Oxidized Phospholipids Binding to Human Aortic Endothelial Cells

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Abstract

Lesions resulting from the uptake of cholesterol through the endothelial cell membrane will eventually cause heart disease, if left untreated, potentially leading to heart attack or stroke. The lesions are a result of a product of the oxidation of the cholesterol, known as Ox-LDL. This research is designed to determine the mechanisms by which such lesions develop. The stimulation by these oxidized lipids to the endothelial cells have proven to be of significant importance in the changing of the biological pathways. The stimulation of the cells is caused by the binding of these oxidized lipids to proteins, signaling changes in the pathways. In order to monitor this upregulation we have synthesized and oxidized both tagged and un-tagged versions of the lipid. After doing so, we performed western blots to determine the molecular weights of lipid-bound proteins which could be signaling changes in the pathways. Lipid-bound proteins were isolated by tagging oxidized lipid with biotin, and pulling down the protein-lipid complex. By isolating the proteins we will be able to cleave and identify the amino acid sequences and determine what proteins have bound. We have also used RT-PCR to monitor the upregulation of genes which have been previously proven to be involved in these changes.