The Role of JAM-A in Inflammatory Bowel Disease: Unrevealing the Ties That Bind

Stefania Vetrano and Silvio Danese
Division of Gastroenterology, IRCCS Istituto Clinico Humanitas, Rozzano, Milan, Italy

Tight junctions (TJ) are junctional proteins whose function is to maintain an intact intestinal epithelial barrier and regulate the paracellular movement of water and solutes. Altered TJ structure and epithelial permeability are observed in inflammatory bowel disease and seem to have an important role in the pathogenesis of these diseases. Junctional adhesion molecule-A (JAM-A) is a protein expressed at tight junctions of epithelial and endothelial cells, as well as on circulating leukocytes. Its function at tight junctions appears to be crucial as an extracellular adhesive molecule in the direct regulation of intestinal barrier function. This review focuses on the role of JAM-A in controlling mucosal homeostasis by regulating the integrity and permeability of epithelial barrier function.

Key words: junctional adhesion molecule-A (JAM-A); inflammatory bowel disease (IBD); tight junctions

Introduction

Ulcerative colitis (UC) and Crohn’s disease (CD) are the two major forms of inflammatory bowel disease (IBD). These diseases are complex and multifactorial as environmental, microbial, genetic, and immune dysregulation lead to relapsing acute inflammation and intestinal tissue damage. Although disease pathogenesis remains unclear, it is evident that defective epithelial barrier function observed in IBD plays a crucial role in disease pathogenesis. The intestinal epithelium, composed of a single layer of epithelial cells, not only creates a selective barrier between luminal antigens and tissue, but also regulates the paracellular movement of solutes and the transport of materials through the epithelium. Epithelial cell dysfunction on the one hand contributes to the passage of antigens from the lumen into the intestinal tissue thereby enhancing the inflammatory process, and on the other hand induces paracellular ion and solute permeability. An intact intestinal barrier is, therefore, critical to normal physiological function and prevention of disease. Gross epithelial damage including ulceration and crypt abscesses can compromise the intestinal barrier function as seen in patients with active IBD. Furthermore increased permeability observed in healthy first degree relatives of patients with CD, implies that barrier dysfunction could be a very early event in the disease process, in contrast to UC, where it seems that altered intestinal permeability may be a consequence of the inflammatory events.

The integrity of the epithelial cell layer is maintained by an apical junctional complex (AJC) composed primarily of tight junctions (TJs) and adherens junctions (AJs). Tight junctions are composed of several different transmembrane and intracellular molecules such as occludin, and molecules pertaining to the claudin family, junctional adhesion molecule family (JAMs), zonula occludens family, and cingulin, and E-cadherin which is associated with the catenins family belongs to AJ proteins.
In contrast to AJs, which are involved in intercellular recognition and the maintenance of cell–cell contacts, TJs assume the important function of semipermeable gates by regulating paracellular solute and water flux and preventing paracellular permeation of dangerous luminal agents. Moreover the appropriate function of the AJC is coordinated by a complex array of signaling proteins which include the Rho and Rap family of GTPases, kinases, and phosphatases.

Numerous studies have reported the disruption of E-cadherin, α-catenin and β-catenin in adherens junctions of the epithelial layer in the inflamed mucosa of patients with IBD. Altered expression of these proteins correlated with the extension of inflammation. In addition to these alterations, morphological and structural changes of tight junctions were also observed in both UC and CD patients. Emerging evidence suggests that altered expression of some tight junction proteins in the intestinal mucosa of IBD patients may be central pathogenic factors involved in defective intestinal barrier. The scope of the current review is focused on newer observations supporting the role of JAM-A (member of JAM family) as a new “gate keeper” of epithelial integrity.

**JAM-A and Tight Junctions**

Junctional adhesion molecules (JAMs) are a family of proteins composed of five members including JAM-A, JAM-B, JAM-C, JAM-4, and JAM-like. All members are localized at intercellular contacts of endothelial and epithelial cells, and via homophilic interactions they participate in the assembly and maintenance of junctions.

JAM-A was the first JAM-family transmembrane protein to be identified. Not only is it positioned at the apical surface at tight junctions of epithelial and endothelial cells of various tissues, but JAM-A is also expressed on circulating platelets, monocytes, lymphocytes, neutrophils, dendritic cells, and spermatozoa. JAM-A can also function as a receptor for reovirus. In addition to homophilic binding, JAM-A has been proven to be capable of heterophilic engagement with other adhesion molecules such as integrins and via this type of ligation is involved in a variety of cellular process including leukocyte transmigration (directing the correct movement of leukocytes toward the site of inflammation) angiogenesis and platelet aggregation.

At intercellular contacts JAM-A associates with the tight-junction components cingulin, occludin, and the PDZ-domain-containing proteins, which act as scaffold proteins in the cytoplasm. The interaction with scaffold proteins may be functionally relevant for linking JAM-A to the cytoskeleton, and to signal transduction pathways for the regulation of cell polarity, growth, and differentiation. In fact, a dramatic change in cell morphology associated with a mislocalization of other junctions proteins was demonstrated in vitro after depletion of JAM-A in human hepatocytes. Similar findings were also reported in human intestinal epithelial cell lines. Its absence altered the polarity of cells and enhanced paracellular permeability. Furthermore, the inhibition of JAM-A with either blocking antibodies or soluble recombinant JAM-A protein prevented the recovery of barrier function of epithelial cells and human corneal endothelium after disruption of TJ. These data suggested that homophilic interaction of JAM-A affects tight-junction formation and stability. Thus, the function of JAM-A at tight junctions appears to be crucial as an extracellular adhesive molecule in the direct regulation of barrier function and important for intracellular cytosolic interactions. Although it is well documented that epithelial barrier function is compromised in the colonic mucosa of IBD patients, mechanisms clarifying this event are still unknown.

Recently, Kurcharzik et al. showed the reduction of some TJ proteins including occludin, JAM-A, claudin-1, and ZO-1 in the inflamed mucosa of patients with IBD. In contrast to occludin, JAM-A, claudin-1, and...
ZO-1 proteins were specifically downregulated at sites of active intestinal inflammation. Similarly, we found that the normal colonic mucosa expressed abundant epithelial JAM-A, while in both UC and CD mucosa its expression was downregulated exclusively in involved tissue. Though occludin is important in the organization of the TJ, its absence at epithelial junctions does not affect both TJ strand formation and intestinal barrier functions, as observed in occludin null mice. To the contrary, JAM-A null mice displayed increased intestinal permeability and reduced transepithelial resistance compared to wild type. Furthermore, we found enhanced altered intestinal permeability during dextran sodium sulfate (DSS)-induced colonic inflammation and determined a massive leukocyte infiltration of the intestinal mucosa, an excessive production of inflammatory cytokines and chemokines in the gut, and strikingly high mortality. Furthermore, a widespread apoptosis of epithelial cells was observed in colonic mucosa of colitic JAM-A null mice, but not in wild-type mice. Blocking JAM-A with the monoclonal antibody reproduced in wild-type mice a similar phenotype to that of JAM-A null mice during DSS treatment. Surprisingly, depletion of commensal bacteria with broad-spectrum antibiotics did not diminish inflammation driven by the administration of DSS in JAM-A null mice, excluding therefore the role of increased bacterial translocation during the treatment. Because JAM-A is not only expressed at epithelial and endothelial junctions, but also on circulating leucocytes, the use of endothelial conditional JAM-A null mice has allowed for the identification of epithelial JAM-A and the exclusion of endothelial/ematopoietic JAM-A in controlling intestinal permeability, suggesting that epithelial JAM-A is essential for the preservation of a well-balanced gut homeostasis.

Laukoetter and colleagues reported similar findings in JAM-A deficient mice. The authors showed that JAM-A null mice display an over-expression of claudin-10 and -15 in the colonic mucosa, and speculated that such upregulation might be responsible for the increased permeability and susceptibility to DSS in JAM-A null mice. Our data has supported these observations. We reported that JAM-A null and wild-type mice have identical expression levels of claudin -1, -2, -3, cingulin, and ZO-1, but not claudin-10 and -15. The claudin proteins create the selective aqueous pores that allow for the paracellular movements of solutes and soluble ions across the epithelium. Thus, the loss of JAM-A results in increased expression of claudin-10 and -15, which could generate the passage of water and ions in intestinal mucosa that, at least partially, could explain the altered intestinal permeability seen in JAM-A null mice.

Some studies have demonstrated that inflammatory processes have differential effects on claudin proteins. An increase in the expression of IL13-dependent pore-forming protein claudin-2 in the intestinal mucosa in patients with UC has been observed. Claudin-2, which appears in a tight junction location throughout the extended crypts of intestinal tissue, has been shown to induce cation-selective channels in the epithelial tight junctions resulting in increased paracellular permeability to sodium. This could help explain the increased ion flux and diarrhea in IBD. It is plausible to believe that increased expression of claudin-10 and -15 could have similar effects in enhancing paracellular movements of ions across the intestinal epithelia in JAM-A null mice (Fig. 1). More studies are needed to determine whether claudin-10 and -15 contribute in alteration of epithelial permeability in patients with IBD.

What drives the downregulation of JAM-A under inflammatory conditions remains to be identified. It is well known that the inflammatory cytokines like TNF-α, IFN-γ, and IL-1β are involved in directly increasing intestinal epithelial permeability and are able to induce the disassembly of TJ in epithelial cells. Although in vitro co-stimulation of epithelial cell lines with INF-γ induced internalization of TJ proteins by macropinocytosis including
Figure 1. Model of epithelial barrier dysfunction in the absence of JAM-A protein in inflammatory bowel disease. In normal conditions, the intestinal barrier is sealed by adherens junctions and tight junction proteins, which regulate paracellular ion and solute permeability and prevent the passage of antigens from the lumen into the intestinal tissue. In inflammatory bowel disease, JAM-A expression is downregulated, but only in the inflamed mucosa. The loss of JAM-A alters intestinal permeability via increased expression of claudin-10 and -15 that create pores favoring the passage of water and ions in the intestinal mucosa. Furthermore, this change in JAM-A expression induces widespread apoptosis of epithelial cells causing erosion and ulceration of the intestinal mucosa.

JAM-A, further studies are required to clarify the mechanisms by which JAM-A is downregulated.\textsuperscript{41} Furthermore, NF-κB has a regulatory role on TJ protein expression.\textsuperscript{40,42} Indeed, mice deficient for NF-κB essential modulator (NEMO), essential for NF-κB activation, display increased epithelial cell apoptosis and permeability leading to the development of severe chronic spontaneous colitis.\textsuperscript{42} Additional studies are needed to investigate whether the NF-κB signaling pathway is involved in the observed downregulation of JAM-A expression and increased epithelial apoptosis in experimental IBD.

Acknowledgments

This work was supported by grants from the Ricerca Ordinaria Finalizzata from the Italian Minister of Health and from the Broad Medical Research program to S.D., and “Premio Tilde Fiorentini” to S.V.

Conflicts of Interest

The authors declare no conflicts of interest.

References


