VERSION 4

APR 29, 2020

OPEN ACCESS

DOI:
dx.doi.org/10.17504/protocols.io.bfskjcw

Protocol Citation: Elizabeth Neumann, Jamie Allen, Maya Brewer, David Anderson, Mark De Caestecker, Danielle Gutierrez, Jeff Spraggins 2020. VU Biomolecular Multimodal Imaging Center (BIOMIC) kidney characterization pipeline for tissues collected through the Cooperative Human Tissue Network (CHTN).
protocols.io
<https://dx.doi.org/10.17504/protocols.io.bfskjcw>

License: This is an open access protocol distributed under the terms of the [Creative Commons Attribution License](https://creativecommons.org/licenses/by/4.0/), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited

Protocol status: Working
We use this protocol and it's working

Created: Apr 29, 2020

Last Modified: Apr 08, 2023

PROTOCOL integer ID:
36396

Keywords: HuBMAP, BIOMIC, Vanderbilt, Multimodal Imaging, IMS, MxIF, Proteomics, Image Registration

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

Elizabeth Neumann¹, Jamie Allen¹, Maya Brewer¹, David Anderson¹, Mark De Caestecker², Danielle Gutierrez¹, Jeff Spraggins¹

¹Vanderbilt University; ²Division of Nephrology, Vanderbilt University Medical Center

VU Biomolecular Multimodal Imaging Center / Spraggins Research Group

Human BioMolecular Atlas Program (HuBMAP) Method Development

Jamie Allen
Vanderbilt University

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

- 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.879hizr6
- 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 performed after step 4.
Antibody labeling: dx.doi.org/10.17504/protocols.io.667hghn
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