STRONG CHILDREN'S RESEARCH CENTER

Summer 2015 Research Scholar

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ABSTRACT

Title: Gene transfer of the Na+, K+-ATPase β1 subunit rescues mice with LPS-induced lung injury and upregulates adherens junction proteins

Background: Acute lung injury (ALI) and Acute Respiratory Distress Syndrome (ARDS) are potentially fatal conditions that are characterized by retention of excess fluid in the alveoli and neutrophil infiltration into the alveolar space. Two cellular components that could play an important role in mitigating these symptoms are ion channels and cellular junctions. The Na^+, K^+-ATPase and the epithelial sodium channel (ENaC) can transport Na+ ions to create a concentration gradient that draws fluid out of the alveolar space. Cellular junctions, such as tight junctions and adherens junctions, keep this fluid from returning to the cells and prevent neutrophil infiltration. In polarized epithelia/endothelia, tight junctions are apically localized and act to regulate small molecule transport in and out of the interstitium. Adherens junctions are basolaterally located anchoring junctions that join neighboring cells through their actin bundles. The major proteins involved in adherens junctions are cadherins and catenins. Cadherins are anchoring proteins that act to hold neighboring cells together, while catenins are scaffolding proteins that help to connect cadherins to actin bundles of the cytoskeleton. Epithelial cells of the alveoli are linked through calcium dependent interactions between E-cadherin molecules. The Dean lab has previously shown that gene transfer of the Na^+,K^+-ATPase 1 subunit, but not 1-ENaC, leads to increased alveolar fluid clearance (AFC), decreased neutrophil infiltration, and upregulation of tight junction proteins.

Objective: The purpose of this study was to determine if 1-ENaC and 1 Na^+,K^+-ATPase gene delivery upregulate adherens junction proteins.

Results: Mice were instilled with lipopolysaccharide (LPS) to induce lung injury. One day later, these mice underwent electroporation-mediated gene transfer of either pcDNA3 (a vector control), 1-ENaC, or 1 Na^+,K^+-ATPase. Lung tissue was harvested 24 hours after gene transfer and protein levels were analyzed by densitometric analysis of western blots, which were