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

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2 Works for me dx.doi.org/10.17504/protocols.io.7hhhj36

VU Biomolecular Multimodal Imaging Center
Human BioMolecular Atlas Program (HuBMAP) Method Development Community

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ABSTRACT

We aim to develop high resolution, chemically informative imaginig 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 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.
Cryosectioning: dx.doi.org/10.17504/protocols.io.7ethjen
Proteomics: dx.doi.org/10.17504/protocols.io.67nhhme

4 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

5 Coat tissue sections with MALDI matrix for IMS analysis.
Matrix Application: dx.doi.org/10.17504/protocols.io.4srgwd6

6 Perform high resolution IMS analysis of matrix coated tissue sections.
IMS: dx.doi.org/10.17504/protocols.io.7gdhjs6

7 Perform fluorescence microscopy to visualize laser ablation spots.

8 Remove MALDI matrix and perform PAS staining.
PAS Staining: dx.doi.org/10.17504/protocols.io.4qngvve

9 Alternatively, MxIF can be peformed after step 4.
Antibody labeling: dx.doi.org/10.17504/protocols.io.667hhhn
MxIF: dx.doi.org/10.17504/protocols.io.665hhg6

10 Registration of autofluorescence images from both IMS and MxIF sections allow for the direct correlation of the two orthogonal approaches.
Registration: https://doi.org/10.1021/acs.analchem.8b02884