Please make sure to do the reading prior to completing the problem set.

Equilibrium Potentials II Lab Notebook

Reading: Equilibrium Potentials II

Name:
Date:
Purpose of Lab:

Question 1: Solve the following problem, assuming that the membrane is in equilibrium.

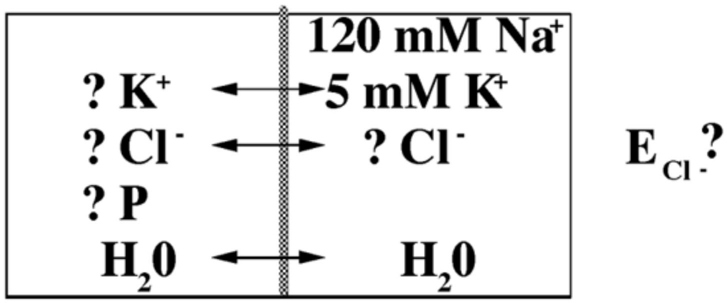

Figure 7: Solving the unknowns can generate a cell that is fully in equilibrium

You must follow the instruction in the reading step by step for this problem. *Please solve for the specific values given in Figure 7, including the value of the membrane potential*. **Please include your work and the results of your calculation in your notebook and check your work.**

Question 2: Solve the following problem, assuming that the membrane is in equilibrium.

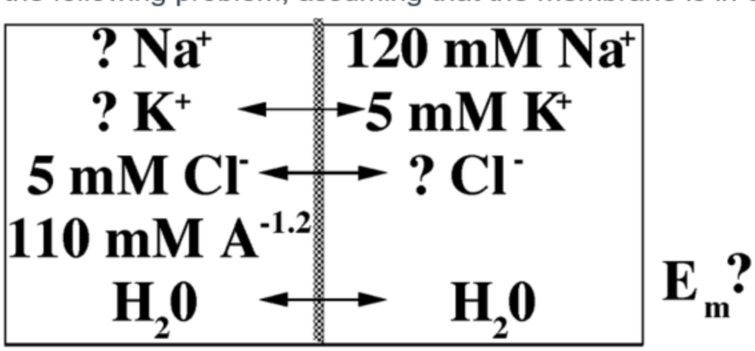

Figure 8: Solving the unknowns can generate a cell that is fully in equilibrium until sodium ions become permeable

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You must follow the instruction in the reading step by step for this problem.. *Please solve for the specific values given in Figure 8, including the value of the membrane potential.* **Please include your work and the results of your calculation in your notebook and check your work.**

Question 3: What would happen if the membrane suddenly also became permeable to sodium ions? **You must follow the instruction in the reading step by step for this problem.** *Use the values of internal and external sodium ions that you computed to find the equilibrium conditions for the cell shown in Figure 8. Is this value similar to the equilibrium membrane potential that you computed for the membrane?* **Please include your work and the results of your calculation in your notebook and check your work.**

Question 4: You must follow the instruction in the reading. Using the values computed for the model cell shown in Figure 8, *estimate the resting potential of the model cell.* Assume that the nerve cell is equally permeable to sodium, potassium, and chloride ions, i.e., $p_{Na^+} = p_{K^+} = p_{Cl^-} = 1$. **Please include your work and the results of your calculation in your notebook and check your work.**

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Simulation Problem 1:

a. Start the Nernst Potential Simulator, and load the file *Problem2A.init* **(note that due to a recent renumbering of the problem set, this file name does not match the problem number!)**. Note that the model membrane is now permeable solely to potassium, and that both Selective Permeability and Electrostatics are active; thus, you will note that the channels in the membrane only allow potassium ions through, and the program changes the probability that ions may cross the membrane based on the membrane potential. The membrane potential is always initialized to 0 mV; you can imagine that there are impermeable charges not shown (such as charged proteins inside the cell) that provide the appropriate charge balance to make the membrane potential initially 0 mV. Before starting the simulation, *please calculate the predicted potential across the membrane using the Nernst equation. Show your work*. Note that the temperature in this simulation is 298 K (which is warmer than we earlier assumed) and that actual concentrations in the simulator are rounded to the nearest whole number of ions. Thus, the value predicted by the simulation (the red line on the plot) may not exactly match your calculation.

b. Now start the simulation briefly, immediately pause it, and click on a red ion on the left side (the intracellular side) so that you can readily track the movements of the ion. Then continue the simulation. *Describe the behavior of the single ion, and of all the potassium ions. Also, explain the changes in the graph of membrane potential (black line), and contrast it to the predicted value (the red line). Predict what will happen over long periods of time. Run the simulation for 5,000 iterations to see if your prediction is correct. Explain the results.* ***Take a screenshot of the simulation for your lab notebook, and be sure to include the Membrane Potential plot in the picture.***

Paste screenshot here. Make sure to label the figure, you can do this by right clicking the image and clicking “Insert Caption”

c. Press the "Reset with Default Settings" button. Note that the zoom window shows channels selectively permeable to potassium, sodium or chloride ions (tinted red, blue, and green, respectively). Before running the simulation, *use the Goldman-Hodgkin-Katz (GHK) equation to calculate the predicted membrane potential* (the simulation provides you the concentrations of the key ions inside and outside of the cell, as well as their permeabilities, which you need to solve the equation). *Show your work*. Again, because of small differences in temperature and concentration, your calculation may differ slightly from the value predicted by the simulator.

d. Briefly start the simulation, pause it, and select a potassium (red), sodium (blue) and chloride (green) ion to track. *Describe the movements of the selected ions and the graph of the membrane potential relative to the predicted potential* (red line). *Predict what will happen over time*. Run the simulation for about 5,000 iterations to see if your prediction is correct. *Explain the results*. ***Again, take a screenshot of the simulation for your lab notebook, and be sure to include the Membrane Potential plot in the picture.***

Paste screenshot here. Make sure to label the figure, you can do this by right clicking the image and clicking "Insert Caption"

e. The simulation represents only a very small number of the very large numbers of ions that are actually present in real nerve cells. To see how this may affect the results, press the "Reset with Default Settings" button, and then, under the File menu, select Load Initial Conditions, and load the file *Problem2B.init* **(note that due to a recent renumbering of the problem set, this file name does not match the problem number!)**. The initial conditions are the same, but the "world's" thickness has been greatly reduced. This would be similar to a dendritic spine surrounded by a limited extracellular space. *Predict what may happen* under these circumstances to the concentration gradients, and to the long term potential across the membrane. Now run the simulation for 5,000 iterations, and see what happens. ***Once more, take a screenshot of the simulation for your lab notebook, and be sure to include the Membrane Potential plot in the picture.***

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After about 5,000 iterations have passed, pause the simulation, *and calculate the new predicted potential* from the measured concentrations (found in the table on the right) and the GHK equation. *Explain the results*, contrasting the mechanisms generating the resting potential with those that generate the Nernst potential for a single ion (parts **a** and **b** of this question). *How is it that the results you obtained are not observed in most nerve cells?*

Simulation Problem 2:

a. Start the Nernst Potential Simulator, and load the file *Problem3A.init* **(note that due to a recent renumbering of the problem set, this file name does not match the problem number!)**. *Predict the final membrane potential using the GHK equation. Show your work.* Note that, as in Problem 1, because of small differences in temperature and concentration, your calculation may differ slightly from the value predicted by the simulator.

b. Now start the simulation briefly, immediately pause it, and click on a potassium (red), sodium (blue) and chloride (green) ion to track them. *Describe the movements of the selected ions and the graph of the membrane potential relative to the predicted potential* (red line). *Predict what will happen over time.* Run the simulation for 10,000 iterations to see if your prediction is correct. ***Take a screenshot for your lab notebook, including the plot of the Membrane Potential.***

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From the actual measured intracellular and extracellular ion concentrations (found in the table on the right) and the GHK equation, *what should the final membrane potential be? What has happened to account for the change in the initial predicted membrane potential, and that which you observe after 10,000 iterations?*

c. In this problem, you will design your own neuron with a unique set of ionic parameters. Devise a set of **concentrations and permeabilities** to represent a nerve cell at rest with a negative resting potential. In your imaginary neuron, movement of **chloride** ions should be primarily responsible for the resting potential (this implies that neither sodium nor potassium ions contribute significantly to the resting potential). *Report the concentration and permeabilities in the table.*

Next, **using the same concentrations**, devise a **different set of permeabilities** to represent a neuron at the peak of the action potential (i.e., at a positive membrane potential in the range of +10 mV to +50 mV, approximately; the action potential is a temporary depolarization of the membrane that neurons use to send signals across long distances; you will learn much more about this in the coming units). Movement of **potassium** ions should be primarily responsible for the depolarization at the peak (this implies that neither sodium nor chloride ions contribute significantly to peak of the action potential). *Record these permeabilities in the table in your lab notebook.*

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Parameters for a Designed Neuron

	Concentration (mM)		Permeability	
	Intracellular	**Extracellular**	**Resting Potential**	**Peak of Action Potential**
K^+				
Na^+				
Cl^-				

Using the GHK equation, *calculate the membrane potential of the neuron you have designed for both sets of permeabilities.* ***Show your work and your answers in your lab notebook.*** If necessary, revise your parameters until the membrane potential meets the criteria stated above for both when the neuron is resting and when it is at the peak of the action potential.

Now test your values in the simulator (both the "Selective Permeability" and "Electrostatics" boxes should be checked). Enter the concentrations and permeabilities for the cell at rest, run the simulation for a few thousand iterations, and ***take a screenshot.*** Next, pause the simulation and change the permeabilities to those you chose for a cell during the action potential. Run the simulation again to see what happens, and ***take another screenshot.*** You can imagine that if you had such a cell with a membrane capable of selectively changing its permeability under certain conditions, you would have a cell capable of very interesting behavior!

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Question 5: Is the squid giant axon in osmotic balance? If not, would it shrink or swell? **Please include your work and the results of your calculation in your notebook and check your work.**

Question 6: Do the particles on either side of the squid giant axon satisfy electroneutrality? **Please include your work and the results of your calculation in your notebook and check your work.**

Question 7: Finally, what are the Nernst or equilibrium potentials of the three permeable ions (sodium, potassium, and chloride)? Based on calculating these potentials, is the squid giant axon in equilibrium, or in a steady state? **Please include your work and the results of your calculation in your notebook and check your work.**

Simulation Problem 3: Using the constant field equation.

A neuron's internal and external concentrations of potassium, sodium and chloride have been measured. The internal concentration of potassium ions is 168mM, of sodium ions is 50mM, and of chloride ions is 41mM. The external concentration of potassium ions is 6mM, of sodium ions is 337mM, and of chloride ions is 340mM. At rest, $pK+:pNa+:pCl-$ is 1:0.019:0.381.

a. *Predict the resting potential V_m from the Goldman-Hodgkin-Katz equation.*

b. *What would be the effect of a tenfold increase in external potassium concentration on the resting membrane potential? What would be the effect of a tenfold decrease in external potassium concentration on the resting membrane potential? Predict the effects using the GHK equation, and confirm your predictions by running the simulation and* ***taking screenshots for your lab notebook*** (when running the simulation, round the external potassium concentration to the nearest integer). *Explain.*

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Discussion