Redeeming an old foe: protective as well as pathophysiological roles for tumor necrosis factor in inflammatory bowel disease

Philip E. Dubé,1,2 Shivesh Punit,1,2 and D. Brent Polk1,2,3

1Saban Research Institute, Children’s Hospital Los Angeles, Los Angeles, California; 2Department of Pediatrics, Children’s Hospital Los Angeles and University of Southern California Keck School of Medicine, Los Angeles, California; and 3Department of Biochemistry and Molecular Biology, University of Southern California Keck School of Medicine, Los Angeles, California

Submitted 11 April 2014; accepted in final form 30 November 2014

Tumor necrosis factor (TNF) is a key regulatory cytokine involved in biological responses ranging from cellular activation and proliferation to cytotoxicity and apoptosis. Recent work has determined that TNF can mediate both beneficial and harmful effects in the gastrointestinal (GI) tract, emphasizing how the physiological role of cytokines can contribute to pathological outcomes. There is a longstanding association between TNF and inflammatory bowel disease (IBD), including Crohn’s disease (CD) and ulcerative colitis (UC), that has led to a multitude of laboratory studies and genome-wide association study approaches. However, the basis for these associations remains uncertain. This review considers the efficacy and mechanism of anti-TNF therapies for inflammatory bowel disease (IBD), including Crohn’s disease (CD) and ulcerative colitis (UC), and its receptors TNFR1 and TNFR2 are major therapeutic targets for inflammatory bowel disease. Research advances have demonstrated that TNF promotes pleiotropic responses in the gastrointestinal (GI) tract. Although in excess TNF can contribute to GI pathology, TNF is also a critical protective factor to promote GI homeostasis following injury and inflammation. Genetic studies using candidate and genome-wide association study approaches have identified variants in TNF or its receptors that are associated with Crohn’s disease or ulcerative colitis in multiple populations. The purpose of this review is to emphasize some of the perplexing insights for TNF coming out of the laboratory and reconcile these with clinical experience.

Address for reprint requests and other correspondence: D. B. Polk, Children’s Hospital Los Angeles, 4650 Sunset Blvd. MS#126, Los Angeles, CA 90027 (e-mail: dbpolk@chla.usc.edu).

The TNF Family

TNF (TNFSF2) is produced as a 26-kDa membrane-bound protein that is subsequently cleaved by TNFα-converting enzyme to generate a soluble 17-kDa protein (41). The membrane-bound (mTNF) and soluble (sTNF) forms of TNF are bioactive and signal through two structurally distinct membrane receptors, TNFR1 (p55/TNFRSF1A) and TNFR2 (p75/TNFRSF1B) (55). TNFR1 and TNFR2 are single-transmembrane glycoproteins, with 28% homology, mostly in their extracellular ligand-binding domains. An important caveat is that most of what we know about the function and signaling capacities of these two receptors is from in vitro systems, and it is not entirely clear how these attributed functions might translate to the in vivo setting in animal models or human patients. Furthermore, in vitro studies have identified TNF concentration-dependent responses that determine receptor selectivity and signaling; however, varying local TNF concentrations in vivo and the contribution of bioactive cell surface mTNF in addition to sTNF confound much of our ability to translate results from in vitro models. As such, many of the contrasting results in this discussion (see below) might reflect differences in experimental systems, and a major obstacle is to determine how TNF or its receptors contribute to physiology or disease in animal or human settings.

Activation of TNFR1 or TNFR2 initiates distinct and overlapping intracellular signaling pathways; an important feature of TNF signaling is that cell fate is determined by the balance between competing pathways downstream of TNFR1 and/or...
TNFR2 (Fig. 1) (5, 11). This balance, which represents a molecular switch for cellular responses, is regulated in a tissue-specific manner and is influenced by disease state and interactions with other cytokines and growth factors. TNFR1-regulated pathways are often considered proapoptotic, involving the adaptor proteins TNFR1-associated death domain and Fas-associated death domain, with subsequent caspase 8 activation (15). Indeed, epithelial TNFR1 expression is both necessary and sufficient to induce intestinal epithelial apoptosis and permeability defects following TNF administration to mice (101). However, paradoxically, TNFR1 can also activate antiapoptotic pathways in GI epithelial cells that can promote cell survival in a context-dependent manner. Numerous pathways are involved in this molecular switch. For example, we have shown that TNFR1-mediated transactivation of epidermal growth factor receptor and activation of kinase suppressor of Ras promote TNFR1-induced cell survival signals in the GI epithelium (38, 108–110). Importantly, a key mechanism in the switch between apoptosis and survival is the cellular compartmentalization of TNFR1: cell surface TNFR1 mediates survival through activation of the NF-κB pathway, whereas endosomal TNFR1 preferentially induces apoptotic pathways (89). Both AP-1- and NF-κB-mediated transcription regulate the prosurvival effects of TNFR1 (21, 44, 54), and, in combination with STAT3, TNFR1 mediates liver regeneration (107).

Similar to TNFR1, TNFR2-mediated pathways also regulate disparate cellular outcomes that may have differential roles in IBD (11). TNFR2 activation enhances proliferation, migration, and wound closure in mouse colonic epithelial cells (17, 43, 61) and proliferation and collagen accumulation in intestinal myofibroblasts (98). Although TNFR2 has been implicated in mediating apoptosis and barrier dysfunction in colonic epithelial cells through a myosin light chain kinase-dependent mechanism (93, 103), it can also signal through the NF-κB and AP-1 pathways to promote cell survival in multiple cell types (11, 22, 75). In immune cells, TNFR2 mediates the activation and proliferation of cytotoxic T cells, thymocytes, and B cells (30, 34, 96). In CD4+ and CD8+ T cells, TNFR2 is a critical costimulator for IL-2 induction and T cell survival. It also enhances antigen-driven T cell responses to T-cell receptor-mediated signaling (47, 48). In contrast, TNFR2 may also serve regulatory and anti-inflammatory roles. TNFR2 regulates the proliferation of regulatory T (Treg) cells and induces their suppressive function and survival (13). Interestingly, TNFR2-specific agonists result in selective apoptosis of autoreactive CD8+ T cells (3). Together, these effects on Treg and CD8+ T cells would be expected to inhibit inflammatory responses.

An Enigmatic Role for TNF in the Pathology of Mouse IBD Models

The first evidence showing a link between TNF and IBD was increased levels of circulating and colonic TNF in children with colonic CD (10, 62). Subsequently, increased levels of

![Diagram of TNF signaling](http://ajpgi.physiology.org/ by 10.220.33.5 on August 5, 2017)
TNF production were observed in CD and UC biopsies (73), and numerous studies have implicated increased TNF in the pathogenesis of IBD. These findings are consistent with the observation that TNF neutralization induces remission in refractory IBD, and, as a result, anti-TNF antibodies have become a mainstay in IBD treatment (77, 95).

Although increased TNF production is associated with multiple different mouse IBD models, it has been difficult to generalize how TNF contributes to disease etiology. Numerous studies have shown a pathogenic role for TNF in animal models of colitis; however, recent data suggest that TNF may be a protective factor for certain aspects of colitis (Table 1). For example, TNF neutralization effectively reduces disease in many studies (7, 29, 49, 52, 71, 87, 90, 91), but it also exacerbates acute injury in the dextran sulfate sodium (DSS) colitis model and is not effective in the IL-10 knockout (IL-10−/−) or oxazolone-induced colitis model (49, 53, 74, 91). A problem with interpreting antibody neutralization results may be indirect effects on cells producing TNF (see below). Interestingly, several studies using TNF knockouts have shown worsening of experimental colitis in the DSS or IL-10−/− model following complete loss of TNF (35, 63, 68). While it is unclear if loss of TNF leads to compensatory changes in other cytokines, there is evidence that TNF administration actually alleviates colitis in oxazolone-treated mice (68). Although these findings, particularly those from acute mucosal damage models, suggest that TNF provides a reparative cue following GI injury or inflammation, the duality of TNF is emphasized by its deleterious role when overexpressed.

To test this idea, the TNFΔARE mouse possesses a deletion in the untranslated regulatory region of TNF mRNA, leading to TNF overproduction and a CD-like phenotype, which is responsive to TNF neutralization (51, 59). While these contradictions may appear puzzling, one must always remember that there are numerous differences between animal models and human disease and that experimental systems are subject to multiple experimental variables that may confound results (100).

Receptor-Dependent Roles for TNF in Mouse IBD Models

The disparate findings for TNF in IBD models may relate to differential receptor-dependent signaling (Table 1). TNFR1 and TNFR2 appear to serve distinct roles in a cell type-specific manner, differentially regulating epithelial- and immune cell-specific responses to injury and inflammation in the GI tract. TNFR1 is ubiquitousy expressed throughout many tissues, including the GI epithelium, mesenchyme, and mucosal immune cells. In contrast, TNFR2 expression is normally restricted to hematopoietic lineages; however, its expression is induced in intestinal epithelial cells during inflammation, in IBD patients, and in murine colitis (61, 103). While loss of TNFR1 in whole body TNFR1−/− mice can ameliorate colitis in mouse models by decreasing epithelial apoptosis (12, 20), other studies have demonstrated a protective and antiapoptotic role for TNFR1 (105). Similarly, although TNFR2 knockout ameliorates disease in some mouse colitis models, TNFR2 can also promote epithelial proliferation and survival (20, 61, 93, 105). To complicate the issue further, an additional report suggests that whole body knockout of TNFR1 or TNFR2 does not affect histopathology or epithelial apoptosis in DSS colitis (92). Nevertheless, TNFR1 is required for disease in the T-bet deficiency colitis model (29). Although certainly confusing, these conflicting data suggest that whole body knockouts of TNFR1 and TNFR2 may be insufficient to determine the precise roles of these receptors in colitis and that a more nuanced approach to investigate the cell type-specific outcomes of TNF signaling is necessary.

Indeed, accumulating evidence suggests that differential cell type-specific roles are critical in determining how TNF influences disease. An intriguing example is from the TNFΔARE mouse model (51). These mice, when crossed to the TNFR1−/− background, develop normally, with no signs of illness. Intriguingly, although gut pathology is only partially attenuated in TNFΔARETNFR2−/− mice, the arthritis phenotype in this model is not affected by loss of TNFR2, suggesting that TNFR2 regulates organ-specific inflammation in response to excess TNF. Since TNFΔARERAG1−/− mice exhibit only minimal gut inflammation, most, but not all, of the pathology in this model likely requires lymphocytes. However, the pathological target of excess TNF in these mice is unclear, since TNFR1/TNFR2 on bone marrow-derived or stromal cells is sufficient to induce disease (50). Moreover, mesenchymal, but not epithelial, TNFR1 is sufficient for disease in TNFΔARE mice, suggesting that TNF acting directly on mesenchymal TNFR1 (e.g., on collagen IV-positive myofibroblasts) is pathogenic, potentially by influencing secondary inflammatory mediators or the epithelial stem cell niche (1, 76). In contrast, the DSS colitis model appears quite different, in that TNFR1 signaling in myeloid cells is protective by indirectly reducing epithelial injury and enhancing restitution, potentially by indirectly activating the mitogen-activated protein kinase and Akt/phosphoinositide-3 kinase pathways in epithelial cells (60). Interestingly, macrophage TNFR1 is required to suppress T cell proliferation and induce macrophage apoptosis; together, these may represent important mechanisms to resolve inflammation in colitis (72, 94). Similarly, TNFR2 may play complex regulatory roles in different immune cell subtypes. Data from adoptive transfer studies using naïve CD4+ T cells to induce colitis in RAG−/− mice show that TNFR2 in CD4+ T cells can be pathogenic (39, 93) or protective (14, 19). The reason behind this discrepancy may involve a differential role for TNFR2 in subsets of differentiated T cells. In particular, TNFR2 stabilizes Treg (FoxP3+ Treg) cells, and TNFR2 in these cells is required for their ability to suppress colitis (14). Furthermore, TNFR2 has the potential to restrict CD8+ T cells in colitis (97); however, this possibility has not been specifically tested. One problem is the lack of studies that have specifically addressed cell type-specific roles for TNFR1- or TNFR2-dependent physiological or pathobiological roles through directed cell-specific knockouts in colitis. Nevertheless, these data emphasize the importance of various distinct immune and stromal cell type-specific responses to TNF in regulating protective vs. detrimental roles in IBD models.

What Do Anti-TNF Drugs Tell Us About the Pathobiological Role of TNF in IBD?

Anti-TNF monoclonal antibodies have become an integral part of therapeutic regimens to induce and maintain remission in IBD (Table 2) (37, 77, 81, 95). Infliximab (Remicade), a chimeric antibody with 25% murine and 75% human sequences, was the first anti-TNF therapy with proven efficacy in
## Table 1. Differential roles of TNF and its receptors in mouse IBD models

<table>
<thead>
<tr>
<th>Model</th>
<th>Intervention</th>
<th>Effect on Disease Severity</th>
<th>Features</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Mucosal damage models</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>DSS</td>
<td>TNF−/−</td>
<td>↑</td>
<td>DSS-induced inflammation is significantly increased in TNF−/− mice compared with WT control (63)</td>
</tr>
<tr>
<td></td>
<td>TNF neutralization</td>
<td>↑</td>
<td>In acute DSS colitis, TNF neutralization worsens disease (49)</td>
</tr>
<tr>
<td></td>
<td>TNF neutralization</td>
<td>↓</td>
<td>In chronic DSS colitis, TNF neutralization reduces severity of colonic inflammation (49)</td>
</tr>
<tr>
<td></td>
<td>TNF neutralization</td>
<td>↓</td>
<td>Orally bioavailable anti-TNF reduces severity of colonic inflammation (7)</td>
</tr>
<tr>
<td></td>
<td>TNFR1/−/−</td>
<td>↑</td>
<td>TNFR1 is protective in DSS-induced colitis (104)</td>
</tr>
<tr>
<td></td>
<td>TNFR2/−/−</td>
<td>↓</td>
<td>TNFR2 is pathogenic in DSS-induced colitis (104)</td>
</tr>
<tr>
<td></td>
<td>TNFR1/−/− or TNFR2/−/−</td>
<td>↑</td>
<td>TNFR1 and TNFR2 are protective in DSS-induced colitis (105)</td>
</tr>
<tr>
<td></td>
<td>TNFR1/−/− in Rag1−/− mice</td>
<td>↑</td>
<td>TNFR1 in myeloid cells confers protection from disease in Rag1−/− background (60)</td>
</tr>
<tr>
<td></td>
<td>TNFR2/−/− in Rag1−/− mice</td>
<td>↑</td>
<td>TNFR2 in myeloid cells worsens disease in Rag1−/− background (60)</td>
</tr>
<tr>
<td></td>
<td>TNF neutralization</td>
<td>↓</td>
<td>TNF neutralization reduces TNBS-induced intestinal mucosal inflammation (91)</td>
</tr>
<tr>
<td></td>
<td>Transgenic human TNF overexpression</td>
<td>↑</td>
<td>Human TNF overexpression causes colitis in the TNBS model (66)</td>
</tr>
<tr>
<td></td>
<td>TNFR1/−/−</td>
<td>↑</td>
<td>TNFR1 is protective in the TNBS model (20)</td>
</tr>
<tr>
<td></td>
<td>TNFR2/−/−</td>
<td>↓</td>
<td>TNFR2 promotes disease in the TNBS model (20)</td>
</tr>
<tr>
<td></td>
<td>TNFR1/−/−, TNFR2/−/− or TNFR1/−/−, TNFR2/−/−</td>
<td>↓</td>
<td>TNFR1 or TNFR2 deficiency protects against TNBS-induced mucosal damage (112)</td>
</tr>
<tr>
<td></td>
<td>or TNFR1/−/−, TNFR2/−/− or TNFR1/−/−, TNFR2/−/−</td>
<td>↓</td>
<td>or TNFR1, but not TNFR2, deficiency protects against mucosal damage (64)</td>
</tr>
<tr>
<td><strong>Immune defect models</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TNE2ARE mutant mice</td>
<td>TNF overproduction</td>
<td>↑</td>
<td>TNF overproduction causes a Crohn’s-like IBD phenotype (51)</td>
</tr>
<tr>
<td></td>
<td>Myeloid or lymphoid cell-specific TNF overexpression</td>
<td>↑</td>
<td>TNF overproduction in hematopoietic cells is required for disease, acting on TNFR1/ TNFR2 in bone marrow or stromal cells (50)</td>
</tr>
<tr>
<td></td>
<td>Epithelial cell-specific TNF overexpression</td>
<td>↑</td>
<td>Epithelial TNF overproduction is sufficient to induce disease (76)</td>
</tr>
<tr>
<td></td>
<td>Epithelial cell-specific TNFR1 expression</td>
<td>→</td>
<td>TNFR1 on epithelial cells alone is not sufficient to induce disease (76)</td>
</tr>
<tr>
<td></td>
<td>Mesenchymal cell-specific TNFR1 expression</td>
<td>↑</td>
<td>TNFR1 on collagen IV-expressing mesenchymal cells is sufficient to induce disease (1)</td>
</tr>
<tr>
<td>Adoptive transfer of naive CD4+ T cells</td>
<td>TNF neutralization</td>
<td>↓</td>
<td>Chronic administration of anti-TNF is required for therapeutic effect (71)</td>
</tr>
<tr>
<td></td>
<td>Blocking sTNF</td>
<td>→</td>
<td>Blocking mTNF, but not sTNF, ameliorates colitis (69)</td>
</tr>
<tr>
<td></td>
<td>TNFR2−/− in CD4+CD45RBhi T cells</td>
<td>↑</td>
<td>TNFR2-deficient CD4+CD45RBhi T cells worsen colitis (19)</td>
</tr>
<tr>
<td></td>
<td>TNFR2−/− in Treg cells</td>
<td>↑</td>
<td>TNFR2-deficient Treg cells do not protect against colitis (14)</td>
</tr>
<tr>
<td></td>
<td>TNFR2−/− in Treg cells</td>
<td>↑</td>
<td>TNFR2-deficient Treg cells do not protect against colitis (40)</td>
</tr>
<tr>
<td></td>
<td>Transgenic TNFR2 overexpression</td>
<td>↑</td>
<td>TNFR2 overexpression in CD4+CD62L+ T cells promotes colitis (39)</td>
</tr>
<tr>
<td></td>
<td>TNFR1−/− in recipients</td>
<td>↓</td>
<td>Rag1−/− TNFR1−/− recipients of WT CD4+CD45RBhi T cells are not protected from colitis (93)</td>
</tr>
<tr>
<td></td>
<td>TNFR2−/− in recipients</td>
<td>↓</td>
<td>Rag1−/− TNFR2−/− recipients of WT CD4+CD45RBhi T cells are partially protected from colitis (93)</td>
</tr>
<tr>
<td><strong>IL-10−/− mice</strong></td>
<td>TNF neutralization</td>
<td>→</td>
<td>Anti-TNF does not affect disease onset or disease course (74)</td>
</tr>
<tr>
<td></td>
<td>Anti-TNF neutralization</td>
<td>→</td>
<td>Anti-TNF does not affect established disease (53)</td>
</tr>
<tr>
<td></td>
<td>TNF−/−</td>
<td>↑</td>
<td>IL-10−/− TNF−/− mice develop more severe colitis and colitis-associated colorectal cancer (35)</td>
</tr>
<tr>
<td><strong>SAMP/Yit mice</strong></td>
<td>TNF neutralization</td>
<td>↓</td>
<td>Pretreatment with a neutralizing anti-TNF antibody prevents development of intestinal inflammation (52)</td>
</tr>
<tr>
<td><strong>T cell receptor mutant mice</strong></td>
<td>TCR−/−</td>
<td>↓</td>
<td>Reduced disease score and epithelial proliferation in TNFR2−/− mice (61)</td>
</tr>
<tr>
<td><strong>TRUC mice</strong></td>
<td>TNF neutralization</td>
<td>↓</td>
<td>TNF neutralization in TRUC mice prevents inflammation (29)</td>
</tr>
<tr>
<td></td>
<td>TNFR1−/−</td>
<td>↓</td>
<td>TNFR1−/− TRUC mice are protected from inflammation (29)</td>
</tr>
<tr>
<td><strong>Epithelial IKKγ−/−</strong></td>
<td>TNFR1−/−</td>
<td>↓</td>
<td>TNFR1 is required for colonic inflammation (65)</td>
</tr>
<tr>
<td><strong>Infectious models</strong></td>
<td>Citrobacter rodentium infection</td>
<td>↑</td>
<td>Exacerbated bacterial load and colonic pathology in TNFR1−/− mice (33)</td>
</tr>
</tbody>
</table>

↑, Increased severity; ↓, decreased severity; →, no change. DSS, dextran sulfate sodium; IBD, inflammatory bowel disease; mTNF, membrane-bound TNF; sTNF, soluble TNF; TNFR1 and TNFR2, TNF receptors 1 and 2; TNBS, trinitrobenzene sulfonic acid; Treg cell, regulatory T cell.
CD and, subsequently, in UC (77, 95). More recently, fully humanized monoclonal antibodies, such as adalimumab (Humira), golimumab (Simponi), and the polyethylene glycol-conjugated certolizumab pegol (Cimzia), have been introduced to improve on the immunogenicity and pharmacokinetics of infliximab (37, 80, 81). These drugs are highly effective in many patients and are becoming increasingly common tools in the therapeutic arsenal for IBD. However, it is important to note that each has fundamental limitations. In addition to the well-known adverse effects for this class of drugs (e.g., increased susceptibility to infection and malignancies) (46), anti-TNF drugs are ineffective to induce acute remission in a small, but significant, number of IBD patients. The incidence of these primary nonresponders ranges from 10% to 40% for CD and from 10% to 70% for UC in adult patients, depending on the particular study, although recent studies tend to show greater efficacy (6, 26). Furthermore, among those patients who initially respond to anti-TNF therapy, ~30–40% lose this response or become intolerant within the 1st yr (16, 36, 83, 88). Although immunogenicity has been blamed for the loss of efficacy in these secondary nonresponders, switching regimens to fully humanized antibodies has not proven beneficial for many of these patients (23, 84). Thus, currently available anti-TNF agents, while clearly important in the therapeutic arsenal for IBD, do not represent a long-term solution for a subset of IBD patients.

To help address whether the efficacy of anti-TNF drugs implies a direct pathogenic role for TNF in IBD patients, it is important to consider the mechanism of anti-TNF drugs and ask if TNF might contribute to GI physiology or pathophysiology in humans. First, there is a discrepancy between the perceived rationale for anti-TNF therapy (i.e., neutralization of pathological levels of TNF production) and the evidence for the mechanism of anti-TNF efficacy in IBD. Three general mechanisms for anti-TNF therapy have been proposed (Fig. 2, Table 2). Initially, anti-TNF therapeutics were proposed to neutralize sTNF and prevent its actions on target cells at sites of inflammation. However, etanercept (Enbrel), a fusion protein of the TNFR2 extracellular domain and the Fc region of human IgG1, is not effective in IBD, despite its ability to neutralize sTNF (82, 86) and its clear efficacy in rheumatoid arthritis. Furthermore, oneccept, a recombinant sTNFR1, which effectively neutralizes sTNF, is also ineffective in CD (78). In contrast, infliximab binds sTNF, as well as mTNF. This dif-
ference supports the notion that binding to mTNF on the cells responsible for TNF production is critical for efficacy in IBD and suggests that additional mechanisms, such as cell-autonomous reverse signaling through mTNF and blocking paracrine mTNF-TNFR2 interactions, are required for efficacy in IBD (2, 58). Although this remains a matter of some debate (18, 67), solely blocking sTNF appears to be ineffective in IBD. Therefore, in addition to directly blocking TNF effects, anti-TNF antibodies likely act as diverse immunomodulators through the indirect modulation of TNF-independent pathways by target-
ing inflammatory cells that are also responsible for TNF production. Thus it is possible that the clinical success of anti-TNF drugs may not necessarily support a major patho-

genic role for TNF in IBD, since the therapeutic efficacy of these drugs may involve blocking multiple inflammatory pathways in addition to TNF. Intriguingly, case reports from the rheumatology field suggest that TNF may provide protective physiological roles for the GI tract. These reports show that anti-TNF drugs can cause paradoxical GI adverse events resembling some aspects of IBD (9, 25, 27, 31, 99, 113). It is interesting to speculate that such paradoxical events might contribute to the pathology in IBD patients who lose response to anti-TNF therapies, although these effects may relate to biological therapy in general and may not relate specifically to a protective role for TNF. Nevertheless, these reports support the notion that TNF contributes to the maintenance of GI homeostasis in humans, in agreement with some preclinical findings in mice; however, this is clearly a field for further investigation. The duality for TNF to be either protective or harmful in the GI tract indicates that it has a complex role with potentially health- and disease-promoting activities in the intestinal and colonic mucosa.

Perspectives on the Genomic Era of Personalized Medicine

To move forward with the next generation of IBD therapies, it is critical that we understand the intricacies of TNF biology in health and disease. While potent approaches have been developed to block TNF effects, these have largely failed to address the unmet needs of primary or secondary nonrespon-ders. On the basis of preclinical data from cell and mouse models, it is possible that TNF and its receptors may play protective physiological roles, suggesting that there may be limitations in the strategy to inhibit TNF. It is unclear why some patients are refractory to anti-TNF therapies, but it is interesting to speculate that this may relate to specific genetic variants of TNF or its receptors. TNF has been identified by genome-wide association studies as a candidate CD risk gene (4, 28), and specific polymorphisms in the promoter region of TNF have been associated with UC and CD (45, 56, 79, 85, 102, 111). It is important to note that the relative risk for disease with these TNF variants appears low and is likely subject to numerous genetic modifiers, which may include coincident NOD2 mutations (102) and population-specific genetic variation (57). In vitro studies suggest that TNF polymorphisms alter the level of TNF transcription by monocytes or lymphocytes; however, the identified disease-associated TNF alleles can either increase or decrease TNF production, suggesting that there is no generalizable role for the genetic control of TNF production in disease risk (8, 56, 102). These findings are likely complicated by the existence of different combinations of these single-nucleotide polymorphisms (i.e., haplotypes) in IBD patients (79), and there is clearly a need for additional well-controlled studies of TNF variants in diverse populations. Together, these data suggest that genetic control of TNF production might contribute to disease risk in certain genetic backgrounds, although there is clearly a need to more fully understand its mechanistic role in IBD pathology or etiology.

Polymorphisms in TNFR1 and, in particular, TNFR2 have been identified as potential candidate risk factors in IBD in small-scale studies. In a European study, a missense polymor-
phism in TNFR2 (T587G) that promotes TNF-stimulated cytokine production was associated with UC, whereas a missense polymorphism in TNFR1 (A36G) was associated with pancolitis in UC patients (70). The data are somewhat more intriguing for CD, in which a missense TNFR2 polymorphism that promotes TNF-induced cytokine production and cytotoxicity in vitro is associated with reduced disease risk (106). Furthermore, various TNFR2 haplotypes have been associated with either increased or decreased CD risk (24, 85). Perhaps somewhat surprisingly, neither TNFR1 nor TNFR2 has been identi-
ed in large-scale genome-wide association studies for IBD (4, 28, 42). While this might reflect a population bias of the studies, it also suggests that functional modification of IBD risk genes by TNFR1 and/or TNFR2, as opposed to polymor-
phisms in TNFR1/TNFR2 per se, might influence disease risk.

Increasing evidence supports the idea that TNF has protective, as well as pathogenic, roles in the gut and that a nuanced approach to this regulatory cytokine is necessary to guide the next generation of IBD therapies. Recent studies have identi-
ied 163 genetic loci that are associated with IBD, including both unique and overlapping candidate susceptibility genes for CD and UC (42). Along with clinical experience demonstrating numerous IBD subtypes, these genetic data are reshaping our appreciation of the diversity of syndromes, perhaps diseases, within IBD. Instead of a simplistic CD-UC dichotomy, IBD more likely represents a spectrum of numerous disorders with distinct functional and genetic etiologies, and TNF likely contributes divergent regulatory and/or pathobiological roles between these subtypes, in a manner similar to TNF’s varying roles in animal IBD models. A major challenge will be to determine how genetic information or, potentially, other factors [e.g., the microbiome (32)] can inform therapeutic decisions in IBD by identifying patients for whom anti-TNF or alternative therapies are more appropriate and effective. Of course, progress toward this goal is only in its infancy, and a more thorough appreciation of the intricate biological roles for TNF and its receptors in GI physiology and pathophysiology is required to inform the rational development of future therapies.

GRANTS

Work by D. B. Polk has been supported by National Institute of Diabetes and Digestive and Kidney Diseases Grants R01 DK-056008, R01 DK-066176, and R01 DK-54993 and Senior Research Awards from the Crohn’s and Colitis Foundation of America. P. E. Dubé is currently supported by a fellowship from the Crohn’s and Colitis Foundation of America and was formerly supported by a fellowship from the Canadian Institutes of Health Research.

DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the authors.
AUTHOR CONTRIBUTIONS

P.D., S.P., and D.B.P. prepared the figures; P.D., S.P., and D.B.P. drafted the manuscript; P.D., S.P., and D.B.P. edited and revised the manuscript; D.B.P. approved the final version of the manuscript.

REFERENCES


factor receptors (Tnfr) in mouse fibroblasts deficient in Tnfr1 or Tnfr2 are tumor necrosis interaction have shaped the genetic architecture of inflammatory bowel disease: the ACCENT I randomised trial. Gut 59: 1541–1549, 2009.


