Non-Canonical Notch signaling also regulates the metabolism and proliferation of CD8 T-cells

Kaitlyn Hawkins, Kristina Larter, Hanh Luu, Samarpan Majumder, Luis Del Valle, Lucio Miele, and Fokhrul Hossain
Department of Genetics, School of Medicine, LSUHSC

Introduction

Triple-negative breast cancer (TBNC) is one of the harder forms of breast cancer to treat due to it lacking three receptors that are essential to the targeted therapy used to treat other forms of breast cancer. The Notch pathway is a protein pathway that has been proven essential in both healthy cells, specifically CD8 T-cells, and cancer cell growth and is generally overexpressed in cancer cells, including TBNC. Its prevalence in TBNC subsequently makes it a focus in finding targeted therapies for TBNC. The Notch pathway is also known to play a role in T-cell activation, which is necessary for the anti-tumor response in the body. With the non-canonical Notch pathway regulating both mitochondrial metabolism in TBNC cells and T-cell activation in CD8 T-cells, attempting to target this pathway in TBNC cells while not affecting CD8+ T-cells is a daunting task.

Objective

The aim of this project is to better understand the role of mitochondrial Notch (non-canonical Notch) in CD8+ T-cell metabolism, cell proliferation, and cytotoxic activity.

Methods

1. Extract CD8+ T-cells from the spleen of C57BL/6 mice, mitochondria from isolated cells, and immunoprecipitate Notch-1 from mitochondrial fraction.

2. Confirm the isolation of mitochondrial Notch-1.

3. Analyze the role of Notch-1 in metabolic processes, cell proliferation, and cytotoxic activity.

Western Blot

Immunofluorescence Assay

Seahorse Assay

T-cell proliferation assay

ELISA Assay

Results

Our results suggest that in addition to the canonical pathway, non-canonical Notch does play a role in metabolic processes, cell proliferation processes, and cytotoxic activity of CD8+ T-cells.

From these findings, future research will be geared toward determining the specific role Notch-1 plays in these necessary biological processes.

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