A Newly Discovered Regulatory CD8 T Cell Network Has The Potential To Regulate And Eliminate Pathogenic CD4 T Cells In Autoimmune Mediated Disease Of The Gut

Crane, CA; Bowser, J; Fasnacht, R; Gardell, JL; Maurer, M; Julien, S; Templeton, ML; Therriault J; Swiderek, KM

Introduction
Several groups have described a subset of immunosuppressive CD8 T cells in inflammatory disease settings, suggesting a protective role for select CD8 T cells in autoimmune disease. We hypothesized that a dysregulated regulatory CD8 T cell (CD8 Treg) network was similarly involved in the pathology of autoimmune diseases of the gut, including Celiac disease, Crohn's, and Ulcerative Colitis. Here, we have demonstrated the presence of this CD8 Treg cell network in Celiac patients and its potential to reduce pathogenic CD4 T cell prevalence.

We characterized a surface marker and functional phenotype associated with CD8 Treg prevalence and consequences of TCR-mediated activation. Using this phenotype, we then confirmed the presence and prevalence in Celiac patient PBMCs and duodenal tissues (Fig. 1). Activation resulted in a rapid and specific cytolytic mechanism of action CD8 Treg cells. Using Celiac disease as platform to introduce a known antigenic trigger for autoimmunity, we defined CD8 Tregs and their activity in patients with other autoimmune diseases, including Crohn's disease and Ulcerative Colitis.

In autoimmunity, regulatory CD8 T cells fail to control expansion of pathogenic CD4 T cells resulting in downstream inflammation

CD8 Treg cells are mobilized by surrogate peptide cocktail and reduce pathogenic CD4 T cells

Celiac patient-derived CD8 Treg cells use a cytolytic mechanism to specifically eliminate pathogenic CD4 T cells and reduce inflammatory cytokines

Methods
• Using antigen induced animal models of experimental autoimmune encephalomyelitis (EAE), as well as Celiac patient-derived PBMCs and tissues, we evaluate CD8 Treg functions
• Methods include flow cytometry, soluble analyte analysis, and immunohistochemistry.
• We describe co-culture assays to define the phenotype, relevant mechanisms of action, dosing, and kinetics of CD8 Treg functions after exposure to gluten peptides

Conclusions
• We describe a novel CD8 T regulatory cell network present in autoimmune mediated gut disorders and other inflammatory disease.
• CD8 T regulatory cells use a cytolytic mechanism to specifically eliminate pathogenic CD4 T cells
• Recovery of CD8 T regulatory cell functions may suppress pathogenic T cells and reduce severity of inflammatory disease
• This network may be targeted by immune-modulating biologics

Acknowledgements:
The authors would like to acknowledge the contributions of Hooke Laboratories, Inc (Lawrence, MA) for conducting animal studies (Figure 1) and Ullswater Inc (Cambridge, MA) for panel development and multiplex immunohistochemical tissue staining (Figure 4).

Contact:
Follow-up questions can be directed to Kristine Szewski, Chief Scientific Officer, Mozart Therapeutics ksszewski@mozart-llc.com or by visiting the website at https://www.mozart-llc.com/

References:

Figure 4 Duodenal tissues from Celiac patients were stained for CD8 Treg markers and quantified to evaluate presence and prevalence before and after gluten consumption. Similar increases are observed in peripheral blood after gluten consumption.

Figure 5 PBMCs from healthy donors, patients with Type 1 Diabetes (T1D), Ulcerative Colitis (UC), Systemic Lupus Erythematosus (SLE), Multiple Sclerosis (MS), Crohn's, or Celiac were evaluated for CD8 Treg prevalence.