inhibited expression of IL23 and IL-1, along with reduced intestinal production of IL22 and fibrosis in chronic TNBS fibrosis mouse model. Suppression of IL23 and IL-1 expression upon mTOR inhibition is associated with an elevated autophagy activity. Ablating the autophagy gene Atg7 in CX3CR1Atg7f/f mice increases expression of IL23 and IL-1, leading to increased IL22 expression and fibrosis. Moreover, we demonstrated that IL22 facilitated the transformation of fibroblasts to myofibroblasts. IL22-pre-exposed fibroblasts become more sensitive to fibrotic reaction to TGF through induction of TGFRIII. Thus, priming of fibroblasts by IL22 represents a linchpin of excessive fibrotic response. Altogether, this study elucidated a signaling cascade underlying intestinal fibrosis in which mTOR/autophagy in CX3CR1+ mononuclear phagocytes regulates the expression of intestinal IL23/IL22 to mediate the fibrotic response. Thus, this cascade could be a pivotal target for alleviation of intestinal fibrosis.

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## MUCOSAL STEM CELL HETEROGENEITY IN PEDIATRIC CROHN'S DISEASE

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Background: Crohn's is an aggressive inflammatory bowel disease driven by interactions of unknown relative significance between the immune system, the microbiome, and the intervening intestinal epithelial barrier. Histopathology analyses and genome-wide association studies (GWAS) have provided renewed focus on the role of the mucosal barrier in this disease.

Methods: We obtained endoscopic biopsies from 30 patients either newly diagnosed with Crohn's, under treatment for Crohn's, or functional controls. Using stem cell cloning technology that maintains cells in their most immature ground state with the potential for conditional differentiation to a mature epithelium1, we analyzed stem cells from each of these cases.

Results: We find that pediatric patients with Crohn's harbor two populations of mucosal stem cells including one that is similar to children without Crohn's and a second distinguished by three epigenetically maintained traits that are potentially pathogenic. These traits include a robust inflammatory gene signature including many implicated by GWAS in the risk for Crohn's, defective secretory cell differentiation impacting barrier function, and a profound homeotic transformation of terminal ileum to proximal gastrointestinal tract. Our epistasis analyses indicate that the homeotic transformation dictates both the inflammatory and barrier defects and underlies the absolute concurrence of these traits across cases. Our analysis of barrier function of epithelia formed from these aberrant stem cells shows their enhanced sensitivity to bacterial components, and the pattern of chemokines and cytokines produced by these cells promotes leukocyte extravasation.

Conclusion: These findings link an epigenetically distinct subpopulation of intestinal stem cells and the defective epithelial they beget to the pathophysiology of Crohn's and may reframe therapeutic strategies in this disease.

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## MUTATIONS IN STXBP3 CONTRIBUTE TO VERY EARLY ONSET OF IBD, IMMUNODEFICIENCY AND HEARING LOSS

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Very early-onset inflammatory bowel disease (VEO-IBD), defined by the onset of IBD before 6 years of age, is often associated with more severe and extensive disease than IBD in older patients. Some VEO-IBD cases have been linked to mutations in primary immunodeficiency genes, which regulate immunity and hyperinflammatory pathways, however the underlying pathophysiological mechanisms are still poorly understood. Here we describe eight patients from four unrelated families manifesting with VEO-IBD, immunodeficiency and severe bilateral sensorineural hearing loss - each carrying either heterozygous or compound heterozygous deleterious mutations in Syntaxin-Binding Protein 3 gene (STXBP3). These mutations interfere with either intron splicing or protein stability, lead to reduced STXBP3 protein expression, which in turn, affect cytotoxic T-Lymphocyte (CTL) and epithelial cell function. STXBP3 knock-down in control CTLs significantly reduces cytotoxic activity, mimicking the patients' CTL defects. Strikingly, forced expression of STXBP3 rescues patient CTL function. Live-cell microscopy analyses show that STXBP3 is required for recycling of RAB11A-containing endosomes to the plasma membrane. Defects in this process prevent the delivery of key effector proteins that are required for granule secretion and epithelial cell polarity. Our results identify STXBP3 as a causal gene for the development of VEO-IBD with associated immunodeficiency and hearing loss.

Figure 1

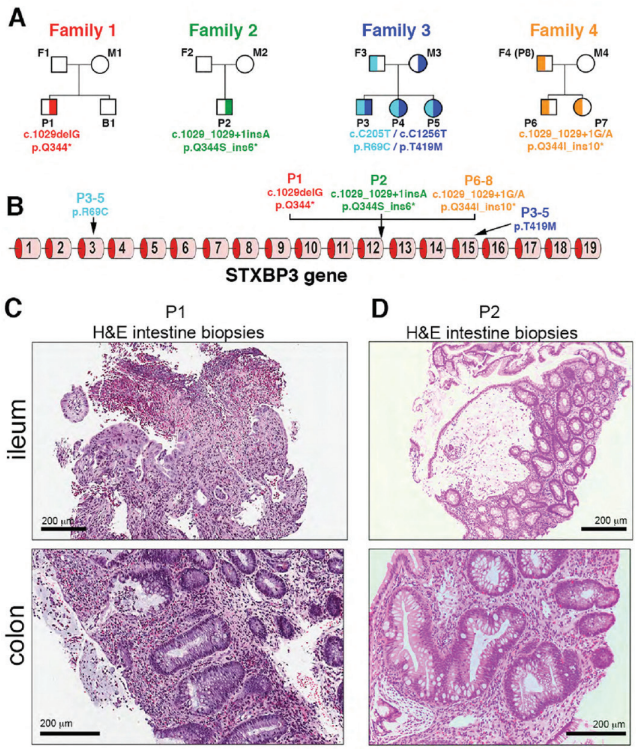


Figure 2