Dr. Nicholas Cianciola, School of Medicine, Case Western Reserve University

"Activation of an alternative cholesterol homeostatic mechanism in NPC"

"Dr. Cianciola's project builds upon an exciting observation he made while a graduate student and will explore the role of a viral protein in facilitating movement of cholesterol from lysosomes. The research has the potential to identify new therapeutic targets to stimulate release of cholesterol from lysosomes in the absence of a functional NPC1 protein."

Dr. Dan Ory, Chairman, Scientific Advisory Board, NNPDF

Lay Summary

Activation of an alternative cholesterol homeostatic mechanism in Niemann-Pick disease type C

Nicholas Cianciola; Sponsor: Cathleen Carlin

Niemann-Pick disease type C (NPC) is a fatal autosomal recessive disorder characterized by progressive neurodegeneration due to the inappropriate storage of cholesterol and other lipids inside the cell. Cells obtain cholesterol in two ways: *de novo* biosynthesis in the endoplasmic reticulum (ER); and uptake of dietary cholesterol *via* LDL particles through a series of internal membrane-bound compartments known collectively as endosomes. NPC is caused by mutations affecting two proteins, NPC1 and NPC2, which coordinate the transfer of dietary cholesterol from endosomes to other parts of the cell. Mutations in either protein cause the buildup of cholesterol in lysosomal storage organelles (LSOs). These mutations also block the transport of cholesterol from endosomes to the ER, where the machinery that regulates the *de novo* synthesis of cholesterol is located. The breakdown in cholesterol transport leads to excessive cholesterol synthesis because the ER no longer senses cholesterol in endosomes, further exacerbating the disease. Despite significant advances in our knowledge of cholesterol trafficking and regulation in the past decade, it is still unknown how dietary cholesterol is transported to the ER.

Human viruses have evolved complex adaptations to ensure their own replication and survival, and the study of the cell biology of host-virus interactions remains a key area of research. Host-virus interactions also bring unique perspectives to fundamental cell biological mechanisms. Recently, I identified a pathway that regulates cholesterol homeostasis independent of NPC proteins. This pathway was identified from studies using a human adenovirus protein called RIDα that interacts with host proteins involved in endosome trafficking. I showed that RIDα expression significantly reduces the number of LSOs, and also decreases overall levels of intracellular cholesterol, in cells with an NPC1 mutation. In addition, a mutant form of the RIDα protein provokes a cholesterol storage phenotype in cells expressing normal NPC proteins. My goal is to use RIDα as a model system to examine a complementary cholesterol trafficking pathway that operates independent of NPC proteins. In the past, human adenovirus has been used to study type 1 diabetes, and tissue-specific expression of a different adenovirus protein prevents diabetes in a mouse model. Similarly, I predict that tissue-specific expression of RIDα will correct neurodegeneration and liver dysfunction in the *npc1-/-* mouse.

Before proceeding with those studies, I will test the hypothesis that RIDα restores cholesterol homeostasis at the level of the ER in addition to alleviating cholesterol storage abnormalities in NPC cells. The first specific aim of the project will determine whether RIDα facilitates transport of cholesterol to the ER in NPC cells. I will use a number of established techniques that test the activation of cholesterol homeostatic processes that rely on delivery of dietary cholesterol to the ER. Second, I will test whether RIDα expression induces formation of direct physical contacts between endosomes and the ER, providing a possible mechanism for cholesterol transfer between these two structures. I plan to use a variety of microscopic techniques to demonstrate the presence of these contacts and to dissect the proteins that may be involved in their formation. These experiments will give us a better understanding of the fundamental mechanism of endosome-to-ER cholesterol transport, and identify novel targets for the design of new NPC therapeutics.