

Presenting Author Name

Dheer Pandey

Presenting Author Category

PhD Student

Research Category

Basic Science

Abstract Title

TRPA1 Mediates Oxidized Phosphatidylcholine-Induced Inflammatory Gene Expression in Human Airway Fibroblasts

Authors

Name	Role	Profession
Dheerendra Pandey	First Author	PhD Candidate
Dedmer Schaafsma	support in writing	Research Associate
Andrew Halayko	Corresponding Author	Professor and Tier 1 Canada Research Chair

Background

Oxidized phospholipids (OxPAPC) are inflammatory bioactive molecules that accumulate in the airways of children with asthma and may contribute to persistent airway inflammation and remodeling. OxPAPC activates transient receptor potential ankyrin 1 (TRPA1), promoting Ca^{2+} influx leads to airway smooth muscle contraction. Mesenchymal cells like airway smooth muscle cells and fibroblast play a critical role in airway inflammation and remodeling, which impair respiratory health in children with asthma. Here, we investigate the role of TRPA1 in OxPAPC-induced inflammatory gene expression and release in human airway fibroblasts (HAFs).

Objective

Determine the role of TRPA1 in OxPAPC induced inflammatory gene expression in human airway fibroblast

Methods

OxPAPC (60 $\mu\text{g/mL}$)-induced Ca^{2+} influx was assessed for 3 min in Fluo-4-loaded primary human airway fibroblasts (HAFs), with or without the TRPA1 inhibitor A967079 (1 μM). OxPAPC- and AITC-induced inflammatory responses were evaluated under normal and Ca^{2+} -free conditions using qRT-PCR (IL-6, IL-8, COX2), Luminex assays (IL-6/IL-8 release), and immunoblotting (COX2). Data were analyzed by one-way ANOVA with Tukey's post hoc test ($N \geq 5$).

Results

OxPAPC induced a rapid increase in cytosolic Ca^{2+} (2.89 $\pm$ 0.69 fold), which was suppressed 79.3 $\pm$ 7% by TRPA1 inhibition (A967079). OxPAPC significantly increased mRNA abundance of IL-6, IL-8, and COX2 (5.83 $\pm$ 1.6-, 31.4 $\pm$ 7.7-, and 20.12 $\pm$ 4.5-fold, respectively) and increases the secretion of the corresponding

cytokines release and COX2 protein abundance. Notably, selective TRPA1 activation with AITC was also sufficient to induce IL-6, but not IL-8 or COX2 mRNA. OxPAPC-induced IL-6, IL-8, and COX2 mRNA was suppressed by TRPA1 inhibition ($30\pm0.3\%$, $61.8\pm3.6\%$, and $89.2\pm4.7\%$) and in the absence of extracellular Ca^{2+} ($68.9\pm1.4\%$, $65.9\pm2.1\%$, and $83.53\pm4.2\%$). However, TRPA1 blockade did not impact OxPAPC-induced inflammatory mediator protein abundance (IL-8, $p=0.07$; IL-6, $p=0.08$; COX2, $p=0.1$).

Conclusion

TRPA1 mediates OxPAPC-induced Ca^{2+} signaling and inflammatory gene expression in HAFs, however, cytokine secretion involves additional TRPA1-independent mechanisms. These findings suggest that OxPAPC affects the function of airway resident cells and could have clinical and therapeutic implications.