IL-33 Induces Genes Involved in Epithelial-Mesenchymal Transition in Intestinal Epithelial Cells

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Introduction
IL-33 is induced in the mucosa of adult and pediatric patients with inflammatory bowel disease. IL-33 has been found to have an indirect role in intestinal epithelial differentiation into goblet cells by inducing IL-13 production from group 2 innate lymphoid cells and T helper 2 cells. However, how IL-33 directly affects epithelial function remains unknown.

Hypothesis
IL-33 directly induces genes involved in epithelial-mesenchymal transition (EMT) in intestinal epithelial cells.

Methods
Colonoids, 3-dimensional primary cultures that contain all types of colon epithelial cells, were generated from mouse colon crypts. RNA sequencing was performed on colonoids exposed to IL-33 for 0, 2, or 6 hours. RT-PCR for the housekeeping gene Gapdh and the EMT genes Cdh2, Zeb1, and Igfbp5 was performed on colonoids exposed to IL-33 for 0, 2, 6, or 24 hours. WT and IL-13−/− mice were given intraperitoneal injections of IL-33. IL-13−/− mice were utilized since IL-13 is known to be induced by IL-33 and act on epithelial cells. Whole colon tissue and colon epithelial cells were isolated and underwent RT-PCR for Gapdh, Cdh2, Zeb1, and Igfbp5.

Results
RNA sequencing of colonoids exposed to IL-33 revealed upregulation of Cdh2, Zeb1, and Igfbp5, three genes involved in EMT. RT-PCR showed a trend toward increase in all three genes. To explore the role in vivo, whole colon tissue and epithelial cells were isolated from WT and IL 13−/− mice treated with IL-33 for 4 days. No changes were seen in these genes in WT whole tissue. In WT epithelial cells, we saw a slight increase in Cdh2 and slight decreases in Zeb1 and Igfbp5, suggesting indirect effects of IL-33 may be affecting gene expression in vivo. In IL 13−/− total tissue, we saw a significant increase in Cdh2 and Zeb1 and an increase in Igfbp5. However, in isolated epithelial cells all genes were decreased, indicating effects of IL-33 on other cell types.

Conclusions
IL-33 can directly affect intestinal epithelial cells by increasing the expression of genes involved in EMT, specifically Cdh2, Zeb1, and Igfbp5. These results can be seen in vitro but may be overcome in vivo by secondary effects of IL-33.

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