VERSION 1

AUG 28, 2017

OPEN ACCESS

DOI:

dx.doi.org/10.17504/protocols.io.imzcc76

Protocol Citation: Kai Song, Jianxin Fu, Jianhua Song, Brett H Herzog, Kirk Bergstrom, Yuji Kondo, J. Michael McDaniel, Samuel McGee, Robert Silasi-Mansat, Florea Lupu, Hong Chen, Harini Bagavant, and Lijun Xia 2017. ultrastructural analysis. **protocols.io** <https://dx.doi.org/10.17504/protocols.io.imzcc76>

MANUSCRIPT CITATION:

[J Biol Chem](#). 2017 Aug 25. pii: jbc.M117.798512. doi: 10.1074/jbc.M117.798512. [Epub ahead of print]

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

Protocol status: Working

Created: Aug 25, 2017

🌐 ultrastructural analysis V.1

Kai Song^{1,2}, Jianxin Fu³, Jianhua Song³, Brett H Herzog³, Kirk Bergstrom³, Yuji Kondo³, J. Michael McDaniel³, Samuel McGee³, Robert Silasi-Mansat³, Florea Lupu³, Hong Chen², Harini Bagavant³, and Lijun Xia³

¹Oklahoma Medical Research Foundation;

²Boston Children's Hospital, United States.;

³Oklahoma Medical Research Foundation, United States.

Kai Song

ABSTRACT

Mouse kidney tissues were harvested, fixed in 4% paraformaldehyde (PFA) and embedded in paraffin. All sections (3 micrometers in thickness) were stained with hematoxylin and eosin (H&E), periodic acid-Schiff (PAS, Thermo Scientific) or Masson's trichrome staining (MTS, Sigma) according to the standard procedures or manufacturer's instructions. For transmission electro micrographs (EM), kidneys were fixed in Karnovsky's fixative containing 2% paraformaldehyde and 2.5% glutaraldehyde in 0.1% cacodylate buffer, pH 7.2 for 2 hrs at room temperature, followed by postfixation with 1% osmunim tetraoxide in cacodylate buffer for 90 min at 4°C and mordanting with 1% tannic acid in cacodylate buffer.. Ultrathin sections were contrasted with uranyl acetate and lead citrate, and examined with a Hitachi H-7600 transmission electron microscope located in the Imaging Core Facility of the Oklahoma Medical Research Foundation. We followed previously described histological scoring methods (33).

PROTOCOL integer ID:
6553

- 1** For transmission electro micrographs (EM), kidneys were fixed in Karnovsky's fixative containing 2% paraformaldehyde and 2.5% glutaraldehyde in 0.1% cacodylate buffer, pH 7.2 for 2 hrs at room temperature,
- 2** followed by postfixation with 1% osmunim tetroxide in cacodylate buffer for 90 min at 4°C and mordanting with 1% tannic acid in cacodylate buffer.
- 3** Ultrathin sections were contrasted with uranyl acetate and lead citrate, and examined with a Hitachi H-7600 transmission electron microscope located in the Imaging Core Facility of the Oklahoma Medical Research Foundation.