

Resource Summary Report

Generated by [dkNET](#) on Sep 14, 2026

Single-nucleus isolation from frozen human lung tissue for single-nucleus RNA-seq

DOI:10.17504/protocols.io.zu8f6zw

Type: Protocol

Proper Citation

Nikita Joshi, Alexander Misharin 2019. Single-nucleus isolation from frozen human lung tissue for single-nucleus RNA-seq. protocols.io dx.doi.org/10.17504/protocols.io.zu8f6zw

Protocol Information

URL: <https://dx.doi.org/10.17504/protocols.io.zu8f6zw>

Authors: Nikita Joshi, Alexander Misharin

Group: Human Cell Atlas Method Development Community

Summary: We aimed to develop a protocol for isolation of the single nuclei from the archival frozen human lung tissue suitable for single cell RNA-seq using standard 10x Genomics chemistry. The protocol utilizes standard nuclei isolation buffer (Nuclei EZ buffer) supplemented with RNase inhibitor and a protease inhibitor. For the homogenization step, we elected to use C Tube and GentleMACS tissue dissociation as a way to standardize the homogenization procedure. To maintain nuclei integrity we skipped washing steps and instead use large volumes of washing buffer and proceed to FACS sorting immediately after lysis for nuclei purification. This protocol produces a good yield of nuclei and diverse libraries, with multiple cell types being detected. While the number of detected genes and UMI per nuclei is lower than for single cell RNAseq, it is sufficient for identification of the specific cell types, including rare populations. Moreover, snRNAseq allowed resolution of the cell types that are intimately integrated into the lung matrix and otherwise hard to resolve: fibroblasts and type 1 alveolar epithelial cells. We thank Oni Basu (UChicago), Luciano Martelotto (Monash University), Nicole Abreu (10x Genomics) and Sharmila Chatterjee (10x Genomics) for their advice.

Affiliations: Northwestern University, Northwestern University

Version: 1

Publication Date: 2019

Proper Citation: Nikita Joshi, Alexander Misharin 2019. Single-nucleus isolation from frozen human lung tissue for single-nucleus RNA-seq. protocols.io

dx.doi.org/10.17504/protocols.io.zu8f6zw

Record Creation Time: 20210329T220520+0000

Record Last Update: 20210329T220848+0000

Data and Source Information

Source: