Lay Summary

For proteins to properly function, they must be properly folded. Improper folding of proteins typically results in their degradation, a process mediated by attachment of a degradation tag called ubiquitin. In some diseases attachment of ubiquitin and subsequent degradation of a functional protein results in pathology. This is true in NPC where ~95% of cases are caused by a mutation in the NPC1 protein, with the NPC1-I1061T mutation being the most prevalent. In patients with the NPC1-I1061T mutation, the NPC1 protein is functional, but is tagged with ubiquitin and subsequently degraded. This causes reduced levels of the NPC1 protein and accumulation of cholesterol. Therefore, it is reasonable to think that inhibiting attachment of ubiquitin to the NPC1 protein will be therapeutic, increasing the levels of NPC1 protein, and decreasing cholesterol accumulation (Figure 1).

One major hurdle in developing compounds that inhibit ubiquitination of specific proteins is understanding how ubiquitin is attached to proteins. During my predoctoral research to date, I have identified how ubiquitin is attached to proteins by the same enzymes that attach ubiquitin to NPC1. I am now ready to begin translating these findings to develop small molecules that prevent attachment of ubiquitin, and the subsequent degradation of select proteins. For these studies I have chosen to focus on attachment of ubiquitin to NPC1-I1061T. In my application I propose to 1) confirm that ubiquitin is attached to NPC1-I1061T in the same way as I have identified for other proteins; and 2) develop small molecules that will prevent the attachment of ubiquitin and subsequent degradation of NPC1-I1061T. Through these studies I plan to initiate the development of a new class of compounds that stabilize specific substrates by preventing ubiquitin from being attached to them, beginning with NPC1.

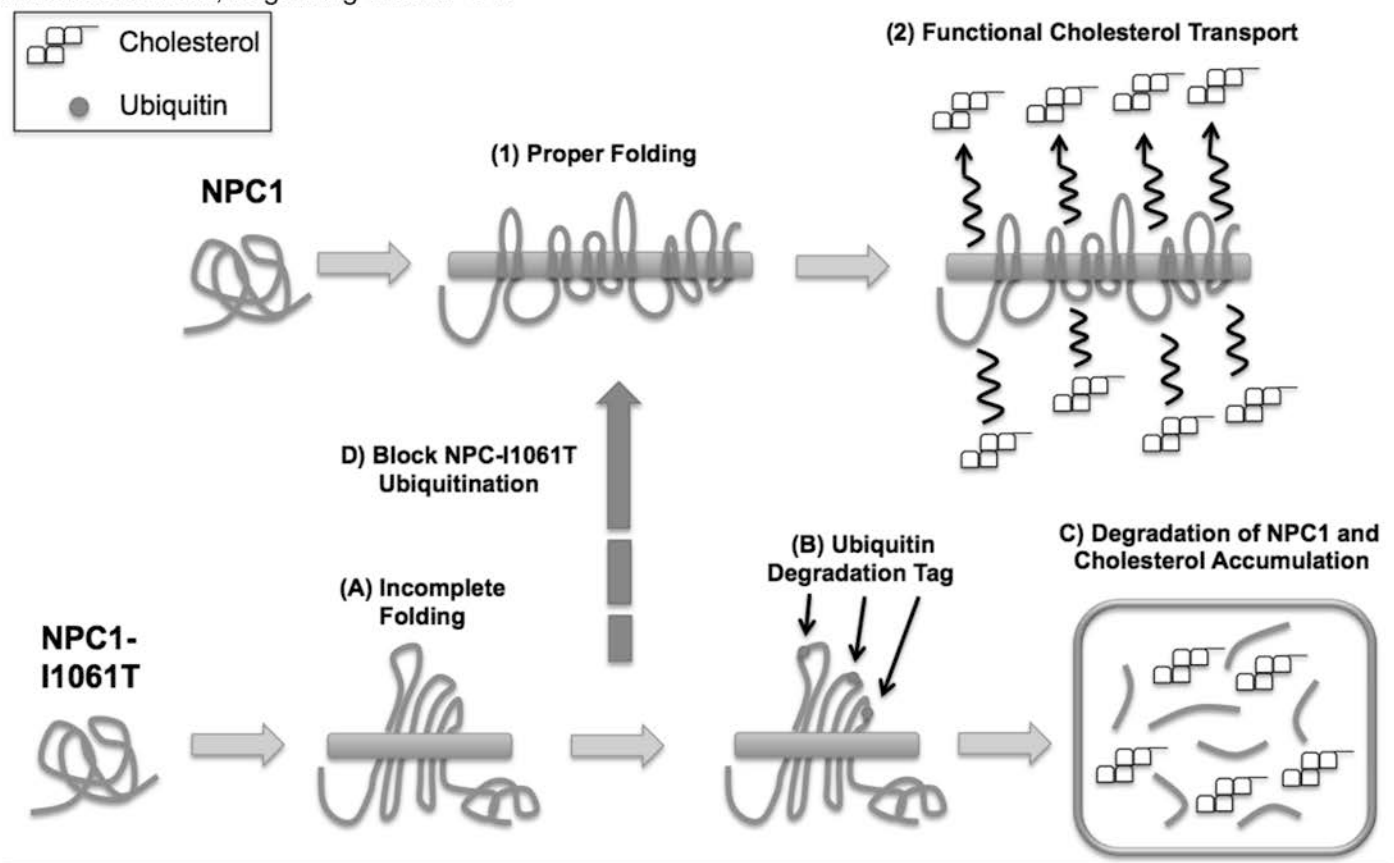

Figure 1: Outline of NPC1 function and degradation. NPC1 is folded (1) and functions normally to transport cholesterol (2) before it is eventually degraded. NPC1-I1061T is incompletely folded (A) ubiquitinated (B) and then degraded before it can function to remove accumulating cholesterol (C). Preventing the ubiquitination of NPC1-I1061T (D) would prevent NPC1-1061T from being degraded and allow it to properly fold, restoring its ability to transport cholesterol (2).