

Note that the specific order of TRUE/FALSE questions may be different in your exam, so be sure to read through each statement carefully.

1. (2 marks) Transport of various substances across the cell membrane is achieved using a number of different mechanisms. Determine whether each of the following statements is TRUE or FALSE. Please circle the correct answer.

- (a)  TRUE or  FALSE | A distinguishing feature of primary active transport is the requirement of ATP.
- (b) TRUE or  FALSE | Facilitated diffusion can involve transport of a substance against its concentration gradient.
- (c)  TRUE or  FALSE | The direction of transport through membrane ion channels depends on the concentration gradient of the ion involved.
- (d)  TRUE or  FALSE | Some types of voltage-gated ion channels can be open or closed at that same level of membrane depolarization.

2. (4 marks) You have just succeeded in recording membrane potential from a previously unidentified neuron in a pond snail. You find that the resting potential is -65mV but that once every few seconds an action potential (AP) is produced. As a first step towards characterizing this neural activity, you consider what you have learned about the control of membrane potential and classically-described APs, and come up with some predictions. Determine whether each of the following statements is TRUE or FALSE. Please circle the correct answer.

- (a) TRUE or  FALSE | If the depolarization phase of the AP involves voltage-gated  $\text{Na}^+$  channels (as in the squid axon), then decreasing extracellular  $\text{Na}^+$  concentration will increase the AP amplitude (i.e. peak voltage during AP).
- (b) TRUE or  FALSE | If the after-hyperpolarization phase (AHP) involves voltage-gated  $\text{K}^+$  channels (as in the squid axon), then decreasing extracellular  $\text{K}^+$  concentration will result in a smaller AHP (i.e. less hyperpolarization).
- (c)  TRUE or  FALSE | If  $\text{Cl}^-$  channels contribute to the resting potential and the  $\text{Cl}^-$  equilibrium potential (Nernst potential) is -30mV, then opening more of these channels could lead to the production of an AP.
- (d)  TRUE or  FALSE | If the after-hyperpolarization phase (AHP) involves voltage-gated  $\text{K}^+$  channels (as in the squid axon), then a drug that delays the closing of these channels will increase the duration of the relative refractory period.

3. (a) (2 marks) The  $\text{Na}^+/\text{K}^+$  ATPase is considered to be an “electrogenic” ion pump. Explain why in one sentence.

*“electrogenic” means that there is a net charge exchange. In this case, the  $\text{Na}^+/\text{K}^+$  ATPase pumps 3  $\text{Na}^+$  out for every 2  $\text{K}^+$  in, resulting in a net loss of positive charge from inside the cell.*

- (b) (2 marks) Ouabain is a drug that blocks the  $\text{Na}^+/\text{K}^+$  ATPase. Assume that for a particular neuron the  $\text{Na}^+/\text{K}^+$  ATPase is always active. Based on the electrogenic properties of this pump, how would this neuron’s membrane potential change after application of ouabain? Briefly explain (in one or two sentences).

*In this case, the “ $\text{Na}^+/\text{K}^+$  ATPase is always active” and it is electrogenic (see above), so that it tends to hyperpolarize the neuron (loss of positive charge). Thus, blocking this action results in a depolarization of the neuron (i.e. it “removes” hyperpolarization)*

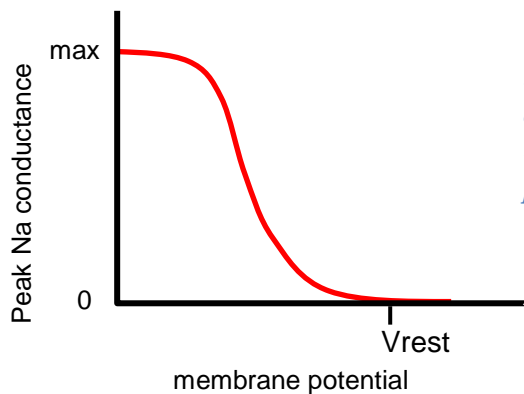
4. (4 marks) While recording from your favorite neuron, you observe brief hyperpolarization events of about 5mV in amplitude. The events last about 100ms and occur about once every second. The resting potential of this neuron is -60mV. You hypothesize that  $\text{Cl}^-$  channels underlie these events, but you know of no specific drugs that block these channels. In the space below, describe **an experimental approach** that will allow you to test your hypothesis. Be sure to explain the predicted outcome(s) of your experiment.

*Under these constraints, the best approach (one with fewest caveats/assumptions), involves manipulating membrane potential ( $V_m$ ). But now you must make an assumption about  $E_{\text{Cl}}$ . If you assume  $E_{\text{Cl}}$  is above  $V_{\text{rest}}$  (i.e. -60mV is this neuron), then the hyperpolarizing events must be due to closing  $\text{Cl}^-$  channels. If you assume  $E_{\text{Cl}}$  is below  $V_{\text{rest}}$ , then the hyperpolarizing events must be due to opening  $\text{Cl}^-$  channels. Now you change  $V_m$  so that it is above  $E_{\text{Cl}}$ , equal to  $E_{\text{Cl}}$  and below  $E_{\text{Cl}}$ . The predicted outcomes of this experiment will depend on whether the events involve opening or closing channels – remember that if you open  $\text{Cl}^-$  channels,  $V_m$  will go towards  $E_{\text{Cl}}$ .*

*It is also possible to manipulate  $E_{\text{Cl}}$  but this involves more assumptions, the most important of which is that  $\text{Cl}^-$  channels do not influence the resting potential. If this is not indicated, the possibility of confounding factors must be considered (due to changing multiple parameters at the same time), and thus predicted outcomes are not as clear.*

5. A colleague tells you about a neuron with an interesting mutation in the voltage-gated Na<sup>+</sup> channel. You notice that the mutation involves several residues in the TM4 domain of the pore-forming region of the  $\alpha$ -subunit. After some reflection, you realize these changes will likely reverse the voltage dependence of this channel. In other words, you think the channel will now be activated by hyperpolarization, rather than depolarization. You also suspect that the time-dependence of activation will be unaffected by the mutation and that the channel is completely de-activated at the resting potential.

(a) (3 marks) Draw a graph showing the expected conductance (activation) for this channel as a function of membrane potential (label carefully). Briefly describe (in the space below) how you could measure this relationship experimentally.



*V<sub>m</sub> is the independent variable. Assume you can isolate the Na<sup>+</sup> current (e.g. block all other channels). Set V<sub>m</sub> to different levels and measure Na<sup>+</sup> current (or conductance). For the graph, you could choose to plot peak or steady state values of current/conductance.*

(b) (3 marks) Propose one possible role this mutant channel might play in the control of membrane potential. Explain your answer in the space below.

*There are many different ways to answer this question. For full marks, we were looking for a clearly presented, logical and functional link to the properties described above. If any assumptions were necessary, they should be clearly indicated.*

*One possible role is to control the level of membrane hyperpolarization. In other words, for any hyperpolarizing stimulus, these channels would open and thus oppose (to some extent) the hyperpolarization. With normal voltage-dependence of inactivation, hyperpolarization would only decrease inactivation levels and thus this effect could be sustained. However, if you assumed that inactivation was also affected by the mutation, such that it increases with hyperpolarization, then the effect would be very brief.*

*Another possibility could involve action potential production in response to a hyperpolarizing stimulus. In this case, you must assume that there is also a population of “normal” Na<sup>+</sup> channels (or some other voltage-gated channel) to support AP production. Then, given a hyperpolarization, the mutant Na<sup>+</sup> channels would open, lead to a depolarization. If deactivation was fast then the depolarization would simply result in a return to V<sub>rest</sub>. However, if you assumed deactivation was slow, then in principle the depolarization could overshoot V<sub>rest</sub> and possibly result in an AP.*