Grant Number: 1R43DK098896-01

Principal Investigator: Gordon Robert Alton, PhD

Project Title: Development of RORγt Immunomodulators Targeting the TH17 Axis in IBD

This research is supported by the National Institute Of Diabetes And Digestive And Kidney Diseases of the National Institutes of Health under Award Number R43DK098896.

Th17 cells are a lineage of T helper cells that have recently been identified as critical mediators of the immunopathology of several human inflammatory disease states, including inflammatory bowel disease (IBD). The orphan nuclear receptor ROR©t has been shown to be the master controller of the differentiation of Th17 cells. RORγt knockout animals are highly resistant to several autoimmune diseases. In the body the intestine has the highest proportion of Th17 cells compared to other organs. With relevance to IBD, substantially reduced numbers of Th17 cells are found in the intestinal lamina propria of animals with RORγt-null T-cells, compared to wild-type.

Antagonism of the transcriptional activity of ROR©t results in blocking the differentiation of $CD4^+$ T-cells to the Th17 cell lineage. Thus, ROR©t antagonists reduce the Th17 cell population at sites of inflammation. Th17 cells secrete large quantities of IL-17A, IL-17F, IL-22, TNF-α and other inflammatory cytokines. ROR©t is important for production of these cytokines and it has been demonstrated that ROR©t antagonists reduce the secretion of these cytokines from pre-existing Th17 cells. Therefore, small molecule antagonists of RORγt will be efficacious in modulating the pathogenesis of IBD by reduction of the Th17 cell population and IL-17A/F production.

Some small molecule tool compounds have been reported that attenuate autoimmune disease in animal models, but none of these possess sufficient drug-like properties to be considered as viable starting points for drug discovery programs. Using our proprietary RORγt BindingSIGHTs (patent pending) structure-guided drug design platform we have performed an *in silico* screen of 22 million compounds obtained from our MANIFOLD virtual compound library (the largest in the industry). Subsequently, more than 1000 commercially available drug-like compounds were tested in cell-based reporter assays for antagonism of RORγt–mediated transcription. Potent hits were further evaluated for selectivity against closely related nuclear receptors.

Some compounds were functionally active to inhibit the *ex vivo* differentiation of human Th17 cells. Based on the success of the preliminary data we propose the following aims:

(1) optimize the pharmacological properties of novel RORγt antagonists using our proprietary BindingSIGHTs drug design platform to guide sophisticated medicinal chemistry;

(2) determine the *ex vivo* T-cell functional activity of RORγt antagonists on human Th17, Th1, Th2 and Treg cells;

(3) evaluate the therapeutic efficacy of RORγt antagonists in animal models of IBD.

Together, these studies will provide highly advanced and novel drug-like chemical matter with the appropriate pharmacological profile to establish the feasibility of our approach. This will ultimately enable subsequent IND-enabling studies and clinical trials.