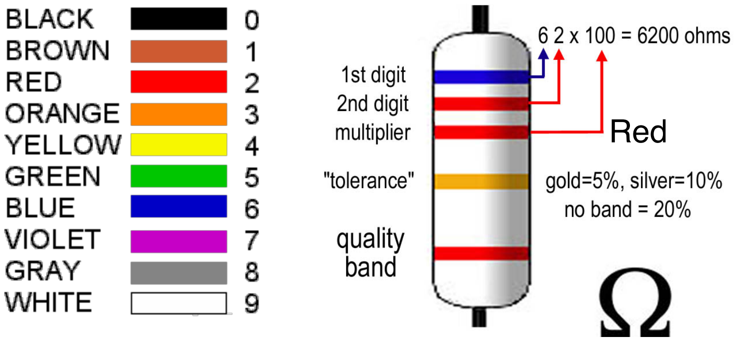
Part I. Circuits & Ohm's Law

1. Use the resistor color code to determine the resistances of your two resistors. Then measure the resistance with the voltmeter

(use the <u>lowest</u> resistance resistor as $R1$)



symbol R_{R1}: ____________

measured R_{R1}: ____________

symbol R_{R2}: ____________

measured R_{R2}: ____________

2. Read on the battery to determine its voltage: symbol V_{total}: ____________

3. Use the voltmeter to measure the battery: **measured** $\mathbf{V_{total}}$: ____________

(set the voltmeter to DC voltage and *choose the correct range* for your measurement)

4. Use Ohm's Law with measured values to calculate the total current through circuit:

measured $\mathbf{R_{Total}}$: __________ $\mathbf{I_{total}}$: ____________________

5. Calculate the voltage drop across each resistor (using *measured* values):

calculated V_{R1}: ____________

calculated V_{R2}: ____________

6. Connect the battery to the <u>outside ends</u> of the resistor chain

7. Measure the voltage across each resistor with your voltmeter:

measured V_{R1}: ____________

measured V_{R2}: ____________

measured V_{R1+2}: ____________

Does V_{R1+2} match the *measured* V_{total}? _____

Part II: Data Acquisition

Using the PowerLab 26T and LabChart software

In this part of the laboratory, we will focus on using the PowerLab 26T to acquire data by measuring voltages across the resistors. We will also learn how to set up the ***stimulus output*** of the PowerLab and control the output using the LabChart software. Finally, we will collect some data and export it to a program called IgorPro by Wavemetrics, in order to analyze and plot data in Part III.

1. Setup the hardware to collect some data. (Note: see picture of adapters on last page)

1) Connect the *DIN to BNC smart adapter* to ***Input 1*** on the PowerLab 26T
2) Connect the *BNC-to-double banana jack* to the *smart adapter*
3) Connect a red and black male *banana plug* with alligator clip to the double banana jack. You will use the alligator clips to connect to various parts of the circuit.

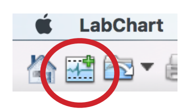
Keep positive (red) and negative/ground (black) colors correct

==> see last page for more info on ground (GND).

4) Connect the alligator clips to either side or R_1
5) **IMPORTANT** Keep the battery connected to the **ends** of the resistor chain

2. Use LabChart to Measure Voltages of the Circuit

1) Read the LABCHART8 SETUP sheet to familiarize yourself with LabChart
2) Open LabChart by clicking on the dock icon. Start a new experiment:



3) Go to *Setup>Channel Settings* and set the *Number of Channels* to 1
4) Set the *Range* to the max voltage you will likely measure (i.e., battery voltage).
5) Record a few seconds of data across each resistor by pressing ***Start*** then ***Stop***.
   Note that the voltage across the resistor is constant, so you will record a flat line.
6) Measure the voltages: V_{R1}:________ V_{R2}:________ V_{R1+R2} :__________

Does V_{R1+2} match *measured* V_{total}? __________

3. Stimulate and manipulate data with LabChart

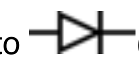
LabChart includes software control for sending an analog voltage pulse from the BNC connectors labelled "*+ Output*" and "*—Output*" on the front panel of the PowerLab 26T.

We will use this "stimulus" output as a voltage source for our mini circuit.

1) UNPLUG the battery
2) Connect the *BNC-to-single banana jack* to the ***+ Output** BNC connector*.
3) Connect the *red banana plug* with alligator clip to the *BNC-banana jack*.
4) Clip the red stimulus wire to outside the highest resistor, R_2.
5) Clip the two *Input 1* connectors across the lowest resistor, R_1.

    Make sure the black (GND) alligator is outside the red *recording* alligator.

6) Why aren't we using the GND of the ***+ Output** BNC*?______________________

    Set the voltmeter to  (***continuity)*** to see if you are right.

7) Go to *Setup>Stimulator...* and setup the Stimulator as follows:

    Baseline = 0 mV; Start Delay = 0.1 s; Repeats = 1; Max Repeat Rate = 1

    Pulse Height (V) = 0.5 V; Pulse Width (duration) = 0.5 s; Marker Channel = off

    (see LabChart8 Setup sheet for details)

8) What is the stimulus pulse voltage? V_{stimulus}: ________________

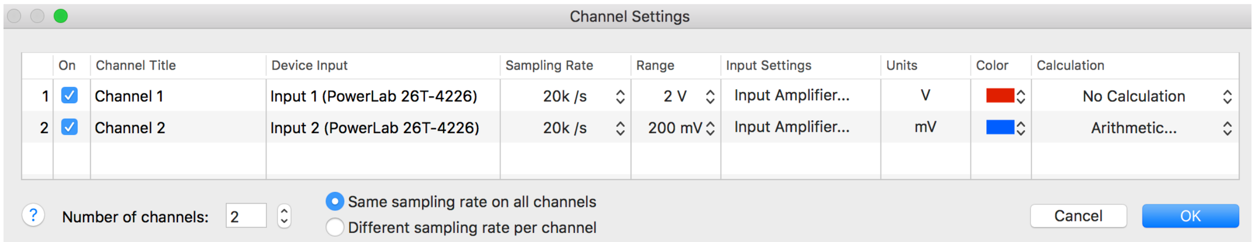
9) Go to *Setup>Sampling...* and set the duration of the experiment:

    Check the "Fixed Duration" box and set to 0.7 seconds

    (this will result in 0.1 s of baseline on either side of the 0.5 s stimulus)

Now lets use the software to also compute the resistance of the resistor R1 using Ohm's Law and the recorded voltage from Channel 1. We'll display this in real time in a second channel window.

10) Add *Channel 2*:

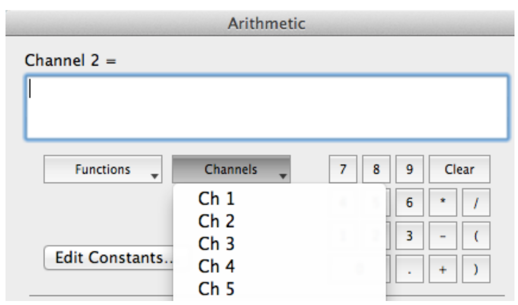


11) Calculate I_{total} from $V_{stimulus}$: I_{total}:_________ (use scientific notation e.g., 5 mA = (5*10^-3)

Use Ohm's Law to write the equation that transforms the recorded V_{R1} from Input 1 into a resistance:

$$R_{R1} = \_\_\_\_\_\_\_\_\_\_\_\_\_\_\_$$

12) Set *Channel 2* to display this Resistance by setting the Calculation to "Arithmetic". Enter your equation in the dialog window. (you can change the units to Ω symbol):



13) Now take a recording and measure the voltage drop across R_1 from Channel 1 and the resistance from Channel 2:

Channel1 V_{R1}:_________ Channel2 R_{R1}__________

IMPORTANT:You will use the ***Channel2*** $\boldsymbol{R_{R1}}$ in Igor for the next half of the lab.

Note: Each time you take a recording, it starts a new "Block" in the LabChart file. Take some time exploring how to view data within each block and jump between blocks using the numbered quick links and X-axis scaling.

4. Recording at multiple stimulus voltages

1) Change *Setup>Stimulator* to use the ***Step mode***.

   We will use Step mode to make 4 incremental stimuli as follows:

   Steps = 4; Step Width = 0.2 s; Start Height = 0 V; End Height = 4 V

Enter the four stimulus voltages: V_{stim1}:_____ V_{stim2}: _____ V_{stim3}: _____ V_{stim4}: ______

Calculate the voltage drop at R1 (V_{R1}) for each stimulus voltage:

Stim1 calc V_{R1}: _____ Stim2 calc V_{R1}: _____ Stim3 calc V_{R1}: _____ Stim4 calc V_{R1}: _____

2) Set the *duration* of Channel 1 to 1 sec (via *Setup>Sampling…*) to record for the length of the stimulus (i.e., 4 steps * 0.2 ms) within the scope window (see LabChart8 Setup sheet)

3) Record and measure the voltage across the R_{R1} resistor at each stimulus voltage:

Stim1 meas V_{R1}: ____ Stim2 meas V_{R1}: ____ Stim3 meas V_{R1}: _____ Stim4 meas V_{R1}: ____

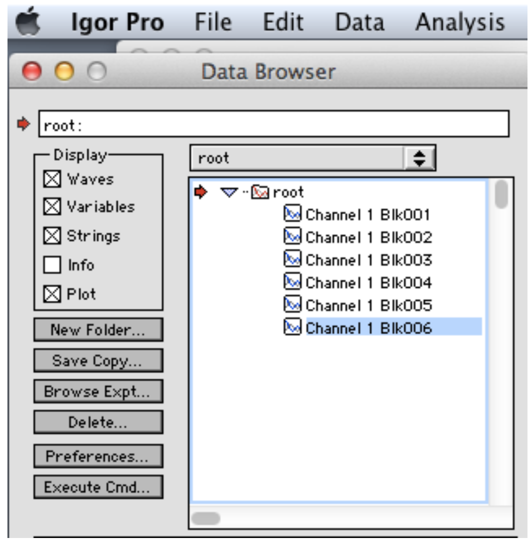
4) Now save the data in Igor format by using *Export As…* select *File Format: Igor*

   **If given the option, select *Channel 1* from the drop-down menu

5) Save your file to the desktop.

Part III: Analysis & Plotting

1) The Igor file is on your desktop. Double-click it and your data will load into *Waves* in Igor (similar to Spreadsheets in Excel). Waves are viewable in the *Data Browser*:



2) Double-click the *Wave* corresponding to the *Block* of data containing your step experiment (likely your last recorded block)

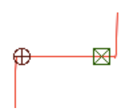
3) You should now see each data point as a column of Y values in "Table0".

4) Now use *Windows>New Graph...* and create a new graph using the Block Wave as the Y-wave and "*-calculated-*" as the X-wave.

5) With our new graph, use *Graph>Show Info* to open a drawer beneath the graph.

6) Place cursors on the graph by dragging the **A** and **B** cursors onto the graph from the drawer.

Place the cursors on one of the steps:

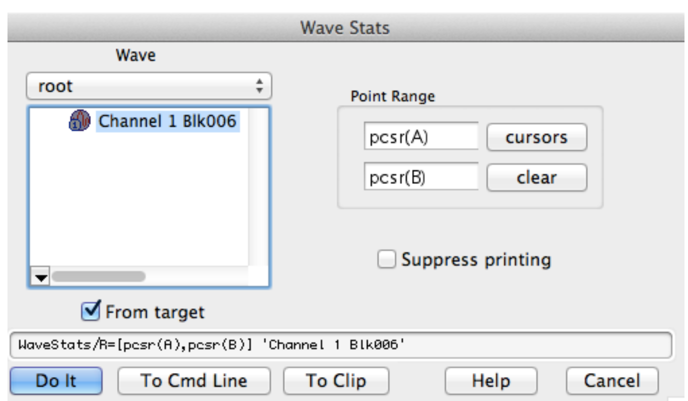


7) Use *Statistics>Wave Stats...* to compute the average voltage of the steps:

Note: Be sure to select "cursors" and check the box for "From target":

Click "*Do It*" and the stats for the Wave will be printed out in the *Command* Window at bottom of screen.

The "V_avg" value listed in the command window is the average voltage value between the cursors.

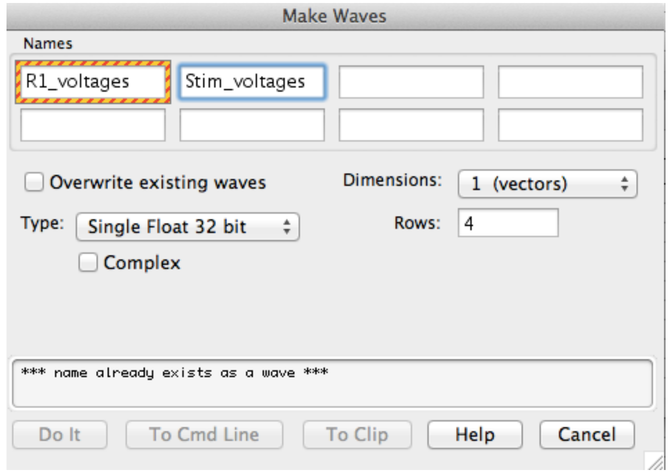


Repeat this process for each stimulus voltage step and record your data in Volts:

V_avg for Step 1 ___________ V_avg for Step 2 ___________

V_avg for Step 3 ___________ V_avg for Step 4 ___________

8) Make two new Waves by clicking *Data>Make Waves...* enter the names as shown and set the **number of rows to 4**.:



9) Go to the Data Browser and open the two new Waves.

10) Enter your 4 step voltages in the "Stim_voltages" wave and the 4 *V_avg* voltages in the "R1_voltages" wave.

11) Now make one more Wave called "R1_currents" . In the Make Waves dialog box, again set the number of rows to 4.

12) Use the Command line in the Command Window to enter the following equation in order to do math on our Waves:

Type: "`R1_currents = R1_voltages/(`type your Channel2-measured R_{R1}`)`"

13) Hit Enter and the calculated values will fill your *R1_currents* wave. Double-click the Wave to confirm the values are shown.

14) Now graph the newly calculated "R1_currents" wave (Y-axis) versus the "Stim_voltages" wave (X-axis).

Remember for resistors in series, I_{R1} is I_{total} for each stimulus voltage!

In Igor, the default for graphs is a line connecting data points, lets see the actual data points:

15) Double-click the line on the graph. Change the *Mode* to "Markers" and select "filled circles" from the drop down menu. Set the marker size to 5.

16) Now lets fit the data with a linear equation. Select *Analysis>Curve Fitting...* Be sure "Function" is "Line" and be sure "From Target" is checked (so that it fits the points in the newly created graph).

The equation for a line is displayed, note what the meaning of "a" and "b" variables

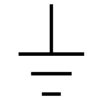
17) Finally, recall what the slope of this line tells you:

Determine the R_{total} for your circuit: _______________

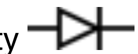
Given R_1 and R_{Total}, determine the R_2 for your circuit: ______________

Does your graphed data and fit line accurately show the Total Resistance? ______

Positive, Negative, and Ground

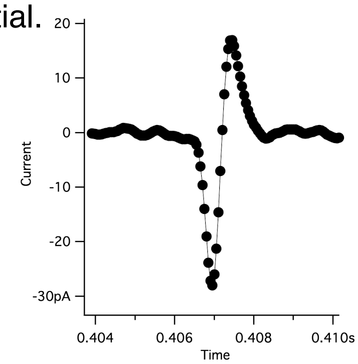


In electrophysiology, we typically think of regions as positive or negative when compared to Ground (abbreviated GND), which is 0 V. Ground is literally the earth (note that the symbol for ground is a stake into earth). The third prong in most outlets, completes a circuit with the ground outside (usually by way of a big copper wire that leads from the electric panel to outside). In the world of recording from neurons, a ground wire is usually placed in the extracellular bath of your experiment—this then is continuous with the third prong in the wall (back in the day it would lead to a metal pipe of a sink in the lab or to a stake outside). BNC connectors typically have a center "positive" terminal, and the surrounding ring is GND (instead of negative).

1. If GND is indeed ground, where can you check for continuity  of equipment in front of you? ______________

Sampling

When you collect data, the instrument is taking measurements (collecting information) at discrete time points. The time interval in between measurements is called the "sampling interval". Therefore, the sampling rate is the inverse of the interval and generally expressed in samples/sec or Hz (1 Herz = 1 cycle/1 second). The rate for sampling is determined by the nature of data being collected. Action Potentials (APs) are only 1 millisecond in length, so your sample rate should be fast enough to take several measurements during the duration of an action potential.



For this laboratory, the sampling interval is 50 μsec

What is the sampling rate: ____________

Is this fast enough sampling for measuring APs? _________

How many samples will be taken during an AP?__________

CONNECTORS

BNCconnector = Bayonet Neill–Concelman connector
Male: Female:

Banana Plug: 

BNC-to-female banana: