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Version 3

VU Biomolecular Multimodal Imaging Center (BIOMIC) kidney characterization pipeline for tissues collected through the Cooperative Human Tissue Network (CHTN) V.3

DOI

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Protocol status: Working

We use this protocol and it's working

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Keywords: HuBMAP, BIOMIC, Vanderbilt, Multimodal Imaging, IMS, MxIF, Proteomics, Image Registration, kidney characterization pipeline for tissue, kidney characterization pipeline, vanderbilt tissue mapping center, vanderbilt tissue mapping center as part, human biomolecular atlas program, atlas of human organ, vu biomolecular multimodal imaging center, cooperative human tissue network, kidney, human organ, such as the kidney, tissue, atlas

Abstract

We aim to develop high resolution, chemically informative imaging methodologies for building an atlas of human organs, such as the kidney.

Scope:

Provide an overview of the methods used by the Vanderbilt Tissue Mapping Center as part of the Human Biomolecular Atlas Program (HuBMAP, NIH Common Fund) and contextualize individual protocols within our larger workflow.



- 1 Collection of post-surgical tissue.
Collection: dx.doi.org/10.17504/protocols.io.7gehjte
- 2 Stabilize and freeze tissues.
Freezing Tissue: dx.doi.org/10.17504/protocols.io.6wghfbw
- 3 Initial Rapid Pathology Assessment of Kidney Tissue
Staining: dx.doi.org/10.17504/protocols.io.4qngvve
Assessment: dx.doi.org/10.17504/protocols.io.9dph25n
- 4 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 a microcentrifuge tube for proteomics analysis.
Cryosectioning: dx.doi.org/10.17504/protocols.io.7ethjen
Proteomics: dx.doi.org/10.17504/protocols.io.67nhhme
- 5 Perform autofluorescence microscopy on all tissue sections before IMS (step 5) or MxIF analysis (step 9)
Autofluorescence: dx.doi.org/10.17504/protocols.io.7e3hjgn
- 6 Coat tissue sections with MALDI matrix for IMS analysis.
Matrix Application: dx.doi.org/10.17504/protocols.io.4srgwd6
- 7 Perform high resolution IMS analysis of matrix coated tissue sections.
IMS: dx.doi.org/10.17504/protocols.io.7gdhjs6
- 8 Perform fluorescence microscopy to visualize laser ablation spots.
Post IMS AF: dx.doi.org/10.17504/protocols.io.879hze6
- 9 Annotation of Lipids from IMS Data
Cal & Annotate: dx.doi.org/10.17504/protocols.io.864hzgw
- 10 Remove MALDI matrix and perform PAS staining.
PAS Staining: dx.doi.org/10.17504/protocols.io.4qngvve
- 11 Alternatively, MxIF can be performed after step 4.
Antibody labeling: dx.doi.org/10.17504/protocols.io.667hhhn
MxIF: dx.doi.org/10.17504/protocols.io.665hhg6



- 12 Registration of autofluorescence images from both IMS and MxIF sections allow for the direct correlation of the two orthogonal approaches.
Registration: <https://dx.doi.org/10.17504/protocols.io.bed2ja8e>
Publication: <https://doi.org/10.1021/acs.analchem.8b02884>
- 13 MALDI imaging mass spectrometry data processing.
Data Processing: <https://dx.doi.org/10.17504/protocols.io.bed3ja8n>
- 14 RNA Assessment from tissue.
RNA extraction: dx.doi.org/10.17504/protocols.io.86nhzde