ABSTRACT

The chemokine receptor CXCR4 and its cognate ligand CXCL12 are constitutively expressed by the gastrointestinal epithelium. The CXCR4/CXCL12 signaling axis regulates intestinal epithelial migration, barrier maturation and restitution, consistent with an in vivo role for this axis in the maintenance of mucosal barrier integrity. To better understand the role of CXCR4 at the gastrointestinal epithelial surface, conditional knockout mice in which CXCR4 expression was ablated specifically in intestinal epithelial cells were generated. Methods: Epithelial specific CXCR4 knockout mice were generated using the Cre-loxP system. Animal weight, development and intestinal morphology were monitored using histological analyses, RT-PCR, immunostaining and immunofluorescence microscopy. Results: RT-PCR, immunofluorescence, and immunofluorescence microscopy confirmed that CXCR4 was specifically deleted in intestinal epithelial cells. Conditional CXCR4 knockout mice developed normally and were phenotypically and morphologically comparable to controls. Chronic colitis induced by three bouts of 3% (w/v) DSS resulted in more consistent GALT formation and re-epithelialization in knockout mice compared to controls. Heterozygous animals demonstrated greater healing of ulcers than control or homozygous CXCR4 deleted animals. Consistent with those data knockdown of CXCR4 in cultured IEC-6 cells evoked more migratory epithelial cells relative to controls. Notably, ERK1/2 phosphorylation was markedly decreased in CXCR4 deleted cells. Moreover, the size of ERK1/2 phosphorylation was reversed from surface colonicin to crypt cell activity in the small intestine of mice, suggesting an important role for ERK signaling in barrier formation and ulcer healing. Conclusions: These data indicate that white enterogenous CXCR4 is a potent motogen, sustained CXCR4 signaling is a key regulator of basal ERK1/2 activity. Conditional deletion of CXCR4 in epithelial cells leads to a reversal in ERK2 activity within the crypt/surface axis. Modification in ERK2 signaling plays key roles in barrier maintenance potentially through proliferation, migration and anoikis at sites of epithelial ulcers.

Repair Through Altered ERK1/2 Targeted Deletion of Intestinal Epithelial CXCR4 Modulates Epithelial Barrier Signaling

Noah Zimmerman, Rebecca A. Vongs, Priscilla A. Johansen, Michael K. Wendt, Nita H. Salzman and Michael B. Dwinell

Department of Microbiology and Molecular Genetics, Medical College of Wisconsin, Milwaukee, WI

ERK 1/2 phosphorylation was altered in both CXCR4 f/f and CXCR4 f/+ mice compared to wild type littermates. A. White CXCR4 f/f mice demonstrate higher ERK and JNK expression in the colon, CXCR4 f/+ mice have less JNK and p38 expression. B. ERK 1/2 is primarily expressed at the villus tip and crypt face in wild type mice, while expression moves to the crypt bottom in the CXCR4 f/f mice. ERK expression was observed in both the villus tip and crypt base in CXCR4 f/+ mice. Images representative of 4 animals each.