

Skeletal Muscle Physiology

OBJECTIVES

1. To define motor unit, twitch, latent period, contraction phase, relaxation phase, threshold, summation, tetanus, fatigue, isometric contraction, and isotonic contraction
2. To understand how nerve impulses trigger muscle movement
3. To describe the phases of a muscle twitch
4. To identify threshold and maximal stimuli
5. To understand the effect of increases in stimulus intensity on a muscle
6. To understand the effect of increases in stimulus frequency on a muscle
7. To demonstrate muscle fatigue
8. To explain the differences between isometric and isotonic muscle contractions

Humans make voluntary decisions to talk, walk, stand up, or sit down. The muscles that make these actions possible are skeletal muscles. Skeletal muscle is generally muscle that is attached to the skeleton of the body, although there are some exceptions—for example, the *obicularis oris*, a muscle around the mouth, never attaches to any skeletal element. Skeletal muscles characteristically span two joints and attach to the skeleton via tendons connecting to the periosteum of the bone.

The Motor Unit and Muscle Contraction

A **motor unit** consists of a motor neuron and all of the muscle fibers it innervates. Motor neurons direct muscles when and when not to contract. A motor neuron and a muscle cell intersect at what is called the *neuromuscular junction*. Specifically, the neuromuscular junction is where the axon terminal of the neuron meets a specialized region of the muscle cell's plasma membrane. This specialized region is called the *motor end-plate*. An action potential (depolarization) in a motor neuron triggers the release of acetylcholine, which diffuses into the muscle plasma membrane (also known as the *sarcolemma*). The acetylcholine binds to receptors on the muscle cell, initiating a change in ion permeability that results in depolarization of the muscle plasma membrane, called an *end-plate potential*. The end-plate potential, in turn, triggers a series of events that results in the contraction of a muscle cell. This entire process is called *excitation-contraction coupling*.

We will be simulating this process in the following activities, only instead of using acetylcholine to trigger action potentials, we will be using electrical shocks. The shocks will be administered by an electrical stimulator that can be set for the precise voltage, frequency, and duration of shock desired. When applied to a muscle that has been surgically removed from an animal, a single electrical stimulus will result in a muscle **twitch**—the mechanical response to a single action potential. A twitch has three phases: the **latent period**, which is the period of time that elapses between the generation of an action potential in a muscle cell and the start of muscle contraction; the **contraction phase**, which starts at the

end of the latent period and ends when muscle **tension** peaks; and the **relaxation phase**, which is the period of time from peak tension until the end of the muscle contraction (Figure 2.1b).

Single Stimulus

Follow the instructions in the Getting Started section at the front of this manual. From the Main Menu, select **Skeletal Muscle Physiology**. You will see the opening screen for the **Single Stimulus** activity, shown in Figure 2.1.

On the left side of the screen is a muscle suspended in a metal holder that is designed to measure any force produced by the muscle. To the right of the metal holder are three pieces of equipment. The top piece of equipment is an oscilloscope screen. When you apply an electrical stimulus to the muscle, the muscle's reaction will be graphically displayed on this screen. Elapsed time, in milliseconds, is measured along the X axis of this screen, while any force generated by the muscle is measured along the Y axis. In the lower right-hand corner of the oscilloscope is a **Clear Tracings** button; clicking the button will remove any tracings from the screen.

Beneath the oscilloscope screen is the electrical stimulator you will use to stimulate the muscle. Note the electrode from the stimulator that rests on the muscle. Next to the **Voltage** display on the left side of the stimulator are (+) and (-) buttons, which you may click to set the desired voltage. When you click on the **Stimulate** button, you will electrically stimulate the muscle at the set voltage. In the middle of the stimulator are display fields for *active force*, *passive force*, and *total force*. Muscle contraction produces *active force*. *Passive force* is generated from the muscle being stretched. The sum of active force and passive force is the *total force*. Also notice a **Measure** button on the stimulator. Clicking this button after administering a stimulus will cause a yellow vertical line to appear. Clicking the (+) or (-) buttons under **Time (msec)** will then allow you to move the yellow line along the X axis and view the active, passive, or total force generated at a specific point in time.

Beneath the stimulator is the data collection box. Clicking on **Record Data** after an experimental run will allow you to record the data in this box. To delete a line of data, click on the data to highlight it and then click **Delete Line**. You may also delete the entire table by clicking **Clear Table**.

ACTIVITY 1

Identifying the Latent Period

Recall that the **latent period** is the period of time that elapses between the generation of an action potential in a muscle cell and the start of muscle contraction.

1. Set the **Voltage** to 6.0 volts by clicking the (+) button on the stimulator until the voltage display reads 6.0.
2. Click **Stimulate** and observe the tracing that results. Notice that the trace starts at the left side of the screen and stays flat for a short period of time. Remember that the X axis displays elapsed time.
3. Click on the **Measure** button on the stimulator. Note that a thin, vertical yellow line appears at the far left side of the oscilloscope screen.

4. Click on the (>) button underneath **Time (msec)**. You will see the vertical yellow line start to move across the screen. Watch what happens in the **Time (msec)** display as the line moves across the screen. Keep clicking the (>) button until the yellow line reaches the point in the tracing where the graph stops being a flat line and begins to rise (this is the point at which muscle contraction starts.) If the yellow line moves past the desired point, you can use the (<) button to move it backwards.

How long is the latent period? _____ msec

Note: If you wish to print your graph, click **Tools** on the menu bar and then click **Print Graph**.

5. Increase or decrease the stimulus voltage and repeat the experiment. (Remember that you can clear the tracings on the screen at any time by clicking **Clear Tracings**.) Record your data here:

Stimulus voltage: _____ V Latent period: _____ msec

Stimulus voltage: _____ V Latent period: _____ msec

Stimulus voltage: _____ V Latent period: _____ msec

Does the latent period change with different stimulus voltages? _____

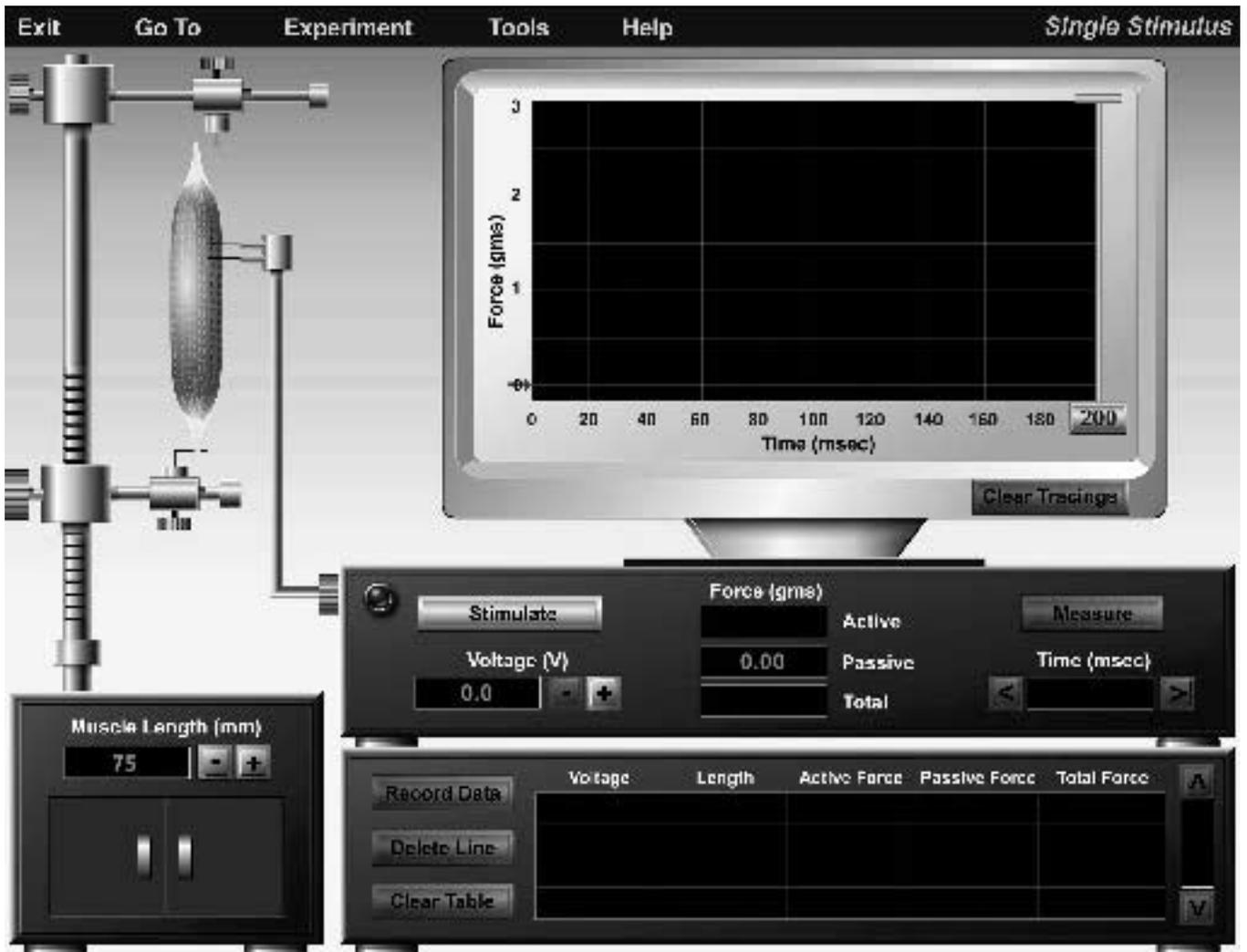
After completing this experiment, click **Clear Tracings** to clear the oscilloscope screen of all tracings. ■

ACTIVITY 2

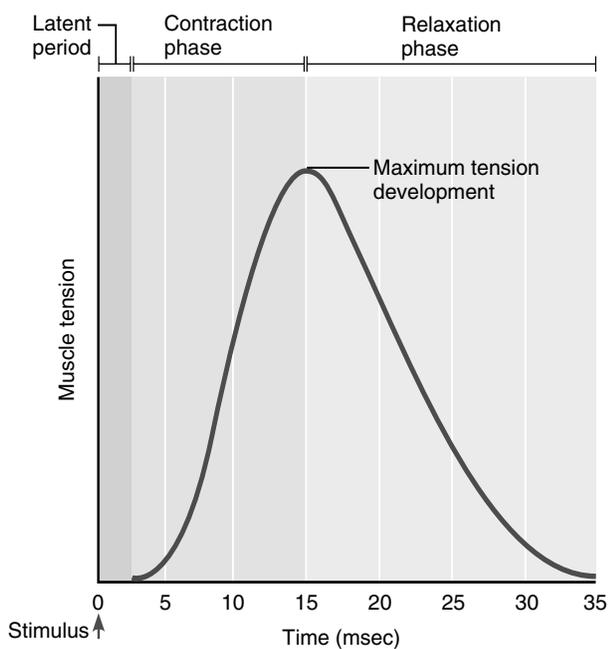
Identifying the Threshold Voltage

By definition, the **threshold** is the minimal stimulus needed to cause a depolarization of the muscle plasma membrane (sarcolemma.) The threshold is the point at which sodium ions start to move into the cell (instead of out of the cell) to bring about the membrane depolarization.

1. Set the **Voltage** on the stimulator to 0.0 volts.
 2. Click **Stimulate**. What do you see in the Active Force display?
-
3. Click **Record Data**.
 4. Increase the voltage to 0.1 volt, then click **Stimulate**. Observe the oscilloscope screen and the Active Force display (on the right side of the stimulator).
 5. Click **Record Data**.
 6. Repeat steps 4 and 5 until you see a number that is greater than 0.00 appear in the Active Force display.



(a)



(b)

FIGURE 2.1 Single stimulus and muscle twitch. (a) Opening screen of the Single Stimulus experiment. (b) The muscle twitch: myogram of an isometric twitch contraction.

7. Print out the graph(s) that you see on the oscilloscope screen by clicking **Tools** at the top of the screen and then selecting **Print Graph**.

What is the threshold voltage? _____ V

How does the graph generated at the threshold voltage differ from the graphs generated at voltages below the threshold?

ACTIVITY 3

Effect of Increases in Stimulus Intensity

In this activity we will examine how additional increases in stimulus intensity (such as additional increases in voltage) affect muscle response.

1. Set the voltage to 0.5 volts and click **Stimulate**. Then click **Record Data**.
2. Continue increasing the voltage by 0.5 volts and clicking **Stimulate** until you have reached 10.0 volts. Observe the **Active Force** display and click **Record Data** after each stimulation. Leave all of your tracings on the screen so that you can compare them to one another. If you wish, you may click **Tools** and then **Print Graph** to print your tracings.
3. Observe your tracings. How did the increases in voltage affect the peaks in the tracings?

How did the increases in voltage affect the amount of active force generated by the muscle?

What is the voltage beyond which there were no further increases in active force? Maximal voltage: _____ V

Why is there a maximal voltage? What has happened to the muscle at this voltage? Keep in mind that the muscle we are working with consists of many individual muscle fibers.

An individual muscle fiber follows the *all-or-none* principle—it will either contract 100% or not at all. Does the muscle we are working with exhibit the *all-or-none* principle? Why or why not?

4. If you wish, you may view a summary of your data on a plotted data grid by clicking **Tools** and then **Plot Data**.
5. Click **Tools** → **Print Data** to print your data. ■

Multiple Stimulus

Click on **Experiment** at the top of the screen and then select **Multiple Stimulus**. You will see a slightly different screen appear (Figure 2.2.) The main change is that a **Multiple Stimulus** button has now been added to the electrical stimulator. This button allows you to start and stop the stimulator as you wish. When you click **Multiple Stimulus**, you'll notice that the button's label changes to **Stop Stimulus**. Clicking on **Stop Stimulus** turns off the stimulator.

ACTIVITY 4

Treppe

Treppe is the progressive increase in force generated when a muscle is stimulated at a sufficiently high frequency. At such a frequency, muscle twitches follow one another closely, with each successive twitch peaking slightly higher than the one before. This step-like increase in force is why *treppe* is also known as the *staircase phenomenon*.

1. Set the voltage to the maximal voltage you established in Activity 3.
2. Click on the **200** button on the far right edge of the oscilloscope screen and slowly drag it as far to the left of the screen as it will go. This will allow you to see a longer span of time displayed on the screen.
3. Click **Single Stimulus** once. You will watch the trace rise and fall. As soon as it falls, click **Single Stimulus** again. Watch the trace rise and fall; when it falls, click **Single Stimulus** a third time.

What do you observe?

4. Click on the **200** button and drag it back to the far right edge of the oscilloscope screen.
5. To print your graphs, click on **Tools** on the menu bar, then click **Print Graph**. ■

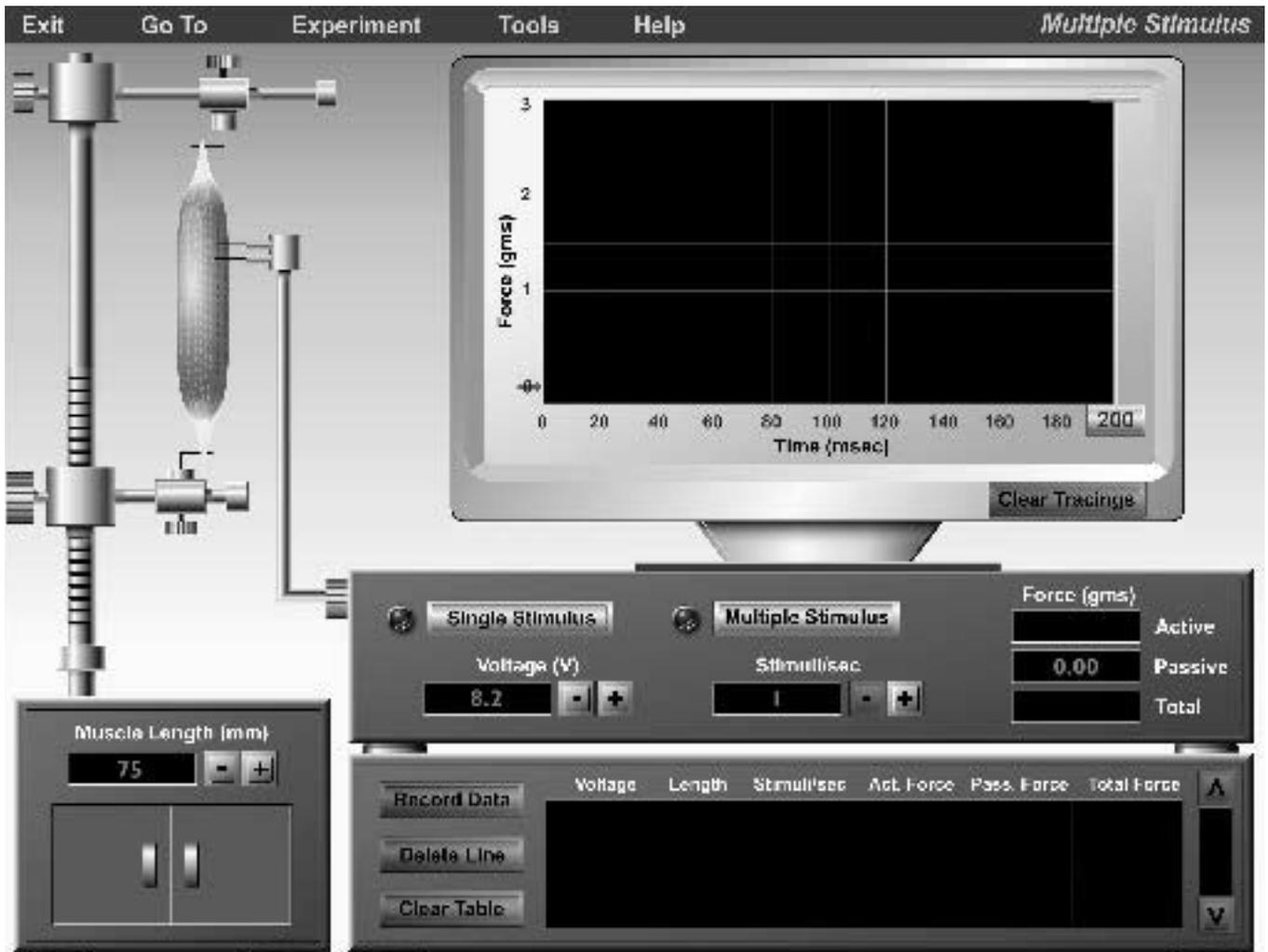


FIGURE 2.2 Opening screen of the Multiple Stimulus experiment.

ACTIVITY 5

Summation

When a muscle is stimulated repeatedly, such that the stimuli arrive one after another within a short period of time, twitches can overlap with each other and result in a stronger muscle contraction than a stand-alone twitch. This phenomenon is known as **summation**. Summation occurs when muscle fibers that have already been stimulated once are stimulated again, before the fibers have relaxed.

1. Set the **Voltage** to the maximal voltage you established in Activity 3.
2. Click on the **Single Stimulus** button and observe the oscilloscope screen.

What is the active force of the contraction? _____ gms

3. Click on the **Single Stimulus** button once. Watch the trace rise and begin to fall. Before the trace falls completely, click **Single Stimulus** again. (You may want to simply click

Single Stimulus twice in quick succession in order to achieve this.)

What is the active force now? _____ gms.

4. Click on **Single Stimulus** and allow the graph to rise and fall before clicking **Single Stimulus** again.

Was there any change in the force generated by the muscle?

5. Click on **Single Stimulus** and allow the graph to rise, but not fall, before clicking **Single Stimulus** again.

Was there any change in the force generated by the muscle?

Why has the force changed?

6. Decrease the voltage on the stimulator and repeat this activity.

Do you see the same pattern of changes in the force generated? _____

7. Next, stimulate the muscle as fast as you can (that is, click on **Single Stimulus** several times in quick succession).

Does the force generated change with each additional stimulus? If so, how?

ACTIVITY 6

Tetanus

In the previous activity we observed that if stimuli are applied to a muscle frequently in quick succession, the muscle generated more force with each successive stimulus. However, if stimuli continue to be applied frequently to a muscle over a prolonged period of time, the muscle force will eventually reach a plateau—a state known as **tetanus**. If stimuli are applied with even greater frequency, the twitches will begin to fuse so that the peaks and valleys of each twitch become indistinguishable from one another—this state is known as *complete (fused) tetanus*. The stimulus frequency at which no further increases in force are generated by the muscle is the *maximal tetanic tension*.

1. Click **Clear Tracings** to erase any existing tracings from the oscilloscope screen.
2. Underneath the **Multiple Stimulus** button, set the **Stimuli/sec** display, located beneath the **Multiple Stimulus** button, to 50 by clicking on the (+) button.
3. Set the voltage to the maximal voltage you established in Activity 3.
4. Click **Multiple Stimulus** and watch the trace as it moves across the screen. You will notice that the **Multiple Stimulus** button changes to a **Stop Stimulus** button as soon as it is clicked. After the trace has moved across the full screen and begins moving across the screen a second time, click the **Stop Stimulus** button.

What begins to happen at around 80 msec?

What is this condition called?

5. Leave the trace on the screen. Increase the **Stimuli/sec** setting to 130 by clicking the (+) button. Then click **Multiple Stimulus** and observe the trace. After the trace has moved across the full screen and begins moving across the screen a second time, click **Stop Stimulus**.

How does the trace at 130 stimuli/sec compare with the trace at 50 stimuli/sec?

What is this condition called?

6. Click **Clear Tracings** to clear the oscilloscope screen.
7. Increase the **Stimuli/sec** setting to 145 by clicking the (+) button. Then click **Multiple Stimulus** and observe the trace. Click **Stop Stimulus** after the trace has swept one full screen. Then click **Record Data**.
8. Repeat step 7, increasing the **Stimuli/sec** setting by 1 each time until you reach 150 stimuli/sec. (That is, set the **Stimuli/sec** setting to 146, then 147, 148, etc.) Be sure to click **Record Data** after each run.
9. Examine your data. At what stimulus frequency is there no further increase in force?

What is this stimulus frequency called?

10. For another view of your data, click **Tools** and then click **Plot Data**.
11. Click **Clear Tracings** to clear the oscilloscope screen. If you wish to print your data, click **Tools** and then **Print Data**.

ACTIVITY 7

Fatigue

Fatigue is a decline in a muscle's ability to maintain a constant force of contraction after prolonged, repetitive stimulation. The causes of fatigue are still being investigated, though in the case of high-intensity exercise, lactic acid buildup in muscles is thought to be a factor. In low-intensity exercise, fatigue may be due to a depletion of energy reserves.

1. Design an experiment that demonstrates fatigue on the oscilloscope screen. Hint: Set **Stimuli/sec** to above 100.

In fatigue, what happens to force production over time?

2. Print out your results by clicking on **Tools**, then **Print Graph**.
3. Click **Tools** → **Print Data** to print your recorded data.

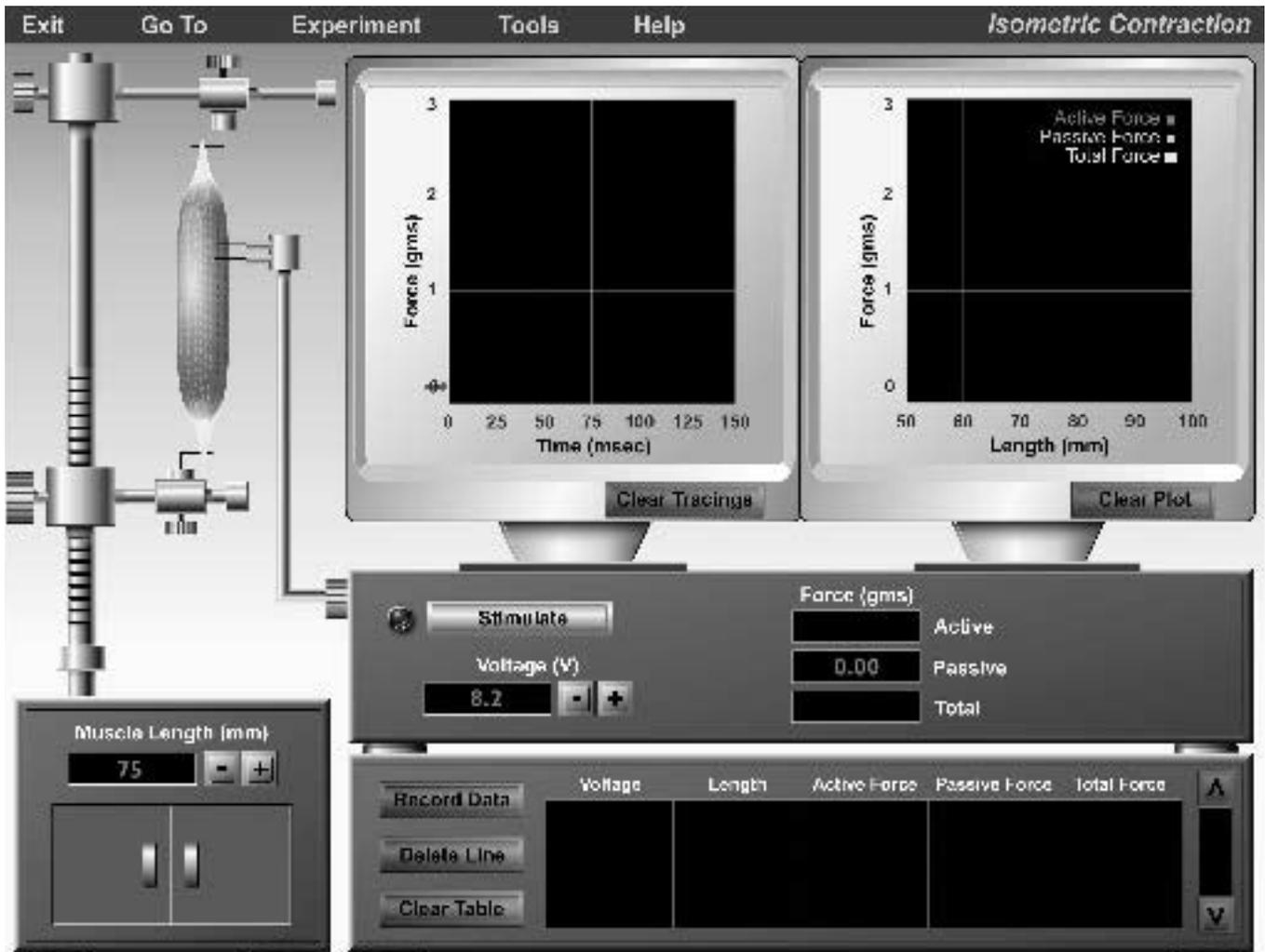


FIGURE 2.3 Opening screen of the Isometric Contraction experiment.

Isometric and Isotonic Contractions

Muscle contractions can be either **isometric** or **isotonic**. When a muscle attempts to move a load that is greater than the force generated by the muscle, the muscle contracts *isometrically*. In this type of contraction, the muscle stays at a fixed length (isometric means “same length”). An example of isometric muscle contraction is when you stand in a doorway and push on the doorframe. The load that you are attempting to move (the doorframe) is greater than the force generated by your muscle, and so your muscle does not shorten.

When a muscle attempts to move a load that is equal to or less than the force generated by the muscle, the muscle contracts *isotonically*. In this type of contraction, the muscle shortens during a period of time in which the force generated by the muscle remains constant (isotonic means “same tension”). An example of isotonic contraction is when you lift a book from a tabletop. The load that you are lifting (the book) is equal to or less than the force generated by your muscle. Your muscle shortens when it contracts, allowing you to lift the book.

We will first examine isometric contraction. Click on **Experiment** at the top of the screen and then select **Isomet-**

ric Contraction. You will see the screen shown in Figure 2.3. Notice that there are now two oscilloscope screens. The screen on the left is basically identical to the ones you worked with in the previous activities. The screen on the right is new. The Y axis is still “Force,” but the X axis is now muscle length. Let’s conduct a practice run to familiarize ourselves with the equipment.

1. Leave the **Voltage** set at 8.2 and click **Stimulate**. You will see indicators of three forces in the right-hand screen. You will see the Passive Force indicator near the bottom of the screen (in green), and the Active Force (in purple) and Total Force (in yellow) indicators together higher up on the screen. Note that the Active Force indicator is seen inside of the Total Force indicator.
2. Click the (+) and (–) buttons beneath **Muscle Length** on the left side of the screen and notice how the muscle may be stretched or shortened.
3. You are now ready to begin the experiment. Click the **Clear Tracings** and **Clear Plot** buttons under each oscilloscope screen. ■

ACTIVITY 8

Isometric Contractions

1. Leave the **Voltage** set at 8.2.
2. On the lower left side of the screen, click the (–) button underneath **Muscle Length** and reduce the length to 50 mm.
3. Click **Stimulate** and observe the results on both oscilloscope screens.
4. Click **Record Data** at the bottom of the screen.
5. Repeat steps 1–5, increasing the muscle length by 10 mm each time (i.e., 60 mm, then 70 mm, then 80 mm, etc.) until you reach 100 mm. Remember to click **Record Data** after each run.
6. Click **Tools** at the top of the screen, then click **Plot Data**. You will see a screen pop up depicting a plotted graph. Be sure that *Length* is depicted on the X axis and *Active Force* is depicted on the Y axis. You may wish to click **Print Plot** at the top left corner of the window to print the graph.

Looking at this graph, what muscle lengths generated the most active force? (provide a range)

____-to-____ mm

At what muscle length does passive force begin to play less of a role in the total force generated by the muscle?

_____ mm

7. Move the blue square bar for the Y axis to *Passive Force*. You may wish to click **Print Plot** at the top left corner of the window to print the graph.

Looking at this graph, at what muscle length does Passive Force begin to play a role in the total force generated by the muscle?

_____ mm

8. Move the blue square bar for the Y axis to *Total Force*. You may wish to click on **Print Plot** at the top left corner of the window to print the graph.

9. Click **Tools** → **Print Data** to print your data.

The graph shows a dip at muscle length = 90 mm. Why is this?

What is the key variable in an isometric contraction?

ACTIVITY 9

Isotonic Contractions

Recall that isotonic contraction occurs when a muscle generates a force equal to or greater than the load it is opposing. In this type of contraction, there is a latent period, followed by a rise in generation of force, followed by a period of time during which the force produced by the muscle remains constant (recall that isotonic means “same tension”). During this plateau period, the muscle shortens and is able to move the load. The muscle is not able to shorten prior to the plateau because enough force has not yet been generated to move the load. When the force generated becomes equal to the load, the muscle shortens. The force generated will be constant so long as the load is moving. Eventually, the muscle will relax and the load will begin to fall. An isotonic twitch is not an all-or-nothing event. If the load is increased, the muscle must generate more force to move it. The latent period will also lengthen, as it will take more time for the necessary force to be generated by the muscle. The speed of the contraction depends on the load the muscle is opposing. Maximal speed is attained with minimal load. Conversely, the heavier the load, the slower the muscle contraction.

Click on **Experiment** at the top of the screen and then select **Isotonic Contraction**. The screen that appears (Figure 2.4) is similar to the *Single Stimulus* screen you worked with in Activities 1–3. Note that fields for Muscle Length and Velocity have been added to the display below the oscilloscope screen, and that the muscle on the left side of the screen is now dangling freely at its lower end. The weight cabinet below the muscle is open; inside are four weights, each of which may be attached to the muscle. Above the weight cabinet is a moveable platform, which you may move by clicking the (+) or (–) buttons under **Platform Height**.

In this experiment, you will be attaching weights to the end of the muscle in order to observe isotonic contraction.

1. The **Voltage** should already be set at 8.2, and the **Platform Height** at 75 mm. If not, adjust the settings accordingly.
2. Click on the **0.5g** weight in the weight cabinet and attach it to the dangling end of the muscle. The weight will pull down on the muscle and come to rest on the platform.
3. Click **Stimulate** and observe the trace. Note the rise in force, followed by a short plateau, followed by a relaxation phase. Note that the Active Force display is the same as the weight that was attached: 0.5 grams.

How much time does it take for the muscle to generate 0.5 grams of force? _____ msec

4. Click **Stimulate** again, watching the muscle and the screen at the same time as best you can. Then click **Record Data**.

At what point in the trace does the muscle shorten?

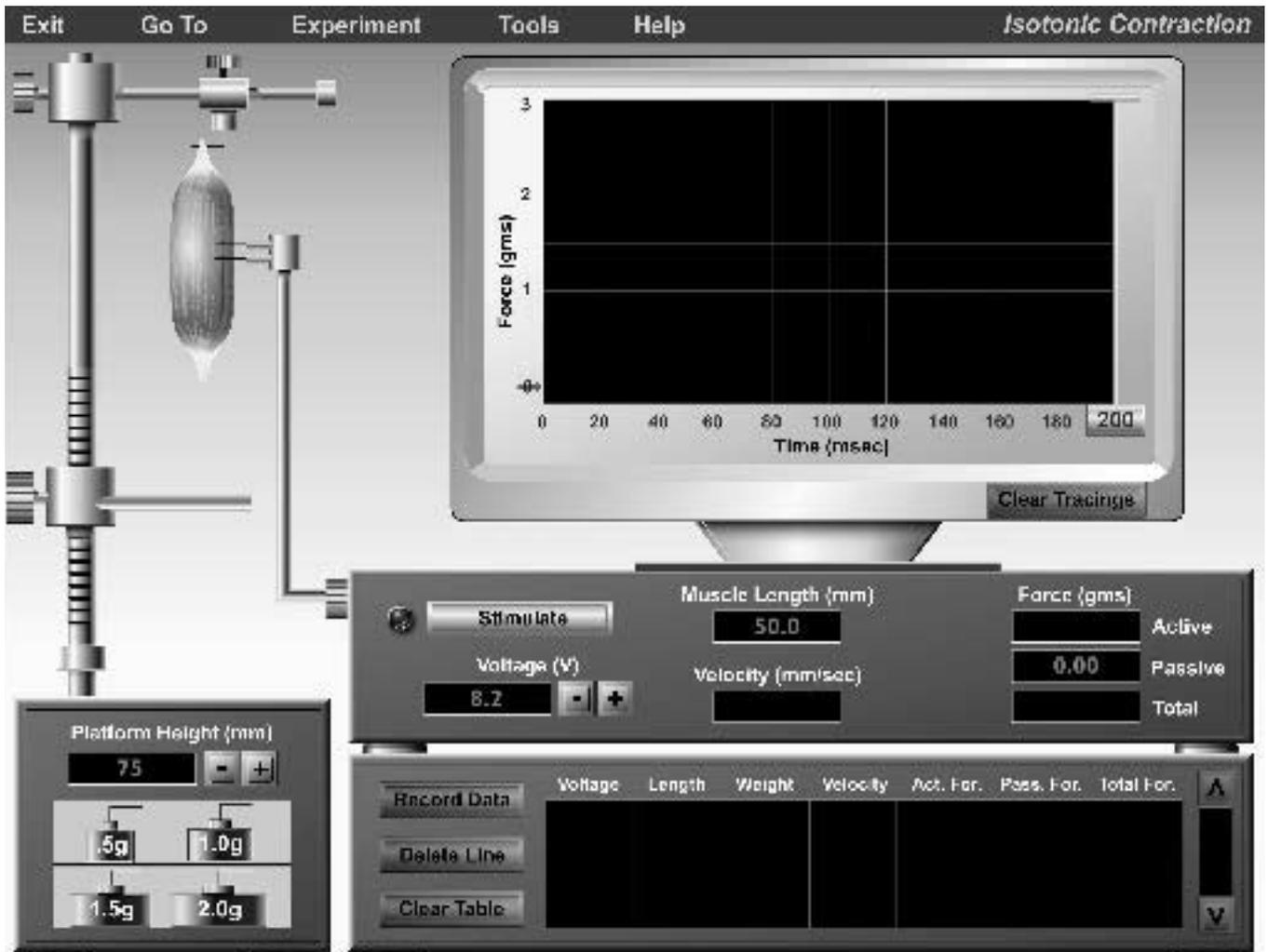


FIGURE 2.4 Opening screen of the Isotonic Contraction experiment.

You can observe from the trace that the muscle is rising in force before it reaches the plateau phase. Why doesn't the muscle shorten prior to the plateau phase?

5. Remove the .5g weight and attach the 1.0g weight. Leave your previous trace on the screen.

6. Click **Stimulate** and then **Record Data**.

Did it take any longer for the muscle to reach the force it needed to move the weight?

How does this trace differ from the trace you generated with the .5g weight attached?

7. Leaving the two previous traces on the screen, repeat the experiment with the two remaining weights. Click **Record**

Data after each result. If you wish to print your graphs, click **Tools** and then select **Print Graph**.

8. When you have finished recording data for all four weights, click **Tools** from the top of the screen and click **Plot Data**.

9. Move the blue square bar for the Y axis to *Velocity* and the blue square bar for the X axis to *Weight*.

Examine the plot data and your numerical data. At what weight was the velocity of contraction the fastest?

_____ gms

What happened when you attached the 2.0g weight to the muscle and stimulated the muscle? How did this trace differ from the other traces? What kind of contraction did you observe?

10. Close the Plot Data window by clicking on the “X” at the top right corner of the window. If you still have a weight attached to the muscle, remove it. Also click **Clear Tracings** to clear the oscilloscope screen.

11. Place the **0.5g** weight on the muscle and adjust the height of the platform to 100 mm. Notice that the platform no longer supports the weight.

12. Click **Stimulate** and observe the muscle contraction tracing. What kind of a trace did you get?

What was the force of the contraction? _____ gms

13. Click **Record Data**. Then repeat steps 12–13 for each of the remaining weights (remember to click **Record Data** after obtaining the results for each weight). Click **Tools** and then **Print Graph** if you wish to print your tracings.

Describe your four tracings and explain what has happened in each of them.

14. Click **Clear Tracings**.

15. Place the **1.5g** weight on the muscle.

16. Adjust the platform height to 90 mm.

17. Click **Stimulate** and then **Record Data**.

18. Repeat steps 16–18 except lower the platform height by 10 mm each time until you reach 60 mm (i.e., set the platform height at 80 mm, then 70 mm, then 60 mm).

19. Click **Tools**, then **Plot Data**.

20. Within the Plot Data window, move the blue square bar for the X axis to *Length* and the blue square bar for the Y axis to *Velocity*.

What muscle length(s) generated the fastest contraction velocity?

22. Close the Plot Data window by clicking the “X” in the upper right corner of the window.

23. Click **Tools** → **Print Data** to print your data. ■

Histology Review Supplement

For a review of skeletal muscle tissue, go to Exercise **H: Histology Atlas and Review** on the **PhysioEx™** website to print out the **Skeletal Muscle Tissue Review** worksheet.