Simulation Exercises and Homework Questions – Passive Membrane Properties

The simulations you will perform in class use the Electrophysiology of the Neuron simulation software. This software simulates whole-cell patch-clamp experiments and it provides very realistic simulations of what the outcomes of actual experiments would look like.

Begin by running the SimCC program. This simulates patch-clamp experiments in the "current-clamp mode", which is to say that the experimental configuration allows you to deliver current to the cell while recording the voltage of the membrane.

1. Simulations: Changing membrane resistance. These exercises will help you to answer these questions from the first homework assignment:

Homework Question 1a: Describe how the membrane voltage response changes as you increase and decrease resistance. What are two things about the voltage response that you notice changing?

Homework Question 1b: What are the most notable differences between the voltage response at lowest resistance (pKleak = 1) and the highest resistance (pKleak = 0.35).

- Open the file StepResistance.cc5 and select Run | Begin. This will produce one "sweep" of data. At the bottom of the screen in red is the current you injected into the cell. At the top is the membrane voltage response.
- Now, increase the membrane resistance. To do this you will decrease a resting potassium (K^+) conductance (i.e., you will close K^+ channels that are normally open at resting potential).
- On the menu, select Parameters | Conductances then change the value for pKleak (for passive K^+ leak) Change this value from 0.5 to 0.45. **Remember, by *decreasing* conductance (closing ion channels) you are *increasing* resistance, and vice-versa**.
- Click OK then at the main menu select Run | Overlay. The "overlay" command will run a new simulation sweep while leaving your previous data on the screen, choosing "Run" will clear all your previous data before starting the sweep.
- Repeat this process for K^+ leak values of 0.4 and 0.35. **Take a screenshot of the superimposed sweeps to include in your homework.**
- Now, repeat this process starting at a pKleak value of 0.5, then using values of 0.6, 0.7, and 1 to simulate *decreasing* membrane resistance.
- Run two sweeps that will superimpose the membrane voltage responses when pKleak is 1.0 and when it is 0.35. Show these traces to your instructor. **Take a screenshot of the superimposed sweeps to include in your homework.**
- Try repeating the above steps but with a negative stimulus current. Go to Parameters | Protocol and make the Injected Current negative.

2. Simulations: Changing membrane capacitance. These exercises will help you to answer this question from the first homework assignment:

Question 2a: Describe what is changing about the membrane voltage response as you change the membrane capacitance. How is this different from what happened when you changed resistance?

- Load the file StepCapacitance.cc5 file (don't save changes to the previous file)
- Run a sweep of data. The outcome should look familiar to you.
- You will now change the cell capacitance by changing the surface area of the cell. Remember – capacitance is a direct function of the surface area of the cell. At the top menu select Other | Misc. parameters. You will be changing the value for "Membr. Area (sq μ)" The initial value is 12500 (which is 1250.0 μm^2) the surface area of a spherical cell 20 μm in diameter. Change this to 28000 (the area of a 30 μm diameter cell) then overlay this sweep on your first one.
- Change the area to 50000 (approximate area of a 40 μm diameter cell) and overlay this response on the previous two. **Take a screenshot of the superimposed sweeps to include in your homework.**

3. **<u>Simulations – responses to a "realistic" stimulus as resistance changes.</u>** In real life, neurons never receive stimulus currents that are square steps. Neurons receive synaptic inputs (in the form of postsynaptic currents). These currents have a distinctive shape. The most common excitatory postsynaptic current (EPSC) is the AMPA receptor current, which is what we will simulate now. These simulations will help you answer this homework question:

Question 3a: Describe what is changing about the membrane voltage response as you increase and decrease resistance. What are two things that you notice changing? How do these data relate to what happened when you were using a square step pulse?

- Load the file EPSCsingle_resistance.cc5 and run a sweep of data. The blue line at the bottom is the simulated EPSC that you are injecting into your cell, and this mimics one excitatory synaptic input to the cell (an AMPA receptor current to be exact). In blue at the top is the membrane voltage response to this EPSC.
- Find out how changing resistance changes the membrane response to this synaptic input. Increase resistance by changing pKleak from 0.5 to 0.45. **Remember, by decreasing conductance (closing ion channels) you are increasing resistance, and vice-versa**. Overlay this sweep on your first sweep.
- Now repeat this process for K^+ leak values of 0.4 and 0.35.

4. **<u>Simulations – responses to a train of synaptic inputs as resistance changes</u>**. Neurons often receive multiple inputs in succession or at the same time. How cells "interpret" multiple inputs is a primary and fundamental level of information processing in the nervous system. You will now examine how the cell's "interpretation" of multiple inputs changes as the membrane passive properties change. These exercises will help you answer these two homework questions:

Question 4a: Describe what happens as you increase resistance and when you decrease resistance.

Question 4b: When does the membrane show greater integration of its inputs over time? When does the membrane show the best fidelity to its inputs? (That is, when does the membrane potential best reproduce the inputs it is receiving).

- Load the two_epsc.cc5 file (don't save the previous file).
- Run one sweep of data, then run three more sweeps of data after changing pKleak to 0.45 then 0.35 then 0.30 (remember – this is the same as increasing resistance). What happens as you increase resistance? **Take a screenshot of the superimposed sweeps to include in your homework.**
- Now run another set of sweeps where you decrease resistance. Start with pKleak at 0.35, then superimpose a sweep with pKleak at 1.0 **Take a screenshot of the superimposed sweeps to include in your homework.**

5. **<u>The big picture</u>** Given all that you have learned, and what you witnessed in these simulations answer these two questions:

Question 5a: How does membrane resistance affect neural function?

Question 5b: How does membrane capacitance affect neural function?

Question 5c: If you had to design a cell that required a very strong input in order to fire, but it had to reach threshold very quickly once that stimulus was present, what kind of cell would you design? (How would you make it have the proper passive properties?)

Question 5d: If you were designing a cell that needed to be very sensitive to small inputs and to sum up many inputs over time, what kind of cell would you design? (How would you make it have the proper passive properties?)

<u>HOMEWORK ASSIGNMENT</u>:

Submit your answers to questions 1-5 as a MS Word or PDF document through Canvas under the assignment "Homework 1".

Where appropriate, include the screenshots you took during your simulation. These are your data figures. Your answers should refer to your data to help support your answers. It might be helpful to annotate these figures (arrows, labels, etc). At the end of the assignment, you must include a "Statement of Contributions" that describes how each team member contributed to the assignment.

Question 1a: Describe how the membrane voltage response changes as you increase and decrease resistance. What are two things about the voltage response that you notice changing?

Question 1b: What are the most notable differences between the voltage response at lowest resistance (pKleak = 1) and the highest resistance (pKleak = 0.35).

Question 2: Describe what is changing about the membrane voltage response as you change the membrane capacitance. How is this different from what happened when you changed resistance?

Question 3: Describe what is changing about the membrane voltage response as you increase and decrease resistance. What are two things that you notice changing? How do these data relate to what happened when you were using a square step pulse?

Question 4a: Describe what happens as you increase resistance and when you decrease resistance.

Question 4b: When does the membrane show greater integration of its inputs over time? When does the membrane show the best fidelity to its inputs? (That is, when does the membrane potential best reproduce the inputs it is receiving).

Question 5a: How does membrane resistance affect neural function?

Question 5b: How does membrane capacitance affect neural function?

Question 5c: If you had to design a cell that required a very strong input in order to fire, but it had to reach threshold very quickly once that stimulus was present, what kind of cell would you design? (How would you make it have the proper passive properties?)

Question 5d: If you were designing a cell that needed to be very sensitive to small inputs and to sum up many inputs over time, what kind of cell would you design? (How would you make it have the proper passive properties?)

Statement of Contributions: Describe how each team member contributed to this final product.

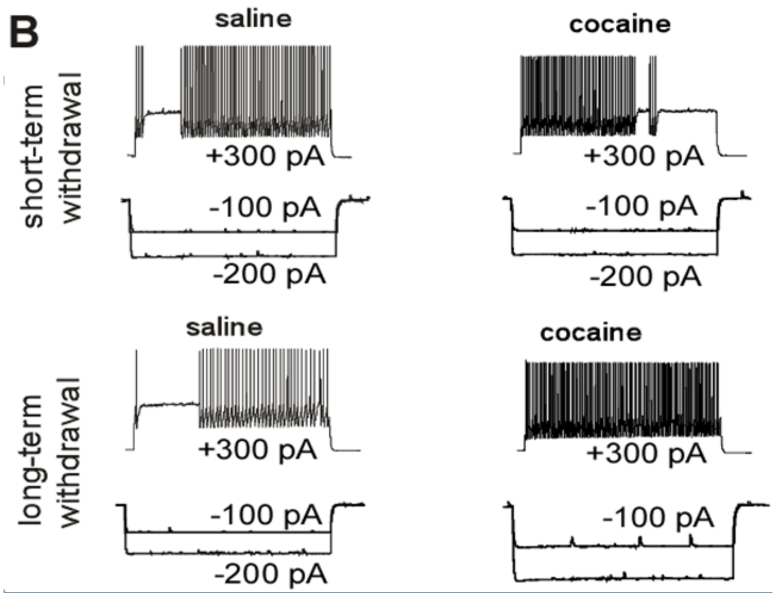
Why are we doing this? Take a look at some questions from prior exams on the next page

From Exam 1 Fall 2013

The figure below is taken from an article published in the Journal of Neurophysiology in June 2013: Campanac, E., Hoffman, D. A., (2013). Repeated cocaine exposure increases fast-spiking interneuron excitability in the rat medial prefrontal cortex. *J. Neurophysiol*. 109**,** 2781-2792.

The experimenters injected one group of rats with cocaine every day for five days, while another group received saline control injections on the same days. They then recorded from neurons in the rats' prefrontal cortex (a region of the brain associated with planning). Some recordings were made at 3-6 days after the last injection (the short-term withdrawal group) while other recordings were made 10-13 days after the last injection (the long-term withdrawal group).

Question 3: (5points) What do the cell's responses to the -100 pA and -200 pA step currents in Panel B tell you about the effects of long-term withdrawal on the neurons' passive properties?



From Exam 1 Fall 2012

Question 11. (10 points) At right is a figure from an article in The Journal of Neuroscience. The experimenters recorded from neurons in the cortex of normal mice (wild type) and mice that were missing the gene for a particular type of ion channel (HCN1 KO). The experiment delivered a train of five excitatory postsynaptic currents to the cell and measured membrane potential (MP). [redacted] From the data shown in Panel D, what do you think were the effects of the drug bicuculline in cells from the HCN1 KO mice? Be sure to explain your answers.