

Activity 4.2.4: The Laws of Contraction

Introduction

You have now seen how big muscle groups work. We know that muscles contract and get shorter, but what does that really mean? How is it possible that our muscles constantly change size? Imagine you could explore deep into the structure of a muscle. You zoom in on the fiber and focus on the smallest muscle unit, the sarcomere. On the most basic level, muscle is made up of a special set of proteins that when triggered change in a way that causes this shortening. But what triggers this change?

In this experiment, you will stimulate the contraction of actual skeletal muscle tissue. Observe the reaction of muscle to various solutions and use your results to determine the optimal energy requirements for contraction. In the next activity, you will use your findings as well as additional research to provide a snapshot of what actually happens when you zoom in on a contracting muscle.

Equipment

* *Carolina Biological Supply* *ATP Muscle Kit*
* Glycerinated skeletal muscle
* Various ATP/salt solutions
* Microscope slides and coverslips
* Plastic Petri dishes
* Distilled water
* Test tubes and test tube rack
* Disposable transfer pipettes (1ml)
* Laboratory journal
* Teasing needles (from dissection kits) or straight pins
* Forceps or tweezers
* Millimeter ruler
* Microscope
* Safety goggles
* Lab apron
* Dissecting microscope (optional)

Procedure

1. Observe the reaction of muscle fibers to five different solutions:

* 0.25% ATP in distilled water
* 0.25% ATP in salt solution
* 0.10% ATP in salt solution
* 0.05% ATP in salt solution
* Salt solution, no ATP

1. Using your knowledge of ATP, predict which solution you think will cause the greatest percent contraction in the muscle fibers. Which do you think will cause little or no change in the muscle? Record your predictions in your lab book.
2. Create a data table in your lab notebook that displays the following headings: Type of solution, Length before Solution (mm), Length after Solution (mm) and % Contraction.
3. Label test tubes and microscope slides 1,2,3,4, and 5.
4. Have one partner continue with Step 6 to prepare the solutions and the other partner begin Steps 7-9 to prepare the muscle fibers.
5. Notice that stock vials of the ATP and salt solutions are available at the front table or on each lab bench. To avoid crowding, take a sample of each solution back to your lab station. Some mixing is required to prepare solutions in the required concentration. Mix the solutions you will need for the experiment in the labeled test tubes and take your five tubes back to you station.

* **Test tube 1 (0.25% ATP in distilled water)**- 5 drops from dropper bottled labeled *ATP only (Aqueous Solution)*
* **Test tube 2 (0.25% ATP in salt solution)**- 5 drops from dropper bottled labeled *ATP plus salt solution*
* **Test tube 3 (0.1% ATP in salt solution)**- 2 drops from dropper bottle labeled *ATP only* and 3 drops from dropper bottle labeled *Salt solution*
* **Test tube 4 (0.05% ATP in salt solution)**- 1 drop from dropper bottle labeled *ATP only* and 4 drops from dropper bottle labeled *Salt solution*
* **Test tube 5 (salt solution, no ATP)**- 5 drops from dropper bottle labeled *Salt solution*

1. Note that the skeletal muscle provided is still wound as thick fibers. Isolate the muscle into thin single strands before you begin your experiment.
2. Place the microscope slide or Petri dish containing a segment of skeletal muscle tissue (in glycerol) on the stage of a microscope on low power. You can also complete the following steps using a dissecting microscope or a simple magnifying glass.
3. Looking through the microscope, use a teasing needle or straight pin to carefully pull apart the segment and release very thin strands. You may not be able to get single strands, but make sure that the pieces you remove are not more than four fibers thick and do not exceed 0.2mm in diameter.
4. Drip a **tiny** drop of glycerol from the Petri dish on microscope slide #1. The amount of glycerol that is transferred with the strands should only be enough to keep them moist. Transfer two to three of your thinnest strands to the slide, placing the fibers on the drop of glycerol.
5. Separate the strands, lay them out straight and do NOT cover them with a coverslip.
6. Carefully place the slide under a microscope and measure the length of the strands with a millimeter ruler held next to the slide. Record the measurements in your data table.
7. Flood the strands with several drops of solution 1. Observe the reaction of the fibers.
8. After thirty seconds, remeasure the strands.
9. Calculate the percent contraction of the strand using the following equation:

Original length – Final length

Original length **x** 100 = % contraction

1. Record the calculation and the final measurement in your laboratory journal.
2. If you have multiple fibers on your slide, calculate the percent difference for each strand and then take an average of these values.
3. Note if the fibers have changed in width.
4. Carefully place slide #1 off to the side.
5. Repeat the experiment using the remaining slides and new myofibers for each of the remaining four solutions.
6. Record all data and calculations in your laboratory journal.
7. When you have completed all five trials, carefully transfer one contracted fiber (from any solution) to a clean microscope slide. Mount the strand to a slide with a coverslip.
8. Examine the strand under both low and high magnification. In your laboratory journal, draw what you see. Note the striations in the myofibers.
9. Repeat steps 20 and 21 using a fiber that did not contract. Draw what you see and record your observations in your lab book. Compare the distance between the bands in the relaxed muscle to the distance you observed in the contracted muscle.
10. Answer the conclusion questions.

Conclusion

1. When you see the glycerinated muscle fibers shorten, what do you think is actually happening in the muscle cells? Think about what you just saw under the microscope.
2. Did your muscle samples eventually relax and return to the length they were before contraction? Thinking back to the muscles rules and how your know muscles work, why does this make sense?
3. Which solution produced the greatest percent contraction of the muscles? What does this tell you about the requirements for contraction?
4. Think about the way in which organ systems work together to kick your leg or swing your arm. Besides the skeletal and the muscular system, what other systems do you think are involved in moving the human machine?