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Human Knee Meniscus Collection Protocol for scRNA-seq

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Protocol status: Working

We use this protocol and it's working

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compartment, scrna



Abstract

Meniscus is taken from the medial knee compartment is collected for scRNA-seq processing. The attached image indicates where the meniscal tissue is collected from.

Materials

Sterile drapes Sterile gauze Autoclaved tools Disposable #21 scalpels **DPBS** DMEM Antibiotic-Antimycotic 50mL conical tubes

Protocol materials

- DMEM with L-Glutamine 4.5g/L Glucose and Sodium Pyruvate Fisher Scientific Catalog #MT-10-013-CV
- Antibiotic-Antimycotic (100X) Thermo Fisher Scientific Catalog #15240062
- X PAXgene Tissue FIX Qiagen Catalog #765312
- X PAXgene Tissue STABILIZER Qiagen Catalog #765512

Before start

Knee blocks are shipped on wet ice and received within 48 hours post-mortem from certified tissue banks. The average time from receiving the sample to harvesting cartilage from both knees is about 2 hours. Pictures are taken of knee tissues and macroscopic scoring of the cartilage is completed.



- 1 Prepare harvesting area with sterile drapes, tools and gauze. All harvesting is completed within an aseptic environment.
- Wipe down the knee blocks with 95% ethanol prior to opening the joint capsule.
- Once the joint capsule is opened, the femur and tibia are disarticulated and menisci are resected.
- A full-thickness (sagittal) piece of medial meniscus is cut out using a #21 scalpel; the piece measures about 1.60cmX15mmx8mm and weighs around 1g. This is collected into a 50mL tube that is filled with 40mLs of
 - DMEM with L-Glutamine 4.5g/L Glucose and Sodium Pyruvate Fisher Scientific Catalog #MT-10-013-CV
 - + 1% Antibiotic-Antimycotic (100X) Thermo Fisher Scientific Catalog #15240062 , then placed in wet ice until the cell isolation protocol for scRNAseq begins.

Note

See Description Section for image for the area of interest.