KLF-5 extends its fingers to desmosomes: The next frontier for enteric epithelial research? Running title: KLF-5 extends its fingers to desmosomes: a new frontier?

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Pathologic alterations in intestinal epithelial permeability can be a cardinal feature of inflammatory bowel disease (IBD). A monolayer of intestinal epithelial cells (IECs) and the junctions that seal the paracellular spaces between them serve as the body’s primary barrier against luminal pathogens, provide a means for selective permeability of nutrients and water, and enable the establishment of an immunological environment through cross-talk with enteric flora (2). Aberrance in the integrity of intestinal cell junctions, which include tight junctions, adherens junctions, desmosomes and gap junctions, contributes to increased intestinal permeability and the pathogenesis of inflammation in patients with IBD (3, 4). Numerous prior studies have focused on the roles of tight junctions and adherens junctions. The current study instead highlights the direct role of Kruppel-like factor 5 (KLF5) through desmoglein-2 (DSG2), a component of desmosome complexes, in intestinal epithelial barrier function.

KLF5 is a widely expressed zinc-finger transcription factor known for its critical roles in cellular proliferation and tissue morphogenesis. The authors had previously demonstrated that KLF5 is important for the proliferation of mouse intestinal epithelial stem cells (5). KLF5 has more recently been recently implicated in the maintenance of enteric barrier function (6) and has been shown to be protective in a murine model of colitis (7). Corroborative of these data, genome-wide association studies have demonstrated lower KLF5 expression in colonic tissue from patients with IBD (8). The mechanism by which KLF5 regulates colonic barrier function, however, has not been determined.

Desmosomes exhibit their intercellular adhesive capacities through two types of cadherin transmembrane proteins, desmocollins and desmogleins. Human intestinal epithelial cells primarily express desmoglein-2 (DSG2) and desmocollin-2 (DSC2). Several lines of evidence suggest a role for desmosomal cadherins, particularly DSG2, in IBD pathogenesis. Abundance and intensity of immunofluorescence staining for DSG2 is significantly reduced in regions of inflamed mucosal tissue of individuals with IBD (3). Further, corticosteroid-sensitive patients with ulcerative colitis exhibit greater DSG2 staining in colonic biopsies compared to corticosteroid-refractory patients (9). Intestinal barrier function is impaired in Caco-2 cells after administration of a DSG2 antibody (10), in line with their suspected role in epithelial barrier maintenance.

In the current issue, Liu and colleagues provide a novel perspective on the regulation of intestinal barrier function by KLF5 through DSG2 (1). The authors first examined this interaction utilizing two mouse models of intestine-specific Klf5 knockout: a constitutive...
knockout mouse model (Klf5-floxed : Villin-Cre) and a tamoxifen-inducible knockout mouse model (Klf5-floxed : Villin-CreERT2). By analysis of colonic transcript levels of genes associated with cell junctions, the investigators discovered significantly decreased levels of the desmosomal cadherin Dsg2. The correlation between levels of KLF5 and DSG2 was confirmed by immunohistochemistry staining which showed decreased staining of DSG2 in tissues that had concurrently diminished KLF5 staining. Using transmission electron microscopy, the authors further demonstrated stark morphological differences in the desmosome complexes of cells from Klf5-inducible knockout mice, including a decreased number of microvilli on the apical membranes, decreased desmosome numbers and an enlarged intercellular space between adjacent plasma membranes.

The authors correlated the in vivo findings with an in vitro cell culture model using the Caco-2 BBBe cell line. They established an inducible KLF5 knockdown cell line whereby treatment with doxycycline rendered inhibition of KLF5 expression. The investigators showed that doxycycline- treated cells exhibited a similar pattern of desmosome morphological alterations to those seen in the Klf5-floxed : Villin-Cre and the Klf5-floxed : Villin-CreERT2 mouse models. They further found that the cell monolayers exhibited increased permeability, as determined by decreased transepithelial electrical resistance and an increased level of FITC-dextran pass-through concentration. As exhibited in the in vivo models, the investigators found that protein levels of DSG2 were significantly decreased, further confirming the link between KLF5 and DSG2. Intriguingly, DSG2 overexpression in the KLF5 knockdown cell line partially rescued the impairments in barrier function.

Liu et al. beautifully demonstrated a novel influence of KLF5 on the intestinal epithelium through its interaction with the desmosomal cadherin, DSG2. The investigators comprehensively evaluated their findings in two in vivo models and confirmed the mechanisms involved by utilizing an in vitro KLF5 knockdown cell line. Importantly, they evaluated a wide array of cell junction components, including cadherins, claudins and occludins, and only found highly significant differences in DSG2, suggesting a highly specific action for KLF5.

The authors illustrated an important ability of DSG2 overexpression to partially reverse the epithelial permeability alterations caused by KLF5 knockdown. Because this rescue was only partial, however, KLF5 may act on additional factors or through other
pathways to modulate epithelial barrier and function. Nonetheless, these findings are
demonstrative of a unique, novel pathway that may be harnessed to influence epithelial
barrier function.

The findings in this article may be relevant to disease states where intestinal barrier
permeability can be a precipitant of the underlying pathophysiology. Intestinal
permeability has been reported as a sensitive prognostic indicator for relapse in patients
with Crohn’s disease (11), and multiple studies have revealed that intestinal epithelial
structure and permeability are dysfunctional in at least a subset of individuals with IBD
(12, 13). It would thus be interesting to determine whether KLF5 KO mice develop
intestinal inflammation and, if so, the respective roles of intestinal barrier function and
the enteric microbiota. Further, intestinal barrier permeability has been found to be
increased in subsets of patients with other GI conditions like irritable bowel syndrome
(14, 15). There are currently no FDA-approved therapies that target the intestinal
epithelial barrier. This study thus adds significantly to our arsenal of knowledge geared
towards understanding the mechanisms underlying epithelial permeability and may
pave the way towards novel interventions for disease states where epithelial
permeability is a contributing factor.


