Bi150 Problem Set 2

Due: Tuesday, October 18th 2011 at 4:30 P.M.
At the "Bi 150 Box"
3rd floor of Kerckhoff in front of Room 326

(The building may be locked after 5 P.M.)

INSTRUCTIONS

Please:
1) Turn in your work with this cover page.
2) Use separate sheets of paper for the answer to each question, so that grading can proceed in parallel
3) Write or type your answers neatly.
4) Put your name on each page of your answers.

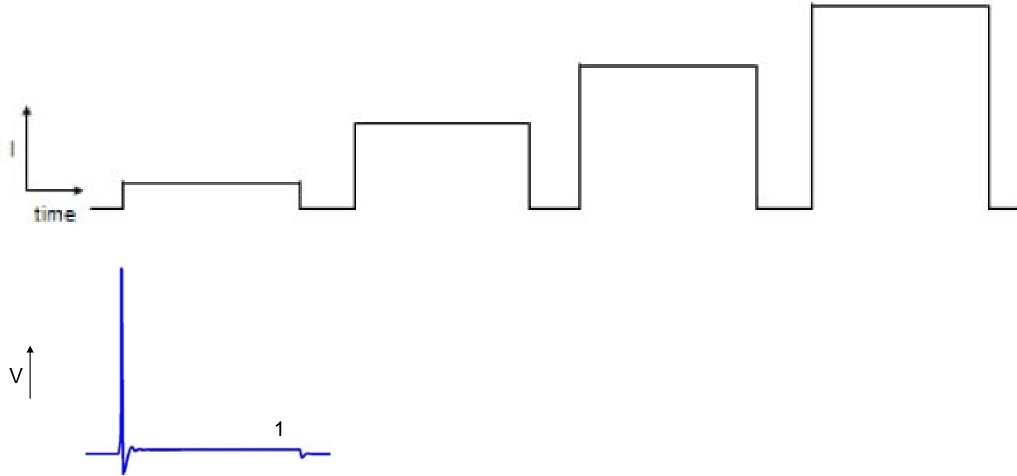
Name: ____
Section #: ____
Mail Code: ____
TA Name: ____
Date and Time turned in: _______________________________________
Number of pages including this one:_______________________________

There are 3 questions.
Grade and Comments:
1____________________
2____________________
3____________________

Total: _______________

Problem 1. Action potentials (1.5 points)

A. (0.5 point) Generation



a. In a typical neuron's cell body, the experimenter gives a series of current pulses (top trace). The smallest pulse gives an axon potential, as indicated (bottom trace). Draw the remaining part of the bottom trace, showing the qualitative and approximate trend in number and shape of action potentials.

b. The generation of action potentials is a regenerative or positive-feedback process. Explain the mechanism of positive feedback during action potential generation.

B. (1.0 point) Propagation

This question deals with the influence of an axon's diameter on its basic properties.

Use the simulation program on the website:
http://nerve.bsd.uchicago.edu/nerve1.html
(You may need to restart your browser occasionally)

Use the "V vs x" option. Click on "axon parameters" and make "Max gNa" and "Max gK" zeros, to examine passive properties of membrane.

Open "Stimulus" and make the duration of current injection long enough (e.g. 20 msec). You may also need to change "amplitude" of the current pulse and "Total Time" of simulation accordingly.

Start the simulation, then you will see spatial voltage change in the axon induced by tonic current injection.

a. Describe the change in the spatial decay of membrane potentials:
When you increase or decrease “Radius” in “axon parameters”.
When you increase or decrease “IntR” in “axon parameters”.

b. (Without simulation) When the axon’s diameter is doubled (length is infinite), how much will the g_m (membrane conductance per length) change? How much do c_m (membrane capacitance per length. The axon is **not a coaxial cable), and r_a (axial resistance per length) change? What about the length constant? Explain.

c. (Use simulation) Use the “V vs t” option. Click on “reset parameters” often to ensure that you are using the default parameters.

Set the blue and red electrodes 4 cm apart; measure the conduction velocity between them.

Measure the conduction velocity when the diameter of the axon has changed (other parameters are same; you may need to adjust the stimulus current). Draw the graph of “diameter”(x-axis) vs. “velocity” with at least 5 data points over the range 10 μm to 1 mm. What is the best power law for this graph?

d. (Without simulation) Among vertebrates, axon diameters (10-20 μm, maximum) are much less than that of a squid axon. However, some vertebrate axons still have a high propagation velocity, up to ~30 m/sec. Describe a mechanism that vertebrate axons utilize to achieve such high speed. Diagram the current flow in axons with and without this mechanism, and explain how this mechanism increases action potential velocity. What is the effect on energy cost?

e. (Use simulation) Some inherited human disorders, such as epilepsy and cardiac arrhythmia, are caused by defects in sodium channel inactivation. Sodium channel inactivation becomes slower or incomplete. You can mimic this by using the “concentrations” control to add pronase to the intracellular solution (100 μg/ml). This proteolyzes the inactivation flap. Draw and label the waveform of a “normal” and “mutant” AP. Explain the effect that you observe.

Problem 2. Synaptic Transmission (1.5 points)

A. (0.3 point) Write the following events in the correct sequence as they occur at the neuromuscular junction of the California olive tree rat (*Rattus pasadenesis*) during normal synaptic transmission.

Some events overlap others. Write "overlap" if you think an event overlaps with the events immediately preceding and following on the list. *start here.

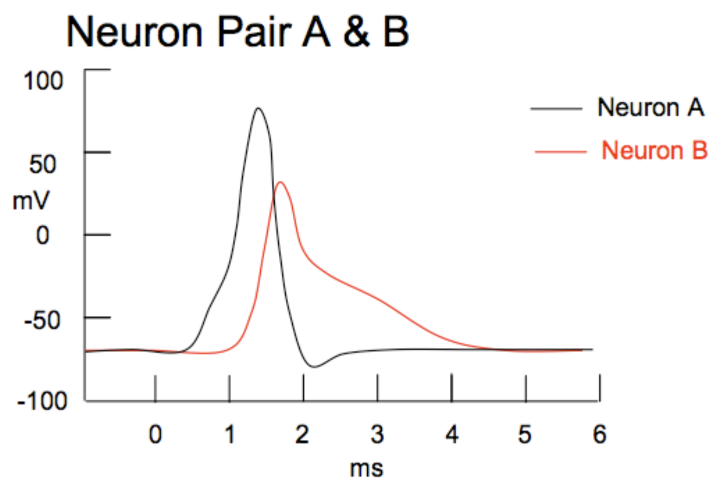
1) Action potential arrives at the presynaptic terminal.
2) Muscle fiber action potential.
3) Na^+ enters, and K^+ exits, through the same channels.
4) There is a local increase in intracellular Ca^{2+} concentration near the Ca^{2+} channels.
5) ACh is hydrolyzed to acetate plus choline.
6) Choline is transported into the presynaptic terminal.
7) ACh molecules fill the synaptic vesicles
8) ACh is released into the synaptic cleft.
*9) The synaptic vesicles become acidic.
10) Na^+ is pumped out and K^+ is pumped into the presynaptic terminal and muscle fiber.
11) ACh binds to ACh-gated ion channels on the postsynaptic membrane.
12) ACh diffuses within the synaptic cleft.
13) Muscle fiber reaches threshold depolarization.
14) ACh-gated ion channels open.
15) ACh-filled vesicles fuse with the presynaptic membrane.
16) Ca^{2+} enters the presynaptic terminal through voltage gated Ca^{2+} channels.

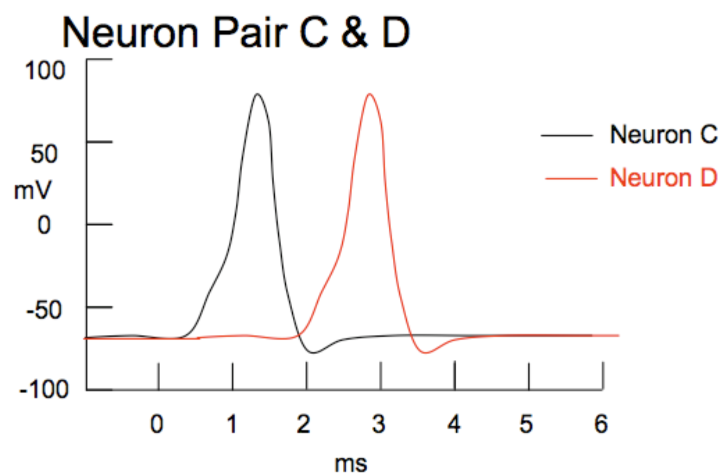
B. (0.6 point) Describe how the events above would differ for a spine synapse, for instance in the hippocampus of *R. pasadenesis.* Name four differences.

C. (0.6 point) You are in lab recording directly from the cell bodies of two different neuron pairs (A&B; C&D) in the hippocampus of *R. pasadenesis*. Neuron A forms a synapse on neuron B, and neuron C forms a synapse on neuron D. You induce a stimulus in neurons A and C. The traces you record are shown in the diagram.

a. State the main difference between these two synapses and among the complement of channels in the cells. What evidence do you have to support your conclusion?

b. Describe some other important differences between these two types of synapses.





Problem 3. Transmitter Release (1.5 points)

A. (0.5 point) Briefly describe the events that are involved in neurotransmitter vesicle docking, fusion and recycling. In your answer, please mention the roles of the following proteins: synaptotagmin, VAMP, syntaxin, SNAP-25, NSF, SNAP and Rab proteins.

B. (1.0 point) You are in lab recording from neuron pairs, each connected by a glutamatergic synapse, in four different mutant animals and in a wild type animal. In each animal a different protein involved in neurotransmitter release is mutated.

The mutations are:

a. Mutation of the voltage-gated Ca^{2+} channels of the presynaptic terminal. The mutant channels have a lower opening threshold.

b. Mutation of the Ca^{2+} binding sites of synaptotagmin: the mutant does not bind Ca^{2+}.

c. Mutation of plasma membrane glutamate transporter to inhibit its function. Please note that several redundant processes maintain intracellular [glutamate] at ~ 10 mM, vs CSF concentrations of ~ 1 μM.

d. Knockout of NSF

You stimulate the presynaptic cell and record from the postsynaptic neuron of each pair.

For each mutation above, please describe the following:

i. At the first stimulus, how does the EPSP you record from the postsynaptic neuron differ from the EPSP recorded from a wild type animal? Does it increase, decrease, stay comparatively the same, or is there no EPSP generated at all?

ii. After five stimulations of the presynaptic cell in quick succession, how does the EPSP you record from the mutant animal differ from that in the same experiment on a wild type animal?

Explain your reasoning in each case.