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Collection and shipment of specimen for single-nuclei RNA sequencing (snRNA-Seq)

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Cellular Senescence Net…

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We use this protocol and it's working

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Abstract

These samples are part of the JAX-Sen project in the SenNet Consortium. Here we provide details on specimen collection and shipment to the Robson laboratory at The Jackson Laboratory for Genomic Medicine (JAX-GM) in Farmington, CT for its processing for single-nuclei RNA-sequencing (snRNA-seq).

Reagents and Materials

1
- 2mL Cryovials/ screw-cap tubes
- ice cold 1X PBS
- Kimwipes
- Petri dishes
- Liquid Nitrogen
- Dry ice
- Tweezers (clean, sterile)

Quality Key Points:

2
- The tissue specimen should be kept at 4 degrees Celsius and RNase-free until snap-freezing and maintained at -80 degrees C or on dry ice thereafter, until it is processed at JAX.
- It is crucial to not store the tissue specimen at RT to avoid freeze-thaw cycles, any cell death, and tissue and/or RNA degradation.

Procedure:

3 **Timeline:** The daily shipping deadline at Jax BH is 12:00 noon. So, harvest the tissues before and closer to noon.

4 **Collection/Harvest:**

4.1 Animal was euthanized via cervical dislocation.

4.2 Animal was pinned to a necropsy tray which remained on wet ice throughout the harvest.

4.3 Animal was perfused with 20ml cold PBS.

4.4 Heart, pancreas, and kidney was collected and the specimen was collected and the specimen was rinsed with ice-cold 1x PBS in a Petri dish to remove blood and other debris.

4.5 Pat the specimen dry using clean Kimwipes and freeze as quickly as possible after harvest.

4.6 To flash freeze (snap freeze), place the tissue in a cryo-tube and submerge the tube in liquid nitrogen or immerse deeply in dry ice.

4.7 Wait at least 2-3 minutes for the tissue to completely freeze, and transfer the tube containing the tissue to -80 degrees C.

4.8 Keep at -80 degrees Celsius thereafter, until shipping.

5 **Shipment:**

5.1 Place sample tubes in a plastic box (cardboard boxes insulate the samples from the cold ice) or in double Ziplock bags after checking that they are completely sealed.

5.2 Ship the sample box on dry ice (O/N shipping) to:
The Jackson Laboratory for Genomic Medicine,
Farmington, CT, 06032