Laboratory 4 - Human Muscle Physiology

Introduction

About 40% of the total body mass of a human is skeletal muscle. Skeletal muscle is intimately associated with the skeletal system and, combined, these muscles and bones are responsible for supporting and moving the body. While all skeletal muscle fibers have sarcomeres and the same banded appearance, different muscles can function in different ways. For example, some are relatively weak and fatigue resistant, while others are strong but fatigue quickly. These features may be explained in terms of biochemical properties.

The muscle fibers found in most mammalian skeletal muscles are either fast- or slow- twitch types. Each type contains a different type of myosin, with different rates of ATPase activity and cross-bridge binding. Within the group of fast-twitch fibers, there are fibers that use both glycolysis and oxidative phosphorylation. There are also fast-twitch fibers that just use glycolysis; this group tends to need less oxygen and is much stronger than the fibers that also use phosphorylation. However, these stronger glycolytic fibers breakdown glucose very inefficiently; a burst of contractile activity uses most of the available glucose, causes lactic acid to accumulate, and leads to fatigue more quickly.

Even though a motor neuron synapses with many muscle fibers, all the muscle fibers connected to that motor neuron are the same type. So, the stimulation of one particular motor neuron will cause contractions of only one type of muscle fiber. Motor neurons that supply the weak, slow, oxidative fibers have the lowest threshold. Those that innervate fast, intermediate-strength oxidative fibers have higher thresholds. And those that supply the fast, strong, glycolytic only fibers have the highest threshold. By increasing the amount of activity coming from the brain more motor neurons are progressively activated, and more of the stronger muscle fibers are brought into the response. All of this occurs through a single synapse on each fiber.

When a muscle tries to lift any weight, the muscle first shortens to put tension on the tendons which hold the muscle to the bones. Development of this tension before movement occurs takes time, known as the latency period, which is directly proportional to the weight attached to the muscle. Once the tension exceeds the weight of the object, any further muscle contraction produces a shortening of the muscle and a movement of the weight. The time that a muscle is in its active state (contracting) is finite; so, muscles have less time to shorten when they move heavier weights.

In this experiment, students will use a specialized displacement transducer, the striated muscle transducer, to demonstrate the effect of increasing the stimulus strength on the strength of contraction of a muscle, the effect of increasing weight on twitch amplitude and work of a preloaded muscle, and the effect of increasing the frequency of stimulation on the contraction strength and muscle fatigue.

This experiment involves the application of stimulus pulses to the skin on the back of the hand to produce a twitch in the muscles that move the subject's little finger. The stimulus pulses used in this experiment are generated by a high voltage stimulus isolator. While the sensation of evoked muscle contractions may initially feel peculiar, electrical stimulation of the muscles in the hand is safe and painless. Standard safety precautions need to be observed. Persons with poor cardiac function, pacemakers, or any other condition that can be aggravated by electrical stimulation should not volunteer for this experiment.

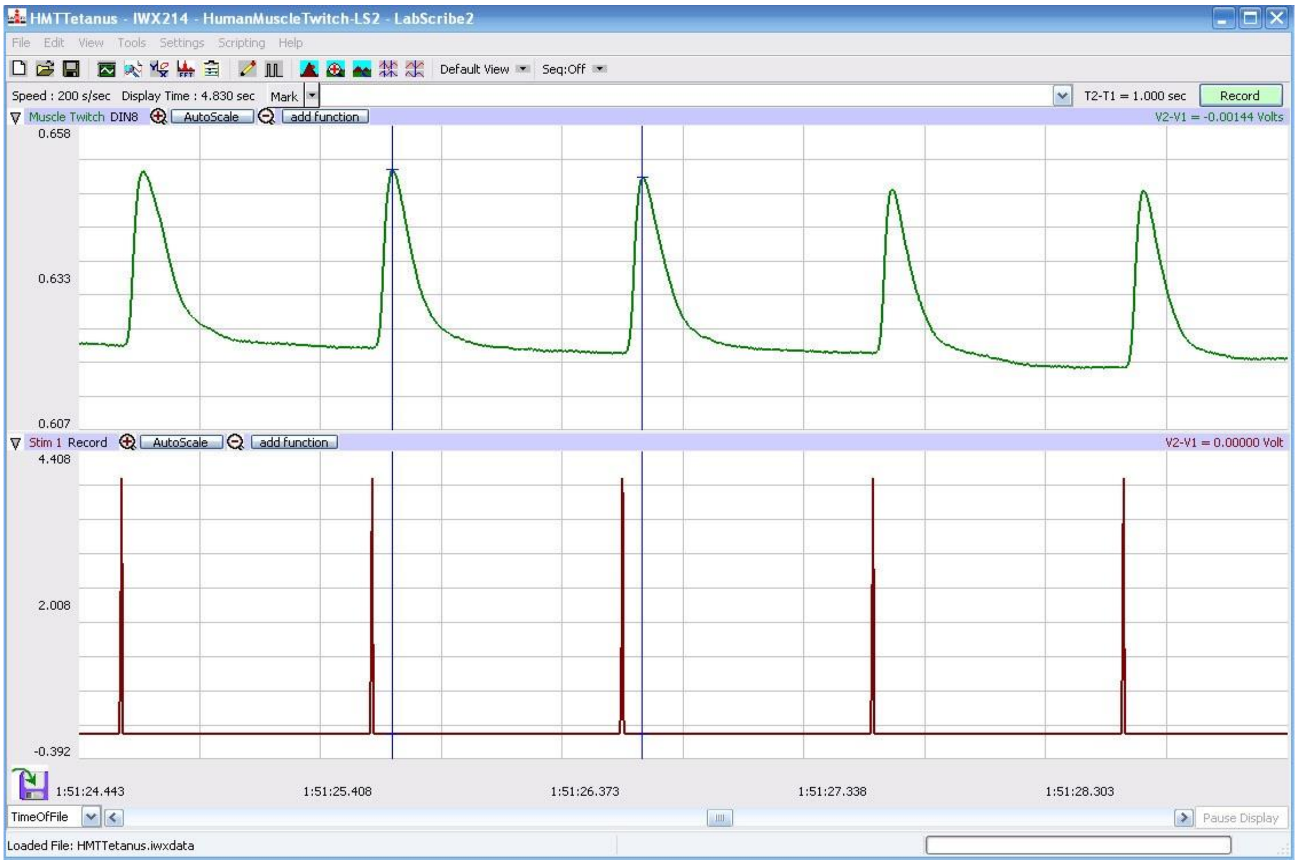
Change the location of the stimulating electrodes if the subject finds the stimulus to be painful. Changing subjects might be necessary, if the subject is experiencing discomfort.

Follow the instructors directions in lab to pull up an instruction sheet for how to set up your equipment.

Exercise 1: Properties of a Finger Twitch

Procedure

1. Instruct the subject to relax and place the hand that is used for the experiment on the bench, with its palm up.
2. Using the instructions given in class try to get a stimulus that records a tracing similar to the one below:



- Twitch Amplitude. To measure the amplitude of the twitch, place one cursor on the baseline at the point just before the twitch starts to develop, and the second cursor on the peak of the twitch. On the Finger Twitch channel, the value for V2-V1 is the amplitude of that twitch

- Contraction Time, which is the time it takes the amplitude of the twitch to rise to its peak. To measure the contraction time of the twitch, keep the cursors in the same positions used to measure the twitch amplitude. On the Finger Twitch channel, the value for T2-T1 is the contraction time of that twitch.

Relaxation Time, which is the time it takes the amplitude of the twitch to return to baseline. To measure the relaxation time of the twitch, place one cursor on the peak of the twitch, and the second cursor at the point where the amplitude returns to the baseline. On the Finger Twitch channel, the value for T2-T1 is the relaxation time of that twitch.

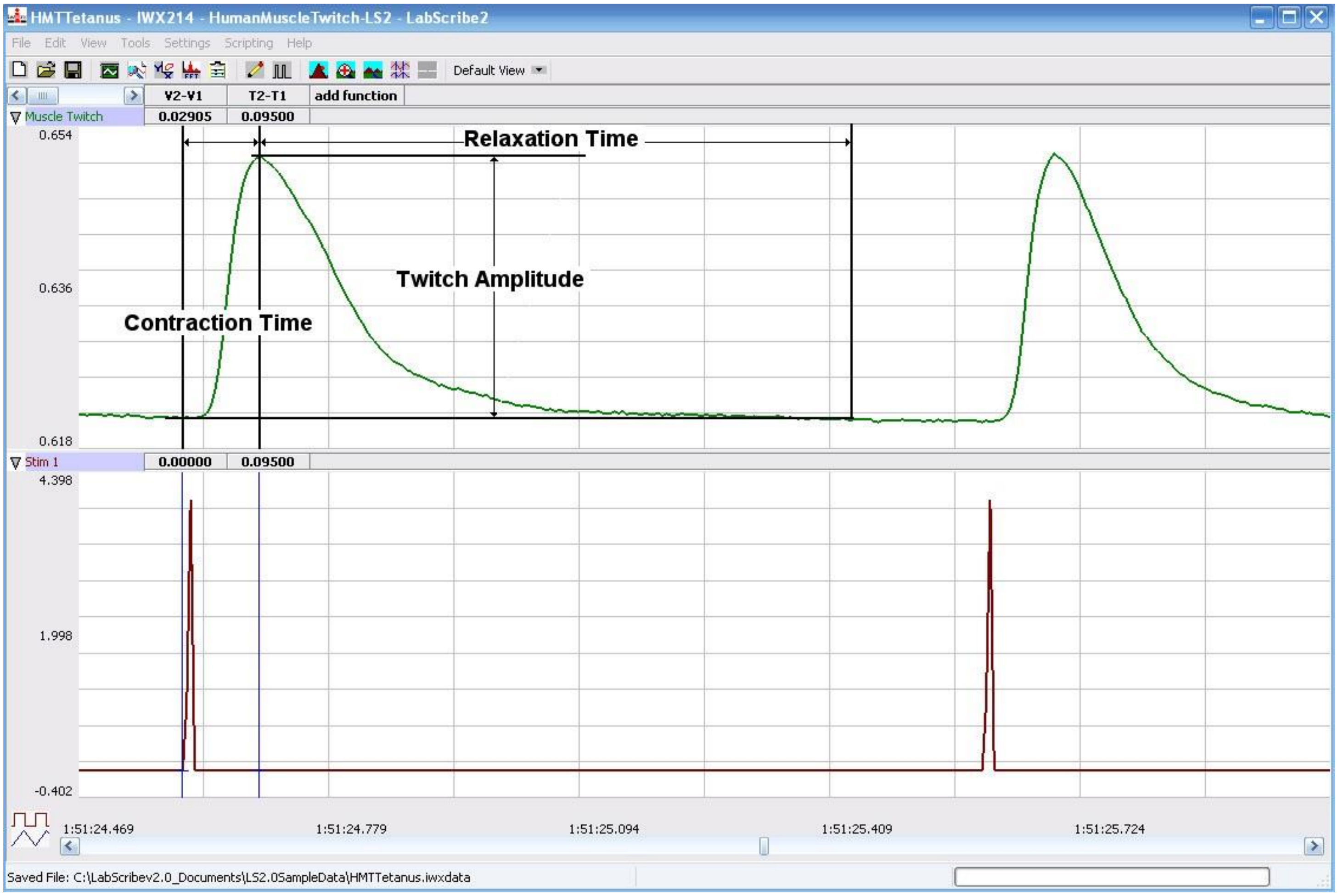


Figure HM-4-L4: Finger twitches stimulated by maximal current at a frequency of 1 Hz.

Question

1. It is not shown in the picture above, but there is a small amount of time between the beginning of the stimulus and the beginning of the contraction. This is called the "Lag Time". What is happening at the muscle fiber level to cause the lag time?

2. What is happening at the muscle fiber level during the contraction time? How does this correlate to what you know about a power-stroke?

3. What is happening at the muscle fiber level during the relaxation time?

4. In a single muscle twitch, why is the relaxation time longer than the contraction time?

5. If an individual has hypercalcemia, would you expect to see an increase in the rate of contraction? If so, where in the motor unit would we see this change manifested?

Exercise 2: The Effect of Weight on Finger Twitches

Aim: To determine the amount of work performed by the finger with different weights, or loads, attached to the finger.

Procedure

1. Attach a thread to the last joint of the finger being used in the experiment. The thread should be long enough to go across the ring, middle, and index fingers of the same hand, and also allow the weight to hang over the edge of the table. Attach a weight to the other end of the thread.
2. Have the subject move his or her hand to the edge of the table, so the palm is up and the thread with the weight is hanging along the side of the table.
3. Type the weight of the object, that is on the thread, in the Mark box that is to the right of the Mark button. Click the Record button on the LabScribe Main window. Press the Enter key on the keyboard to mark the recording. Continue to record for 15 seconds. Click the Stop button.

6. Add another 10 gram weight to end of the thread and repeat.

7. Continue to add weight to the thread in 10 gram increments. Repeat Step 5 each time you add weight to the thread until the amplitude of the finger twitch is undetectable.

Data Analysis

1. Scroll to the beginning of the recording for this exercise. Locate the section of data in which finger twitches appeared when the stimulus threshold was reached. Click AutoScale to maximize the size of the twitch on the window.
2. Use the Display Time icons to adjust the Display Time of the Main window to show a segment with four or five twitches on the Main window. The twitches can also be selected by:
   - Placing the cursors on either side of the selected twitches; and
   - Clicking the Zoom between Cursors button on the LabScribe toolbar to expand the segment of twitches to the width of the Main window.
3. Record the amplitudes (V2-V1) of three twitches recorded at each weight.

5. For each weight, determine the average amplitude of three finger twitches, the contraction, relaxation and lag time.

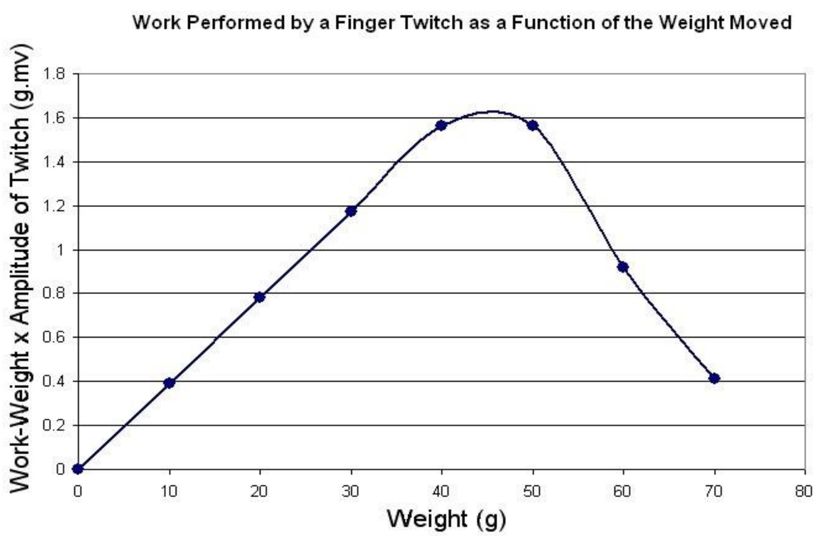
6. Use the average twitch amplitude at each weight to calculate the work performed on that weight. For this exercise, use the following equation:

Work = Weight (g) x Average Amplitude of Twitch (mv) with that Weight

Questions

1. How do the amplitudes of the finger twitches with different weights compare to each other?

2. Why did the amount of work decrease when heavier weights were used?



Exercise 3: Stimulus-Response

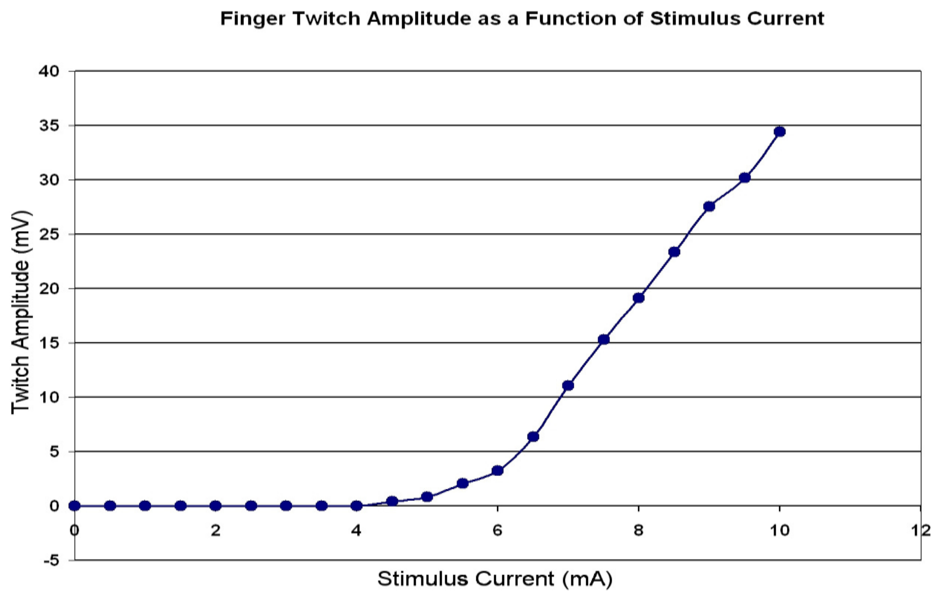
Aim: To examine the effect of stimulus current on the amplitude of finger movement.

Procedure

1. Remind the subject to relax and place the hand that is used for the experiment on the bench, with its palm up.
2. Check the stimulus parameters set in the stimulator control panel on the LabScribe Main window. The duration of the stimulus pulses should be 5 milliseconds (ms), their frequency should be 1 Hz, and the number of pulses should be 0 (zero). Setting the number of pulses to zero makes the stimulus isolator fire continuously. If the values of these parameters were changed, click the Apply button on the Stimulator control panel to effect the change
3. Make sure the Amplitude knob on the front of the SI-200 is set to zero; or change the values in the stimulus control panel for the IXTA. Type Zero in the Mark box that is to the right of the Mark button. Click the Record button on the LabScribe Main window. Press the Enter key on the keyboard to mark the recording.
4. While recording, type 0.5 mA in the Mark box that is to the right of the Mark button. Increase the current output by 0.5mA (rotating the Amplitude knob half a turn or changing amplitude in the stimulator control panel). Press the Enter key on the keyboard to mark the recording. Record at this stimulus current for ten to fifteen seconds.
5. Repeat Step 4, in increments of 0.5 mA, until the stimulus current reaches the maximal level. Click the Stop button.

Data Analysis

1. Scroll to the beginning of the data for this exercise. Locate the segment of the finger twitches that occurred at threshold stimulus current. Click AutoScale to maximize the size of the twitch on the window. For any stimulus currents that did not cause finger movement, report the amplitude of the finger twitch as zero (0).
2. Use the Display Time icons to adjust the Display Time of the Main window to show a segment with four or five twitches on the Main window. The twitches can also be selected by:
   - Placing the cursors on either side of the selected twitches; and
   - Clicking the Zoom between Cursors button on the LabScribe toolbar to expand the segment of twitches to the width of the Main window.
3. Click on the Analysis window icon in the LabScribe toolbar or select Analysis from the Windows menu to transfer the data displayed in the Main window to the Analysis window.
4. Use the same techniques used in Exercise 1 to measure and record the amplitudes (V2-V1) of three twitches recorded for each stimulus current that causes a response.
5. For each stimulus current, determine the average amplitude of three finger twitches.



Questions

1. **At which stimulus current did the finger first show a twitch? Why didn't a finger twitch occur at a lower stimulus current?**
2. **What is the mechanism for increased twitch amplitude with increased current?**
3. **Above a certain stimulus current, the amplitude of the finger twitch no longer increases. Why does this happen?**

Exercise 4: Summation and Tetanus

Aim: To monitor the contraction and relaxation of the finger twitch in relation to the stimulus frequency.

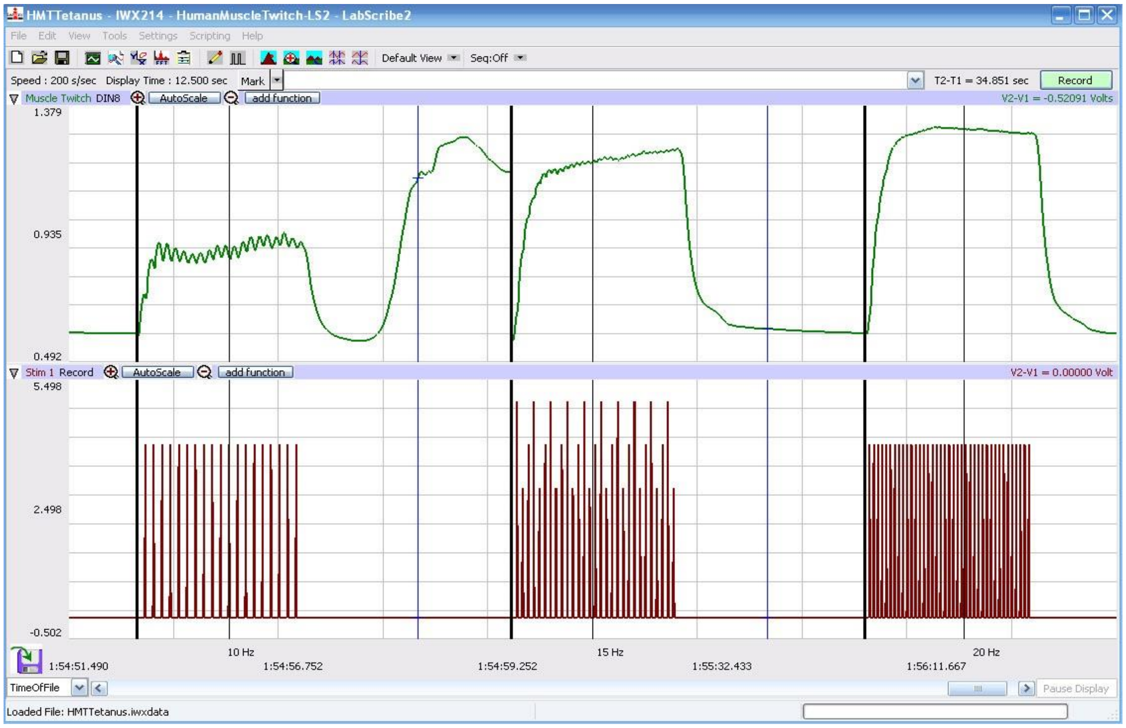
Procedure

1. Remind the subject to relax and place the hand that is used for the experiment on the bench, with its palm up.
2. The duration of the stimulus pulses should be 5 milliseconds (ms), their frequency should be 1 Hz, and the number of pulses should be set to 15.
3. During the course of this exercise, the stimulus frequency and the number of stimulus pulses will be changed to demonstrate summation and tetanus. After any stimulus parameter is changed, click the Apply button on the Stimulator control panel to effect the change.

5. Type 1 Hz in the Mark box to the right of the Mark button. Click the Record button on the LabScribe Main window. Press the Enter key on the keyboard. Click Stop when the stimulus pulse stop firing. There should be fifteen twitches on the recording.
6. Change the stimulus frequency to 2 Hz. Go to the Stimulator control panel on the LabScribe Main window, and change the value in the stimulus frequency box, labeled F(Hz), from 1 to 2. Click the up arrow in the box to increase the frequency. The frequency can also be changed by typing the new value into box. Click the Apply button to put any frequency change into effect.
7. Repeat Step 5 with the stimulus frequency set to 2. Type 2 in the Mark box that is to the right of the Mark button.
8. Repeat Step 5 for stimulus frequencies of 3, 4, 5, 10, 15, and 20 Hz. At higher frequencies, you will also need to increase the number of pulses sent to the subject's finger to see the effects of summation and tetanus. For example, it may take a few seconds to see the complete effect of tetanus at 20 Hz. So, the total number of pulses may need to be set to 40 or higher to see the complete effect. Always remember to click the Apply button to effect changes.
9. When performing the experiment at the highest frequency keep recording after complete tetanus is reached. Remove the cable from the stimulator of the to stop the stimulus isolator from firing. Continue to record as the tension in the muscles of the finger begins to relax.
10. When the muscles in the finger are relaxed, click Stop to halt the recording.
11. Select Save in the File menu.

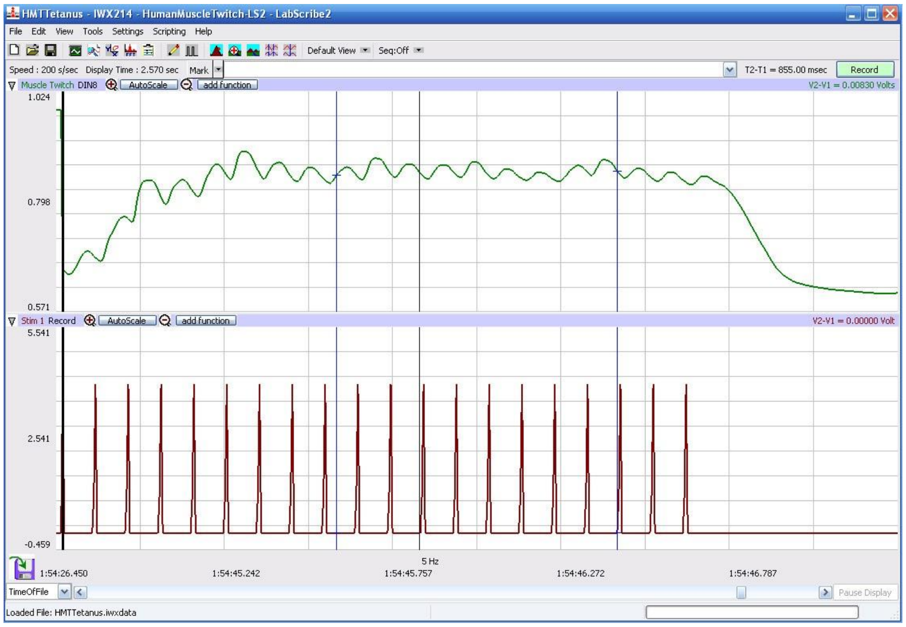
Data Analysis

1. Scroll to the beginning of the data for this exercise. Locate the segment of the finger twitches that occurred at a frequency of 1 Hz. Click AutoScale to maximize the size of the twitches on the window.
2. Use the Display Time icons to adjust the Display Time of the Main window to show the first four or five twitches at that frequency on the Main window. The twitches can also be selected by:
    - Placing the cursors on either side of the selected twitches; and
    - Clicking the Zoom between Cursors button on the LabScribe toolbar to expand the segment of twitches to the width of the Main window.



Amplitudes of finger twitches at 10, 15, and 20 Hz from left to right. Recordings from 10 and 15 Hz show incomplete tetanus. Complete tetanus occurs at 20 Hz.

3. Click on the Analysis window icon in the LabScribe toolbar or select Analysis from the Windows menu to transfer the data displayed in the Main window to the Analysis window.
4. Use the same techniques used in Exercise 1 to measure and record the amplitudes (V2-V1) of two adjacent twitches recorded at a stimulus frequency of 1 Hz.
5. Move to the segment of data recorded at the next stimulus frequency. If the twitches at this frequency are all about the same amplitude, measure and record the amplitudes of two adjacent twitches.
6. Repeat Step 5, until the recording of the finger twitches show summation. Summation occurs when the time between twitches is not long enough to allow the muscle to completely relax to its baseline level of tension. Another twitch or contraction, following in quick succession to the first, will add on the first twitch at the current level of tension for the first twitch. The third twitch will add on to the current tension of the second twitch, and so on. Since the twitches were maximal, this summation is a form known as mechanical summation, which can be seen in



Summation of finger twitches occurring at a stimulus frequency of 5Hz. Cursors are place on the first twitch and the tallest twitch.

7. At stimulus frequencies where summation occurred, measure and record the amplitude of the first twitch in the series and the amplitude of the tallest twitch in the series.
8. If a level of relatively constant tension occurs during contractions at high frequency, the phenomenon is called tetanus. If small relaxations are still detectable along the level of constant tension, a state of incomplete tetanus exists. If there are no detectable relaxations along the region of constant tension, a state of complete tetanus exists.

Questions

1. **In order to increase the force of a muscle fiber, neurons have the ability to summate. Explain what summation is, and the mechanism of both Temporal and Spatial Summation.**
2. **In relation to muscle fiber contractions, what is tetanus? Why does this occur?**
3. **What changes would you expect to see in the amplitude of a twitch undergoing summation or tetanus, compared to a single muscle twitch. Explain your reasoning.**
4. **Once the stimulation for a muscle finer is removed, the muscle fibers will relax. Did you notice that the relaxation time for a fiber that has experienced tetanus is much longer than for a fiber that has not? Explain the mechanism for this.**

5. During complete tetanus (and while a stimulus is still being applied), why do you see a gradual decline in the tension exerted by the muscle fibers?

6. A muscle can often maintain a moderate level of active tension for long periods of time, even though many of its fibers become fatigued. Why does this occur?