Interaction of Genetic, Environmental and Immune Factors in the Pathogenesis of Inflammatory Bowel Diseases

Yehuda Chowers MD

Department of Gastroenterology, Sheba Medical Center, Tel Hashomer, Israel
Affiliated to Sackler Faculty of Medicine, Tel Aviv University, Ramat Aviv, Israel

Key words: irritable bowel syndrome, ulcerative disease, Crohn's disease, genetic factors, environment, interleukin

IMAJ 2002;4:815–817

Ulcerative colitis and Crohn's disease are heterogeneous chronic inflammatory disorders of the intestine. Despite intensive research, the etiology of UC and CD is still illusive. However, the clinical and epidemiologic characteristics, as well as experimental studies in humans and animal models have allowed for the development of a paradigm of factors that might be involved in the development of chronic intestinal inflammation. For example, a number of animal models were described in which genes involved in the regulation of the immune response were knocked out. These included genes of either pro-inflammatory cytokines such as interleukin-2 [1] or an anti-inflammatory cytokine such as IL-10 [2] [for review see ref. 3]. Regardless of the specific gene that was knocked out, the intestinal inflammation developed only in animals whose intestine was populated with the normal indigenous mouse flora. No inflammation developed in germ-free animals [3]. These observations suggest that an uncontrolled immune response towards the normal gut flora may play a role in intestinal inflammation in these animal models. A number of observations support the role of intestinal bacteria in the pathogenesis of CD as well. These include the efficacy of antibiotic treatment in CD [4], the beneficial clinical response following surgical fecal diversion [5,6], the presence of cellular immune response against resident flora [7], and the induction of mucosal lesions following exposure of healed mucosa to fecal extracts [8,9]. Based on these different studies, it is hypothesized that chronic intestinal inflammation may develop as the result of a combination of genetic factors that predispose carriers to environmental factors, particularly intestinal bacteria, which elicit an aggressive inflammatory response that causes the tissue damage.

In this issue of *IMAJ*, Karban et al. [10] provide a comprehensive review of genetic components that are thought to be involved in inflammatory bowel disease. The authors reviewed the epidemiologic studies that suggest a significant role for the genetic background as a factor in the etiology of IBD, as well as studies related to specific genetic loci that are linked to either UC or CD. Special emphasis was placed on the recent finding of an association between mutations within the NOD2/CARD15 locus and CD. This finding is of particular interest since it may provide a long-sort linkage between a specific genetic background and bacterial factors that contribute to the pathogenesis of CD. The NOD2/CARD15 protein is expressed selectively in monocytes and belongs to a family of proteins that are homologous to the mammalian apoptotic activating factor 1 and the *C. elegans* Ced-4 proteins. These proteins share structural homology with each other and are regulators of cell apoptosis that may be induced by external stimuli such as stress [11]. The NOD proteins are composed of an N-terminal domain, which is responsible for downstream activation of signal transduction pathways, a central nucleotide-binding oligomerization domain (NOD), and a C-terminal domain that contains a leucine-rich repeat [12]. This part of the NOD2/CARD15 protein binds lipopolysaccharide, an event that leads to the formation of a dimer and subsequent activation of the nuclear factor kappa-B pathway. The activation of NFKB is a central cellular event for inducing the transcription and secretion of multiple pro-inflammatory cytokines that are thought to mediate the tissue damage in CD. Importantly, the mutations within the NOD2/CARD15 protein were described within the leucine rich repeat, i.e., the LPS-binding domain of the protein [13,14]. Indeed, when functional studies were performed, COS cells transfected with mutated NOD2/CARD15 constructs failed to activate NFKB in response to LPS as compared to cells that were transfected with control wild-type NOD2/CARD15 protein [14]. However, the current paradigm suggests that CD patients have an increased immune response to intestinal bacteria. How could these findings be reconciled with each other? One possibility is that the lack of an efficient innate immune response by the monocytes that express a mutated NOD2/CARD15 protein towards intestinal bacteria may stimulate an exaggerated adapted immune response by intestinal T cells. Alternatively, the mutation may result in gain of function that can potentially induce inflammation despite the inability of the protein to bind LPS [14]. Finally, the NOD2/CARD15 protein may perform a yet undefined cellular function whose absence leads to a defect in the control of the inflammatory process. Further studies are needed to explain this apparent paradox.

UC = ulcerative colitis
CD = Crohn's disease
IL = interleukin

IBD = inflammatory bowel disease
NFKB = nuclear factor kappa-B
LPS = lipopolysaccharide

Despite the potential association between NOD2/CARD15 and the pathogenesis of CD, the puzzle is far from complete. As the authors noted, both in the two initial studies in which the mutations were initially described [13,14] and in a subsequent study that evaluated the frequency of the frameshift mutation 3020insC [15], it is clear that the mutations within the NOD2/CARD15 gene do not account for the entire genetic predisposition for CD. The relative low frequency of the mutations in the patient population and the calculated low penetrance of the gene suggest that other genetic loci are involved as well. Furthermore, the NOD2/CARD15 mutation may be of different importance in different patient populations. For example, conflicting results were obtained in other studies in which the frequency of the mutation was correlated with different phenotypes of CD. Thus, whereas one group of investigators found no correlation between the presence of mutation within the NOD2/CARD15 gene and specific disease phenotypes [16], two other groups noted a specific association between such phenotypes and mutations within the NOD2/CARD15 locus. Both in a British cohort and in a European cohort, mutations within the NOD2/CARD15 locus were associated with ileal but not colonic disease [17,18]. Furthermore, in the UK study, patients with frameshift mutations or compound heterozygotes/homozygotes presented at an earlier age [17]. Taken together, additional functional and epidemiologic studies are needed to evaluate the role of NOD2/CARD15 in the pathogenesis of CD in different ethnic groups and in patient populations with different disease characteristics.

The association between NOD2/CARD15 proteins and CD was demonstrated using both the candidate gene approach and genome-wide scanning. Although the association between bacteria and the genetic factor received substantial attention due to the aforementioned findings, other environmental factors were noted and are unexplained at this point. Such factors include, among others, the protective effect of smoking [19] and appendectomy [20] in UC, and the potential association of oral contraceptives with the development of IBD [21]. Results of therapeutic trials may provide mechanistic insight into disease pathogenesis as well. Examples from such studies are the beneficial effect of nicotine in UC [22] and probiotic treatment in pouchitis [23]. These observations may serve as the starting point for research of additional genetic loci based on the target gene approach and hypothetical pathogenic mechanisms by which these specific factors can be linked to inflammation or its down-regulation.

The final damage to the intestine is caused by the immune response, which is hypothesized to result from the combination of genetic and environmental factors. Although both CD and UC result from chronic uncontrolled inflammation, they differ in other aspects of the disease, such as the nature of the inflammatory process, the distribution of disease, and the associated disease complications. For example, the inflammation in Crohn's disease is thought to be mediated mainly by Th1 type cytokines [24], whereas the Th2 type cytokine IL-5 is found in higher levels in ulcerative colitis [25]. In addition, therapeutic trials using anti-tumor necrosis factor antibodies to treat CD patients suggest that other pro-inflammatory cytokines might play an important role in intestinal inflammation. However, not all patients respond to such treatments [26], suggesting that the secretion of these cytokines may be regulated differently in different patient subpopulations. Such studies, which are based on specific immunotherapy and the fine definition of the immune response, constitute the basis for additional research that can link these observations to the different IBD patient populations.

In conclusion, in light of the lack of knowledge regarding the precise etiology of IBD, future research will be aimed at finding the link between the environmental, immunologic and genetic arms that appear to interweave in their pathogenesis.

References

1. Sadlack B, Merz H, Schorle H, Schimpl A, Feller AC, Horak I. Ulcerative colitis-like disease in mice with a disrupted interleukin-2 gene. *Cell* 1993;75:253–61.
2. Kuhn R, Lohler J, Rennick D, Rajewsky K, Muller W. Interleukin-10-deficient mice develop chronic enterocolitis. *Cell* 1993;75:263–74.
3. Blumberg RS, Saubermann LJ, Strober W. Animal models of mucosal inflammation and their relation to human inflammatory bowel disease. *Curr Opin Immunol* 1999;11:648–56.
4. Scribano ML, Prantera C. Medical treatment of active Crohn's disease. *Aliment Pharmacol Ther* 2002;16(Suppl 4):35–9.
5. Yamamoto T, Allan RN, Keighley MR. Effect of fecal diversion alone on perianal Crohn's disease. *World J Surg* 2000;24:1258–62.
6. Winslet MC, Andrews H, Allan RN, Keighley MR. Fecal diversion in the management of Crohn's disease of the colon. *Dis Colon Rectum* 1993;36:757–62.
7. Duchmann R, Kaiser I, Hermann E, Mayet W, Ewe K, Meyer zum Buschenfelde KH. Tolerance exists towards resident intestinal flora but is broken in active inflammatory bowel disease (IBD). *Clin Exp Immunol* 1995;102:448–55.
8. Harper PH, Lee EC, Kettlewell MG, Bennett MK, Jewell DP. Role of the faecal stream in the maintenance of Crohn's colitis. *Gut* 1985;26: 279–84.
9. D'Haens GR, Geboes K, Peeters M, Baert F, Penninckx F, Rutgeerts P. Early lesions of recurrent Crohn's disease caused by infusion of intestinal contents in excluded ileum. *Gastroenterology* 1998;114:262–7.
10. Karban A, Eliakim R, Brant SR. Genetics of inflammatory bowel disease. *IMAJ* 2002;4:798–802.
11. Inohara N, Nunez G. The NOD: a signaling module that regulates apoptosis and host defense against pathogens. *Oncogene* 2001;20:6473–81.
12. Ogura Y, Inohara N, Benito A, Chen FF, Yamaoka S, Nunez G. Nod2, a Nod1/Apaf-1 family member that is restricted to monocytes and activates NF-kappaB. *J Biol Chem* 2001;276:4812–18.
13. Hugot JP, Chamaillard M, Zouali H, et al. Association of NOD2 leucine-rich repeat variants with susceptibility to Crohn's disease. *Nature* 2001;411:599–603.
14. Ogura Y, Bonen DK, Inohara N, et al. A frameshift mutation in NOD2 associated with susceptibility to Crohn's disease. *Nature* 2001;411:603–6.
15. Hampe J, Cuthbert A, Croucher PJ, et al. Association between insertion mutation in NOD2 gene and Crohn's disease in German and British populations. *Lancet* 2001;357:1925–8.
16. Murillo L, Crusius JB, van Bodegraven AA, Alizadeh BZ, Pena AS. CARD15 gene and the classification of Crohn's disease. *Immunogenetics* 2002;54:59–61.
17. Ahmad T, Armuzzi A, Bunce M, et al. The molecular classification of the clinical manifestations of Crohn's disease. *Gastroenterology* 2002;122: 854–66.
18. Cuthbert AP, Fisher SA, Mirza MM, et al. The contribution of NOD2 gene mutations to the risk and site of disease in inflammatory bowel disease. *Gastroenterology* 2002;122:867–74.

19. Calkins BM. A meta-analysis of the role of smoking in inflammatory bowel disease. *Dig Dis Sci* 1989;34:1841–54.
20. Andersson RE, Olaison G, Tysk C, Ekbom A. Appendectomy and protection against ulcerative colitis. *N Engl J Med* 2001;344:808–14.
21. Godet PG, May GR, Sutherland LR. Meta-analysis of the role of oral contraceptive agents in inflammatory bowel disease. *Gut* 1995;37:668–73.
22. Pullan RD, Rhodes J, Ganesh S, et al. Transdermal nicotine for active ulcerative colitis. *N Engl J Med* 1994;330:811–15.
23. Gionchetti P, Rizzello F, Venturi A, et al. Oral bacteriotherapy as maintenance treatment in patients with chronic pouchitis: a double-blind, placebo-controlled trial. *Gastroenterology* 2000;119:305–9.
24. Romagnani P, Annunziato F, Baccari MC, Parronchi P. T cells and cytokines in Crohn's disease. *Curr Opin Immunol* 1997;9:793–9.
25. Fuss IJ, Neurath M, Boirivant M, et al. Disparate CD4+ lamina propria (LP) lymphokine secretion profiles in inflammatory bowel disease. Crohn's disease LP cells manifest increased secretion of IFN-gamma, whereas ulcerative colitis LP cells manifest increased secretion of IL-5. *J Immunol* 1996;157:1261–70.
26. Targan SR, Hanauer SB, van Deventer SJ, et al. A short-term study of chimeric monoclonal antibody cA2 to tumor necrosis factor alpha for Crohn's disease. Crohn's Disease cA2 Study Group. *N Engl J Med* 1997;337:1029–35.

Correspondence: Dr. Y. Chowers, Dept. of Gastroenterology, Sheba Medical Center, Tel Hashomer 52621, Israel.
Phone: (972-3) 530-2679
Fax: (972-3) 530-3160
email: ychowers@post.tau.ac.il