Characterization of Epithelial Cells Derived Extracellular Vesicles for Studying Upper Respiratory Tract Inflammation

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Airway diseases are known for the excessive production of mucus and inflammatory levels. Nonetheless, most of the current literature has been on intracellular pathways rather than on the extracellular communication process. Extracellular vesicles (EVs), on the other hand, have been identified as significant elements in cell-to-cell communications, possibly corresponding with inflammation in the airway microenvironment. The aim of the current study is to isolate, identify, assess the connection to upper-airway inflammation, as well as the capabilities for the different EV isolation methods. The A-549 cells were grown in culture, while the EVs for these cells were isolated in the culture medium through size-exclusion chromatography (SEC), and in the precipitation-based separation (PBS) protocol. The isolated EVs were identified through nanoparticle tracking analysis (NTA) for size concentration, bicinchonic acid (BCA) assay for protein analysis, in combination with western blot analysis for the identification of the protein present in the isolated EVs. Both of the isolation techniques provided samples of the EVs in the correct size parameters. On the other hand, the isolation technique, SEC, provided lower purity with a larger number of EVs, while the PBS isolation provided higher purity amounts.

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