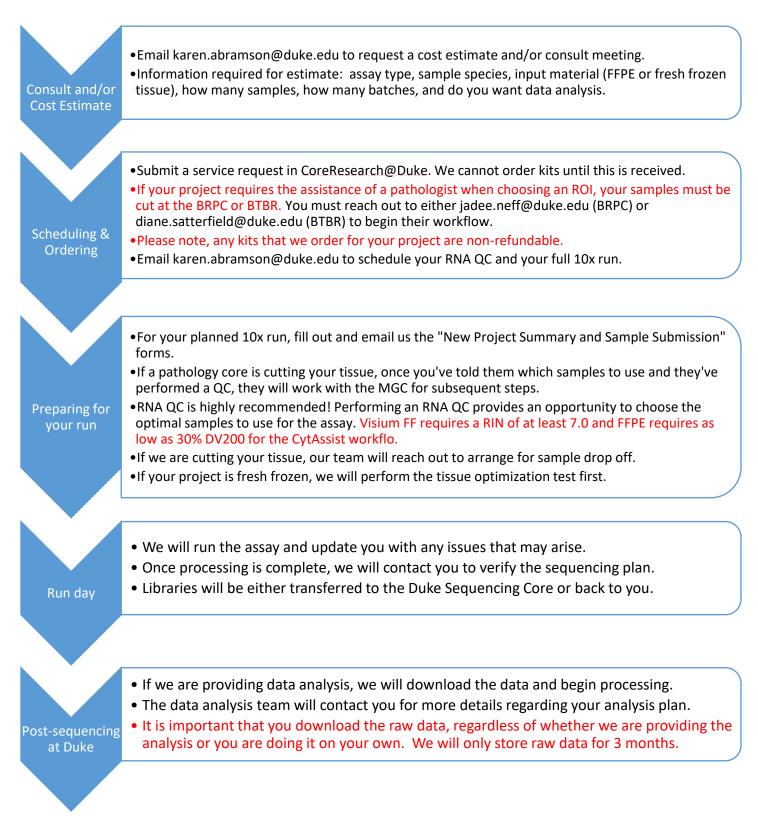
# Duke Molecular Genomics Core 10x Genomics Visium Project Workflow



## **Duke Molecular Genomics Core**

## **10x Genomic Visium Sample Preparation Guidelines**

### It is essential that you follow the sample preparation protocol provided by 10x Genomics® and the MGC.

The 10x Genomics<sup>®</sup> Visium fresh frozen assay uses poly-a capture technology to bind mRNA to the surface of a slide. How far the RNA travels from its original position is affected by how long the tissue is permeabilized. In order to get the most accurate spatial information and high-quality data, a tissue optimization assay should be performed to determine permeabilization time. Ideally this test is done on all tissues that may differ in pathology or phenotype. If this is impractical and you request to only test one representative tissue, we cannot guarantee the permeabilization will be optimized across the range of phenotypes/disease pathology in other tissues and this may affect the data.

The MGC does not have a pathologist on staff, any project requiring a pathologist to identify a region of interest must either be cut in your own lab or work with a pathology core. The BRPC or BTBR has been trained to cut slides for Visium and have workflows in place.

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BTBR (Diane Satterfield: diane.satterfield@duke.edu)

The quality of RNA in a sample is greatly affected by collection processing, storage and age of sample. It is recommended for new tissue collection to reduce time between collection of tissue and preservation. Extended time between collection and preservation will greatly reduce quality. Always keep fresh tissues on ice while processing. FFPE samples should be stored at 4C to best preserve quality.

It is recommended to optimize freezing protocols to reduce cracking, crystallization or tissue distortion. Following 10x's recommended guidelines for freezing is a good place to start.

### The best way to ensure good quality data is to strictly follow the provided sample preparation guidelines.

The full 10x Genomics' Tissue Preparation Guide can be found here:

CytAssist FFPE:

https://www.10xgenomics.com/support/spatial-gene-expression-ffpe/documentation/workflows/cytassist-ffpe/steps/tissue-prep/visium-cyt-assist-spatial-gene-expression-for-ffpe-tissue-preparation-guide

### Fresh Frozen:

https://www.10xgenomics.com/support/spatial-gene-expression-fresh-frozen/documentation/steps/tissue-prep/visium-spatial-protocols-tissue-preparation-guide

Please don't hesitate to ask questions! We're here to help! Please email <u>DMPI-MGC@dm.duke.edu</u> or <u>karen.abramson@duke.edu</u>.