

PROTOCOL FOR MUSCLE & NERVE BIOPSIES

The Department of Pathology at the University of Louisville, in Louisville, Kentucky, currently offers histochemical evaluation of muscle biopsy specimens and electron microscopic evaluation, if indicated. Proper handling of the muscle biopsy, especially proper freezing, will help ensure a correct diagnosis for the patient; therefore, the following protocol should be observed.

Due to the limitations of our laboratory, we DO NOT accept fresh specimens containing INFECTIOUS AGENTS: such as, but not limited to, hepatitis, human immunodeficiency virus (HIV), tuberculosis (TB), or Creutzfeldt-Jakob disease (CJD).

MUSCLE BIOPSY

1. **SCHEDULING** - Notify the Special Procedures Laboratory (SPL), (502) 852-5519, 24 hours in advance of the biopsy. It is the responsibility of the surgeon or his office, the surgery department where the biopsy is being performed, and the submitting Pathology Laboratory to see that the muscle biopsy is scheduled with SPL.
2. **TRANSPORTATION** - The courier from the UofL Pathology Department will pick up muscle biopsies from the hospitals within the Medical Center complex. All other hospitals should send the specimen, *on wet ice*, via a local courier. **ALL SPECIMENS MUST BE LABELED WITH THE BIOHAZARD LABEL AS PER THE KENTUCKY BLOODBORNE PATHOGENS STANDARD #1910.1030. ALL SPECIMENS MUST BE LABELED WITH THE PATHOLOGY ACCESSION NUMBER.**
3. **AVAILABLE** - Biopsies may be scheduled between 8:00AM and 2:00 PM, Monday through Friday, excluding holidays. To properly handle 'emergency' or otherwise unscheduled biopsies, please contact SPL.
4. **MUSCLE** - The biopsy specimen for frozen section enzyme histochemistry should be UNCLAMPED and UNDAMAGED (e.g., tied to tongue blades or squeezed with forceps.), UNCAUTERIZED, NOT PREVIOUSLY DISSECTED, and NOT CHOPPED. It should be placed on gauze that has been *slightly* dampened with saline. A muscle biopsy 7-10 mm in diameter is optimal, comparable to a pencil eraser. **DO NOT place muscle, at any time, in or near any gross area containing the liquid or fumes of formalin, glutaraldehyde, or alcohols as these will alter the enzymes.**
5. **PATIENT INFORMATION** – As required by the College of American Pathologists (CAP), specimens must be accompanied with a completed requisition form, which includes patient identification, hospital numbers, pathology accession number, appropriate clinical information, and patient history. Billing and insurance information is also necessary.

7. **CANCELATION** – Should a scheduled muscle biopsy be canceled, please notify SPL as soon as possible.
8. **STAINS** - The stains routinely performed for diagnosis include: H&E, modified Gomori trichrome, oil red O, Congo red, PAS, PAS-D, ATPase (4.3, 9.4), SDH, NADH, COX, PFK, NE, APP, MPP, and AD.
9. **SNAP FREEZE** - Specimen will be frozen and stored at -70 °C according to CAP guidelines.
10. **REPORT** - In general, **a verbal or written report will be available within 4 working days of receipt of the specimen.**

OTHER INFORMATION

1. **Indications for muscle biopsy**
 - a. Unexplained muscle weakness.
 - b. Unexplained muscle pain.
 - c. Evaluation of connective tissue disorder.
 - d. Abnormality of muscle contraction such as myotonia or hypotonia.
2. **Before biopsy** - Before a muscle biopsy, certain points should be considered. The muscle selected for biopsy should be the least affected but definitely affected. Severely fibrotic or atrophic muscle should be avoided. Sites which have been previously traumatized by biopsy, injection, electromyography, or trauma should be avoided. Determine if muscle is to be obtained for special enzyme histochemistry studies, biochemical analysis, electron microscopy, routine paraffin processing, all of these, or any combination of these.

NERVE BIOPSY

1. Nerve biopsies should contain the same information/paperwork as muscle biopsies.
2. Nerve biopsies are to be sent fresh on barely moistened saline gauze.
3. Stains include: H&E, Masson trichrome, LFB/PAS, Congo red, and neurofilament via immunohistochemistry.
4. Electron microscopy with toluidine blue thick-sections will be performed.