

## TOPIC HIGHLIGHT

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# Novel genetic markers in inflammatory bowel disease

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## Abstract

Genetic factors play a significant role in determining inflammatory bowel disease (IBD) susceptibility. Epidemiologic data support genetic contribution to the pathogenesis of IBD, which include familial aggregation, twin studies, racial and ethnic differences in disease prevalence. Linkage studies have identified several susceptibility genes contained in different genomic regions named IBD1 to IBD9. Nucleotide oligomerization domain (NOD2) and human leukocyte antigen (HLA) genes are the most extensively studied genetic regions (IBD1 and IBD3 respectively) in IBD. Mutations of the NOD2 gene are associated with Crohn's disease (CD) and several HLA genes are associated with ulcerative colitis (UC) and CD. Toll like receptors (TLRs) have an important role in the innate immune response against infections by mediating recognition of pathogen-associated microbial patterns. Studying single-nucleotide polymorphisms (SNPs) in molecules involved in bacterial recognition seems to be essential to define genetic backgrounds at risk of IBD. Recently, numerous new genes have been identified to be involved in the genetic susceptibility to IBD: NOD1/Caspase-activation recruitment domains 4 (CARD4), Chemokine ligand 20 (CCL20), IL-11, and IL-18 among others. The characterization of these novel genes potentially will lead to the identification of therapeutic agents and clinical assessment of phenotype and prognosis in patients with IBD.

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**Key words:** Genetic; Inflammatory bowel disease; Human leukocyte antigen; Nucleotide oligomerization domain; Toll like receptors; Susceptibility

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## INTRODUCTION

Inflammatory bowel disease (IBD) includes ulcerative colitis (UC) and Crohn's disease (CD) which are characterized by chronic illness of unknown etiology; however, its development is influenced by genetic, environmental and immunological factors<sup>[1]</sup>.

Epidemiological studies suggest that genetic susceptibility is a major contributing factor to IBD. Molecular data from total genome scans and from candidate gene studies have led to the identification of genetic determinants of susceptibility and disease phenotype of UC and CD. The primary goal of genetic research is to identify genetic variants within specific genes which could modify homeostasis and increase disease susceptibility. There is growing attention to the innate immune response and the interaction between genetic factors and bacterial flora, or pathogen-associated molecular patterns in order to understand the contribution of environmental factors to disease susceptibility, as well as the phenotype based on a more precise molecular basis of disease pathogenesis. Clinical impact of the genetic findings has helped in understanding the heterogeneity of IBD in location, age at onset, clinical course and predicting response to conventional treatment.

## GENETIC MODEL

Clinical and epidemiological data do not support a simple Mendelian model of inheritance for IBD. In its place CD and UC are considered to be complex polygenic diseases. Two major methods for identification of genes in complex multifactorial diseases are used, the positional cloning method based on association studies and the candidate gene analysis. Linkage analysis allows scanning of the whole genome studying the co-segregation of the disease with a marker within families, constituting an important method where allele sharing between affected sibling pairs is used. An alteration of the observed ratio of sharing contrasting the expected is interpreted as evidence for linkage with a particular marker. On the other hand, the candidate gene analysis attempts to determine the

Table 1 Major regions and genes in IBD

Region	Localization	Involved genes
IBD1	Chromosome 16	NOD2/CARD15, IL-4R, CD11B
IBD2	Chromosome 12	Vitamin D receptor (VDR), STAT6, Interferon $\gamma$ , $\beta$ 7 integrine.
IBD3	Chromosome 6	Major histocompatibility complex (MHC): Class I, II, III.
IBD4	Chromosome 14	T- Lymphocyte receptor (TCR) and Leukotriene B4
IBD5	Chromosome 5	Organic cations transporter (OCTN), <i>Drosophila long disc homologue gene 5</i> (DLG5), <i>Multidrug resistant gene</i> (MDR1), IL-6, CD14
IBD6	Chromosome 19	Thromboxane A2, Leukotriene B4, ICAM-1
IBD7	Chromosome 1	Transforming growth factor Beta (TGF $\beta$ ), TNF $\alpha$ receptors.
IBD8	Chromosome 16	Under research
IBD9	Chromosome 9	CCR-5, CCR9, IL-12

NOD2/CARD15: Nucleotide oligomerization domain 2/Caspase-activation recruitment domains 15; STAT6: signal transducer and activator of transcription-6; ICAM-1: intracellular adhesion molecule 1; CCR5: CC-chemokine receptor5.

importance of specific genes in disease pathogenesis, using case-control cohorts or trios of affected progeny with both parents. The allelic frequencies or the transmission of a single-nucleotide polymorphism (SNP) towards affected progeny is studied and the differences between patients and controls might point towards the implication of a particular gene in the pathogenesis of the disease under investigation. It also includes positional candidate genes which are found in areas of linkage defined by genome screening. As CD and UC are likely to share some susceptibility genes, it has been proposed a genetic model of UC and CD where the two are polygenic disorders, sharing some susceptibility *loci*, but differing in others<sup>[2]</sup>. Linkage studies support this concept because some *loci* appear to interfere with susceptibility to IBD, which have also been implicated in the abnormal immune response and those susceptibility genes could interfere with the disease phenotype such as extension, need for procto-colectomy, extraintestinal manifestations, as well as the response to different treatments. From most of the genome-wide scans performed in IBD, a number of susceptibility regions on several chromosomes have been found<sup>[3-6]</sup> and according to their initial date of reporting and independent confirmations, the regions on such chromosomes have been renamed as IBD1-9 (Table 1).

## IBD1

Accurate mapping of the IBD1 locus has led to identification of the underlying gene called the NOD2/Caspase-activation recruitment domains 15 (CARD15) gene located on the pericentromeric region on the long arm of chromosome 16 (16p12.3) extending to 16q13<sup>[7-10]</sup>. Several studies have demonstrated the identification of NOD2/CARD15 gene within the IBD1 locus as a susceptibility gene in CD, suggesting that approximately 25%-30% of the genetic susceptibility in CD can be explained by mutations in NOD2/CARD5 though most of these studies have shown no association between

NOD2 mutations and susceptibility to UC<sup>[11,12]</sup>. Thirty non-conservative polymorphisms have been identified within the gene that are associated with CD and only three are common (Arg702Trp, Gly908Arg and Leu1007insC). The three common variants, however, account for approximately 82% of the mutated alleles<sup>[13]</sup>. Nevertheless, these mutations seem to have different effects on the risk of developing CD: Arg702Trp, Gly908Arg and a deletion in the last 33 aminoacids Leu1007finsC, which are present in 43% of the patients with CD (10%-30% is heterozygous and 2%-15% is homozygous for these mutations)<sup>[8-10]</sup>. These variants in NOD2 have been associated with certain clinical features of CD<sup>[14]</sup>. The NOD2 contribution seems to be stronger in Ashkenazi Jewish population who has a higher frequency of the Gly908Arg NOD2 variant. In relation to phenotypic expression and translation into the clinic, some associations between NOD2 mutations and earlier-onset disease in adult populations, fistulizing disease, fibrostenosing disease behavior and increased risk of need for surgery in children have been described<sup>[15-18]</sup>.

CARD gene codes for a protein expressed in several cells of innate immunity, epithelial cells and Paneth cells<sup>[11,12]</sup>. This protein consists of two N-terminal CARD, a central nucleotide-binding domain and a C-terminal leucine-rich-repeat region (LRR). It has been reported that CARD15 is implicated in the recognition of a bacterial product peptidoglycan-derived muramyl dipeptide (MDP) that enters into the cytosol via a transporter protein hPepT1 and interacts with the LRR of NOD2. Mutations within the leucine-rich region are associated with CD, as mutations within the nucleotide-binding domain are associated with granulomatous diseases<sup>[19]</sup>. Through the recognition of MDP, secretion of alpha-defensins is stimulated for protection against microbial invasion. In CD patients, a diminished expression of alpha-defensins has been found in those carriers with NOD2 mutation<sup>[20,21]</sup>. It is known that through this recognition of bacterial products the nuclear factor kappa B (NF $\kappa$ B) activation is regulated<sup>[10,22]</sup>.

Evidence show that NOD2 protein acts as an important regulator of NF $\kappa$ B activation in response to the Toll-like receptor (TLR) 2 activation system leading to its down regulation<sup>[23,24]</sup>. However in carriers of mutant protein, this process does not occur and proinflammatory cytokines are produced with a Th1 profile<sup>[25,26]</sup>.

## IBD2

This region is located on chromosome 12 showing greater linkage evidence in UC compared with CD. A number of possible candidate genes have been investigated including signal transducer and activator of transcription-6 (STAT6), INF $\gamma$ , metalloproteinase (MMP18), Vitamin D receptor (VDR) and  $\beta$ 7 integrin family that could be associated with the susceptibility to IBD. Parkes *et al*<sup>[27]</sup> found that IBD2 appears to make a major contribution to UC susceptibility but has only a relatively minor effect on CD.

### Vitamin D receptor

The Vitamin D receptor (VDR) is a member of a steroid receptor family and mediates the effects of the active metabolite 1.25 (OH)<sub>2</sub> vitamin D<sub>3</sub> by regulating

transcription of a number of different genes. It is synthesized by activated macrophages. It is expressed by monocytes and activated B and T lymphocytes. It activates monocytes and macrophages but suppresses lymphocyte proliferation and immunoglobulin production, and also inhibits transcription factor NF $\kappa$ B, and the production of IL-2, IL-12 and interferon  $\gamma$ . 1,25 (OH) $_2$ D $_3$  is the form of vitamin D that binds to the VDR and inhibits experimental autoimmunity<sup>[28,29]</sup>. Vitamin D deficiency and VDR deficiency have been shown to exacerbate chronic IBD in IL-10 knock out mice<sup>[29,30]</sup>. Absence of the VDR results in mice that are extremely susceptible to chemical injury in the gut<sup>[30]</sup>. The linked SNPs found at the 3' end of VDR are: *BsmI*, *ApaI*, *TaqI*, and the exon 2 splice site *FokI* polymorphism<sup>[31,32]</sup>. *FokI* polymorphism has been associated with osteoporosis, *TaqI* polymorphism with the risk of prostate cancer<sup>[33]</sup> and recently homozygotes for the *TaqI* allele have been shown to have altered susceptibility to a variety of infectious diseases<sup>[34]</sup>. Simmons *et al*<sup>[35]</sup> studied 403 European Caucasian patients with IBD, and found significantly more homozygotes for the *TaqI* polymorphism among patients with CD than in patients with UC or controls, providing evidence for a genetic association between CD susceptibility and a gene that lies within one of the candidate regions determined by linkage analysis. Dressner-Pollak *et al*<sup>[36]</sup> found that *BsmI* VDR gene polymorphism is associated with increased susceptibility to UC in Israeli Ashkenazi patients with UC contrasting with *TaqI* polymorphisms that favor susceptibility to CD. It seems that in the absence of the VDR, inflammation in the gut is increased, colonic epithelial cell proliferation is dysregulated, and the host tissue fails to satisfactorily maintain gastrointestinal integrity following chemical insult. These data identify vitamin D as a key regulator of gastrointestinal homeostasis and an important player in regulation of the innate immune response.

### IFN- $\gamma$

IFN- $\gamma$  seems to be specifically important in CD pathogenesis as suggested by case control studies that showed elevated levels of IFN- $\gamma$  production in the mucosa in patients with CD, but not in UC<sup>[37]</sup>. Data indicate that patients with relapsing perianal fistulizing disease have an increased production of IFN- $\gamma$  measured by *in vitro* cell cultures<sup>[38]</sup>. Cytokine genotyping study showed that IFN- $\gamma$  (+874T/A) polymorphism is found in an increased proportion of patients with fistulizing CD<sup>[39]</sup> probably related to the reduction of tissue repair and migratory potential in fibroblasts apparently influenced by IFN- $\gamma$  in CD patients<sup>[40]</sup>.

### IBD3

Major histocompatibility complex (MHC) genes are located in this region, specifically on the short arm of the human chromosome 6.

With a candidate gene approach the MHC is the most extensively studied region. Two meta-analyses have been carried out to scan for IBD regions that are common for all populations<sup>[5,41]</sup> in which the highest evidence for linkage to IBD was achieved at the IBD3 locus. There are 3 classes

of MHC genes: I, II and III. The antigenic recognizing process in T-lymphocytes from the antigen presenting cells is achieved through the antigenic recognition associated with the MHC gene product<sup>[42]</sup>.

### HLA class I

Some not classical genes related to the class I genes such as MHC class I chain-related gene A (MICA) and MHC class I-related chain B (MICB), are expressed in the basolateral cells in the gastric epithelium, fibroblasts, endothelial and dendritic cells. It is known that its expression rises during viral and bacterial infections<sup>[43]</sup>. Some genetic studies in patients with IBD have found associations with MICA-A6 and HLA-B52 in Japanese patients with UC<sup>[44]</sup>, MICA\*010 and HLA-B\*1501 in English patients with fistulous CD<sup>[23]</sup>. MICA and MICB bind to an activating receptor natural killer group 2D (NKG2D) which is expressed on NK cells, T cells and macrophages and the interactions between these receptors may directly stimulate cell cytotoxicity as well as providing costimulation for NK and T cell activation. Several MICA alleles have been shown to alter the binding affinity with NKG2D suggesting they may exert a functional effect on immune activation. In contrast to HLA class II, HLA class I genes show a weak and inconsistent role in IBD. The functional significance of these polymorphisms and the nature of selective forces maintaining them are still being elucidated.

### HLA class II

Class II genes are located on the centromeric pole of the short arm of the human chromosome 6 and include HLA-DR, DP and DQ *loci* expressed in a dimeric glycoprotein only in macrophages, activated T-lymphocytes, B-lymphocytes, dendritic, epithelial and endothelial cells, playing a central role in the immune response. Polymorphisms in these molecules are concentrated around specific pockets of the binding groove that interact with critical side-chains or anchor residues of peptides. The different HLA molecules may bind preferentially to different peptides, or bind the same peptide with varying affinities. In IBD the molecular mimicry may exist between the peptides derived from bacterial luminal flora and from self antigens present in the gut, leading to the generation of auto reactive T cells and contributing to disease pathogenesis. The mechanism of cross reactivity is supported by the identification of murine MHC-restricted CD4+ T cells reactive to enteric bacterial antigens that are able to induce colitis by adoptive transfer<sup>[45]</sup>.

In a meta-analysis made by Stokkers *et al*<sup>[46]</sup>, positive associations between UC and HLA-DR2, HLA-DRB1\*1502, HLA-DR9 and HLA-DRB1\*0103 were found. A study from Mexican population found that HLA-DRB1\*0103 allele was associated with UC and its severe manifestations such as colectomy and pancolitis, while HLA-DRB1\*15 allele was only associated with pancolitis in Mexican patients with UC.

### HLA class III

These genes are located on the 1100 kb section between class I and II genes inside the MHC, and contain about 70



genes. The complement gene block is inherited as a genetic unit known as complotype. Each complotype codifies for the synthesis of complement classic pathway C2, C4A, C4b factors, and alternative pathway B factor, which may suggest that alterations within the region might affect the host's defense system and introduce a complement deficiency. This raises attention when TNF $\alpha$  is thought to play an important role in the pathogenesis of IBD, acting as a potent proinflammatory cytokine with elevated serum and tissue levels in patients with IBD<sup>[47-49]</sup>, and evidence show that there are specific genetic polymorphisms involving TNF $\alpha$  that influence the amount of cytokine produced. Bouma *et al.*<sup>[50]</sup> and Louis *et al.*<sup>[51]</sup> studied the allelic frequency of TNF $\alpha$  gene polymorphisms at -308 position finding that polymorphism in allele 2 was decreased in UC patients as compared to normal controls. It was also found that patients with pancolitis had increased frequency in the TNF-C haplotype<sup>[52]</sup>. In a Mexican population with UC, the presence of TNF\*2 allele was associated with the presence of this disease as compared with healthy subjects (23.7% vs 3%,  $P = 0.00002$ ; OR = 10.1; 95% CI = 2.69-26.8)<sup>[53]</sup>. In Mexican patients with UC, an association was found between complotype SC30 (Bf\*S-C2\*C-C4A\*3-C4B\*0) and UC<sup>[54]</sup>, which might suggest that activation of complement system could interfere with the disease pathogenesis.

## IBD4

This locus is located on chromosome 14. Evidence for linkage to the adjacent D14S261 and D14S283 *loci* on chromosome 14q11-12 satisfied criteria for confirmed linkage and this region was designated IBD4 locus<sup>[55]</sup>. Vermeire *et al.*<sup>[56]</sup> in a genome wide scan in a 149 Belgian IBD affected families cohort, demonstrated the existence of IBD4 on 14q11 as a susceptibility *loci*. The IL-25 gene is located within this susceptibility region at 14q11.2. The IL-25 gene is located within this susceptibility region at 14q11.2.

## IL-25

Interleukin-25 (IL-25) is a newly identified proinflammatory cytokine that has been shown to promote Th2 responses by inducing cytokines such as IL-4, IL-5 and IL-13, implicated in the initiation of type 2 cytokine-dependent immunity to gastrointestinal infection and limiting proinflammatory cytokine production and chronic intestinal inflammation. IL-25-deficient knockout mice failed to develop a type 2 immune response or eradicate infection. Moreover, chronically infected IL-25 (-/-) mice developed severe infection-induced intestinal inflammation identifying a role for IL-25 in limiting pathologic inflammation at mucosal sites in the gastrointestinal tract<sup>[57]</sup>. Still more evidence is needed to conclude a precise role of this gene in the IBD susceptibility.

## IBD5

The IBD5 locus on chromosome 5q31-33<sup>[58]</sup> contains the cytokine cluster and is a candidate region for IBD. The IBD5 risk haplotype has been associated with CD, although there have been some suggestions of a weak

association with UC as well. Phenotypically this locus has been associated with earlier onset of disease as well as perianal disease<sup>[59-62]</sup>.

## Carnitine/Organic cation transporter genes

The organic cation transporter genes OCTN1 and OCTN2 are within a single haplotype block (block 7) of the IBD5 locus and some mutations have been reported within these: L503F (rs1050152) and G-207C (rs2631367) in the *SLC22A4* (OCTN1) and *SLC22A5* (OCTN2) genes, respectively, which are associated with the development of CD and also an association with susceptibility to UC has been reported<sup>[63]</sup>. The presence or combination of these mutations constitutes the TC haplotype, which is associated with ileal, colonic and perianal disease<sup>[64,65]</sup>. Associations between TC haplotype variants and CD affected sites have been shown in genotype-phenotype studies<sup>[66,67]</sup>. According to some studies, 1672C→T missense substitution in *SLC22A4* and the -207G→C transversion in the *SLC22A5* promoter contribute to disease susceptibility by impairing OCTN activity or expression respectively<sup>[68]</sup>. The risk associated with the OCTN-TC haplotype seems to be only observed in homozygotes and not in heterozygotes<sup>[60,63,69]</sup>, so the carriage of the homozygous OCTN-TC haplotype is likely to be associated with a higher relative risk for colonic disease. The association of the OCTN polymorphisms with CD phenotypes shows a higher frequency of the OCTN-TC haplotype in patients with colonic involvement compared with exclusive ileal disease<sup>[60]</sup>. It has been reported a moderate increase in the frequency of the TC haplotype among patients without fistulas or stenosis and this is compatible with the negative association with ileal involvement, showing a tendency towards a lower-frequency of ileocecal resection in the presence of at least one OCTN-TC haplotype and might explain the absence of colonic involvement<sup>[64]</sup>. The impaired eradication of luminal pathogens results in a persistent infection which may constitute a possible mechanism causing IBD.

## Drosophila long disc homologue 5 gene

Drosophila long disc homologue 5 gene (DLG5) on chromosome 10q22-23 is a member of the membrane associated guanylate kinase gene family which encodes cell scaffolding proteins and seems to play a role in the maintenance of intestinal epithelial cells, and its mutations have been involved in a rise in intestinal permeability<sup>[70]</sup>. DLG5 is a widely expressed protein found in many tissues such as the placenta, small bowel, colon, heart, skeletal muscle, liver and pancreas and it is important in signal transduction and epithelial cell integrity. Four haplotypes have been identified associated with IBD in a European cohort<sup>[71]</sup>. Haplotype A is characterized by the presence of an insertion of thirteen pairs in exon 26. It has been shown to be protective in some case control studies<sup>[72]</sup>, however it is substantially undertransmitted in people with IBD<sup>[62]</sup>. The haplotype characterized by the haplotype-tagging SNP G113A called Haplotype D, was found substantially overtransmitted in patients with IBD controversially contradictory with another<sup>[73]</sup> Belgian study where the D haplotype involving the 113A variant was shown to be undertransmitted in flamish patients with

IBD. These apparently contradictory results might yet be compatible with the possibility that DLG5 has a small effect in IBD with heterogeneity in its effect.

#### **ATP-binding cassette or multidrug resistant gene**

The multidrug-resistance (MDR1) gene is located on the long arm of chromosome 7 and consists of 29 exons. The total length is 209 kb and 6326 bp. Its product, the P-glycoprotein (Pgp), a member of the ATP binding cassette family, is an integral membrane protein which functions as an energy-dependent efflux pump and reduces the intracellular concentrations of toxins and xenobiotics<sup>[74]</sup>. Studies show evidence for natural single nucleotide polymorphisms (SNPs) of MDR1 gene and their effects on drug efficiency, toxicity, distribution, absorption and elimination. Two main polymorphisms or variants of this gene have been described, C3435T and G2677T which are associated with IBD in some populations<sup>[75,76]</sup> and have also been related with the expression of glycoprotein P-170. Variant C3435T was related with the presence of pancolitis in patients with UC in Scotland<sup>[77]</sup>. However, the frequency of SNPs is low and is different among populations, with the exception of three SNPs in exon 12 (C1236T), exon 20 (G2677T/A) and exon 26 (C3435T), and some of them are correlated with different diseases and clinical characteristics<sup>[78]</sup>. Glucocorticoid is a potent inhibitor of the T cell activation and a highly effective treatment for IBD<sup>[79]</sup>. Over-expression produces three molecular mechanisms of glucocorticoid resistance: increase of P-gp and decrease of cytoplasmatic glucocorticoid, dysfunction at the level of glucocorticoid receptor and activation of NFκB, resulting in inhibition of glucocorticoid receptor transcriptional activity. Cucchiara *et al*<sup>[80]</sup> investigated the predisposition and response to medical therapy of TNFα and MDR1 genes in 200 pediatric patients with CD and 186 UC patients and 347 adults as a control. The 308A allele of the TNF-α gene was increased in both patients with CD and UC, strongly suggesting this polymorphism carries a significant reduction in response to steroid therapy.

#### **IL-6 (-174G/C) polymorphism**

IL-6 is a well-studied IBD candidate gene and its polymorphism has been associated with the site of disease in CD. IL-6 levels are higher in patients with active CD as compared to patients with active UC and normal controls<sup>[81,82]</sup>. A study from Cantor *et al*<sup>[39]</sup> demonstrated a relationship between IL-6 genotype and the site of CD, showing that patients with the high producer of IL-6 genotype were more likely to have colonic CD. In CD patients IL-6 concentrations also correlate with the disease activity, response to treatment and rate of relapse.

#### **IBD6**

In a Canadian linkage scan, a linkage peak of genome-wide scan on chromosome 19p was identified and appeared to confer susceptibility to both CD and UC<sup>[61]</sup>. Two independent genome-wide linkage studies also determined evidence for linkage to this region and two other meta-analyses of all published genome-wide scans<sup>[5,41,83]</sup> identified

evidence that supports the existence of a locus conferring susceptibility to IBD in chromosomal region 19p, currently known as the IBD6 locus. In order to identify IBD susceptibility alleles in the 19p region two candidate genes DDXL and intracellular adhesion molecule 1 (ICAM-1) were examined in a case-control study with CD and UC patients but no association with either UC or CD was found in 3 single nucleotide polymorphisms in DDXL gene, however a significant association was found between ICAM-1 K469 homozygosity and CD as well as E469 and fistulating disease<sup>[84]</sup>.

#### **IBD7**

Located on the short arm of chromosome 1, IBD7 is thought susceptibility genes are residing in this locus. One of these codifies for the transforming growth factor beta 2 (TGF-beta 2) which is a cytokine present in human and bovine milk and plays a critical role in the development of tolerance, prevention of autoimmunity, and in anti-inflammatory responses and is also a potent inhibitor of intestinal epithelial cell (IEC) growth and stimulates IEC differentiation<sup>[85-87]</sup>. McKaig and colleagues<sup>[88]</sup> studied the expression of TGF-beta isoforms in isolated and cultured primary human intestinal myofibroblasts from normal controls as well as from UC and CD patients, and determined the responsiveness of these cells to TGF-beta isoforms. Proliferation of myofibroblasts in CD patients was significantly greater than that of myofibroblasts derived from normal and ulcerative colitis tissue, suggesting that it may be related to the development of intestinal strictures, seen frequently as a major feature in CD. The anti-inflammatory attributes of TGF-β3 may be evidenced in a study of children with active intestinal Crohn's disease, who were treated with an oral polymeric diet rich in TGF-β2 as the sole source of nutrition for eight weeks and it was associated with mucosal healing and a down-regulation of mucosal pro-inflammatory cytokines mRNA in both the terminal ileum and colon<sup>[89]</sup>. However, further investigation on this locus is needed to determine the level of significance related to the pathogenesis of IBD.

#### **IBD8**

This gene is located on the short arm of human chromosome 16. There has been evidence of a second chromosome 16 locus (IBD8) independent of NOD2 that overlaps IBD1 on the pericentromeric short arm<sup>[90]</sup>, but yet no studies have been performed for the identification on this locus.

#### **IBD9**

The CC-chemokine receptor 5 (CCR5) gene located on chromosome 3p21 coincides with this IBD susceptibility locus identified by genome-wide scanning<sup>[91]</sup>. The CCR5 is the receptor for regulated and normal T-cell expressed and secreted (RANTES), a natural pro-inflammatory cytokine. A 32-bp deletion (A32) in the CCR5 gene results in a nonfunctional receptor found with a high

frequency in Caucasians. They found an association between CCR5delta32 homozygosity and the presence of anal lesions in CD patients with statistical significance<sup>[92]</sup>. Several genes located in these regions are still under research (Table 1).

## GENES INVOLVED IN THE INNATE IMMUNE RESPONSE

### Toll like receptors

Rising evidence suggests an essential role of the enteric bacterial flora in the pathogenesis of IBD. Rather than a passive barrier, the intestinal epithelium is an active participant in the mucosal immune response through its expression of proinflammatory genes, secretion of inflammatory cytokines, and recruitment of inflammatory cells in response to pathogenic bacteria and their products<sup>[93]</sup>. IBD has been increasingly thought to result from an aberrant interaction between the environment and the genetically susceptible host. Specifically, several lines of evidence point to a deregulation of the immune response to a commensal or uncharacterized pathogenic bacterium in the gut<sup>[94]</sup>. Animal models have demonstrated that genes involved in the regulation of the immune response are likely to play a crucial role in the genetic predisposition to IBD<sup>[95]</sup>. The family of Toll-like receptors (TLR) recognizes pathogen-associated molecular patterns and activates signal transduction pathways of the innate immune response genes including NFκB<sup>[95]</sup>. The SNPs involved in bacterial recognition are becoming essential in understanding individual responses to bacterial components and defining genetic backgrounds at risk of IBD.

**Toll-like receptor 4:** The toll-like receptor 4 (TLR4) gene is located on the long arm of human chromosome 9 and it identifies lipopolysaccharides (LPS) on gram-negative bacteria. It has been found strongly upregulated in IBD, and it binds to LPS together with CD14 and by internalization prevents inappropriate NFκB activation<sup>[96]</sup>. The TLR4 Asp299Gly polymorphism has been associated with CD and UC in a Belgian study<sup>[97]</sup>. On the other hand this SNP was exclusively related to CD in other series<sup>[97-102]</sup> and TLR4 polymorphism Thr399Ile was exclusively associated with UC in others<sup>[99]</sup>. A lipid A-mimetic CRX-526 with antagonistic activity for TLR4, is known to block the interaction of LPS with the immune system, therefore, CRX-526 can prevent the expression of proinflammatory genes stimulated by LPS *in vitro*. This disturbed activation of the innate immune system by bacterial antigens may be crucial in some patients with IBD.

**TLR1, TLR2, and TLR6:** Pierik *et al.*<sup>[100]</sup> studied the nonsynonymous polymorphisms in other TLR genes in IBD. They found no SNP was involved in disease susceptibility, and a number of variants influenced the disease phenotype, however, they found a positive association between TLR1 R80T and TLR2 R753G and pancolitis in UC. TLR2 and its cofactors TLR1 and TLR6 are involved in the initial immune response to bacteria and

recognition of peptidoglycan. This TLR2 is required for recognition of Gram-positive and mycobacterial pathogen-associated molecular patterns (PAMPs) including bacterial lipopeptide lipoteichoic acid (LTA), peptidoglycan (PGN) and the mutations associated are involved in severe mycobacterial infections<sup>[103-109]</sup>. Further studies have shown that combinations of TLR molecules are required for recognition of certain PAMPs and that specifically, combined expression of TLR2 and TLR6 is required for recognition of PGN<sup>[110-114]</sup>.

It is suggested that TLR1 may be regulated diversely in inflammation to down-regulate or enhance the response to certain TLR2 ligands and that a relative absence of TLR2 protein expression may be important in preventing chronic proinflammatory cytokine secretion in response to commensal Gram-positive bacteria in the gut<sup>[95]</sup>.

**TLR5:** TLR5 gene is located on the short arm of human chromosome 1 and is responsible for recognizing a protein named flagellin which is found in intestinal bacteria<sup>[113]</sup>. Lodes *et al.*<sup>[113]</sup> observed through serological studies a strong response to flagellin in multiple animal models of colitis and synergism has been identified between NOD2 and TLR5 signaling<sup>[114]</sup>. The dominant negative variant of TLR5 (TLR5-stop) seems to protect against the development of CD and results in significant reduction of IgA and IgE circulating antibodies against flagellin<sup>[115]</sup>, suggesting that pharmacological blockade of TLR5 has potential in the treatment of CD.

### NOD1/CARD4 gene

Located on chromosome 7p14, NOD1/CARD4 gene is one of the three human NOD-LLR proteins that has similar structure to NOD2/CARD15, having only one CARD domain, a central NOD domain and a leucine rich repeat region (LLR). Its function is the recognition of gram negative bacterial products such as γ-glutamine diaminopimelic acid and plays a role in colonic epithelial defense against the intracellular pathogens *E.coli* and *Shigella flexneri*. Its effector domain is associated with Ripk2 (a CARD-containing interleukin-1 beta converting enzyme-associated kinase) mediating NFκB activation. In a recent study of 556 patients with IBD (294 CD and 252 UC), an association between the variant rs695857 in nucleotides 30, 258 and 950 of NOD1 and the development of IBD was found. Another variant known as rs2907748 in nucleotides 30, 246 and 263 was also associated with the presence of UC and CD and even with the early onset of the disease (< 25 years)<sup>[116]</sup>. These genetic variants of NOD1 have shown to be associated with disease susceptibility supporting that impaired local immunity might influence bacterial proliferation and aberrant immune responses in the host.

### CCL20

CC-chemokine ligand 20 (CCL20) gene is located on the short arm of human chromosome 2 and codifies for the CCL20 cytokine ligand, which is responsible for the chemoattraction of immature dendritic cells that express CCR6 receptor on the intestinal epithelium and on Peyer's plaques<sup>[117]</sup>, and also attracts memory T lymphocytes.



Microarray analysis and PCR-RT quantification have shown a rise in the expression of mRNA from IBD biopsies with inflammation compared to normal biopsies<sup>[118]</sup>. A study made in Korean UC patients showed that the expression of CCL20 was significantly up-regulated in the peripheral blood mononuclear cells compared with those of normal healthy controls. Interestingly, untreated UC groups expressed higher levels of CCL20 mRNA than treated UC and normal control groups, therefore suggesting that CCL20 could be modulated by anti-inflammatory drugs<sup>[119]</sup>.

### Interleukin 11

IL-11 mediates anti-inflammatory effects and is able to downregulate LPS-induced NF $\kappa$ B activation. The IL-11 gene is therefore a good candidate involved in genetic predisposition to IBD. Klein *et al*<sup>[120]</sup> evaluated the role of IL-11 in IBD, finding decreased expression and a failure to downregulate NF $\kappa$ B expression that could play an important role in the pathogenesis of UC.

### Interleukin 18

IL-18 is a pleiotropic cytokine that induces the production of IFN $\gamma$  and also regulates Th2 cytokines. It seems to be an important cytokine involved in the pathogenesis of IBD, apparently because SNPs at the 5'-end of IL-18 gene might be closely related to the etiology of IBD. Takagawa *et al*<sup>[121]</sup> found that IL-18 gene promoter polymorphisms may be related to the extent of disease in UC patients.

### Interleukin 23

IL-23 is a heterodimeric cytokine composed of a p19 subunit and the p40 subunit of IL-12. It is produced by macrophages and dendritic cells, and activates memory T cells. Interleukin-12 (IL-12) is composed of p35 and p40 subunits and acts as an important factor for the differentiation of naive T cells into T-helper type 1 CD4<sup>+</sup> lymphocytes secreting interferon-gamma. Therefore it has been reported that IL-12 is crucial for T-cell-dependent immune and inflammatory responses through the use of IL-12 p40 gene-targeted mice and neutralizing antibodies against p40<sup>[122-127]</sup>. Apparently IL-12 is a key factor driving Th1 responses and IFN production in the initial phases of an immune response, but conversely IL-12 may play a subsequent immunoregulatory role in late-stage inflammation at a point when IL-23 strongly supports the inflammatory process. IL-23 induces the production of IL-17 by a unique subset of memory T cells. IL-17 is known to stimulate fibroblasts, endothelial cells, macrophages and epithelial cells to secrete multiple pro-inflammatory mediators<sup>[128]</sup> and the local production of IL-17 causes site-specific activation of inflammatory cells<sup>[129-132]</sup>. Dendritic cells found in the lamina propria of the small intestine were described as constitutively expressing IL-23<sup>[131]</sup>, whereas IL-23 regulates a highly potent T cell-derived cytokine that has major actions on the immune system. IL-23 specifically stimulates memory CD4<sup>+</sup>T cells contrasting the IL-12 which is a stimulant for naive CD4<sup>+</sup> T cells<sup>[129,130]</sup>. Studies with IL-23 deficient mice show that IL-23 is essential for the manifestation of intestinal inflammation and a dominant role for IL-23 over

IL-12 in central nervous system and joint autoimmune inflammation has been described. These findings point to IL-23, but not IL-12, as the necessary mediator for organ specific autoimmune diseases development. Furthermore, the absence of IL-12 results in more severe disease, reflected in elevated and prolonged expression of proinflammatory cytokines. Yen and colleagues<sup>[132]</sup> reported that the activation of tissue-homing memory T cells by IL-23 is responsible for chronic inflammatory disease.

## CONCLUSION

Genetic research in IBD has provided knowledge about the complexity and heterogeneity of the disease and started to correlate the interactions between genetic and environmental risk factors in IBD; however, the complex genetic background that allows the development of IBD is not fully understood.

Understanding the pathways in which genetic factors influence IBD will uncover pathogenesis of the disease, offer more accurate diagnosis and ultimately lead to the breakthrough of better new drugs and therapies. Most of the important advances toward understanding this process have been identification of specific genetic associations with IBD, which will shed new light on future research of IBD.

## REFERENCES

- 1 Podolsky DK. Inflammatory bowel disease. *N Engl J Med* 2002; **347**: 417-429
- 2 Satsangi J, Morecroft J, Shah NB, Nimmo E. Genetics of inflammatory bowel disease: scientific and clinical implications. *Best Pract Res Clin Gastroenterol* 2003; **17**: 3-18
- 3 Hugot JP, Laurent-Puig P, Gower-Rousseau C, Olson JM, Lee JC, Beaugier L, Naom I, Dupas JL, Van Gossum A, Orholm M, Bonaiti-Pellie C, Weissenbach J, Mathew CG, Lennard-Jones JE, Cortot A, Colombel JF, Thomas G. Mapping of a susceptibility locus for Crohn's disease on chromosome 16. *Nature* 1996; **379**: 821-823
- 4 Hampe J, Schreiber S, Shaw SH, Lau KF, Bridger S, Macpherson AJ, Cardon LR, Sakul H, Harris TJ, Buckler A, Hall J, Stokkers P, van Deventer SJ, Nürnberg P, Mirza MM, Lee JC, Lennard-Jones JE, Mathew CG, Curran ME. A genomewide analysis provides evidence for novel linkages in inflammatory bowel disease in a large European cohort. *Am J Hum Genet* 1999; **64**: 808-816
- 5 Williams CN, Kocher K, Lander ES, Daly MJ, Rioux JD. Using a genome-wide scan and meta-analysis to identify a novel IBD locus and confirm previously identified IBD loci. *Inflamm Bowel Dis* 2002; **8**: 375-381
- 6 Barmada MM, Brant SR, Nicolae DL, Achkar JP, Panhuysen CI, Bayless TM, Cho JH, Duerr RH. A genome scan in 260 inflammatory bowel disease-affected relative pairs. *Inflamm Bowel Dis* 2004; **10**: 15-22
- 7 Ogura Y, Bonen DK, Inohara N, Nicolae DL, Chen FF, Ramos R, Britton H, Moran T, Karaliuskas R, Duerr RH, Achkar JP, Brant SR, Bayless TM, Kirschner BS, Hanauer SB, Nuñez G, Cho JH. A frameshift mutation in NOD2 associated with susceptibility to Crohn's disease. *Nature* 2001; **411**: 603-606
- 8 Hugot JP, Chamaillard M, Zouali H, Lesage S, Cézard JP, Belaiche J, Almer S, Tysk C, O'Morain CA, Gassull M, Binder V, Finkel Y, Cortot A, Modigliani R, Laurent-Puig P, Gower-Rousseau C, Macry J, Colombel JF, Sahbatou M, Thomas G. Association of NOD2 leucine-rich repeat variants with susceptibility to Crohn's disease. *Nature* 2001; **411**: 599-603
- 9 Hampe J, Cuthbert A, Croucher PJ, Mirza MM, Mascheretti

- S, Fisher S, Frenzel H, King K, Hasselmeyer A, MacPherson AJ, Bridger S, van Deventer S, Forbes A, Nikolaus S, Lennard-Jones JE, Foelsch UR, Krawczak M, Lewis C, Schreiber S, Mathew CG. Association between insertion mutation in NOD2 gene and Crohn's disease in German and British populations. *Lancet* 2001; **357**: 1925-1928
- 10 **Hampe J**, Frenzel H, Mirza MM, Croucher PJ, Cuthbert A, Mascheretti S, Huse K, Platzner M, Bridger S, Meyer B, Nürnberg P, Stokkers P, Krawczak M, Mathew CG, Curran M, Schreiber S. Evidence for a NOD2-independent susceptibility locus for inflammatory bowel disease on chromosome 16p. *Proc Natl Acad Sci USA* 2002; **99**: 321-326
  - 11 **Lesage S**, Zouali H, Cézard JP, Colombel JF, Belaiche J, Almer S, Tysk C, O'Morain C, Gassull M, Binder V, Finkel Y, Modigliani R, Gower-Rousseau C, Macry J, Merlin F, Chamaillard M, Jannot AS, Thomas G, Hugot JP. CARD15/NOD2 mutational analysis and genotype-phenotype correlation in 612 patients with inflammatory bowel disease. *Am J Hum Genet* 2002; **70**: 845-857
  - 12 **Rosenstiel P**, Fantini M, Bräutigam K, Kühbacher T, Waetzig GH, Seegert D, Schreiber S. TNF-alpha and IFN-gamma regulate the expression of the NOD2 (CARD15) gene in human intestinal epithelial cells. *Gastroenterology* 2003; **124**: 1001-1009
  - 13 **Gutierrez O**, Pipaon C, Inohara N, Fontalba A, Ogura Y, Prosper F, Nuñez G, Fernandez-Luna JL. Induction of Nod2 in myelomonocytic and intestinal epithelial cells via nuclear factor-kappa B activation. *J Biol Chem* 2002; **277**: 41701-41705
  - 14 **Török HP**, Glas J, Lohse P, Folwaczny C. Alterations of the CARD15/NOD2 gene and the impact on management and treatment of Crohn's disease patients. *Dig Dis* 2003; **21**: 339-345
  - 15 **Harton JA**, Linhoff MW, Zhang J, Ting JP. Cutting edge: CATERPILLER: a large family of mammalian genes containing CARD, pyrin, nucleotide-binding, and leucine-rich repeat domains. *J Immunol* 2002; **169**: 4088-4093
  - 16 **Kobayashi KS**, Chamaillard M, Ogura Y, Henegariu O, Inohara N, Nuñez G, Flavell RA. Nod2-dependent regulation of innate and adaptive immunity in the intestinal tract. *Science* 2005; **307**: 731-734
  - 17 **Wehkamp J**, Harder J, Weichenthal M, Schwab M, Schäffeler E, Schlee M, Herrlinger KR, Stallmach A, Noack F, Fritz P, Schröder JM, Bevins CL, Fellermann K, Stange EF. NOD2 (CARD15) mutations in Crohn's disease are associated with diminished mucosal alpha-defensin expression. *Gut* 2004; **53**: 1658-1664
  - 18 **Ogura Y**, Inohara N, Benito A, Chen FF, Yamaoka S, Nuñez G. Nod2, a Nod1/Apaf-1 family member that is restricted to monocytes and activates NF-kappaB. *J Biol Chem* 2001; **276**: 4812-4818
  - 19 **Hisamatsu T**, Suzuki M, Reinecker HC, Nadeau WJ, McCormick BA, Podolsky DK. CARD15/NOD2 functions as an antibacterial factor in human intestinal epithelial cells. *Gastroenterology* 2003; **124**: 993-1000
  - 20 **McDonald C**, Chen FF, Ollendorff V, Ogura Y, Marchetto S, Lécine P, Borg JP, Nuñez G. A role for Erbin in the regulation of Nod2-dependent NF-kappaB signaling. *J Biol Chem* 2005; **280**: 40301-40309
  - 21 **Yamamoto-Furusho JK**, Barnich N, Xavier R, Hisamatsu T, Podolsky DK. Centaurin beta1 down-regulates nucleotide-binding oligomerization domains 1- and 2-dependent NF-kappaB activation. *J Biol Chem* 2006; **281**: 36060-36070
  - 22 **Watanabe T**, Kitani A, Murray PJ, Strober W. NOD2 is a negative regulator of Toll-like receptor 2-mediated T helper type 1 responses. *Nat Immunol* 2004; **5**: 800-808
  - 23 **Ahmad T**, Armuzzi A, Bunce M, Mulcahy-Hawes K, Marshall SE, Orchard TR, Crawshaw J, Large O, de Silva A, Cook JT, Barnardo M, Cullen S, Welsh KI, Jewell DP. The molecular classification of the clinical manifestations of Crohn's disease. *Gastroenterology* 2002; **122**: 854-866
  - 24 **Büning C**, Genschel J, Bühner S, Krüger S, Kling K, Dignass A, Baier P, Bochow B, Ockenga J, Schmidt HH, Lochs H. Mutations in the NOD2/CARD15 gene in Crohn's disease are associated with ileocecal resection and are a risk factor for reoperation. *Aliment Pharmacol Ther* 2004; **19**: 1073-1078
  - 25 **Kugathasan S**, Collins N, Maresco K, Hoffmann RG, Stephens M, Werlin SL, Rudolph C, Broeckel U. CARD15 gene mutations and risk for early surgery in pediatric-onset Crohn's disease. *Clin Gastroenterol Hepatol* 2004; **2**: 1003-1009
  - 26 **Sun L**, Roesler J, Rösen-Wolff A, Winkler U, Koch R, Thürigen A, Henker J. CARD15 genotype and phenotype analysis in 55 pediatric patients with Crohn disease from Saxony, Germany. *J Pediatr Gastroenterol Nutr* 2003; **37**: 492-497
  - 27 **Parkes M**, Barmada MM, Satsangi J, Weeks DE, Jewell DP, Duerr RH. The IBD2 locus shows linkage heterogeneity between ulcerative colitis and Crohn disease. *Am J Hum Genet* 2000; **67**: 1605-1610
  - 28 **Cantorna MT**, Mahon BD. Mounting evidence for vitamin D as an environmental factor affecting autoimmune disease prevalence. *Exp Biol Med* (Maywood) 2004; **229**: 1136-1142
  - 29 **Cantorna MT**, Munsick C, Bemiss C, Mahon BD. 1,25-Dihydroxycholecalciferol prevents and ameliorates symptoms of experimental murine inflammatory bowel disease. *J Nutr* 2000; **130**: 2648-2652
  - 30 **Froicu M**, Weaver V, Wynn TA, McDowell MA, Welsh JE, Cantorna MT. A crucial role for the vitamin D receptor in experimental inflammatory bowel diseases. *Mol Endocrinol* 2003; **17**: 2386-2392
  - 31 **Morrison NA**, Qi JC, Tokita A, Kelly PJ, Crofts L, Nguyen TV, Sambrook PN, Eisman JA. Prediction of bone density from vitamin D receptor alleles. *Nature* 1994; **367**: 284-287
  - 32 **Gross C**, Eccleshall TR, Malloy PJ, Villa ML, Marcus R, Feldman D. The presence of a polymorphism at the translation initiation site of the vitamin D receptor gene is associated with low bone mineral density in postmenopausal Mexican-American women. *J Bone Miner Res* 1996; **11**: 1850-1855
  - 33 **Wood RJ**, Fleet JC. The genetics of osteoporosis: vitamin D receptor polymorphisms. *Annu Rev Nutr* 1998; **18**: 233-258
  - 34 **Ingles SA**, Ross RK, Yu MC, Irvine RA, La Pera G, Haile RW, Coetzee GA. Association of prostate cancer risk with genetic polymorphisms in vitamin D receptor and androgen receptor. *J Natl Cancer Inst* 1997; **89**: 166-170
  - 35 **Simmons JD**, Mullighan C, Welsh KI, Jewell DP. Vitamin D receptor gene polymorphism: association with Crohn's disease susceptibility. *Gut* 2000; **47**: 211-214
  - 36 **Dresner-Pollak R**, Ackerman Z, Eliakim R, Karban A, Chowers Y, Fidler HH. The BsmI vitamin D receptor gene polymorphism is associated with ulcerative colitis in Jewish Ashkenazi patients. *Genet Test* 2004; **8**: 417-420
  - 37 **Noguchi M**, Hiwatashi N, Liu Z, Toyota T. Enhanced interferon-gamma production and B7-2 expression in isolated intestinal mononuclear cells from patients with Crohn's disease. *J Gastroenterol* 1995; **30** Suppl 8: 52-55
  - 38 **Agnholt J**, Dahlerup JF, Buntzen S, Tøttrup A, Nielsen SL, Lundorf E. Response, relapse and mucosal immune regulation after infliximab treatment in fistulating Crohn's disease. *Aliment Pharmacol Ther* 2003; **17**: 703-710
  - 39 **Cantor MJ**, Nickerson P, Bernstein CN. The role of cytokine gene polymorphisms in determining disease susceptibility and phenotype in inflammatory bowel disease. *Am J Gastroenterol* 2005; **100**: 1134-1142
  - 40 **Leeb SN**, Vogl D, Gunkel M, Kiessling S, Falk W, Göke M, Schölmerich J, Gelbmann CM, Rogler G. Reduced migration of fibroblasts in inflammatory bowel disease: role of inflammatory mediators and focal adhesion kinase. *Gastroenterology* 2003; **125**: 1341-1354
  - 41 **van Heel DA**, Fisher SA, Kirby A, Daly MJ, Rioux JD, Lewis CM. Inflammatory bowel disease susceptibility loci defined by genome scan meta-analysis of 1952 affected relative pairs. *Hum Mol Genet* 2004; **13**: 763-770
  - 42 **Yamamoto-Furusho JK**. Immunogenetics of chronic ulcerative colitis. *Rev Invest Clin* 2003; **55**: 705-710
  - 43 **Das H**, Groh V, Kuijl C, Sugita M, Morita CT, Spies T, Bukowski JF. MICA engagement by human Vgamma2Vdelta2 T cells enhances their antigen-dependent effector function. *Immunity* 2001; **15**: 83-93
  - 44 **Seki SS**, Sugimura K, Ota M, Matsuzawa J, Katsuyama Y,



- Ishizuka K, Mochizuki T, Suzuki K, Yoneyama O, Mizuki N, Honma T, Inoko H, Asakura H. Stratification analysis of MICA triplet repeat polymorphisms and HLA antigens associated with ulcerative colitis in Japanese. *Tissue Antigens* 2001; **58**: 71-76
- 45 **Powrie F**, Leach MW, Mauze S, Caddle LB, Coffman RL. Phenotypically distinct subsets of CD4<sup>+</sup> T cells induce or protect from chronic intestinal inflammation in C. B-17 scid mice. *Int Immunol* 1993; **5**: 1461-1471
- 46 **Stokkers PC**, Reitsma PH, Tytgat GN, van Deventer SJ. HLA-DR and -DQ phenotypes in inflammatory bowel disease: a meta-analysis. *Gut* 1999; **45**: 395-401
- 47 **Yamamoto-Furusho JK**, Uscanga LF, Vargas-Alarcón G, Ruiz-Morales JA, Higuera L, Cutiño T, Rodríguez-Pérez JM, Villarreal-Garza C, Granados J. Clinical and genetic heterogeneity in Mexican patients with ulcerative colitis. *Hum Immunol* 2003; **64**: 119-123
- 48 **Murch SH**, Lamkin VA, Savage MO, Walker-Smith JA, MacDonald TT. Serum concentrations of tumour necrosis factor alpha in childhood chronic inflammatory bowel disease. *Gut* 1991; **32**: 913-917
- 49 **Reimund JM**, Wittersheim C, Dumont S, Muller CD, Baumann R, Poindron P, Duclos B. Mucosal inflammatory cytokine production by intestinal biopsies in patients with ulcerative colitis and Crohn's disease. *J Clin Immunol* 1996; **16**: 144-150
- 50 **Bouma G**, Xia B, Crusius JB, Bioque G, Koutroubakis I, Von Blomberg BM, Meuwissen SG, Peña AS. Distribution of four polymorphisms in the tumour necrosis factor (TNF) genes in patients with inflammatory bowel disease (IBD). *Clin Exp Immunol* 1996; **103**: 391-396
- 51 **Louis E**, Satsangi J, Roussomoustakaki M, Parkes M, Fanning G, Welsh K, Jewell D. Cytokine gene polymorphisms in inflammatory bowel disease. *Gut* 1996; **39**: 705-710
- 52 **Bouma G**, Crusius JB, García-González MA, Meijer BU, Hellemans HP, Hakvoort RJ, Schreuder GM, Kostense PJ, Meuwissen SG, Peña AS. Genetic markers in clinically well defined patients with ulcerative colitis (UC). *Clin Exp Immunol* 1999; **115**: 294-300
- 53 **Yamamoto-Furusho JK**, Uscanga LF, Vargas-Alarcón G, Rodríguez-Pérez JM, Zuñiga J, Granados J. Polymorphisms in the promoter region of tumor necrosis factor alpha (TNF-alpha) and the HLA-DRB1 locus in Mexican mestizo patients with ulcerative colitis. *Immunol Lett* 2004; **95**: 31-35
- 54 **Yamamoto-Furusho JK**, Cantú C, Vargas-Alarcón G, Andrade F, Zuñiga J, Rodriguez J, Boom R, Granados J. Complotype SC30 is associated with susceptibility to develop ulcerative colitis in Mexicans. *J Clin Gastroenterol* 1998; **27**: 178-179
- 55 **Duerr RH**, Barmada MM, Zhang L, Pfützer R, Weeks DE. High-density genome scan in Crohn disease shows confirmed linkage to chromosome 14q11-12. *Am J Hum Genet* 2000; **66**: 1857-1862
- 56 **Vermeire S**, Rutgeerts P, Van Steen K, Joossens S, Claessens G, Pierik M, Peeters M, Vlietinck R. Genome wide scan in a Flemish inflammatory bowel disease population: support for the IBD4 locus, population heterogeneity, and epistasis. *Gut* 2004; **53**: 980-986
- 57 **Owyang AM**, Zaph C, Wilson EH, Guild KJ, McClanahan T, Miller HR, Cua DJ, Goldschmidt M, Hunter CA, Kastelein RA, Artis D. Interleukin 25 regulates type 2 cytokine-dependent immunity and limits chronic inflammation in the gastrointestinal tract. *J Exp Med* 2006; **203**: 843-849
- 58 **McGovern DP**, Van Heel DA, Negro K, Ahmad T, Jewell DP. Further evidence of IBD5/CARD15 (NOD2) epistasis in the susceptibility to ulcerative colitis. *Am J Hum Genet* 2003; **73**: 1465-1466
- 59 **Rioux JD**, Silverberg MS, Daly MJ, Steinhart AH, McLeod RS, Griffiths AM, Green T, Brettin TS, Stone V, Bull SB, Bitton A, Williams CN, Greenberg GR, Cohen Z, Lander ES, Hudson TJ, Siminovitch KA. Genomewide search in Canadian families with inflammatory bowel disease reveals two novel susceptibility loci. *Am J Hum Genet* 2000; **66**: 1863-1870
- 60 **Armuzzi A**, Ahmad T, Ling KL, de Silva A, Cullen S, van Heel D, Orchard TR, Welsh KI, Marshall SE, Jewell DP. Genotype-phenotype analysis of the Crohn's disease susceptibility haplotype on chromosome 5q31. *Gut* 2003; **52**: 1133-1139
- 61 **Vermeire S**, Pierik M, Hlavaty T, Claessens G, van Schuerbeeck N, Joossens S, Ferrante M, Henckaerts L, Bueno de Mesquita M, Vlietinck R, Rutgeerts P. Association of organic cation transporter risk haplotype with perianal penetrating Crohn's disease but not with susceptibility to IBD. *Gastroenterology* 2005; **129**: 1845-1853
- 62 **Walters TD**, Silverberg MS. Genetics of inflammatory bowel disease: current status and future directions. *Can J Gastroenterol* 2006; **20**: 633-639
- 63 **Giallourakis C**, Stoll M, Miller K, Hampe J, Lander ES, Daly MJ, Schreiber S, Rioux JD. IBD5 is a general risk factor for inflammatory bowel disease: replication of association with Crohn disease and identification of a novel association with ulcerative colitis. *Am J Hum Genet* 2003; **73**: 205-211
- 64 **Török HP**, Glas J, Tonenchi L, Lohse P, Müller-Myhsok B, Limbersky O, Neugebauer C, Schnitzler F, Seiderer J, Tillack C, Brand S, Brünner G, Jagiello P, Epplen JT, Griga T, Klein W, Schiemann U, Folwaczny M, Ochsenkühn T, Folwaczny C. Polymorphisms in the DLG5 and OCTN cation transporter genes in Crohn's disease. *Gut* 2005; **54**: 1421-1427
- 65 **Noble CL**, Nimmo ER, Drummond H, Ho GT, Tenesa A, Smith L, Anderson N, Arnott ID, Satsangi J. The contribution of OCTN1/2 variants within the IBD5 locus to disease susceptibility and severity in Crohn's disease. *Gastroenterology* 2005; **129**: 1854-1864
- 66 **Russell RK**, Drummond HE, Nimmo ER, Anderson NH, Noble CL, Wilson DC, Gillett PM, McGrogan P, Hassan K, Weaver LT, Bisset WM, Mahdi G, Satsangi J. Analysis of the influence of OCTN1/2 variants within the IBD5 locus on disease susceptibility and growth indices in early onset inflammatory bowel disease. *Gut* 2006; **55**: 1114-1123
- 67 **Pelteková VD**, Wintle RF, Rubin LA, Amos CI, Huang Q, Gu X, Newman B, Van Oene M, Cescon D, Greenberg G, Griffiths AM, St George-Hyslop PH, Siminovitch KA. Functional variants of OCTN cation transporter genes are associated with Crohn disease. *Nat Genet* 2004; **36**: 471-475
- 68 **Newman B**, Gu X, Wintle R, Cescon D, Yazdanpanah M, Liu X, Peltekova V, Van Oene M, Amos CI, Siminovitch KA. A risk haplotype in the Solute Carrier Family 22A4/22A5 gene cluster influences phenotypic expression of Crohn's disease. *Gastroenterology* 2005; **128**: 260-269
- 69 **Mirza MM**, Fisher SA, King K, Cuthbert AP, Hampe J, Sanderson J, Mansfield J, Donaldson P, Macpherson AJ, Forbes A, Schreiber S, Lewis CM, Mathew CG. Genetic evidence for interaction of the 5q31 cytokine locus and the CARD15 gene in Crohn disease. *Am J Hum Genet* 2003; **72**: 1018-1022
- 70 **Peeters M**, Geypens B, Claus D, Nevens H, Ghooys Y, Verbeke G, Baert F, Vermeire S, Vlietinck R, Rutgeerts P. Clustering of increased small intestinal permeability in families with Crohn's disease. *Gastroenterology* 1997; **113**: 802-807
- 71 **Stoll M**, Corneliussen B, Costello CM, Waetzig GH, Mellgard B, Koch WA, Rosenstiel P, Albrecht M, Croucher PJ, Seegert D, Nikolaus S, Hampe J, Lengauer T, Pierrou S, Foelsch UR, Mathew CG, Lagerstrom-Fermer M, Schreiber S. Genetic variation in DLG5 is associated with inflammatory bowel disease. *Nat Genet* 2004; **36**: 476-480
- 72 **Daly MJ**, Pearce AV, Farwell L, Fisher SA, Latiano A, Prescott NJ, Forbes A, Mansfield J, Sanderson J, Langelier D, Cohen A, Bitton A, Wild G, Lewis CM, Annesse V, Mathew CG, Rioux JD. Association of DLG5 R30Q variant with inflammatory bowel disease. *Eur J Hum Genet* 2005; **13**: 835-839
- 73 **Friedrichs F**, Brescianini S, Annesse V, Latiano A, Berger K, Kugathasan S, Broeckel U, Nikolaus S, Daly MJ, Schreiber S, Rioux JD, Stoll M. Evidence of transmission ratio distortion of DLG5 R30Q variant in general and implication of an association with Crohn disease in men. *Hum Genet* 2006; **119**: 305-311
- 74 **Bodor M**, Kelly EJ, Ho RJ. Characterization of the human MDR1 gene. *AAPS J* 2005; **7**: E1-E5
- 75 **Brant SR**, Panhuysen CI, Nicolae D, Reddy DM, Bonen DK, Karaliukas R, Zhang L, Swanson E, Datta LW, Moran T,

- Ravenhill G, Duerr RH, Achkar JP, Karban AS, Cho JH. MDR1 Ala893 polymorphism is associated with inflammatory bowel disease. *Am J Hum Genet* 2003; **73**: 1282-1292
- 76 **Glas J, Török HP, Schiemann U, Folwaczny C.** MDR1 gene polymorphism in ulcerative colitis. *Gastroenterology* 2004; **126**: 367
- 77 **Ho GT, Nimmo ER, Tenesa A, Fennell J, Drummond H, Mowat C, Arnott ID, Satsangi J.** Allelic variations of the multidrug resistance gene determine susceptibility and disease behavior in ulcerative colitis. *Gastroenterology* 2005; **128**: 288-296
- 78 **Wang D, Sadée W.** Searching for polymorphisms that affect gene expression and mRNA processing: example ABCB1 (MDR1). *AAPS J* 2006; **8**: E515-E520
- 79 **Farrell RJ, Kelleher D.** Glucocorticoid resistance in inflammatory bowel disease. *J Endocrinol* 2003; **178**: 339-346
- 80 **Cucchiara S, Latiano A, Palmieri O, Canani RB, D'Incà R, Guariso G, Vieni G, De Venuto D, Riegler G, De'Angelis GL, Guagnozzi D, Bascietto C, Miele E, Valvano MR, Bossa F, Annese V.** Polymorphisms of tumor necrosis factor- $\alpha$  but not MDR1 influence response to medical therapy in pediatric-onset inflammatory bowel disease. *J Pediatr Gastroenterol Nutr* 2007; **44**: 171-179
- 81 **McAlindon ME, Mahida YR.** Pro-inflammatory cytokines in inflammatory bowel disease. *Aliment Pharmacol Ther* 1996; **10** Suppl 2: 72-74
- 82 **Mahida YR, Kurlac L, Gallagher A, Hawkey CJ.** High circulating concentrations of interleukin-6 in active Crohn's disease but not ulcerative colitis. *Gut* 1991; **32**: 1531-1534
- 83 **Ma Y, Ohmen JD, Li Z, Bentley LG, McElree C, Pressman S, Targan SR, Fischel-Ghodsian N, Rotter JL, Yang H.** A genome-wide search identifies potential new susceptibility loci for Crohn's disease. *Inflamm Bowel Dis* 1999; **5**: 271-278
- 84 **Low JH, Williams FA, Yang X, Cullen S, Colley J, Ling KL, Armuzzi A, Ahmad T, Neville MJ, Dechairo BM, Walton R, Lench NJ, Jewell DP.** Inflammatory bowel disease is linked to 19p13 and associated with ICAM-1. *Inflamm Bowel Dis* 2004; **10**: 173-181
- 85 **Marek A, Brodzicki J, Liberek A, Korzon M.** TGF- $\beta$  (transforming growth factor- $\beta$ ) in chronic inflammatory conditions - a new diagnostic and prognostic marker? *Med Sci Monit* 2002; **8**: RA145-RA151
- 86 **Yamagiwa S, Gray JD, Hashimoto S, Horwitz DA.** A role for TGF- $\beta$  in the generation and expansion of CD4+CD25+ regulatory T cells from human peripheral blood. *J Immunol* 2001; **166**: 7282-7289
- 87 **Lee YJ, Han Y, Lu HT, Nguyen V, Qin H, Howe PH, Hocevar BA, Boss JM, Ransohoff RM, Benveniste EN.** TGF- $\beta$  suppresses IFN- $\gamma$  induction of class II MHC gene expression by inhibiting class II transactivator messenger RNA expression. *J Immunol* 1997; **158**: 2065-2075
- 88 **McKaig BC, Hughes K, Tighe PJ, Mahida YR.** Differential expression of TGF- $\beta$  isoforms by normal and inflammatory bowel disease intestinal myofibroblasts. *Am J Physiol Cell Physiol* 2002; **282**: C172-C182
- 89 **Fell JM, Paintin M, Arnaud-Battandier F, Beattie RM, Hollis A, Kitching P, Donnet-Hughes A, MacDonald TT, Walker-Smith JA.** Mucosal healing and a fall in mucosal pro-inflammatory cytokine mRNA induced by a specific oral polymeric diet in paediatric Crohn's disease. *Aliment Pharmacol Ther* 2000; **14**: 281-289
- 90 **Brant SR, Shugart YY.** Inflammatory bowel disease gene hunting by linkage analysis: rationale, methodology, and present status of the field. *Inflamm Bowel Dis* 2004; **10**: 300-311
- 91 **Satsangi J, Parkes M, Louis E, Hashimoto L, Kato N, Welsh K, Terwilliger JD, Lathrop GM, Bell JL, Jewell DP.** Two stage genome-wide search in inflammatory bowel disease provides evidence for susceptibility loci on chromosomes 3, 7 and 12. *Nat Genet* 1996; **14**: 199-202
- 92 **Rector A, Vermeire S, Thoelen I, Keyaerts E, Struyf F, Vlietinck R, Rutgeerts P, Van Ranst M.** Analysis of the CC chemokine receptor 5 (CCR5) delta-32 polymorphism in inflammatory bowel disease. *Hum Genet* 2001; **108**: 190-193
- 93 **De Jager PL, Franchimont D, Waliszewska A, Bitton A, Cohen A, Langelier D, Belaiche J, Vermeire S, Farwell L, Goris A, Libioulle C, Jani N, Dassopoulos T, Bromfield GP, Dubois B, Cho JH, Brant SR, Duerr RH, Yang H, Rotter JL, Silverberg MS, Steinhart AH, Daly MJ, Podolsky DK, Louis E, Hafler DA, Rioux JD.** The role of the Toll receptor pathway in susceptibility to inflammatory bowel diseases. *Genes Immun* 2007; **8**: 387-397
- 94 **Melmed G, Thomas LS, Lee N, Tesfay SY, Lukasek K, Michelsen KS, Zhou Y, Hu B, Arditi M, Abreu MT.** Human intestinal epithelial cells are broadly unresponsive to Toll-like receptor 2-dependent bacterial ligands: implications for host-microbial interactions in the gut. *J Immunol* 2003; **170**: 1406-1415
- 95 **Strober W, Lúdvíksson BR, Fuss IJ.** The pathogenesis of mucosal inflammation in murine models of inflammatory bowel disease and Crohn disease. *Ann Intern Med* 1998; **128**: 848-856
- 96 **Kopp EB, Medzhitov R.** The Toll-receptor family and control of innate immunity. *Curr Opin Immunol* 1999; **11**: 13-18
- 97 **Ouburg S, Mallant-Hent R, Crusius JB, van Bodegraven AA, Mulder CJ, Linskens R, Peña AS, Morrè SA.** The toll-like receptor 4 (TLR4) Asp299Gly polymorphism is associated with colonic localisation of Crohn's disease without a major role for the *Saccharomyces cerevisiae* mannan-LBP-CD14-TLR4 pathway. *Gut* 2005; **54**: 439-440
- 98 **Török HP, Glas J, Tonenchi L, Mussack T, Folwaczny C.** Polymorphisms of the lipopolysaccharide-signaling complex in inflammatory bowel disease: association of a mutation in the Toll-like receptor 4 gene with ulcerative colitis. *Clin Immunol* 2004; **112**: 85-91
- 99 **Fort MM, Mozaffarian A, Stöver AG, Correia Jda S, Johnson DA, Crane RT, Ulevitch RJ, Persing DH, Bielefeldt-Ohmann H, Probst P, Jeffery E, Fling SP, Hershberg RM.** A synthetic TLR4 antagonist has anti-inflammatory effects in two murine models of inflammatory bowel disease. *J Immunol* 2005; **174**: 6416-6423
- 100 **Pierik M, Joossens S, Van Steen K, Van Schuerbeek N, Vlietinck R, Rutgeerts P, Vermeire S.** Toll-like receptor-1, -2, and -6 polymorphisms influence disease extension in inflammatory bowel diseases. *Inflamm Bowel Dis* 2006; **12**: 1-8
- 101 **Franchimont D, Vermeire S, El Housni H, Pierik M, Van Steen K, Gustot T, Quertinmont E, Abramowicz M, Van Gossum A, Devière J, Rutgeerts P.** 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* 2004; **53**: 987-992
- 102 **Gazouli M, Mantzaris G, Kotsinas A, Zacharatos P, Papalambros E, Archimandritis A, Ikononopoulos J, Gorgoulis VG.** Association between polymorphisms in the Toll-like receptor 4, CD14, and CARD15/NOD2 and inflammatory bowel disease in the Greek population. *World J Gastroenterol* 2005; **11**: 681-685
- 103 **Schwandner R, Dziarski R, Wesche H, Rothe M, Kirschning CJ.** Peptidoglycan- and lipoteichoic acid-induced cell activation is mediated by toll-like receptor 2. *J Biol Chem* 1999; **274**: 17406-17409
- 104 **Takeuchi O, Hoshino K, Kawai T, Sanjo H, Takada H, Ogawa T, Takeda K, Akira S.** Differential roles of TLR2 and TLR4 in recognition of gram-negative and gram-positive bacterial cell wall components. *Immunity* 1999; **11**: 443-451
- 105 **Yoshimura A, Lien E, Ingalls RR, Tuomanen E, Dziarski R, Golenbock D.** Cutting edge: recognition of Gram-positive bacterial cell wall components by the innate immune system occurs via Toll-like receptor 2. *J Immunol* 1999; **163**: 1-5
- 106 **Lien E, Sellati TJ, Yoshimura A, Flo TH, Rawadi G, Finberg RW, Carroll JD, Espevik T, Ingalls RR, Radolf JD, Golenbock DT.** Toll-like receptor 2 functions as a pattern recognition receptor for diverse bacterial products. *J Biol Chem* 1999; **274**: 33419-33425
- 107 **Opitz B, Schröder NW, Spreitzer I, Michelsen KS, Kirschning CJ, Hallatschek W, Zähringer U, Hartung T, Göbel UB, Schumann RR.** Toll-like receptor-2 mediates *Treponema glycolipid* and lipoteichoic acid-induced NF- $\kappa$ B

- translocation. *J Biol Chem* 2001; **276**: 22041-22047
- 108 **Means TK**, Jones BW, Schromm AB, Shurtleff BA, Smith JA, Keane J, Golenbock DT, Vogel SN, Fenton MJ. Differential effects of a Toll-like receptor antagonist on Mycobacterium tuberculosis-induced macrophage responses. *J Immunol* 2001; **166**: 4074-4082
  - 109 **Jones BW**, Means TK, Heldwein KA, Keen MA, Hill PJ, Belisle JT, Fenton MJ. Different Toll-like receptor agonists induce distinct macrophage responses. *J Leukoc Biol* 2001; **69**: 1036-1044
  - 110 **Ozinsky A**, Smith KD, Hume D, Underhill DM. Co-operative induction of pro-inflammatory signaling by Toll-like receptors. *J Endotoxin Res* 2000; **6**: 393-396
  - 111 **Hajjar AM**, O'Mahony DS, Ozinsky A, Underhill DM, Aderem A, Klebanoff SJ, Wilson CB. Cutting edge: functional interactions between toll-like receptor (TLR) 2 and TLR1 or TLR6 in response to phenol-soluble modulin. *J Immunol* 2001; **166**: 15-19
  - 112 **Hayashi F**, Smith KD, Ozinsky A, Hawn TR, Yi EC, Goodlett DR, Eng JK, Akira S, Underhill DM, Aderem A. The innate immune response to bacterial flagellin is mediated by Toll-like receptor 5. *Nature* 2001; **410**: 1099-1103
  - 113 **Lodes MJ**, Cong Y, Elson CO, Mohamath R, Landers CJ, Targan SR, Fort M, Hersherberg RM. Bacterial flagellin is a dominant antigen in Crohn disease. *J Clin Invest* 2004; **113**: 1296-1306
  - 114 **Netea MG**, Ferwerda G, de Jong DJ, Jansen T, Jacobs L, Kramer M, Naber TH, Drenth JP, Girardin SE, Kullberg BJ, Adema GJ, Van der Meer JW. Nucleotide-binding oligomerization domain-2 modulates specific TLR pathways for the induction of cytokine release. *J Immunol* 2005; **174**: 6518-6523
  - 115 **Gewirtz AT**, Vijay-Kumar M, Brant SR, Duerr RH, Nicolae DL, Cho JH. Dominant-negative TLR5 polymorphism reduces adaptive immune response to flagellin and negatively associates with Crohn's disease. *Am J Physiol Gastrointest Liver Physiol* 2006; **290**: G1157-G1163
  - 116 **McGovern DP**, Hysi P, Ahmad T, van Heel DA, Moffatt MF, Carey A, Cookson WO, Jewell DP. Association between a complex insertion/deletion polymorphism in NOD1 (CARD4) and susceptibility to inflammatory bowel disease. *Hum Mol Genet* 2005; **14**: 1245-1250
  - 117 **Iwasaki A**, Kelsall BL. Localization of distinct Peyer's patch dendritic cell subsets and their recruitment by chemokines macrophage inflammatory protein (MIP)-3alpha, MIP-3beta, and secondary lymphoid organ chemokine. *J Exp Med* 2000; **191**: 1381-1394
  - 118 **Puleston J**, Cooper M, Murch S, Bid K, Makh S, Ashwood P, Bingham AH, Green H, Moss P, Dhillon A, Morris R, Strobel S, Gelinas R, Pounder RE, Platt A. A distinct subset of chemokines dominates the mucosal chemokine response in inflammatory bowel disease. *Aliment Pharmacol Ther* 2005; **21**: 109-120
  - 119 **Lee HJ**, Choi SC, Lee MH, Oh HM, Choi EY, Choi EJ, Yun KJ, Seo GS, Kim SW, Lee JG, Han WC, Park KI, Jun CD. Increased expression of MIP-3alpha/CCL20 in peripheral blood mononuclear cells from patients with ulcerative colitis and its down-regulation by sulfasalazine and glucocorticoid treatment. *Inflamm Bowel Dis* 2005; **11**: 1070-1079
  - 120 **Klein W**, Tromm A, Griga T, Fricke H, Folwaczny C, Hocke M, Eitner K, Marx M, Duerig N, Eppelen JT. A polymorphism in the IL11 gene is associated with ulcerative colitis. *Genes Immun* 2002; **3**: 494-496
  - 121 **Takagawa T**, Tamura K, Takeda N, Tomita T, Ohda Y, Fukunaga K, Hida N, Ohnishi K, Hori K, Kosaka T, Fukuda Y, Ikeuchi H, Yamamura T, Miwa H, Matsumoto T. Association between IL-18 gene promoter polymorphisms and inflammatory bowel disease in a Japanese population. *Inflamm Bowel Dis* 2005; **11**: 1038-1043
  - 122 **Berg DJ**, Davidson N, Kühn R, Müller W, Menon S, Holland G, Thompson-Snipes L, Leach MW, Rennick D. Enterocolitis and colon cancer in interleukin-10-deficient mice are associated with aberrant cytokine production and CD4(+) TH1-like responses. *J Clin Invest* 1996; **98**: 1010-1020
  - 123 **Davidson NJ**, Hudak SA, Lesley RE, Menon S, Leach MW, Rennick DM. IL-12, but not IFN-gamma, plays a major role in sustaining the chronic phase of colitis in IL-10-deficient mice. *J Immunol* 1998; **161**: 3143-3149
  - 124 **Neurath MF**, Fuss I, Kelsall BL, Stüber E, Strober W. Antibodies to interleukin 12 abrogate established experimental colitis in mice. *J Exp Med* 1995; **182**: 1281-1290
  - 125 **Cua DJ**, Sherlock J, Chen Y, Murphy CA, Joyce B, Seymour B, Lucian L, To W, Kwan S, Churakova T, Zurawski S, Wiekowski M, Lira SA, Gorman D, Kastelein RA, Sedgwick JD. Interleukin-23 rather than interleukin-12 is the critical cytokine for autoimmune inflammation of the brain. *Nature* 2003; **421**: 744-748
  - 126 **Murphy CA**, Langrish CL, Chen Y, Blumenschein W, McClanahan T, Kastelein RA, Sedgwick JD, Cua DJ. Divergent pro- and antiinflammatory roles for IL-23 and IL-12 in joint autoimmune inflammation. *J Exp Med* 2003; **198**: 1951-1957
  - 127 **Langrish CL**, Chen Y, Blumenschein WM, Mattson J, Basham B, Sedgwick JD, McClanahan T, Kastelein RA, Cua DJ. IL-23 drives a pathogenic T cell population that induces autoimmune inflammation. *J Exp Med* 2005; **201**: 233-240
  - 128 **Fossiez F**, Banchereau J, Murray R, Van Kooten C, Garrone P, Lebecque S. Interleukin-17. *Int Rev Immunol* 1998; **16**: 541-551
  - 129 **Oppmann B**, Lesley R, Blom B, Timans JC, Xu Y, Hunte B, Vega F, Yu N, Wang J, Singh K, Zonin F, Vaisberg E, Churakova T, Liu M, Gorman D, Wagner J, Zurawski S, Liu Y, Abrams JS, Moore KW, Rennick D, de Waal-Malefyt R, Hannum C, Bazan JF, Kastelein RA. Novel p19 protein engages IL-12p40 to form a cytokine, IL-23, with biological activities similar as well as distinct from IL-12. *Immunity* 2000; **13**: 715-725
  - 130 **Trinchieri G**. Proinflammatory and immunoregulatory functions of interleukin-12. *Int Rev Immunol* 1998; **16**: 365-396
  - 131 **Becker C**, Wirtz S, Blessing M, Pirhonen J, Strand D, Bechthold O, Frick J, Galle PR, Autenrieth I, Neurath MF. Constitutive p40 promoter activation and IL-23 production in the terminal ileum mediated by dendritic cells. *J Clin Invest* 2003; **112**: 693-706
  - 132 **Yen D**, Cheung J, Scheerens H, Poulet F, McClanahan T, McKenzie B, Kleinschek MA, Owyang A, Mattson J, Blumenschein W, Murphy E, Sathe M, Cua DJ, Kastelein RA, Rennick D. IL-23 is essential for T cell-mediated colitis and promotes inflammation via IL-17 and IL-6. *J Clin Invest* 2006; **116**: 1310-1316

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