Catenins in the Esophageal Epithelium in Eosinophilic Esophagitis

Functional Consequences of CAPN14-Mediated Proteolysis of Catenins in the Esophageal Epithelium in Eosinophilic Esophagitis

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Background

Eosinophilic esophagitis (EoE) is a food-related chronic, allergic disorder that affects 1 in 2000 individuals and costs the United States about 1 billion dollars annually to treat and manage. EoE is characterized by chronic esophageal inflammation that can make it difficult and painful to swallow. In children, this inflammatory process leads to decreased appetite, resulting in inadequate nutrition and ultimate failure to thrive. Patients must be placed on a restricted diet and undergo numerous endoscopies throughout their lifetime, resulting in a low quality of life. As a cellular and molecular hallmark of EoE, impaired epithelial barrier function, increased proliferation, and altered differentiation in the esophageal epithelium.

Methods

First, EoE cells were cultured with or without IL-13, a cytokine shown to upregulate expression of CAPN14. We generated protein lysates from these cells and conducted SDS-PAGE and western blot analysis to determine if IL-13 affected CTNNA1 production. Next, we co-expressed CTNNA1 and alpha catenin in HEK293T cells by transfecting them with mammalian expression constructs. We then generated protein lysates and conducted SDS-PAGE and western blot analysis. Additionally, we used immunofluorescence staining to observe the pattern of alpha-catenin in patients with active EoE.

Results

IL-13 induction of CAPN14 in EPC2 cells has no effect on endogenous CTNNA1

Alpha catenin expression was unaffected when co-transfected with CAPN14 in HEK293T cells

Patient biopsy lysates exhibit no change in CTNNA1 quantity in patients with active EoE

Immunofluorescence staining of patient biopsies demonstrate no change in CTNNA1 expression pattern in patients with active EoE

Conclusions

- In EPC2 cells incubated with Ca²⁺ and IL-13, there was no significant decrease in CTNNA1 quantity or molecular weight regardless of time control.
- In HEK293T cells that were co-transfected with CAPN14 and CTNNA1, there was no decrease in CTNNA1 quantity or molecular weight, regardless of the presence of ionomycin.
- In patient biopsy lysates there was no decrease in CTNNA1 quantity or expression pattern in patients with active EoE.

Future Directions

- Determine if proteolytic products of CTNNA1 identified in preliminary screening studies are sufficient to promote barrier dysfunction.
- Explore alternative targets of CAPN14
- Preliminary lab data suggest several alternative targets.
- One target of interest is Annexin A1, a protein involved in anti-inflammatory responses and an inhibitor of the NF-kB signaling pathway.
- Annexin A1 is significantly decreased on the RNA level in patients with active EoE, and preliminary lab data demonstrates there may be lower protein levels in patients with active EoE.

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