

Experiment AM-1: Skeletal Muscle, Weight and Work

Equipment Required

PC Computer

IWX/214, USB cable, power supply

DT-475 Displacement transducer

A-BST-100 Stimulating electrodes

Female BNC to Dual Banana Adapter

Ring stand and clamps

Femur clamp

Thread

6" Ruler

Set of weights and pan

Amphibian Ringer's solution (See appendix)

Start the Software

1. Click on LabScribe
2. Click Settings → Animal Muscle → SkeletalMuscle-Weight-Work
3. Once the settings file has been loaded, click the **Experiment** button on the toolbar to open any of the following documents:
 - Appendix
 - Background
 - Labs
 - Setup (opens automatically)

DT-475 and Stimulus Electrode Setup

1. Locate the following items in the iWorx kit: DT-475 displacement transducer ([Figure AM-1-S1](#)); A-BST-100 bipolar stimulator cable ([Figure AM-1-S2](#)); Male double banana-female BNC adapter ([Figure AM-1-S3](#)).



Figure AM-1-S1: The DT-475 displacement transducer.



Figure AM-1-S2: The A-BST-100 bipolar stimulating electrode.

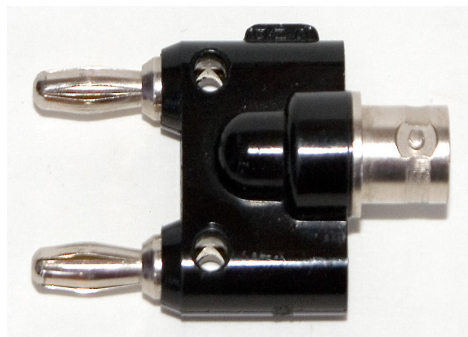


Figure AM-1-S3: Female BNC to Dual Banana Adapter.

2. Plug the DIN8 connector of the DT-475 into the Channel 3 input ([Figure AM-1-S4](#)).
3. Plug the male double banana-female BNC adapter into the positive (red) and negative (black) banana jacks of the IWX/214 stimulator. The banana plug that goes into the negative (black) stimulator output is identified by a tab, embossed with the letters GND (ground), on that side of the adapter.
4. Attach the BNC connector of the A-BST-100 bipolar stimulator cable to the adapter on the stimulator outputs.

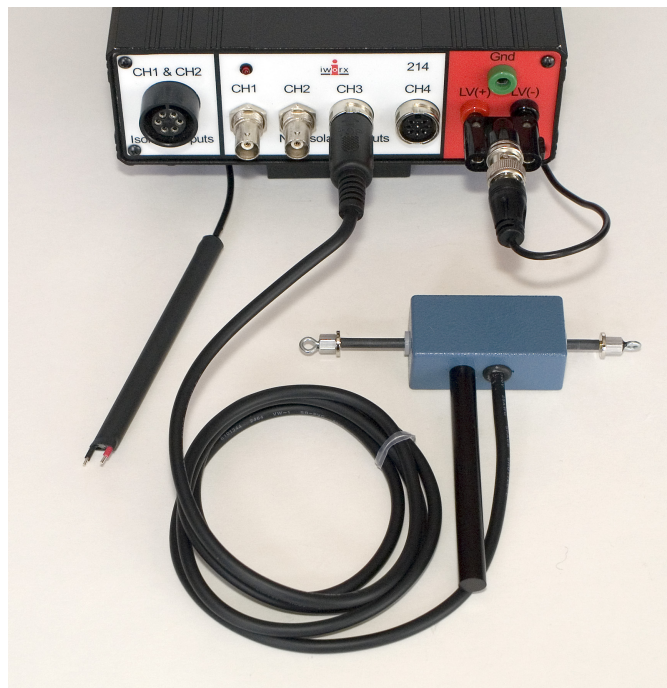


Figure AM-1-S4: The DT-475 displacement transducer and the A-BST-100 bipolar stimulating electrode connected to the IWx/214.

Note: *The DT-475 and the A-BST-100 can be connected to any of the iWorx recording devices. This illustration is just an example.*

The Dissection

1. Place a frog in ice water for 15 minutes. Double pith the frog as soon as it is removed from the ice water.
2. Remove the skin from the legs by making an incision through the skin around the entire lower abdomen. Cut the connections between the skin and the body—especially around the base of the pelvic girdle. Use stout forceps to pull the skin off the frog in one piece (like a pair of pants).
3. Place the frog in a dissection tray with its dorsal side up.

Note: *Moisten the exposed limbs of the frog with Ringer's solution every five minutes or so.*

4. Identify the Gastrocnemius muscle on the lower leg.
5. Use a glass hook to separate the Gastrocnemius muscle from the bone and other muscles of the lower leg.
6. Use scissors to free the Achilles tendon from the connective tissue around the heel of the foot. Double up a 24" piece of thread. Firmly tie the doubled thread around the Achilles tendon, leaving the ends of the thread long enough to attach the muscle to the displacement transducer.

Note: Isolate as much tendon as possible, since it will be used to attach the muscle to the transducer.

7. Cut the Achilles tendon as close to the bottom of the foot as possible, so the thread is still attached to the Gastrocnemius muscle.
8. Move the Gastrocnemius muscle away from the rest of the lower leg. Cut the tibia just below the knee to separate the rest of the lower leg from the preparation. Rinse the preparation with Ringer's solution to moisten the tissue and rinse away any blood.
9. Dissect away the muscles of the upper leg and expose the femur. Use a stout pair of scissors to cut the femur close to the pelvis. Rinse the preparation with Ringer's solution to moisten the tissue and rinse away any blood.

The Preparation

1. Use the femur clamp to mount the preparation on the ringstand ([Figure AM-1-S5](#)).
2. Attach the thread on the Achilles tendon to the upper eyelet on the rod of the displacement transducer.
3. Use a paper clip to attach the weight pan to the lower eyelet on the rod.
4. Adjust the femur clamp and the displacement transducer so the thread from the Achilles tendon to the transducer rod is vertical.

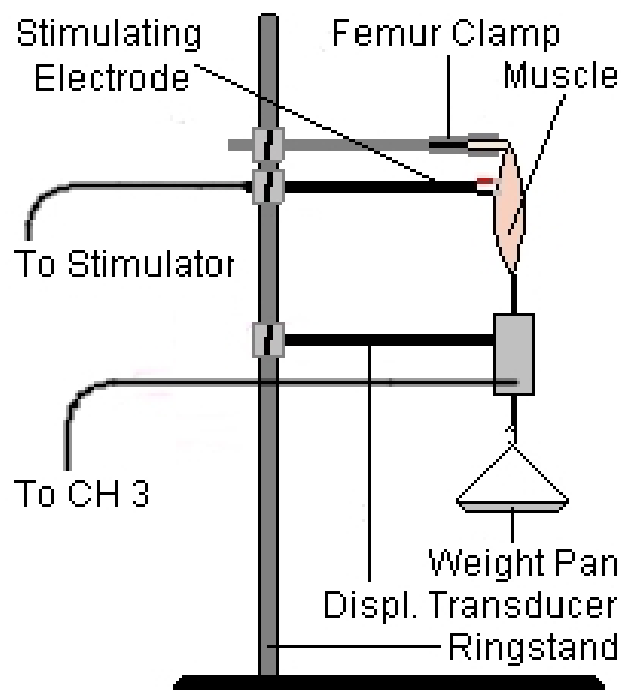


Figure AM-1-S5: The arrangement of the equipment used to evoke and record contractions from the frog Gastrocnemius muscle.

5. To prevent the weight in the pan from stretching the muscle, the knob on the upper end of the rod should be resting on the bushing in the top of the transducer case.
6. Position the stimulating electrodes so they lay against the muscle about midway between the knee and the tendon. The two electrodes should not touch one another.
7. Place two nickels (10 g) in the weight pan.

Warning: The muscle preparation used in this experiment is functional for a limited period of time. If the muscle is bathed periodically in Ringer's solution, it will work for about four hours. To conserve time, complete all the exercises in the experiment before analyzing the data.