In case of severe rectal disease, an ultralow Hartmann procedure or a proctocolectomy can be performed. The entire colon is extracted through an enlarged right lower quadrant trocar incision, where the ileostomy is constructed (Fig. 14.19).

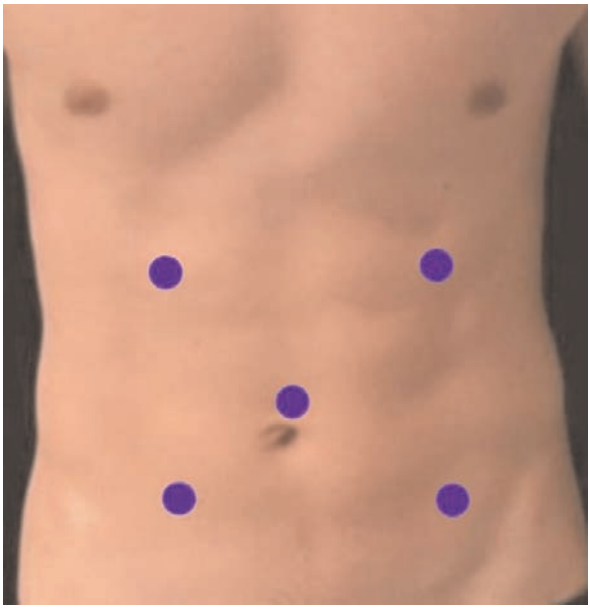

Fig. 14.19 Laparoscopic total colectomy. Five trocars are used, with extraction of the colon through an enlargement of a lower right quadrant trocar incision

Laparoscopic Restorative Proctocolectomy

Five or six trocars 5/10/12 mm are used for a total proctocolectomy and double-stapled IPAA. The rectum is transected with an articulate linear stapler 1–2 cm. above the dentate line. The colorectal specimen is extracted through an enlarged right lower quadrant trocar incision. The distal 25-40 cm of ileum is looped with 12- to 20-cm limbs and a standard J pouch is constructed. Care must be taken to inspect the anastomotic staple lines. The anvil is inserted through the apex of the ileal pouch. The abdominal incision is sutured and the peritoneum re-established. The circular stapler is introduced into the anus and the center rod advanced. The pouch is brought down to the pelvic floor, the anvil is connected to the pin, and the staples released. Finally, a loop ileostomy is eventually created.

References

1. Travis SPL, Stange EF, Lémann M et al (2006) European evidence based consensus on the diagnosis and management of Crohn's disease: current management. Gut 55(Suppl 1):i16-i35
2. Bernstein CN, Blanchard JF, Rawsthorne P, Yu N (2001) The prevalence of extraintestinal diseases in inflammatory bowel disease: a population-based study. Am J Gastroenterol 96:1116-1166
3. Michelassi F, Balestracci T, Chappell R, Block GE (1991) Primary and recurrent Crohn's disease. Experience with 1379 patients. Ann Surg 214:230-240

4. Broe PJ, Bayless TM, Cameron JL (1982) Crohn's disease: are enteroenteral fistulas an indication for surgery? Surgery 91:249-253
5. Present DH, Rutgeerts P et al (1999) Infliximab for the treatment of fistulas in patients with Crohn's disease. N Engl J Med 340:1398-1405
6. Sands BE, Anderson FH, Bernstein CN et al (2004) Infliximab maintenance therapy for fistulizing Crohn's disease. N Engl J Med 350:876-885
7. Ribeiro MB, Greenstein AJ, Yamazaki Y, Aufses AH Jr (1991) Intra-abdominal abscess in regional enteritis. Ann Surg 213:32-36
8. Garcia JC, Persky SE, Bonis PA, Topazian M (2001) Abscesses in Crohn's disease: outcome of medical versus surgical treatment. J Clin Gastroenterol 32:409-412
9. Gutierrez A, Lee H, Sands BE (2006) Outcome of surgical versus percutaneous drainage of abdominal and pelvic abscesses in Crohn's disease. Am J Gastroenterol 101:2283-2289
10. Greenstein AJ, Sachar DB, Mann D et al (1987) Spontaneous free perforation and perforated abscess in 30 patients with Crohn's disease. Ann Surg 205:72-76
11. Kronberger IE, Graziadei IW, Vogel W (2006) Small bowel adenocarcinoma in Crohn's disease: a case report and review of literature. World J Gastroenterol 12:1317-1320
12. Glehen O, Lifante JC, Vignal J et al (2003) Small bowel length in Crohn's disease. Int J Colorectal Dis 18:423-427
13. Krause U, Ejerblad S, Bergman L (1985) Crohn's disease. A long-term study of the clinical course in 186 patients. Scand J Gastroenterol 20:516-524
14. Softley A, Myren J, Clamp SE et al (1988) Factors affecting recurrence after surgery for Crohn's disease. Scand J Gastroenterol Suppl 144:31-34
15. Fazio VW, Marchetti F, Church M et al (1996) Effect of resection margins on the recurrence of Crohn's disease in the small bowel. A randomized controlled trial. Ann Surg 224:563-573
16. Hamilton SR, Reese J, Pennington L et al (1985) The role of resection margin frozen section in the surgical management of Crohn's disease. Surg Gynecol Obstet 160:57-62
17. Gasche C, Scholmerich J, Brynskov J et al (2000) A simple classification of Crohn's disease: report of the Working Party for the World Congresses of Gastroenterology, Vienna 1998. Inflamm Bowel Dis 6:8-15
18. Ikeuchi H, Kusunoi M, Yamamura T (2000) Long-term results of stapled and hand-sewn anastomoses in patients with Crohn's disease. Dig Surg 17:493-496
19. Tersigni R, Alessandroni L, Barreca M et al (2003) Does stapled functional end-to-end anastomosis affect recurrence of Crohn's disease after ileocolonic resection? Hepatogastroenterology 50:1422-1425
20. Milsom JW, Hammerhofer KA, Bohm B et al (2001) Prospective, randomized trial comparing laparoscopic vs. conventional surgery for refractory ileocolic Crohn's disease. Dis Colon Rectum 44:1-9
21. Simillis C, Yamamoto T, Reese GE et al (2008) A meta-analysis comparing incidence of recurrence and indication for reoperation after surgery for perforating versus nonperforating Crohn's disease. Am J Gastroenterol 103:195-205
22. Bernell O, Lapidus A, Hellers G (2000) Risk factors for surgery and postoperative recurrence in Crohn's disease. Ann Surg 231:38-45
23. Yamamoto T (2005) Factors affecting recurrence after surgery for Crohn's disease. World J Gastroenterol 11:3971-3979
24. Ozuner G, Fazio VW, Lavery IC et al (1996) How safe is strictureplasty in the management of Crohn's disease? Am J Surg 171:57-61
25. Tichansky D, Cagir B, Yoo E et al (2000) Strictureplasty for Crohn's disease: meta-analysis. Dis Colon Rectum 43:911-919
26. Fearnhead NS, Chowdhury R, Box B et al (2006) Long-term follow-up of strictureplasty for Crohn's disease. Br J Surg 93:475-482

27. Poggioli G, Laureti S, Pierangeli F et al (2003) A new model of strictureplasty for multiple and long stenoses in Crohn's ileitis: side-to-side diseased to disease-free anastomosis. Dis Colon Rectum 46:127-130
28. Michelassi F, Upadhyay GA (2004) Side-to-side isoperistaltic strictureplasty in the treatment of extensive Crohn's disease. J Surg Res 117:71-78
29. Shatari T, Clark MA, Yamamoto T et al (2004) Long strictureplasty is safe and effective as short strictureplasty in small-bowel Crohn's disease. Colorectal Dis 6:438-441
30. Tonelli F, Fedi M, Paroli GM et al (2004) Indications and results of side-to-side isoperistaltic strictureplasty in Crohn's disease. Dis Colon Rectum 46:127-130
31. Barwood N, Platell C (1999) Case report: adenocarcinoma arising in a Crohn's stricture of the jejunum. J Gastroenterol Hepatol 14:1132-1134
32. Yamamoto T, Fazio VW, Tekkis PP (2007) Safety and efficacy of strictureplasty for Crohn's disease: a systematic review and meta-analysis. Dis Colon Rectum 50:1968-1986
33. Maykel JA, Hagerman G, Mellgren AF et al (2006) Crohn's colitis: the incidence of dysplasia and adenocarcinoma in surgical patients. Dis Colon Rectum 49:950-957
34. Collins PD, Mpofu C, Watson AJ, Rhodes JM (2006) Strategies for detecting colon cancer and/or dysplasia in patients with inflammatory bowel disease. Cochrane Database Syst Rev CD000279
35. Tekkis PP, Purkayastha S, Lanitis S et al (2006) A comparison of segmental vs. subtotal/total colectomy for colonic Crohn's disease: a meta-analysis. Colorectal Dis 8:82-90
36. Bernell O, Lapidus A, Hellers G (2001) Recurrence after colectomy in Crohn's colitis. Dis Colon Rectum 44:647–654
37. Andersson P, Olaison G, Hallbook O, Sjodahl R (2002) Segmental resection or subtotal colectomy in Crohn's colitis? Dis Colon Rectum 45:47-53
38. Fichera A, McCormack R, Rubin MA et al (2005) Long-term outcome of surgically treated Crohn's colitis: a prospective study. Dis Colon Rectum 48:963-969
39. Braveman JM, Schoetz DJ Jr, Marcello PW et al (2004) The fate of the ileal pouch in patients developing Crohn's disease. Dis Colon Rectum 47:1613-1619
40. Brown CJ, Maclean AR, Cohen Z et al (2005) Crohn's disease and indeterminate colitis and the ileal pouch-anal anastomosis: outcomes and patterns of failure. Dis Colon Rectum 48:1542-1549
41. Hartley JE, Fazio VW, Remzi FH et al (2004) Analysis of the outcome of ileal pouch-anal anastomosis in patients with Crohn's disease. Dis Colon Rectum 47:1808-1815
42. Reese GE, Lovegrove RE, Tilney HS et al (2007) The effect of Crohn's disease on outcomes after restorative proctocolectomy. Dis Colon Rectum 50:239-250
43. de Oca J, Sanchez-Santos R, Rague JM et al (2003) Long-term results of ileal pouch-anal anastomosis in Crohn's disease. Inflamm Bowel Dis 9:171–175
44. Mylonakis E, Allan RN, Keighley MR (2001) How does pouch construction for a final diagnosis of Crohn's disease compare with ileoproctostomy for established Crohn's proctocolitis? Dis Colon Rectum 44:1137–1142
45. Regimbeau JM, Panis Y, Pocard M et al (2001) Long-term results of ileal pouch-anal anastomosis for colorectal Crohn's disease. Dis Colon Rectum 44:769–778
46. Broering DC, Eisenberger CF, Koch A et al (2001) Strictureplasty for large bowel stenosis in Crohn's disease: quality of life after surgical therapy. Int J Colorectal Dis 16:81-87
47. Tan JJ, Tjandra JJ (2007) Laparoscopic surgery for Crohn's disease: a meta-analysis. Dis Colon Rectum 50:576-585
48. Maartense S, Dunker MS, Slors JF et al (2006) Laparoscopic-assisted versus open ileocolic resection for Crohn's disease: a randomized trial. Ann Surg 243:143-153
49. Eshuis EJ, Polle SW, Slors JH et al (2008) Long-term surgical recurrence, morbidity, quality of life, and body image of laparoscopic-assisted vs. open ileocolic resection for Crohn's disease: a comparative study. Dis Colon Rectum 51:858-867

Crohn's Disease in Children

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S. Cucchiara, L. Stronati

Introduction

Although Crohn's disease occurring in adults and in childhood share the same immuno-pathogenetic mechanism and genetic background, as well as diagnostic modalities and therapeutic strategies, aspects pertaining to all these variables make the disorder in children a peculiar entity. Children with Crohn's disease represent a unique patient population to investigate the initial intestinal immune response and to characterize the genotype-phenotype relationship, the natural history of the disease, the intervention of environmental factors, and the early introduction of therapeutic immunomodulators and biologicals to alter the natural course of the disease.

This chapter emphasizes the specificities of Crohn's disease in childhood.

Epidemiology

Although the exact incidence and prevalence of inflammatory bowel disease (IBD) (Crohn's disease, ulcerative colitis, indeterminate colitis) in developed countries unknown, recent studies suggest an increasing frequency both in pediatric and adult populations. The increase in the incidence and prevalence of IBD is part of the world-wide emergence of chronic immunomediated inflammatory diseases that is strongly related to social and economic development. It is now estimated that roughly 100,000 children and adolescents are affected by IBD in the USA [1]. Heyman and colleagues of the Pediatric IBD Consortium have reported prospective data from more than 1300 prevalent and index cases of IBD, from six referral pediatric gas-

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troenterology centers in the USA, over a 3-year period [2]. According to these data, ulcerative colitis (UC), Crohn's disease (CD), and indeterminate colitis (IC) were equally prevalent among the youngest (0–2 years) age groups, and definitive diagnoses were made in 1% of patients before they reached one year of age. UC was more common (47%) in younger (3–5 years) patients, whereas the incidence of CD rose with age, reaching 66% in the 13–17 years strata; the prevalence of IC fell from 33% in the youngest cohort to 9% in the older age strata. Recently, an IBD registry from a northwest region of France (EPIMAD) identified 404 consecutive pediatric CD patients during a 5-year period (73% of all pediatric cases: UC 23% and IC 4%) [3]. The mean annual incidence of pediatric CD, standardized to age and sex, was 2.6 per 100,000.

Disease location of IBD seems to differ by patient age, such that the proportion of UC, CD, or IC patients with isolated colonic disease decreases with age. More than 63% of children < 8 years of age had isolated colonic disease, irrespective of the IBD type, whereas only 35% of those ≥ 8 years had isolated colonic disease. It is widely agreed that early-onset IBD (0–5 years) seems to represent a disease phenotype that is distinct from disease of later onset (> 5 years). A recent report from the Pediatric IBD Consortium Registry analyzing uniform data, collected between January 2000 and November 2003, from 989 consecutive CD patients confirmed this peculiarity. In particular, an initial classification as UC or IC was more common among the 0–5 years of age group, and rectal bleeding was a more common presenting feature in this group than in patients 6–17 years of age.

Familial aggregation studies have documented a significantly greater relative risk for developing IBD among first-degree relatives of affected individuals compared to the general population. From data of the US Pediatric IBD Consortium it was observed that, overall, 29% of IBD patients had one or more family members with IBD. Indeed, a family history of IBD was noted in 3% of siblings, 9% of parents, and 22% of second-degree relatives. The most striking evidence that genetic factors are involved into the mechanisms of IBD comes from twin studies. Disease concordance has been found to be as high as 58% and 18% for monozygotic twins with CD and UC, respectively, while the concordance for dizygotic twins is 0–10% for both diseases. Monozygotic twins are also commonly concordant for the same types of IBD, suggesting that there are genetic factors specific for CD and UC.

A family history of IBD appears to be more common in CD than in UC patients such that the sibling risk ratio (i.e., the risk of a sibling developing disease compared to the risk within the general population) is 30–40 for CD and 10–20 for UC [4]. There is also evidence suggesting that early-onset CD represents a distinct and more aggressive phenotype than disease in subjects older than 20 years. It has been observed that disease occurring in younger patients is associated with a greater family history of IBD and a greater need for surgery than disease occurring in the older age group. There is also a high rate of concordance for disease location and disease behavior among affected relatives with CD. In addition, offspring of affected CD parents tend to develop CD at an early age, with a greater extent of the disease. Thus, the concept of genetic anticipation for CD has been proposed even though it has not been fully confirmed.

Genetics

Complex disorders such as CD are controlled by multiple risk factors that interact with each other. The currently proposed genetic model for CD phenotypes outlines the complex interactions between environmental factors and genetic determinants, which together result in clinical expression of the disease along the gut of genetically predisposed individuals. Almost all of the genes that have been associated with susceptibility to CD are related to the innate and adaptive immune response of the intestinal mucosa to commensal microbiota as well as to the biology of the intestinal epithelium [5–7].

Crohn's Disease Susceptibility Loci: IBD1 (CARD15/NOD2)

Recently, several genetic associations have been identified in IBD, mainly in CD. The first susceptibility gene for CD to be identified is located on chromosome 16 (IBD1) and was named *CARD15/NOD2*. Its product is a cytoplasmic protein involved in microbial sensing [8,9]. Three mutations of this gene (R702W, G908R, and 1007fsinsC) are considered independent risk factors for CD in Caucasians but not in Asians and African Americans. Individuals with one of the disease-associated alleles have a 2- to 4-fold increased risk for developing CD, whereas homozygous or compound heterozygous carriers have an up to 40-fold increase in the genotype-relative risk. However, the known polymorphisms of *CARD15/NOD2* are not sufficient neither necessary for the development of CD; indeed, most CD patients do not exhibit CARD15/NOD2 mutations and the estimated penetrance is only 5–10%. The genotype-phenotype of individuals with CARD15/NOD2 variants seems to be associated with a younger age at onset, ileal involvement, and a tendency to develop strictures and/or fistulae. Interestingly, children with the 1007fsinsC mutation have a six-fold increased risk for developing a stricture phenotype requiring surgery.

Pediatric-onset CD is associated with more extensive colitis and less ileitis than is the case in adult-onset CD. These differences in disease site by age suggest a different genotype or different host responses, such as decreased ileal susceptibility or increased susceptibility of the colon. Recent data have shown that young children without NOD2/CARD15 mutations have an isolated distribution of their colonic disease, suggesting that this phenotype is associated with genes that lead to a specific phenotype of early-onset disease [10].

Other Candidates Genes of Interest

Several studies identified linkage with a region at chromosome 5q31-q33 (IBD5), specifically with a length of approximately 250 kb, between susceptibility for CD and early age of onset. This region contains a number of potentially interesting genes, including several encoding immunoregulatory cytokines. Two functionally relevant mutations in genes coding for the carnitine/organic cation transporter (OCTN), locat-

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ed on the IBD5 locus, were shown to be associated with CD. OCTNs help in transporting, distributing, and eliminating charged organic cations (e.g., those found in drugs, xenobiotics, endogens metabolites) and thus are important for intracellular homeostasis. In addition, they transport the endogenous amine carnitine, an essential cofactor for lipid metabolism, assisting in β-oxidation, and playing an important role in cellular energy production. The T substitution of C_{1672} in exon 9 of the SLC22a4 gene encoding OCTN1 and a G to C substitution at position-207 in the promoter region of SLC22a5, encoding OCTN2, were identified. Together these polymorphisms form a two-allele risk haplotype associated with CD susceptibility. Gene-gene interaction with mutations in the CARD15 gene has been suggested as well.

Recently, the DLG5 gene was recognized as a novel susceptibility gene for CD and IBD. DLG5 is important in maintaining epithelial structure, and genetic variants could result in impaired intestinal permeability. Two haplotypes have been implicated in IBD and CD susceptibility. A single nucleotide polymorphism (SNP) resulting in an amino acid substitution R30Q was positively associated with IBD and CD patients in a case–control study. A second haplotype, identified by several other tagging SNPs, was found to be protective for IBD. Gene–gene interaction with CARD15 in CD was detected by a significant difference in association with the 113A variant in patients carrying the CARD15 risk alleles compared with patients not carrying these alleles. However, only one published study has been able to reproduce these results, and initial enthusiasm has been discouraged by several studies from Europe and Japan that failed to show any association of DLG5 with IBD. As for OCTN, large population-based case–control studies are needed to elucidate the role of DLG5 in IBD susceptibility.

The genome-wide association (GWA) study involves rapidly scanning markers across the complete sets of DNA, or genomes, of many people to find genetic variations associated with a particular disease. This approach has revealed a highly significant association between CD susceptibility and variation in the gene encoding the interleukin (IL)-23 receptor (IL-23R). IL-23 is a cytokine closely related to IL-12 and is known to be critical in activating several pathologic inflammatory reactions. Several reports indicate that variations in the IL-23R gene have a protective effect for developing CD. Other loci identified by GWA studies are: the autophagy-related 16-like 1 (ATG16L1) gene on chromosome 2q37, a locus on chromosome 5p13 that may influence expression of prostaglandin E receptor 4 (subtype 4) (PTGER4), and a region on chromosome 10q21 with no known genes that is flanked on the centromeric side by a zinc-finger gene (ZNF365) and on the telomeric side by a predicted protein (c10orf 22) and the early growth response 2 (EGR2) gene, which also encodes a zinc-finger protein.

Natural History

The EPIMAD Registry (France) and the US Pediatric IBD Consortium Registry have generated interesting data on the presentation and course of CD in childhood [2,3].

Both registries outlined that early onset (0–5 years of age) CD represents a unique phenotype, marked both by predominant colonic involvement and a stronger positive family history compared to later-onset CD. Interestingly, compared to patients with later-onset CD, a greater proportion of early-onset CD patients are more likely to have been initially diagnosed with UC or IC. A dramatic change in disease behavior seems to occur in pediatric CD during a follow-up period of almost a decade, generally evolving from non-penetrating, non-structuring disease (B1) to the structuring (B2) or penetrating phenotype (B3). Specifically, the B2 and B3 phenotypes increase from 25% to 44% and from 4% to 15% respectively, whereas the B1 phenotype decreased from 71% to 41% between the time of diagnosis and the last follow-up. Perianal disease (abscesses or fistulae) was detected in almost 9% of patients at diagnosis and in 27% at follow-up. The risk for the development of complications such as abscesses, fistulae, strictures, or perianal fissures is greater in the older age group than in the early-onset group. Interestingly, the risk for the occurrence of compression, fractures/osteopenia/osteoporosis, and growth failure do not differ with age of diagnosis. The cumulative probability of surgery was 34% after 5 years. Stricturing and penetrating behaviors of the disease at diagnosis, growth retardation, and complications during follow-up were associated with an increased risk for surgery that did not differ according to CD localization or the presence of perianal disease at diagnosis. From data of EPIMAD Registry it is evident that corticosteroids are associated with an increased risk of recourse to surgery, whereas immunomodulators were associated with a decreased risk. Recent data from the US Pediatric IBD Collaborative Group Registry indicated that approximately 10% of newly diagnosed pediatric patients with CD have perianal fistulas and/or abscesses at the time of diagnosis; most of them will satisfactorily respond within 1 year to medical therapy alone. Of 276 patients, 41 had perianal lesions: 13 with skin tags and fissures, 28 with fistulae and/or abscesses. The latter resolved by 1 year in 20 cases whereas 8 of these patients had chronic recurrent perianal disease lasting more than 1 year following the diagnosis. Characteristically, patients with perianal disease are more likely to receive earlier antibiotics, biologics, and immunomodulators.

Growth Failure

Growth failure occurs in 15–40% of children with CD and it may precede clinical evidence of bowel disease by years [11]. In CD, temporary growth failure occurs in up to 50% of affected children and stunting into adulthood may involve 15–30% of patients. Furthermore, final height remains below the fifth percentile in 7–30% of patients. While static height measurement may be misleading to define growth disturbance, a shift from higher to lower percentiles on a growth chart of height reflects accuracy height velocity (which can be expressed either as a centile or as a standard deviation score for age and gender). It is known that linear growth occurs along the growth hormone (GH)–insulin-like growth factor 1 (IGF-1) axis, which is affected by adequate nutrient support [12]. Briefly, GH, secreted by the pituitary gland, stimu-

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lates the release of IGF-1, which in turn stimulates the proliferation of chondrocytes at the epiphyseal plate, with growth of the long bones. Circulating IGF-1 is found in complexes with IGF binding proteins (IGFBPs). Factors that increase IGFBP-1 may decrease the bio-availability of IGF-1: IFGBP-1 expression is increased by insulopenia and starvation as well as corticosteroid therapy. Our current understanding of the pathogenesis of growth retardation indicates that the etiology is multifactorial, with the inflammatory process per se directly implicated, in addition to nutritional deficiencies and prolonged corticosteroid therapy [13]. Interestingly, tumor necrosis factor (TNF)-α, a Th1-related cytokine, has a key role in cytokine-mediated growth retardation, synergistically inhibiting, together with IL1β, the growth plate, increasing IL-6 transcription, and possibly decreasing circulating leptin levels.

Promoting growth and pubertal development has become one of the main expectations and targets in the therapy of pediatric CD patients [14]. Different treatment modalities may improve growth providing they achieve suppression of active disease and inflammation. There is a widely agreed view that suppression of inflammation, an adequate maintenance strategy to minimize the relapse rate, and intensive nutritional support are the key therapeutic endpoints that should be combined in order to support growth in children with CD [12]. Nutritional therapy alone (consisting of either polymeric or elemental formulae) is able to achieve mucosal healing and promote growth in a short-term period, whereas a long-term nutritional maintenance strategy has yet to be designed [15]. Consecutive cycles of exclusive nutritional therapy intermingled with periods of a normal or low-antigen diet, may offer an appropriate long-term approach to minimize relapses and maintain remission as well as an optimal growth rate. Corticosteroids are the key therapy responsible for drug-induced growth failure and every effort should be made to avoid their chronic use (mainly because they do not alter the course of the disease), while immunomodulators, which have become the standard of care in children with CD because of their value as steroid-sparing medications, have not been evaluated on a long-term basis as growth promoting drugs. Interestingly, a large prospective randomized trial in childhood (REACH study) examined the role of infliximab (IFX) (a monoclonal anti-TNFα antibody) as a maintenance medication in 103 CD children over a period of 46 weeks [16]. The height status of patients with at least a 1-year delay in bone age was assessed, and the patient's height *z*-score calculated. The latter is a measure of the deviation of the patient's height from the expected height of an age- and sex-matched population. The scores for these patients improved significantly at both week 30 (mean improvement in *z*-score of 0.3) and week 54 (mean improvement in *z*-score of 0.5). Previously, Walters et al. evaluated 32 children and adolescents with CD who were treated with IFX because of chronically active disease despite immunomodulatory and prior corticosteroid therapy [17]. The mean SD score for height at the beginning of IFX was -1.4 for patients in Tanner stage 1–3, after IFX; height improved by a mean of 0.5 in 70% patients with a Tanner pubertal stage < 4. Nevertheless, growth abnormalities seem to persist in most children with CD, despite improvement in disease activity and the frequent use of immunomodulators and biologics [18]. Future strategies should be aimed at minimizing or even eliminating the early use of corticosteroids–the main factor associated with poorer growth outcome.

Diagnosis

Figure 15.1 presents a simplified diagnostic algorithm of pediatric IBD. The gold standard for the diagnosis of IBD remains endoscopic evaluation with tissue histology. However, less invasive tools with increased sensitivity and specificity, such as serological markers, videocapsule endoscopy, and advanced imaging modalities, are being developed [19].

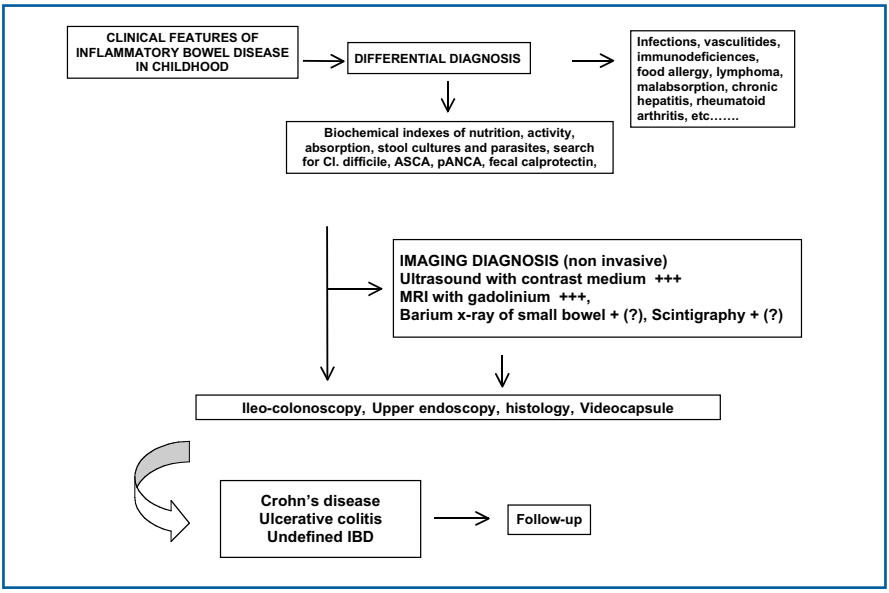

Fig. 15.1 Diagnostic algorithm of inflammatory bowel disease in childhood

Gastrointestinal Endoscopy

All children evaluated for CD undergo both esophagogastroduodenoscopy and ileocolonoscopy at initial diagnostic work-up. These procedures are performed under conscious sedation (with the administration of oral or intravenous midazolam, a benzodiazepine with hypnotic action) or general anesthesia (in infants and small children and in those with risk factors). The presence of inflammatory changes in the esophagus, stomach, or duodenum is common in UC and CD; however, the presence of granuloma is specific for CD, whereas the presence of focally enhanced gastritis is common in CD, but non-specific and is also detected in UC. In general, gastroenterological endoscopic and histologic criteria distinguishing CD from UC are shared by adult and pediatric patients; however, there are features that are peculiar to the pediatric age. These can be summarized as follows:

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- The diagnosis of CD is unequivocal if there are clear radiographic and/or endoscopic features of small bowel involvement, non-caseating granulomas on endoscopic mucosal biopsies, or evidence of severe perianal disease (fissures, fistulae). When CD only involves the colon and granulomas are not present on biopsies, the diagnosis is rather difficult: the differentiation of CD colitis from UC can be established primarily by the endoscopist, through the observation at initial endoscopy of focal discontinuous inflammation, deep fissuring ulcers, and aphthous lesions superimposed on a background of normal colonic mucosa.
- In general, over-reliance on histological interpretation at the expense of an appropriate combination with clinical and gross endoscopic features, can result in the inappropriate classification of UC patients as instead having CD, on the basis of non-specific mucosal inflammatory changes. Similarly, a reliance only or mainly on the visually determined morphology of the colon without an appropriate tissue sampling may carry a risk of failing to identify granulomatous inflammation (which would change the diagnosis from UC to CD).
- Although colonic and ileal biopsies of patients with CD share many of the same features of chronic colitis, mucosal changes in CD colitis (particularly early in the disease course) can be patchy and show subtle or slight features of chronicity. The earliest discernable lesion of CD is typically a focal active colitis with a lymphoid aggregate, which corresponds to the endoscopic aphthous erosions. The predictive value of focal active colitis for the development of CD has been debated: it has been suggested that almost one third of children with this feature develop CD, the remaining having infectious colitis or an idiopathic inflammation.
- It is widely agreed that backwash ileitis is seen in approximately one-fourth of UC patients with inflammation involving the entire colon (pancolitis) and that ileal inflammation is very rare in UC limited to the left colon. Ileal erythema and granularity can characterize the backwash ileitis (usually for the most distal 10 cm) and lymphoid nodules can be obvious; however, no linear ulcerations, deep fissures, or areas of cobblestoning are seen and the inflammation consists of a mild mixed infiltrate without crypt distortion, atrophy, or epithelial changes. Furthermore, imaging tools show that in backwash ileitis the terminal ileum is of normal caliber and without stenosis or cobblestoning.
- While UC is classically defined as a diffuse continuous disease beginning in the rectum and extending proximally without skip areas, “patchy colitis” and “relative rectal sparing” are often observed in children with new-onset UC and are also seen in treated colitis; thus, the criterion of “rectal sparing” as discriminatory between CD and UC has less value in childhood than in adult IBD.
- While epidemiological studies in adults report a prevalence of IC of 5–10%, pediatric studies cite a prevalence of up to 30%. It is common belief that pediatric gastroenterologists are more familiar than adult gastroenterologists with the diagnosis of IC. Nevertheless, the current literature lacks a precise definition of IC such that there is wide variability in the use of this term and a paucity of studies reporting long-term follow-up of these patients. Criteria for diagnosing IC or, as recently suggested, IBD unclassified (IBDU) can be summarized as follows: a history of chronic colitis compatible with both the diagnosis of CD or UC; colitis that cannot

definitively be declared as CD or UC based on clinical history, physical examination, endoscopic appearance, histologic findings, and imaging studies; endoscopy and histopathology either inconclusive or divergent with regard to the diagnosis of UC or CD; exclusive inflammation of the large bowel and neither endoscopic nor histologic findings typical for CD or UC. IBDU can only be diagnosed after a full diagnostic work-up. This must include colonoscopy with intubation of the terminal ileum, upper gastrointestinal endoscopy, and small bowel follow-through or MRI of the ileum. The diagnosis of IBDU is suggested by the histology, which shows acute and chronic inflammation with architectural changes confined to the colon, the absence of abnormalities suggesting lymphocytic or allergic colitis, or CD, a normal small bowel follow-through or MRI of the ileum, and no definite classification of CD or UC possible with histology. If the clinician decides to classify a patient as having IBDU, it is crucial that the medical records outline the clinical data that prompted the diagnosis. At the same time it should be stated that at some point following the diagnosis these patients will benefit from additional endoscopic and histologic evaluation to determine whether features prompting the IBDU diagnosis have changed and to attempt to make a definitive diagnosis of CD or UC.

Videocapsule Endoscopy

This diagnostic tool (VCE) is increasingly being used in the detection of obscure small bowel lesions and now has a proven role in the identification of CD lesion of the ileum. Although most cases of IBD are diagnosed without difficulty, there are cases of CD in which standard diagnostic tools do not reveal features specific to the disease. Usually, these individuals have isolated small bowel disease that is not detected with other modalities. VCE may be of help in these situations, even if prospective pediatric studies comparing VCE and other tools are not available. Studies in adults have shown that the diagnostic yield of VCE is similar to that of push enteroscopy in patients with suspected CD. The drawbacks of VCE are the cost of the test and the potential risk of capsule impaction in strictured areas of the small bowel. Thus, contrast radiography or MRI of the small bowel is recommended in order to exclude the presence of strictures. Future studies are warranted to determine whether capsule endoscopy should be done as a routine examination in patients with newly diagnosed colitis and normal diagnostic imaging findings.

Imaging-Based Diagnosis

Direct imaging of the gastrointestinal tract is essential to accurately stage the location, extent, and type (mucosal versus transmural) of intestinal inflammation. Accurate, inexpensive, non-invasive studies in the evaluation of IBD would represent a significant advance in identifying and measuring disease activity. Small intestinal contrast ultrasonography (US) has been shown to be as sensitive as a small bowel series with barium in detecting bowel abnormalities in CD. The technique has gained

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considerable acceptance among pediatricians and pediatric gastroenterologists due to its non-invasive nature as well as its capability to detect mucosal abnormalities and to document complications of CD, such as abscess formation. High-resolution probes can also detect bowel wall thickening, fibro-fatty infiltration of the mesentery, enlarged nodes, and prestenotic dilation. Interestingly, Doppler US can be of value in depicting areas of inflammatory hyperperfusion. Perfusion velocity is higher in the small bowel and colon in CD than in normal controls, but actual values seem to not well correlate with disease activity indexes. In a diagnostic algorithm of children with suspected CD, small bowel US can be proposed as non-invasive initial screening prior to more invasive diagnostic procedures, such as colonoscopy.

Although computed tomography (CT) is an excellent tool for detecting disease and disease complications, such as abscess, its use is limited by the significant radiation exposure. Magnetic resonance imaging (MRI) is an emerging technique for the evaluation of small bowel CD [20]. Advantages over conventional barium studies include the ability to detect extraluminal complications as well as the absence of ionizing radiation. The advent of powerful gradient systems has substantially improved image quality in ultrafast magnetic resonance sequences. Within this context, MRI of the small bowel has been developed, providing an alternative imaging modality to CT. Inherent MRI properties include excellent soft-tissue contrast and three-dimensional capabilities, which may be of importance in studies of the small intestine. Sufficient luminal distension to guarantee the accurate detection of individual lesions has so far been achieved only with a certain amount of an oral iso-osmotic water solution (polyethyleneglycol). A lack of consistent optimal luminal distension downgrades the diagnostic efficacy of the technique and limits its focus to assessing moderate or extensive wall thickening and the presence of stenosis, as well as documenting extraintestinal complications. In children with suspected IBD, gadolinium MRI (G-MRI) has shown a sensitivity of 94% and a specificity of 92% in terms of diagnostic accuracy. This tool seems also to be very sensitive in detecting perianal disease, by showing abscesses and fistulae, and thus aids surgeons in caring for these complications. Because of its non-invasive nature and its inherent properties, MRI is currently contributing to the assessment of CD activity. MRI features that are reported to reflect or measure disease activity include mucosal hyperemia, intramural edema, transmural ulceration, wall thickening and enhancement, and mesenteric involvement, in terms of either vascular engorgement (the comb sign) or the presence of inflammatory mesenteric lymph nodes. A number of studies suggest that the activity of CD can be assessed using these MRI parameters.

There is new evidence that positron emission tomography (PET) scanning can be helpful in measuring CD activity. PET has been assessed in both children and adults and has shown excellent sensitivity for detecting active bowel inflammation, but with poor specificity in some studies. PET alone appears sufficient for the evaluation of UC, but PET/CT provides considerably more information over PET alone in the evaluation of CD. Current clinical applications for PET in IBD include its use in the early evaluation of IBD, especially in children who may not tolerate an invasive test such as colonoscopy; and in differentiating between a flare of IBD versus the onset of a non-inflammatory process causing similar symptoms in patients with known IBD.

Many unanswered questions remain, but PET appears to be a promising tool in the non-invasive evaluation of IBD.

Serological Markers

Immune reactivity to specific microbial antigens has been reported in the sera of IBD patients and has been used to discriminate between UC and CD and to predict the severity of the disease course. Different biomarkers are commercially available for this type of analysis: anti-*Saccharomyces cerevisiae* (ASCA) IgG and IgA, anti-*E. coli* outer membrane porin C (OmpC) IgA, anti-*Pseudomonas fluorescens* CD-related protein (I2) IgA, anti-flagellin (C Bir-1) IgA, and anti-perinuclear anti-neutrophil (pANCA) IgG with DNAse sensitivity used to improve specificity. The use of an antibody panel consisting of pANCA, ASCA, and Anti-OmpC identified 65% of children with CD and 76% of children with UC, with a specificity of 94%. In other reports, a sequential serodiagnostic strategy was accurate in differentiating IBD from non-IBD in 84% of patients presenting with non-specific symptoms suggestive of IBD but with a normal physical examination. A positive test sequence was predictive of IBD in 90% of patients, whereas a negative sequence predicted the absence of disease in 80%. Higher ASCA levels are associated with younger age of disease onset, stricturing and penetrating disease, and the need for surgery, in both adults and children with CD. It was recently shown that the frequency of complicated CD (internal penetrating or structuring) increased with an increasing number of serologic immune responses, and the odds of developing complicated disease were highest in the cohort of patients positive for anti-I2, anti-OmpC, anti-CBir-1, and ASCA. In a Canadian study that looked at the utility of serological markers in predicting complications and surgery in children, it was shown that ASCA positivity increased the hazard of developing a complication (abscess or fistula) at any time during the disease course in patients with pediatric-onset CD.

Management

Great progress has been made in the medical management of IBD over the past decade [21]. This has largely resulted from better use of "older" medications as well as the emergence of newer therapies, particularly biological agents [22]. Current expectations from the therapy of CD in children can be summarized as follows:

- Inducing clinical remission
- Maintaining clinical remission
- Improving the quality of life
- Promoting growth and pubertal development
- Mucosal healing
- Reducing costs related to hospitalization and surgery
- Minimizing complications due to the disease and to therapy

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To date there is no single therapy that meets all of these requirements. In the following, we describe some of the characteristics of the current medical treatment modalities in relation to their respective roles for CD.

5-Aminosalicytates

The use of these drugs in pediatric CD is largely derived from reports of adult gastroenterology studies, whereas no large controlled trials are available for pediatric populations. These drugs are mainly used in the long-term maintenance of remission, whereas their use in the active disease is limited to patients with mild disease localized to short segments of the gut. Reports from the literature have often been contradictory in terms of the effectiveness of 5-aminosalicylates in maintaining a medically induced remission. There is evidence that mesalamine (3 g/day) and azathioprine have the same efficacy in preventing postoperative clinical recurrence in adults, with no statistical difference at 24 months following segmental resection.

Corticosteroids

These agents are used in the majority of children with CD who present with moderate to severe disease activity [23]. The acute responses are excellent, with over 80% responding; however these drugs are unable to alter the natural course of the disease. Data from 109 children from a multicenter observational registry started in 2002 indicate that at 1 year of follow-up, 61% were corticosteroid responsive, 31% corticosteroid dependent, and 8% required surgery [24]. Growth impairment at diagnosis increased the risk of corticosteroid dependence or surgery at 1 year. Almost all these patients received immunomodulators, which are commonly used both as steroid-sparing agents and as an "exit strategy" for those patients started on corticosteroids.

Budesonide, a corticosteroid that is taken orally and released in the distal small bowel and proximal colon, acts locally and is thought to have fewer systemic side effects. The ideal candidates to receive budesonide are patients with mild to moderate ileal-right colonic disease; in this group, treatment with 9 mg budesonide daily was superior to 4 g mesalamine/day in inducing remission at 8 weeks (69 vs. 45%) and 16 weeks (62 vs. 36%). Budesonide (6 mg/day) used to maintain remission induced by 9 mg budesonide was shown to be better than placebo at 3 months, but no difference between the two groups was noted regarding long-term relapse rates (almost 60%). In pediatric patients, suppression of the pituitary–adrenal axis has been seen with budesonide use, and it is not clear whether growth velocity is maintained in the normal range in children on long-term therapy with this agent.

Immunomodulators

In a landmark study on 6-mercaptopurine (6-MP) treatment of pediatric CD, children

with moderate to severe disease were randomized to receive corticosteroids plus 6-mercaptopurine or corticosteroids plus placebo, and the clinical course was monitored over 2 years [25]. While the response at 3 months was similar in the two groups (almost 90% improvement), the 6-MP group was much more likely to be in remission at 18 months, requiring significantly less corticosteroids than the placebo group. A recent report from the Pediatric IBD Collaborative Research Group Registry Database evaluated, in a prospective observational fashion, the effects of immunomodulator (IM) use in newly diagnosed pediatric CD, comparing outcomes of "early" vs. "late" initiation of therapy [26]. Moderate/severe disease patients treated with IM were compared for variables such as outcomes of remission, corticosteroid use, IFX therapy, hospitalizations, and CD-related surgery based on the timing of initiation of IM therapy. It was noted that 80% of children with newly diagnosed moderate to severe CD received IM within 1 year and that early IM use was associated with reduced corticosteroid exposure and fewer hospitalizations per patient. The current prevailing attitude among pediatric gastroenterologists is to start IM therapy early in the course as a strategy to reduce corticosteroid use and hospitalization rate; however, criteria for selecting patients candidates for early IM use have not yet been clearly defined. This is important if we consider that azathioprine (which is metabolized to 6-MP) and 6-MP may take up to 3 months to be efficacious. Caution is needed in the use of azathoprine or 6-MP. Approximately 0.3 and 11% of the population is homozygous or heterozygous, respectively, for a deficiency in thiopurine methyltransferase (TPMT), an enzyme system mitigating accumulation of the metabolite 6-thioguanine, associated with severe bone marrow suppression. A commercial assay is now available to measure TPMT before starting therapy and can guide the clinician. However, bone marrow suppression may occur even in subjects with normal TPMT activity, and frequent blood count monitoring is required. It is suggested to interrupt or reduce the IM dose when the lymphocyte count decreases to < 1200/mmc. Other side effects of clinical relevance are hepatitis (reversible), pancreatitis (idiosyncratic), vasculitis, and an increased risk of infection. The chronic use of these drugs has been calculated to increase the risk of lymphoma by four-fold compared with the general population. Methotrexate has been used with the same success as IM in CD, although there are no large studies in pediatric IBD patients. However, methotrexate is thought to be a particularly useful agent in children failing azathioprine/6-MP or with intolerable side effects from those agents.

Biologicals

Infliximab (IFX) is a chimeric monoclonal antibody (IgG1) that neutralizes both circulating and cell membrane bound TNF-α and causes apoptosis of mononuclear cells in the intestinal lamina propria. The drug has been used worldwide in many pediatric patients even prior to the FDA approval for its pediatric use in May 2006. IFX is the most widely used biological agent, both for inflammatory luminal disease and for enterocutaneous and rectovaginal fistulizing disease, in inducing and maintaining remission [27]. Before the recently published REACH study (a controlled multicen-

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ter randomized trial), IFX was investigated in pediatric CD through open observational studies, some of them retrospective. These reported the ability of the drug to induce and maintain remission in a high percentage of patients resistant to conventional treatment, mainly consisting of corticosteroids and IM therapy. The REACH study reported a response rate of 89% at 10 weeks following a three-dose induction schedule of 5 mg/kg/dose at 0, 2, and 6 weeks in patients with moderate to severely active disease despite therapy with corticosteroids and IMs. It is now clear (also from the results of REACH) that for patients successfully responding to an induction regimen of IFX, scheduled maintenance infusions (5 mg/kg every 8 weeks) rather than episodic on-demand therapy results in better outcomes. This scheduled strategy is also associated with less formation of neutralizing antibodies (responsible for a lack of or reduced response to the drug) and a significantly reduced incidence of side effects. It is of clinical relevance that mucosal healing has been commonly demonstrated following IFX therapy, and there is a commonly agreed view that mucosal healing is strongly related to prolonged periods of remission and to decreased hospitalization rate. Extraintestinal manifestations of IBD, such as pyoderma gangrenosum, vascultis, uveitis, erythema nodosum, and arthritis, have been successfully treated with IFX. IFX is contraindicated in patients with active tuberculosis (TBC) or any other serious infection, such as undrained abscess, opportunistic infection (e.g., herpes zoster, cytomegalovirus), a history of demyelinating disease, malignancy, or congestive heart failure. Before starting IFX therapy, patients should be carefully checked for tbc and for underlying active viral infections. Among the drug's side effects, the most common are acute infusion reactions (flushing, chest pain, chills, dyspnea, and pruritus), which occur approximately following 5% of infusions. More serious reactions (hypotension and oxygen desaturation) are less common (0.1%). Infusion reactions can be controlled by slowing the rate of IFX infusion and with the administration of anthistamines and corticosteroids. Features of delayed hypersensitivity (arthralgias, myalgias, fever, rash, pruritus, and headache) occurring 3–14 days following an infusion have been reported occasionally. Despite the fact that most clinical trials did not report an increased rate of infection, postmarketing data have shown serious bacterial and fungal infections. It has been observed that concomitant corticosteroid therapy may be a greater risk factor for infection and other morbidities. The association between IFX use and the development of malignancies (in particular those of a lymphoproliferative nature) has been controversial. To date, there are no sound data suggesting a serious increased incidence of malignancy in IFX-treated patients compared to the general population.

Interestingly, data from the Pediatric IBD Collaborative Research Group Registry show that IFX maintenance therapy is a durable and effective treatment that is associated with prolonged corticosteroid withdrawal over a 3-year follow-up period.

Currently, biological agents are indicated for children with moderate-to-severe CD that has shown unresponsiveness to conventional treatment, whereas their use as first-line strategy is still debated and not yet officially approved. As basic research reveals new signaling pathways underlying the inflammatory cascade in the intestinal wall and new inflammatory mediators are identified, additional biological agents will no doubt become available for use in CD patients.

Recent reports of increasing numbers of mainly adolescents and young adults with an aggressive and commonly fatal form of lymphoma, hepatosplenic T cell lymphoma (HSTCL), has raised great concern among pediatric gastroenterologists caring for patients with IBD [28]. These cases have influenced therapeutic attitudes in the management of severe IBD in children, and widely accepted strategies (i.e., combination IMs and biologicals) are now being questioned. In contrast, a maintenance strategy of monotherapy with biologics without concomitant IM administration is gaining acceptance [29]. It should, however, be remembered that several cases of HSTCL have been described in patients receiving only IMs and no biologicals.

There are two other anti-TNFα agents, adalimumab (ADA) and certolizumab, for which controlled trial have shown efficacy. Response and remission rates are similar to those following IFX. Due to the fully humanized nature of these antibodies, allergic reactions may be less common, but antibody formation has been observed. The very few studies on ADA in pediatric patients are retrospective and with a short follow-up period. Recently, however, a North American multicenter retrospective study that included 100 CD children evaluated the short- and long-term efficacy of ADA, showing high rates of clinical response and remission. Most of the patients were treated with a baseline schedule of 80/40 mg or 160/80 mg (subcutaneous injections at 0 and 2 weeks, and maintenance every other week). ADA was successful in patients naive to biologicals as well as in those with a loss of response to IFX.

Nutritional Therapy

As an alternative to corticosteroids, nutritional therapy has been reported to induce remission in almost two-thirds of patients with active CD [31]. This treatment traditionally has consisted of elemental (amino acid mixture) or semi-elemental (hydrolyzed) formulae given continuously through a nasogastric tube. Recently, however, polymeric formulae with a better palatability and lower cost than semi-elemental or elemental formulae have been proposed for treating active CD. A course of 6–8 weeks of nutritional therapy alone is able to induce remission in almost 80% of children, without the addition of drugs and no regular food allowed. In patients with growth failure or in those with features of malnutrition and in whom withdrawal of corticosteroids is suggested, nutritional therapy should be offered. Unfortunately, many children are unable to orally consume adequate sufficient amounts of formula and thus require a nasogastric tube. Moreover, most patients will relapse following the cessation of primary enteral feeding, and maintenance strategies are required; these can include repeated cycles of nutritional therapy or IM addition.

The mechanisms by which nutritional therapy is active include bowel rest, low-fat content of the administered diets, a change in the commensal microbiota profile. The low antigen does not seem to explain the effectiveness since polymeric formulae containing whole proteins are as efficacious as elemental and semi-elemental formulae [32].

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Antiobiotics/Probiotics

Antibiotics have proven to be useful and efficacious in certain clinical settings. Metronidazole is used to treat perirectal fistula, although recurrence rates are high and toxicity (e.g., paresthesias) often limit its use. Ciprofloxacin is also used in this setting, but data are limited. In perianal disease characterized by abscesses and fistulae, the combined parenteral administration of metronidazole and ciprofloxacin is given prior to the beginning of biological therapy. Metronidazole has also been shown to decrease the likelihood of endoscopic recurrence at 3 months and clinical recurrence at 1 year following ileal resection and primary anastomosis in patients with CD.

Probiotics have not been reproducibly shown to alter the natural history of CD. They have been shown to be helpful in the prevention and treatment of pouchitis in UC patients following colectomy, whereas reports in CD patients are controversial and generally not enthusiastic.

References

1. Rufo PA, Bousvaros A (2007) Challenges and progress in pediatric inflammatory bowel disease. Curr Opin Gastroenterol 23:406-412
2. Heyman MB, Kirschner BS, Gold BD et al (2005) Children with early-onset inflammatory bowel disease (IBD): analysis of a pediatric IBD consortium registry. J Pediatr 146:35-40
3. Vernier-Massouille G, Balde M, Salleron J et al (2008) Natural history of pediatric Crohn's disease: a population-based cohort study. Gastroenterology 135:1106-1113
4. Biank V, Broeckel U, Kugathasan S (2007) Pediatric inflammatory bowel disease: clinical and molecular genetics. Inflamm Bowel Dis 13:1430-1438
5. Barrett JC, Hansoul S, Nicolae DL et al (2008) Genome-wide association defines more than 30 distinct susceptibility loci for Crohn's disease. Nat Genet 40:955-962
6. Cho JH (2008) The genetics and immunopathogenesis of inflammatory bowel disease. Nat Rev Immunol 8:458-66
7. Heap GA, van Heel DA (2009) The genetics of chronic inflammatory diseases. Hum Mol Genet 18:R101-R106
8. Stronati L, Negroni A, Merola P et al (2008) Mucosal NOD2 expression and NF-kappaB activation in pediatric Crohn's disease. Inflamm Bowel Dis 14:295-302
9. Kobayashi KS, Chamaillard M, Ogura Y, et al (2005) Nod2-dependent regulation of innate and adaptive immunity in the intestinal tract. Science 307:731-734
10. Levine A, Kugathasan S, Annese V et al (2007) Pediatric onset Crohn's colitis is characterized by genotype-dependent age-related susceptibility. Inflamm Bowel Dis 13:1509-1515
11. Heuschkel R, Salvestrini C, Beattie RM et al (2008) Guidelines for the management of growth failure in childhood inflammatory bowel disease. Inflamm Bowel Dis14:839-849
12. Shamir R, Phillip M, Levine A (2007) Growth retardation in pediatric Crohn's disease: pathogenesis and interventions. Inflamm Bowel Dis 13:620-628
13. Paganelli M, Albanese C, Borrelli O et al (2007) Inflammation is the main determinant of low bone mineral density in pediatric inflammatory bowel disease. Inflamm Bowel Dis 13:416-423
14. Bousvaros A, Sylvester F, Kugathasan S et al (2006) Challenges in pediatric inflammatory bowel disease. Inflamm Bowel Dis 12:885-913

15. Borrelli O, Cordischi L, Cirulli M et al (2006) Polymeric diet alone versus corticosteroids in the treatment of active pediatric Crohn's disease: a randomized controlled open-label trial. Clin Gastroenterol Hepatol 4:744-753
16. Hyams J, Crandall W, Kugathasan S et al (2007) Induction and maintenance infliximab therapy for the treatment of moderate-to-severe Crohn's disease in children. Gastroenterology 132:863-873
17. Walters TD, Gilman AR, Griffiths AM (2007) Linear growth improves during infliximab therapy in children with chronically active severe Crohn's disease. Inflamm Bowel Dis 13:424-430
18. Pfefferkorn M, Burke G, Griffiths A et al (2009) Growth abnormalities persist in newly diagnosed children with crohn disease despite current treatment paradigms. J Pediatr Gastroenterol Nutr 48:168-174
19. Bousvaros A, Antonioli DA, Colletti RB et al (2007) Differentiating ulcerative colitis from Crohn disease in children and young adults: report of a working group of the North American Society for Pediatric Gastroenterology, Hepatology, and Nutrition and the Crohn's and Colitis Foundation of America. J Pediatr Gastroenterol Nutr 44:653-674
20. Paolantonio P, Ferrari R, Vecchietti F et al (2009) Current status of MR imaging in the evaluation of IBD in a pediatric population of patients. Eur J Radiol 69:418-424
21. Grossman AB, Baldassano RN (2008) Specific considerations in the treatment of pediatric inflammatory bowel disease. Expert Rev Gastroenterol Hepatol 2:105-124
22. Rutgeerts P, Vermeire S, Van Assche G (2009) Biological therapies for inflammatory bowel diseases. Gastroenterology 136:1182-97
23. Benchimol EI, Seow CH, Steinhart AH et al (2008) Traditional corticosteroids for induction of remission in Crohn's disease. Cochrane Database Syst Rev 16(2):CD006792
24. Markowitz J, Hyams J, Mack D et al (2006) Corticosteroid therapy in the age of infliximab: acute and 1-year outcomes in newly diagnosed children with Crohn's disease. Clin Gastroenterol Hepatol 4:1124-1129
25. Markowitz J, Grancher K, Kohn N et al (2000) A multicenter trial of 6-mercaptopurine and prednisone in children with newly diagnosed Crohn's disease. Gastroenterology 119:895-902
26. Punati J, Markowitz J, Lerer T et al (2008) Effect of early immunomodulator use in moderate to severe pediatric Crohn disease. Inflamm Bowel Dis 14:949-954
27. Rufo PA, Bousvaros A (2006) Current therapy of inflammatory bowel disease in children. Paediatr Drugs 8:279-302
28. Rosh JR, Gross T, Mamula P et al (2007) Hepatosplenic T-cell lymphoma in adolescents and young adults with Crohn's disease: a cautionary tale? Inflamm Bowel Dis 13:1024-1030
29. Cucchiara S, Escher JC, Hildebrand H et al (2009) Pediatric inflammatory bowel diseases and the risk of lymphoma: should we revise our treatment strategies? J Pediatr Gastroenterol Nut 48:257-267
30. Zachos M, Tondeur M, Griffiths AM (2007) Enteral nutritional therapy for induction of remission in Crohn's disease. Cochrane Database Syst Rev 24(1):CD000542
31. Issa M, Binion DG (2008) Bowel rest and nutrition therapy in the management of active Crohn's disease. Nutr Clin Pract 23:299-308
32. El-Matary (2009) Enteral nutrition as a primary therapy of Crohn's disease: the pediatric perspective. Nutr Clin Pract 24:91-97

Perianal Disease

16

L. Alessandroni

Introduction

Perianal Crohn's disease (PACD) refers to the involvement of the perianal and perineal region in the inflammatory process of the disease. Perianal disease is a frequent complication of Crohn's disease (CD), can result in morbidity and low quality of life, and its management is difficult and controversial [1]. Despite recent advances in the diagnosis and treatment of PACD, the primary goals of management remain unchanged: relief of symptoms, control of sepsis, and avoidance of permanent stoma. The complete healing of perianal lesions remains a goal for the future.

The incidence of perianal disease in Crohn's patients, as reported in the literature, ranges from 15 to 43%. In one-third of patients, it is associated with ileal disease, and in one-half of the patients with colorectal disease [2]. The incidence of PACD in rectal involvement of the disease increases to 90%. In two population-based studies [3,4], the incidence of PACD was 23% (Stockholm County study) and 38% (Olmsted County study).

The transmural inflammation characteristic of CD often results in the formation of fistulas [5]. Perianal fistulas may arise from inflamed anal glands and/or penetration of ulcers of the rectum or anal canal. PACD most commonly presents with perianal abscesses and fistulas, causes significant morbidity, and significantly jeopardizes patients' quality of life. Luminal and cutaneous bacteria may play a role in the development and maintenance of fistulas in CD [6]. Perianal fistulas represent the first manifestation of CD in approximately 10% of patients and in 5% they represent the only symptom of the disease [7]. Anovulvar and rectovaginal fistulas occur in 3–10% of women affected by CD and are challenging to treat. PACD may also present with anal fissures and ulcers, skin tags and hemorrhoids, fecal incontinence, anal stenosis, and, rarely, anal cancer [8].

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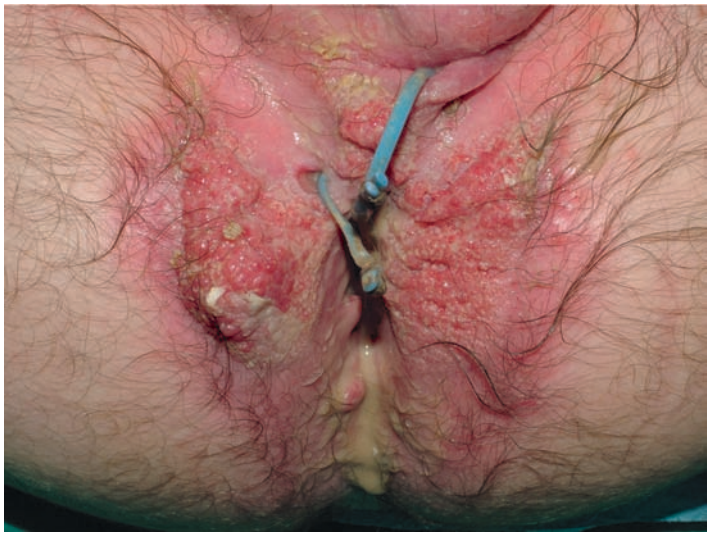

Fig. 16.1 Complex perianal Crohn's disease, with multiple fistulas and abscesses, ulcerated skin tags, and disruption of sphincters

PACD is a predictive factor for the development of severe CD [9] and is frequently associated with intestinal fistulization, especially in the presence of Crohn's colitis [10]. Perianal disease can be highly disabling for patients, in terms of symptoms, hospitalization, and the need for surgery (Fig. 16.1). Significant variables for disabling PACD are age at onset below 40 years, presence of perianal lesions at diagnosis, and a requirement for steroids for treating the first flare [11].

Diagnosis

Accurate evaluation of PACD, especially perianal fistulas, is essential to providing effective and safe treatment for these patients. Clinicians should have a high index of suspicion for PACD in any patient suffering from CD, and a high risk of suspicion of CD in patients with complex and recurrent fistulas, edematous skin tags, or anal/perianal ulcers [12]. An accurate anal and perianal examination should be performed routinely in all patients with CD, inspecting the region for evidence of indurations, erythema, fistulous openings, multiple or eccentric fissures, ulcerations, edematous skin tags, pus discharge, pain, fluctuance, stenosis, and masses. Examination under anesthesia (EUA) demands surgical attendance, constitutes the gold standard for anatomical definition, and can be supplemented with imaging techniques (Fig. 16.2).

In the presence of known or suspected PACD, diagnostic tools are frequently focused on defining the type, location, and nature of perianal involvement. Diagnostic tools have improved over the last decade, which has changed the goal of

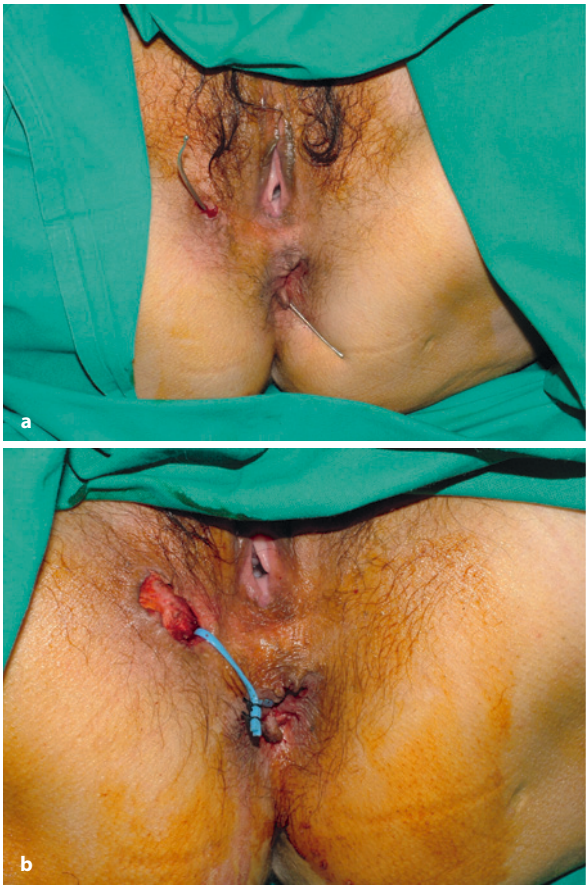

Fig. 16.2 Examination under anesthesia (EUA) in perianal Crohn's disease: **a** identification of the fistula tract; **b** placement of loose seton

treatment from symptomatic improvement to complete cessation of drainage and, in selected cases, to fistula healing. An accurate understanding of fistula anatomy is essential, and the first priority of the treating physician is to define the anatomy of the PACD. Incomplete or inadequate identification of the fistula and the associated pathology can result in poor clinical outcomes [2].

Transanal endoscopic ultrasound (EUS) is a highly sensitive technique for the detection of abscesses and fistula tracts, especially in combination with the use of hydrogen peroxide, but the interpretation of the examination is user-dependent and it cannot be used in patients with severe anal tenderness [13,14]. A new modality of EUS is static transperineal ultrasound, in which an ultrasound transducer is applied directly on the perineal skin. The procedure is less invasive than the former and can be used also in patients with anal pain and stenosis [15].

In the pre-treatment assessment of PACD, colonoscopy is essential to identify active colorectal disease, to perform biopsies on lesions of the colon-rectum and anal canal, and to view internal openings of fistula tracts. Fistulography, contrast enema,

and defecography are helpful in localizing fistula tracts, associated abscesses, or secondary fistulas, but they cannot define the relationships with the anal sphincter, and their accuracy is < 50%. Fistulography is painful and can disseminate septic content; therefore, this technique should no longer be performed [16]. Classically, computed tomography is considered not useful in the diagnosis of PACD, as it has an accuracy of < 60% and the spatial resolution of the pelvis is poor [17].

Pelvic and perineal magnetic resonance imaging (MRI), performed with phased-array or an endoanal coil, can be used to classify perianal fistulas and shows high diagnostic accuracy [18]. A prospective study found that the accuracy of MRI, EUS, and EUA in classifying PACD was 87, 91, and 91%, respectively [19]. The authors suggested that a combination of MRI or EUA with EUS provided an accuracy of 100%. Buchanan et al. [20] reported a linear trend in the proportion of correctly classified tracts of 90% for MRI, 81% for endoanal ultrasound, and 61% for EUA.

Classification

At present, there are no validated anatomical or clinical classifications of PACD. The most commonly used anatomical classification of fistulas is that proposed by Parks et al. in the mid-1970s [21], in which the external anal sphincter is the anatomical point of reference for the description of fistulous tracts. Perianal fistulas can be classified as *low* (below the dentate line) or *high* (above the dentate line).The fistula is further considered *superficial* if the tract lies below the internal and external anal sphincter; *intersphincteric*, if the fistula tracks between the internal and external anal sphincters; *trans-sphincteric*, if the fistula tracks from the intersphincteric space through the external anal sphincter; *suprasphincteric*, in which the fistula leaves the intersphincteric space over the top of the puborectalis and penetrates the levator muscle before tracking down the skin; and *extrasphincteric,* if the fistula tracks outside of the external sphincter and penetrates the levator muscle into the rectum (Table 16.1). Superficial, intersphincteric, and low trans-sphincteric fistulas, without extensions and/or abscesses and anal strictures, are considered *simple*; those that are high trans-sphincteric, suprasphincteric, extrasphincteric, multiple, and recurrent are considered *complex* [22]. Complex fistulas carry a worse prognosis in relation to the need for surgery and its extent, but no medical trials have stratified fistulas regarding their complexity.

The Cardiff classification is a TNM-like classification in which each major manifestation of PACD, (ulcer, fistula, and strictures) is graded along a scale of 0–2. A modification of the original classification added associated anal lesions, intestinal location of CD, and activity of PACD [23]. Nonetheless, this classification has never gained widespread acceptance in clinical practice.

The most comprehensive and frequently applied measure of PACD is the perianal disease activity index (PDAI), which evaluates five categories affected by perianal disease, including discharge, pain, sexual activity, type of disease, and degree of induration [24]. Each category is graded on a 5-point scale, ranging from no symptoms (score 0) to severe symptoms (score 4). However, the PDAI has yet to be validated.

Table 16.1 Patterns of main lesions at first examination under anesthesia (EUA) of 180 patients with perianal Crohn's disease (Surgical Department, San Camillo Hospital, Rome, Italy, 1998–2008)

Lesions	N (%)
Fistulas	
Superficial/intersphincteric	17 (9.5)
Trans-sphincteric	81 (45.0)
Extrasphincteric/suprasphincteric	11 (6.2)
Anovulvar/rectovaginal	18 (10.0)
Rectourethral	2 (1.1)
Multiple	6 (3.3)
Abscesses	
Ischiorectal	8 (4.4)
Horseshoe	5 (2.8)
Perirectal/supralevator	4 (2.2)
Hemorrhoids/skin tags	6 (3.3)
Ulcerations/fissures	19 (10.5)
Stenosis	3 (1.7)
Total	180

In the Montreal modification of the Vienna classification, PACD was added as separate sub-classification, because it is not ever associated with internal fistulizing disease [25].

Medical Treatment

State of the Art

Treatment options combine medical and surgical management. A multi-disciplinary approach, involving the surgeon, gastroenterologist, radiologist, pathologist, nutritionist, and, naturally, the patient, is essential for successful treatment of PACD.

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However, the nature of CD is such that it is currently unrealistic to expect any therapy to be curative. Instead, the main goal of both medical and/or surgical treatments is to improve and maintain patients' quality of life. The short- and long-term management of PACD depends on knowledge of the type and location of the lesions and the severity of the symptomatology. In the St Mark's Hospital's experience, perianal fistulas in CD need a median of three surgical treatments and 2.6 years to heal [26].

Active PACD is often associated with active luminal disease. The initial aim should be to treat active disease and sepsis. For complex PACD, the therapeutic approach involves definition of the anatomy, support of the patient's nutritional state, as well as medical and potentially surgical treatment. For medical treatment, the use of antibiotics (metronidazole, ciprofloxacin), immunosuppressive agents (azathioprine, 6-mercaptopurine), and infliximab has grade A relevance in evidence-based reviews and guidelines [27,28]. Metronidazole and/or ciprofloxacin are appropriate first-line treatments for simple sepsis and fistulas, as adjuvant therapy for the local and general sepsis caused by the fistula, and as maintenance treatment in medically or surgically treated PACD [29–31]. The disease generally responds to antibiotics after 6–8 weeks and treatment is usually continued for 3–4 months. Due to the need for prolonged administration of these drugs, attention must be paid to potential adverse effects. Moreover, due to the risk of persistent neuropathy even after metronidazole is stopped, the drug should be discontinued [22]. In a prospective open-label trial, antibiotics were used as a bridge therapy with azathioprine. Better long-term results were observed in the group treated with immunosuppressants after antibiotic treatment [32]. No long-term healing data support the use of antibiotics as maintenance treatment. A recent randomized pilot study compared metronidazole, ciprofloxacin, and placebo [33]; albeit in the absence of statistical significance, ciprofloxacin resulted in more responses and remissions of PACD as well as better tolerance. The effect of combination treatment with ciprofloxacin and infliximab was assessed in another trial [34], which found that the outcome of fistula healing was not improved by the addition of ciprofloxacin to infliximab, compared with infliximab alone. In conclusion, antibiotics are effective for improving the symptoms of the disease but rarely induce complete healing, and relapse is the rule after withdrawal of these drugs [28].

Azathioprine (1.5–2.5 mg/kg/day) or 6-mercaptopurine (0.75–1.5 mg/kg/day) is potentially effective for the treatment of simple fistulas, in the absence of active sepsis [35]. A meta-analysis of five controlled trials published in the 1970s and 1980s demonstrated that 54% of the 41 patients with PACD who received immunosuppressive therapy responded vs. 21% of the patients in the placebo group [36]. Unfortunately, the treatment of PACD was not a primary goal in those studies.

Infliximab is a chimeric murine and human monoclonal antibody to tumor necrosis factor alpha (TNF-α), a cytokine implicated in the mucosal inflammatory process of CD. In a placebo-controlled study, intravenous injection of infliximab (5 mg/kg), administered as a 3-dose regimen at 0, 2, and 6 weeks, was able to control perianal sepsis and to induce closure of the draining fistulas' external orifices [37]. In this study, the primary efficacy end-point was an achievement of at least 50% reduction from baseline in the number of draining fistulas for at least two consecutive clinical

evaluations, with the observation of no draining fistulas upon gentle digital compression. Complete closure of all fistulas was considered a secondary end-point. The primary end-point was achieved in 68% of patients compared with 26% of those in the placebo control group ($P = 0.0002$), while the secondary end-point was demonstrated in 46% vs. 13% of patients, respectively ($P = 0.001$).

In the ACCENT 2 trial, maintenance therapy with a 5 mg/kg dose of infliximab every 8 weeks prolonged the response time [38]. A prolonged response was observed when immunosuppressive agents were included in the protocol [39]. Infliximab is an important drug with some adverse effects, but it has been accepted with a recommendation of grade A as a second-line treatment of severe fistulizing PACD by the British Society of Gastroenterology [27], the American Gastroenterological Association [40], and the European Crohn's and Colitis Organisation [28]. In case of failure of infliximab, the use of azathioprine or 6-mercaptopurine, with antibiotics as adjunctive treatment, is the approach of choice.

Adalimumab was tested in three randomized, double-blind, placebo-controlled trials: CLASSIC 1 [41], GAIN [42], and CHARM [43]. In the two first trials, the drug was not found to be superior to placebo for the treatment of perianal fistulas. In the latter trial, adalimumab was associated with improved fistula healing, as determined at a follow-up of 56 weeks.

The randomized data on the effects of certolizumab pegol in fistula closure are ineffective, and additional investigations are required [44,45].

Current Treatment Approaches

Several treatments have been proposed and studied in uncontrolled case series, such as CsA, enteral or parenteral nutrition, mycophenolate mofetil, methotrexate, thalidomide, granulocyte colony-stimulating factor (sargramostim), methotrexate, and hyperbaric oxygen, but none are recommended for standard practice [28, 40].

The use of calcineurin inhibitors (cyclosporine, tacrolimus) was investigated in an uncontrolled case series. A small placebo-controlled trial showed that oral tacrolimus (0.2 mg/kg/day for one month) was effective at improving but not healing perianal fistulas in CD; in addition, serious adverse effects were reported in the tacrolimus group [46].

A recent Japanese multi-center, randomized, double-blind trial compared oral spherical adsorptive carbon (AST-120) with placebo in the treatment of intractable Crohn's perianal fistulas. The drug has been hypothesized to reverse abnormalities in the luminal environment and in the gut microflora, with the improvement of intestinal immune responses and mucosal repair. Improvement rates were 37% in the treated group vs. 10% in the placebo group, with good tolerance of the drug [47], but long-term results are not yet available.

Local treatment with metronidazole or tacrolimus ointment appeared to be effective in the control of active anal ulcerations in CD, but the efficacy of these drugs in the presence of fistulas is unclear [48,49].

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Surgical Treatment

State of the Art

Surgical treatment of PACD can be divided into two main categories, *urgent treatment*, which is mainly aimed at controlling perineal sepsis by adequate drainage, and *elective treatment* of the perianal sequelae of perianal sepsis. The rate of perianal fistulas requiring surgical treatment is nearly 30% [50]. Due to the absence of controlled trials, the role of surgery in PACD has yet to be defined, and the grade of recommendation remains C in all guidelines [27,28].

In an urgent and emergent setting, EUA is required for diagnosing the source of the sepsis and for draining simple or deep abscesses without concomitant fistulotomy. If a fistula is reached, a loose seton is inserted through the fistula tract to ensure continuous drainage. Premature removal of the seton should be avoided because of the risk of recurrent sepsis [51,52].

Elective surgery includes procedures for non-fistulous complications, such as dilatation of anorectal strictures, and, more commonly, for the treatment and repair of perianal fistulas not responsive to medical treatment. Treating perianal fistulas in CD is challenging because of poor wound healing and the risk of incontinence. Selection of the surgical approach depends on the type of fistula, previous surgical treatments, the severity of symptoms, and current continence.

The rates of spontaneous fistula closure are low, ranging from 6 to 13% in placebo-treated patients in randomized studies [37,38]. Spontaneous closure increases to 50% in simple fistulas [53]. Asymptomatic simple perianal fistulas and low anovulvar fistulas do not need surgical treatment. In the presence of symptomatic simple fistulas, fistulotomy or loose seton placement, associated with antibiotics, is the approach of choice.

A fistulotomy or fistulectomy should be performed only on low and simple fistulas, in patients without active rectal disease, with well-controlled proximal luminal disease, and with adequate continence. The fistula tract should be identified and probed throughout its length. Following preoperative and intraoperative assessment, the fistula is laid open (fistulotomy) or excised (fistulectomy) and the tract is curetted. Anovaginal and rectovaginal fistulas should not be treated with fistulotomy in PACD, even with low fistulas, due to the risk of incontinence and damage to the rectovaginal septum [54]. Simple fistulas, in the absence of active rectal involvement by CD, have an excellent response to fistulotomy, with healing rates of 70–100% [55].

Some studies have demonstrated impaired wound healing after fistulotomy or fistulectomy in the presence of active rectal disease [7]. Several authors recommend a more conservative approach in such patients, with the placement of a loose non-cutting seton rather than fistulotomy, as it provides adequate drainage of the fistula tract and may be left in place for a long time, until the inflammatory process becomes quiescent [55,56]. A fistulotomy or fistulectomy should not be performed in patients with complex perianal fistulas because of the significant risk of anal incontinence, due to transection of both the internal and external anal sphincters.

The best results and outcome have been achieved when surgical and medical therapies are used in conjunction. This allows for the control of fistula healing during medical treatment. In fact, the rate of abscess formation during treatment with infliximab is high (11–15%) and the rate of prolonged fistula closure is relatively low (36%) [37,38,57]. Two studies demonstrated that patients submitted to EUA before infliximab treatment were significantly less likely to have fistula recurrence compared with patients who did not require surgical drainage [58,59]. The combination of endovenous infliximab and seton drainage was shown to heal the fistulas in up to 47% of patients [60], but the long-term healing rates are low [61].

Occasionally, temporary diverting ileostomy or colostomy is required to control symptoms and to induce the remission of severe perianal lesions [62,63]. A diverting stoma can rapidly restore the quality of life in highly symptomatic patients [28]. However, restoring the intestinal passage carries a high risk of recurrent fistulas and sepsis, possibly resulting in a decreased quality of life compared to treatment by fecal diversion.

Active intestinal disease should be appropriately treated with the aim of reducing diarrhea, which irritates the perineal area. Some authors have suggested that, if PACD persists in the presence of active luminal disease, resection of the involved bowel can improve the perineal manifestations—but practical use of this approach is controversial [64,65] and PACD alone is not an indication for intestinal resection.

In extremely severe cases resistant to both medical and surgical therapy, proctectomy or proctocolectomy, with permanent ileostomy, may be required [66]. The risk of permanent fecal diversion is substantial in patients with complex PACD, active colorectal disease, and/or fecal incontinence, while there is no or only very low risk in patients with PACD who require surgery for small bowel disease or segmental colon resection [67]. In complex PACD, diversion before proctectomy may allow healing of perianal lesions and it avoids the risk of a persistent non-healed perineal wound, frequently associated with the procedure.

The endorectal advancement flap is a surgical technique for repairing complex fistulas, with preservation of a continent sphincter. Closure of the fistula's internal opening is achieved using a rectal flap; therefore, the procedure is not possible in the presence of active rectal disease [68, 69]. The main indications of the procedure are anterior and rectovaginal fistulas [70]. The reported average success rate in patients treated with endorectal advancement flap, as reported in the indexed literature, is about 50%, ranging from 25 to 100%.

In patients with rectovaginal symptomatic fistulas, surgery is usually necessary. According to the ECCO's guidelines, rectovaginal fistulas not responding to conservative treatment should be submitted to an advancement flap and/or diverting stoma if they are associated with major symptomatology [28].

Current Treatment Approaches

Instillation of fibrin glue, a blood product derived from the activation of thrombin to a fibrin clot, is a simple procedure for sealing perianal fistulas. The fibrin clot induces tissue-healing processes while being gradually absorbed by fibrinolysis. In a

series of mixed cases, success rates of 30–70% were reported [71]. However, fibrin glue does not have an established role in the treatment of PACD [72].

A bioprosthetic extracellular matrix plug, made from lyophilized porcine intestinal collagen, has been proposed for the closure of anal fistula tracks, with an 86% healing rate of CD-associated fistulas and a reoperation rate of 28% [73,74]. The success rate of the plug was reduced in fistulas with multiple tracts [75].

Recently, gracilis muscle transposition has been proposed as a therapeutic option, in experienced centers, to treat complex perianal fistulas. Wexner et al. [76] reported a 33% healing rate in CD-associated complex fistulas.

Cornillie et al. [77] demonstrated that the clinical efficacy of infliximab is based on the drug's local anti-inflammatory and immunomodulatory effects in the bowel mucosa, down-regulating adhesion molecules within the lamina propria but without generalized suppression of systemic immune functions in CD patients. Therefore, infiltration of infliximab through the mucosal barrier should enhance its effectiveness. Based on these data, Lichtiger [78] proposed the local injection of infliximab as an alternative to systemic infusion in patients with mild to moderate PACD refractory to antibiotics and/or immunosuppressants. Nine patients were treated with a circumferential and intra-fistula injection of infliximab, with a 50% complete response, 33% partial response, and 17% no response. Poggioli et al. [79] modified the technique, employing a wide fistulectomy and injection of infliximab in the internal orifice to avoid false closure of the tract. The authors thus treated 15 patients with severe PACD resistant to previous surgery and medical treatments. Complete healing of the fistulae, tested with probe examination, was achieved in 66% of the patients while 20% had a partial response, with control of sepsis but persisting pus discharge; the condition of 14% of the patients remained unchanged. The authors concluded that local injection of infliximab is effective in patients with complex perianal disease untreatable with surgery alone and unable to receive intravenous infliximab. In another study [80], local infliximab was administered to 11 patients with PACD. After a mean follow-up of 10.5 months, 36.4% of the patients were in remission (complete cessation of fistula drainage), 54.5% had a partial response (reduction in fistula drainage of 50% or more), and 9.1% remained unchanged.

The use of autologous stem cells, obtained from fat and injected around fistula tracts, was recently proposed. The rate of fistula healing after treatment with local injection of stem cells plus fibrin glue, in a phase II study, was about 70% in 14 patients with complex PACD [81]. A randomized multi-center trial (FATT-1) is currently ongoing.

Evaluation of the Response

Within the indexed literature, various definitions have been used to define fistula healing. No widely accepted and validated definitions or scores are available for assessing the response of PACD to various treatments, with high variability from study to study [82]. The more commonly used clinical classification to therapy is that

of Present et al. [37], which defines healing as a cessation of drainage despite gentle pressure. This evaluation can vary from investigator to investigator, and other authors have designed their own scoring systems [79].

According to ECCO statements, in the evaluation of the response to medical or surgical treatment in routine practice, clinical assessment is usually sufficient. In the setting of clinical trials, MRI alone or in combination with clinical assessment is considered mandatory [28].

PACD and Carcinoma

Fistula-associated anal carcinoma must be suspected in all patients with long-standing PACD. The presence of carcinoma along a perianal fistula tract is rare but must be considered in patients with PACD, and must be cured with aggressive surgical and adjuvant chemoradiotherapy [83].

References

1. Rutgeerts P (2004) Review article: treatment of perianal fistulising Crohn's disease. Aliment Pharmacol Ther 20 (Suppl 4):106–110
2. Nielsen OH, Rogler G, Hanholer D, Thomsen OØ (2009) Diagnosis and management of fistulising Crohn's disease. Nat Clin Pract Gastroenterol Hepatol, In press
3. Hellers G, Bergstrand O, Ewerth S, Holstrom B (1980) Occurrence and outcome after primary treatment of anal fistulae in Crohn's disease. Gut 21:525–527
4. Schwartz D, Loftus EV Jr, Tremaine W et al (2002) The natural history of fistulising Crohn's disease in Olmsted County, Minnesota. Gastroenterology 122:525–527
5. Judge TA, Lichtenstein GR (2004) Treatment of fistulising Crohn's disease. Gastroenterol Clin North Am 33:421–454
6. West RL, Van der Woude CJ, Endtz HP et al (2005) Perianal fistulas in Crohn's disease are predominantly colonized by skin flora: implications for antibiotic treatment? Dig Dis Sci 50:1260–1263
7. Nordgren S, Fasth S, Hulten L (1992) Anal fistulas in Crohn's disease: incidence and outcome of surgical treatment. Int J Colorectal Dis 7:214–218
8. Williamson PR, Hellinger MD, Larach SW, Ferrara A (1995) Twenty-year review of the surgical management of perianal Crohn's disease. Dis Colon Rectum 38:389–392
9. Loly C, Belaiche J, Louis E (2008) Predictors of severe Crohn's disease. Scand J Gastroenterol 4:1–8
10. Sachar DB, Bodian CA, Goldstein ES et al (2005) Is perianal Crohn's disease associated with intestinal fistulisation? Am J Gastroenterol 100:1547–1549
11. Beaugerie L, Seksik P, Nion–Larmurier I et al (2006) Predictors of Crohn's disease. J Gastroenterology 130:650–656
12. Bonheur JL, Braunstein J, Korelitz BI et al (2008) Anal skin tags in inflammatory bowel disease:new observations and a clinical review. Inflamm Bowel Dis 14:1436–1439
13. Orsoni P, Barthet M, Portier F et al (1999) Prospective comparison of endosonography, magnetic resonance imaging and surgical findings in anorectal fistula and abscess complicating Crohn's disease. Br J Surg 25:1–20

14. Sloots C, Felt-Bersma RJ, Poen AC et al (2001) Assessment and classification of fistula-in-ano in patients with Crohn's disease by hydrogen peroxide enhanced transanal ultrasound. Int J Colorectal Dis 16:292–297
15. Kleinubing H Jr, Jannini JF, Malafaia O et al (2000) Transperineal ultrasonography: a new method to image the anorectal region. Dis Colon Rectum 43:1572–1574
16. Weisman RI, Orsay CP, Pearl RK, Abcarian H (1991) The role of fistulography in fistula-in-ano. Dis Colon Rectum 34:181–184
17. Schratter–Sehn AU, Lochs H, Vogelsang H et al (1993) Endoscopic ultrasonography versus computed tomography in the differential diagnosis of perianorectal complications of Crohn's disease. Endoscopy 25:582–586
18. Van Assche G, Vanbeckevoort D, Bielen D et al (2003) Magnetic resonance imaging of the effects of infliximab on perianal fistulising Crohn's disease. Am J Gastroenterol 98:332–339
19. Schwartz DA, Wiersema MJ, Dudiak KM et al (2001) A comparison of endoscopic ultrasound, magnetic resonance imaging, and exam under anaesthesia for evaluation of Crohn's perianal fistulas. Gastroenterology 121:1064–1072
20. Buchanan GN, Halligan S, Bartram CI et al (2004) Clinical examination, endosonography, and MR imaging in preoperative assessment of fistula in ano: comparison with outcome-based reference standard. Radiology 233:674–681
21. Parks AG, Gordon PH, Hardcastle JD (1976) A classification of fistula-in-ano. Br J Surg 63:1–2
22. Schwartz DA, Pemberton JH, Sandborn WJ (2001) Diagnosis and treatment of perianal fistulas in Crohn's disease. Ann Intern Med 135.906–918
23. Hughes LE (1992) Clinical classification of perianal Crohn's disease. Dis Colon Rectum 35:928–932
24. Irvine EJ (1995) Usual therapy improves perianal Crohn's disease as measured by a new disease activity index. J Clin Gastroenterol 20:27–32
25. Satsangi J, Silverberg MS, Vermeire S, Colombel JF (2006) The Montreal classification of inflammatory bowel disease: controversies, consensus, and implications. Gut 55:749–753
26. Bell SJ, Williams AB, Wiesel P et al (2003) The clinical course of fistulising Crohn's disease. Aliment Pharmacol Ther 17:1145–1151
27. Carter MJ, Lobo AJ, Travis SPL (2004) Guidelines for the management of inflammatory bowel disease in adults. Gut 53 (Suppl V):v1–v16
28. Caprilli R, Gassull MA, Escher JC et al (2006) European evidence based consensus on the diagnosis and management of Crohn's disease: special situations. Gut 55:i36–i58
29. Brandt LJ, Berstein LH, Baley SJ et al (1982) Metronidazole therapy for perineal Crohn's disease:a follow-up study. Gastroenterology 83:383–387
30. Jacobovits J, Schuster MM (1984) Metronidazole therapy for Crohn's disease and associated fistulae. Am J Gastroenterol 79:533–540
31. Solomon MJ, McLeod RS, O'Connor BJ et al (1993) Combination ciprofloxacin and metronidazole in severe perianal Crohn's disease. Can J Gastroenterol 7:571–573
32. Dejaco C, Harrer M, Waldhoer T et al (2003) Antibiotics and azathioprine for the treatment of perianal fistulas in Crohn's disease. Aliment Pharmacol Ther 18:1113–1120
33. Thia KT, Mahadevan U, Feagan BG et al (2009) Ciprofloxacin of metronidazole for the treatment of perianal fistulas in patients with Crohn's disease: a randomized, double blind, placebo-controlled pilot study. Inflamm Bowel Dis 15:17–24
34. West RL, Van der Woude CJ, Hansen BE et al (2004) Clinical and endosonographic effect of ciprofloxacin treatment of perianal fistulae in Crohn's disease with infliximab: a doubleblind placebo-controlled study. Alimen Pharmacol Ther 20:1329–1336
35. Korelitz BI, Adler DJ, Mendhelsohn RA et al (1993) Long-term experience with 6-mercaptopurine in the treatment of Crohn's disease. Am J Gastroenterol 88:1198–1205
36. Pearson DC, May GR, Fick GH et al (1995) Azathioprine and 6-mercaptopurine in Crohn's disease. A meta-analysis. Ann Intern Med 122:132–142

37. Present DH, Rutgeerts P, Targan S et al (1999) Infliximab for the treatment of fistulas in patients with Crohn's disease. N Eng J Med 340:1398–1405
38. Sands BE, Anderson FH, Bernstein CN et al (2004) Infliximab maintenance therapy for fistulizing Crohn's disease. N Eng J Med 350:876–885
39. Ochsenkuhn T, Goke B, Sackmann M (2002) Combining infliximab with mercaptopurine/azathioprine for fistula therapy in Crohn's disease. Am J Gastroenterol 97:2022–2025
40. Sandborn WJ, Fazio VW, Feagan BG, Hanauer SB (2003) AGA technical review on perianal Crohn's disease. Gastroenterology 125:1508–1530
41. Hanauer SB, Sandborn WJ, Rutgeerts P et al (2006) Human anti-tumor necrosis factor monoclonal antibody (adalimumab) in Crohn's disease:the CLASSIC-I trial. Gastroenterology 130:323–333
42. Sandborn WJ, Rutgeerts P, Enns R et al (2007) Adalimumab induction therapy for Crohn's disease previously treated with infliximab:a randomized trial. Ann Intern Med 146:829–838
43. Colombel JF, Sandborn WJ, Rutgeerts P et al (2007) Adalimumab for maintenance of clinical response and remission in patients with Crohn's disease:the CHARM trial. Gastroenterology 132:1641–1657
44. Sandborn WJ, Feagan BG, Stoinov S et al (2007) Certolizumab pegol for the treatment of Crohn's disease. N Eng J Med 357:228–238
45. Schreiber S, Khaliq-Kareemi M, Lawrence IC et al (2007) Maintenance therapy with certolizumab pegol for the treatment of Crohn's disease. N Eng J Med 357:239–250
46. Sandborn WJ, Present DH, Isaacs KL et al (2003) Tacrolimus for the treatment of fistulas in patients with Crohn's disease:a randomized, placebo–controlled trial. Gastroenterology 125:380–388
47. Fukuda Y, Takazoe M, Sugita A et al (2008) Oral spheric adsorptive carbon for the treatment of intractable anal fistulas in Crohn's disease: a multicenter, randomized, double-blind, placebo-controlled trial. Am J Gastroenterol 103:1721–1729
48. Stringer EE, Nicholson TJ, Armstrong D (2005) Efficacy of topical metronidazole (10 percent) in the treatment of anorectal Crohn's disease. Dis Colon Rectum 48:970–974
49. Hart AL, Plamondon S, Kamm MA (2007) Topical tacrolimus in the treatment of perianal Crohn's disease. Inflamm Bowel Dis 13:245–253
50. Williams JG, Farrands PA, Williams AB et al (2007) The treatment of anal fistula: ACPGBI position statement. Colorectal Dis 9 (Suppl 4):18–50
51. Makowiec F, Jehle EC, Starlinger M (1995) Clinical course of perianal fistulas in Crohn's disease. Gut 37:696–701
52. Scott HJ, Northover JMA (1996) Evaluation of surgery for perianal Crohn's disease. Dis Colon Rectum 39:1039–1043
53. Halme L, Sainio AP (1995) Factors related to frequency, type, and outcome of anal fistulas in Crohn's disease. Dis Colon Rectum 38:55–59
54. Saclarides TI (2002) Rectovaginal fistula. Surg Clin North Am 1261–1272
55. Williams JG, MacLeod CA, Rothenberger DA, Goldberg SM (1991) Seton treatment of high anal fistulae. Br J Surg 78:1159–1161
56. Larson DW Pemberton JH (2004) Current concepts and controversies in surgery for IBD. Gastroenterology 126:1611–1619
57. Domenech E, Hinojosa J, Nos P et al (2005) Clinical evolution of luminal and perianal Crohn's disease after inducing remission with Infliximab:how long should patients be treated? Aliment Pharmacol Ther 22:1107–1113
58. Regueiro M, Mardini H (2003) Treatment of perianal fistulizing Crohn's disease with infliximab alone or as an adjunct to exam under anaesthesia with seton placement. Inflamm Bowel Dis 9:98–103
59. Topstad DR, Panaccione R, Heine JA et al (2003) Combined seton placement, infliximab infusion, and maintenance immunosuppressives improve healing rate in fistulising anorectal

Crohn's disease:a single center experience. Dis Colon Rectum 46:577–583
60. Talbot C, Sagar PM Johnston MJ et al (2005) Infliximab in the surgical management of complex fistulating anal Crohn's disease. Colorectal Dis 7:164–168
61. Hyder SA, Travis SP, Jewell DP et al (2006) Fistulating anal Crohn's disease:results of combined surgical and Infliximab treatment. Dis Colon Rectum 49:1837–1841
62. Burman JM, Thompson M, Cooke WT, Alexander–Williams J (1971) The effects of diversion of intestinal contents on the progress of Crohn's disease of the large bowel. Gut 2:11–15
63. Frizelle FA, Santoro GA, Pamberton JH (1996) The management of perianal Crohn's disease. Int J Colorectal Dis 11:227–237
64. Heuman R, Bolin T, Sjodhal R et al (1981) The incidence and course of perianal complications and arthralgia after intestinal resection with restoration of continuity for Crohn's disease. Br J Surg 68:528–530
65. Orkin BA, Telander RL (1985) Effect of intra-abdominal resection or fecal diversion of perianal Crohn's disease in pediatric Crohn's disease. J Pediatr Surg 20:343–347
66. Fichera A, McCormack R, Rubin MA et al (2005) Long-term outcome of surgically treated Crohn's colitis: a prospective study. Dis Colon Rectum 48:936–939
67. Mueller MH, Geis M, Glatzle J et al (2007) Risk of faecal diversion in complicated perianal Crohn's disease. J Gastrointest Surg 11:529–537
68. Joo JS, Weiss EG, Nogueras JJ, Wexner SD (1998) Endorectal advancement flap in perianal Crohn's disease. Am Surg 64:157–160
69. Kodner IJ, Mazor A, Shemesh EI et al (1993) Endorectal advancement flap repair of rectovaginal and other complicated anorectal fistulas. Surgery 114:682–690
70. O'Leary DP, Durdey P, Milroy CE (1998) Definitive repair of anovaginal fistula in Crohn's disease. Ann Royal Coll Surg Engl 64:147–150
71. Zmora O, Mizrhai N, Rotholtz N et al (2003) Fibrin glue sealing in the treatment of perineal fistulas. Dis Colon Rectum 46:584–589
72. Loungnarath R, Dietz DW, Mutch MG et al (2004) Fibrin glue treatment of complex anal fistulas has low success rate. Dis Colon Rectum 47:432–436
73. Committee Consensus Statement (2008) The Surgisis® AFPTM anal fistula plug: report of a consensus conference. Colorectal Dis 10:17–20
74. Schwandner O, Stadler F, Dietl O et al (2008) Initial experience on efficacy in closure of cryptoglandular and Crohn's transphinteric fistulas by the use of the anal fistula plug. Int J Colorectal Dis 23:319–324
75. O'Connor L, Champagne BJ, Ferguson MA et al (2006) Efficacy of anal fistula plug in closure of Crohn's anorectal fistulas. Dis Colon Rectum 49:1569–1573
76. Wexner D, Ruiz DE, Genua J et al (2008) Gracilis muscle interposition for the treatment of rectourethral, rectovaginal, and pouch-vaginal fistulas: results in 53 patients. Ann Surg 248:39–43
77. Cornillie F, Dhealy D, D'Haems G et al (2001) Infliximab induces potent anti-inflammatory and local activity, but no systemic immune suppression in patients with Crohn's disease. Alimen Pharmacol Ther 15:463–473
78. Lichtiger S (2001) Healing of perianal fistulae by local injection of antibody to TNF. Gastroenterology 120(Suppl):A3154
79. Poggioli G, Laureti S, Pierangeli F et al (2005) Local injection of infliximab for the treatment of perianal Crohn's disease. Dis Colon Rectum 48:768–774
80. Asteria CR, Ficari F, Bagnoli et al (2006) Treatment of perianal fistulas in Crohn's disease by local injection of antibody to TNF-alpha accounts for a favourable response in selected cases: a pilot study. Scand J Gastroenterol 41:1064–1072
81. Garcia-Olmo D, Herreros D, Pascual I et al (2009) Expanded adipose-derived stem cells for the treatment of complex perianal fistula: a phase II clinical trial. Dis Colon Rectum 52:79–86
82. Irvine EJ (1995) Usual therapy improves perianal Crohn's disease as measured by a new dis-

ease activity index. J Clin Gastroenterol 20:27–32
83. Gaertner WB, Hagerman GF, Finne CO et al (2008) Fistula-associated anal adenocarcinoma:good results with aggressive therapy. Dis Colon Rectum 51:1061–1067

Crohn's Disease and Pregnancy

17

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Introduction

Crohn's disease (CD) can involve any area of the gastrointestinal tract, but the distal small intestine, colon, and anal-rectal region are most often affected. The disease consists of a granulomatous inflammatory process of the bowel mucosa extending to the deeper layers, with possible transmural involvement. About 30% of patients require surgery during the first year after the diagnosis is made; thereafter 5% of all patients per year require surgery. Complications such as fistula formation and perineal communication interfere with vaginal delivery.

Crohn's disease can occur at any age, but it is most common between the ages of 15 and 30 and thus often affects women of childbearing age. While women with CD can expect to conceive successfully, carry to term, and deliver a healthy baby, many issues arise about the impact of CD on pregnancy.

Effects on Fertility

Men with CD who are taking sulfasalazine may be temporarily infertile because in 60% sperm count is decreased. This effect is reversed within 2 months of drug suspension; switching to 5-ASA drugs is advisable. Proctocolectomy in men can cause impotence, but this is rare.

Crohn's disease seems to affect children more severely than adults. A child with CD may have slower growth and delayed sexual development. In addition, nutritional complications are common in CD, which, in women, can lead to ovulatory problems.

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Active CD raises the risk of miscarriage. While the overall infertility rate (12%) is similar to that seen in the general population, patients who had active disease at the time of conception have a high rate (35%) of spontaneous abortion. Some patients with extensive perienteric inflammation experience complications due to concomitant adnexal involvement [1].

Women who have had ileostomies may have lower fertility rates. According to one review of the literature, post-operative infertility occurs in 48% of women surgically treated for ulcerative colitis [2]. This is presumably due to scarring of the fallopian tubes. There are similar reports of infertility in CD patients. Indeed, major surgery in pregnant women with CD for acute complications of the disease may carry a 50% risk of miscarriage.

Pregnancy Planning

There is general agreement that conception should be avoided when CD is active [2]; the presence of diarrhea and abdominal pain are good indicators. If the disease is in remission at the time of conception, the chance of a normal baby is 85% for those with ulcerative colitis (UC) and 83.5% for those with CD, comparable to the rates in the general population.

As in the general population, tobacco consumption should be avoided.

Steroid therapy should be modified to the lowest dose possible or avoided entirely if pregnancy is a possibility. Vitamins can be useful and should not be discontinued if a pregnancy occurs. Adequate intake of folic acid prevents neural tube birth defects such as spina bifida. Sulfasalazine blocks the absorption of folic acid.

Methotrexate can cause abortion and skeletal abnormalities; it should be discontinued 3 months prior to conception, if possible. It should not be taken during pregnancy either by a woman who is trying to conceive or by the potential father.

Ileostomy patients should wait for a year after surgery to become pregnant in order to reduce the risk of the ileostomy dropping or becoming blocked during pregnancy.

Sparse observations indicate that intestinal disease has a smoother course during pregnancy in patients who had previously undergone surgery for disease treatment [3].

Effects of Crohn's Disease on Pregnancy

The rate of preterm deliveries is higher in women with a subsequent diagnosis of CD than in controls and the birth weight of children of mothers who later developed CD is lower than that of controls. Most likely, factors that might predispose to preterm delivery (inflammation, nutritional deficiencies, immunological or neurological disturbances) were already present in the future CD patients or certain etiological factors may predispose both to CD and to preterm delivery and low birth weight.

There is a strong association between CD and the risk of adverse birth outcomes.

Particularly, large cohort studies [4,5] have shown two- to three-fold increased risks of low birth weight (LBW) and preterm birth in women with inflammatory bowel disease (IBD). LBW and preterm birth are closely associated with increased early and late childhood mortality and morbidity. This may be due to the presence of inflammatory mediators, which stimulate uterine contractions by facilitating the production of prostaglandins. Preterm birth incidence was higher in iatrogenically induced births, possibly because of concerns regarding fetal or maternal well-being [6,7].

The incidence of most pregnancy complications was comparable [3] between patients with CD and controls, but the incidence of poor maternal weight gain differed significantly between the two groups.

Effects of Pregnancy on Crohn's Disease

A study of 70 pregnancies [8] using the Harvey-Bradshaw index indicated that, during pregnancy, disease activity is mildly but significantly lower than during the year preceding pregnancy and during the year following delivery. Likewise, the use of therapeutic drugs was significantly lower during pregnancy. Incidentally, the reduction of tobacco consumption during pregnancy in smokers with CD may play an important role in this improvement.

For patients with CD in remission at conception, about a quarter will suffer disease relapse in the following 12 months, no different than non-pregnant CD patient. For patients with active disease at the time of conception, one-third will improve and one-third will worsen. Flare-up of CD was seen in 13 out of 65 women evaluated (20%) [3]. The rate of relapse in pregnant women with CD is about 25% if they are in remission at the time of conception, according to Danish pregnancy registry data [4]. Patients who had active CD at the time of conception continued to have symptoms and their disease mostly failed to go into satisfactory remission despite therapy.

Genetic differences between mother and child might correlate with disease activity. Testing the DNA of mothers and children has shown that, the more genetically different the baby is from the mother, the better the mother feels during the pregnancy.

Effects on the Fetus

The greatest differences in a comparison of patients with CD and normal controls with respect to neonatal outcomes were in terms of birth weight and birth weight percentile [3]. Overall, the rate of small for gestational age (SGA) births in the CD group was 24.6%, compared with only 1.5% in the control group. Ileal CD was a statistically significant predictor of SGA birth, whereas previous bowel resection trended toward statistical significance.

An often cited [1–6] meta-analysis of 12 prior studies encompassing 3,907 patients with IBD, including 1,952 with CD (63%), and 320,531 controls reported a

1.87-fold increase in the incidence of prematurity. In addition, the incidence of LBW was more than twice that of normal controls. Women with IBD were 1.5 times more likely to undergo caesarean section. The risk of congenital abnormalities was found to be 2.37-fold higher. The greater risk of LBW and caesarean section was significant only in patients with CD. When the analysis was restricted to the higher-quality studies, there was no greater risk of congenital abnormalities.

Management of Crohn's Disease in Pregnancy

Some CD diagnostic procedures can be performed during pregnancy at any time, e.g., colonoscopy, sigmoidoscopy, upper endoscopy, rectal biopsy, and abdominal ultrasound. However, X-ray and computed tomography (CT) scans should be avoided. Magnetic resonance imaging (MRI) appears to be safe during pregnancy, but a risk associated with the procedure cannot be ruled out.

The incidence of poor maternal weight gain differed significantly between patients with CD and normal controls [3], emphasizing the importance of maintaining a healthy, well-balanced diet during pregnancy. Supplements, vitamins, and minerals in conjunction with a regular diet are advisable.

Symptoms of IBD, such as diarrhea or constipation, can increase the risk of hemorrhoids. The typical forms of hemorrhoid management is permissible (Kegel exercises, keeping the anal area clean, avoiding sitting and standing for long periods of time and heavy or moderate lifting, and using suppositories or creams for treatment).

Physicians should treat a severe flare-up of IBD that occurs during an unplanned pregnancy very aggressively. Achieving remission is important to help ensure that the pregnancy is as healthy as possible.

Drug Therapy during Pregnancy

Pharmacological therapy for CD during pregnancy is similar to that for non-pregnant patients. Patients maintained in remission by way of pharmacological therapy should continue their medications throughout pregnancy [9].

According to the FDA's Drug Categories, drugs administered in pregnancy are classified as follows: (A) studies in pregnant women have not shown increased fetal risk; (B) animal studies show no harm to the fetus but there are no adequate studies in pregnant women *or* animal studies show an adverse effect, but well-controlled studies in pregnant women do not show a fetal risk; (C) animals have shown adverse effects but there are no study on pregnant women *or* no data are available for animal or human pregnancies; (D) studies in pregnant women have demonstrated risk to the fetus; (X) studies in animals or pregnant women have demonstrated positive evidence of fetal abnormalities.

Some drugs commonly used for the treatment of IBD are safe for use by pregnant

women (category B). These are: drugs of the aminosalicylate class (5-ASA drugs), sulfasalazine, forms of esalamine (mesalamine, balsalazide, olsalazine), and prednisone, which is the only form of cortisone with very limited transplacental passage.

The immunosuppressive drugs azathioprine (Imuran, category D) and 6-mercaptopurine (purinethol or 6-MP, category D) do cross the placenta. Nonetheless, they are recommended, with caution, by some physicians for women patients during pregnancy who experience a serious disease flare-up. Danish birth registry data show greater risk of preterm birth and congenital abnormalities among patients prescribed AZA/6-MP throughout their pregnancies. In this group, the risk of preterm birth and congenital abnormalities was 4.2 and 2.9, respectively. These drugs are prescribed in more severe cases, so disease activity may have been a confounding factor [5,10].

Cohort studies examining the effect of 6-MP on conception and during pregnancy are less worrisome. One study, involving 155 patients (79 female) with 325 pregnancies, included patients who took 6-MP but stopped the drug before conception (84 conceptions from 40 women and 44 men). This group was compared with patients who conceived on 6-MP (61 pregnancies from 24 women and 37 men), patients who stayed on 6-MP from conception to term (15 conceptions), and patients who had not been exposed to 6-MP prior to pregnancy (165 pregnancies from 92 women and 73 men). Among these groups, there was no statistical difference in conception failures (defined as a spontaneous abortion), abortion secondary to a birth defect, major congenital malformations, neoplasia, or increased infections.

Methotrexate (category X) and thalidomide (category X) are two immunosuppressive drugs that absolutely should not be used during pregnancy as they have serious effects on an unborn child. Methotrexate can cause abortion and skeletal abnormalities, and thus should be discontinued 3 months prior to conception, if possible. Thalidomide is well-known for causing limb defects as well as other major organ complications in a fetus.

Biological drugs, such as infliximab (Remicade, category C) and adalimumab (Humira) seem to be safe for use during pregnancy as well.

Vitamins and supplements should be continued during pregnancy. Sulfasalazine must be associated with folic acid supplementation.

Antibiotics are used to treat bacterial overgrowth in the small intestine caused by stricture, fistulas, or prior surgery. For this common problem, ampicillin, cephalosporin (category A), sulfonamides, or metronidazole can be used. Metronidazole (Flagyl, category B) may not be safe for the fetus after the first trimester. Although one study showed that metronidazole did not cause birth defects in the first trimester, no long-term studies have been conducted. Tetracyclines are best avoided during pregnancy.

Due to the potential side effects of antidiarrheal medications, such as Imodium and Lomotil, pregnant women should avoid these drugs, especially during the first trimester. Use of narcotic antidiarrheals, such as codeine, also should be avoided because of possible newborn addiction.

Drug Therapy and Breastfeeding

Drugs in the aminosalicylate class (5-ASA drugs) allow safe breastfeeding; however, since all show anti-folic-acid effects, supplementation might be in order.

Breastfeeding while taking steroids in moderate to high doses mandates monitoring of the baby by a pediatrician. Breastfeeding should not be allowed during methotrexate treatment. Other immunomodulators and immunosuppressives should be used with care. Biological drugs such as infliximab (Remicade) and adalimumab (Humira) do not appear to be secreted in breast milk [9].

Obstetrical Management

Therapeutic abortions should not be advised, even as a therapeutic option for IBD flare-up during pregnancy. Concern about disease complications or the teratogenic effect of drug treatment are not justified and the presence of a congenital abnormality should be documented [7,10].

Delivery Timing and Route

Elective preterm birth is still the most important single cause of perinatal mortality and morbidity and should be avoided if possible [6].

Women with IBD were 1.5 times more likely to undergo caesarean section such that the rates of the procedure were higher in women with Crohn's disease (20.9%) than in the general population (15%) [11].

Patients with inactive perianal disease had no relapse during 1 year of follow-up postpartum. Of 39 vaginal deliveries in women with no known perianal disease, only one ultimately showed evidence of disease within 1 year of postpartum follow-up. It can thus be concluded that patients with either no history or inactive perianal disease at the time of giving birth have are at very low risk of relapse. Cesarean section is not justified in these patients.

Women who become pregnant and have had previous surgery for IBD, such as colectomy, ileostomy, or one of the newer operations, e.g., continent ileostomy (Kock pouch) or ileoanal anastomosis, should be able to have a normal vaginal delivery, which is preferable to a Caesarean section unless there are obstetrical reasons to perform the latter. Women with active perineal disease (e.g., rectovaginal fistulas or abscesses) are strongly recommended to undergo Caesarean section.

During the course of the pregnancy, enlargement of the abdomen may cause some problems with the size of the ileostomy, necessitating a larger appliance. Also, a prolapse of the ileostomy may occur. As the pregnancy progresses, however, a woman who has undergone ostomy surgery should avoid wearing a belt, which can increase

abdominal pressure and cause further prolapse of the ileostomy. At the end of the third trimester, the ileostomy may be displaced by the uterus, leading to partial obstruction of the small intestine. This may require the induction of labor earlier than the expected date of delivery.

Disease Inheritance

Crohn's disease does run in families, especially in Jewish families whereas Africans seem less affected. There is no sex prevalence. Children who have one parent with CD have only a 7–9% lifetime risk of developing the condition, and just a 10% risk of developing a form of IBD. If both parents have IBD, this risk is increased to about 35% [1,5,10].

Individuals with CD have a 20% chance of having an affected relative.

References

1. Katz JA (2004) Pregnancy and inflammatory bowel disease. Curr Opin Gastroenterol 20:328–332
2. Sela HY, Rojansky N, Hershko AY (2005) Reproduction and ulcerative colitis: a review. J Reprod Med 50:361–366
3. Moser MA, Okun NB, Mayes DC et al (2000) Crohn's disease pregnancy, and birth weight. Am J Gastroenterol 95:1021-1026
4. Fonager K, Sorensen HT, Olsen J et al (1998) Pregnancy outcome for women with Crohn's disease: A follow-up study based on linkage between national registries. Am J Gastroenterol 93:2426–2430
5. Dominitz JA, Young JC, Boyko EJ (2002) Outcomes of infants born to mothers with inflammatory bowel disease: a population based cohort study. Am J Gastroenterol 97:641–648
6. Elbaz G, Fich A, Levy A et al (2005) Inflammatory bowel disease and preterm delivery. Int J Gynaecol Obstet 90:193–197
7. Bush MC, Patel S, Lapinski RH et al (2004) Perinatal outcomes in inflammatory bowel disease. J Matern Fetal Neonatal Med 15:237–241
8. Agret F, Cosnes J, Hassani Z et al (2005) Impact of pregnancy on the clinical activity of Crohn's disease. Aliment Pharmacol Ther 21:509:513
9. Mottet C, Juillerat P, Pittet V et al (2007) Pregnancy and breastfeeding in patients with Crohn's disease. Digestion 76:149-160
10. Ludvigsson JF, Ludvigsson J (2002) Inflammatory bowel disease in mother or father and neonatal outcome. Acta Paediatr 91:145–151
11. Lnyckyj A, Blanchard JF, Rawsthorn PI et al (1999) Perianal Crohn's disease and pregnancy: role of the mode of delivery. Am J Gastroenterol 94:3274–3278

Rheumatological Manifestations of Crohn's Disease

18

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Historical Background

The correlation between inflammatory bowel disease (IBD) and articular involvement has long been well known, but a nosographic classification was proposed only in the early 1970s, when two clinical forms of chronic intestinal inflammation were identified: Crohn's disease (CD) and ulcerative colitis (UC). Both conditions were considered to be responsible for the peculiar forms of arthritis seen in a subset of patients with seronegative spondyloarthritides (SnSp) (Fig. 18.1) [1]. Numerous clinical and experimental findings subsequently drew strong attention to the enormous relevance of the relationship between IBD and either axial or peripheral articular inflammations [2,3].

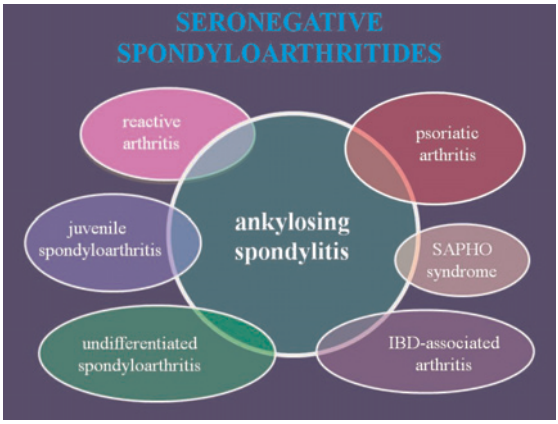

Fig. 18.1 linical conditions belonging to the group of seronegative spondyloarthritides (SnSp). *IBD,* inflammatory bowel disease; *SAPHO*, synovitis, acne, pustulosis, hyperostosis, osteitis

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The first reports on the correlation between ankylosing spondylitis (AS) and IBD originated in the mid-1980's and were of particular interest, as they described a higher incidence of *Klebsiella pneumoniae* in the stool of subjects with active AS [4]. This finding allowed researchers to propose that antigenic fractions of gram-negative bacteria passed through the intestinal wall and could evoke an immune response able to cross-react with cellular structures of the antigen, due to molecular mimicry with histocompatibility antigen HLA-B27. According to this hypothesis, consequent complement activation would explain why the inflammation became chronic; however, why the inflammatory reaction specifically localizes to articular and periarticular structures was unclear.

During the same period of time, ileocoloscopy performed in a population of patients with SnSp showed the presence of lesions with variable degrees of inflammation; in some cases, these lesions could not be distinguished from the ones typically caused by CD. Surprisingly, it was shown that in 75% of patients with a diagnosis of AS intestinal lesions could be detected at the histological level; these lesions, although sometimes minimal, were indicative of an inflammatory state [5].

In the following years, a possible correlation with inflammatory arthropathies was ascertained for other bowel diseases as well, including Whipple disease and bypass arthritis.

Arthropathies Associated with Crohn's Disease: General Considerations

Crohn's disease is associated with many extraintestinal manifestations, many of which are clinically more significant than the intestinal disease itself. The incidence of symptomatic arthropathies associated with CD reportedly ranges from 4 to 29% [2,6,7]. A genetic susceptibility has been recognized; these patients show a familial pattern for rheumatic diseases and for inflammatory conditions involving the bowel. There is no gender difference with respect to frequency. Rheumatological manifestations seem to be significantly more frequent when the disease is limited to the colon [8].

Joint involvement in CD belongs to the field of enteroarthritides, a term used to identify arthropathies caused by or associated with bowel diseases. Enteroarthritides, a subset of SnSp, include the medium and major joint arthritides, mainly of the lower extremities, small joint arthritis of the hand and foot, sacroiliitis (SI), AS, and enthesitis. The pathogenetic factors that explain the association of arthropathies with CD are unknown, but an important role seems to be played by autoimmune mechanisms, including: loss of tolerance to autologous bowel flora in response to a persistent local inflammatory stimulus, autoantigen display, aberrant recognition of self, immunopathogenetic autoantibodies against cellular antigens shared by bowel and joints, and the presence of microbial antigens or toxins able to trigger molecular mimicry [9].

It is clinically remarkable that in many patients the intestinal manifestation is so mild (or even absent) that it cannot be detected. However, when present, signs and

symptoms of intestinal involvement frequently consist of abdominal pain and diarrhea containing mucus, pus, or traces of blood. These may appear years before the articular symptoms but sometimes joint involvement precedes the onset of bowel disease and is thus its first symptom [10]. For this reason, early identification of articular manifestations as symptoms of CD is critical for appropriate treatment choice and to avoid mistakes in disease management.

From the anatomopathological viewpoint, intestinal lesions almost always appear before articular lesions. The latter consist of non-specific synovitis with synovial cell hyperplasia, fibrin deposits, increased vascularization, and the presence of cell infiltrates rich in neutrophils, lymphocytes, plasma cells, and macrophages. In some cases, there is also a granulomatous component with epithelioid giant cells that appear very similar to those seen in the intestinal mucosa.

When the articular and the intestinal components are clinically evident, episodes of acute colitis are often associated with recurrent joint involvement, sometimes in an acute form, although the two components may be clinically independent. The relationship between the severity of the intestinal inflammation and that of the arthropathy is questionable, but there does seem to be a correlation between the extent of bowel disease and the severity of joint involvement. Surgical treatment of the intestinal disease yields no resolution of the peripheral arthritis, in contrast to what happens in UC.

Arthropathies Associated with Crohn's Disease: Diagnostic Aspects

To establish a diagnosis, it is necessary to perform instrumental tests in all cases of arthritis and SpSn not yet reported in the nosography or difficult to classify, with the aim of assessing bowel conditions, especially if the patient's medical history reveals signs and symptoms suggesting intestinal disease. When a classical radiograph of the digestive tract and double-contrast barium enema are not sufficiently informative, the only means to disclose even minimal signs of CD is an endoscopic examination. Laboratory diagnostics are performed to detect biohumoral abnormalities suggestive of an inflammatory state and its relevant severity.

Depending on the case, instrumental diagnostics in rheumatology are based on conventional radiology, bone scintigraphy, CT, MRI, and ultrasonography. A classical categorization of arthropathies associated with CD is axial vs. non-axial (or peripheral) arthritides, depending on the location of the involved joint. Axial arthropathies include SI and AS and are often associated with the presence of antigen HLA B-27. Non-axial (or peripheral) arthropathies are further categorized as type 1 (or peripheral oligoarticular) if no more than four joints are involved, and type 2 (or peripheral polyarticular) if at least five joints are involved. Discriminating factors between the two types also include genetic susceptibility, prevalence, size of the involved joints, as well as the duration of acute episodes and their correlation with bowel disease activity and other extra-articular manifestations (Table 18.1). In contrast to UC, the prevalence of arthropathy shows a less important correlation with the

Table 18.1 Characteristics of arthropathies associated with Crohn's disease

	Oligoarticular peripheral arthropathy (type 1)	Polyarticular peripheral arthropathy (type 2)	Axial arthropathy
Prevalence	~29%	~20%	~12% Up to 50% for asymptomatic sacroiliitis
HLA genetics	B-27	B-27, B-35, DR-B1*0103	B-27
Most affected joints	Large joints	Small joints	Sacroiliac, spine
Course	Transient	Persistent	Trend to progression
Association with bowel disease activity	Yes	No	No
Other associated extra-intestinal manifestations	Erythema nodosum, uveitis	Uveitis	Uveitis
Onset vs. bowel	Generally at the same time	Generally at the same time	Generally before

extent and severity of arthritis and with the frequency of other extraintestinal manifestations.

Oligoarticular Peripheral Arthropathy (Type 1)

Type 1 arthropathy is episodic and recurrent in nature and mainly involves the large joints of the inferior limbs, in particular those subject to load or intense and continuous strain. The most frequently involved joints are the knee and ankle, while the elbow, wrist, and shoulders are less often affected. The onset of arthritis and intestinal disease is usually contemporary.

In the initial stages, there is an acute arthritis, which reaches a peak within 48 h after onset, synchronously with the recurrence of acute enteritis. The involved areas become hot and swollen; active and passive mobilization make spontaneous pain more intense. Analysis of the synovial fluid shows non-specific signs of inflammation, but since CD can cause septic arthritis as a complication, a laboratory culture is recommended. In addition, type 1 peripheral arthropathy is frequently associated

with other extraintestinal manifestations, including erythema nodosum and uveitis. Resolution of the arthropathy occurs after several weeks, without permanent consequences, but in 10% of patients there is a tendency to chronicity.

When the presence of an intestinal disease is uncertain or difficult to diagnose, it is necessary to consider the possibility of a rheumatic disease of inflammatory (rheumatoid arthritis, other subsets of SnSp), degenerative (osteoarthritis), metabolic (gout, chondrocalcinosis), infective (septic arthritis), or other (steroid- and non-steroid-induced osteonecrosis, osteodystrophy) nature.

Polyarticular Peripheral Arthropathy (Type 2)

Type 2 arthropathy mainly affects the small joints of the hands and feet, almost always symmetrically. As is the case in type 1, the onset of arthropathy and intestinal disease are usually contemporary. Also as in type 1, the polyarticular arthritis is typically seronegative, non-erosive, and non-deforming.

In type 2 arthropathy there is a dissociation between objective signs of joint involvement, which might be minimal, and pain symptoms, which can be intense. The disease may last for several months and its course is independent of the intestinal disease activity phase. It may be associated with uveitis but not with other extraintestinal manifestations.

In case of doubt as to the diagnosis, then osteoarthritis, rheumatoid arthritis, other subsets of SnSp, and iatrogenic forms originating from the use of azathioprine and mesalazine or the discontinuation of long-term steroid therapy must be excluded. The differential diagnosis between type 2 arthropathy and rheumatoid arthritis can be particularly difficult when metacarpophalangeal and metatarsophalangeal articulations are affected. In these cases, two useful typical signs of enteroarthritis are the weak tendency to bone fusion and the presence of reactive bone neoformation.

Axial Arthropathies

This group include SI and AS. The onset of either one usually occurs before the onset of enteritis and does not correlate with the course of the disease.

In many CD patients (up to 50%), the asymptomatic form of SI may be detected radiologically (Fig. 18.2) [11]. The less-frequent symptomatic form presents as pain in the sacroiliac joints, possibly spreading to the most inferior area of the lumbar and pelvic regions. Mobilization tends to relieve pain symptoms, which also may be present at night. The application of digital pressure on the sacroiliac joints causes acute pain sensation. In CD patients, it is important to evaluate any involvement of the sacroiliac joints through appropriate diagnostic imaging exams. SI may be monolateral and asymmetric; it is not associated with HLA-B27.

AS and its relevant concomitant peripheral arthropathies tend to evolve inde-

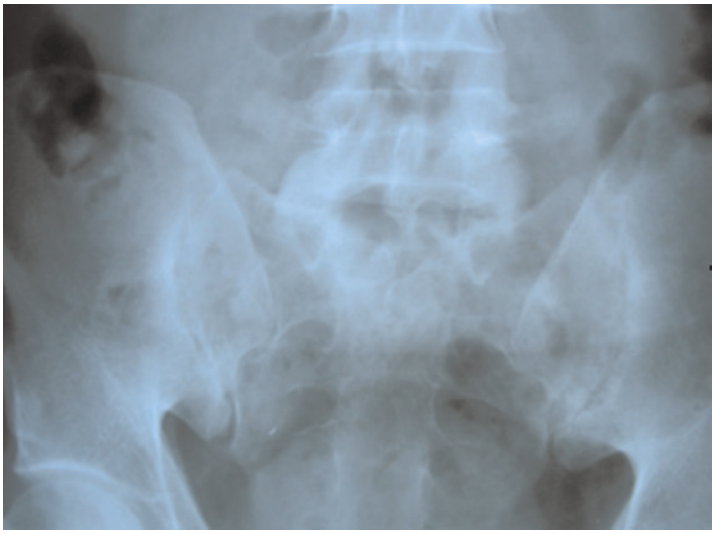

Fig. 18.2 Asymmetric sacroiliitis at the beginning phase in a patient with Crohn's disease

pendently of the course of the intestinal disease and its surgical treatment. The onset tends to occur at young age, presenting as morning stiffness and persistent pain in the lumbar area during both night and day time. There is a tendency to for the pain to spread, involving the pelvis and thighs as well. It may be associated with enthesitis (of the plantar fascia, Achilles tendon, or patellar ligament), uveitis, and peripheral arthritides involving, often aggressively and destructively, the hips, knees, and shoulders. The objective examination shows kyphotic posture of the lumbar spine, absence of common lumbar lordosis, and more or less marked limitation of spinal movements, flexion in particular. In AS the association with the antigen HLA-B27 is strong, possibly as high as in 75% of patients [12]. However this association is weaker than in patients with AS not associated with bowel disease, and it is independent from the concomitant presence of SI. Another differentiating factor is a lower male/female ratio. The onset of AS not associated with HLA-B27 usually occurs after the onset of bowel disease, and under these circumstances the latter seems to have a less favorable course, with frequent intestinal complications.

The reference instrumental diagnostic methodology to detect abnormalities in the sacroiliac region is MRI, which can reveal marrow edema. However, other diagnostic imaging techniques may be helpful as well. Erosions of the surfaces of the sacroiliac joints can be seen by CT, while entheseal erosions due to insertion enthesitis, modifications in the sacroiliac regions, squaring of the vertebral bodies, bone proliferation along the vertebral margins, and marginal syndesmophytes are visualized by conventional radiology. All of these findings are the most typical signs of AS (Fig. 18.3). Bone scintigraphy shows areas of hypercaptation suggestive of an ongoing inflammatory state.

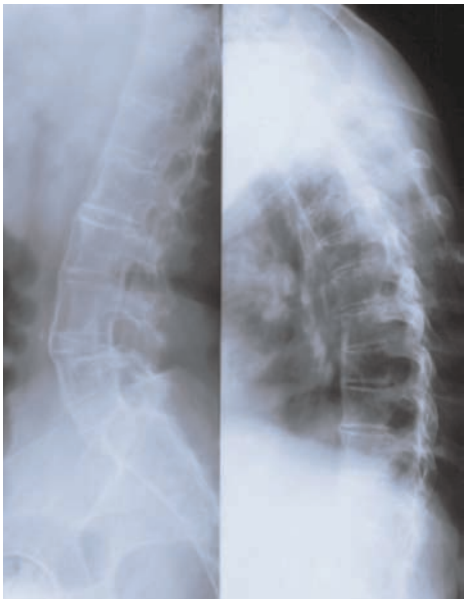

Fig.18. 3 Severe spondylitis of the dorsolumbar tract in a patient with Crohn's disease. Note the ossification of the anterior and posterior longitudinal ligaments, fusion of the lumbar interapophyseal joints, and partial intersomatic synostosis

Arthropathies in Children and Adolescents

The frequency of enteroarthritis in children and adolescents reportedly ranges between 7.5 and 19%. Arthropathies may involve the peripheral and the axial joints [13,14]. Axial forms are associated with HLA-B27. They may be progressive despite treatment of the intestinal disease [15].

Peripheral arthritis has the same frequency in men as in women. It is more often oligoarticular, and the inferior limbs are its preferred site. Its onset typically coincides with the onset of intestinal disease. Most episodes of peripheral arthritis happen during phases of active CD, last less than 2 months, and their course parallels that of the bowel disease.

SI, which can be associated with enthesitis, peripheral arthritis, or neither, is much less frequent than peripheral arthritis. AS is even less frequent and occurs predominantly in male patients.

Treatment of Arthropathies

Treatment also includes measures addressed at the intestinal disease and consists of pharmacological and non-pharmacological therapy. Drug intervention includes non-opioid analgesics, steroidal and non-steroidal anti-inflammatory drugs, classical disease-modifying anti-rheumatic drugs (DMARDs), and biological agents.

Analgesics should be used when pain symptoms are mild. Traditional anti-inflammatory agents and specific COX-2 inhibitors are useful when pain is more severe. The length of treatment should be short in order not to worsen the bowel disease. A recent, and reassuring, randomized controlled study found that the COX-2 specific inhibitor celecoxib is safe in patients with UC in remission if taken for no more than 2 weeks [16]. Intra-articular therapy with steroids, possibly repeated, may be evaluated when joint involvement is single, prevalent, or resistant to systemic treatment.

The most effective DMARD is sulfasalazine. Among the biological agents, the most compelling and well-established results have been obtained with the TNF-α inhibitor infliximab, which was shown to induce remission in bowel disease and in joint symptoms [17,18].

A more or less intensive rehabilitation physical therapy is specifically indicated in AS together with drug treatment. It may also be indicated in the other forms of arthritis, depending on the patient, and mainly serves to alleviate pain. Resting of the joints and the prescription of orthotics may be part of the overall approach.

References

1. Wright V, Moll JMH (eds) (1976) Seronegative polyarthritis. Holland Publishing Company, Amsterdam
2. Dekker-Sayes BJ, Meuwissen SG, Van Den Berg-Loonen EM et al (1978) Ankylosing spondylitis and inflammatory bowel disease. II. Prevalence of peripheral arthritis, sacroiliitis, and ankylosing spondylitis in patients suffering from inflammatory bowel disease. Ann Rheum Dis 37:33–35
3. Rynes RI, Volastro PS, Bartholomew LE (1984) Exacerbation of B27 positive spondyloarthropathy by enteric infections. J Rheumatol 11:96–97
4. Ebringer A, Ghuloom M (1986) Ankylosing spondylitis, HLA-B27, and Klebsiella: cross reactivity and antibody studies. Ann Rheum Dis 45:703–704
5. Mielants H, Veys EM, Cuvelier C et al (1987) Significance of gut inflammation in the seronegative spondyloarthropathies. Clin Exp Rheum 5 Suppl 1:S81-S87
6. Das KM (1999) Relationship of extraintestinal involvement in inflammatory bowel disease. Dig Dis Sci 44:1–13
7. Orchard TR, Wordsworth BP, Jewell DP (1998) Peripheral arthropaties in inflammatory bowel disease: their articular distribution and natural history. Gut:42 387–391
8. Repiso A, Alcántara M, Muñoz–Rosas C et al (2006) Extraintestinal manifestations of Crohn's disease: prevalence and related factors. Rev Esp Enferm Dig 98:510–517
9. Ardizzone S, Sarzi Puttini P, Cassinotti A, Bianchi Porro G (2008) Extraintestinal manifestations of inflammatory bowel disease. Dig Liver Dis 40 Suppl 2:S253-S259
10. Mader R, Segol O, Adawi M et al (2005) Arthritis or vasculitis as presenting symptoms of Crohn's disease. Rheumatol Int 25:401–405
11. Fornaciari G, Salvarani C, Beltrami M et al (2001) Musculoskeletal manifestations in inflammatory bowel disease. Can J Gastroenterol 15:339–403
12. Steer S, Jones H, Hibbert J et al (2003) Low back pain, sacroiliitis, and the relationship with HLA–B27 in Crohn–s disease. J Rheumatol 30:518–522
13. Farmer RG, Michener WM (1979) Prognosis of Crohn's disease with onset in childhood or adolescence. Dig Dis Sci 24:752–757

14. Hamilton JR, Bruce GA, Abdurrahman M, Gall DG (1979) Inflammatory bowel disease in children and adolescent. Adv Pediatr 26:311–41
15. Holden W, Orchard T, Wordsworth P (2003) Enteropathic arthritis. Rheum Dis Clin N Am 29:513–530
16. Sand born WJ, Stemson WF, Brinson J et al (2006) Safety of celecoxib in patients with ulcerative colitis in remission:a randomized, placebo-controlled, pilot study. Clin Gastroenterol Hepatol 4:203–211
17. Van den Bosch F, Kruithof E, De Vos M et al (2000) Crohn's disease associated with spondyloarthropathy:effect of TNF-a blockade with infliximab on the articular symptoms. Lancet 356:1821–1822
18. De Keyser F, Baeten D, Van den Bosch F et al (2003) Infliximab in patients who have spondyloarthropathy:clinical efficacy, safety, and biological immunomodulation. Rheum Dis Clin North Am 29:463–479

Cutaneous Manifestations

19

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Introduction

Cutaneous lesions are well-recognized extraintestinal manifestations of inflammatory bowel disease (IBD), specifically, Crohn's disease (CD) and ulcerative colitis (UC). These extraintestinal manifestations, associated with intestinal symptoms, contribute to the impaired quality of life of patients with CD. During the course of the disease, a great variety of cutaneous lesions may develop, many of which are secondary to granulomatous cutaneous disease, reactive skin eruptions, undernourishment, pharmacological treatment, and other associated conditions [1]. The occurrence of these manifestations varies widely but the mean incidence is around 10% [2]. The cutaneous abnormalities that accompany IBD can be viewed as a spectrum of changes resulting from leukocyte migration to different sites of the skin. Diagnosis of the cutaneous manifestations of CD is made on clinical grounds, based on their characteristic features, and the exclusion of other specific skin disorders; biopsy is rarely appropriate or necessary [3]. Whether the cutaneous manifestations can be considered as real extraintestinal patterns of CD or simply represent associations between different autoimmune diseases is still not clear.

Erythema Nodosum

The frequency of erythema nodosum (EN), the most common cutaneous lesion in CD, ranges from 3% up to 15% [4-6]. EN is also seen in other diseases, such as streptococcal infections, sarcoidosis, and reactions to various drugs. Some subsets

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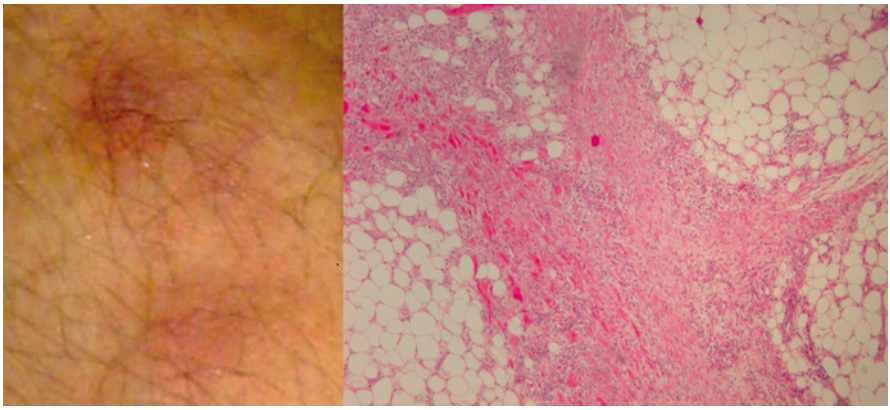

Fig. 19.1 Erythema nodosum: gross appearance and histological aspect. (Courtesy of Roberto Pisa, MD, and Liborio Manente, MD, Department of Pathology, San Camillo-Forlanini Hospitals)

of patients are more susceptible to the development of EN: female sex, isolated colonic involvement, and the presence of arthritis [6,7]. EN tends to occur during the first two years of the disease and may recur in approximately 50% of patients. It appears with characteristic raised, tender, red or violet, warm subcutaneous nodules of 1–5 cm in diameter (Fig. 19.1). Within few days of onset, the lesions usually evolve into red-brown to purple macules. EN commonly affects the extensor surfaces of the extremities, mainly the anterior tibial area, and can be accompanied by fever and leukocytosis. Usually, the lesions are associated with the activity of the intestinal disease, and many patients can predict a flare of CD by the antecedent finding of EN [1]. Biopsy is not usually necessary; when performed, the histology shows a nonspecific focal panniculitis, with an initial neutrophilic infiltrate that progresses to a predominance of both mononuclear and histiocytic giant cells [8]. The process involves the subcutaneous tissues and relatively spares the overlying dermis. EN is the most common form of panniculitis and represents an immunologic response to a number of different antigens. The differential diagnosis of EN includes metastatic CD, in which ulcerating nodules may appear at any site and the histology of which includes non caseating granulomas. Treatment is mainly directed toward the primary disease and systemic corticosteroids are usually required. In resistant and relapsing cases, immunomodulation with azathioprine and/or anti-TNF antibodies may be added, but such measures are rarely needed based on the presence of EN only [3,9].

Pyoderma Gangrenosum

While a serious extraintestinal manifestation of CD, the frequency of PG is, fortunately, < 2% [4,10]. The lesions are often preceded by trauma at the same site, a phenomenon known as “pathergy.” PG is also associated with other diseases, such as systemic

bacterial infections, vasculitis, chronic hepatitis, arthritis, AIDS, tumors, and myeloproliferative disorders [9]. PG may be related to an imbalance of cytokines and to other inflammatory responses mechanisms seen in active CD. The lesions can occur anywhere on the body, including the genitals, but the commonest site is adjacent to stomas, and in 50–60% of cases may be multiple [7]. Initially, they take the form of single or multiple erythematous papules or pustules, but subsequent necrosis of the dermis leads to the development of deep excavating ulcerations with purulent material inside, usually sterile on culture unless secondary infection has occurred. The healed lesions leaves a hyperpigmented area or cribiform scar. The disease can often resemble squamous cell carcinoma and usually requires biopsies for diagnosis (Fig. 19.2). The histological findings are nonspecific, and either neutrophilic or mononuclear cell infiltrates are observed, depending on the biopsy site and the stage of the lesion [11]. PG is a chronic persistent skin disorder that, if extensive, is often resistant to therapy; it tends to occur more often in the setting of active disease, mostly in colonic CD. PG onset can precede the onset of intestinal symptoms but can also develop after the removal of diseased bowel [12]. Initial treatment is aimed at controlling the intestinal inflammation and at preventing cutaneous superinfections. The optimal schedule of medical treatment has yet to be established. High doses of corticosteroids are considered the most effective treatment for PG. Intravenous cyclosporine or tacrolimus are often effective in refractory cases; however, there are no available

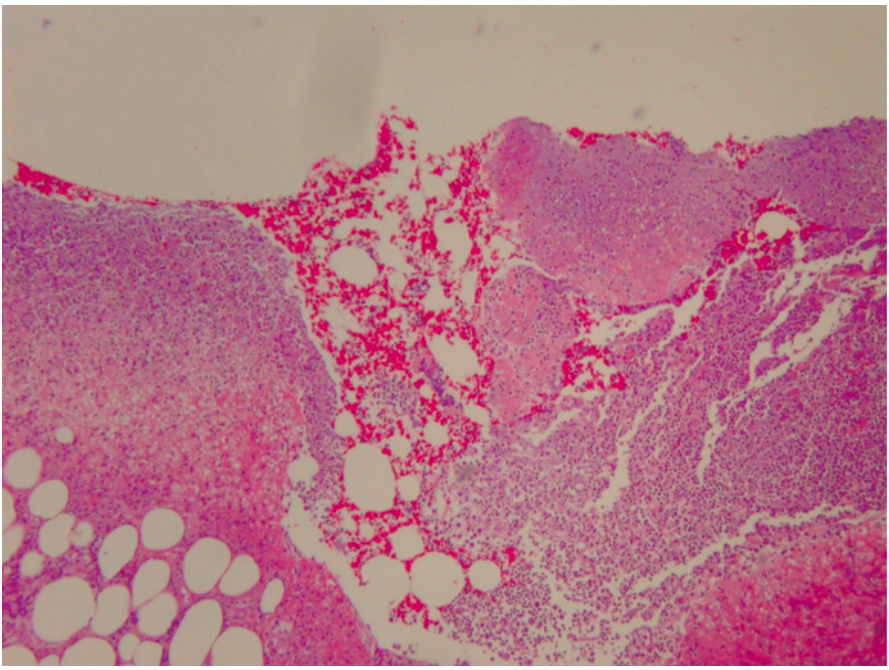

Fig. 19.2 Pyoderma gangrenosum: histological aspect. (Courtesy of Roberto Pisa, MD, and Liborio Manente, MD, Department of Pathology, San Camillo-Forlanini Hospitals)

randomized trials to support their use and these drugs have significant side effects [3]. Infliximab has shown its value in uncontrolled studies and in a small controlled trial vs. placebo [13-15]. The side effect profile and efficacy of infliximab compared with other drugs are such that many clinicians now consider it the preferred treatment for established PG [3]. A recent case report showed the successful use of adalimumab in a patient with fistulizing CD and PG [16]. Local therapy includes topical antibacterial agents and steroids. Surgical debridement of PG only promotes further ulceration due to pathergy phenomena and should thus be avoided [1]. For both PG and EN, recurrent lesions or cluster of lesions may be observed.

Sweet's Syndrome

Also known as acute febrile neutrophilic dermatosis, Sweet's syndrome is associated with a number of diseases. It is characterized by the sudden eruption of multiple erythematous plaques, 1–2 cm in diameter, accompanied by fever, arthralgia, and leukocytosis. This syndrome is rare and is characterized histologically by a dense, diffuse neutrophilic infiltrate and dermal edema [17]. Its relationship with the activity and extent of CD is unknown. The syndrome usually responds rapidly to corticosteroid therapy [18]. In recurrent disease, a number of treatments (colchicines, cyclosporine, dapsone, etc) have been described.

Other Cutaneous Abnormalities

Several case reports of other, rare, cutaneous abnormalities occurring in CD have been published.

Epydermolysis bullosa acquisita is an autoimmune subepidermal blistering occasionally observed in Crohn's colitis and in other diseases, such as rheumatoid arthritis, diabetes mellitus, malignancies, and systemic infections; immunosuppressive therapy is often required [19].

Dermatomyositis have been described in cases of active Crohn's colitis; corticosteroids are usually rapidly effective [1].

Recently, a few cases of *cutaneous periarteritis nodosa* have been described in patients with CD; the syndrome differs from systemic periarteritis by the absence of visceral involvement [20].

Acrodermatitis enteropathica, a rare psoriasiform erythema, also has been described in CD and may be related to malabsorbtion and malnutrition [9].

Psoriasis is seen more frequently in patients with CD and UC than in the general population; the appearance and response to therapy are the same as for the non-IBD population [1]. In patients with psoriasis resistant to common treatments, infliximab can be used, but recently some cases of psoriasis induced by the drug have been reported [21].

Oral mucosal lesions, such as aphthous stomatitis, cobblestone-like lesions, and pyostomatitis vegetans, have been described in CD and should be differentiated from sporadic stomatitis and glossitis both of which can be present in these patients [22].

Metastatic Crohn's Disease

So called *"metastatic" Crohn's disease* (MCD) is a non-neutrophilic cutaneous lesion. This term is applied when a non-caseating granulomatous cutaneous reaction develops in sites anatomically separate from the gastrointestinal tract. These lesions can be found in any location and may take the form of papule, nodule, or plaque, ulcerated or not ulcerated, solitary or multiple, and are usually painful and tender to touch. The activity of MCD seems to be independent from the activity of intestinal CD. The simultaneous presence of perianal CD is more frequent in females and children. Associated intestinal symptoms are present in one-third of adults and one-half of children [23]. Diagnosis is made by skin biopsy and is also aimed at differentiating MCD from other lesions, such as sarcoidosis and hidradenitis suppurativa. The most common histopathologic pattern is a non-suppurative granulomata with a slight cuff of lymphocytes in a nodular or diffuse pattern with a mixed inflammatory infiltrate, often rich in eosinophils [24]. Immunosuppressive therapy and infliximab have been used successfully in selected cases of MCD [25].

References

1. Warner AS, MacDermott P (1996) Extraintestinal manifestations. In: Prantera C, Korelitz B (eds) Crohn's disease. Dekker, New York, pp 467–488
2. Ephgrave K (2007) Extraintestinal manifestations of Crohn's disease. Surg Clin North Am 87:673–680
3. Caprilli R, Gassull MA, Escher JC et al (2006) European evidence based consensus on the diagnosis and management of Crohn's disease: special situations. Gut 55(Suppl 1):i36–38
4. Greenstein AJ, Janowitz HD, Sachar DB (1976) The extraintestinal complications of Crohn's disease and ulcerative colitis. A study of 700 patients. Medicine 55:401–412
5. Bernstein CN, Blanchard JF, Rawsthorne P, Yu N (2001) The prevalence of extraintestinal diseases in inflammatory bowel disease. A population based study. Am J Gastroenterol 96:1116–1122
6. Freeman HJ (2005) Erythema nodosum and pyoderma gangrenosum in 50 patients with Crohn's disease. Can J Gastroenterol 19:603–606
7. Farhi D, Cosnes J, Zizi N et al (2008) Significance of erythema nodosum and pyoderma gangrenosum in inflammatory bowel diseases. Medicine 87:281–293
8. Requena L, Yus E (2008) Erythema nodosum. Dermatol Clin 26:425–438
9. Tavarela Veloso F (2004) Review article: skin complications associated with inflammatory bowel disease. Alimen Pharmacol Ther 220(Suppl 4):50–53
10. Schwaegerle SM, Bergfeld WF, Snitzer D, Tidrick TR (1988) Pyoderma gangrenosum: a review. J Am Acad Dermatol 18:559–568

11. Su WPD, Schroeter AL, Perry HO, Powell PC (1986) Histopathologic and immunopathologic study of pyoderma gangrenosum. J Cutan Pathol 13:223–230
12. Wollina V (2002) Clinical management of pyoderma gangrenosum. Am J Clin Dermatol 3:14–58
13. Ljung T, Staun M, Grove O et al (2002) Pyoderma gangrenosum associated with Crohn's disease: effect of TNF alpha blockade with infliximab. Scand J Gastroenterol 37:1108–1110
14. Regueiro M, Valentine J, Plevy S et al (2003) Infliximab for treatment of pyoderma gangrenosum associated with inflammatory bowel disease. Am J Gastroenterol 98:1824–1826
15. Brooklyn T, Dunnill G S, Shetty A et al (2006) Infliximab for the treatment of pyoderma gangrenosusm: a randomised, double-blind placebo-controlled trial. Gut 55:505–509
16. Zold E, Nagy A, Devenyi K et al (2009) Successful use of adalimumab for treating fistulizing Crohn's disease with pyoderma gangrenosum: two birds with one stone. World J Gastroenterol 15:2293–2295
17. Cohen PR (2007) Sweet's syndrome. A comprehensive review of an acute febrile neutrophilic dermatosis. Orph J Rare Dis 2:34–62
18. Kemmett D, Hunter JA (1990) Sweet's syndrome: a clinicopathologic review of twenty-nine cases. J Am Acad Dermatol 23:503–507
19. Ray TL, Levine JB, Weiss W, Ward PA (1982) Epidermiolysis bollosa acquisita and inflammatory bowel disease J Am Acad Dermatol 6:242–252
20. Komatsuda A, Kinoshita K, Togashi M et al (2008) Cutaneous polyrteritis nodosa in a patient with Crohn's disease. Mod Reumathol 18:639–642
21. Takahashi H, Hashimoto Y, Ishida-Yamamoto A et al (2007) Psoriasiform and pustular eruption induced by infliximab. J Dermatol 34:468–472
22. Plauth M, Jensu H, Meyle G (1991) Oral manifestations of Crohn's disease: an analysis of 79 cases. J Clin Gastroenterol 13:29–37
23. Palamaras I, El-Jabbour J, Pietropaolo N et al (2008) Metastatic Crohn's disease: a review. J Eur Acad Dermatol Venereol 22:1033–1043
24. Emanuel PO, Phelps G (2008) Metastatic Crohn's disease: a histopathologic study of 12 cases. J Cutan Pathol 35:4657–4661
25. Konrad A, Seibold F (2003) Response of cutaneous Crohn's disease to infliximab and methotrexate. Dig Liver Dis 35:351–356

Ocular Manifestations

20

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Introduction

Crohn's disease (CD), or regional enteritis, is a segmentary granulomatous inflammatory bowel pathology of unknown etiology. although autoimmunity is probably involved. In addition, there are extraintestinal manifestations, such as arthritis, cutaneous lesions, and hepatic, renal, and ocular involvement. The incidence of the latter, manifested as uveitis, scleritis, and episcleritis, is about 2–9% of the cases of CD [1–4]. Moreover, 30–40% of patients with scleritis also have a systemic autoimmune disease, such as rheumatoid arthritis, Wegener granulomatosis, and inflammatory bowel disease (IBD) [5], and 2% of patients with uveitis may develop IBD; indeed uveitis may even precede IBD onset. Nearly 40% of patients with IBD-associated uveitis are positive for HLA B27. Arthritis in CD may develop as peripheral arthropathies or as ankylosing spondylitis. Among the latter, nearly 70% are HLA-B27 positive and 50% of the patients in this group develop ocular involvement, mainly uveitis. In addition to these immunological pathologies in patients with CD, an infection of the eye, mycotic endophthalmitis, may develop as a complication in patients receiving parenteral nutrition to treat malabsorption or following bowel surgery.

Ocular Manifestations

Scleritis and Episcleritis

Scleritis or episcleritis may appear in CD as a chronic, painful inflammatory condition and is usually of the non-nodular type. It ranges from a benign self-limited

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Table 20.1 Classification of episcleritis and scleritis

Episcleritis	Scleritis
Simple	Anterior (diffuse, nodular, necrotizing)
Nodular	Posterior

process to a fulminant course resulting in scleral necrosis [6]. The term "episcleritis" refers to inflammation of the episclera, the loose highly vascularized connective tissue that lies between Tenon's capsule and the sclera superficially.

Diagnosis of scleritis or episcleritis is simply clinical. Slit-lamp examination better defines the biomicroscopic aspect of the inflammation, distinguishing between scleritis and episcleritis. The use of red-free diffuse light is very helpful for studying and better-defining the involvement of the conjunctival vessels. The clinical classification of episcleritis and scleritis is described in Table 20.1. Therapy is based on the use of eyedrops containing NSAID, particurarly indomethacin, although some patients are prescribed topical steroids. Steroid eyedrops must gradually be tapered to avoid disease relapse. In severe cases, oral indomethacin may be useful (25–50 mg/die).

The prognosis of scleritis-episcleritis in patients with IBD usually is good and is rarely complicated by uveitis, keratitis, or retinal detachment.

Uveitis

Two patterns of IBD associated uveitis are recognized [7]. The first is bilateral, chronic, and indolent, with a moderate involvement of the posterior ocular structures. The second is acute, unilateral, and recurrent, resembling the HLA-B27-related uveitis. Slit-lamp examination is necessary for the diagnosis (Table 20.2) and shows corneal precipitates, exudation of proteins (Tyndall phenomenon or flare), and the presence of cells in the aqueous and vitreous fluids. These are signs of the breakdown of the blood-aqueous barrier, typical of intraocular inflammation.

In patients with CD, chronic anterior uveitis is more common in those with inflammation of the colon and in those in whom the disease is accompanied by arthritis. Patients with the chronic form of the disease complain of blurred vision without redness or pain. Slit-lamp examination shows no or only a mild conjunctival reaction (white eye), corneal precipitates (larger than those in acute uveitis), flare, and cells in the aqueous and vitreous fluids. Rarely, the retina and optic nerve are involved. A routine ophthalmic evaluation is required in every patient with CD because chronic uveitis is often asymptomatic but if untreated it may become complicated by the development of severe ocular lesions, such as cystoid macular edema, secondary glaucoma, and complicated cataract.

Acute uveitis, by contrast, is characterized by conjunctival injection (red eye) and pain.

Table 20.2 Biomicroscopic signs of anterior uveitis

Conjunctival injection (perikeratic or diffuse)
Corneal precipitates (small medium large)
Aqueous flare/fibrinous clots
Aqueous cells/hypopyon
Intraocular pressure changes (hypotony or hypertony)
Posterior synechiae
Iris nodules (granulomatous uveitis)
Iris atrophy (herpetic uveitis)

Slit-lamp examination shows small diffuse corneal precipitates that cause dusting of the corneal endothelium and a more severe inflammatory reaction in the aqueous, with fibrinous clots and sometimes hypopyon, which results from a dense accumulation of inflammatory cells that causes them to sediment. When the concentration of proteins in the aqueous is very high, adherences between the iris and the anterior capsule of the lens can form (posterior synechiae).

Therapy of uveitis in CD, as in other types of uveitis, is based on topical, oral, or periocular steroids together with mydriatics. For acute fibrinous uveitis atropine 1% eyedrops associated with tropicamide and phenylephrine 10% is necessary. In chronic uveitis, atropine may not be necessary and a mild mydriatic may be sufficient. Among the topical steroids, dexamethasone or betamethasone eyedrops can be used. No specific therapy exists.

Mycotic Endophthalmitis

As noted in the Introduction, mycotic endophthalmitis [8] is a complication of parenteral nutrition in patients treated for malabsorption or following bowel surgery. In such cases, *Candida albicans* is usually the causative agent. Infectious fungal elements, via the bloodstream, reach the eye from fungal colonies on indwelling venous catheter tips. Floaters, localized scotomata, ciliary injection, ocular pain, and photophobia are the typical symptoms of mycotic endophthalmitis. However, sometimes patients are too ill to pay attention to the onset of symptoms and a routine ophthalmic examination is necessary.

Fundus examination discloses the early signs of endophthalmitis, most often, an intense focal inflammation in the inner choroid. The characteristic appearance of a candidal lesion in the fundus is a creamy-white round chorioretinal lesion with over-

lying vitreous inflammation of varying intensity. Such lesions may be present in both eyes. Prompt diagnosis, based on ophthalmic examination, and treatment are mandatory in these patients to avoid blindness. The isolation of *Candida* species from the catheter and from blood confirms the diagnosis but is not necessary to initiate therapy, which is based on oral or intravenous antifungal agents. The preferred drug is fluconazole; amphotericin B may be administered intravenously but we prefer intravitreal injection to potentiate the action of fluconazole when necessary [9]. Vitrectomy may be necessary in eyes with severe vitreous involvement and disease that progresses despite intensive medical therapy.

References

1. Hopkins DJ, Horan E, Burton IL et al (1974) Ocular disorders in a series of 332 patients with Crohn's disease. Br J Ophthalmol 58:732–737
2. Mintz R, Feller FR, Bahr RL, Shah SA (2004) Ocular manifestations in inflammatory bowel disease. Inflamm Bowel Dis 10:135–139
3. Yilmaz S, Aydemir E, Maden A, Unsal B (2007) The prevalence of ocular involvement in patients with inflammatory bowel disease. Int J Colorectal Dis 22:1027–1030
4. Knox DL, Schachat AP, Mustonen E (1984) Primary, secondary and coincidental ocular complications of Crohn's Disease. Ophthalmology 91:163–173
5. Lin P, Bhullar SS, Tessler HH, Goldstein DA (2008) Immunologic markers as potential predictors of systemic autoimmune disease in patients with idiopathic scleritis. Am J Ophthalmol 145:463–471
6. McCluskey PJ Wakefield D (1995) Scleritis and episcleritis. In: Pepose JS, Holland GN, Wilhelmus KR (eds) Ocular infection & immunity. Mosby, St Louis, Missouri, pp 642–662
7. Pivetti Pezzi P (1987) Le flogosi uveali. Masson Italia Editore, Milan
8. Pettit TH, Edwards JE, Purdy EP, Bullock JD (1995) Endogenous fungal endophthalmitis. In: Pepose JS, Holland GN and Wilhelmus KR (eds) Ocular infection & immunity. Mosby, St Louis, Missouri, pp 1262–1285
9. Pivetti Pezzi P, Tamburi S, Bozzoni F et al (1992) Endoftalmite micotica endogena: possibilità e limiti attuali della terapia farmacologica. Boll Ocul 2:255–264

Hepatopathologies

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L. Nauri

Introduction

Crohn's disease (CD) is a chronic inflammatory bowel disease (IBD) of unknown etiology that predominantly affects young adults. It can involve any part of the gastrointestinal tract, most commonly, the terminal ileum, the colon, or both, and its course is characterized by exacerbations and remissions. It may also involve organs outside the gastrointestinal tract. Some of these extraintestinal manifestations are true components of CD while others are complications caused by malnutrition, chronic inflammation, or the side effects of therapy, which make the differential diagnosis accordingly difficult [1,2]. Moreover, the extraintestinal manifestations may or may not correlate with disease activity but nonetheless can have a high impact on the quality of life, morbidity, and mortality of CD patients.

The association between colonic ulceration and liver disease was first made by Thomas in 1873 [3] and was confirmed by Lister in 1899 [4]. In the following years, the close relationship between IBD and hepatobiliary disorders became well-established. The prevalence of liver disease in CD patients varies in different series due to the number of patients included in the study, the severity of the disease, and the methods used to identify liver damage. It may be as high as 50% in patients with active, severe CD requiring surgery (malnutrition, sepsis, fistulae, transfusions with risk of viral infections) while in one study it was documented in 10% of 100 unselected patients with CD. A study from the Mayo Clinic showed that CD patients may have normal liver function tests despite the presence of primary sclerosing cholangitis on cholangiography [5–8].

Gastroenterologists are often faced with the problem of abnormal liver-enzyme levels in patients with CD. The first step in the evaluation of such patients is to

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repeat the test to confirm the results and to determine the degree of the elevation. If confirmed, the patient should be evaluated for [9,10]:

1. liver disease not correlated with CD (chronic hepatitis B and C, alcohol abuse, hemochromatosis, Wilson's disease, α1-antitrypsin deficiency);
2. causes likely related to CD through common pathogenetic mechanisms (primary sclerosing cholangitis, autoimmune hepatitis and overlap syndrome, liver granuloma);
3. complications of the disease (non-alcoholic fatty liver, cholelithiasis, amyloidosis, liver abscesses);
4. drug-related hepatitis or cholestasis (sulfosalazine, glucocorticoids, azathioprine, mercaptopurine, thioguanine, methotrexate, cyclosporine), nodular regenerative hyperplasia of the liver, veno-occlusive hepatic disease, and reactivated latent infections, specifically hepatitis B virus particularly after biological treatment of CD (infliximab).

Primary Sclerosing Cholangitis

Primary sclerosing cholangitis (PSC) is a chronic progressive disorder of unknown etiology that is characterized by inflammation and fibrosis in the biliary tree (intra- and extrahepatic). PSC may progress to secondary biliary cirrhosis and liver failure and is associated with a high risk of cholangiocarcinoma. It probably has the same immune pathogenetic mechanism as CD, although there is a poor association with the stage of bowel disease. PSC also may appear several years after intestinal resection.

Delbert identified the syndrome for the first time in 1924 [11]. In the past, PSC was generally diagnosed in its late stage, when the patient was very ill and markedly jaundiced. More recently, non-surgical diagnostic tests, such as endoscopic retrograde cholangiopancreatography (ERCP) and magnetic resonance imaging (MRI), have become available, such that the syndrome can now be recognized earlier, sometimes even in the asymptomatic stage. PSC can be defined in terms of involvement of the biliary system. The global form involves the bile ducts both inside and outside the liver and is characterized by cholangiographic abnormalities of the intra- and extrahepatic bile ducts. Typical findings are seen on liver biopsy. Other forms of the disease are large-duct PSC and PSC of the small bile ducts, which, as their names imply, involve principally, but not exclusively, the large and small biliary duct systems. Patients with normal ERCP and MRI who have histological findings typical of PSC seem to have a relatively benign course of the disease: 12% of these patients develop classical PSC and there are no reports of cholangiocarcinoma in this population.

Until the introduction of ERCP and MRI into clinical practice, PSC was considered to be rare; however, it is now recognized as the most common chronic cholestatic condition in adults. Although the real frequency of PSC is unknown, the literature reports its occurrence in 1–3.5% of CD patients [7,12]. An association of PSC with CD is generally seen in patients who have extensive colonic or ileocolonic disease or in pediatric populations. The association between PSC and IBD differs among populations;

in northern Europe and in the United States the incidence is 70–90%, whereas it is less common in Italy and Spain (30/40%) and much less common in Japan (20%). Although several characteristic the outcome as well as prognostic factors of PSC patients from Unites States, Japan and European countries appeared to be similar. PSC in Italy mainly follows a benign course while the incidence of Cholangiocarcinoma (CCA) in PSC appeared to be lower in Japan [13–15]. PSC may be present despite normal levels of cholestasis-related enzymes; conversely, these enzymes may be altered in the absence of PSC. The disease may affect people at any age, but it is more often seen in men younger than 45 years and later in women. While smoking seems to protect against PSC, it may increase the risk for cholangiocarcinoma. PSC probably develops through an autoimmune mechanism, as yet unknown. The patient has hypergammaglobulinemia, elevated IgM, autoantibodies (most typical findings include atypical anti-neutrophil cytoplasmic antibodies, pANCA), activated complement, intracellular cytokines, aberrant HLA expression in the biliary epithelium, and high tumor necrosis factor (TNF)-α levels. The portal infiltrates are mostly formed by T cells (CD4 and CD8).

The histological findings may range from normality to frank biliary cirrhosis, with the typical appearances comprising portal inflammation, concentric "onion skin" periductal fibrosis, and periportal fibrosis developing into septal and bridging necrosis. These findings are seen in 30% of PSC patients.

More than half of patients with CD-associated PSC are diagnosed during symptom-free stages and only because of the known association of the two diseases. The presence of liver test abnormalities should prompt an MRI and possibly a liver biopsy. In light of the risks of serious complications following ERCP, such as cholangitis, pancreatitis, perforation, or bleeding, the procedure is normally restricted to therapeutic applications in patients with increased cholestasis. Endoscopic treatments of strictures can improve cholestasis and pruritus and is especially aimed at strictures located in the common bile and main hepatic ducts. Current endoscopic treatments consist of stricture dilatation by bougienage or balloon, brush cytology, forceps biopsy, intraductal ultrasound, and direct visualization of the biliary tree.

Symptom-free patients are not guaranteed a mild condition, as up to 20% of patients with asymptomatic PSC are in the cirrhotic stage at the time of diagnosis [16]. A symptomatic patient usually has anorexia, nausea, pruritus, weight loss, nonspecific abdominal pain, jaundice, and irregular fever. Cholangitis is unusually found at diagnosis and instead is known to follow a surgical procedure or ERCP.

Physical examination is normal in around 50% of patients or it may reveal the characteristics of cholestatic liver disease (hepatomegaly, splenomegaly, jaundice, etc.). The diagnosis of PSC is mainly based on demonstrating the presence of segmentary stricture of the bile ducts separated by areas with a normal or dilated lumen. Usually, both the intrahepatic and extrahepatic bile ducts are involved, but sometimes only one or the other is affected [17,18]. Even if the gold standard in lesion recognition is ERCP, we prefer MRI as it is non-invasive, provides high-quality images, may allow recognition of liver growths (cholangiocarcinoma), and has excellent sensitivity and specificity with no complications.

The natural history of PSC is variable and the disease can therefore evade diag-

nosis. If symptomatic, it corresponds to a fluctuating progressive illness that evolves over 12–21 years. Normally, small-duct PSC is more benign, with less frequent evolution to cholangitis and cholangiocarcinoma. Four stages have been identified: preclinical, asymptomatic with laboratory cholestasis-related changes, symptomatic (jaundice, choluria, pruritus, asthenia, cholangitis, hepatosplenomegaly, and histologically evident liver cirrhosis), and terminal (ascites, portal hypertension, encephalopathy, bleeding from esophageal varices). Cholangiocarcinoma may contribute to the rapid development of terminal-stage disease and is the most severe complication of PSC. The progress of cholangiocarcinoma is not related to the severity of the liver disease and has to be suspected in all patients with IBD in order to spare the patient the need for liver transplantation. The combination of ultrasonography, computed tomography (CT), ERCP, bile cytology, and MRI may allow an early diagnosis of PSC and thereby improve patient survival.

Patients with CD may develop every kind of liver disease that can be detected by laboratory or clinical changes. The gold standard for the diagnosis of all these diseases is liver biopsy.

Nash-Nafld

In 1980, Ludwig et al. [19] coined the term “NASH” to designate a liver condition that mimics alcohol-related hepatitis but which occurs in people who are not alcohol abusers. This condition is diagnosed based on laboratory changes suggesting liver disease, ultrasound and histological findings indicative of fatty change in the liver, hydropic degeneration, inflammation, and the possible presence of Mallory’s hyaline or centrolobular-pericellular fibrosis. These lesions are usually asymptomatic and may progress to fibrosis and cirrhosis. They are more common in patients with IBD and are probably associated with malnutrition and with the increased import of free fatty acids into the liver as a result of abdominal lipolysis, which may develop during weight loss. The pathogenesis of the disease may be explained by passage of bacteria or their products from the damaged intestinal lumen, jejunal diverticulosis, jejunoileal bypass, bowel bacterial overgrowth, and increased mucosal permeability.

Amyloidosis

Amyloid type AA deposition is a rare complication of CD. Secondary amyloidosis is usually associated with infections and chronic inflammatory conditions but rarely with CD (0.99%). CD-associated amyloidosis is three times more common in males. This complication has to be looked for in patients with intestinal and extraintestinal suppurative foci, which lead to increased synthesis of serum amyloid P (SAP) glycoprotein as a result of defective catabolism (worsened by proinflammatory cytokines, particularly TNF-α).

Autoimmune hepatitis, liver granulomas, and liver lesions related to drugs used in the treatment of CD can be found in these patients. Both anamnestic findings of alterations of liver enzymes post-treatment and liver biopsy may permit the differential diagnosis.

Liver Abscess

Liver abscess in patients with CD is challenging and the diagnosis is often delayed because the symptoms and signs are similar to manifestations of exacerbations of CD [20]. As a result, many patients are treated with increasing doses of corticosteroids or even undergo surgery for resection. Consequently, several authors now advise that liver abscesses should be actively searched for, either by ultrasound or CT scan, in patients with CD resistant to conventional treatment.

Gallstones

Patients with CD of the small bowel have an increased incidence of gallstones (13–34% vs. 5% of the general population and of patients with CD confined to the colon) [21]. The increased rate of gallstone formation in patients with inflammation or absence of the terminal ileum is most likely due to the reduction in bile-salt absorption. The relative increase in biliary cholesterol and its precipitation in the gallbladder lead to the increased formation of gallstones [22].

Conclusions

Crohn's disease is a systemic disorder that may involve every site of the body. In addition, the possibility of contributing disease complications as side effects of treatment always should be considered.

From the original descriptions over a century ago of liver disease associated with CD (and IBD), much progress has been made in understanding the relationship between the two conditions, but in addition to answers there are more questions. The association of CD and PSC, autoimmune hepatitis, overlap syndrome, and a wide range of diseases that can be well detected with liver biopsy has been established. However, the cause, consequence, and coincidence remain to be elucidated. From the hepatopathological point of view, PSC is the most intriguing extraintestinal manifestation of CD. Due to the high risk of malignancy, surveillance strategies are essential, but they have yet to be designed and evaluated. Since hepatocellular carcinoma is a disease without any curative options apart from liver transplantation, it has to be regarded as the greatest challenge in hepatology today.

The development of multiple new drugs, and especially biological therapies, that intervene in the biological pathways involved in CD, implies that their effectiveness

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in the treatment of CD patients has to be studied over the long term. This is essential to verify the possible reduction in hepatic damage (especially PSC and cholangicarcinoma) in patients with CD. In another section of this book, the activation of hepatic infections (reactivation of latent infection of hepatitis B virus) and other side effects of these drugs is discussed [23].

References

1. Solis Herruzo JA, Solis–Munoz P (2007) Hepatobiliary manifestations of inflammatory bowel disease. Rev Esp Enferm Dig 99:525–542
2. Weismuller TJ, Wedemeyer J, Kubicka S et al (2008) The challenges in primarysclerosing cholangitis – Aetiopathogenesis, autoimmunity, management and malignancy. J Hepatol 48:38–57
3. Thomas CH (1873) Ulceration of the colon with a much enlarged fatty liver. Trans Pathol Soc Phil 4:87–88
4. Lister JD (1899) A specimen of diffuse ulcerative colitis with secondary diffuse hepatitis. Trans Pathol Soc Lon 50:130–134
5. Perrett AD, Higgins G, Johnston HH et al (1971) The liver in Crohn's disease. Q J Med 40:187–209
6. Atkinson AJ, Carrol WW (1964) Sclerosing cholangitis association with regional enteritis. JAMA 188:183–184
7. Rasmussen HH, Fallingborg JF, Mortensen PB et al (1997) Hepatobiliary dysfunction and PSC in patients with Crohn's disease. Scan J Gastroenterol 32:604–610
8. Okada H, Mizuno M, Yamamoto K, Tsuji T (1966) Primary sclerosing cholangitis in Japanese patients: association with IBD. Acta Med Okayama 50:227–235
9. Chapman RW, Angus PW (1999) The effect of gastrointestinal disease on the liver and biliary tract. In: Oxford textbook of clinical hepatology. Oxford medical publications. Oxford University Press, Oxford New York Tokyo, pp 1686–1691
10. Pratt DS, Kaplan MM (2000) Evaluation of abnormal liver-enzime results in asymptomatic patients. N Engl J Med 342:1266–1271
11. Delbert (1924) Bull Mem Soc Chir Paris 50:1144–1146
12. Angulo P, Maor–Kendler Y, Lindor KD (2002) Small–duct PSC: a long-term follow-up study. Hepatology 35:1494–1500
13. Escorell A, Pares A, Rodes J et al (1994) Epidemiology of primary sclerosing cholangitis in Spain. J Hepatol 21:789–791
14. Okolicsanyi L, Fabris L, Viaggi S et al (1996) Primary sclerosing cholangitis: clinical presentation, Natural history of prognostic variables: an italian multicenter study. Eur J Gastroenterol Hepatol 8:685–691
15. Tanaka A, Takamori Y, Toda G et al (2008) Outcome and prognostic factors of 391 Japanese patients with primary sclerosing cholangitis. Liver Int 28(7):983–998
16. Broome U, Olsson R, Loof L et al (1966) Natural history and prognostic factors in 305 Swedish patients with PSC. Gut 38:610–615
17. Chapman RW, Arborgh BA, Rhodes JM et al (1980) PSC: review of its clinical features, cholangiography, and hepatic histology. Gut 21:870–877
18. MacCarty RL, LaRusso NF, Wiesner RH, Ludwig J (1983) PSC: findings on cholangiography and pancreatography, Radiology 149:39–44
19. Ludwig J, Viggiano RT, McGill DB (1980) Nonalcoholic steatohepatitis: Mayo Clinic experiences with a hitherto unnamed. Mayo Clinic Proc 55:342–348

20. Margalit M, Elinav H, Ilan Y, Shalit M (2004) Liver abscess in IBD. J Gastr Hepatol 19:1338–1342
21 Fraquelli M, Losco A, Visentin S et al (2001) Gallstone disease and related risk factors in patients with CD. Arch Intern Med 161:2201–2204
22. Dowling RH , Bell DG, White J (1972) Lithogenic bile in patients with ileal dysfunction. Gut 13:415–420
23. Millonig G, Kern M, Ludwiczek O et al (2006) subfulminant hepatitis B after infliximab in CD; need for HBV screening? World J Gastroenterol 12:974–976

Infectious Complications

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N. Petrosillo, C.M.J. Drapeau

Introduction

Infections are strongly associated with Crohn's disease (CD), both in their etiopathogenesis and in their clinical course [1]. The hypothesis that infective agents are linked to the onset of CD is supported by several clinical and experimental observations, although a direct causality remains to be demonstrated. In particular, bacteria such as *Mycobacterium avium* subsp. *paratuberculosis* (MAP), adherent invasive *Escherichia coli* (AIEC), and *Listeria* spp. have been cited as possible causal agents of CD [1–3]. This conclusion is based on the association of MAP DNA and CD, reported in two meta-analyses [4,5]. Moreover, experimental infections of animals have demonstrated that MAP is capable of causing inflammatory bowel disease [2]. Other investigations have reported the presence of AIEC in CD biopsies and a mechanism of attachment to the diseased bowel [2,3].

A series of independent observations stressed the association between infections and CD onset, including Crohn's-like pathology in patients with chronic granulomatous disease and chronic variable immunodeficiency; impaired phagocytic function at biopsy sites of Crohn's patients; clinical improvement with antibiotic treatment; and the positive results of genome association studies of CD and genes that govern innate immune recognition of intracellular bacteria [2]. However, a nested case-control study conducted by Bernstein et al. [6], in which polymerase chain reaction (PCR) was carried out with mucosal samples from patients with CD and controls, found no association between CD and infection with MAP.

Three theories are currently under consideration regarding the "infectious" etiopathogenesis of CD: (1) a reaction to a persistent intestinal infection; (2) a defec-

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tive mucosal barrier to luminal microorganisms, and (3) a deregulated immune response of the host to luminal agents [7]. Although the role of infections in the onset of CD is still hypothetical, it is nonetheless clear that infection can play an important role in the clinical course of CD. Particularly, infections can be responsible for the relapse of the disease; they can also represent a complication of the disease itself or of its pharmacologic and surgical treatment. They may also indicate the ineffective immunization of patients with CD who are thus at risk of developing what are otherwise vaccine-preventable illnesses [1]. Infectious complications of CD are caused by multiple infectious agents, including bacteria, viruses, and fungi. The aim of this chapter is to describe the epidemiological and clinical features of the infectious complications of CD.

Infections and the Relapse of Crohn's Disease

Infections can lead to relapse episodes of CD by activating the gastrointestinal immune system. Multiple organisms have been associated with such episodes, including bacteria, viruses, and parasites.

Bacterial Infections

The most frequently reported bacterial infections associated with CD are *Clostridium difficile*, enteropathogenic *E. coli*, *Salmonella* spp., *Shigella* spp., and *Campylobacter* spp. [1]. Among them, *Cl. difficile* is considered the most important pathogen, with a prevalence of 10.9/1000 hospitalized patients with CD, compared to 4.8/1000 patients without inflammatory bowel disease (IBD) [8]. The higher prevalence of *Cl. difficile* infections among patients with CD is attributable to the use of immunomodulator drugs, the colonic involvement itself, or both conditions. In the study conducted by Nguyen et al., only CD patients with small bowel involvment had infection rates that approached those of general medical hospitalized patients without IBD [8].

The clinical importance of infections in the relapse of CD requires prompt identification of potential pathogens in stool in order to prevent unnecessary treatment with immunosuppressors. Clinicians should perform microbiological examination of fresh specimens of stool (generally three samples) by microscopy and culture, and, in patients with clinical relapse of CD, also test for *Cl. difficile* toxins. The isolation of *Cl. difficile* in a CD patient without clinical symptoms like diarrhea or active colitis is of uncertain clinical importance since it likely represents a colonization, for which antibiotic treatment is not indicated [9].

Viral Infections

The viral agents responsible for CD relapse belong to the Herpesvirus family: Epstein-Barr virus (EBV), cytomegalovirus (CMV), and herpesvirus (HSV) [1]. The available information on CD relapse of viral origin is mainly based on studies of CMV infection. It is well known that CMV infection is a generally benign, self-limited disease among immunocompetent adults; however, in immunocompromised patients, CMV infection is associated with serious complications, including colitis. CMV infection can be primary or secondary to the reactivation of latent infection and can develop in CD patients following immunosuppressive therapy, thus leading to the worsening of CD colitis, steroid-refractory disease, and toxic megacolon [1]. Nevertheless, as is the case in bacterial infections, the detection of viral agents in the bowel mucosa and/or in other biological fluids does not necessary imply active viral infection. Moreover, the results of diagnostic tests for CMV infection should be interpreted based on the clinical picture in order to distinguish CMV infection from CMV-related disease.

Currently, the multiple diagnostic tests available for CMV diagnosis include: (1) cultures of CMV from blood, urine, saliva, and other biological fluids; (2) histological examination of tissue samples (colonic mucosa); (3) serological tests (IgM and IgG antibodies); (4) CMV antigenemia; and (5) PCR to detect CMV DNA in blood and in stool [1]. The most sensitive diagnostic tests for CMV infection with digestive tract involvement in CD patients are histological examination of mucosal tissue and PCR to detect CMV in colonic biopsies [1].

The indications for CMV therapy in patients with relapse of CD and CMV infection are still controversial. Clinical evidence suggests antiviral treatment with ganciclovir only in those patients with significant CMV DNA in the mucosa [1].

Parasitic Infections

Amoebiasis has been cited as a frequent cause of CD relapse in endemic areas, with patients receiving corticosteroid therapy more predisposed to fatal complications of infections [10].

Infectious Complications of CD

Patients with CD have an increased incidence of infectious complications and of infection-related death. Possible explanations for the increased susceptibility to infections are defective innate immunity and malnutrition [1]. Actually, CD itself predisposes the patient to sepsis, which is frequently related to abscess and fistulae formation. The pathogenesis of sepsis in these cases is likely related to the transmur-

al inflammatory process, which favors bacterial translocation and spread from the bowel lumen to the extraluminal space [1].

Abscess formation develops in nearly 20% of patients with CD and can involve multiple organs and tissue spaces [11]. Moreover, CD carries a significant burden in terms of extraintestinal manifestations, particularly hepatobiliary pathologies such as pericholangitis, sclerosing cholangitis, granulomatous hepatitis, and cholelithiasis. It is well known that most of those conditions can predispose to infectious complications, such as cholangitis, cholecystitis, and, less frequently, to liver abscess. Pyogenic liver abscess is considered a relatively rare complication of CD, with an estimated incidence of 114–297/100,000 patients with CD. It is more common in younger patients (mean age 36.5 years) and usually occurs secondary to infections of the biliary tract (cholecystitis, cholangitis) or to infections of organs that are drained by the portal vein, especially in the setting of appendicitis, diverticulitis, and loss of integrity of the bowel wall [12]. Other risk factors for liver abscess include long-term steroid treatment, metronidazole treatment, inflammatory and perforating diseases, fistulae and intra-abdominal abscesses, malnutrition, immunological abnormalities, and surgical interventions [12,13].

The diagnosis of liver abscess is often difficult due to the lack of specificity of the symptoms, which can mimic CD reactivation (high fever, chills, sweating, right upper abdominal pain, nausea, vomiting, weight loss), and the delay in diagnosis is generally associated with poor prognosis. Instrumental investigations for liver abscess include abdominal ultrasonography (USG) and computed tomography (CT), The final diagnosis can be performed by USG or CT-guided percutaneous aspiration and drainage of the abscess. In liver abscess, hemocultures are positive only in 50% cases, whereas culture of purulent drainage fluid is positive in nearly 90% of cases, the most frequent bacterial causal agents being *Streptococcus* spp. or aerobic and anaerobic gram-negative bacilli. Antibiotic therapy should be based on the results of microbiological cultures, when available, and should be administered for at least 10–14 days [12,13].

Apart from the hepatobiliary infectious complications of CD, other extraintestinal manifestations have been described in the literature and deserve to be mentioned due to their unusual clinical presentation. In particular, many vertebral and paravertebral infections have been described in association with enteric fistula [14,15]. Kavia et al. [16] reported a case of isolated discitis in a patient with apparent lack of fistulization from the gut to the infected disc. Other authors have reported cases of presacral abscess and vertebral osteomyelitis, and of spondylodiscitis. Of note, spondylodiscitis is also a rare extraintestinal rheumatological manifestation of CD, which demands differential diagnosis with bacterial spondylodiscitis as an infectious complication of CD [17]. Maggiore et al. [14] reported a case of meningitis and epidural abscess associated with CD-related fistulizing distal ileum, due to communication to the extradural space of the lumbar spine from a psoas abscess. Aguas et al. [13] reported a case consisting of both septic thrombophlebitis of the superior mesenteric vein and pylephlebitis in a CD patient who at onset had multiple liver abscesses. The diagnosis of pylephlebitis should be considered in any patient with evidence of intra-abdominal infection and high-grade bacteremia. Instrumental

diagnostic exams include color Doppler and CT scan. Broad-spectrum antibiotic therapy should be administered for al least 6 weeks.

Among the viral infectious complications of CD, CMV colitis has been more frequently reported [18], although it is likely related to steroid therapy.

Therapy-Related Infectious Complications of CD

Drug-Therapy-Related Infections

Drugs currently used in the treatment of CD include immunosuppressive agents, such as thiopurine and methotrexate, anti-tumor-necrosis-factor (TNF) agents (infliximab), anti-IL-12p40 antibody, anti-α_4 integrins (natalizumab), and steroids. Each of these can be used alone or in combination with the others, and each has its own mechanism of action and targets within the immune system. The latter property accounts for differences in the type and severity of immunosuppression and thus in the related infections that can arise as treatment complications. Infections related to immunosupressive therapy are mainly of the opportunistic type and include tuberculosis (TB). Moreover, CD patients are frequently administered antibiotics, which is a well-known risk factor for colonization and/or infection with other bacteria, especially those that are antibiotic-resistant, and with fungi. Unfortunately, the exact incidence of infections related to immunosuppression in CD patients is still unknown.

Among the previously cited immunosuppressive agents, Infliximab, which is an immunomodulatory agent that is increasingly used for the treatment of refractory fistulizing CD, appears to be associated with the greatest risk of infection, most likely because of its long half-life and its induction of monocyte apoptosis [19].

Despite its high efficacy, infliximab is associated with the occurrence of severe infections, such as TB, endemic mycoses, and intracellular bacterial infections. Baldin et al. [20] found that in France the incidence of TB among patients treated with infliximab is higher (8–24/10,000) than in the general population (1.1/10,000) [20]. Moreover, the authors observed that TB occurred more frequently within the first 12 weeks of infliximab treatment, and around the time of the third infusion.

Together with the diagnostic and therapeutic management of CD patients with active TB, we must also consider the diagnosis and treatment of patients with latent TB, with disease onset possibly before the initiation of infliximab therapy. Indeed, the presence of quiescent mycobacteria, which defines latent TB, represents a considerable risk factor for TB reactivation once infliximab treatment is started. Consequently, the screening of all the patients for latent TB before infliximab therapy is mandatory. Baldin et al. [20] formulated "*National guidelines on the prevention and management of TB during or after infliximab treatment*", and López-San Román et al. [21] provided recommendations to which clinicians can readily refer. Of note, according to Denis et al. [22], infliximab therapy can be restarted in TB patients if adequate anti-TB treatment has been completed.

Interestingly, regarding the use of screening tests for latent TB, some authors [23]

recommend in-vitro specific blood tests rather than the tuberculin skin test for the diagnosis of latent tuberculosis before anti-TNF therapy, due to the former's higher sensitivity and specificity, at least in patients with immune-mediated inflammatory diseases.

Infections related to infliximab therapy also result in other granulomatous and non-granulomatous bacterial infections, many of which have been described in the literature. The former include *Mycobacterium xenopi* [24], cutaneous nocardiosis [25], listerosis [26,27], coccidioidomycosis, and histoplasmosis [28], and the latter staphylococcal liver abscess [29], staphylococcal sepsis [30], and pulmonary actinomycosis [31].

Among viral infections, subfulminant hepatitis B [32], varicella [33,34], adenovirus pneumonia [35], and disseminated CMV infection [36] have been reported. Of note, infiximab also has been associated with EBV-related lymphoma [37]. Fungal infections include *Pneumocistis jiroveci* (carinii) pneumonia [38,39], systemic candidiasis [40], aspergillosis [41,42], both disseminated and localized (fungal nasal septal abscess due to *Aspergillus flavus*), and histoplasmosis [42].

According to the literature, the use of azathioprine is associated with varicella pneumonia, whereas anti-IL-12p40 antibody has been related to a high risk of infections caused by *Salmonella* spp. and *Mycobacterium* spp. [43].

Treatment of CD patients with the biological agent natalizumab treatment is reportedly linked to the occurrence of progressive multifocal leukoencephalopathy [45]. Steroid usage has been generally associated with an increased incidence of particular types of infections, such as viral infections, TB, and fungal infections. Interestingly, Nausheen et al. [46] described a case of Q fever in a CD patient under chronic steroid therapy. Additionally, clinicians should be aware of the possible reactivation of latent viral infections in CD patients under immunosuppressive therapy (e.g., CMV, EBV, HSV) or increased viral replication with organ damage (e.g., hepatitis B and C viruses).

Surgical-Therapy-Related Infections

Indications for surgical treatment in patients with CD include infectious and non-infectious conditions (abscess formation, primary bacterial peritonitis, and secondary bacterial peritonitis). However, surgery represents an additional risk factor for infectious complications. Indeed, patients with CD are at higher risk for post-operative complications such as surgical site infections (SSI). In a prospective surveillance study conducted by Uchino et al. [47] among IBD patients, the incidence of incisional SSI was 26.3% in those with CD, which is considerably higher than that reported in the general population for abdominal surgery [48,49]. Moreover, in the same study [47], the authors found that IBD surgery was an independent risk factor for incisional SSI (OR = 2.59; 95% CI = 1.34–5.03).

According to CDC guidelines [50], SSI are classified as incisional/superficial, incisional/deep, and organ/space infections, but sepsis also can be considered as a post-operative complication if it is demonstrated to occur following surgical intervention. Organ/space SSI includes post-operative peritonitis, also defined as second-

ary peritonitis, which often requires additional surgical treatment. Secondary peritonitis is generally considered a nosocomial infection; therefore, antibiotic treatment should also cover antibiotic-resistant bacteria.

A possible explanation for the higher risk of SSI in CD patients is the burden of microbial colonization with nosocomial pathogens. This occurs during the frequent hospitalizations and at follow-up medical visits. Indeed, microbial colonization is a well-established risk factor for subsequent bacterial infections, such as SSI [51]. Moreover, CD patients have an impaired immune response, related to defective innate immunity and to malnutrition, both of which potentially increase the risk of SSI. Nonetheless, Kunitake et al. [52] did not find an association between infliximab therapy and an increased rate of cumulative postoperative complications, including SSI.

Infectious Complications of CD Related to Ineffective Immunization

Patients with CD under immunosuppressive therapy are likely to develop ineffective immune responses following vaccination. Consequently, these patients are at higher risk of infections related to vaccine-preventable diseases [53] (e.g. hepatitis B, varicella). Moreover, Melmed et al. [53] found that immunizations are still uncommonly administered by clinicians to patients with IBD, despite the presence of these known and significant risk factors. Sands et al. [54] formulated guidelines for immunizations in patients with IBD that could be a useful tool for clinicians.

Conclusions

This chapter has provided an overview of the infectious complications of CD, with their respective pathogenetic mechanisms. It is clear that CD patients are at higher risk for developing infectious complications of all types; therefore, clinicians should be aware of the need and possibility of preventing potentially fatal infectious complications such as tuberculosis. The literature provides extensive information on the prevention and treatment of infections in patients with IBD. The available recommendations and guidelines are a useful tool for clinicians in the diagnosis and clinical management of the infectious complications of patients with CD.

References

1. Irving PM, Gibson PR (2008) Infections and IBD. Nat Clin Pract Gastroenterol Hepatol 5(1): 18–27
2. Lowe AM, Yansouni CP, Behr MA (2008) Causality and gastrointestinal infections: Koch, Hill, and Crohn's. Lancet Infect Dis 8(11):720–726
3. Barnich N, Darfeuille-Michaud A (2007) Role of bacteria in the etiopathogenesis of inflammatory bowel disease. World J Gastroenterol 13(42): 5571–5576

4. Feller M, Huwiler K, Stephan R et al (2007) Mycobacterium avium subspecies paratuberculosis and Crohn's disease: a systematic review and meta–analysis. Lancet Infect Dis 7(9):607–613
5. Abubakar I, Myhill D, Aliyu SH, Hunter PR (2008) Detection of Mycobacterium avium subspecies paratuberculosis from patients with Crohn's disease using nucleic acid-based techniques: a systematic review and meta-analysis. Inflamm Bowel Dis 14(3):401–410
6. Bernstein CN, Nayar G, Hamel A, Blanchard JF (2003) Study of animal–borne infections in the mucosas of patients with inflammatory bowel disease and population–based controls. J Clin Microbiol 41(11):4986–4990
7. De Hertogh G, Geboes K (2004) Crohn's disease and infections: a complex relationship. MedGenMed 10(3):14
8. Nguyen GC, Kaplan GG, Harris ML, Brant SR (2008) A national survey of the prevalence and impact of Clostridium difficile infection among hospitalized inflammatory bowel disease patients. Am J Gastroenterol 103(6):1443–1450
9. Sunenshine RH, McDonald LC (2006) Clostridium difficile-associated disease: new challenges from an established pathogen. Cleve Clin J Med 73(2):187–197
10. Abbas MA, Mulligan DC, Ramzan NN et al (2000) Colonic perforation in unsuspected amebic colitis. Dig Dis Sci 45(9):1836–1841
11. Farmer RG, Hawk WA, Turnbull RB (1975) Clinical patterns in Crohn's disease: a statistical study of 615 cases. Gastroenterology 68:627–635
12. Karaca C, Pinarbaşi B, Danalioğlu A et al (2004) Liver abscess as a rare complication of Crohn's disease: a case report. Turk J Gastroenterol 15(1):45–48
13. Aguas M, Bastida G, Nos P et al (2007) Septic thrombophlebitis of the superior mesenteric vein and multiple liver abscesses in a patient with Crohn's disease at onset. BMC Gastroenterol 7:22
14. Maggiore R, Miller F, Stryker S, Buchman AL (2004) Meningitis and epidural abscess associated with fistulizing Crohn's disease. Dig Dis Sci 49(9):1461–1465
15. Zapata E, Cosme A, Ojeda E et al (2006) Psoas abscess complicating Crohn's disease: review of 5 cases. Rev Esp Enferm Dig 98(5):393–395
16. Kavia S, Gilbert JM (2004) Crohn's disease and discitis. J R Soc Med 97(10):484–485
17. Zöld E, Barta Z, Zeher M (2007) Spondylodiscitis representing as the very first sign of Crohn's disease. Inflamm Bowel Dis 13(8):1058–1059
18. Olsen S, Gilbert J (2004) Cytomegalovirus infection in Crohn's colitis. J R Soc Med 97(7):335–336
19. Crum NF, Lederman ER, Wallace MR (2005) Infections associated with tumor necrosis factor-alpha antagonists. Medicine (Baltimore) 84(5):291–302
20. Baldin B, Dozol A, Spreux A, Chichmanian RM (2005) Tuberculosis and infliximab treatment. National surveillance from January 1, 2000, through June 30, 2003. Presse Med 12(5):353–357
21. López–San Román A, Obrador A, Fortún J et al; Grupo Espanol de Trabajo en Enfermedad de Crohn y Colitis Ulcerosa (GETECCU) (2006) Recommendations on tuberculosis and treatment of inflammatory bowel disease with infliximab, 2006 update. Gastroenterol Hepatol 29(2):81–84
22. Denis B, Lefort A, Flipo RM et al (2008) Long-term follow-up of patients with tuberculosis as a complication of tumour necrosis factor (TNF)–alpha antagonist therapy: safe re-initiation of TNF-alpha blockers after appropriate anti–tuberculous treatment. Clin Microbiol Infect 14(2):183–186
23. Sellam J, Hamdi H, Roy C et al; RATIO (Research Axed on Tolerance of Biotherapies) Study Group (2007) Comparison of in vitro-specific blood tests with tuberculin skin test for diagnosis of latent tuberculosis before anti-TNF therapy. Ann Rheum Dis 66(12):1610–1615
24. Majoor CJ, Schreurs AJ, Weers-Pothoff G (2004) Mycobacterium xenopi infection in an immunosuppressed patient with Crohn's disease. Thorax 59(7):631–632

25. Singh SM, Rau NV, Cohen LB, Harris H (2004) Cutaneous nocardiosis complicating management of Crohn's disease with infliximab and prednisone. CMAJ 171(9):1063–1064
26. Ramanampamonjy RM, Laharie D, Bonnefoy B et al (2006) Infliximab therapy in Crohn's disease complicated by Listeria monocytogenes meningoencephalitis. Gastroenterol Clin Biol 30(1):157–158
27. Molina J, Núñez O, Beceiro I et al (2003) Rhomboencephalitis due to Listeria monocytogenes as a complication of Crohn's disease. Gastroenterol Hepatol 26(7):457–458
28. Wallis RS, Broder MS, Wong JY et al (2004) Granulomatous infectious diseases associated with tumor necrosis factor antagonists. Clin Infect Dis 38(9):1261–1265
29. Patel TR, Patel KN, Boyarsky AH (2006) Staphylococcal liver abscess and acute cholecystitis in a patient with Crohn's disease receiving infliximab. J Gastrointest Surg 10(1):105–110
30. Herrlinger KR, Borutta A, Meinhardt G et al (2004) Fatal staphylococcal sepsis in Crohn's disease after infliximab. Inflamm Bowel Dis 10(5):655–656
31. Cohen RD, Bowie WR, Enns R et al (2007) Pulmonary actinomycosis complicating infliximab therapy for Crohn's disease. Thorax 62(11):1013–1014
32. Millonig G, Kern M, Ludwiczek O et al (2006) Subfulminant hepatitis B after infliximab in Crohn's disease: need for HBV-screening? World J Gastroenterol 12(6):974–976
33. Lemyze M, Tavernier JY, Chevalon B, Lamblin C (2003) Severe varicella zoster pneumonia during the course of treatment with azathioprine for Crohn's disease. Rev Mal Respir 20:773–776
34. Tougeron D, Mauillon J, Tranvouez JL (2006) Severe varicella infection during treatment with infliximab for Crohn's disease. Gastroenterol Clin Biol 30(12):1410–1413
35. Ahmad NM, Ahmad KM, Younus F (2007) Severe adenovirus pneumonia (AVP) following infliximab infusion for the treatment of Crohn's disease. J Infect 54(1):e29–32
36. Helbling D, Breitbach TH, Krause M (2002) Disseminated cytomegalovirus infection in Crohn's disease following anti-tumour necrosis factor therapy. Eur J Gastroenterol Hepatol 14(12):1393–1395
37. Bai M, Katsanos KH, Economou M et al (2006) Rectal Epstein–Barr virus-positive Hodgkin's lymphoma in a patient with Crohn's disease: case report and review of the literature. Scand J Gastroenterol 41(7):866–869
38. Velayos FS, Sandborn WJ (2004) Pneumocystis carinii pneumonia during maintenance anti-tumor necrosis factor-alpha therapy with infliximab for Crohn's disease. Inflamm Bowel Dis 10(5):657–660
39. Kaur N, Mahl TC (2007) Pneumocystis jiroveci (carinii) pneumonia after infliximab therapy: a review of 84 cases. Dig Dis Sci 52(6):1481–1484
40. Belda A, Hinojosa J, Serra B et al (2004) Systemic candidiasis and infliximab therapy. Gastroenterol Hepatol 27(6):365–367
41. Alderson JW, Van Dinter TG Jr, Opatowsky MJ, Burton EC (2005) Disseminated aspergillosis following infliximab therapy in an immunosuppressed patient with Crohn's disease and chronic hepatitis C: a case study and review of the literature. MedGenMed 21)7(3):7
42. Walker R, Gardner L, Sindwani R (2007) Fungal nasal septal abscess in the immunocompromised patient. Otolaryngol Head Neck Surg 136(3):506–507
43. Jain VV, Evans T, Peterson MW (2006) Reactivation histoplasmosis after treatment with anti–tumor necrosis factor alpha in a patient from a nonendemic area. Respir Med 100(7):1291–1293
44. Fieschi C, Allez M, Casanova JL (2005) High risk of infectious disease caused by salmonellae and mycobacteria infections in patients with Crohn disease treated with anti-interleukin-12 antibody. Clin Infect Dis 40(9):1381
45. Berger JR (2007) Progressive multifocal leukoencephalopathy. Handb Clin Neurol 85:169–183
46. Nausheen S, Cunha BA (2007) Q fever community-acquired pneumonia in a patient with Crohn's disease on immunosuppressive therapy. Heart Lung 36(4):300–303

47. Uchino M, Ikeuchi H, Tsuchida T et al (2009) Surgical site infection following surgery for inflammatory bowel disease in patients with clean-contaminated wounds. World J Surg 33(5):1042–1048
48. Watanabe A, Kohnoe S, Shimabukuro R et al (2008) Risk factors associated with surgical site infection in upper and lower gastrointestinal surgery. Surg Today 38(5):404–412
49. Wilson J, Ramboer I, Suetens C: HELICS–SSI working group (2007) Hospitals in Europe Link for Infection Control through Surveillance (HELICS). Inter-country comparison of rates of surgical site infection: opportunities and limitations. J Hosp Infect 65 Suppl 2:165–170
50. Mangram AJ, Horan TC, Pearson ML et al (1999) Guideline for prevention of surgical site infection, 1999. Centers for Disease Control and Prevention (CDC) Hospital Infection Control Practices Advisory Committee. Am J Infect Control. 27(2):97–132
51. Muñoz P, Hortal J, Giannella M et al (2008) Nasal carriage of S. aureus increases the risk of surgical site infection after major heart surgery. J Hosp Infect 68(1):25–31
52. Kunitake H, Hodin R, Shellito PC et al (2008) Perioperative treatment with infliximab in patients with Crohn's disease and ulcerative colitis is not associated with an increased rate of postoperative complications. J Gastrointest Surg 12(10):1730–1736
53. Melmed GY, Ippoliti AF, Papadakis KA et al (2006) Patients with inflammatory bowel disease are at risk for vaccine–preventable illnesses. Am J Gastroenterol 101(8):1834–1840
54. Sands BE, Cuffari C, Katz J et al (2004) Guidelines for immunizations in patients with inflammatory bowel disease. Inflamm Bowel Dis 10(5): 677–692

Kidney Disease

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M. Rosa, S. Di Giulio

Introduction

Crohn's disease (CD) potentially affects many organs, including, albeit rarely, the kidney. The most frequent renal manifestations are calcium oxalate stones and their complications; secondary amyloidosis; acute tubular necrosis related to volume depletion caused by diarrheal fluid losses or inadequate fluid intake, or both. Tubulo-interstitial nephritis is another common finding and, among patients with CD, the most common cause is drug-related nephritis. However, case reports in the literature suggest that granulomatous interstitial nephritis is also an extraintestinal manifestation of CD. Glomerulonephritis has been reported in CD patients as well.

Early recognition of renal complications, despite their relatively infrequent occurrence, is nonetheless very important to prevent the development of irreversible renal damage. There is abundant evidence showing that renal function must be monitored, also with respect to potential treatment-related nephrotoxicity.

Urinary Stones

The prevalence of nephrolithiasis among patients with CD is higher than in the general population. Nephrolithiasis can lead to obstructive uropathy or progressive nephrocalcinosis [1]. One study showed that 7–15% of patients with CD of the terminal ileum have episodes of renal stones, with an even higher frequency (28%) in those who have had >100 cm of ileum resected [2].

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Most of the renal stones in CD patients are composed of calcium oxalate (CaOx), but there is also a marked increase in the tendency to form uric acid stones. This is especially the case in patients who have undergone resection of the colon and ileostomy, as the loss of bicarbonate in the ileostomy effluent leads to formation of acidic urine, which coupled with the low urine volume, decreases the solubility of uric acid, causing crystallization and stone formation. Thus, the prevention of stone formation requires treatment with alkalizing agents to raise the urine pH to about 6.5. In addition, urine volume should be increased, which increases the solubility of uric acid and prevents crystallization [2,3].

Patients with small bowel resection may develop steatorrhea; if the colon is present, these patients are thus at risk of hyperoxaluria [3]. The increase in oxalate absorption and its subsequent excretion are due to the binding of free calcium to fatty acids in the intestinal lumen, which allows more free oxalate to be absorbed; moreover, colonic permeability to small molecules such as oxalate is increased because of exposure of the colon to non-absorbed bile salts. Normally, calcium binds oxalate in the intestinal lumen to form an insoluble complex, thereby leaving less oxalate available for absorption. However, with steatorrhea, intraluminal calcium preferentially binds fatty acids, leaving more soluble oxalate available for absorption [3].

The absence of intestinal oxalate-degrading bacteria, *Oxalobacter formigens* (OF), has been identified as a possible cause of hyperoxaluria in patients with inflammatory bowel diseases [4]. Recurrent stone-formers are significantly less likely to be colonized with OF than controls, but this appears to be due to antibiotic use. Studies in animal and in humans have shown that gut colonization with OF can decrease urinary oxalate levels. However, it remains to be determined whether gut colonization with oxalate-metabolizing bacteria can reduce stone formation [5].

Factors other than urinary supersaturation with CaOx have been implicated in stone development; for example, gastrointestinal surgery results in lowered urinary levels of anti-lithogenic substances, such us magnesium and citrate, which normally act as inhibitors of CaOx crystallization [6]. The formation of magnesium citrate in the renal tubule is thought to reduce the reabsorption of citrate, thereby increasing urinary citrate excretion. In magnesium-deficient states, a greater proportion of citrate is available for reabsorption, resulting in lowered urinary citrate levels. The tendency to magnesium deficiency and hypomagnesuria in CD patients thus contributes to their hypocitraturia [2].

CD patients share a tendency to chronic volume contraction due to loss of water and salt in diarrheal stool, which leads to decreased urine volumes. In turn, the chronically low urine volume adds to the risk of renal damage from stone crystal formation. Bowel disease patients with a tendency to stone formation also frequently have reduced creatinine clearance compared to stone formers without bowel disease [1]. Chronic extracellular fluid volume depletion that is hemodynamically mediated chronically reduces the glomerular filtration rate (GFR) [1].

To facilitate the detection, evaluation, and management of chronic kidney disease, guidelines of the National Kidney Foundation recommend assessment of the GFR by the estimated-GFR (e-GFR), which is based on serum creatinine concentration and demographic and clinical variables, including age, sex, ethnicity, and body size using estimating equations such as those derived from the Modification of Diet in Renal Disease (MDRD) Study [7].

Current guidelines define chronic kidney disease as kidney damage or a GFR < 60 ml/min per 1.73 m^2 for 3 months or more, regardless of cause. Kidney damage is usually demonstrated by markers such as albuminuria [8]. However, clinicians should be aware of the limitations of the MDRD Study equation and other GFR-estimating equations in apparently healthy individuals with low GFR estimates and in patients with low creatinine generation, including those with muscle wasting or reduced meat intake. If greater accuracy is needed in such cases, then clearance can be measured using exogenous filtration markers (inulin or iothalamate) or creatinine clearance.

Hyperoxaluria therapy involves a low-fat, low-oxalate diet, attempts to increase urine volume, and the administration of agents such as calcium to bind oxalate in the gut lumen. The correction of hypocitraturia and hypomagnesuria are also helpful [9]. Magnesium and citrate administration must result in correcting urinary, rather than serum, levels to normal. This may be achieved by magnesium replacement alone or by additional citrate replacement therapy [2,10]. The administration of lactic acid bacteria at high concentrations can influence urinary oxalate excretion, reducing urinary supersaturation levels and thus potentially preventing urinary stone formation [11].

Hyperoxaluria appears to be an important predisposing factor for the deposition of calcium oxalate crystals in the renal interstitium [12]. Nephrocalcinosis occurs when calcium precipitates in conjunction with either oxalate or phosphate. There have been several reports of this severe complication in patients with malabsorption and steatorrhea following jejunoileal bypass surgery, in those undergoing extensive resection of the small intestine in association with short bowel syndrome; however, the exact incidence of nephrocalcinosis is not known [12]. CT scanning is the most sensitive imaging technique to detect nephrocalcinosis. Even in mild cases, plain X-ray may reveal nephrocalcinosis as small deposits of calcium salts in the renal calyces. Nephrocalcinosis also may be diagnosed incidentally or may present as a typical tubulointerstitial nephropathy, with low-grade proteinuria (< 1 g/day), a bland abnormality of urine sediment, or an unexplained rise in serum creatinine. Patients may have a history of nephrolithiasis. Crystal extrusion from the obstructed tubules has been proposed as the cause of interstitial nephritis. Crystals evoke an inflammatory response leading to fibrosis, a loss of nephrons, and, eventually, to chronic renal failure [13].

Interstitial fibrosis is a common histologic finding and appears most striking near sites of crystal deposition in renal biopsy specimens. Chronic inflammatory cells often surround areas of scarring and widespread tubular destruction may be seen.

If unrecognized, oxalate interstitial nephritis inevitably leads to severe and progressive renal failure. This cause of renal failure may be underestimated and should be systematically searched for in all patients with malabsorption [14].

Tubulo-interstitial Nephritis

Acute and chronic interstitial nephritis may occur in CD. Acute interstitial nephritis (AIN) can be associated with drug therapy; while enteric hyperoxaluria is a possible cause of chronic interstitial nephritis, but rarely described [12]. Most patients with or

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without chronic interstitial nephritis have recurrent episodes of acute renal failure associated with volume depletion, prerenal azotemia, and, occasionally, AIN or acute tubular necrosis [12]. Many drugs cause AIN, including several antimicrobial agents, nonsteroidal anti-inflammatory drugs, anticonvulsants, diuretics, and proton pump inhibitors [15,16]. AIN is difficult to diagnose on clinical basis, as the symptoms are nonspecific, including oliguria, malaise, anorexia, nausea, and vomiting. Acute renal failure is usually present and eosinophilia, eosinophiluria, proteinuria and/or hematuria can be often detected [15].

Drug-related tubulo-interstitial nephritis should be confirmed by histology. Indications for renal biopsy include advanced renal failure and a lack of spontaneous recovery following cessation of drug therapy after 15 days. Light microscopy generally shows prominent inflammation with an interstitial infiltrate of lymphocytes, plasma cells, and eosinophils. The mechanism of drug-induced AIN is unknown, but an immunological etiology is suspected. For this reason, drugs that modulate the immune response, especially corticosteroids, have been used in the treatment of AIN [17].

The 5-aminosalicylates (5-ASA) are among the many groups of drugs frequently used in the treatment of patients with CD. Epidemiological studies have shown that patients taking 5-ASA preparations are at an increased risk of renal disorders, and there have been several case reports of nephrotoxicity in patients with inflammatory bowel disease receiving 5-ASA therapy. The potential nephrotoxicity of mesalazine and sulfasalazine seems to be similar [18,19].

The incidence of nephrotoxicity in patients receiving 5-ASA therapy is reported to be less than 0.5% [19]; however, this adverse drug reaction may be under recognized and underreported [19]. Morbidity is high and most of the case reports describe the development of a severe, chronic, and progressive interstitial nephritis [18,20].

Clinical evidence suggests that nephrotoxicity most often occurs within the first 12 months but in some patients disease presentation can be delayed and first occurs after several years. Accordingly, renal function should be regularly monitored for the duration of therapy [21,22].

Clinical manifestations of 5-ASA-related nephrotoxicity include enzymuria (such as β-*N*-acetyl-D-glucosaminidase) and microalbuminuria, both of which can be detected in early stages of renal damage. Tubular proteinuria (such as α1-microglobulin) is considered an extraintestinal manifestation of IDB irrespective of 5-ASA treatment. Moreover, other clinical presentations are: interstitial nephritis, glomerulonephritis (possibly secondary), hypersensitivity reactions involving the liver and kidney, and acute renal failure. Unlike classic drug-induced interstitial nephritis, the symptoms and signs of 5-ASA-related nephrotoxicity are few and nonspecific. It most frequently has an insidious onset and takes the form of an indolent, severe, chronic, and progressive interstitial nephritis. Consequently, detection of this condition may be delayed for many months [20,21].

The exact mechanism of the induction of interstitial nephritis is unknown. However, type 1 hypersensitivity reactions are unlikely since fever, arthralgia, eosinophilia, and skin rashes are uncommon [21]. A delayed, cell-mediated response, resembling that described for other nonsteroidal anti-inflammatory drugs is the most likely mechanism [22].

5-ASA treatment should be withdrawn when renal impairment appears in a patient with CD after controlling for the causes described above. If this does not lead to an improvement in GFR, then renal biopsy should be considered, as it is the only way to distinguish between interstitial nephritis and glomerulonephritis associated with CD.

Steroids and azathioprine have been used in patients with renal impairment due to mesalazine-associated interstitial nephritis, but evidence for a beneficial role is anecdotal and uncontrolled. Partial improvement or even complete recovery of renal function after steroid administration has been reported by several authors [22]. However, other studies have been unable to demonstrate a beneficial effect of these immunosuppressive drugs [22]. Nevertheless, a trial of high-dose steroids (60 mg/day or 1 mg day/kg for up to 3 months) may be recommended in patients whose renal function does not improve after drug withdrawal alone [20].

Proton pump inhibitors (PPIs) are a widely prescribed class of drugs. Although well tolerated, there have been numerous case reports and a recent case series implicating PPIs in acute idiosyncratic interstitial nephritis. As noted above, the most common symptoms are non-specific and include oliguria, malaise, anorexia, nausea, and vomiting. Some symptoms of AIN might initially be confused with the illness the drug was originally prescribed for, leading to a delay in diagnosis. With the ever increasing use of PPIs, this complication is likely to become more prevalent. Promoting awareness of AIN will facilitate earlier diagnosis and management of this potentially harmful condition, which, if detected early, is quickly reversible [23].

Granulomatous chronic interstitial nephritis has been described in patients with CD [22]. Renal biopsy revealed severe chronic tubulo-interstitial nephritis with the formation of non-caseous epithelioid granulomas containing multinucleated giant cells. It was suggested that granulomatous interstitial nephritis is related to the granulomatous interstitial nephritis of CD but not to the use of 5-ASA preparations [24]. Furthermore, 5-ASA preparations such as mesalazine may cause tubulo-interstitial nephritis without granuloma. Granulomatous chronic interstitial nephritis may be a renal extraintestinal manifestation of CD [25].

Secondary Amyloidosis

Amyloid A amyloidosis, also referred to as secondary amyloidosis, is a rare but potentially fatal complication of CD, occurring in 0.5–6% of these patients. However, despite its low prevalence renal failure due to renal amyloidosis is one of the most common causes of death in CD patients [26,27]. Secondary amyloidosis is associated with increased hepatocyte production of the acute-phase reactant serum amyloid A (SAA); this process may be stimulated by the release of cytokines (e.g., interleukin-1) from activated macrophages. Cleavage of SAA by circulating monocytes/macrophages results in the generation of smaller, proteolytic fragments of SAA, called AA proteins, that can then be deposited and stored extracellularly in tissues as insoluble fibrils.

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Inflammatory bowel disease, primarily CD, with suppurative infections, rheumatoid arthritis, familial Mediterranean fever; and chronic infections are the diseases that most frequently evolve to AA amyloidosis. The underlying disease usually is longstanding, and active inflammation typically is present when amyloidosis becomes evident [26]. While the gastrointestinal tract, liver, autonomic nervous system, and, less frequently, the heart, are sites of AA amyloid deposition, the kidneys are most frequently affected in AA amyloidosis. Thus, renal disease is a frequent early manifestation of the systemic amyloidoses and often is the major source of morbidity for individuals with these disorders [28]. With respect to CD, nephropathy is the most serious and potentially fatal manifestation of systemic amyloidosis [29]. Ongoing deposition of amyloid in the kidney results in proteinuria and progressive loss of renal function. While the underlying mechanism of organ dysfunction in the amyloidoses seems to be disruption of tissue architecture by amyloid deposits, several observations suggest that amyloidogenic precursor proteins, folding intermediates, and protofilaments have toxicities that directly contribute to the disease manifestations.

The diagnosis of amyloidosis requires histologic demonstration of amyloid deposits. This usually is accomplished by staining of tissue samples with Congo red dye. Amyloid stained with Congo red has an orange-red appearance under light microscopy and produces apple-green birefringence under polarized light. Amyloid can be found anywhere in the kidney of affected CD patients, but glomerular deposition typically predominates. By light microscopy, glomerular amyloid appears as amorphous material in the mesangium and capillary loops of the glomeruli. Amyloid deposition in the tubulointerstitium produces tubular atrophy and interstitial fibrosis. In a small proportion of patients, glomerular deposition is scant or absent and the amyloid is confined to the tubulointerstitium or the vasculature. Irrespective of the distribution of amyloid, Congo red staining produces the disease-defining birefringence under polarized light [28].

The clinical manifestations of renal disease vary with the site of involvement. Proteinuria and nephrotic syndrome, as well as renal insufficiency, are common. Proteinuria ranges from subnephrotic to massive, with urinary protein excretion rates as high as 20–30 g/24 h. The urinary protein is composed mostly of albumin. Proteins loss > 3 g/24 h usually is accompanied by other markers of the nephrotic syndrome. Hypoalbuminemia can be profound, and edema often is severe and refractory to diuretics. The multisystem nature of systemic amyloidosis can contribute to the difficulty of managing fluid retention. When amyloid is confined to the tubulointerstitium or vasculature, proteinuria is minimal and a reduced GFR is the principal clinical manifestation [28].

Renal impairment tends to progress less rapidly when tubulointerstitial rather than glomerular deposition predominates. Vascular involvement often is accompanied by hypertension, an otherwise uncommon feature of amyloidosis.

The diagnosis of crescentic glomerulonephritis should be suspected in a patient with renal amyloidosis who develops acute renal failure in association with an active urine sediment. The optimal therapy for this condition is not known. A regimen consisting of pulse methylprednisolone and immunosuppressive agents, similar to that used in idiopathic crescentic glomerulonephritis, has been beneficial in isolated

cases. Although less common, heavy tubular deposition, potentially leading to signs of tubular dysfunction, such as type 1 (distal) renal tubular acidosis, or polyuria due to nephrogenic diabetes insipidus have been reported. Amyloidosis can be considered an infiltrative diseases and often cause enlargement of the kidneys. However, in most patients, the kidneys are of normal size, as seen on imaging studies, and the absence of enlarged kidneys should not exclude a diagnosis of amyloidosis. Clear relationships between the extent of amyloid deposition, as determined by kidney biopsy, and the severity of clinical manifestations have not been demonstrated. Urinary protein excretion or GFR decline cannot be predicted on the basis of biopsy findings [28].

The current strategy for the treatment of AA amyloidosis is firmly based on knowledge of the underlying pathogenetic mechanisms and aimed at reducing the amyloid precursor (SAA) load by intensive anti-inflammatory/immunosuppressive therapy or anticytokine (TNF-α, IL-1β) or interleukin-6 blockade therapy; when applicable, any existing infectious focus (surgery, antimicrobial drugs) should be eliminated [30].

Colchicine has also been tried in some forms of secondary amyloidosis, based upon the ability of this drug to decrease experimental amyloid formation. It is most successful in patients with familial Mediterranean fever, particularly if given early in the disease course and before the development of renal dysfunction. A good response to colchicine in amyloidosis secondary to inflammatory bowel diseases has been reported [31].

Emerging strategies focus on dissolution of the amyloid deposits using small molecules that either interact with the glycosaminoglycans or the fibril component of the deposits, or deplete the amyloid P component. Eprodisate binds to the glycosaminoglycan-binding site on amyloid fibrils, thus targeting amyloid fibril polymerization and tissue deposition [32]. Successful treatment can lead to stabilization of renal function, a reduction in protein excretion, and partial resolution of amyloid deposits (as assessed by scintigraphy). These benefits are primarily seen in patients in whom the serum SAA concentration is kept within the normal range [33].

There have been reported cases of patients with systemic amyloidosis associated with CD who showed improvement of renal function and proteinuria and a decrease of serum amyloid A protein levels after treatment with the anti-TNF antibody infliximab [29].

Patients who progress to end-stage renal disease are treated with either dialysis or renal transplantation. The latter may offer the best prospect for patients with CD who developed amyloidosis and end-stage renal failure [34].

Glomerulonephritis

Glomerulonephritis is a rarely reported extraintestinal manifestation of CD that occurs in the setting of active bowel inflammation. Circulating immune complexes are found in nearly half the patients, while serum complement usually is normal. Renal failure with hematuria and/or proteinuria, even nephrotic range proteinuria,

may be present at the time of diagnosis of glomerulonephritis. The disease most often improves in parallel with treatment of the gastrointestinal disorder [35].

The histologic findings are varied and include membranoproliferative glomerulonephritis, mesangioproliferative glomerulonephritis, membranous nephropathy, IgA nephropathy, and IgM nephropathy. Glomerulonephritis caused by antiglomerular basement membrane antibody has been described in association with CD and also with a case of thin basement membrane disease [36].

References

1. Parks JH, Worcester EM, O'Connor RC et al (2003) Urine stone risk factors in nephrolithiasis patients with and without bowel disease. Kidney Int 63:255–265
2. McConnell N, Campbell S, Gillanders I et al (2002) Risk factors for developing renal stones in inflammatory bowel disease. BJU Int 89:835-841
3. Worcester EM (2002) Stones from bowel disease. Endocrinol Metab Clin North Am 31:979-999
4. Kumar R, Ghoshal UC, Singh G et al (2004) Infrequency of colonization with Oxalobacter formigenes in inflammatory bowel disease: possible role in renal stone formation. J Gastroenterol Hepatol 19:1403-1409
5. Siva S, Barrack ER, Reddy GP et al (2009) A critical analysis of the role of gut Oxalobacter formigenes in oxalate stone disease. BJU Int 103:18–21
6. Viana ML, Pontes RM, Garcia WE et al (2007) Crohn's disease and kidney stones: much more than coincidence? Arch Gastroenterol 44:210-214
7. National Kidney Disease Education Program. Information of Health Professionals 2006 Creatinine Standardization Program. Accessed at http://www.nkdep.nih.gov/professionals/gfr_calculators/index.htm
8. National Kidney Foundation (2002) K/DOQI clinical practice guidelines for chronic kidney disease: evaluation, classification, and stratification. Kidney Disease Outcome Quality Initiative. Am J Kidney Dis 39:S1-266
9. Cirillo M, Iudici M, Marcarelli F et al (2008) Nephrolithiasis in patients with intestinal diseases. G Ital Nefrol 25:42-48
10. Pak CY (2008) pharmacotherapy of kidney stones. Expert Opin Pharmacother 9:1509-1518
11. Lieske JC, Goldfarb DS, De Simone C et al (2005) Use of a probiotic to decrease enteric hyperoxaluria. Kidney Int 68:1244-1249
12. Braden GL, O'Shea MH, Mulhern JG (2005) Tubulointerstitial diseases. Am J Kidney Dis 46:560-572
13. Khan SR (2004) Crystal-induced inflammation of the kidneys: results from human studies, animal models, and tissue-culture studies. Clin Exp Nephrol 8:75-88
14. Tintillier M, Pochet JM, Blackburn D et al (2001) Hyperoxaluria: an underestimated cause of rapidly progressive renal failure. Acta Clin Belg 56:360-363
15. Rossert J (2001) Drug-induced acute interstitial nephritis. Kidney Int 60:804-817
16. Härmark L, van der Wiel HE, de Groot MC et al (2007) Proton pump inhibitor-induced acute interstitial nephritis. Br J Clin Pharmacol 64:819-823
17. González E, Gutiérrez E, Galeano C et al (2008) Early steroid treatment improves the recovery of renal function in patients with drug-induced acute interstitial nephritis. Kidney Int 73:940-946
18. Van Staa TP, Travis S, Leufkens HG et al (2004) 5-aminosalicylic acids and the risk of renal disease: a large British epidemiologic study. Gastroenterology 126:1733-1739

19. Gisbert JP, González-Lama Y, Maté J (2007) 5-Aminosalicylate and renal function in inflammatory bowel disease: a systematic review. Inflamm Bowel Dis 13:629-638
20. Corrigan G, Stevens PE (2000) Review article: interstitial nephritis associated with the use of mesalazine in inflammatory bowel disease. Aliment Pharmacol Ther 14:1-6
21. Tadic M Grgurevic I, Scukanec-Spoljar M et al (2005) Acute interstitial nephritis due to mesalazine. Nephrology 10:103-105
22. Arend LJ, Springate JE (2004) Interstitial nephritis from mesalazine: case report and literature review. Pediatr Nephrol 19:550-553
23. Sierra F, Suarez M, Rey M et al (2007). Systematic review: proton pump inhibitor-associated acute interstitial nephritis. Aliment Pharmacol Ther 26:545-553
24. Joss N, Morris S, Young B et al (2007) Granulomatous interstitial nephritis. Clin J Am Soc Nephrol 2: 222-230
25. Archimandritis AJ, Weetch MS (1993) Kidney granuloma in Crohn's disease. BMJ 307:540-541
26. Béji S, Kaaroud H, Ben Moussa F et al (2004) Renal amyloidosis complicating the outcome of chronic inflammatory colitis. Presse Med 33:862
27. Lovat LB, Madhoo S, Pepys MB et al (1997) Long term survival in systemic amyloid A amyloidosis complicating Crohn's disease. Gastroenterology 112:1362–1365
28. Dember LM (2006) Amyloidosis-associated kidney disease. J Am Soc Nephrol 17:3458–7341
29. Park YK, Han DS, Eun CS (2008) Systemic amyloidosis with Crohn's disease treated with infliximab. Inflamm Bowel Dis 14:431-432
30. Petterson T Konttinen YT, Maury CP (2008) Treatment strategies for amyloid A amyloidosis. Expert Opin Pharmacother 9:2117-2128
31. Garrido Serrano A, Guerrero Igea FJ, Hierro Guilmain C et al (2001) Good response to colchicine in amyloidosis secondary to inflammatory bowel diseases. Gastroenterol Hepatol 24:196-198
32. Dember LM (2009) Modern treatment of amyloidosis: unresolved questions. J Am Soc Nephrol 20:469-472
33. Gillmore JD, Lovat LB, Persey MR et al (2001) Amyloid load and clinical outcome in AA amyloidosis in relation to circulating concentration of serum amyloid A protein. Lancet 358:24-29
34. Sherif AM, Refaie AF, Sobh MA et al (2003) Long-term outcome of live donor kidney transplantation for renal amyloidosis. Am J Kidney Dis 42:370-375
35. Takemura T, Okada M, Yagi K et al (2002) An adolescent with IgA nephropathy and Crohn disease: pathogenetic implications. Pediatr Nephrol 17:863-866
36. Shaer AJ, Stewart LR, Cheek DE et al (2003) IgA antiglomerular basement membrane nephritis associated with Crohn's disease: a case report and review of glomerulonephritis in inflammatory bowel disease. Am J Kidney Dis 41:1097-1109

Crohn's Disease and Cancer

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F. Calabrò, C.N. Sternberg

Introduction

Inflammatory bowel disease (IBD) is a group of inflammatory conditions of both the large and small intestine. The major types of IBD are Crohn's disease (CD) and ulcerative colitis (UC). CD is also known as granulomatous colitis, regional enteritis, and ileitis. It is an inflammatory disease that may affect any part of the gastrointestinal tract from the mouth to the anus, causing a wide variety of symptoms. Because the symptoms of CD are similar to other intestinal disorders, such as irritable bowel syndrome and UC, it can be difficult to diagnose. In CD, all layers of the intestine may be involved, and normal healthy bowel can be found between sections of diseased bowel.

CD affects men and women equally and runs in some families. About 20% of patients with CD have a close relative with some form of IBD, with the highest risk occurring in individuals with siblings who have the disease. CD is most often diagnosed between the ages of 20 and 30. People of Jewish heritage have a higher risk, and African Americans a lower risk for developing CD.

In CD, nonpathogenic, commensal intestinal bacteria are thought to trigger a chronic deregulated immune response against mucosal barrier function [1,2], such that maintenance of the latter by adaptive and innate immunity is disrupted. The relationship between specific types of bacteria and CD is, however, unclear.

CD was first described in a report by Crohn and Rosenberg in 1925. Patients with all forms of IBD have an increased risk of developing intestinal cancer, and colorectal cancer in these patients has long been recognized.

In contrast to UC, in which the risk of colorectal cancer has been thoroughly investigated [3], the risk of intestinal malignancy in CD is less clearly defined. There

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is substantial variation among studies that have attempted to quantify the risk of colorectal cancer in CD. Methodological challenges relate to the heterogeneous nature of the disease, the extent of colonic inflammation (many patients have no colonic involvement), and surgical treatment, in which some of the at-risk tissue is removed.

Therapy

Treatment for patients with CD has changed over time. While traditional medication with 5-aminosalicylates is believed to provide some protection against colorectal cancer [4], immunosuppressive agents, including methotrexate and azathioprine, are carcinogenic [5]. Tumor necrosis factor (TNF) antagonists have been increasingly used in the treatment of CD, particularly for patients with severe disease. However, emerging data have pointed to the associated risk of lymphoma with these agents [6].

Few data are available in which the overall outcome of patients with CD who then develop colon cancer has been assessed. Likewise, studies assessing the particular treatment options in these patients are lacking.

Crohn's Disease and the Risk of Colorectal Cancer

Colorectal Cancer

Several publications have offered estimates of colorectal cancer risk in patients with CD. Gyde and colleagues reported the relative risk of colorectal cancer in Crohn's colitis to be 23.8, compared to 4.3 in the general population [7]. A landmark study from Sweden determined a relative risk of colorectal cancer of 5.6 for those with exclusively colonic involvement, compared to a relative risk of 3.2 for patients with ileocolitis and 1.0 for patients with ileal involvement only [8]. A subset analysis revealed that patients whose disease was diagnosed prior to age 30 had a higher relative risk than patients diagnosed at an older age, similar to patients with UC.

A meta-analysis of 12 hospital-based and population-based studies of colorectal cancer in CD revealed an overall relative risk of 2.5 (95% CI, 1.3–4.7) [9]. In the subset of patients with colonic disease, the risk rose to 4.5 (95% CI, 1.3–14.9), while for patients with ileal disease only, the risk was not significantly different from that of the general population. The cumulative risk of colorectal cancer for all patients with CD, regardless of disease distribution was 2.9% at 10 years, 5.6% at 20 years, and 8.3% after 30 years of disease.

Jess et al. carried out a meta-analysis restricted to population studies of intestinal cancer risk in CD [10]. Six papers met the inclusion criteria and reported varying estimates of relative risk of colorectal cancer, ranging from 0.9 to 2.2, with a pooled estimate of 1.9 (95% CI, 1.4–2.5).

In a study from the UK, patients with extensive CD were compared to those with

extensive UC with regard to colorectal cancer risk [11]. The results were similar, with a relative risk of developing colorectal cancer of 18 for Crohn's colitis and 19 for UC. The cumulate risk of colorectal cancer was 8% at 22 years for patients with CD vs. 7% at 22 years for patients with UC.

A Canadian matched-cohort study of 5529 patients with IBD found the incidence rate ratio (IRR) for colon cancer to be similarly increased in patients with UC (IRR = 2.75, 95%CI: 1.91–3.97) and CD (IRR = 2.64, 95% CI: 1.69–4.12). For rectal cancer, the IRR of patients with UC increased (IRR = 1.90, 95% CI: 1.05–3.43) whereas this was not the case in patients with CD (IRR= 1.08, 95% CI: 0.43 – 2.70). This study reinforced the findings that CD and UC patients share a similar risk of colorectal cancer. This is consistent with the finding that patients with CD have many of the same risk factors for developing colorectal cancer as patients with UC, including a younger age at diagnosis, a greater extent of colonic involvement, and longer duration of disease [12].

In contrast to the results from these studies, Jess et al. found no statistical increase in the incidence of colorectal cancer in 692 patients diagnosed with either UC or CD between 1940 and 2001, who resided in Olmsted County, Minnesota [13]. In this retrospective study, the standardized incidence ratio for colorectal cancer of patients with CD was 1.9 (95% CI: 0.7–4.1).

Although the incidence of colorectal cancer appears to be elevated in patients with IBD, there is substantial geographic variation. The reasons for these variations are unclear but may involve a combination of genetic factors, diet, and differences in the use of medications and/or preventive strategies [3].

While it is technically difficult to determine the exact risk of colorectal cancer associated with CD, it is generally accepted that patients with CD of the colon are at an increased risk for dysplasia and that this risk is related to the cumulative effect of colonic inflammation.

Small-Bowel Adenocarcinoma

Carcinomas of the small bowel are uncommon, representing only 1–5% of all gastrointestinal tract malignancies. Most cancers of the small bowel in CD are adenocarcinomas, usually in the terminal ileum or jejunum. The most common presentation of small-bowel cancer is intestinal obstruction. Other important symptoms include diarrhea, weight loss, and abdominal fistulae, but these symptoms are also found in CD.

The association of CD with small-bowel carcinoma is uncommon and to date only some 130 cases have been reported in the literature, since the first description of this entity in 1956 [14]. Nonetheless, the risk of small-bowel carcinoma in patients with CD is 60-fold higher than in the general population [15,16]. It was suggested that, surgically by-passed bowel segments are at particularly high risk of developing small bowel adenocarcinoma [17]. Most ileal carcinomas in CD are located in the strictures and are often diagnosed postoperatively [18,19].

Other interesting characteristics of small-bowel carcinomas are adjacent metapla-

sia, adenoma, and epithelial dysplasia, all of which underline the importance of further research with respect to the sequence of dysplasia in ileal adenocarcinoma in relation to CD [20,21].

Risk factors for small intestinal carcinoma in CD are a chronic active course, with stricture formation, fistulas, and onset of disease before the age of 30 years [19,22]. Surgery should be considered if the fistulae or strictures cannot be adequately examined, or symptoms substantially worsen.

Carcinoid Tumors

The coexistence of other intestinal neoplasms, including carcinoid tumors, in patients with CD has also been reported. To date, only a few case reports and case series have reported an association of carcinoid tumors in patients with CD, and evidence of an increased risk of carcinoid tumors in patients with CD is mixed.

In a retrospective study, the incidence of carcinoid tumor in patients with CD was 4 of 111 (3.6%) [23]. In comparison, only 3 of the 1999 patients (0.25%) who had an appendectomy also had an incidental appendiceal carcinoid tumor identified in the pathologic specimen. Thus, CD was associated with an increased incidence of carcinoid tumor, with an overall risk of 14.9 (95%, CI 2.5–102.5, $p<0.0001$). In all four patients, the tumor did not occur in an area of inflammation, which suggests that the development of carcinoid tumors in these patients is secondary to distant mediators rather than to local inflammatory effects from adjacent CD. Possible mediators include interferon (IFN)-γ or TNF-α . In addition, interleukin (IL)-12 has been implicated in both CD and in carcinoid tumors and may play a role in the development of these tumors [24]. In conclusion, the incidence of carcinoid tumors in patients with CD seems to be increased. Routine screening is not, however, indicated because the overall risk is quite low and in most cases the disease tends to have a benign course.

Risk Factors

Patients with UC and those with CD appear to have a similar risk of developing colorectal cancer. Nonetheless, studies have shown that certain patients with IBD may be at greater risk of developing cancer than others. Overall, factors that appear to increase the risk of colorectal cancer in patients with IBD include disease duration, severity, extent, family history of colorectal cancer, primary sclerosing cholangitis, age at diagnosis, degree of histologic inflammatory activity, and presence of dysplasia of any grade. The cumulative incidence of colorectal cancer has been shown to increase with increased duration of disease. The extent of the colitis is an independent risk factor for the development of colorectal cancer. The greater the amount of colonic surface involved with colitis, the greater the colorectal cancer risk. However, different criteria are sued to classify the extent of colitis.

Endoscopic and histologic evidence of inflammation are valid alternative criteria, particularly in high-risk patients. There are no studies that have correlated colorectal cancer risk with histopathologic extent of disease, even though microscopic evidence of colitis is arguably a better indicator of disease extent than either endoscopic or radiographic changes.

A positive family history of sporadic colorectal cancer is a risk factor for colon cancer in the general population. IBD patients with a family history of colorectal cancer have at least a two-fold higher risk of developing colorectal cancer [25]. Both UC and CD patients with primary sclerosing cholangitis are at particularly high risk for developing colorectal neoplasia [26]. Young age at onset of colitis has also been reported to be an independent risk factor for colorectal cancer in some studies [3]. Nonetheless, it is somewhat difficult to compare the exact relative risk for these various factors because of differences in the patient populations and in the analytical methods used.

Clinical Features of Colorectal Cancer in Crohn's Disease

Compared with sporadic colorectal carcinoma, colorectal cancers arising in patients with CD have several distinguishing features. These cancers generally affect individuals at a younger age compared to the general population. The carcinomas typically progress from flat non-polypoid dysplasia rather than arising as sporadic carcinoma. They also have a higher proportion of mucinous and signet ring cell histology, and the likelihood of finding two or more synchronous primaries is higher [27].

There is no agreement in the literature as to the anatomical distribution of colorectal cancer in CD patients. Some studies have in fact suggested a predominance of cancer in the right colon [28], whereas others have reported a high incidence of rectosigmoid carcinomas [29]. One study compared the clinicopathologic features of colorectal neoplasia complicating the two types of IBD. The median age at diagnosis of cancer, the frequency of mucinous adenocarcinoma, and the frequency of dysplasia adjacent to and distal from the cancer were similar in patients with UC and CD. All neoplastic lesions occurred in areas affected by IBD [30]. The frequently segmental topography of CD has, in the past led, to an approach favoring segmental resection of the area involved by the cancer. According to the results of that study, a more aggressive approach should be considered for patients with colorectal cancer and CD. But it is evident that further research is needed to clarify this issue.

Pathogenesis of Colorectal Cancer in Crohn's Disease

Molecular Pathology of Cancer in Crohn's Disease

The exact mechanism by which chronic inflammation results in carcinogenesis is unclear. Persistent inflammation is believed to cause increased cell proliferation as

well as oxidative stress and ultimately the development of dysplasia [31,32]. In addition, it is postulated that many of the molecular alterations responsible for sporadic colorectal cancer also play a role in the colitis-associated colon carcinogenesis. These events include microsatellite instability, inhibition of regulatory genes via hypermethylation of the promoter regions, and loss of adenomatous polyposis coli (APC), p53, and k-ras tumor suppressor functions. However, the timing and sequence of events differs from what is usually observed in patients with sporadic colorectal cancer. Whereas APC loss is considered an early development in the adenoma-carcinoma sequence of sporadic colorectal cancer, it is much less frequent and usually occurs late in the colitis-associated dysplasia-carcinoma sequence [33]. Conversely, p53 mutations, which represent the final mutation that transforms adenoma into carcinoma in sporadic tumors, occur early in patients with colitis and are often detected in a non-dysplastic mucosa [34].

Dysplasia in Crohn's Disease

Dysplasia represents a premalignant phase during which therapeutic intervention can prevent or minimize the complications associated with invasive cancer. An understanding of the definitions, diagnostic challenges, and natural history of dysplasia in CD is therefore essential. Although few studies have evaluated dysplasia in CD, some data support a dysplasia-carcinoma sequence [20,29]. Dysplasia is defined as unequivocal tumor of the epithelium confined to the basement membrane, without invasion of the lamina propria [35]. Dyplastic lesions can be classified as raised or flat based on their endoscopic appearance. Flat dysplasia is classically thought to be endoscopically invisible and is detected only on random biopsy specimens. Some studies, however, have demonstrated that newer generation colonscopes with higher optical resolution allow many of these lesions to be visualized by standard white-light endoscopy [36]. Elevated lesions that are endoscopically visible but not amenable to endoscopic resection are often referred to as DALMs (dysplasia-associated lesion or mass), a term attributable to the high rate of synchronous malignancy associated with these lesions. A newer term, ALM (adenoma-like lesion or mass), has been introduced to describe the finding of a polypoid lesion resembling a sporadic adenoma and located in an area of the colon not involved by chronic colitis. A standardized classification system introduced by Riddell [35] divides dysplasia into four categories: indefinite dysplasia, low-grade dysplasia (LGD), high-grade dysplasia (HGD), and cancer. Although this system has been widely accepted, its limitations include poor inter-observer agreement and intra-observer reliability even among expert gastrointestinal pathologists [37].

Dysplasia of any grade is associated with a risk of concurrent colorectal cancer in patients with IBD. The rate of progression from LGD to HGD or cancer has been evaluated in patients with UC who have undergone colectomy. In a review of ten prospective surveillance trials, 43% of patients who underwent colectomy because of DALM had coexistent colorectal cancer; 42% of patients with HGD and 16% of

patients with LGD who underwent immediate colectomy had synchronous colorectal cancer [38]. More recently, Rutter et al., from St Mark Hospital, reported that 20% of patients with LGD who proceeded to colectomy were found to have concurrent adenocarcinoma while 39.1% who were followed-up for LGD progressed to subsequent HGD or colorectal cancer [39]. The reported frequency of dysplasia in mucosa adjacent to colorectal cancer in patients with CD ranges from 40% [40] to 100% [29].

Management of Dysplasia

The management of dysplasia should rely on knowledge of its natural history. A description of the inflammation-dysplasia-carcinoma sequence facilitates our understanding of the molecular alterations involved in IBD-associated colorectal cancer. It is important, however, to recognize that this process does not necessarily occur in a systematic and sequential progression, from inflammation to indefinite dysplasia, to LGD, to HGD, and ultimately to invasive carcinoma.

Cancer can develop without any apparent preceding dysplasia and the natural history of LGD has been described to regress or to progress to cancer without necessarily evolving first into HGD [41]. This unpredictable course of dysplasia in IBD complicates efforts to develop molecular or histologic markers of neoplastic progression or future cancer risk.

Surveillance

The best proof that surveillance with colonoscopy effectively reduces colorectal cancer mortality would be a prospective randomized controlled trial in which patients with longstanding IBD undergo colonscopic surveillance whereas controls matched for a similar risk profile would not. However, due to ethical, financial, and practical limitations, this type of study is not likely to occur and we must, therefore, rely on retrospective studies. To date, only one practice-based surveillance study has been reported [42]. In that study, 259 patients with chronic Crohn's colitis were followed in a prospective colonoscopic surveillance program. Screening examination detected definite dysplasia or cancer in 18 patients (7%); 13 had LGD, 2 had HGD, and 3 had cancer. On surveillance examinations, dysplasia or cancer was found in an additional 30 patients (14%): 22 had LGD, 4 had HGD, and 4 had cancer. The cumulative risk of detecting an initial finding of any definite dysplasia or cancer after a negative screening colonoscopy was 25% by the tenth surveillance examination. The cumulative risk of detecting an initial finding of flat HGD or cancer after a negative screening colonoscopy was 7% by the ninth surveillance examination. While additional data are needed, a UC-based surveillance strategy seems reasonable [43]. An initial screening colonoscopy should be performed 8–10 years after the onset of symptoms attributable to CD.

Patients in whom at least one-third of the colon is involved are considered to have

extensive colitis. Those patients with left-sided or extensive colitis who have a negative screening examination should continue periodic surveillance at an interval of every 1 or 2 years. Any finding suspicious for dysplasia should be discussed with the pathologist. If the suspicion of dysplasia is high, surveillance with a repeat exam in 3–6 months may be indicated.

The management of LGD is a subject of debate among experts, with no clear consensus on optimal management. Patients should be informed regarding the risks and benefits of immediate surgery vs. colonoscopic surveillance [44]. If LGD is detected in a discrete polyp that can be resected and there is no flat dysplasia adjacent to the polyp, surveillance can be continued. If LGD is detected in a flat mucosa, surgery should be seriously considered.

The finding of flat HGD should prompt referral for immediate total proctocolectomy due to the high possibility of concurrent or subsequent malignancy. If HGD is diagnosed in an adenoma-like polyp and the affected area can be completely removed without evidence of flat dysplasia, surveillance can be continued.

A review assessed the role of endoscopic surveillance in prolonging life by permitting earlier detection of colon cancer or dysplasia in patients with IBD [45]. The conclusion was that there is no clear evidence that surveillance with colonoscopy prolongs survival in patients with extensive colitis. While cancers tend to be detected at an earlier stage in patients who undergo surveillance, and these patients have a correspondingly better prognosis, lead-time bias could contribute substantially to this apparent benefit.

As with any medical recommendation, the decision regarding surveillance vs. surgery should be personalized and discussed with the patient. Hopefully, surveillance strategies will become more defined as more knowledge of the natural history of dysplasia in CD will be obtained.

Surgery

The timing of surgery in CD is based on the evaluation of several factors, such as the failure of medical treatment, complications due to CD or to pharmacological therapy, growth retardation, and the development of dysplasia or cancer. An important question that remains to be clarified is whether patients with dysplasia or cancer in the setting of segmental Crohn's colitis should undergo segmental resection of the involved area or proceed to a more extensive UC-like approach. The same criteria as in UC apply regarding the indications for prophylactic colectomy [46]. When proctocolectomy is required, patients are most often advised to undergo permanent ileostomy, as there is a high rate of functional failure if ileal pouch reconstruction is performed. Only in highly selected patients with Crohn's colitis can ileal pouch reconstruction be recommended with the possibility of successful disease control and possibly cancer prevention [47].

Another dilemma in patients with Crohn's colitis is the management of strictures. A stricture in the colon of a patient with UC usually means an underlying malignan-

cy, especially if the stricture is causing symptoms and is located in the proximal colon [48]. However, since the majority of strictures in Crohn's colitis are benign, the patient can often be managed conservatively and followed with annual surveillance and biopsy if the lesion can be traversed with a standard pediatric colonscope. In the setting of longstanding CD, consideration should be given to surgical resection of the stricture due to the increased risk of cancer [43].

Chemoprevention

Despite the relative protection afforded by surveillance colonoscopies, there are still patients with IBD who develop colorectal cancer. This raises the question whether chemoprevention can reduce the risk of colorectal cancer in IBD patients. Chemoprevention refers to the use of chemical compounds to prevent, halt, or reverse the development of cancer. The goal of chemoprevention should be to reduce colorectal cancer risk, allowing for less frequent surveillance examinations and a reduction in the number of invasive cancers.

The bulk of evidence in favor of chemoprevention in IBD relates to the use of 5-aminosalicylates (5-ASA). Unfortunately, no prospective data exist and retrospective studies have yielded mixed results with regard to the protective effect of these drugs. Multiple studies have assessed the utility of 5-ASA in preventing dysplasia and cancer in patients with UC, whereas fewer studies have assessed this endpoint in patients with CD. A meta-analysis consisting of nine case-control and cohort studies in UC patients revealed a pooled odds ratio of 51% (95% CI, 0.38–0.69) for the development of dysplasia or cancer in patients who regularly used 5-ASA medications [49]. This is in contrast to a retrospective case-controlled study in 25 patients with IBD and colorectal cancer, which demonstrated no association between 5-ASA use and a reduction in colorectal cancer risk [50].

The mechanisms by which 5-ASA exerts its anti-inflammatory effects have not been completely elucidated. Proposed mechanisms include modulation of inflammatory cytokine production [51], inhibition of cyclo-oxygenase [52], inhibition of inducible nitric oxide synthase (an important final effecter of mucosal injury in IBD) [53], inhibition of nuclear factor κB (a transcription factor responsible for the expression of multiple genes involved in inflammatory responses and the promotion of carcinogenesis via the blockade of apoptosis) [54], activation of the peroxisome proliferator-activated receptor-γ (a nuclear receptor highly expressed in the colon that plays a key role in bacterial induced inflammation) [55]. These mechanisms underlie the anti-inflammatory effect of 5-ASA, and some may also contribute to their chemopreventive effects.

While other medications have been explored as potential chemopreventive agents, none have yielded satisfactory results. Currently, insufficient data preclude the use of folates, corticosteroids, NSAIDs, calcium, statins, and immunomodulators as chemoprotective agents against colorectal cancer.

Anti-TNF Therapy and the Risk of Malignancies

Infliximab is an anti-TNF agent that has been shown to be effective in the treatment of luminal and fistulizing CD. Induction and maintenance therapy with the drug was introduced into practice in 1998 [56,57]. Randomized controlled trials for the treatment of luminal CD have shown that two other anti-TNF agents, adalimumab and certolizumab, are also effective [58,59].

Due to their proven efficacy, TNF-antagonists are widely used in CD, but concerns have been raised about their long-term side effects. A meta-analysis in patients with rheumatoid arthritis treated with anti-TNF therapy found an increased risk of malignancies [60]. In a multicenter matched pair study, 404 patients with CD treated with infliximab were matched with 404 patients who had never received infliximab. The goal of the study was to assess whether infliximab is associated with an increased risk of cancer. Among the 404 patients treated with infliximab, cancer was diagnosed in nine (2.22%) while among the 404 not-treated patients, seven developed tumors (1.73%) (OR 1.33, 95% CI 0.46–3.84), $p = 0.40$). Thus, the incidence of tumors in CD associated with inflixumab use was comparable to that in patients who had never received infliximab.

A recent meta-analysis of placebo-controlled trials evaluated the efficacy and safety profile of TNF antagonists for CD [61]. In the overall analysis, there was no difference in the frequency of malignancies between anti-TNF and control groups (0.24 vs. 0.39%, respectively, 95% CI, 0.45–0.18). In the subgroup analysis, there was no difference between anti-TNF and control groups in short-term and long-term induction trials and in maintenance trials.. These data need to be interpreted with caution because of several limitations. Patients in clinical trials might not represent those seen in clinical practice; the control group usually had anti-TNF exposure; and follow-up might not be sufficiently long for serious events such as malignancy to occur.

Systemic Treatment of Colorectal Cancer in Patients with Crohn's Disease

Only one retrospective trial has compared the overall outcome for patients who have colon cancer and IBD with that of patients who have colorectal cancer alone. No difference in 5-year survival was found between 290 patients who had colorectal cancer and IBD, and age- and sex matched patients who had colorectal cancer but not IBD [62].

Furthermore, no data are available to assess the specific treatment options in patients who have CD and develop colorectal cancer. Patients with IBD who develop colorectal cancer are known to be at higher risk for developing severe diarrhea during chemotherapy, due to the toxic effect of the cytotoxic drug or to a flare of the underlying bowel disease. In patients with CD who have undergone a partial bowel resection, increasing diarrhea could indicate active IBD. For decades, treatment with

fluoropyrimidines has been the backbone of therapy; however, effective chemotherapeutics agents and targeted drugs have increasingly proven useful. These include fluorouracil (5-FU), a fluorinated pyrimidine, that acts primarily by inhibiting thymidylate synthetase and thereby impairs nucleotide synthesis and DNA replication. Almost 25 years after its development, 5-FU remains the central drug of colon cancer therapy. In one small retrospective study [63], 19 patients with colorectal cancer and IBD (10 patients had CD) were treated at the Memorial Sloan-Kettering Cancer Center with 5-FU based chemotherapy. Of these 19 patients, 53% experienced dose-limiting diarrhea. This is much higher than the rate of 5–15% that is typically associated with this treatment in randomized trials. The degree of active IBD prior to treatment had no effect or predictive value on the development of diarrhea. These observations have led investigators to recommend that patients with CD and gastrointestinal malignancies should be administered full-treatment doses of 5-FU but should be monitored carefully for toxicity effects.

Irinotecan is a semisynthetic topoisomerase-I inhibitor that causes the persistence of DNA strand breaks during replication. It is a prodrug that is hydrolyzed in the liver into the active compound SN-38. The most common dose-limiting toxicity is delayed-onset diarrhea, which occurs in up to 33% of treated patients. No reports are available for the use of irinotecan in patients with IBD, but one would expect a high rate of dose-limiting diarrhea and sepsis. Therefore, this drug should be used judiciously or avoided in patients with CD.

Oxaliplatin is a platinum compound that induces DNA damage by forming DNA adducts or cross-links and inducing apoptosis. It acts synergistically with 5-FU with regards to its therapeutic efficacy but also in relation to myelosuppression and diarrhea. The incidence of severe diarrhea in regimens containing oxaliplatin and 5-FU seems to be less than that observed with irinotecan and 5-FU. Therefore, this combination seems a more appropriate choice for patients with CD.

Bevacizumab is a humanized monoclonal antibody that targets vascular endothelial growth factor (VEGF), which is responsible for the stimulation of blood vessel growth in tumors. It has been approved by the US Food and Drug Administration and the EMEA in Europe for first-line treatment of advanced colorectal cancer in combination with any 5-FU-containing regimen. Because of the possibility of intestinal perforation associated with the use of this drug, it is necessary to be more cautious and it should be avoided in patients with CD.

Cetuximab is a recombinant chimeric monoclonal antibody that binds specifically to the epidermal growth factor receptor (EGFR) on normal and tumor cells, thus competitively inhibiting the binding of epidermal growth factor (EGF) and related ligands. Cetuximab is effective as second-line treatment, either alone or in combination with an irinotecan-containing regimen, in patients whose disease has progressed following treatment with another irinotecan-containing regimen. The observed paucity of severe gastrointestinal side effects in patients treated with cetuximab may be due to the drug's selectivity for its target and the relative quiescence of the targeted pathway in homeostatic adult tissue. During acute or chronic inflammation, these pathways may be activated to induce processes of tissue regeneration, repair, and growth. There are some data suggesting that the EGF pathway plays an important role

in repairing the inflamed colon. Theoretically, EGFR inhibition could exacerbate the severity of underlying disease. Despite these theoretical concerns, cetuximab is well-tolerated in patients with IBD.

There is no symptom-based method of distinguishing chemotherapy-induced diarrhea from IBD-associated diarrhea. For patients with CD who undergo chemotherapy for colorectal cancer and develop diarrhea, it is prudent to assume that there has been a disease flare and thus to treat with aminosalicylates. Immunosupressive drugs such as corticosteroids, 6-mercaptopurine, or TNF antagonists should be avoided during chemotherapy because they may lead to a higher incidence of hematological toxicity.

Conclusions

Patients with CD and IBD have an increased risk of colon cancer. However, having CD does not automatically imply the development of cancer; in fact, 90% of IBD patients do not develop cancer. While there is a link between CD and an increased chance of developing cancer, this risk is especially increased in patients who have had IBD for an extensive period of time, such as 8–10 years, and disease affecting the entire colon. If only a small part of the colon is involved and the disease has not been present for a long period of time, patients are less likely to develop cancer.

It may be difficult to detect colon cancer in Crohn's sufferers as its early symptoms often mimic those of IBD. Diarrhea and rectal bleeding are common symptoms in CD patients and may not cause concern. Regular colonoscopies should be performed in patients who have had CD for extended time periods. Colonoscopy with multiple biopsies is currently the most reliable method available.

Optimizing strategies for CD patients with colorectal cancer who undergo chemotherapy requires further research specifically for these patients, highlighted by an integrative multidisciplinary approach with collaboration among oncologists, gastroenterologists, and surgeons.

References

1. Schreiber S, Rosenstiel P, Albrecht M et al (2005) Genetics of Crohn disease, an archetypal inflammatory barrier disease. Nat Rev Genet 6(5):376-388
2. Sartor RB (2006) Mechanisms of disease: Pathogenesis of Crohn's disease and ulcerative colitis. Nat Clin Pract Gastroenterol Hepatol 3(7):390-407
3. Eaden JA, Abrams KR, Mayberry JF (2001) The risk of colorectal cancer in ulcerative colitis: A meta-analysis. Gut 48(4):526-535
4. van Staa TP, Card T, Logan RF et al (2005) 5-Aminosalicylate us and colorectal risk in inflammatory bowel disease: A large epidemiological study. Gut 54(11):1573-1578
5. Baumgart DC, Sandborn WJ (2007) Inflammatory bowel disease: Clinical aspects and established and evolving therapies. Lancet 369:1641-1657
6. Blomqvist P, Feltelius N, Löfberg R et al (2001) A 10-year survey of inflammatory bowel dis-

eases-drug therapy, costs and adverse reactions. Aliment Pharmacol Ther 15:475-481
7. Gyde SN, Prior P, Macartney JC et al (1980) Malignancy in Crohn's disease. Gut 21:1024-1029
8. Ekbom A, Helmick C, Zack M et al (1990) Increased risk of large-bowel cancer in Crohn's disease with colonic involvement. Lancet 336:357-359
9. Canavan D, Abrams KR, Mayberry J (2006) Meta-Analysis colorectal and small bowel cancer risk in patients with Crohn's disease. Aliment Pharmacol Ther 23:1097-1104
10. Jess T, Gamborg M, Matzen P et al (2005) Increased risk of intestinal cancer in Crohn's disease: A meta-analysis of population-based cohort studies. Am J Gastroenterol 100:2724-2729
11. Gillen CD, Walmsley RS, Prior P et al (1994) Ulcerative colitis and Crohn's disease: a comparison of the colorectal cancer risk in extensive colitis. Gut 25:1590-1592
12. Bernstein CN, Blanchard JF, Kliewer E et al (2001) Cancer risk in patients with inflammatory bowel disease: a population-based study. Cancer 91:854-862
13. Jess T, Loftus EV Jr, Velayos FS et al (2006) Risk of intestinal cancer in inflammatory bowel disease: a population-based study from Olmsted County, Minnesota. Gastroenterology 130:1039-1046
14. Koga H, Aoyagi K, Hizawa K et al (1999) Rapidly and infiltratively growing Crohn's carcinoma of the small bowel: serial radiologic findings and a review of the literature. Clin Imaging 23:298-301
15. Solem CA, Harmsen WS, Zinsmeister AR et al (2004) Small intestinal adenocarcinoma in Crohn's disease: a case-control study. Inflamm Bowel Dis 10:32-35
16. Jess T, Winther KV, Munkholm P et al (2004) Intestinal and extra-intestinal cancer in Crohn's disease: follow-up of a population-based cohort in Copenhagen County, Denmark. Aliment Pharmacol Ther 19:287-293
17. Barwood N, Platell C (1999) Case report: adenocarcinoma arising in a Crohn's stricture of the jejunum. J Gastroenterol Hepatol 14:1132-1134
18. Marchetti F, Faxio VW, Ozuner G (1996) Adenocarcinoma arising from a structureplasty site in Crohn's disease. Report of a case. Dis Colon Rectum 39:1315-1321
19. Partridge SK, Hodin RA (2004) Small bowel adenocarcinoma at a strictureplasty site in a patient with Crohn's disease: report of a case. Dis Colon Rectum 47:778-781
20. Sigel JE, Petras RE, Lashner BA et al (1999) Intestinal adenocarcinoma in Crohn's disease: a report of 30 cases with a focus on coexisting dysplasia. Am J Surg Pathol 23:651-655
21. Petras RE, Mir-Madjlessi SH, Farmer RG (1987) Crohn's disease and intestinal carcinoma. A report of 11 cases with emphasis on associated epithelial dysplasia. Gastroenterology 93:1307-1314
22. Christodoulou D, Skopelitou AS, Katsanos KH et al (2002) Small bowel adenocarcinoma presenting as a first manifestation of Crohn's disease: report of a case, and a literature review. Eur J Gastroenterol Hepatol 14:805-810
23. West NE, Wise PE, Herline AJ et al (2007) Carcinoid tumors are 15 times more common in patients with Crohn's disease. Inflamm Bowel Dis 13:1129-1134
24. Gordon, JN, MacDonald TT (2005) Osteopontin: a new addition to the constellation of cytokines which drive T helper cell type 1 responses in Crohn's disease. Gut 54:1213-1215
25. Askling J, Dickman PW, Karlén P et al (2001) Family history as risk factor for colorectal cancer in inflammatory bowel disease. Gastroenterology 120:1356-1362
26. Jayaram H, Satsangi J, Chapman RW (2001) Increased colorectal neoplasia in chronic ulcerative colitis complicated by primary sclerosing cholangitis: Fact or fiction? Gut 48:430-434
27. Xie J, Itzkowitz SH (2008) Cancer in inflammatory bowel disease. World J Gastroenterol 14:378-389
28. Stahl TJ, Schoetz DJ Jr, Roberts PL et al (1992) Crohn's disease and carcinoma: increasing justification for surveillance? Dis Colon Rectum 35:850-856

29. Hamilton SR (1985) Colorectal carcinoma in patients with Crohn's disease. Gastroenterology 89:398-407
30. Svrcek M, Cosnes J, Beaugerie L et al (2007) Colorectal neoplasia in Crohn's colitis: a retrospective comparative study with ulcerative colitis. Histopathology 50:574-583
31. Itzkowitz SH, Yio X (2004) Inflammation and cancer IV. Colorectal cancer in inflammatory bowel disease: The role of inflammation. Am J Physiol Gastrointest Liver Physiol 287:G7-17
32. Sato F, Shibata D, Harpaz N et al (2002) Aberrant methylation of the HPP1 gene in ulcerative colitis-associated colorectal carcinoma. Cancer Res 62:6820-6822
33. Redston MS, Papadopoulos N, Caldas C et al (1995) Common occurrence of APC and K-ras gene mutations in the spectrum of colitis-associated neoplasias. Gastroenterology 108:383-392
34. Burmer GC, Rabinovitch PS, Haggitt RC et al (1992) Neoplastic progression in ulcerative colitis: Histology, DNA content, and loss of a p53 allele. Gastroenterology 103:1602-1610
35. Riddell RH, Goldman H, Ransohoff DF et al (1983) Dysplasia in inflammatory bowel disease: standardized classification with provisional clinical applications. Hum Pathol 14:931-968
36. Rubin DT, Rothe JA, Hetzel JT et al (2007) Are dysplasia and colorectal cancer endoscopically visible in patients with ulcerative colitis? Gastrointest Endosc 65:998-1004
37. Odze RD, Goldblum J, Noffsinger A et al (2002) Interobserver variability in the diagnosis of ulcerative colitis-associated dysplasia by telepathology. Mod Pathol 15:379-386
38. Bernstein CN, Shanahan F, Weinstein WM (1994) Are we telling the truth about surveillance colonoscopy in ulcerative colitis? Lancet 343:71-74
39. Rutter MD, Saunders BP, Wilkinson KH et al (2006) Thirty-year analysis of a colonoscopic surveillance program for neoplasia in ulcerative colitis. Gastroenterology 130:1030-1038
40. Connell WR, Sheffield JP, Kamm MA et al (1994) Lower gastrointestinal malignancy in Crohn's disease. Gut 35:347-352
41. Itzkowitz SH, Harpaz N (2004) Diagnosis and management of dysplasia in patients with inflammatory bowel diseases. Gastroenterology 126:1634-1648
42. Friedman S, Rubin PH, Bodian C et al (2008) Screening and surveillance colonoscopy in chronic Crohn's colitis: results of a surveillance program spanning 25 years. Clin Gastroenterol Hepatol 6:993-998
43. Itzkowitz SH, Present DH, Crohn's and Colitis Foundation of America Colon Cancer in IBD Study Group (2005) Consensus conference: colorectal cancer screening and surveillance in inflammatory bowel disease. Inflamm Bowel Dis 11:314-321
44. Zisman TL, Rubin DT (2008) Colorectal cancer and dysplasia in inflammatory bowel disease. World J Gasroenterol 14:2662-2669
45. Collins PD, Mpofu C, Watson AJ et al (2006) Strategies for detecting colon cancer and/or dysplasia in patients with inflammatory bowel disease. Cochrane Database Syst Rev CD000279
46. Alos R, Hinojosa J (2008) Timing of surgery in Crohn's disease: a key issue in the management. World J Gastroenterol 14:5532-5539
47. Morpurgo E, Petras R, Kimberling J et al (2003) Characterization and clinical behaviour of Crohn's disease initially presenting predominantly as colitis. Dis Colon Rectum 46:918-924
48. Gumaste V, Sachar DB, Greenstein AJ (1992) Benign and malignant colorectal strictures in ulcerative colitis. Gut 33:938-941
49. Velayos FS, Terdiman JP, Walsh JM (2005) Effect of 5-aminosalicylate us on colorectal cancer and dysplasia risk: a systematic review and metaanalysis of observational studies. Am J Gastroenterol 100:1345-1353
50. Bernstein CN, Blanchard JF, Metge C et al (2003) Does the use of 5-aminosalicylates in inflammatory bowel disease prevent the development of colorectal cancer? Am J Gastroenterol 98:2784-2788

51. Zimmerman MJ, Jewell DP (1996) Cytokines and mechanisms of action of glucocorticoids and aminoslaicylates in the treatment of ulcerative colitis and Crohn's disease. Aliment Pharmacol Ther 10(Suppl 2):93-98
52. Allgayer H (2003) Review article: mechanisms of action of mesalazine in preventing colorectal carcinoma in inflammatory bowel disease. Aliment Pharmacol Ther 18(Suppl 2):10-14
53. Hasko G, Szabo C, Németh ZH et al (2001) Sulphasalazine inhibits macrophage activation: Inhibitory effects on inducible nitric oxide synthase expression, interleukin-12 production and major histocompatibility complex II expression. Immunology 103:473-478
54. Greten FR, Eckmann L, Greten TF et al (2004) IKKbeta links inflammation and tumorigenesis in a mouse model of colitis-associated cancer. Cell 118:285-296
55. Rousseaux C, Lefebvre B, Dubuguoy L et al (2005) Intestinal anti-inflammatory effect of 5-aminosalicylic acid is dependent on peroxisome proliferator-activated receptor-gamma. J Exp Med 201:1205-1215
56. Hanauer SB, Feagan BG, Lichtenstein GR (2002) Maintenance infliximab for Crohn's disease: the ACCENT I randomised trial. Lancet 359:1541-1549
57. Sands BE, Anderson FH, Bernstein CN et al (2004) Infliximab maintenance therapy for fistulizing Crohn's disease. N Engl J Med 350:876-885
58. Hanauer SB, Sandborn WJ, Rutgeerts P et al (2006) Human anti-tumor necrosis factor monoclonal antibody (adalimumab) in Crohn's disease: the CLASSIC-I trial. Gastroenterology 130:323-333, quiz 591
59. Sandborn WJ, Feagan BG, Stoinoy S et al (2007) Certolizumab pegol for the treatment of Crohn's disease. N Engl J Med 357:228-238
60. Bongartz T, Sutton AJ, Sweeting MJ et al (2006) Anti-TNF antibody therapy in rheumatoid arthritis and the risk of serious infections and malignancies: systematic review and meta-analysis of rare harmful effects in randomized controlled trials. JAMA 295:2275-2285
61. Peyrin-Biroulet L, Deltenre P, De Suray N et al (2008) Efficacy and safety of tumor necrosis factor antogonists in Crohn's disease: meta-analysis of placebo-controlled trials. Clin Gastroenterol Hepatol 6:644-653
62. Delaunoit T, Limburg PJ, Goldberg RM et al (2006) Colorectal cancer prognosis among patients with inflammatory bowel disease. Clin Gastroenterol Hepatol 4:335-342
63. Tiersten A, Saltz LB (1996) Influence of inflammatory bowel disease on the ability of patients to tolerate systemic fluorouracil-based chemotherapy. J Clin Oncol 14:2043-2046

Crohn's Disease and Malignant Lymphomas

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Introduction

Crohn's disease (CD) and ulcerative colitis (UC) are the two major subtypes of inflammatory bowel disease (IBD). Hematological changes, including anemia, hypercoagulable state, thrombocytosis, leukocytosis, and acute or chronic forms of leukemia, have been reported in IBD patients [1]. Patients with IBD are also suspected of being at increased risk of malignant lymphomas, especially non-Hodgkin's lymphoma (NHL), as of other inflammatory disorders, namely rheumatoid arthritis (RA), Sjögren syndrome, and systemic lupus erythematosus (SLE) [2]. In all these conditions, as in IBD, the respective role of chronic inflammation and of immunosuppressive treatment in the development of lymphomas has not been elucidated, although the role of the immune system in the development of malignant disorders, especially lymphomas, is well recognized.

The prevalent distribution of lymphoma in the tracts of inflamed bowel suggests a causal relationship with IBD. Lymphoma might develop as a result of the prolonged stimulation of mucosa-associated lymphoid tissue (MALT). This could also explain the increased risk of NHL in RA patients who do not receive immunosuppressive therapy [3]. But intestinal lymphoma can also masquerade as IBD. In fact, the gastrointestinal tract is the most common site of extranodal lymphomas, and the duration of IBD at the time of lymphoma diagnosis is often quite short [4]. In other words, lymphoma may be already present when IBD is diagnosed or may mimic the clinical picture of IBD.

Agents with proven efficacy in the treatment of IBD include azathioprine (AZA), 6-mercaptopurine (6-MP) and methotrexate (MTX) [5,6]. New biological agents [7] have been recently introduced, in particular infliximab. Concerns regarding infec-

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tious and malignant sequelae related to the long-term effects of these drugs have emerged; in particular, an actual risk of lymphoma has been attributed to their prolonged use [8].

In a large population-based study [9] of 6,000 patients with CD, 10,000 with UC and more than 60,000 control subjects randomly selected from non-IBD patients, the relative risk (RR) of lymphoma in either of the patient groups was found not to reach statistical significance. Another large study [10] of over 47,000 Swedish patients with CD or UC found 264 hematopoietic cancers during follow-up, which corresponds to a borderline significant 20% increased risk in both UC and CD; myeloid leukemia occurred significantly more often than expected. Recently, Hemminki et al. [11] published a study on 21,788 hospitalized CD patients throughout Sweden. In this group, 1,424 patients developed cancer, but the highest standardized incidence ratio (SIR) for all patients was reported for small intestinal cancer (SIR 13.82) followed by cancers of the colon, liver, testis, and kidney and by NHL (SIR 2.54), with ten-fold more cases of NHL than of Hodgkin's disease (HD). The lymphoma risk was also higher than in other studies. Unfortunately, there are no data on the medications used by the study patients. A meta-analysis [12] quantified the risk of malignancies in 60,122 patients with CD. Compared with the normal population, there was an increased risk of small bowel, colorectal, and extra-intestinal cancers as well as of lymphoma (RR 1.42). Whether the severity and duration of disease contribute to the risk of lymphoma is matter of discussion.

A profound immunodeficiency is associated with the loss of Epstein-Barr virus (EBV)-specific cytotoxic T-lymphocytes and may result in EBV-driven malignant proliferation. Also in IBD, some reports have pointed to a potential association between the use of thiopurines and the development of EBV-related lymphomas. Dayarsh et al. [13] identified 18 IBD patients diagnosed with lymphoma between 1985 and 2000. Six of them were on AZA or 6-MP. Pathology specimens showed evidence of EBV in five of the six patients treated with thiopurines but in only two of the ten patients not on immunosuppressive agents. Moreover, EBV DNA and EBV gene products have been observed in the mucosal samples of patients with IBD who had gastrointestinal lymphomas [14]. Several population-based studies have examined the risk of lymphoma in patients with IBD who had been treated with AZA and MP, reporting conflicting results. A meta-analysis of six cohort studies meeting severe inclusion criteria was recently carried out by Kandiel et al. [15]. Among the 11 lymphomas, there were nine cases of NHL and two of HD, compared to the expected 2.6 cases overall. Of the nine NHLs, four originated in the bowel and one in the central nervous system. Lewis et al. [16] used a Markov decision analysis model to show that the benefits of AZA treatment clearly outweigh the risk. This benefit was greatest in young patients, who have the lowest baseline risk of lymphoma and the greatest life expectancy in the absence of a CD-related death. Farrel et al. [4] analyzed 782 patients, including 238 who were on immunosuppressive therapy. Four patients, all with prior immunosuppressive therapy, had NHL (three cases were intestinal and one was mesenteric) as compared with no cases of NHL among the remaining 544 IBD patients without prior immunosuppression (SIR 31.2; p=0.0001).

A number of adverse events have been associated with infliximab, including lym-

phomas. RA studies offer meaningful data. A recently published meta-analysis [17] reports 24 malignancies (10 malignant lymphomas) among 3,493 RA patients treated with antibodies to tumor necrosis factor (TNF), while there were only two malignancies (and no lymphomas) among the 1,512 control patients. Malignancies were significantly more common in patients treated with high doses than with low doses of anti-TNF antibodies. In contrast, studies on IBD offer no evidence. In a retrospective review [18] of 117 patients with CD treated with infliximab, no case of cancer was reported. In another retrospective analysis [19], the medical records of 500 CD patients treated with infliximab were reviewed. Nine newly diagnosed cancers were reported but only two patients developed lymphoma (1 NHL and 1 HD). In a Swedish population-based cohort study, 212 IBD patients treated with infliximab were observed for 28 months [20]. Half the patients received concomitant therapy with AZA, 6-MP, or corticosteroids. Three cases of lymphoma were reported in CD patients (1.4%), one was a NK-cell lymphoma and two were B-cell lymphomas. The incidence rate of lymphoma in this cohort was slightly higher than the population-based incidence. No increase in lymphoma incidence was reported by a study from Italy [21], in which 404 CD patients treated with infliximab were compared with 404 matched CD patients who had never received the biological agent. The post-marketing surveillance registry [22] enrolled 6,290 patients, 3,179 of whom received infliximab while 3,111 were treated with other therapies. There was no statistically difference in terms of incidence of all malignancies, including lymphoma. Patients treated with infliximab had more severe CD but the rates of mortality and neoplasias were similar to those in patients treated with other therapies. Moreover, the risk of lymphoma should be weighted also against other endpoints. Siegel et al. [23] analyzed the quality of life in patients treated with infliximab. Their complex simulation model showed that the infliximab-treated group would experience more quality-adjusted life-years than the standard therapy group.

Concerning the pathological findings, recent data have focused on the association of IBD, infliximab, and hepatosplenic T-cell lymphoma [24–26], a rare form of aggressive NHL that comprise 5% of peripheral T-cell NHLs and often occurs in young adult males, either de-novo or as secondary tumor in immunocompromised patients. Ten patients, age 12 to 32 years, were described in a recent review by Rosh et al. [26]. The number of infliximab infusions ranged from 1 to 24. All ten patients were on concomitant immunosuppression with AZA or 6-MP. Nine were affected by CD. Six patients died of their lymphoma. It is not clear whether the apparently increased incidence of hepatosplenic T-cell lymphoma was due to infliximab, 6-MP, AZA or a combination of the three drugs.

Overall, the data show that, in patients with CD, there is probably a slightly increased risk of lymphoma. The risk seems to be lesser than in autoimmune diseases such as RA and SLE [27]. However, the question whether the risk of developing a lymphoma is related to the treatment itself, to disease severity, to concomitant immunomodulation, or to all these factors in combination has been largely ignored.

Once a diagnosis of lymphoma has been made in a patient with IBD, treatment should be instituted. While this is usually done by the hematologist or the oncologist, gastroenterologists should be aware of the risks and possible advantages of

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chemotherapy, radiotherapy, or immunotherapy in these patients. In the literature, however, the issue of treatment in IBD-associated lymphomas has received no attention. We have data for other autoimmune disorders, such as RA, for which standard chemotherapy, such as CHOP, alone or in association with the monoclonal anti-CD20 antibody (rituximab), is reported to be well tolerated and often (77% of cases) induces a remission also of the underlying rheumatoid disease [27].

In our practice, patients with IBD and associated lymphoma receive standard treatment regimens. The exact strategy largely depends on lymphoma histology and clinical stage. The CHOP regimen is considered the standard of care in aggressive NHL, and CVP in indolent clinicopathological types [28]. As in non-IBD-associated lymphomas, rituximab is added as it seem to improve chemotherapy results. Once the lymphoma has been diagnosed, the staging carefully accomplished, and treatment planned, immunosuppressive therapy for IBD (e.g., AZA, 6-MP, MTX) should be stopped to avoid the toxic effects incurred by combining the two treatments.

The use of high-dose myeloablative chemo-radiotherapy followed by transplantation of the autologous or allogeneic hematopoietic stem cells deserves special consideration [29,30]. This treatment is often given to eradicate malignant lymphomas in patients with poor-prognosis disease, but is able to induce a durable response also in those with IBD. In one study, 18 patients with CD underwent allogeneic transplantation for concomitant hematological disorders [31]. At the time of transplantation, six patients were in remission of CD and 12 had moderate to severe disease, with a history of CD complications including fistulae, obstructions, and extraintestinal manifestations. The majority of patients received cyclophosphamide and total body irradiation (TBI) as conditioning regimen. Following transplantation, 11 patients achieved CD remission. There were two transplant-related deaths. At the last follow-up reported, seven out of 18 were off immunosuppressive drugs.

The results achieved in seven patients with CD who received an autologous transplantation for treatment of an underlying lymphoma, acute leukemia, or breast cancer have been described in the literature [32,33]. Prior to hematopoietic stem cell transplantation, the majority of these patients had active IBD with complications necessitating surgery. All transplanted patients achieved clinical CD remission, with a median follow-up of 60 months. At the time of this writing, only two of these patients continued their medications. These data suggest that high-dose therapy results in CD remission without a need for ongoing immunosuppressive treatment and with a reasonable hope for cure. They also encourage the use of autologous transplantation for the treatment of CD itself [34].

References

1. Caspi O, Polliack A, Klar R et al (1995) The association of inflammatory bowel disease and leukemia – coincidence or not? Leuk Lymphoma 17:255–262
2. Leandro MJ, Isenberg DA (2001) Rheumatic diseases and malignancy – is there an association? Scand J Rheumatol 30:185–188
3. Gridley G, Mc Laughlin JK, Ekbom A et al (1993) Incidence of cancer among patients with rheumatoid arthritis. J Natl Cancer Inst 85:307–311

4. Farrell RJ, Ang Y, Kileen P et al (2000) Increased incidence of non-Hodgkin's lymphoma in inflammatory bowel disease patients on immunosuppressive therapy but overall risk is low. Gut 47:514–519
5. Pearson DC, May GR, Fick GH et al (1995) Azathioprine and 6-mercaptopurine in Crohn disease. A meta-analysis. Ann Intern Med 123:132–142
6. Feagan BG, Fedorak RN, Irvine EJ et al (2000) A comparison of methotrexate with placebo for the maintenance of remission in Crohn's disease. North American Crohn's Study Group Investigators. N Engl J Med 342:1627–1632
7. Cominelli F (2004) Cytokine-based therapies for crohn's disease – new paradigms. N Engl J Med 351:2045
8. Aithal GP, Mansfield JC (2001) The risk of lymphoma associated with inflammatory bowel disease and immunosuppressive treatment. Aliment Pharmacol Ther 15:1101–1108
9. Lewis JD, Bilker WB, Brensinger C et al (2001) Inflammatory bowel disease is not associated with an increased risk of lymphoma. Gastroenterology 121:1080–1088
10. Askling J, Brandt L, Lapidus A et al (2005) Risk of haematoepoietic cancer in patients with inflammatory bowel disease. Gut 54:617–622
11. Hemmiki K, Li X, Sundquist J et al (2009) Cancer risk in Crohn disease patients. Ann Oncol 20:574–580
12. Voon Roon AC, Reese G, Teare J et al (2007) The risk of cancer in patients with Crohn's disease. Dis Colon Rectum 50:839–855
13. Dayharsh GA, Loftus EV, Sandborn WJ et al (2002) Epstein-Barr virus-positive lymphoma in patients with inflammatory bowel disease treated with azathioprine or 6-mercaptopurine. Gastroenterology 122:72–77
14. Wakefield AJ, Fox JD, Sawyerr AM et al (1992) Detection of herpesvirus DNA in the large intestine of patients with ulcerative colitis and Crohn's disease using the nested polymerase chain reaction. J Med Virol 38:183–190
15. Kandiel A, Fraser AG, Korelitz BI et al (2005) Increased risk of lymphoma among inflammatory bowel disease patients treated with azathioprine and 6-mercaptopurine. Gut 54:1121–1125
16. Lewis JD, Schwartz JS, Lichtenstein GR (2000) Azathioprine for maintenance of remission in Crohn's disease: benefits outweigh the risk of lymphoma. Gastroenterology 118:1018–1024
17. Bongartz T, Sutton AJ, Sweeting MJ et al (2006) Anti-TNF antibody therapy in rheumatoid arthritis and the risk of serious infections and malignancies:systematic review and meta-analysis of rare harmful effects in randomized controlled trials. JAMA 295:2275–2285
18. Kinney T (2003) Immunomodulators and "on demand" therapy with infliximab in Crohn's disease: clinical experience with 400 infusions. Am J Gastroenterol 98:608–612
19. Colombel JF, Loftus EV Jr, Tremaine WJ et al (2004) The safety profile of infliximab in patients with Crohn's disease:the Mayo Clinic experience in 500 patients. Gastroenterology 126:19–31
20. Ljung T, Karlen P, Schmidt D et al (2004) Infliximab in inflammatory bowel disease: clinical outcome in a population based cohort from Stockholm County. Gut 53:849–853
21. Biancone L, Orlando A, Kohn A et al (2006) Infliximab and newly diagnosed neoplasia in Crohn's disease: a multicentre matched pair study. Gut 55:228–233
22. Lichtestein GR, Feagan BG, Cohen RD et al (2006) Serious infections and mortality in association with therapies for Crohn's disease: TREAT registry. Clin Gastroenterol Hepatol 4:621–630
23. Siegel CA, Hur C, Korzenik JR et al (2006) Risks and benefits of infliximab for the treatment of Crohn's disease. Clin Gastroenterol Hepatol 4:1017–1024
24. Mittal S, Milner BJ, Johnston PW et al (2006) A case of hepatosplenic gamma-delta T-cell lymphoma with a transient response to fludarabine and alemtuzumab. Eur J Haematol 76:531–534

25. Thayu M, Markowitz JE, Mamula P et al (2005) Hepatosplenic T-cell lymphoma in an adolescent patient after immunomodulator and biologic therapy for Crohn's disease. J Pediatr Gastroenterol Nutr 40:220–222
26. Rosh J R., Gross T, Mamula P, Griffiths A, Hyams J. (2007) Hepatosplenic T-cell lymphoma in adolescents and young adults with Crohn's disease:a cautionary tale? Inflamm Bowel Dis 13:1024–1030
27. Wöhrer S, Troch M, Zwerina J et al (2007) Influence of rituximab, cyclophosphamide, doxorubicin, vincristine and prednisone on serologic parameters and clinical course in lymphoma patients with autoimmune diseases. Ann Oncol 18:647–651
28. Michallet AS, Coiffier B (2009) Recent developments in the treatment of aggressive non-Hodgkin lymphoma. Blood Rev 23 (1):11–23
29. Maloney D (2008) Allogeneic transplantation following non-myeloablative conditioning for aggressive lymphoma. Bone Marrow Transplant (Suppl 1):535–536
30. Ladetto M, De Marco F, Benedetti F et al (2008) Prospective, multicenter randomized GITMO/IIL trial comparing intensive (R-HDS) versus conventional (CHOP-R) chemoimmunotherapy in hig-risk follicular lymphoma at diagnosis:the superior disease control of R-HDS does not translate into an overall survival advantage. Blood 111:4004–4013
31. Fortun PJ & Hawkey CJ. (2008) The role of stem cell transplantation in inflammatory bowel disease. Autoimmunity 41 (8):654–659
32. Castro J, Bentch H, Smith L et al (1996) Prolonged clinical remission in patient with inflammatory bowel disease (IBD) after high dose chemotherapy (HDC) and autologous bood stem cell transplantation (ABSCT). Blood 88 (Supplement):133 A
33. Anumakonda V, Hayee B, Chung-Faye (2007) Remission and relapse of Crohn's disease following autologous haematopoietic stem cell transplantation for non-Hodgkin's lymphoma. Gut 56:1325
34. Oyama Y, Craig RM, Traynor AR et al (2005) Autologous hematopoietic stem cell transplantation in patients with refractory Crohns disease. Gastroenterology 128:552–563

Psychological and Social Aspects

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Context and History

Psychological factors can have an impact on every medical condition, if only because a patient's state of mind will affect how assiduously he or she will take the prescribed medications. Psychophysiological phenomena vary in their importance according to the disease, and are arguably strongest in the so-called functional disorders. In irritable bowel syndrome, for example, symptoms are highly vulnerable to the effects of psychological distress on bowel motility and on the threshold for perceiving internal phenomena as painful. But such mechanisms can be important in "organic" conditions as well – watching your national soccer team play in the World Cup can bring on a myocardial infarction [1] – so the severity of Crohn's disease (CD) does not exclude an impact of psychological factors on onset and clinical evolution.

The first description of CD happened to coincide with a period of enthusiasm for psychosomatics, fueled by researchers armed with pathophysiological sophistication and psychoanalytic theory. Ulcerative colitis (UC) was already considered a prime "psychosomatic" disease, and CD was promptly added to the list.

For many years, the clinical literature supporting this concept was beset by methodological limitations and hampered by excessive enthusiasm, so it was no surprise when in the 1970s and 1980s enthusiasm for biomedical reductionism led to a denial of any effects of psychological factors in inflammatory bowel disease (IBD) [2]. Such swings are frequent in modern medicine – good examples are the roles of dietary fiber in diverticulosis and of dietary calcium in urolithiasis.

During the 1990s the story changed again. Studies carried out with modern scientific acumen began to produce evidence that psychological factors have a role in IBD after all, at the same time that mind-body interactions were gaining legitimacy through advances in fields such as psychoneuroimmunology [3].

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Many physicians and IBD patients believe that psychological factors contribute to the clinical evolution of the disease [4, 5]. Patients in particular are often convinced that oscillations in their disease are tightly linked to stress or distress, out of proportion to the modest effect documented in the scientific literature.

This chapter will attempt to provide gastrointestinal surgeons with a perspective on the relation between body and psyche in CD, by reviewing the effect of psychological factors on animal models of IBD; examining similar research in human subjects; discussing the impact of CD on patients' lives; and reviewing research and expert experience on the role of professional psychological support in the treatment of these patients.

Stress in Animal Models of Inflammatory Bowel Disease

Exposing laboratory animals to psychological stressors can, by itself, induce bowel inflammation [6], and stress can have a strong potentiating effect on experimental colitis induced by chemical agents such as trinitrobenzenesulfonic acid, dinitrobenzenesulfonic acid, or dextran sulfate sodium [7]. In an elegant series of experiments, Collins and co-workers induced an experimental colitis in rodents using high-dose di- or trinitrobenzenesulfonic acid, allowed the animals to recover, and then attempted to trigger relapses using lower doses. Restraint and noise stress proved to act as potentiators, enabling inflammation to develop after exposure to what would otherwise have been sub-threshold levels of the drugs [7,8].

Mechanisms of the effects of stress on gut inflammation in animal models have recently been authoritatively reviewed [9]. Increased gut permeability to intestinal antigens and bacteria is a key mediator [10,11], with involvement of corticotropin-releasing hormone, mast cells, and cholinergic nerves [12].

Early separation of young mice and rats from the mother – considered as a murine model of depression – leads to increased gut permeability to bacterial antigens and to enhanced vulnerability to chemical colitis in adulthood [13,14], suggesting a role for early experiences in intestinal inflammation that occurs in later life.

Conclusion: Physical and psychological stressors consistently increase intestinal permeability in animals, potentially causing inflammation themselves and/or facilitating colitis induced by chemical agents.

The Effect of Psychological Factors on Crohn's Disease

Laboratory Stressors

In an elegant laboratory study, IBD patients and healthy adults were exposed for 5 days to a cold pressor stressor, with rectal biopsies performed before and after the stress period. The colonic mucosa of both subject groups reacted with inflammatory

changes, including activation and degranulation of mucosal mast cells, mitochondrial damage to epithelial cells, and mucosal protein oxidation. These changes were more marked in IBD patients than in healthy subjects, with no difference between those with UC and CD [15].

In another study, patients with inactive UC underwent a psychological stress procedure followed by rectal biopsy. Mucosal blood flow decreased after stress, while mucosal inflammatory indicators, including TNF-α release and reactive oxygen metabolite production, increased substantially [16].

Initial Diagnosis

Psychosocial factors play little role in the origins of CD, judging from a prospective study that used population-wide registries to compare all 21,062 parents who had experienced the death of a child in Denmark between 1980 and 1996 to parents whose children had not died [17]. This extremely traumatic stressor did not add to the risk of a new diagnosis of either UC or CD over the following 6–23 years, nor did it lead to an increased number of hospitalizations for IBD.

A recent well-performed case-control study of newly diagnosed patients did find an excess of stressful life events during the months before disease onset in patients with CD, which was statistically accounted for by increased levels of two possible mediators: depressive symptoms and anxiety [18]. Stress and distress may therefore accelerate the initial inflammatory process in a person with preclinical disease – or may simply bring the date of diagnosis forward due to the hypervigilance to somatic phenomena that is typical of anxiety and depression.

Disease Course

Patients often recall having had high stress levels [19] or stressful life events [20] before periods of active disease, but such retrospective data are limited by the possibility of confounding by recall bias and by the distressing effects of the disease itself.

More convincingly, several groups have evaluated psychosocial factors among CD patients during periods of relative disease inactivity and tracked the subsequent course of the disease over time. Mittermaier and colleagues found that depressive symptoms at baseline increased the risk of exacerbation over the subsequent 18 months [21]. Mardini and colleagues found that depressive symptoms worsened disease activity after a lag period of 2–3 months [22]. Bitton and colleagues found a strong trend for depression to predict subsequent exacerbation, at a $p = 0.10$ level [23]. Perhaps the most convincing evidence comes from Persoons et al., who studied 100 patients undergoing treatment with infliximab [24]. Major depression at baseline, found in 29% of subjects, substantially increased the risk of failure to achieve or maintain remission: 71% of depressed patients vs. 30% of non-depressed patients failed to enter remission at 4 weeks, and 100% vs. 73% required retreatment with infliximab within 9 months.

In these studies, the main psychological culprit in CD seems to be depression, whereas stress *per se* seems to have little impact. One large prospective study of a mixed IBD population found no impact of stressful life events on either patient group at a lag time of 1–3 months [25], while Bitton found no direct association between either perceived stress or minor life problems and subsequent disease activity [23].

It is likely that only certain CD patients are vulnerable to psychological factors, just as stress triggers headaches in only a subgroup of migraine sufferers [26].

Studies in Ulcerative Colitis

The evidence of an impact of psychological factors is stronger for patients with UC than for those with CD. The retrospective or case-control literature is somewhat mixed, with some studies reporting an association of recalled life stress with disease activity [19,27] and others not [20,28,29], but studies with a stronger methodology have found more consistent associations. Patients with UC in clinical remission are more likely to show endoscopic signs of activity if they are experiencing high levels of life stress [30], and when patients are enrolled in clinical remission life stress has been found reliably to predict subsequent disease activity [31,32]. This association may be quite strong: if a patient with UC is in the upper tertile for stress, his or her risk of experiencing an exacerbation both in the medium term (6–8 months) and the long term (up to 5 years) is triple that of a similar patient with lower stress levels [31].

Maunder and colleagues examined the impact of various enduring patient characteristics on the stress–activity relationship in UC, documenting that the link between stress and inflammation differs in biological subgroups of patients. Disease activity was significantly correlated with depressive symptoms and with health anxiety in patients who lacked the pANCA antibody, whereas neither correlation was significant in pANCA-positive patients [33]. Patients who had atypical autonomic responses to an acute psychological laboratory stressor (recall of a stressful life event) had a higher level of disease activity 7–37 months later [34]; contrary to expectations, what characterized the high-risk patients was a particularly sluggish rather than a particularly brisk vagal response to stress [34].

Studies of patients with one form of IBD may be relevant to the other form, but there appears to be some specificity: life stress rather than depression is the major exacerbating agent in UC patients. Long-term or cumulative stress may be more important than acute stressful events, which have been reported to have a short-term triggering effect by some [32] but not most investigators [31,35,36]. Several groups have reported, similarly, that anxiety and depression are not followed by flares of UC during the subsequent month or so [31,36,37].

Methodological Issues

It is difficult to perform reliable studies of mind-body interactions in patients with CD. The disease is typically severe and disruptive of daily life, has no known cure,

and complete remission is unusual – even a patient meeting the standard research definition, a Crohn's disease activity index (CDAI) <150, may have diarrhea and abdominal pain. Patients with more aggressive disease are more prone to exacerbations and are also more subject to psychological distress, a confounding effect that makes it difficult to have complete faith in associations detected between distress and subsequent disease course. Furthermore, disease activity in CD patients must be gauged largely on the basis of self-reported symptoms, leaving room for confounding by the sort of recall bias that has been termed "effort after meaning," in which the patient attempts to make sense out of otherwise inexplicable health events [38]. Even a prospective relationship between distress and subsequent disease activity can be confounded by the residual psychological impact of previous disease activity.

Ulcerative colitis is easier to study: disease activity can be determined objectively with proctoscopy, and for most patients periods of disease activity alternate with periods of complete clinical remission. This means subjects can be evaluated during remission and then followed until the disease rears its head again. A comparably convincing study of CD would have to enroll patients immediately following complete surgical resection and follow them over time; such a study has as yet not been performed, although Persoons' study of infliximab-induced remission comes close [24].

Conclusion: Psychological distress, especially depression, can have a deleterious effect on the course of CD.

Personality

Previously, personality factors, especially immaturity, dependency, and obsessional traits, were considered to be important in the precipitation and recurrence of UC [39] and a conclusion that was subsequently extended to CD. This concept has faded, and the current consensus is that while a patient's personality structure may influence his or her adjustment to the disease [40], any personality traits associated with IBD are likely to be caused by the illness [2].

An impact of CD on durable personal traits is plausible. Symptoms often begin during adolescence, forcing patients back into dependent roles during an impressionable period whose defining characteristic is the search for independence, and then further impairing their self-esteem by feelings of being different and by the spectre of fecal incontinence – which, incidentally, can dictate an "obsessional" preoccupation with locating the nearest toilet. In practice, aside from some increase in dependency [41] and obsessiveness [28,42], most studies have found remarkably little difference between the personality profiles of IBD patients and controls [5,43].

Another personality trait sometimes thought to bring a particular vulnerability to psychosomatic phenomena is "alexithymia." Alexithymics appear to be in the grip of strong emotions but cannot recount, or recognize, their own sadness, anger, fear, or tenderness, which therefore possibly leads to chronic activation of the sympathetic nervous system and to the transformation of emotions into bodily signals [44]. Patients with CD, like individuals with other chronic medical conditions, score rela-

tively high on scales of alexithymia [45]. But as with obsessiveness, alexithymic traits may develop in reaction to the illness experience: one method of coping with chronic disease may be to turn one's attention away from potentially overwhelming emotional reactions.

Conclusion: The personality of CD patients is similar to that of the general population, aside from some impact of disease experience.

Psychophysiology of Crohn's Disease: Mechanisms and Mediators

Permeability

Current models of the pathophysiology of IBD assign a key role to enhanced intestinal permeability [46], which facilitates access of luminal dietary and, especially, bacterial antigens to immune effector cells in the bowel wall [47]. Increased permeability has been reported to be a warning of imminent exacerbation in CD [48] and to be a marker of genetic vulnerability in healthy relatives [49]. As discussed in the section on animal models, effects on intestinal permeability constitute a major mechanism by which stress initiates or exacerbates colitis in laboratory animals [8,11].

The impact of psychological factors on jejunal permeability and function has been studied in intubated human subjects. In one study, a psychological stressor (dichotomous listening) significantly reduced mean jejunal net water absorption, changed mean net sodium and chloride absorption to secretion, and slowed transit time [50]. This effect on jejunal water secretion was later replicated using a physical stressor (hand in ice water); pain also increased luminal release of mast cell mediators [51].

In a recent study using a similar methodology, researchers were able to demonstrate that previous life experiences can affect intestinal reactions to an acute laboratory stressor [52]. Healthy women were intubated and the functioning of a 20-cm jejunal segment was monitored during intermittent ice-water immersion of one hand. Intestinal secretion and permeability to albumin increased in the group as a whole, but particularly in women who reported substantial stress in their lives. This differential response occurred despite the absence of any difference between the low- and high-stress groups in heart rate, blood pressure, adrenocorticotrophic hormone, or cortisol.

Using a different model, in-vitro incubation of colonic biopsy specimens from healthy volunteers, mucosal permeability was shown to be increased by exposure to the classic stress mediator, corticotropin releasing factor [53].

Classic Stress Pathways

The hypothalamic-pituitary-adrenal axis seems to be an important mediator of stress-induced permeability increases in animal models [6,54,55], and its potential role in the effects of psychological factors on clinical inflammatory bowel disease is supported by the demonstration of corticotropin releasing factor in patients' colonic tissues [55,56].

One of the effects of corticotropin in the intestinal wall is to stimulate mast cells. Mast cell activation mediates both stress stimulation of bacterial mucosal adherence [57] and colitis [6] in animal models, and activation has been shown to occur in the colonic and jejunal mucosa of humans subjected to laboratory stressors [15,51] or exposed in vitro to corticotropin-releasing hormone [53].

Immune Reactivity

In stress facilitation of experimental animal colitis, the immune system plays a crucial role. Stress-increased gut permeability allows an exaggerated immune response to external immune stimuli [13], and the promotion of relapse by stress after chemically induced colitis is immune-dependent, requiring the participation of sensitized CD4+ lymphocytes. Susceptibility to reactivation by stress could not be produced in animals that had undergone resection of one key immune effector organ, the thymus, and could be adoptively transferred [8].

Immune hyper-reactivity is a fundamental mechanism in the pathophysiology of UC and CD [58], with inadequate suppression of the immune response to bacterial toxins originating inside the gut and with elevation of local and circulating cytokines during clinical activity [59]. For 30 years it has been known that psychological factors can lead to profound deregulation of the immune system, largely through the mediation of autonomic nerves that innervate lymphoid tissue [3]. While the best-known effects on immune function are suppressive, stress and stress mediators also heighten the immune response in certain models, increasing the levels of proinflammatory cytokines in blood and body tissues and stimulating innate immunity [60–64], and thus could intensify the anomalous immune reactivity involved in IBD. Acute psychological stress in the laboratory has in fact been shown to induce intensified systemic and mucosal pro-inflammatory responses in patients with IBD [15,16,65]. Immune stimulation in response to stress could have the evolutionary benefit of improving the organism's defenses against bacterial infection, at the expense of exacerbating inflammatory conditions such as IBD (which are less of a danger, in the wild, than infection) [66].

Patients with chronic inflammatory diseases have anomalous patterns of lymphocyte activation in response to life stress [67]. For example, during exam period, interleukin (IL)-5 was increased in asthmatic students but not in healthy controls [68], and IL-6 levels following a mental calculation stress test were higher in UC patients than in controls [69].

Bacterial Flora

The intestinal flora, abnormal and possibly etiopathogenic in CD [70,71], may be affected by psychological factors. In young monkeys, the stress of separation from the mother decreased the concentration of lactobacilli in the colon, creating an opportunistic environment for pathogenic forms such as *Shigella* and *Campylobacter*

species, with the development of diarrhea in some animals [72]. In vitro, catecholamines can stimulate the growth of *Escherichia coli*, *Yersinia enterocolitica*, and *Pseudomonas aeruginosa*, and can promote the production of Shiga-like toxins by *E. coli* 0157:H7 [73,74]. Stress might therefore influence the composition of luminal bacterial flora in the direction of heightened pathogenicity.

Bacterial adherence to the mucosa, furthermore, which is a prerequisite for the penetration of antigens into the gut wall, is also increased by chronic stress in animals [57].

Autonomic and Psychological Effects on Symptoms

Stress, depression, and anxiety could have nonspecific effects on CD symptoms due to autonomic discharge affecting intestinal motility [75], especially given the frequency of irritable-bowel-syndrome-like phenomena in patients with IBD [76]. Anxious and depressed individuals also have a notoriously high level of attention and concern for bodily phenomena [77] and may therefore report greater symptomatology at a given level of objective pathology.

Behavioral Mediators

The inflammatory process in CD can potentially be affected by smoking and by the consumption of medications such as nonsteroidal antiinflammatory drugs and 5-aminosalicylate derivatives [78–80]. Patient behaviors could therefore mediate the influence of psychosocial factors on CD, since distressed people are likely to smoke more cigarettes, consume more analgesics, and neglect their regimen of prophylactic medication. Many CD patients report that specific foods can precipitate their symptoms [81], possibly reflecting idiosyncratic factors in immune stimulation by various food antigens. Such patients might eat less prudently during periods of stress. More speculatively, the neglect of dental hygiene could modify the gut flora and thereby affect patterns of cytokine secretion [82].

Conclusion: Psychological factors can affect the inflammatory process in CD by inducing immune hyperreactivity, increasing intestinal permeability, and altering the nature and mucosal handling of the gut flora, via mediators including mast cells, the hypothalamic-pituitary-adrenal axis, and behavioral changes; they can also worsen symptoms through effects on motility and subjective perception.

Impact of Crohn's Disease on Psychological State and Quality of Life

Psychological Distress

Physicians and surgeons who treat patients with CD are not always aware of how much the disease disrupts their patients' lives.

Inflammatory bowel disease flares bring understandable psychological distress [5,45,83,84]. In contrast to UC patients, who may be distressed during acute episodes but otherwise enjoy remarkably good psychological health [85], CD patients frequently continue to suffer from depression, anxiety, and even panic disorder when their disease is relatively quiescent [84,86,87]. This difference is related to disease patterns [88]: in UC, symptomatic episodes are often interspersed with periods of complete well-being, and the patient knows that at worst colectomy can provide a cure. CD is incurable, and it is more likely to give unrelenting symptoms – leaving less space for contemplation and distancing – and to require medications, such as corticosteroids, which can have central nervous system side effects.

The psychological impact of CD does seem to decrease with time, as patients learn to live with their disease, and it may be buffered to some extent by social support [89].

Health-Related Quality of Life

The use of specifically developed scales allows comparison of the quality of life of people with various medical conditions. On the Sickness Impact Profile, CD moderately impairs functioning, on a par with angina pectoris but less than rheumatoid arthritis. The impact of UC is considerably less, comparable to that of moderate obesity [90]. All realms are affected: sleep and rest, physical functioning, psychosocial functioning, emotional behavior, and social interaction [90].

Other aspects of disease impact are better elicited by disease-specific questionnaires. Of particular interest for Crohn's disease is the Rating Form for IBD Patient Concerns, which evaluates 25 concerns commonly expressed by patients with IBD regarding disease impact, intimacy, complications, and "stigmata" such as smelling bad [91].

The top concerns of CD patients are fatigue, medication effects, and the uncertain nature of the disease; also important are having an ostomy bag, undergoing surgery, being a burden on others, reaching one's full potential, pain, and – even in countries with no economic barriers to health care – financial problems [91].

In an international comparison, scores varied greatly among countries [92], with patients from southern Europe tending to report greater overall concern than those from northern Europe. The level of concern among Italian patients was second only to that of Portuguese patients, and higher than among patients from the USA, Canada, Israel, Sweden, Austria, or France.

Bringing a patient into prolonged remission is probably the most effective way of improving health-related quality of life; conversely, the outcome of medical or surgical means can be affected by patients' psychological characteristics [24,93].

Conclusion: Crohn's disease often causes psychological distress and disease-related concern, and worsens many aspects of patients' quality of life.

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Psychological Support and Psychotherapy

Approaches and Efficacy

Clinicians should recognize that their CD patients may be experiencing depression, anxiety, or difficulty coping, and should be generous with sympathetic understanding. Sometimes this is not enough, however, and as many as one in three CD patients seek psychological counseling, largely because of the limitations in lifestyle and social interactions imposed by the disease [94]. Physicians might consider suggesting psychologically oriented support to patients who seem to be coping poorly or are excessively dependent on others, depressed, under severe life stress, or lacking a social network [95].

In referring CD patients for counseling, treating physicians should try to choose therapists familiar with the medical aspects of the condition, in part to avoid the risk of inappropriate psychologizing. Psychologists may overestimate the role of psychological factors, especially since the disease involves diarrhea and can be confused with irritable bowel syndrome.

Most trials of psychologically oriented interventions in CD patients have reported encouraging improvements in subjective distress [96–99], in patients' sense of control in coping with their disease [93,100], and in psychosocial functioning [101]. Another study found cognitive-behavioral group therapy to improve depression and anxiety in patients with mixed inflammatory bowel disease [102]. Benefits may be temporary, or patients' distress may resolve spontaneously. In one group of UC patients, an intensive 10-week stress management and self-care intervention improved anxiety, overall psychological distress, and several aspects of health-related quality of life, but by 12 months the advantages over the control group had disappeared [103].

Not all interventions have been successful. In an ambitious multicentric German study, psychodynamic psychotherapy and relaxation training were offered to unselected patients with CD for 1 year. This approach was surprisingly ineffective at improving depression, anxiety, or health-related quality of life, suggesting problems with delivery, choice of intervention, indiscriminate patient selection, or therapist expertise [104]. A study of supportive-expressive group psychotherapy, provided by experienced therapists, similarly failed to help mixed IBD patients suffering from depression or anxiety [105].

Sometimes what will best meet the needs of a distressed patient is not a psychotherapist but a patient support group (such as the Crohn's and Colitis Foundation of America or the Associazione per le Malattie Infiammatorie Croniche dell'Intestino). Self-management and education programs have also proved popular with patients and may decrease health care utilization, though they have not been documented to affect depression, anxiety, or health-related quality of life [106,107]; summer camps for children with IBD have similarly shown promise [108].

Conclusion: Patient support groups or cognitive-behavioral therapy may be particularly helpful for patients with problems coping with their CD; for patients whose disease is the trigger for bringing out deeper conflicts, in-depth psychotherapy may be indicated.

Practical Experience

The aims of psychotherapy for CD patients are psychological integration of a chronic disease, with the limitations it puts on physical functioning and self-image; acceptance of long-term treatment including medications; balancing relations with emotions; finding a personal path that may include the courage to attempt things previously avoided because of illness; letting go of resentment and dependency; and redefining life goals. Patient age, disease duration, and symptom severity do not, in our experience, hinder the therapeutic path, but mental rigidity, isolation, and lack of basic trust can constitute important obstacles.

These challenges in the psychotherapy of patients with IBD have been conceptualized by Robert Maunder as the "adaptive triad" [93]. In his words, "First, illness brings uncertainty. This may be decreased by information gathering, but residual uncertainty remains, in IBD, regarding prognosis and consequences. Second, illness brings major change, especially in the form of loss. Irreducible uncertainty and loss may lead to distress. The final adaptive challenge is the enduring suffering that results from symptoms, uncertainty, distress, and loss. Psychotherapy may be valuable at each of these stages, but the form of psychotherapy varies with the specific adaptive challenge at hand — from information gathering, through facilitated mourning, to more intensive psychodynamic or existential interventions" [109].

Our gastroenterology unit considers a psychologist's services an integral part of the treatment of CD. All newly diagnosed patients are seen during their first hospitalization, and continuing outpatient sessions are offered to those who desire them after discharge. This approach is particularly valuable with young patients, who characteristically distance themselves psychologically from their diagnosis and from the need for testing and treatment; they tend to delegate communication with the physicians to their families, and therapeutic decisions passively to the medical staff. Patients will often express to the psychologist their fantasy that, "If I don't think about the disease, it doesn't exist." Allowing these thoughts to be expressed and elaborated helps the patient to handle doubts and fears more successfully and facilitates a therapeutic alliance with the treating physicians, ensuring the patient will not be lost to follow-up or put up passive resistance to treatment.

Patients generally also welcome the chance to talk with a psychologist during exacerbations, especially during the first years of their disease. They say they find the sessions to be a space where they can express feelings of mistrust and anger, fears that "nothing will ever change," and ambivalent hope that surgery will provide them with a definitive solution. This paves the way for their taking charge, of being capable of encountering their physicians – who they otherwise tend to see either as saviors or as persecutors – as aware, responsible adults.

Patients about to face intestinal resection and temporary or permanent ostomies are also offered the opportunity to talk with a psychologist, in collaboration with the hospital's surgery unit. These sessions often occur in more or less emergency circumstances, when their content is strongly conditioned by a context of particularly aggressive disease with heavy repercussions on the patient's quality of life. In this setting, surgery is inevitably viewed as liberation. In emergencies, there may not be

enough time for the issues to be fully grasped before the surgery takes place, with fears related to self-image and self-esteem being expressed preoperatively but fully addressed only later.

For the new ostomy patient, anguish arises on the one hand from demolition of a part of their body, even if only an invisible intestinal segment, and on the other hand from viewing the ostomy bag as a shameful external extension of something that belongs inside. The body surface that divides the outer world from the inner world is literally imperiled – a metaphor, one might add, for all sorts of other psychological fragilities.

Often during a postoperative session the patient will lift his or her clothes to show the psychologist the ostomy and the bag, all the while intently observing the psychologist, who understands the patient's need for assurance that this object of shame can be seen, and can be accepted, in another's gaze.

Conclusion: Professional psychological support should be offered to all CD patients who have been newly diagnosed, who have high levels of psychological distress, or who are facing an ostomy.

Potential to Influence Disease Course

Psychological support in CD aims to alleviate distress and increase the patient's ability to cope. But some workers in the field have articulated a more ambitious goal: to improve the course of the disease. A capacity of psychosocial manipulations to influence the disease is not entirely implausible, since psychological interventions have striking benefits in patients with irritable bowel syndrome [110,111] and may even be able to improve the prognosis of breast cancer [112].

A laboratory study of hypnosis provides suggestive evidence: when systemic and rectal mucosal parameters were examined in 17 UC patients before and after a single 50-min session of trance with anti-inflammatory suggestion imagery, hypnosis reduced the median serum IL-6 concentration by 53%, and decreased several components of the mucosal inflammatory process (release of SP, histamine, and IL-13) as well as mucosal blood flow [113].

The published literature on psychologically oriented interventions in CD gives modest support to an effect on disease course [95]. In the multicentric German trial, psychotherapy patients had fewer hospital days and sick leave during the following year [114] but the chief measures of somatic course were unaffected [115]. Another study suggested that stress management can improve fatigue and abdominal pain at 6- and 12-month follow-ups [116]. Milne and colleagues compared a combined approach of patient education, stress management, and relaxation training with treatment as usual and found a hint of improvement in the CDAI [96]. "Hard" measures of outcome have not proven substantially different in CD patients than in controls in any trials of CD [99,115] or UC [103].

Shortcomings in these studies leave the question open. Some interventions have been relatively unsuccessful even in improving the patients' psychological state [104], making it improbable that they could influence disease activity for the better.

Other studies have included among their subjects patients with low psychological distress levels and/or long-quiescent disease, for whom psychologically oriented interventions are unlikely to give a detectable benefit and could even, in fact, be considered a waste of time. The psychoanalytic therapies used in the German study might be less appropriate for unselected medical patients than approaches more oriented to coping in the here and now.

Conclusion: While psychological therapies have not been shown reliably to improve the course of CD, carefully chosen interventions focused on vulnerable patient subgroups with high levels of stress or psychiatric symptomatology might be more successful.

Conclusions

Crohn's disease often has a serious and sometimes devastating effect on patients' personal, work, and social lives and their ability to function. The consequent anxiety and depression can, in turn, affect a CD patient's drive and ability to care for him/herself. There is also evidence, supported by animal models, for a contribution of psychological factors, especially depressive symptoms, to disease course. All CD patients should be offered access to professional psychological support, ideally beginning at the time of first diagnosis, primarily to relieve distress but also with the hope of breaking a potentially vicious cycle of exacerbation/distress/exacerbation.

Recognition of the psychological factors in CD can have a number of positive spinoffs, such as encouraging medical personnel to listen sympathetically to their patients and convincing distressed patients of the potential value of non-pharmacological therapeutic approaches. But there is also a danger of negative repercussions. In particular, overemphasis on psychological factors can lead to attribution of all disease swings to "stress" and stigmatization of the patient as "psychosomatic." One physician-patient said, "When I dared expose myself to close friends and relate distress over worsening symptoms, I felt vulnerable to statements like, 'Well, it does sound like your job has been stressful lately' " [117]. The way to ensure an appropriate balance between two excessive poles, biomedical reductionism and blaming the victim, is to embrace what has been called the biopsychosocial model and accept, rather than flee from, the complexity of multidirectional interactions among symptoms, pathophysiology, and psychological, social, immunological, behavioral, and genetic factors [118].

References

1. Wilbert–Lampen U, Leistner D, Greven S et al (2008) Cardiovascular events during World Cup soccer. N Engl J Med 358(5):475–483
2. Aronowitz R, Spiro HM (1988) The rise and fall of the psychosomatic hypothesis in ulcerative colitis. J Clin Gastroenterol 10(3):298–305

3. Ader R, Felten DL, Cohen N (eds) (2006) Psychoneuroimmunology. 4th edn. Academic, San Diego
4. Mitchell CM, Drossman DA (1987) Survey of the AGA membership relating to patients with functional gastrointestinal disorders. Gastroenterol 92:1282–1284
5. Robertson DAF, Ray J, Diamond I, Edwards JG (1989) Personality profile and affective state of patients with inflammatory bowel disease. Gut 30:623–626
6. Wilson LM, Baldwin AL (1999) Environmental stress causes mast cell degranulation, endothelial and epithelial changes, and edema in the rat intestinal mucosa. Microcirculation 6(3):189–198
7. Collins SM, McHugh K, Jacobson K et al (1996) Previous inflammation alters the response of the rat colon to stress. Gastroenterol 111(6):1509–1515
8. Qiu BS, Vallance BA, Blennerhassett PA, Collins SM (1999) The role of CD4+ lymphocytes in the susceptibility of mice to stress–induced reactivation of experimental colitis. Nat Med 5:1178–1182
9. Gareau MG, Silva MA, Perdue MH (2008) Pathophysiological mechanisms of stress–induced intestinal damage. Current Mol Med 8:274–281
10. Velin AK, Ericson AC, Braaf Y et al (2004) Increased antigen and bacterial uptake in follicle associated epithelium induced by chronic psychological stress in rats. Gut 53(4):494–500
11. Soderholm JD, Perdue MH (2001) Stress and the gastrointestinal tract II. Stress and intestinal barrier function. Am J Physiol Gastrointest Liver Physiol 280(1):G7–G13
12. Saunders PR, Hanssen NP, Perdue MH (1997) Cholinergic nerves mediate stress-induced intestinal transport abnormalities in Wistar–Kyoto rats. Am J Physiol 273(2 Pt 1):G486–G490
13. Barreau F, Ferrier L, Fioramonti J, Bueno L (2004) Neonatal maternal deprivation triggers long term alterations in colonic epithelial barrier and mucosal immunity in rats. Gut 53:501–506
14. Varghese AK, Verdú EF, Bercik P et al (2006) Antidepressants attenuate increased susceptibility to colitis in a murine model of depression. Gastroenterol May 130(6):1743–1753
15. Farhadi A, Keshavarzian A, Van de Kar LD et al (2005) Heightened responses to stressors in patients with inflammatory bowel disease. Am J Gastroenterol August 100(8):1796–1804
16. Mawdsley J, Macey M, Feakins R et al (2006) The efect of acute psychological stress on systemic and rectal mucosal measures of inflammation in ulcerative colitis. Gastroenterol 131:410–419
17. Li J, Nørgrd B, Precht DH, Olsen J (2004) Psychological stress and inflammatory bowel disease: a follow–up study in parents who lost a child in Denmark. Am J Gastroenterol 99(6):1129–33
18. Lerebours E, Gower–Rousseau C, Merle V et al (2007) Stressful life events as a risk factor for inflammatory bowel disease onset: a population-based case-control study. Am J Gastroenterol 102(1):122–131
19. von Wietersheim J, Kohler T, Feiereis H (1992) Relapse-precipitating life events and feelings in patients with inflammatory bowel disease. Psychother Psychosom 58:103–112
20. Paar GH, Bezzenberger U, Lorenz–Meyer H (1988) Über den Zusammenhang von psychosozialem Streb und Krankheitsactivität bei Patienten mit Morbus Crohn und Colitis ulcerosa. Z Gastroenterologie 26:648–657
21. Mittermaier C, Dejaco C, Waldhoer T et al (2004) Impact of depressive mood on relapse in patients with inflammatory bowel disease: a prospective 18–month follow–up study. Psychosom Med Jan–Feb 66(1):79–84
22. Mardini HE, Kip KE, Wilson JW (2004) Crohn's disease: a two–year prospective study of the association between psychological distress and disease activity. Dig Dis Sci Mar 49(3):492–497
23. Bitton A, Dobkin PL, Edwardes MD et al (2008) Predicting relapse in Crohn's disease: a biopsychosocial model. Gut 57:1386–1392
24. Persoons P, Vermeire S, Demyttenaere K et al (2005) The impact of major depressive disorder on the short- and long-term outcome of Crohn's disease treatment with infliximab. Ali-

ment Pharmacol Ther Jul 22(2):101–110
25. Duffy LC, Zielezny MA, Marshall JR et al (1991) Lag time between stress events and risk of recurrent episodes of inflammatory bowel disease. Epidemiology 2(2):141–145
26. Vandenbergh V, Amery WK, Waelkens J (1987) Trigger factors in migraine: a study conducted by the Belgian Migraine Society. Headache 27:191–6
27. Fava GA, Pavan L (1976) Large bowel disorders II, Psychopathology and alexithymia. Psychother Psychosom 27:100–105
28. Helzer JE, Stillings WA, Chammas S et al (1982) A controlled study of the association between ulcerative colitis and psychiatric diagnosis. Dig Dis Sci 27(6):513–518
29. Riley SA, Mani V, Goodman MJ, Lucas S (1990) Why do patients with ulcerative colitis relapse? Gut 31:179–183
30. Levenstein S, Prantera C, Varvo V et al (1994) Psychological stress and disease activity in ulcerative colitis: a multidimensional cross-sectional study. Am J Gastroenterol 89:1219–1225
31. Levenstein S, Prantera C, Varvo V et al (2000) Stress and exacerbation in ulcerative colitis: a prospective study of patients enrolled in remission. Am J Gastroenterol 95(5):1213–1220
32. Bitton A, Sewitch M, Peppercorn MA et al (2003) Psychosocial determinants of relapse in ulcerative colitis: A longitudinal study. Am J Gastroenterol 98:2203–2208
33. Maunder R, Greenberg G, Hunter J et al (2006) Psychobiological subtypes of ulcerative colitis: pANCA status moderates the relationship between disease activity and psychological distress. Am J Gastroenterol 101:2546–2551
34. Maunder RG, Greenberg GR, Nolan RP et al (2006) Autonomic response to standardized stress predicts subsequent disease activity in ulcerative colitis. European Journal of Gastroenterology and Hepatology 18:413–420
35. Vidal A, Gomez–Gil E, Sans M et al (2006) Life events and inflammatory bowel disease relapse: a prospective study of patients enrolled in remission. Am J Gastroenterol 101:775–781
36. North CS, Alpers DH, Helzer JE et al (1991) Do life events or depression exacerbate inflammatory bowel disease? A prospective study. Ann Int Med 114:381–386
37. Greene BR, Blanchard EB, Wan CK (1994) Long-term monitoring of psychosocial stress and symptomatology in inflammatory bowel disease. Behav Res Ther 32(2):217–226
38. Brown GW, Harris TO (1978) Social origins of depression: a study of psychiatric disorder in women. Tavistock, London
39. Daniels GE (1944) Nonspecific ulcerative colitis as a psychosomatic disease. Med Clin N A. 28:593–602
40. Gazzard BG, Price HL, Libby GW, Dawson AM (1978) The social toll of Crohn's disease. Br Med J. 2:1117–1119
41. Arapakis G, Lyketsos CG, Gerolymatos K et al (1986) Low dominance and high intropunitiveness in ulcerative colitis and irritable bowel syndrome. Psychother Psychosom 46:171–176
42. Bellini M, Tansella M (1976) Obsessional scores and subjective general psychiatric complaints of patients with duodenal ulcer or ulcerative colitis. Psychol Med 6:461–467
43. Helzer JE, Chammas S, Norland CC et al (1984) A study of the association between Crohn's disease and psychiatric illness. Gastroenterol 86:324–330
44. Taylor G, Bagby R, Parker J (1997) Disorders of affect regulation: alexithymia in medical and psychiatric illness. Cambridge University Press, Cambridge
45. Porcelli P, Leoci C, Guerra V et al (1996) A longitudinal study of alexithymia and psychological distress in inflammatory bowel disease. J Psychosom Res 41(6):569–573
46. Jorgensen J, Ranlov PJ, Bjerrum PJ et al (2001) Is an increased intestinal permeability a valid predictor of relapse in Crohn disease? Scand J Gastroenterol Suppl 36(5):521–527
47. Sartor RB (1997) Pathogenesis and immune mechanisms of chronic inflammatory bowel diseases. Am J Gastroenterol 92(12 Suppl):5S–11S
48. Arnott I, Kingstone K, Ghosh S (2000) Abnormal intestinal permeability predicts relapse in inactive Crohn disease. Scand J Gastroenterol Nov 35(4):1163–1169

49. Hollander D, Vadheim CM, Brettholz E et al (1986) Increased intestinal permeability in patients with Crohn's disease and their relatives: a possible etiologic factor. Ann Int Med 105:883–885
50. Barclay G, Turnberg L (1987) Effect of psychological stress on salt and water transport in the human jejunum. Gastroenterol Jul 93(1):91–97
51. Santos J, Saperas E, Nogueiras C et al (1998) Release of mast cell mediators into the jejunum by cold pain stress in humans. Gastroenterol 114(4):640–648
52. Alonso C, Guilarte M, Vicario M et al (2008) Maladaptive intestinal epithelial responses to life stress may predispose healthy women to gut mucosal inflammation. Gastroenterol 135:163–172
53. Wallon C, Yang P–C, Keita AV et al (2008) Corticotropin–releasing hormone (CRH) regulates macromolecular permeability via mast cells in normal human colonic biopsies in vitro. Gut 57:50–58
54. Meddings JB, Swain MG (2000) Environmental stress-induced gastrointestinal permeability is mediated by endogenous glucocorticoids in the rat. Gastroenterol 119:1019–1028
55. Mikocka–Walus AA, Turnbull DA, Andrews JM et al (2008) The effect of functional gastrointestinal disorders on psychological comorbidity and quality of life in patients with inflammatory bowel disease. Aliment Pharmacol Ther 28:475–483
56. Kawahito Y, Sano H, Mukai S et al (1995) Corticotropin releasing hormone in colonic mucosa in patients with ulcerative colitis. Gut 37(4):544–551
57. Soderholm J, Yang P, Ceponis P et al (2002) Chronic stress induces mast cell–dependent bacterial adherence and initiates mucosal inflammation in rat intestine. Gastroenterol 123(4):1099–1108
58. Sartor R (1995) Current concepts of the etiology and pathogenesis of ulcerative colitis and Crohn's disease. Gastroenterol Clin North Am 24(3):475–507
59. Ligumsky M, Simon PL, Karmeli F, Rachmilewitz D (1990) Role of interleukin 1 in inflammatory bowel disease-enhanced production during active disease. Gut 31:686–689
60. Dhabhar FS, McEwen BS (1996) Stress-induced enhancement of antigen-specific cell-mediated immunity. J Immunol 156(7):2608–2615
61. Sternberg EM, Hill JM, Chrousos GP et al (1989) Inflammatory mediator-induced hypothalamic–pituitary–adrenal axis activation is defective in streptococcal cell wall arthritis- susceptible Lewis rats. Proc Natl Acad Sci USA 86(7):2374–2378
62. Ackerman KD, Martino M, Heyman R et al (1998) Stressor-induced alteration of cytokine production in multiple sclerosis patients and controls. Psychosom Med 60(4):484–491
63. Zhou D, Kusnecov AW, Shurin MR et al (1993) Exposure to physical and psychological stressors elevates plasma interleukin 6: relationship to the activation of hypothalamic-pituitary-adrenal axis, Interleukins, signal transduction, and the immune system-mediated stress response. Endocrinology 133(6):2523–2530
64. Lyte M, Nelson SG, Thompson ML (1990) Innate and adaptive immune responses in a social conflict paradigm. Clin Immunol Immunopathol 57(1):137–147
65. Milunsky A (1980) Prenatal detection of neural tube defects, VI. Experience with 20,000 pregnancies. JAMA 244(24):2731–2735
66. Sternberg EM, Chrousos GP, Wilder RL, Gold PW (1992) The stress response and the regulation of inflammatory disease. Ann Int Med 117(10):854–666
67. Geenen R, Godaert GL, Heijnen CJ, Vianen ME et al (1998) Experimentally induced stress in rheumatoid arthritis of recent onset: effects on peripheral blood lymphocytes. Clin Exp Rheumatol 16(5):553–559
68. Kang D, Coe C, McCarthy D et al (1997) Cytokine profiles of stimulated blood lymphocytes in asthmatic and healthy adolescents across the school year. J Interferon Cytokine Res 17(8):481–487
69. Kuroki T, Ohta A, Aoki Y et al (2007) Stress maladjustment in the pathoetiology of ulcerative colitis. J Gastroenterol Jul 42(7):522–527

70. Sartor RB (1997) Enteric microflora in IBD: Pathogens or commensals? Inflamm Bowel Dis 3(3):230–235
71. Frank D, St Amand A, Feldman R et al (2007) Molecular-phylogenetic characterization of microbial community imbalances in human inflammatory bowel diseases. Proc Natl Acad Sci USA 104(34):13780–13785
72. Bailey MT, Coe CL (1999) Maternal separation disrupts the integrity of the intestinal microflora in infant rhesus monkeys. Dev Psychobiol 35(2):146–155
73. Lyte M, Ernst S (1992) Catecholamine induced growth of gram negative bacteria. Life Sci 50(3):203–212
74. Lyte M, Arulanandam BP, Frank CD (1996) Production of Shiga-like toxins by Escherichia coli O157:H7 can be influenced by the neuroendocrine hormone norepinephrine. J Lab Clin Med 128(4):392–398
75. Prugh DG (1951) The influence of emotional factors on the clinical course of ulcerative colitis in children. Gastroenterol 18(3):339–354
76. Bayless TM, Harris ML (1990) Inflammatory bowel disease and irritable bowel syndrome. Med Clin N A 74(1):21–9
77. Barsky AJ (1992) Amplification, somatization, and the somatoform disorders. Psychosomatics 33(1):28–34
78. Tysk C, Lindberg E, Jarnerot G, Floderus–Myrhed B (1988) Ulcerative colitis and Crohn's disease in an unselected population of monozygotic and dizygotic twins. A study of heritability and the influence of smoking. Gut 29:990–996
79. Felder J, Korelitz B, Rajapakse R et al (2000) Effects of nonsteroidal antiinflammatory drugs on inflammatory bowel disease: a case-control study. Am J Gastroenterol 95(8):1949–1954
80. Nigro G, Angelini G, Grosso S et al (2001) Psychiatric predictors of noncompliance in inflammatory bowel disease: psychiatry and compliance. J Clin Gastroenterol 32(1):66–68
81. Levenstein S, Prantera C, Luzi C, D'Ubaldi A (1985) Low residue or normal diet in Crohn's disease: a prospective controlled study in Italian patients. Gut 26:989–993
82. Deinzer R, Forster P, Fuck L et al (1999) Increase of crevicular interleukin 1beta under academic stress at experimental gingivitis sites and at sites of perfect oral hygiene. J Clin Periodontol 26(1):1–8
83. Tarter RE, Switala J, Carra J et al (1987) Inflammatory bowel disease: psychiatric status of patients before and after disease onset. Int J Psychiatry Med 17(2):173–181
84. Andrews H, Barczak P, Allan RN (1987) Psychiatric illness in patients with inflammatory bowel disease. Gut 28:1600–1604
85. Esler MD, Goulston KJ (1973) Levels of anxiety in colonic disorders. N Engl J Med 288(1):16–20
86. Simren M, Axelsson J, Gillberg R et al (2002) Quality of life in inflammatory bowel disease in remission: the impact of IBS–like symptoms and associated psychological factors. Am J Gastroenterol 97(2):389–396
87. Guthrie E, Jackson J, Shaffer J et al (2002) Psychological disorder and severity of inflammatory bowel disease predict health–related quality of life in ulcerative colitis and Crohn's disease. Am J Gastroenterol 97(8):1994–1999
88. Drossman DA, Leserman J, Mitchell CM et al (1991) Health status and health care use in persons with inflammatory bowel disease: a national sample. Dig Dis Sci 36:1746–1755
89. Sewitch M, Dobkin P, Abrahamowicz M et al (2001) Psychological distress, social support and disease activity in inflammatory bowel disease. Am J Gastroenterol 96(5):1470–1479
90. Drossman DA, Patrick DL, Mitchell CM et al (1989) Health–related quality of life in inflammatory bowel disease: functional status and patient worries and concerns. Dig Dis Sci 34(9):1379–1386
91. Drossman DA, Leserman J, Li Z et al (1991) The rating form of IBD patient concerns: a new measure of health status. Psychosom Med 13(6):701–712

92. Levenstein S, Li Z, Almer S et al (2001) Cross-cultural variation in disease–related concerns among patients with inflammatory bowel disease. Am J Gastroenterol 96(6):1822–1830
93. Maunder R, Esplen MJ (1999) Facilitating adjustment to inflammatory bowel disease: a model of psychosocial intervention in non-psychiatric patients. Psychother Psychosom 68(5):230–240
94. Miehsler W, Weichselberger M, Offerlbauer–Ernst A et al (2008) Which patients with IBD need psychological interventions? A controlled study. Inflamm Bowel Dis 14:1273–1280
95. von Wietersheim J, Kessler H (2006) Psychotherapy with chronic inflammatory bowel disease patients: a review. Inflamm Bowel Dis 12(12):1175–1184
96. Milne B, Joachim G, Niedhardt J (1986) A stress management programme for inflammatory bowel disease patients. J Adv Nurs 11(5):561–567
97. Pickering TG, Schnall PL, Schwartz JE, Pieper CF (1991) Can behavioural factors produce a sustained elevation of blood pressure? Some observations and a hypothesis. J Hypertens Suppl 9(8):S66–S68
98. Mussell M, Bocker U, Nagel N et al (2003) Reducing psychological distress in patients with inflammatory bowel disease by cognitive–behavioural treatment: exploratory study of effectiveness. Scand J Gastroenterol 38(7):755–762
99. Smith GD, Watson R, Roger D et al (2002) Impact of a nurse–led counselling service on quality of life in patients with inflammatory bowel disease. J Adv Nurs 38(2):152–160
100. Schwarz SP, Blanchard EB (1991) Evaluation of a psychological treatment for inflammatory bowel disease. Behav Res Ther 29(2):167–177
101. Gold N, Issenman R, Roberts J, Watt S (2000) Well-adjusted children: an alternate view of children with inflammatory bowel disease and functional gastrointestinal complaints. Inflamm Bowel Dis 6(1):1–7
102. Díaz Sibaja M, Comeche Moreno M, Mas Hesse B (2007) Protocolized cognitive–behavioural group therapy for inflammatory bowel disease. Rev Esp Enferm Dig 99(10):593–598
103. Langhorst J, Mueller T, Luedtke R et al (2007) Effects of a comprehensive lifestyle modification program on quality-of-life in patients with ulcerative colitis: a twelve-month follow-up. Scand J Gastroenterol 42(6):734–745
104. Keller W, Pritsch M, Von Wietersheim J et al (2004) Effect of psychotherapy and relaxation on the psychosocial and somatic course of Crohn's disease: main results of the German Prospective Multicenter Psychotherapy Treatment study on Crohn's Disease. J Psychosom Res 56(6):687–696
105. Maunder R, Esplen M (2001) Supportive-expressive group psychotherapy for persons with inflammatory bowel disease. Can J Psychiatry 46(7):622–626
106. Kennedy AP, Nelson E, Reeves D et al (2004) A randomised controlled trial to assess the effectiveness and cost of a patient orientated self management approach to chronic inflammatory bowel disease. Gut 53:1639–1645
107. Waters B, Jensen L, Fedorak R (2005) Effects of formal education for patients with inflammatory bowel disease: a randomized controlled trial. Can J Gastroenterol 19(4):235–244
108. Shepanski M, Hurd L, Culton K et al (2005) Health–related quality of life improves in children and adolescents with inflammatory bowel disease after attending a camp sponsored by the Crohn's and Colitis Foundation of America. Inflamm Bowel Dis 11(2):164–170
109. Levenstein S, Maunder R, Coe C (2003) Psychological factors in organic gastrointestinal disease. I. Inflammatory bowel disease. Available at: http://www.psychosomatic.org/ed_res/ed_res_IBD_powerpoint.htm
110. Guthrie E, Creed F, Dawson D, Tomenson B (1991) A controlled trial of psychological treatment for the irritable bowel syndrome. Gastroenterol 100(2):450–457
111. Payne A, Blanchard EB (1995) A controlled comparison of cognitive therapy and self-help support groups in the treatment of irritable bowel syndrome. J Cons Clin Psych 63:779–786
112. Spiegel D, Kraemer DC, Bloom JR, Gottheil E (1989) Effect of psychosocial treatment on

survival of patients with metastatic breast cancer. Lancet 14:888–891
113. Mawdsley JE, Jenkins DG, Macey MG et al (2008) The effect of hypnosis on systemic and rectal mucosal measures of inflammation in ulcerative colitis. Am J Gastroenterol 103:1460–1469
114. Deter HC, Keller W, von Wietersheim J et al (2007) Psychological treatment may reduce the need for healthcare in patients with Crohn's disease. Inflamm Bowel Dis 13(6):745–752
115. Jantschek G, Zeitz M, Pritsch M et al (1998) Effect of psychotherapy on the course of Crohn's disease. Results of the German prospective multicenter psychotherapy treatment study on Crohn's disease. German Study Group on Psychosocial Intervention in Crohn's Disease. Scand J Gastroenterol 33(12):1289–1296
116. Garcia–Vega E, Fernandez–Rodriguez C (2004) A stress management programme for Crohn's disease. Behav Res Ther 42(4):367–383
117. Spiro HM (1990) Six physicians with inflammatory bowel disease. J Clin Gastroenterol 12(6):636–642
118. Engel GL (1977) The need for a new medical model: a challenge for biomedicine. Science. 196(4286):129–136

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