Preparation of Renal Biopsy Specimens for Light, Immunofluorescence and Electron Microscopy

Department of Pathology, UCSF, (415) 353-2673

TISSUE SAMPLING:

In order to secure sufficient tissue for all technical procedures, we recommended that two renal biopsy cores be taken and subdivided as in the example illustrated below for light(LM), immunofluorescence(IF}, and electron microscopy(EM}:

If possible use a stereomicroscope to evaluate the core and identify the areas with glomeruli.

Two Cores taken:

| Core 1 | cortical end | medullary end | I |
|--------|--------------|---------------|---|
| | EM: 1/3 Core | LM: 2/3 Core | |
| Core 2 | | | |
| | IF: 1/3 Core | LM: 2/3 Core | |

One core taken

| cortical end | | me | dullary end |
|-----------------|----------------|---------------|-------------|
| EM : 2mm | IF: 2mm | LM: Remainder | |

TISSUE PRESERVATION:

Portions are divided and placed in small vials of fixatives, i.e. 4-5 ml in a 10 ml vial, as outlined below.

Critical Tissue-Handling Notes:

- Care must be taken not to cross-contaminate the EM and IF solutions.
- Contamination may occur from using the same tool to transfer tissues to each fixative.
- It is preferable to use separate tools for transferring tissues into EM and IF solutions.
- Take the immunofluorescence specimen first since EM or LM fixatives will adversely affect the immunofluorescence tests.
- Likewise, avoid transferring tissue from IF fixative to the EM vial or using the same forceps without cleaning them. If fixative may adversely affect the EM test.

a.

1. Light Microscopy (LM):

a. Place the tissue in 10% Neutral Buffered Formalin (NBF) for shipment to our laboratory for light microscopy processing.

2. Immunofluorescence Microscopy (IF):

- Preferred Method:
 - i. Place the tissue for immunofluorescence in immunotransport solution.
 - 1. Zeuss IF Transport Fluid, Wampole Laboratories, phone 800-257-9525.
 - ii. Vials of transport solution will be supplied by our lab upon request.
 - iii. Use of this solution eliminates the problems associated with handling and transport of frozen tissue, i.e., desiccation or thawing of tissue.
 - iv. Refrigerate the specimen until shipment to the lab.
 - v. Do not freeze the tissue in this media or store longer than 4 days in IF Fixative.

-OR-

b. Alternate Method

- i. If no transport solution is available:
 - 1. Place the tissue covered by a drop of OCT compound directly on a small piece of aluminum foil, fold the foil around the tissue and tightly fold over the edges 2-3 times to seal the tissue and prevent desiccation.
 - 2. Immediately freeze on dry ice, and place in a Styrofoam carton of dry ice for shipment to our laboratory.
 - 3. Once the tissue has been frozen, it must not be allowed to thaw.
 - 4. Vials containing the LM and EM tissues must be kept separated from the carton containing the frozen IF sample to avoid freezing them.

3. Electron Microscopy (EM):

- a. The specimen for EM should be placed in EM fixative as soon as possible to minimize artifact.
- b. The sample should be 3-5 mm (needle core length) in aggregate.
- c. The tissue should be refrigerated until shipment to our laboratory.
- d. Buffered glutaraldehyde (EM fixative) may be obtained from our laboratory upon request.
- e. Vials of ready-to-use EM fixative are also available from commercial vendors.

SHIPMENT OF TISSUE:

- Messenger or taxicab (same day),
- Federal Express (First Overnight, in order to meet rapid-processing cutoff at 10 AM),
- US Mail (2-4 days: least desirable choice)

Address:

| When using FedEx, UPS or Taxi | When sending by US Mail |
|--------------------------------|--------------------------------|
| UCSF | UCSF |
| Electron Microscopy Laboratory | Electron Microscopy Laboratory |
| Pathology Department | Pathology Department, Box 0102 |
| 513 Parnassus Ave, Room S-570 | 505 Parnassus Ave, Room M-545 |
| San Francisco, CA 94143 | San Francisco, CA 94143 |
| | |

Phone: 415-353-2673 (call for fixatives)

FAX: 415-514-3403

Request Form and Instructions download: https://pathology.ucsf.edu/consultation-services/renalpath

NOTE: We recommended that you contact our laboratory so that we will be aware that the specimen is coming.

Request Form, Patient demographics and authorizations may be faxed ahead for faster registration and accessioning.