Animal Models of Inflammatory Bowel Diseases: Illuminating the Pathogenesis of Colitis, Ileitis and Cancer

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Key Words

Colitis • Ileitis • T cells • Inflammatory bowel diseases • Cytokines

Abstract

Background/Aims: Inflammatory bowel diseases (IBD) are chronic relapsing diseases of unknown origin. In spite of improved options for therapy, many patients with IBD have an impaired quality of life and require hospitalization or surgery. Animal models of IBD might help to obtain new insights into the pathogenesis of these diseases and may be used to test innovative approaches for therapy. **Methods:** Review of the literature using PubMed. **Results:** Numerous new animal models of IBD have been developed in recent years. Although none of these models provides a full reflection of the IBD pathogenesis in humans, they have provided fascinating new insights into the pathogenesis of gut inflammation. In addition to models of acute and chronic colitis, recent studies have described new models of chronic ileitis and IBD-associated cancer. Furthermore, various new approaches for IBD therapy have been developed in animal models, including strategies against proinflammatory cytokines, signaling pathways and adhesion molecules. **Conclusion:** New animal models of IBD have illuminated some aspects of IBD pathogenesis and suggest new avenues for therapy of IBD.

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Inflammatory bowel diseases (IBD) are idiopathic chronic disorders of the intestine. Patients with IBD suffer from abdominal cramping, diarrhea, rectal bleeding and weight loss. These complaints are due to local intestinal inflammation [1]. While inflammation in ulcerative colitis is restricted to the large bowel (continuous inflammation), inflammation in Crohn's disease can occur anywhere in the intestinal tract (discontinuous inflammation). Additional differences between Crohn's disease and ulcerative colitis can be found at the histologic level. Crohn's disease is characterized by a transmural and sometimes granulomatous inflammation. In contrast, inflammation in ulcerative colitis usually affects mucosal and submucosal layers only. However, some histopathological features are similar in both diseases, including edema, loss of goblet cells, hyperplasia of crypt cells and ulcerations. Complications of IBD include fistula formation, abscess development, stenoses and cancer. In spite of improved therapeutic options in recent years, it is not surprising that many patients with IBD have to undergo surgery due to complications of the disease.

An important limitation in understanding the pathophysiology of IBD is the availability of suitable animal models [2]. However, in recent years a large number of animal models have been developed [3–10]. These models have greatly advanced our knowledge about the

pathogenesis of acute and chronic intestinal inflammation. In fact, there are currently more than 50 animal models of IBD available (see table 1 for selected animal models of IBD). Schematically, animal models of IBD can be classified into several categories based on the mode of induction of gut inflammation. In some models, such as the cotton-top tamarin model, spontaneous mutations cause chronic gut inflammation. In addition, numerous studies have used inducible models of colitis where agents such as DSS, TNBS, oxazolone or PG-PS are given to mice. These models have the advantage of causing inflammation in inbred mouse strains with a normal immune system. Furthermore, there are adoptive transfer models where CD4+ or CD8+ T cells are adoptively transferred to immunocompromised mouse strains such as RAG or SCID knockout mice. The prototypical model in this group is the so-called CD4+CD45Rbhigh transfer model in which naïve T cells are given to immunodeficient hosts. This model has the advantage of high reproducibility of T cell-dependent chronic intestinal inflammation. Finally, there are genetically engineered models of IBD including transgenic mice, conditional knockout mice and general deleter mice. Examples consist of IL-7 and STAT4 transgenic mice, LysMCre STAT3 knockout mice, and IL-10 knockout mice. Depending on the penetrance of the phenotype, these models can be reliably used to assess the development of gut inflammation and to study the effects of therapeutic interventions.

Several key principles of IBD pathogenesis have been discovered by studying animal models [2]. For instance, it was found that T cells are the key players controlling the development of chronic intestinal inflammation. Deficiency of CD4+ T cells or targeting of CD4+ T cell function have been used to prevent or treat chronic intestinal inflammation in several animal models, thus highlighting a role of T cells in disease pathogenesis. In addition, it was found that the commensal bacterial flora drives intestinal inflammation and inflammation was abrogated in several models when mice were raised under germ-free conditions. Furthermore, alterations of the intestinal barrier function appear to be important in mediating chronic intestinal inflammation. It is believed that such alterations of barrier function permit translocation of commensal bacteria into the bowel wall where bacterial antigens are taken up by professional antigen-presenting cells followed by antigen presentation to T cells and T cell-mediated gut inflammation. Furthermore, animal models of IBD have highlighted that genetic alterations may strongly influence the development of chronic intestinal inflammation, as shown for instance by studies in various knockout models of colitis. These findings suggest that many different roads exist that predispose to the development of chronic intestinal inflammation. Along those lines, IBD might be considered as a final common clinical pathway of inflammation that is triggered initially by various different bacteria in a genetically susceptible host [1, 2].

Table 1. Selected animal models of IBD

Spontaneous models
Cotton-top tamarin
SAMP1/Yit
C3H/HeJBir mouse substrain
Chemically-induced models
DSS, AOM/DSS
Oxazolone
TNBS (DNBS)
Peptidoglycan polysaccharide
Immune complex
Indomethacin
Carrageenan
Acetic acid
Cyclosporin A
Genetically engineered models (transgenic animals)
STAT4 transgenic (CMV promoter)
IL-7 transgenic (SR promoter)
HLA-B27/β ₂ -microglobulin transgenic
HSV-thymidine kinase (GFAP astroglia-specific promoter)
<i>N-cadherin</i> dominant negative
Gp39 overexpression
<i>TGF-RII</i> dominant negative (villin IEC-specific promoter)
Genetically engineered (knockout mice)
<i>STAT3</i> (<i>LysMCre</i> conditional KO)
<i>TGFβ-1</i>
<i>ITF</i>
<i>IL-2/IL-2R</i>
<i>IL-10</i>
<i>TNF^{DARE}</i>
<i>TCRα/β</i>
<i>Caspase-8</i> (IEC specific)
<i>FADD</i> (IEC specific)
<i>NEMO</i> (IEC specific)
<i>Gai2</i>
<i>Keratin-8</i>
<i>Mdr1α</i>
<i>CRF2-4</i>
<i>WASP</i>
<i>XPB-1</i>
<i>T-bet(-/-)RAG2(-/-)(TRUC)</i>
T cell transfer models
CD45RB-high cells into RAG/SCID mice
CD62L-high cells into RAG/SCID mice
CD45RB cells transfer into Tgε26
Hsp60 CD8+ T cells

In the following paragraphs, some of the most frequently used animal models of IBD and some recently developed models are described [3–10]. Among the inducible models, the dextran sodium sulfate (DSS) model is very frequently used. In this model, mouse strains are given cycles of DSS in the drinking water resulting in acute or chronic intestinal inflammation. While acute DSS colitis may occur in the absence of T cells, chronic DSS colitis is markedly augmented by T lymphocytes. This model is relatively cheap, easy to use and has provided important new insights into IBD pathogenesis. In particular, it was found that the intestinal epithelial cells as part of the intestinal barrier are severely altered in DSS colitis, thus highlighting the role of these cells in IBD pathogenesis. In addition, various new approaches for treatment of IBD have been developed in this model. Finally, several investigators have combined this model with the procarcinogen azoxymethane to induce colitis-associated tumors in the colon. Tumor development has been shown to depend on the presence of T cells and pro-inflammatory cytokines such as IL-6. Thus, based on these results, growth of IBD-associated cancer appears to be driven by the same T cells that cause chronic intestinal inflammation.

A frequently used genetically engineered model consists of the IL-10-deficient (IL-10^{-/-}) mouse strain. It was described that mice with a targeted disruption of the IL-10 gene suffer from spontaneous pancolitis within several months after birth. Importantly, mice raised under germ-free conditions do not develop colitis. The genetic background of mice was identified as modifier of disease activity, as the penetrance and severity of colitis was markedly different between individual mouse strains. Histopathological analysis of these mice revealed chronic inflammation characterized by mononuclear cell infiltration and epithelial hyperplasia. T cells producing pro-inflammatory cytokines were subsequently identified as a key regulator of chronic colitis. Nowadays, the IL-10 knockout model is a well-established murine model of IBD. In this model, a Th1-mediated chronic colitis is observed that can be treated with anti-Th1-targeted therapies such as anti-IFN- γ or anti-IL-12 p40 antibodies. Disadvantages are the low penetrance of the phenotype in some mouse strains and the heterogeneity of disease onset in individual mice.

Among the T cell transfer models, the CD4+ CD45Rb^{high} transfer model is most frequently used. In this model, a subgroup of naive T cells is given to immunodeficient hosts such as RAG or SCID mice that lack T cells. This model has been well characterized in

recent years. It is believed that transferred T cells reach the gut where they are exposed to antigens of the commensal microflora. Subsequently, Th1 and Th17 cell differentiation takes place resulting in accumulation of T-bet and ROR γ T-positive cells, disruption of T cell homeostasis and chronic intestinal inflammation 3–6 weeks after T cell transfer. While the colon is affected in this model, usually little or no inflammation of the small intestine is found. Histopathological assessment of the colon shows superficial or transmural inflammation with epithelial cell hyperplasia, erosions and dense infiltrates of mononuclear leukocytes. The reconstituted mice demonstrate varying degrees of weight loss and diarrhea due to chronic colitis. This model has allowed a precise characterization of cytokines and adhesion molecules as well as of cell types causing gut inflammation. In particular, Th1 and Th17 cells were found to induce disease, while regulatory T cells may suppress intestinal inflammation. Based on these observations, regulatory T cells may be considered as a novel approach for therapy of IBD.

A new group of animal models has been shown to induce chronic inflammation in the small bowel or terminal ileum. Prototypical examples of this group of mouse models consist of the FADD and caspase-8 conditional knockout mice. In these mice, key regulatory proteins involved in the TNF signaling pathway were selectively inactivated in intestinal epithelial cells by homologous recombination and crossing the floxed mice to villin-Cre mice. It was found that mice with a conditional deletion of caspase-8 in intestinal epithelial cells spontaneously develop chronic inflammation in the terminal ileum within weeks after birth and are highly susceptible to colitis. As Crohn's disease most frequently affects the terminal ileum (terminal ileitis), these findings suggest that this mouse strain might provide some clues to the pathogenesis of gut inflammation in this disease. Subsequent studies demonstrated that caspase-8-deficient mice have reduced numbers of goblet cells and lack Paneth cells, indicating altered antimicrobial immune cell functions of the intestinal epithelium. This reduction in cell numbers was caused by induction of Rip3-dependent necroptosis and cell death upon TNF signaling. Importantly, augmented necroptosis of Paneth cells was also noted in the terminal ileum in Crohn's disease, highlighting a key role of necroptosis in the pathogenesis of this disease. Although future studies on the pathogenesis of this disease are required, this model has unequivocally identified a new pathway of gut inflammation with high relevance to IBD in humans.

In summary, numerous studies have defined new animal models of IBD in recent years [2–10]. Although none of the currently available models reflects all aspects of IBD in humans (e.g. spontaneous flares of disease), these models have greatly advanced our understanding of IBD pathogenesis. Moreover, they have led to the identification of novel pathways that are important for the design of new therapies of IBD.

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Disclosure Statement

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