NAME ____

MCB 166 TAKE HOME EXAM October 5, 2017

This exam consists of six questions, each has four to five parts. Please try to clearly show your line of reasoning and/or work. On the other hand, please do try to keep your answers reasonably brief.

Remember to put your name at the top and to hand in this page along with your exam. Please start your solution to each question on a separate page, write your name on every page, and staple your pages together. PLEASE WRITE YOUR NAME AND ANSWERS LEGIBLY.

You are allowed to consult any resource you find useful, but please respect the Honor Code and make sure the work you hand in is your own. Please do not collaborate with fellow students on the exam. You are welcome to ask us questions of clarification, but not questions directly related to the problems in the exam. You are welcome to use the Discussions page on bCourses for this purpose.

Exams are due at the start of class (11 AM) on Tuesday, October 10.

Good luck!

Scores:

(1)---------------------------(15 points total)

(2)---------------------------(25 points total)

(3)---------------------------(15 points total)

(4)---------------------------(25 points total)

(5)---------------------------(20 points total)

(6)----------------------------(20 points total)

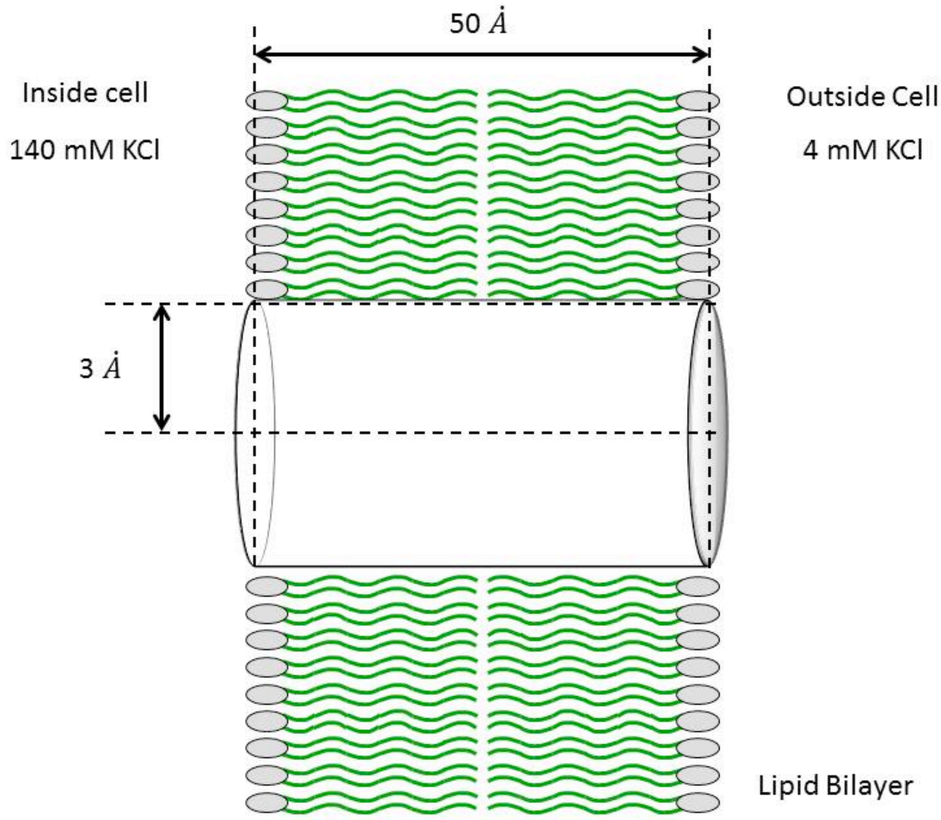
Total ----------------------- (MAX pts possible = 120)

1. Consider the simple cylindrical model of a K+ permeable ion channel shown in the figure.

   A. Starting with Fick's first law and using the numbers given below, calculate the following for the K+ channel shown in the figure:

      1. The flux through the channel in ions/second
      2. The single channel current
      3. The single channel conductance, assuming that the channel is selective for potassium

   B. What other factors might need to be included in the above calculation to more accurately describe flux through an ion channel?

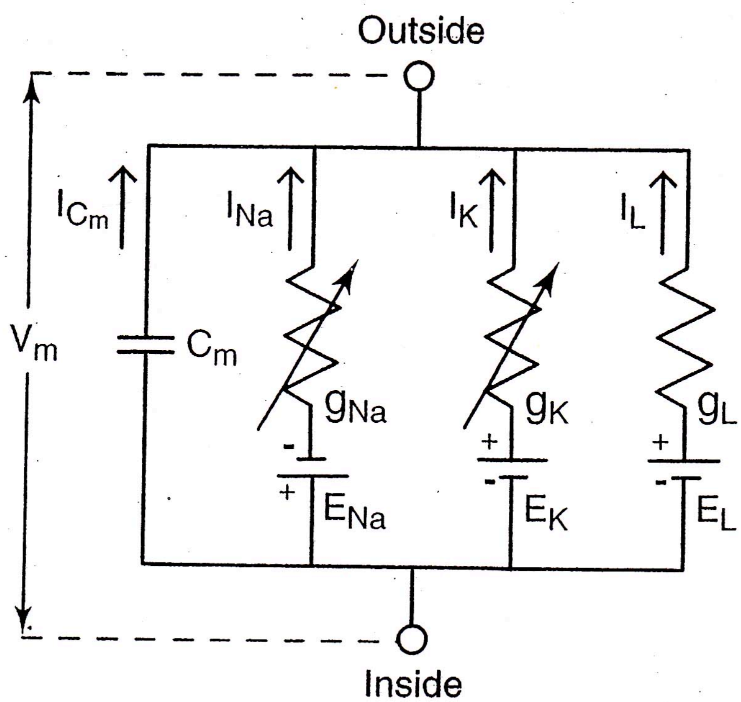


Numbers needed for calculation (numbers in figure are also needed):

$D_{K+} = 1.4 \times 10^{-5}$ cm^2/sec

(RT)/(ZF) = 25 mV

2. The figure below shows the equivalent circuit representation of a giant axon (as used in the Hodgkin-Huxley picture).



First we will consider the axon at rest, so that the three conductances are in the ratio:

$$G_K : G_{Cl} : G_{Na} = 1 : 0.1 : 0.03$$

** assume gL=gCl; EL=ECl

We have not included the Na-K ATPase pump in the figure, but we will include it in a later problem. The arrows represent the voltage dependence of the conductances, which will also be used in a later question.

(a) Derive a formula for the membrane resting potential in terms of the conductances, G_k, G_{Na}, G_{Cl} and the reversal potentials, E_k, E_{Na}, E_{Cl}. That is, state the condition of membrane current for which the steady-state resting potential prevails.

(b) Under steady-state conditions, the formula you have derived (or looked up in the notes) gives the membrane potential in terms of concentrations and conductances. We will now consider an axon of a marine invertebrate, such as the squid. At rest, the only permeable ions are Na^+, K^+, and Cl^- The major internal and external ion concentrations are :

$$[K]_o = 20 \text{ mM}; \quad [Na]_o = 490 \text{ mM}; \quad [Cl]_o = 500 \text{ mM};$$

$$[K]_i = 400 \text{ mM}; \quad [Na]_i = 50 \text{ mM}; \quad [Cl]_i = 32.8 \text{ mM}$$

Assuming the three pathways are perfectly selective, calculate the reversal potentials, E_k, E_{Na}, E_{Cl}. You may use the value $kT/e = 25.9$ mV at T =300 °K (23° C), normal room temperature.

(c) Calculate the membrane potential at room temperature.

(d) In reality, this creature lives at the chilly ocean temperature of 6.3 °C. Recalculate the membrane potential for this more natural condition.

(e) At the peak of the action potential, there is a 45-fold increase in the sodium conductance with no change in potassium or chloride conductance. What potential is reached at peak activity? How does this compare with the sodium Nernst potential for the given concentrations? What is the amplitude of the action potential (from rest)?

3. Now we wish to correct the membrane potential for the presence of a Na-K ATPase pump. This equivalent circuit has two parallel constant-current sources, $I_{P\text{-}Na}$ and $I_{P\text{-}K}$. This is an electrogenic pump defined by

$$(1)\ I_{P\text{-}Na} = -\ 3/2\ I_{P\text{-}K}.$$

The three leak currents are given by:

$$(2) I_{L\text{-}K} = G_K\ (V\text{-}E_K),\quad (3) I_{L\text{-}Na} = G_{Na}\ (V\text{-}E_{Na}),\ (4) I_{L\text{-}Cl} = G_{Cl}\ (V\text{-}E_{Cl}).$$

(a) State the conditions for the membrane to be in ionic equilibrium in the presence of the pump. In question 2, the total current was set equal to zero. Now the individual ionic species must be in equilibrium too. How do these conditions change the three reversal potentials? Are they still equal to the Nernst potentials for the individual ions? (hint: the answer is not the same for all of the ions).

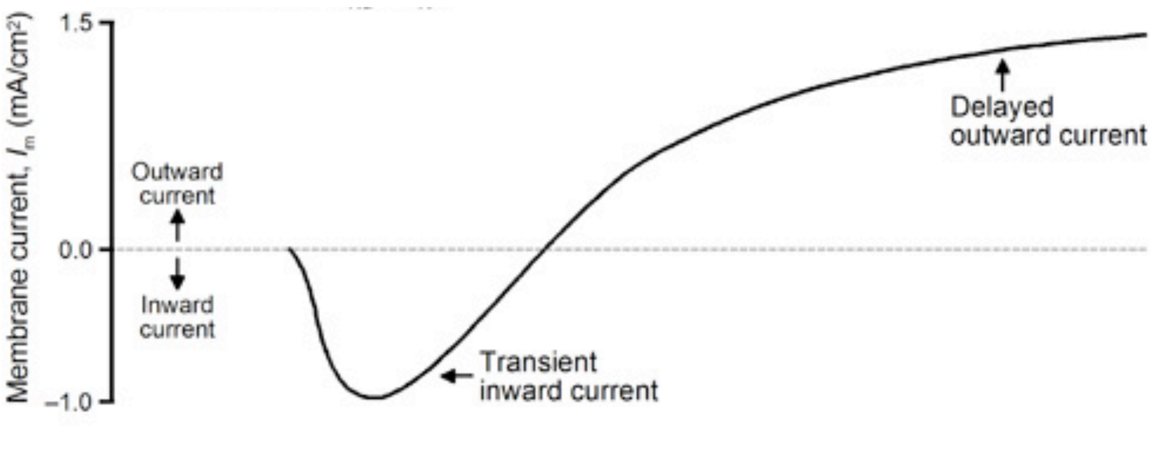
(b) Derive a formula for the membrane potential corrected for the pump. The derivation follows what was done in class notes for the constant-field model, but now we use the linear current-voltage relations of Eqns. 2 – 4 (as in PSet # 2, Problem #3). The result is surprisingly simple.

(c) Calculate the resting potential at room temperature when the pump is operative. Does the pump depolarize or hyperpolarize the membrane potential? Explain your answer.

(d) In the worked example in Pset 2, we calculated the membrane potential (with pump on) but we ignored the Cl^- current. Recalculate V_{rest} without the Cl^- terms (but with the pump on). The answer may surprise you. Explain why despite the relatively high permeability to Cl^- ions, ignoring the Cl^- terms has so little effect.

4. Voltage Clamp.

Under voltage-clamp conditions, the membrane is depolarized from rest by a 60 mV step of potential of 5 msec duration (60 mV positive to the resting potential). The figure shows the ionic current that flows in response to this pulse.



(a) Sketch the individual Na and K components of the current, and describe a way by which the current can be separated into these two components. (There is more than one way.)

(b) Sketch the two components of time-dependent conductance, $g_{Na}(t)$ and $g_K(t)$, corresponding to the currents, and explain how the conductances are determined experimentally.

(c) Explain in a short paragraph how the time-dependent conductances observed experimentally lead to the Hodgkin-Huxley gating variables m, h, and n.

(d) Why are the gating variables m and n raised to powers in the Hodgkin-Huxley description? Explain why the on response to a depolarizing pulse exhibits a time delay and the off transient does not. You can explain things in terms of the concerted action of gating subunits in opening a channel.

(5) Action potentials and propagation

(a) What happens during an action potential to make the membrane potential increase regeneratively. You may describe this process in terms of a cycle of events, but also explain the regenerative cycle in a few short sentences.

(b) Describe the sequence of steps by which an action potential propagates without loss. Draw a diagram of an axon indicating the site where the peak action-potential currents flow across the membrane. Show the lines of current flow to unexcited parts of the cable. Explain how these local currents cause the action potential to propagate.

(c) Sketch the Na^+ and K^+ currents that flow during an action potential. (Hint: Look up how g_{Na} and g_K vary with time during the action potential -- given in the class notes -- and then recall that $I_{Na} = g_{Na}(V - V_{Na})$, $I_K = g_K(V-V_K)$, and that the voltage V(t) is varying according to the action-potential shape. You can do this qualitatively, but justify your answer. A common mistake is to confuse the currents that flow during the action potential with the voltage-clamp currents that flow in response to a depolarizing pulse.

(d) Explain the following action-potential phenomena in terms of the Hodgkin-Huxley variables, m, h and n. Write the relevant variable for each of the phenomenon listed below, and explain your choice in a sentence or two.

(i) Sharp rise in voltage in response to a short-duration stimulus of 30 mV above rest.

(ii) Initial decline in voltage after the action potential reaches its peak.

(iii) Hyperpolarizing voltage for a few msec after the action potential has subsided

(iv) Anode-break effect in which a hyperpolarizing conditioning pulse prior to application of the stimulus causes the threshold to get lower.

(6) Gated Ion Channels

Gated ion channels are membrane proteins capable of undergoing rapid conformation changes between conducting and nonconducting states. Consider a simple channel in which the gating is effected by a single charged group that moves during the conformation change. When the channels are in steady state under an applied transmembrane electric field, the energy difference between the open and closed state is given by

$$\Delta W(V) = -Q_g (V - V_0),$$

where Q_g is the effective charge that moves across the membrane during the gating transition, and $\Delta W_{conf} = Q_g V_0$ represents some intrinsic voltage-independent energy difference between the two conformations.

(a) Assume that the open and closed states distributed according to a Boltzmann distribution,

$$\underline{N}_{open} / \underline{N}_{closed} = \exp(-\Delta W/kT)$$

Derive an expression for the steady state voltage dependent conductance for a collection of N gated channels ($N = N_{open} + N_{closed}$) having a single-channel conductance, γ .

(b) Sketch the curve of steady-state voltage-dependent conductance, $G_{ss}(V)$. Indicate the maximum value of $G_{ss}(V)$, and the voltage V_0. What fraction of the channels is open at V_0 ?

(c) Say the channel is Na^+ selective, so that the current voltage relation of an open channel is

$$I = \gamma (V - V_{Na}).$$

(Note: For this part of the problem, you can assume $\gamma = G_{ss}(V)$.)

Assume $V_0 = 20$ mV, $kT/Q_g = 5$ mV, $[Na]_I = 50$ mM , $[Na]_0 = 500$mM. Sketch the steady-state current-voltage curve. Indicate the reversal potential and any region of negative conductance. (ie. $dI/dV < 0$).

(d) Briefly state how any region of negative conductance for the current voltage curve of an Na + channel is significant for generating an action potential.