Pre-Laboratory Assignment: (Before you arrive to Lab)

Summary of Experiment:

In this physiology lab, you will be analyzing extended force production by muscles and observing muscle fatigue. Simultaneously, you will be analyzing electrical activity within the muscle using electromyography.

Your GTA will be the subject and will be gathering and recording data according to the procedures below using two devices: An **Electromyograph (EMG)** that measures electrical activity within muscle, and a **Hand Dynamometer** that measures force produced by "gripping" the device.

As a student, you will be using a software program, **Graphical Analysis Pro (GAPro)** installed upon your devices (computer or tablet, it is multi-platform friendly). Using the GAPro software you will import the data and you will also interpret the data.

While you will not actually be doing the procedures yourself, you are responsible to fully understand all procedures detailed below. You are also expected to understand all data analysis and properly interpret data as demonstrated within these documents and by your GTA during lab. Any of this information may be tested on the upcoming Practical Exam.

Install GAPro:

We will be using an application from Vernier Software and Technology, called Graphical Analysis (GA) and upgrading it to Graphical Analysis Pro (GAPro) to implement our Lab G Muscle Physiology Lab. You will first need to install GA on your computer or device.

We strongly encourage you to utilize a computer for this application, the use of a keyboard and mouse make the data easier to analyze. You will need to be able to share your screen during the lab within Zoom so it needs to be installed on the same device where you are attending the Zoom Meeting. You will also need to be able to take screenshots on this device.

Download and Install the free program - Graphical Analysis (GA): You can use this link, The download buttons are on the main page: https://www.vernier.com/product/graphical-analysis-4/

Or, download at the App Store ("Vernier Graphical Analysis" – NOT the GW Version).

After downloading and then installing GA, you will upgrade to GAPro.

1. Open up the GA Program (it may show up as "Vernier Graphical Analysis" in your applications list)

2. Upgrade to GAPro: Select the Shield Icon in the upper right and Submit the GAPro License Key.

Once GAPro is upgraded, you are done with this technology portion of the pre-lab. Be sure to close it and make sure you know how to re-open the application. When you arrive to your lab you will need to be able to open the application.
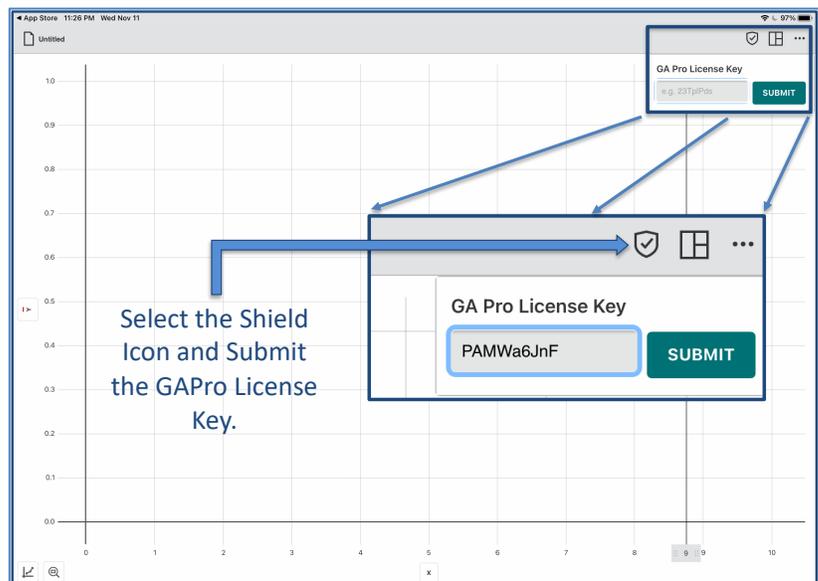
Attendance:

This is a lab where attendance is <u>absolutely required</u> during your Zoom lab meeting. You will not be given any credit for the lab if you do not demonstrate your attendance during lab, regardless of whether you turn in the assignment. The simplest way to do this is to just turn on your camera and microphone and interact with your GTA. You may be asked to share your screen within Zoom if you are not using a video camera.

Demonstrate Your Work With Screen-Shots:

You will need to be able to demonstrate your work by taking screen-shots on your device. Make sure that you know how to do this on the device that you will be using and that you can copy and paste those shots into a secondary document.

Pre-Lab Quiz:

At the beginning of the lab, after you complete the Lab F quiz and before you start this physiology lab, you will complete a BI301L G2 Pre-Lab Quiz on D2L. A reasonable understanding of the material above and below will make it simple to be fully successful on the Quiz.

Summary of Important Principles Necessary for this Lab:

Spinal Cord

Please read the information below carefully. Use your textbook or other resources to clarify those concepts that are not clear to you.

<u>Objectives</u>: In the Virtual Muscle Physiology Lab you will …

- Understand the principles of skeletal muscle force production and skeletal muscle fatigue.
- Learn how to use a Hand Dynamometer to measure the force produced by flexion of the entire hand ("grip force") as well as the force developed between the thumb and index finger ("pinch force").
- Learn to measure electrical activity produced in the muscles associated with grip and pinch force using electromyography (EMG).
- Understand how force production correlates with the EMG signal.
- Understand how force decreases during fatigue and associate this with the factors that affect the extent of fatigue.
- Analyze and interpret how the EMG signal changes during fatigue.
- Review the muscles that produce the forces in this experiment.

Figure 1. A slice of spinal cord depicting two somatic motor neurons innervating two separate motor units. (From Figure 11.6 Saladin)

<u>Background</u>: Voluntary skeletal muscle contraction is the result of communication between the brain and individual muscle fibers of the musculoskeletal system. The process begins within the motor cortex of the brain that stimulates somatic motor neurons originating within the spinal cord or brainstem. The axons of these motor neurons travel within peripheral nerves to innervate specific muscles. Within the muscle, each axon divides into multiple branches each of which innervates a specific muscle fiber at a neuromuscular junction on that fiber. The combination of the motor neuron, all of the muscle fibers that it innervates, and the neuromuscular junctions is known as a motor unit (Fig. 1).

Contraction of muscle begins with action potentials within the alpha motor neurons and these consist of a rapid depolarization and repolarization of the neuron membrane. This activates the neuromuscular junction and causes an action potential to also occur throughout the muscle fiber that subsequently causes contraction to occur.
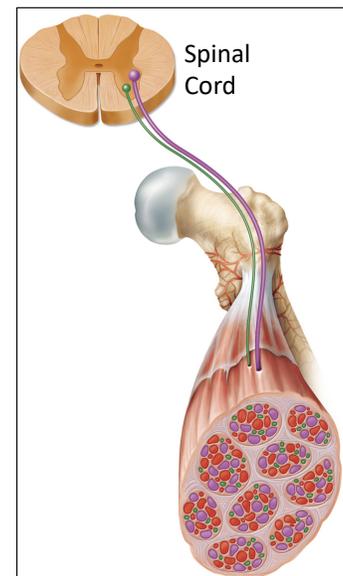
The force that a whole muscle generates during contraction depends upon several factors. The primary factors are:

- How many separate motor units are recruited.
- The size (how many muscle fibers) of the recruited motor units.
- The action potential frequency that is stimulating each of the recruited motor units.

Greater numbers of motor units, recruitment of larger motor units, or higher frequencies of action potentials lead to greater force production within the entire muscle or synergistic muscle group.

Force generation also depends upon the types of motor units that are recruited. Each motor unit has only one muscle fiber type associated with it. Type I muscle fibers have greater endurance but do not generate as great of force. Type II muscle fibers generate greater force but fatigue more rapidly.

Force generation is normally approximately proportional to the sum of all of the action potentials that are occurring within a muscle, unless muscle is fatigued and at that point each action potential does not elicit as great of force production. This combined electrical activity can be measured and quantified with an electromyogram (EMG) sensor placed superficially over the active muscles.

Force Production: Force is exerted by the muscles and this force is transmitted to tendons and to the bones upon which the muscles insert. Force production can be measured using a dynamometer. A hand dynamometer (Fig. 2) can be used to measure "Grip Force". This force is produced by a combination of flexor muscles of the hand and forearm that are utilized when gripping an object. Force production is measured in Newtons (N).

Electromyography (EMG): This procedure is used to quantify the electrical activity within a muscle. This activity reflects the combined action potentials that pass through all of the active muscle fibers that are stimulating contraction. The electrical activity in the axons leading to the muscle is actually proportionally small and negligible in comparison.

Figure 2. Hand Dynamometer

The amplitude of the EMG measure is roughly proportional to the force that the muscle exerts as more muscle fibers stimulated at higher action potential frequencies yield greater force production. EMG can be utilized as either surface EMG with electrode patches applied to the skin overlying muscle (Fig. 3), or intramuscular EMG which can utilize needle electrodes inserted in to the muscles themselves. Intramuscular EMG can be far more accurate and specific to a muscle group but is more invasive. Surface EMG is non-invasive but is less accurate and reflects all of the combined muscle activity underlying the electrodes. Surface EMG has a more difficult time detecting activity of deeper muscles and subcutaneous adipose can interfere with the signal as well.

Figure 3. Electrode patches used for surface electromyography (EMG) to measure muscle activity during hand and finger flexion.

The electrical signal is monitored by an EMG/EKG Sensor (Fig. 4) that is attached to the surface electrodes. Electrical activity is
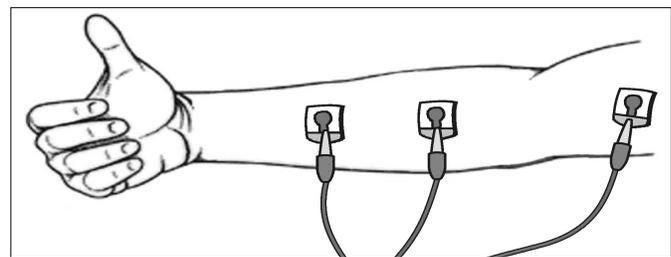
Figure 4. EKG Sensor used for Electromyography (EMG)

measured in milliVolts (mV).

<u>Muscle Fatigue</u>: Muscle fatigue is indicated by a decrease in force production, but also can be indicated by a decrease in practically any performance measure such as velocity of contraction or power output. Fatigue of the muscles that enable gripping and the decrease in force production can be measured by the hand dynamometer.

Muscle fatigue is easily observed, but the reasons that it occurs are complex and task dependent. That is to say that muscle fatigues for different reasons under different conditions. Central fatigue occurs due to a decrease in nervous stimulation due to lack of motivation or other more complex mechanisms where the central nervous system limits the number of motor units that are stimulated or limits the action potential frequency to motor units. This is thought to be an inherent protective mechanism to prevent the muscle itself from fatiguing. True muscle fatigue is known as "peripheral" muscle fatigue and is indicated by a decrease in force despite continued nervous stimulation or even increased nervous stimulation. This type of fatigue can occur due to the breakdown of any point in the chain of events leading to muscle EMG contraction following stimulation of the neuromuscular junction. EMG can be utilized to differentiate central fatigue from true muscle fatigue.

<u>Note</u>: You will also need to use a fillable PDF for submitting this assignment. "Preview" works well within the Mac and iPad world, while Adobe Acrobat (the free version) is probably the best tool within Windows or Chrome. Please install those beforehand.

Table of Contents

Equipment Setup

Locate Necessary Equipment:

- Hand Dynamometer GoDirect Sensor (Fig. 2)
- EMG (EKG) GoDirect Sensor (Fig. 4)
- Confirm that you have at least 3 Tab Electrodes (Fig. 5) to perform the experiment.
- Confirm that you have access to a tape-measure and a timer. Practice using the timer.

Figure 4. EKG Sensor used for Electromyography (EMG)

Figure 2. Hand Dynamometer

Attach Alligator Clips to Electrodes Here

Figure 5. Tab Electrodes to be attached to EMG sensor alligator clips.

GAPro Setup:

<u>Note</u>: If the Hand Dynamometer and EMG Sensor are directly connected, you can skip these steps.

- Be sure that the Bluetooth setting on your computer/device is ON.
- Power on the Hand Dynamometer and the EMG Sensor and they should flash red if properly charged and not already connected (green indicates they are already connected. If they are not charged, they should be plugged in via their USB cables.
- Open GAPro (see PreLab Document for Instructions on installing GAPro). The screen should initially resemble Fig. 6.
- Connect both devices using the Sensor Setup button. Proximity Connect will detect both devices (Fig. 7) if they are flashing red. They will quickly power down automatically if not connected, simply turn the power on again if that happens.
- Choose "Connect" for both devices, but don't hit "Done" yet. They should both flash green when connected.
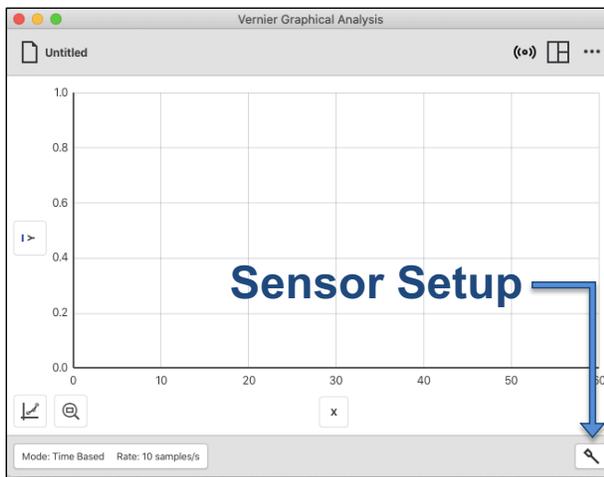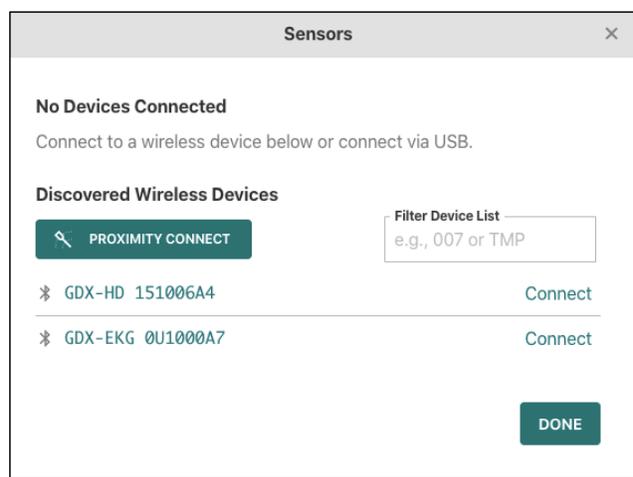

Figure 6. Initial GAPro Screen

Figure 7. GAPro Proximity Connect via BlueTooth.

- Select "Sensor Channels" for the EMG Sensor (GDX-EKG).
  - Select both "EMG" and "EMG Rectified" instead of the default of "EKG" (Fig. 8).
  - Select "Done". Note: You can return to the Sensor Setup button if you forget to do this initially.
- Your screen should resemble Fig. 9. Squeezing the dynamometer or moving around the leads should cause the readouts at the bottom of the screen to change, confirming that everything is working properly.
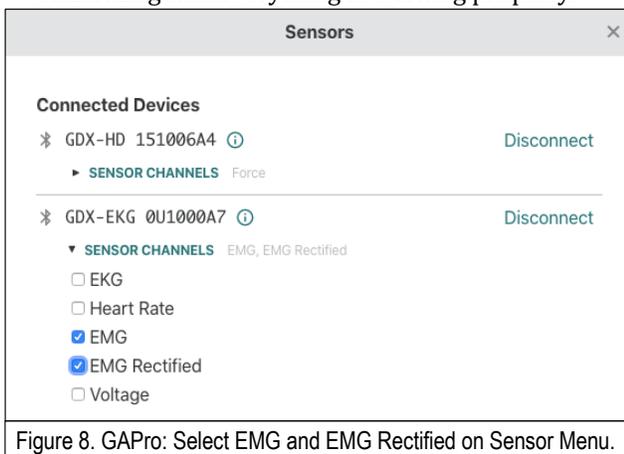

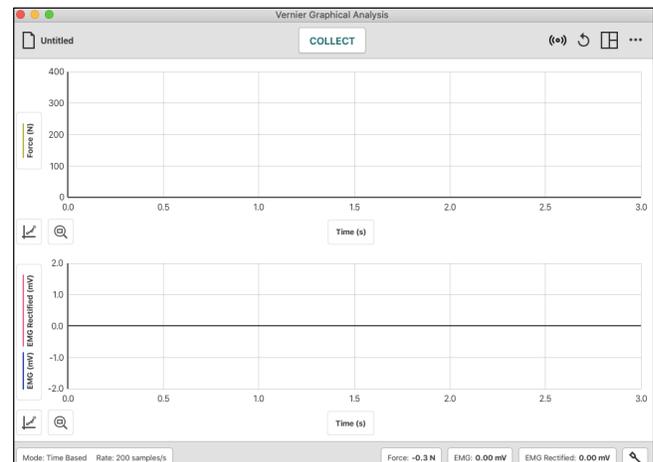Figure 8. GAPro: Select EMG and EMG Rectified on Sensor Menu.

Figure 9. GAPro - Ready to Collect Data.

- Reset Rate and Duration: Click on the "Mode" Button on the bottom left. Change The "Rate" to 100 per second (sampling frequency) and the Duration ("End Collection after") to 60 seconds (Fig. 10).
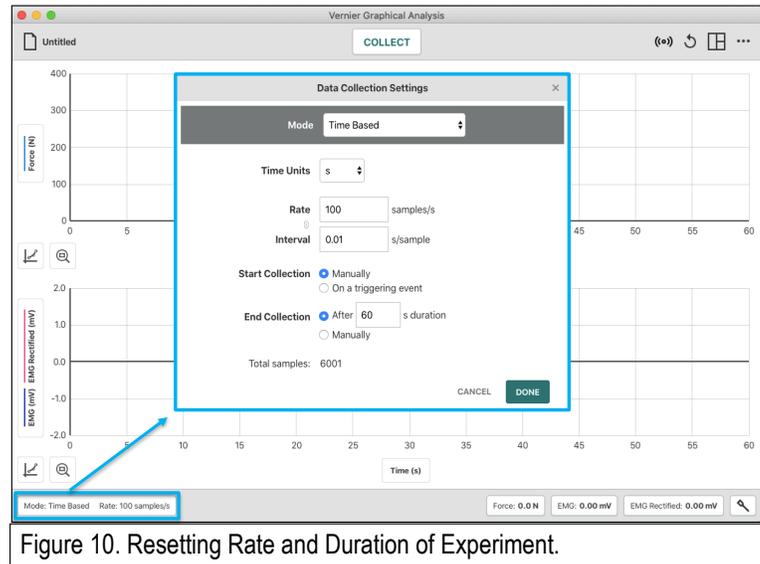
- You are now ready to collect data.

Figure 10. Resetting Rate and Duration of Experiment.

Prepare to Gather Data:

1. Select one subject from your group who will perform <u>ALL</u> experiments. Do not attempt this experiment if you suffer from arthritis, or other conditions of the hand, wrist, forearm, or elbow. Inform your instructor of any possible health problems that might be exacerbated if you participate in this exercise.
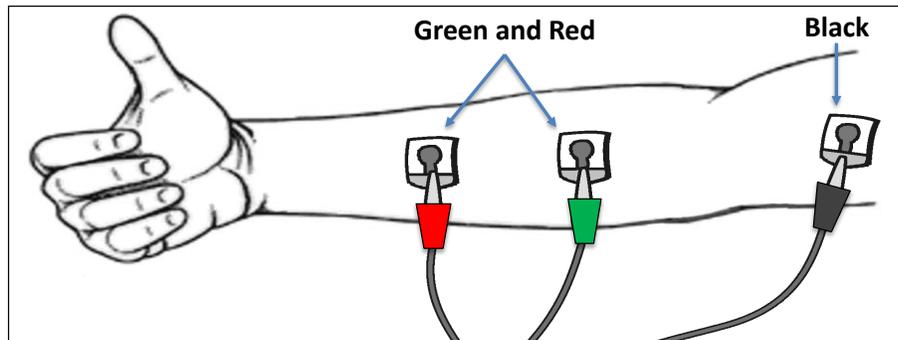

Green and Red   **Black**

Figure 11. Attach electrode tabs at indicated areas to measure electromyography (EMG).

2. Attach Electrode Tabs:

   a. Remove adhesive backing from three electrode tabs and adhere to the skin of the forearm of the dominant hand, as shown in Figure 11.

   b. Clean skin with alcohol swabs before placing electrode stickers. Two tabs should be place onto the ventral forearm, 5 cm and 10 cm from the medial epicondyle along an imaginary line connecting the epicondyle and the middle finger. The third should be placed on the medial surface of the upper arm (Fig. 9).

   c. Have the subject sit in a chair and attach the green and red leads to the forearm tabs in either order. Attach the black lead to the upper arm, this is the ground electrode. Electrical activity is measured in the areas between the red and green electrodes and this electrical activity is dominated by the electrical activity of the muscle in that area between the electrodes. Background electrical noise is subtracted from this signal using the ground.

   <u>Note</u>: Your EMG signal will be most clear if the leads are not tangled. Keep each lead wire parallel and straight to the other wires as much as possible to minimize electrical noise.
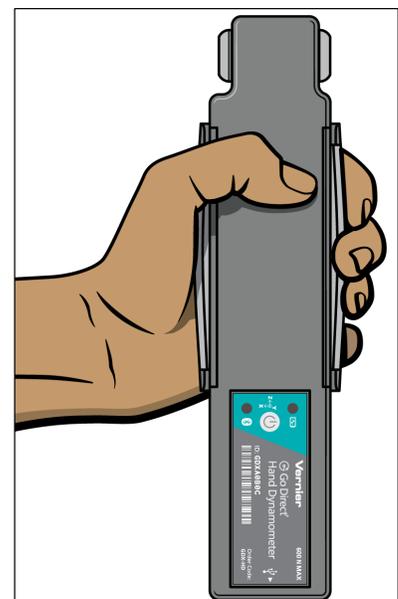
3. Practice Holding and Gripping the Hand Dynamometer (Fig. 13)

4. Zero the readings for the Hand Dynamometer in the same position.

   a. Hold the Hand Dynamometer along the sides, in an upright position NOT in contact with the lighter blue pressure panels.

   b. When the readings stabilize, Choose the "Force" Button at the bottom of the screen and select "Zero" from the pop up menu (Fig. 12).

5. Have the subject sit turned away from the data collection equipment, there should be no visual feedback of the force they are producing.

6. The subject should have their back straight and feet flat on the floor.

7. The elbow should be at a 90° angle, with the arm supported by the knee underneath.

8. The subject should use the <u>dominant hand</u> initially.

9. **Keep the subject in the same position for all 3 experiments below. They may move around between experiments in order to assist with data analysis.**

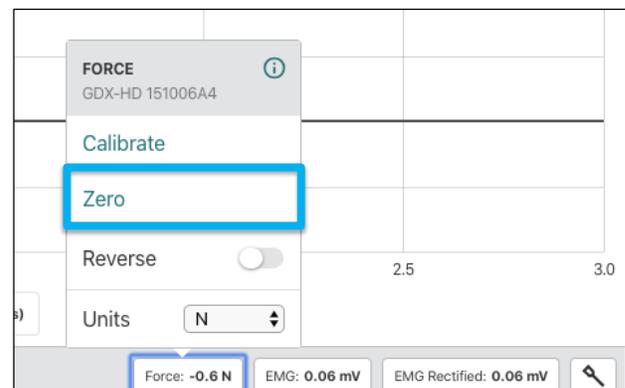
Figure 12. Gripping the Dynamometer.

FORCE
GDX-HD 151006A4

Calibrate

Zero

Reverse

Units   N

Force: -0.6 N   EMG: 0.06 mV   EMG Rectified: 0.06 mV

Figure 13. Select "Zero" on the Force Button to Zero the Dynamometer.

Experiment 1: Continuous Force Production

Data Collection:

1. Have a timer ready.

2. All ready? Click "Collect" on GAPro and start the timer and instruct the subject to "Get Ready" to grip with maximum force but do not start gripping yet.

3. At approximately 5 seconds into the data collection, alert the subject to "Begin Gripping" the dynamometer with full strength for 40 seconds. The subject should attempt to exert maximum effort throughout the data-collection period. Encourage the subject throughout the period.

4. After 40 seconds of "Gripping", instruct the subject to stop gripping but to remain seated with the dynamometer in the same position.

5. Wait for data acquisition to finish until it completes 60 seconds of data collection.

Experiment 2: Continuous Force Production by the Thumb & Index finger

1. EMG data will not be utilized in this experiment, electrodes may be removed.

2. Instruct the <u>same subject</u> to prepare to maximally pinch the fingertip force transducers at the end of the dynamometer between the thumb and index finger (Fig. 15). Continue to use the <u>dominant hand</u>.

3. Begin data collection as in Experiment 1. At approximately 5 seconds into the data collection, alert the subject to begin pinching the dynamometer with full strength and begin timing this for 40 seconds. The subject should attempt to exert maximum effort throughout the data-collection period. Encourage the subject throughout the period.

4. After 40 seconds instruct the subject to stop pinching and to remain seated with the dynamometer in the same position.

Figure 15. "Pinching" the Dynamometer.

Experiment 3: Maximum Force Production by Non-Dominant Hand

1. Using the same procedures, repeat the experiment for the opposite non-dominant hand in the **same subject** long enough to calculate maximum force production (F_{MAX}). Hit "STOP" when finished. You do not need to do the entire 60 seconds of force production and data collection for the non-dominant hand.

Export and Import Data

Saving and Exporting Data:

- Under "Untitled" in the upper Left Choose "Save As"
- Save your File to a known location on your computer.
- Email and Attach the File to your Group

Import Data:

- Save the Email Attachment to a known location on your computer.
- <u>NOTE</u>: double-clicking and opening the file directly does not work well.
- Within GAPro Go To "Untitled" → "Open"
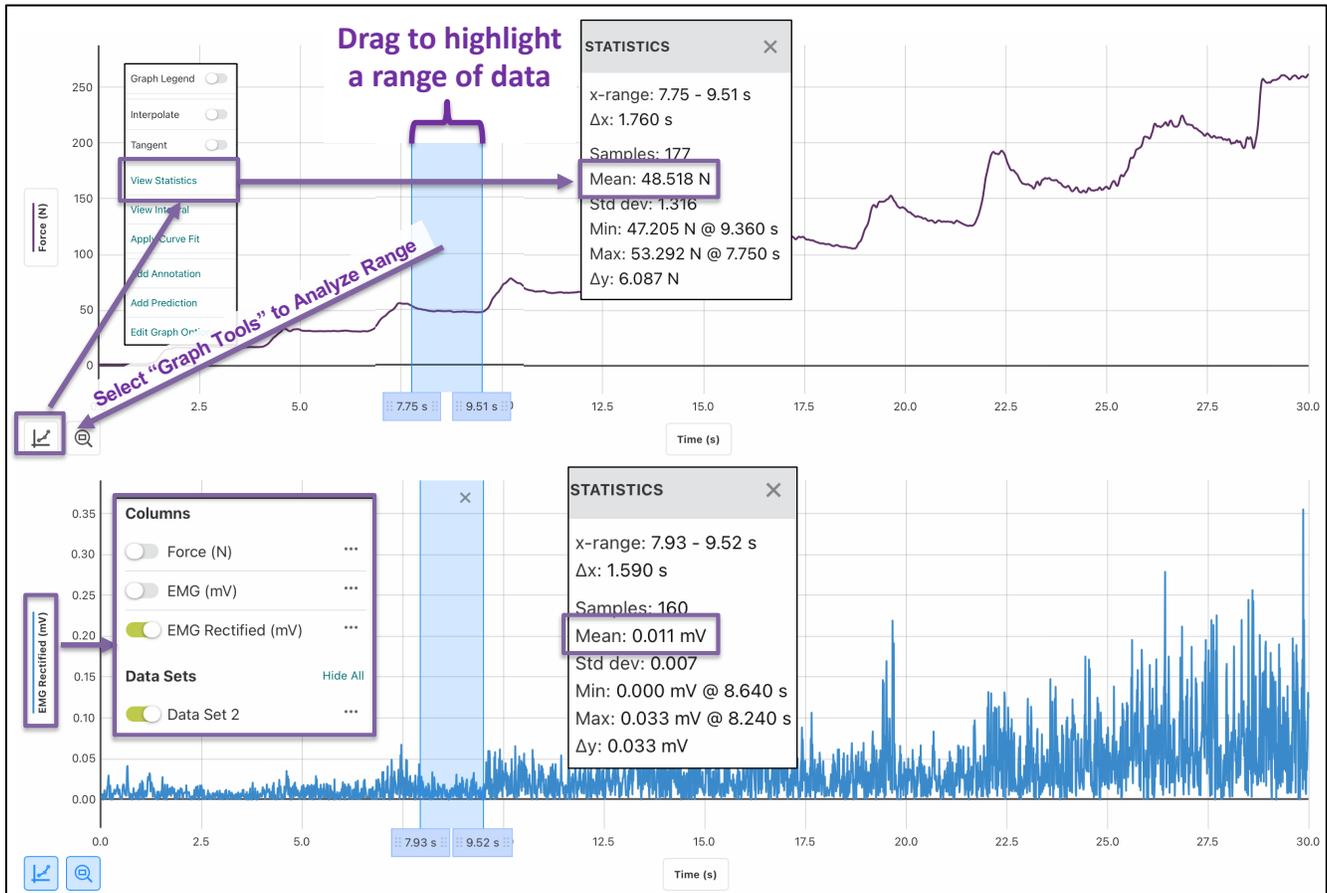- Navigate to the known location on your computer and open the file.
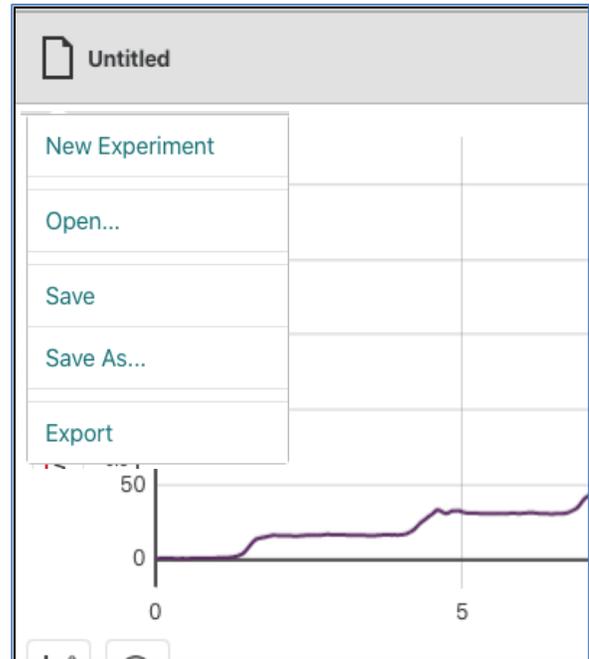
Data Analysis & Recording Data

How To Analyze Data: General Instructions

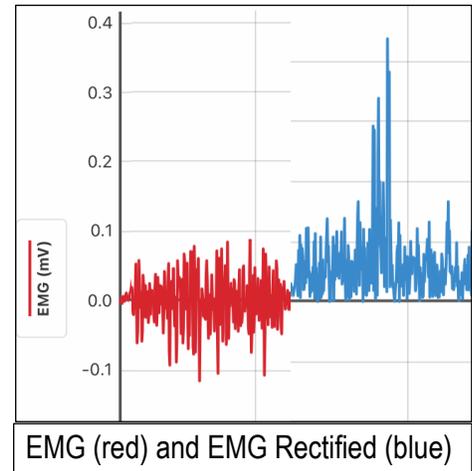### Measuring Mean Force (N):

from Hand Dynamometer (see diagram below)

- Highlight a Range of Data and select the "Graph Tools" Button on the Left
- Select "View Statistics"
- Record the Mean (N)

Measuring Mean EMG (mV):

From EMG Sensor

- Select Y-Axis Label: This Brings up the option to view EMG and/or EMG Rectified.
- Note The differences between the two Signals
  - EMG has deflections both Above and Below Zero
  - EMG Rectified is the Absolute Value of All signals
- Recording Data: Select EMG Rectified ONLY
  - Highlight a Range of Data and select the "Graph Tools" Button on the Left
  - Select "View Statistics"
  - Record the Mean (mV)
  - EMG Signal

EMG (red) and EMG Rectified (blue)

Measuring the Change in Force over Time (N/sec):

- Change in Force over Time (The Slope of Change): from Hand Dynamometer (see diagram below)
  - Select a Wider Range of Data and select the "Graph Tools" Button on the Left
  - Select "Apply Curve Fit" and select the default Fit of "Linear"
  - Record "m": The Slope of the Line.
  - Note: This relationship has a positive slope of + 7.861 N/sec. You will record a negative slope in experiments 1 and 2.
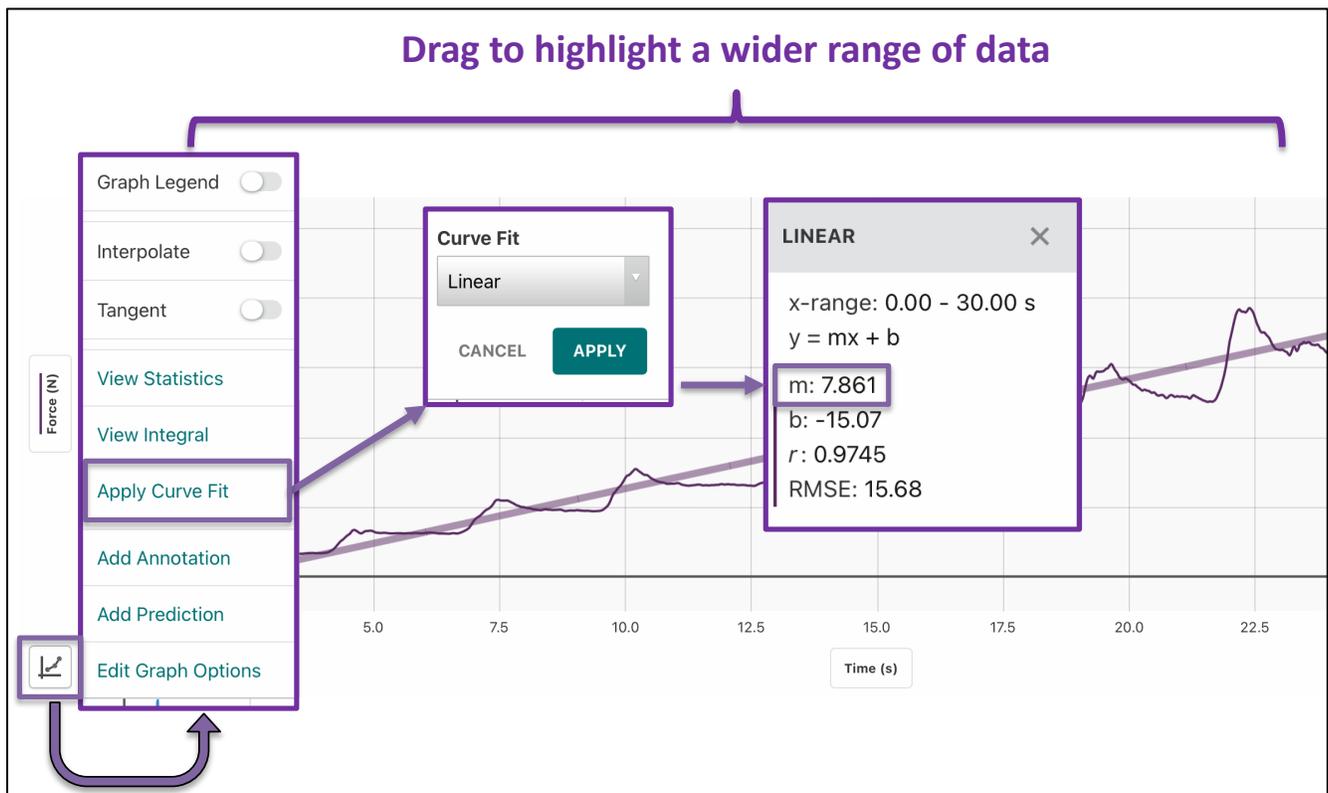
Experiment 1 Data Analysis:

- Be sure you are looking at the data from Experiment 1. On the Y-Axis click on "Force (N)" and set to "Data Set 1".
- Do the same for EMG Data – Set to "Data Set 1".
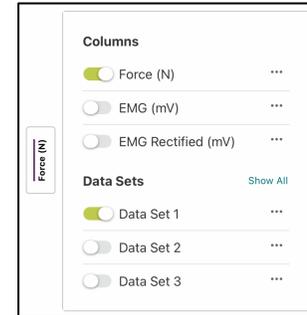
1. Measures to analyze and record from the force output from the dynamometer. Record in worksheet, Table 1a.

    a. F_{MAX} (N): Maximum Grip Force. Assess this as the mean of the first five seconds of effort.

    b. F_{FAT} (N): Fatigued Maximum Grip Force. Assess this as the mean of the last five seconds of effort.

    c. Absolute Rate of Fatigue (N / sec): This is the rate at which force decreased during the experiment. Calculate the slope of this relationship as an index of the absolute rate of fatigue (decrease in force per second): Select the entire 40 seconds of contraction data, and apply a linear curve (ex. m = -1.34 N / sec).

Figure 14. Schematic representation of force data from Experiment 1.
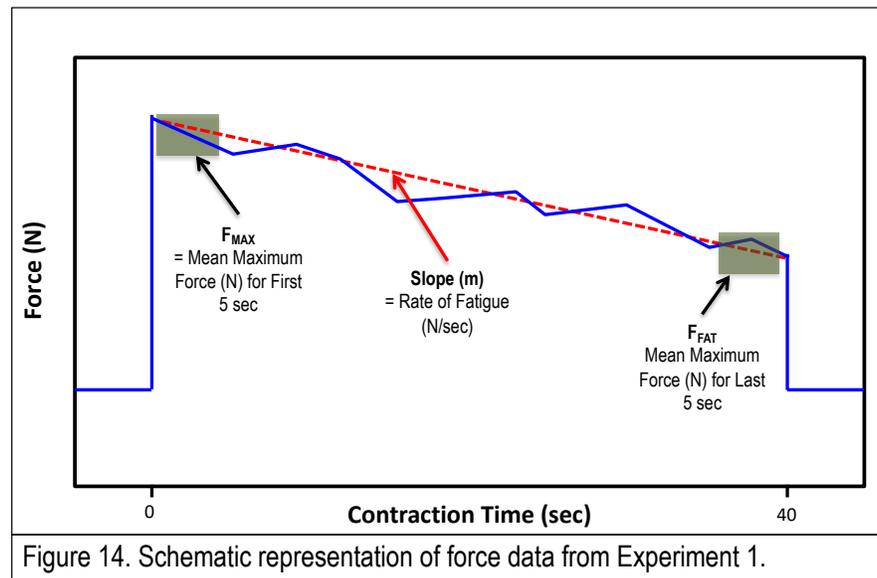
2. Measures to analyze and record from <u>EMG Rectified</u> output. Record in worksheet, Table 1a.

    a. V_{MAX} (mV): Assess this as the mean of the first five seconds of effort during F_{MAX}.

    b. V_{FAT} (mV): Fatigued Maximum Grip Force. Assess this as the mean of the last five seconds of effort during F_{FAT}.

3. Measures to calculate from the data above. In worksheet (Table 1b), show your work and record answers.

    a. Relative Rate of Fatigue: Calculate an index of the relative rate of fatigue, the rate of fatigue as the change in grip force as a percentage of maximum force.

    Relative Rate of Fatigue (% / sec) = Absolute Rate of Fatigue (N / sec) / F_{max} * 100

    b. Calculate the proportional decrease in the force production between maximum and fatigue:

    ΔF (% change) = [(F_{MAX} – F_{FAT}) / F_{MAX}] * 100

    c. Calculate the proportional increase in the EMG activity between maximum and fatigue (Note: this would be a positive number if the EMG signal increases during fatigue, and a negative number if the EMG signal decreases during fatigue):

    ΔV (% change) = [(V_{FAT} – V_{MAX}) / V_{MAX}] * 100

4. Answer the other questions in your worksheet for Experiment when time allows.

Experiment 2 Data Analysis:

- Be sure you are looking at the data from Experiment 2. On the Y-Axis click on "Force (N)" and set to "Data Set 2". Turn off "Data Set 1".

1. No EMG analysis in Experiment 2.
2. Analyze the measures of Force in Table 2 (F_{MAX}, F_{FAT}, Absolute Rate of Fatigue) from your collected data as in Experiment 1.
3. Calculate the additional measures (Relative Rate of Fatigue and ΔF) as in Experiment 1.
4. Answer the other questions in your worksheet for Experiment 2.

Experiment 3 Data Analysis:

- Be sure you are looking at the data from Experiment 3. On the Y-Axis click on "Force (N)" and set to "Data Set 3". Turn off "Data Set 2".

1. Record F_{MAX} in worksheet.
2. Answer the other questions in your worksheet for Experiment 3.

- **You may ask your GTA for help with your analyses at any point throughout the lab.**

- **Use the provided resources to answer the questions within the worksheet, you may ask your GTA for guidance.**

- **You may not ask your classmates for their answers.**

- **You should not discuss this lab with students from other lab sections.**

- **You may discuss your work with your GTA before submission.**

- **When you are Finished: Notify your GTA that you have completed the exercise and are ready to submit your worksheet.**

- **Once submitted, confirm with your GTA that your worksheet has been submitted and accepted.**

BI 301L ~ Lab G2 ~ Human A&P Muscle Physiology Lab – Student Worksheet

Name:

Be sure to include the proper units for all of your answers below. **Part 1: Continuous Force Production**

1.   Fill in the measured values* in Table 1a for F_{MAX}, F_{FAT}, Absolute Rate of Fatigue, V_{MAX} and V_{FAT} following the directions in the procedures.

2.   Calculate the other measures and add to Table 1b as in the procedures.

a. Relative Rate of Fatigue (%/sec) - Show work below including units. Note: U*se "soft" returns (shift-enter) to separate lines Show work here:*

b. ΔF (% decrease) - Show work below including units. Show work here:

c. ΔV (% increase) - Show Work Below including units: Show work here:

Table 1a. Grip Strength	
Measure	**Value**
F_{MAX} (N)*	
F_{FAT} (N)*	
Absolute Rate of Fatigue (N/sec)*	
V_{MAX} (mV)*	
V_{FAT} (mV)*	

Table 1b. Grip Strength	
Measure	**Value**
Relative Rate of Fatigue (%/sec)	
ΔF (% change)	
ΔV (% change)	

3. Is the contraction being performed in this exercise an example of a(n) CONCENTRIC, ECCENTRIC, OR

   ISOMETRIC contraction?

4. What muscles are contributing to grip force in this exercise?

5. What muscle(s) contributing to grip force are **primarily** monitored by the EMG? Why?

6. Is any type of muscle fatigue occurring during this 30 sec activity? How did you come to this conclusion?

7. How does the change in force production (between max and fatigue) relate to the change in the EMG activity (between max and fatigue)? What does this indicate about central vs. peripheral fatigue occurring?

Experiment 2: Data and Questions:

8. Fill in the measured values* for Table 2 using the same directions as for experiment 1 regarding F_{MAX}, F_{FAT}, and Absolute Rate of Fatigue.

9. Calculate the other measures in Table 2 as in the procedures using equations from experiment 1.

   a. Relative Rate of Fatigue (%/sec) - Show work below including units:

   b. ΔF (% change) - Show work below including units:

Table 2. Pinch Force		
Measure	**Value**	
F_{MAX} (N)*		
F_{FAT} (N)*		
Absolute Rate of Fatigue (N/sec)*		
Relative Rate of Fatigue (%/sec)		
ΔF (% change)		

10. Based on the F_{MAX} data from Experiments 1 & 2, how does force production compare during gripping vs. pinching?

11. What is the primary reason that accounts for any differences in observed max force production between gripping and pinching?

12. Which group of muscles fatigues more rapidly, the group involved in gripping or the group involved in pinching?Which measure(s) is(are) best used to make this conclusion and why?

13. Based on your answer to #12 above, would you expect that the gripping muscles of the forearm have a higher proportion of Type I or Type II muscle fibers (in comparison to the intrinsic pinching muscles of the hand)? Explain your answer.

Experiment 3: Data and Questions:

14. Fill in the measured value for F_{MAX} in Table 3.

15. How does this datum compare to the dominant hand? What might account for any observed differences?

Table 3. Non-Dominant Grip		
Measure	**Value**	
F_{MAX} (N)*		

16. Save this PDF with your full name as part of the title and email it as an attachment to your GTA.

17. In the same email, also attach a screenshot of your GAPro Screen showing the data set that you are analyzing.

Return this worksheet to your GTA by the end of the period.