MCB 166 Problem Set #4

I. Hodgkin-Huxley axon. Conditions for a potassium negative conductance.

The HH equations for a space-clamped squid giant axon are given below. Tetrodotoxin (TTX), the puffer-fish poison is known to block axonal Na channels. When TTX is introduced into the bathing medium, the axon I_{Na} will be blocked and I_K and I_L will remain unchanged. Ordinarily, the nerve would be incapable of generating action potentials. We wish to consider an axon bathed in TTX for two different conditions of external K concentration—normal $[K]_o$, for which $V_K = -75$ mV, and high $[K]_o$, for which $V_K = 0$.

The conductance, $\bar{g}_K$ might also vary with changed external K, but for the sake of simplicity, we will assume $\bar{g}_K$ does not change.

(a) In a voltage clamp experiment, the membrane potential is held for a long time at a potential V = -100 mV, and then a depolarizing step of 60 mV to V=-40 mV is applied for 30 msec. By integrating the equation for n(t) for the appropriate boundary conditions, solve for the time course of $\bar{g}_K$. Show mathematically that the rising phase of the conductance has an inflection point but that the falling phase does not. This delayed onset but undelayed fall is what Hodgkin and Huxley originally required for the time-dependent conductances. You may assume $n_\infty(-100) = 0$, $\tau_n(-100) = 5$ msec throughout.

(b) Using the plot of part (a) sketch the current I_K for this voltage pulse for both conditions of $[K]_o$ (ie. $V_K = -75$ and $V_K = 0$). Compare the currents during rising and falling phases of conductance for the two conditions. Is I_K outward for both conditions? Explain. The current transient at the termination of the pulse is called the tail current.

(c) For the two conditions of external K concentration, plot the steady-state current-voltage relations. Which condition gives a negative conductance? Explain how a negative conductance can come about in the absence of Na ions. (Assume that V_L and $\bar{g}_L$ are not altered by the ionic changes).

(d) Can the system in high $[K]_o$ give an action potential in response to a short shock of current? If not, can you think of a way of producing a potassium action potential with this system? What would an imposed steady current do?

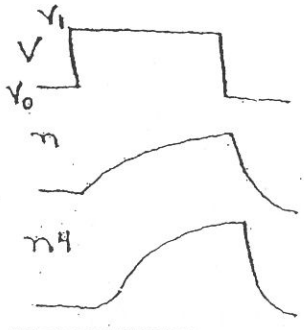
Recall $I = C dV/dt + g_L(V - V_L) + g_K n^4(V - V_K) + g_{Na} m^3(V - V_{Na})$

$dn/dt = (n_\infty(V) - n)/\tau_n(V)$, etc.

$V_L = -60$ mV, $\bar{g}_L = 10$ mS/cm^2 *

$V_K = -75$ mV, $\bar{g}_K = 36$ mS/cm^2

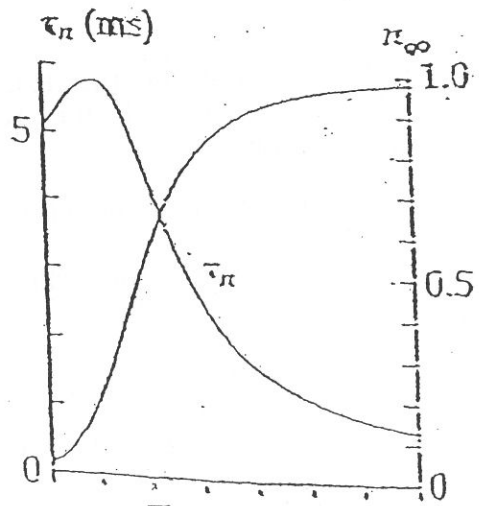
$V_{Na} = +55$ mV, $\bar{g}_{Na} = 120$ mS/cm^2



$$I_K = \bar{g}_K n^4(t)(V - V_K)$$

$$I_{ss} = \bar{g}_K n_\infty^4(V)(V - V_K)$$

$$n(t) = n_\infty(V_0)e^{-t/\tau_n(V_1)} + n_\infty(V_1)(1 - e^{-t/\tau_n(V_1)})$$



* $\bar{g}_L$ artificially large to make phenomenon more visible.

(JW prob. 6.5)

2. An alga *Chara globularis* is known to generate positive-going action potentials. The major ions in both its cytoplasma and the pond water it lives in are Na^+, K^+, and Cl^-, and their concentrations are as follows:

	Cytoplasma (mM)	Pond water (mM)
Na^+	57	0.031
K^+	65	0.046
Cl^-	112	0.040

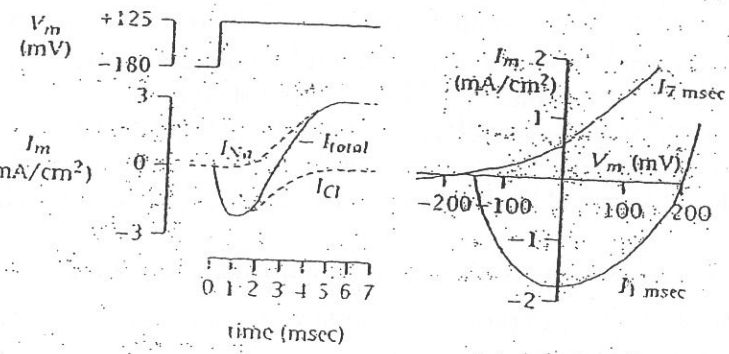
The resting potential of the cell is −182 mV, and the peak amplitude of the action potential is +198 mV.

(a) What is (are) the primary permeable ion(s) for this cell at rest?

(b) What is (are) the primary permeable ion(s) for this cell during

(c) In a pump that pumps both Na^+ and Cl^- into the cell at a ratio of 1 : 1, what is the contribution of this pump to the resting potential of the cell?

(d) The voltage-clamp analysis of this cell reveals the following results:



What are the values of g_{Cl} and g_{Na} at $V_m = -50$ mV and at $V_m = +150$ mV?

3. Which of the Hodgkin-Huxley variables, m,h, or n is primarily responsible each of the following phenomena: Explain your choice in one short sentence

(i) Sharp threshold for excitation (for short-duration stimuli).

(ii) Rapid repolarization at the end of an action potential.

(iii) Undershoot after an action potential, ie. voltage is temporarily hyperpolarized beyond rest.

(iv) Anode-break excitation, ie. membrane has lowered threshold after hyperpolarizing prepulse.

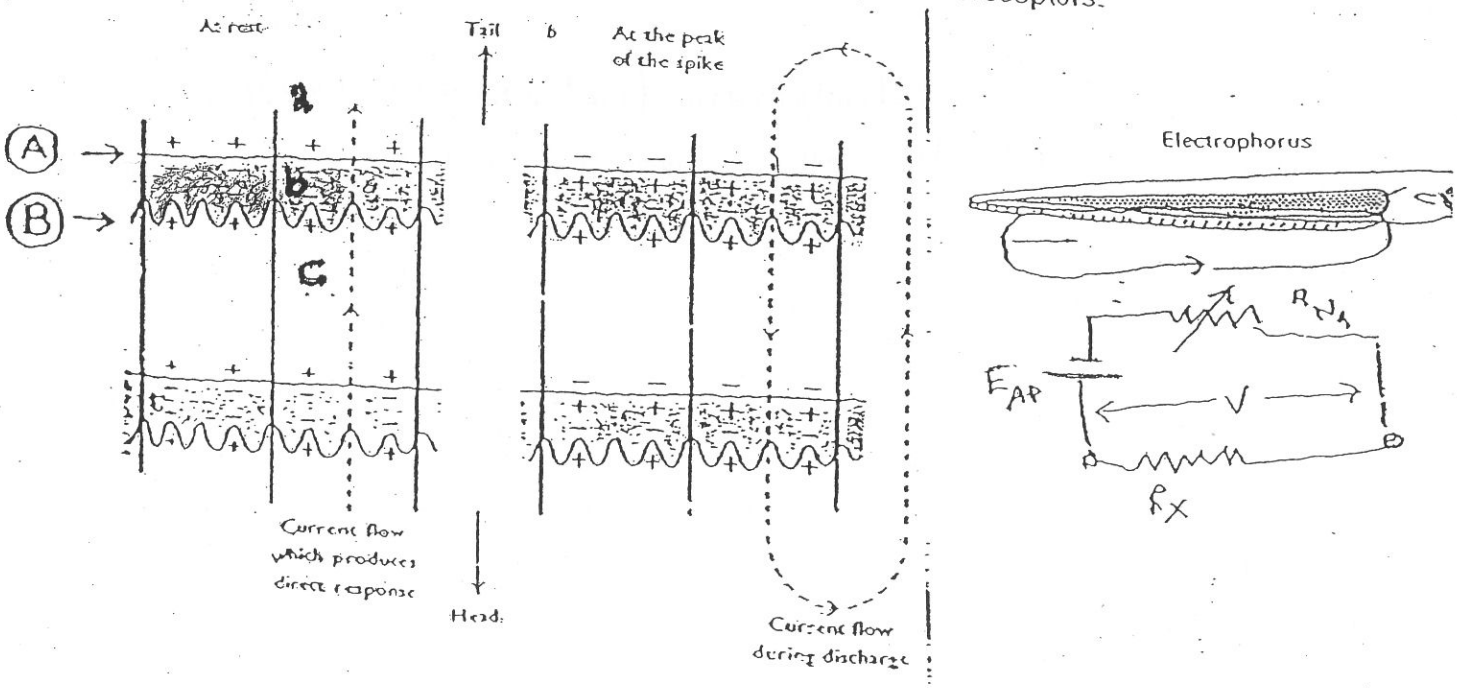
4. The electric organ of the electric eel can generate a 600 volt discharge. It is made up of stacks of about 4000 asymmetric disc-shaped cells called electroplaques. The entire stack is surrounded by an insulating sheath, as shown in Fig. 1. Each elecroplaque has has two different faces -- A and B in the figure. At rest, both faces are permeable to K^+ and have relatively high resistance. At the peak of the action potential, face A has many activated Na^+ channels, and thus has a low resistance and is permeable to Na^+. Face B is transiently activated by postsynaptic (acetylcholine) channels to trigger the action potential in face A. The acteylcholine channel is equally permeable to Na^+ and K^+ and has a reversal potential near zero. During activity this face also shows a low resistance.

(a) Draw an equivalent circuit for the cell at test and at peak activity representing the two membranes in series and Using V_{Na} = +50 mV (inside relative to outside) and V rest = -90 mV (inside relative to outside). Calculate the resting potential and the action potential peak amplitude measured between microelectrodes placed at points a and b, and then between electrodes placed at a and c. [Amplitude of AP $\equiv V_{AP} - V_{REST}$]

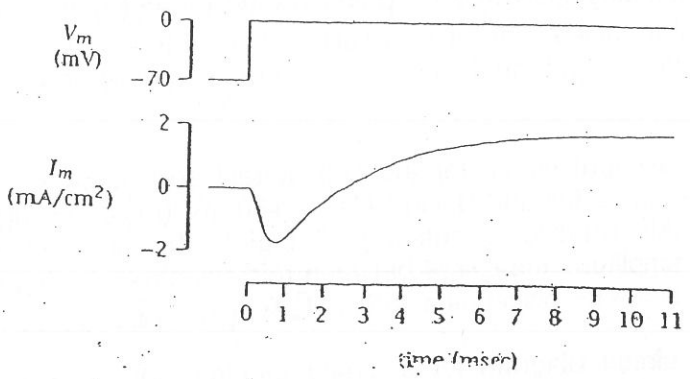
(b) Using the circuit diagram for a single electric cell, draw and label the equivalent circuit for the entire stack of 4000 cells. Calculate the voltage drop across the stack at rest and at the action potential peak. Explain why the electric organ can generate such high voltages (sufficient to light up neon lights in aquariums). Why is it unlikely that a single excitable cell can be designed to generate voltages much higher than a few hundred millivolts?

(c) Complete the equivalent circuit for an electric organ composed of 4000 electroplaques at peak discharge, by sending current through an external resistance, R_x, the resistance of the medium through which the fish swims. Write an expression for the voltage the electric fish can utilize to stun its prey. There are both marine and fresh-water electric fish. Explain why the fresh-water types are more effective at stunning their prey. In fact, marine electric fish have only the excitable synaptic face with no electrically excitable face.

The evolution of these electric organs by natural selection puzzled Darwin. He couldn't understand how the strongly discharging fish evolved from ancestors whose discharge was too weak to be used as a weapon. It is now thought that the weak discharges were used as part of a direction finding system employing electroreceptors.



5. When a normal, healthy squid axon is voltage-clamped in artificial sea water, one obtains the following membrane current record in response to a step change in membrane potential from $V_m = -70$ mV to $V_m = 0$ mV.



Draw similar plots of I_m vs. t (when V_m is stepped from −70 mV to 0 mV) when the recordings are made under each of the following experimental conditions. For each of your plots, explain in one or two sentences how and why your graph differs from that drawn above.

(a) TTX is added to the bath surrounding the axon.

(b) TEA is added to the interior of the axon.

(c) $[Na^+]_{out}$ is adjusted so that $[Na^+]_{out} = [Na^+]_{in}$.

(d) $[K^+]_{out}$ is adjusted so that $[K^+]_{out} = [K^+]_{in}$.

(e) Ouabain, a specific inhibitor of the Na^+-K^+ pump, is added to the bath five minutes before the experiment.

TTX = tetrodotoxin (Na^+-channel blocker)

TEA = tetra ethyl ammonium (K^+-channel blocker)