Muscle Biopsy Tissue Submission Protocol

Please notify the MLabs Client Services Center prior to sending a muscle biopsy specimen. Muscle biopsy collection kits are available from MLabs and will be required in advance of the procedure. MLabs Client Services can be reached at 800.862.7284 or 734.936.2598 (local).

**Preparation Instructions**

1. The muscle biopsy should be taken from the belly of a moderately affected muscle (avoid EMG and injection sites). Ideally, the muscle biopsy is 2.5 – 3.0 cm length by 1.0 cm width for an adult or 2.0 cm length by 0.5 cm width for a child. Divide the muscle into three portions:
   a. 1.5 cm fresh (ideal) tissue portion on a saline-moistened Telfa gauze received on wet ice at refrigerator temperature.
   b. 1.0 cm in 10% neutral buffered formalin (for Histology).
   c. 0.5 cm in glutaraldehyde (for Electron Microscopy).

2. The fresh tissue, once received, will be frozen by MLabs. This specimen is most important for diagnosis as it is used for histochemical stains.

3. If the muscle biopsy is from a child, omit the formalin fixed specimen and provide two fresh (or frozen) specimens along with the glutaraldehyde fixed specimen.

4. Please complete the MLabs Muscle/Renal/Nerve Biopsy Requisition that is provided in the Muscle Biopsy collection kit and send the completed requisition back to MLabs along with the specimens.

**Specimen Handling**

Fresh tissue specimen is preferred provided that the specimen can arrive at MLabs within 4 hours of collection, Monday through Friday 7:00 am – 5:00 pm. Otherwise, the tissue must be frozen (see frozen instructions).

1. **FRESH TISSUE**
   The first piece of muscle, 1.5 cm length by 1.0 cm width will be used for histochemical stains. Do not tease or pinch with forceps or let the tissue dry out. Wrap the tissue in a saline-moistened Telfa pad and place in a screw cap container. Do not immerse the tissue in saline. Container should be kept cold (wet ice) in transit; **do not freeze**.

   If the specimen cannot arrive at MLabs within 4 hours of collection, it must be frozen in the following manner:
2. **FROZEN TISSUE**  
If liquid nitrogen is available:

a. Isopentane method (PREFERRED): Add 50-100 mL of isopentane (2-methylbutane) to a Nalgene or metal beaker. Suspend the beaker in a bath of liquid nitrogen and wait until the isopentane freezes to a white, chalky substance. Remove the beaker from the liquid nitrogen bath. Carefully drop or set the muscle on the frozen isopentane while keeping the muscle fibers straight and running in the same direction. Freeze the muscle for about 10 – 15 seconds. Place the frozen muscle in a Styrofoam container filled with 7 lbs of dry ice or in a -70°C freezer.

b. If isopentane is not available, roll the muscle gently in talc or glove powder, wrap it loosely in a piece of foil (do not flatten the muscle!), and immerse it into liquid nitrogen for 20 – 30 seconds. Place the frozen muscle in a Styrofoam container filled with 7 lbs of dry ice or in a -70°C freezer until ready to ship. **Avoid any thawing of specimen!** If it thaws and re-freezes, the specimen may not be salvageable.

If liquid nitrogen is not available: roll the muscle gently in talc or glove powder, wrap it loosely in a piece of foil (do not flatten the muscle!) and place in a Styrofoam container filled with 7 lbs of dry ice. **Avoid any thawing of specimen!**

3. **FORMalin FIXED TISSUE**  
The second piece of muscle, approximately 1.0 cm length by 1.0 cm width will be used for paraffin sections. Stretch the muscle longitudinally on a piece of tongue blade using sutures tied at either end, then place specimen side down in a container with 10% neutral buffered formalin (NBF). If the muscle biopsy is from a CHILD, omit the formalin fixed specimen; instead provide two fresh (or frozen specimens) along with the glutaraldehyde fixed specimen. Specimen should be sent room temperature, **do not freeze.**

4. **GLUTARALDEHYDE FIXED TISSUE**  
For the third piece of muscle tissue, 0.5 cm for electron microscopy, use a metal clamp or small piece of a sterile wooden tongue blade (size of a toothpick) to suture to a 3 mm thin portion of muscle in situ. Place this sample with the clamp or tongue blade attached in a vial containing 3% glutaraldehyde / 3% formaldehyde. Specimen should be sent at room temperature, **do not freeze.**

**Required Information and Shipping**

1. Label the containers with the patient’s name and a 2nd patient identifier such as a date of birth or a medical record number.
2. Complete the Renal/Muscle/Nerve Biopsy Patient Clinical Requisition and the [MLabs Muscle/Nerve/Renal Requisition](mailto:mlabs.umich.edu) to accompany the specimens.
3. The formalin and glutaraldehyde fixed specimens must be shipped in a room temperature container, separate from cold or frozen material. Care must be taken that these specimens do not freeze. Do not place these in the Styrofoam container with wet or dry ice.

4. Once the formalin and glutaraldehyde specimens have been placed in their liquid containers and the tops secured, place inside the ziploc specimen biohazard bag with the absorbent sheet and seal. Wrap the bag with bubble wrap and place inside transport box. Please be careful to not contaminate the outside of the transport bags, paperwork or box when handling. Transport at room temperature.

5. If shipping by Federal Express, place closed box inside Clinical Pak and seal according to instructions on the Clinical Pak. If shipping with FedEx, call 800.463.3339 or UPS, call 800.742.5877 to schedule pick up. For Friday specimens, clearly indicate Saturday Delivery required.

QUESTIONS?
If you have any questions regarding specimen collection, processing or transportation, please call MLabs Client Services at 800.862.7284. Alternatively, you may call the Nerve/Muscle laboratory directly at 734.764.8170.