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 We use this protocol and it's working

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🌐 HuBMAP | GE/Vanderbilt MALDI IMS and Cell DIVE™ Modality Overview

Nathan Heath Patterson¹, Elizabeth Neumann², Christine Surrette³, Soumya Ghose³, Liz McDonough³, Jamie Allen¹, Jeff Spraggins¹, Fiona Ginty³

¹Vanderbilt University; ²UC Davis; ³GE Research

Human BioMolecular Atlas Program (HuBMAP) Method Development Community

GE R



Liz McDonough
 GE Research

ABSTRACT

This is an overview of all protocols currently in use for the GE/Vanderbilt University Cell DIVE collaboration for the Human BioMolecular Atlas Program (HuBMAP). It includes links to each of the individual protocols that make up this project workflow.

MALDI IMS

1 Collection of post-surgical tissue.

Protocol



NAME

Collection and Post-Surgical Excision of Human Kidney Tissue through the Cooperative Human Tissue Network

CREATED BY

Jamie Allen

PREVIEW

2 Stabilize and freeze tissues.

Protocol



NAME

Freezing Fresh Tissue

CREATED BY

Jamie Allen

PREVIEW

3 Cryosection tissues into micrometer thick sections, alternating between thaw mounting onto indium tin-oxide and positively charged glass slides (proceed to step 4), or collecting several tissue sections within an microcentrifuge tube for proteomics analysis.

Protocol



NAME

Cryostat Sectioning of Tissues for 3D Multimodal Molecular Imaging

CREATED BY

Jamie Allen

PREVIEW

4 Perform autofluorescence microscopy on all tissue sections

Protocol



NAME

Autofluorescence Microscopy Data Acquisition

CREATED BY

Jamie Allen

PREVIEW

5 Perform Matrix Application

Protocol



NAME

Automatic Deposition of CHCA Matrix for MALDI Analysis of Lipids

CREATED BY

Jamie Allen

PREVIEW

6 Perform high resolution IMS analysis of matrix coated tissue sections.

Protocol



NAME

High Resolution Imaging Mass Spectrometry Analysis using Bruker Daltonics Platforms

CREATED BY

Jamie Allen

PREVIEW

7 Obtain autofluorescence microscopy images of tissues after IMS analysis

Protocol



NAME

Post-IMS Autofluorescence Microscopy

CREATED BY

Jamie Allen

PREVIEW

Preparing Sample for MxIF

8 Perform Matrix Removal & Tissue Fixation

Protocol



NAME

Fixation Protocol for Fresh Frozen Tissue Samples (post-MALDI)

CREATED BY

Jamie Allen

PREVIEW

Cell DIVE

9 Characterize antibodies (primary/secondary, direct conjugates, and zenon labelled antibodies) and determine any antigen effects from the Cell DIVE dye inactivation process.

[Cell DIVE™ Platform | Antibody Characterization for Multiplexing](#)

[Cell DIVE™ Platform | Antibody Staining & Imaging](#)

10 Prepare direct conjugates for study.

[Cell DIVE™ Platform | Antibody Purification Chemistry](#)

[Cell DIVE™ Platform | Ab Conjugation: Initial Conjugation & Scale up Conjugation](#)

11 Perform Cell DIVE™ multiplexed data acquisition on the final cohort.

Note

Staining is done manually using a humidity chamber and images are acquired on the Leica Cell DIVE imager utilizing a coverslipless imaging approach