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Sinai SCENT TMC - 10X Xenium

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ABSTRACT

Imagine obtaining histological insights into disease biology and gaining a molecular perspective on the subcellular spatial gene expression of 100s to 1,000s of RNA targets in the same tissue section?

With high-plex Xenium In Situ, you can. Choose from one of our customizable pre-validated panels or create a standalone panel targeted towards the biology of your disease. Since the Xenium workflow is non-destructive, you then have the option to perform H&E and/or immunofluorescent staining on the same section.

IMAGE ATTRIBUTION

10x Genomics

MATERIALS

Deparaffinization _ Decrosslinking.pdf 7MB
Overview.pdf 65KB
Probe Hybridization, Ligation _ Amplification.pdf 7.8MB
Protocol Planner.pdf 1.1MB
Xenium Analyzer.pdf 13.2MB
Xenium Tissue Preparation Guide FFPE.pdf 8.3MB
XeniumTissue Prep Fresh Frozen.pdf 4.9MB

BEFORE START INSTRUCTIONS

It is mandatory for all Xenium Users to fill out the log before using the Xenium instrument. Please fill in the information to the Xenium User Log at least one week before the planned Xenium run and send an e-mail to xenium@mssm.edu to notify others of your plan to use the instrument.

In the Notes column, include information about any irregularities that occurred during the sample processing or instrument initialization.

MANUSCRIPT CITATION:
https://www.10xgenomics.com/platforms/xenium?utm_medium=search&utm_medium=search&utm_source=google&utm_source=google&utm_content=website-page&utm_campaign=7011P000001Pw8ZQAS&utm_campaign=sem-goog-2022-website-page-ra_g-brand-mixed-amer&gclsrc=aw.ds&useroffertype=website-page&userresearcharea=ra_g&userregion=amer&userrecipient=customer&usercampaignid=7011P0000015V8HQAU&gad_source=1&gclid=Cj0KCQiAr8eqBhD3ARIsAIe-buMHK_uV19CzXzEZQVW6NgcSjvhHKSTBepObEVQABOG89jGu6sOqqrYaAk3sEALw_wcB

Protocol status: Working
We have on-site 10X Training, and the demo is working.

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91814

Keywords: Xenium

Overview and Intro

1 Protocol Planner.pdf1.1MB Overview.pdf65KB

Xenium In Situ measures gene expression in tissue sections derived from either formalin fixed and paraffin embedded (FFPE) or fresh frozen (FF) tissue samples placed on Xenium Slides. This Protocol Planner provides an overview of the workflow along with the Xenium Analyzer overview. To enable

efficient planning, a breakdown of key protocol steps and times, list of user-acquired reagents and consumables, and information about supporting documentation that will be available for executing the Xenium Gene Expression workflow is also provided.

Tissue Prep

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Deparaffinization _ Decrosslinking.pdf 7MB
Xenium Tissue Prep Fresh Frozen.pdf 4.9MB
Xenium Tissue Preparation Guide FFPE.pdf 8.3MB

Xenium In Situ for FFPE is designed to measure mRNA in tissue sections derived from formalin fixed & paraffin embedded (FFPE) tissue samples and requires a Xenium slide with intact tissue sections as input. This protocol outlines deparaffinization and decrosslinking of FFPE tissues for use with 10x Genomics Xenium In Situ Gene Expression protocols. Deparaffinized and decrosslinked tissue sections are inputs for the downstream Xenium In Situ Gene Expression - Probe Hybridization, Ligation & Amplification workflow.

Xenium In Situ for Fresh Frozen Tissues is designed to measure mRNA in tissue sections derived from fresh frozen (FF) and embedded tissue samples and requires a Xenium slide with intact tissue sections as input. Proper tissue handling, storage, and preparation techniques preserve the morphological quality of tissue sections and integrity of mRNA transcripts.

This Tissue Preparation Guide provides guidance on:
1. Freezing and embedding tissue samples prior to cryosectioning.
2. Best practices for handling tissue samples and Xenium slides before and after cryosectioning.
3. Hematoxylin and Eosin (H&E) staining to check tissue quality.
4. Cryosectioning of tissue samples and placement of sections on Xenium slides.

Probe Hybridization

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Probe Hybridization, Ligation _ Amplification.pdf 7.8MB

Xenium In Situ Gene Expression assays RNA at the subcellular level by using targeted probes in formalin fixed & paraffin embedded (FFPE) or fresh frozen (FF) tissue sections. FFPE tissue sections placed on Xenium Slides are deparaffinized and decrosslinked as described in Xenium In Situ for FFPE - Deparaffinization & Decrosslinking (Demonstrated Protocol – CG000580). FF tissue sections placed on Xenium slides are fixed and permeabilized as described in Xenium In Situ for Fresh Frozen - Fixation & Permeabilization (Demonstrated Protocol – CG000581).
Pre-designed, add-on custom, or standalone custom probe panels are then added to the tissue. Each circularizable DNA probe contains two regions that hybridize to the target RNA and a third region that encodes a gene-specific barcode. The two ends of the probes bind the target RNA and are ligated to generate a circular DNA probe. Following ligation, the circularized probe is enzymatically amplified, generating multiple copies of the gene-specific barcode for each RNA target.

Xenium slides containing FFPE or FF tissue sections are then loaded for imaging and analysis on the Xenium Analyzer instrument for high-throughput, automated in situ analysis. Fluorescently labeled oligos bind to the amplified DNA probes. Cyclical rounds of fluorescent probe hybridization, imaging, and removal generate optical signatures specific for each barcode, which are converted into a gene identity. Identified transcripts can be visualized using Xenium Explorer software.
This document outlines the protocol for generating Xenium In Situ Gene Expression data from FFPE and FF tissue sections placed on Sample Areas of a Xenium slide.

Xenium Analyzer

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Xenium Analyzer.pdf13.2MB

Xenium In Situ is the next-level in situ solution for subcellular profiling of hundreds of RNA targets. Xenium Analyzer combined with our curated and customizable panels, powerful visualization software, and easy-to-follow workflow is a powerful in situ profiling platform, revealing new insights into cellular structure and function.
Xenium In Situ provides highly sensitive, targeted gene expression information at subcellular resolution for hundreds of RNA targets, in fresh frozen (FF) and formalin fixed & paraffin embedded (FFPE) tissue. Primary analysis to decode image data to transcripts and secondary analysis to segment cells and assign transcripts is performed automatically, directly on-instrument. Using Xenium Explorer, view and explore this on-instrument output to see where cells belonging to clustering results are localized, annotate clusters based on expression patterns, overlay individual transcripts at sub-cellular resolution with morphology images and cell segmentation boundaries, and compare expression between selected regions. The key attributes of Xenium Analyzer are highlighted below.