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VU Biomolecular Multimodal Imaging Center (BIOMIC) kidney characterization pipeline for tissues collected through the Cooperative Human Tissue Network (CHTN) V.4

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VU Biomolecular Multim... Human BioMolecular Atl...

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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

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 Intial 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 an microcentrifuge tube for proteomics analysis.
Cryosectioning: dx.doi.org/10.17504/protocols.io.7ethjen

5 Perform LC based Proteomics on adjacent tissue sections and/or continue to Step 6.
Sample Preparation: dx.doi.org/10.17504/protocols.io.67nhhme
Data Acquisition: dx.doi.org/10.17504/protocols.io.bfsjjncn
Data Analysis: dx.doi.org/10.17504/protocols.io.bfshjnb6

6 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

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

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

9 Perform fluorescence microscopy to visualize laser ablation spots.
Post IMS AF: dx.doi.org/10.17504/protocols.io.879hzr6

10 Annotation of Lipids from IMS Data
Cal & Annotate: dx.doi.org/10.17504/protocols.io.864hzgw

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

12 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**

13 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

14 MALDI imaging mass spectrometry data processing.
Data Processing: https://dx.doi.org/10.17504/protocols.io.bed3ja8n

15 RNA Assessment from tissue.
RNA extraction: **dx.doi.org/10.17504/protocols.io.86nhzde**