# BiBB492: Experimental methods in synaptic physiology

Instructor: Mike Kaplan (mkap@sas.upenn.edu)
Prerequisites: BBB109 or BBB251
Readings: Primary literature (see below).
Supplementary text (optional): From Neuron to Brain (Nicholls, Martin, Wallace and Fuchs)

The immense computational power of nervous systems and their capacity to control behavior are a function of specialized electrical and chemical signaling properties of their elementary units, neurons and glia. In this seminar and lab course, we will use techniques of electrophysiology to directly observe the electrical and synaptic actions of small systems of neurons. Meeting once a week, a small group of students will discuss topics in synaptic physiology from the primary literature while becoming proficient at electrophysiological techniques combining intracellular and extracellular recordings. Focusing on the buccal ganglia of the snail *Heliosoma*, a structure of approximately 120 neurons, we will learn how small networks of neurons work together to produce a patterned output, and how this output can be altered by neuromodulators. Readings from the primary literature will introduce the study of neural networks and neuromodulation, using the buccal ganglia as a specific example. As a midterm assignment, students will prepare and present a short research proposal using this model system, to be evaluated by the class. For the remainder of the course, the class will work together in groups of two on several of these proposals, meeting at the end of each class to pool our data, analyze the results and discuss their significance.

*Requirements:* At the start of each class, we will compare and, when appropriate, pool our data from the previous week, followed by a discussion of the reading. Working in groups of two, each student will be responsible for presenting and leading the discussion of one paper. Students will also submit data from at least one cell in weeks 4-7, to contribute to a class "library" of identified neurons. Finally, students will spend the last part of the course (weeks 15-19) working on the research projects. A final report on the project will serve as a final exam, and will be due one week after the last class.

*Grading:* There will be one quiz in Week 4 and one midterm exam in Week 8 covering the readings and underlying concepts; together these will comprise 30% of the final grade. The presentation of a paper and profile of an identified neuron will each be worth 10%, and the final project will be 25%, with the remainder of the 25% grade reflecting attendance and performance in lab.

| *week* | *date* | |
|---|---|---|
| 1 | Jan 11 | *Lab:* Introduction to equipment; model cells; ground wires; electrodes<br>*Reading (for next time):* Kandel, *Cellular Basis of Behavior*, pg. 36-72. (Some historical perspective on how and why gastropods are valuable for neuroscience.) |
| 2 | Jan 18 | *Analysis and Discussion:* review basic concepts (resting potential, action potential, synaptic transmission) and discuss paper.<br>*Lab:* Intracellular recording I. Using glass electrodes to get a detailed view of individual neurons, focusing on how to get a stable recording and what to do with it if you've got one (input resistance, time constant, data logging).<br>*Reading:* Murphy (2001). *Progress in Neurobio.*. 63: 382-408. (1st part, pp. 382-391: the three-phase feeding cycle in *Helisoma*, and the neural network in the buccal ganglia that mediates it.) |
| 3 | Jan 25 | *Analysis and Discussion:* review neuromodulation, discuss paper.<br>*Lab:* Intracellular recording II. Continue to learn the technique of intracellular recording, and begin to think about how to identify the phase and identity of buccal neurons.<br>*Reading:* Murphy (2001). *Progress in Neurobio.*. 63: 382-408. (2nd. part, 391-396. Transmitters and interneurons of the three phases of the "normal" buccal rhythm.) |

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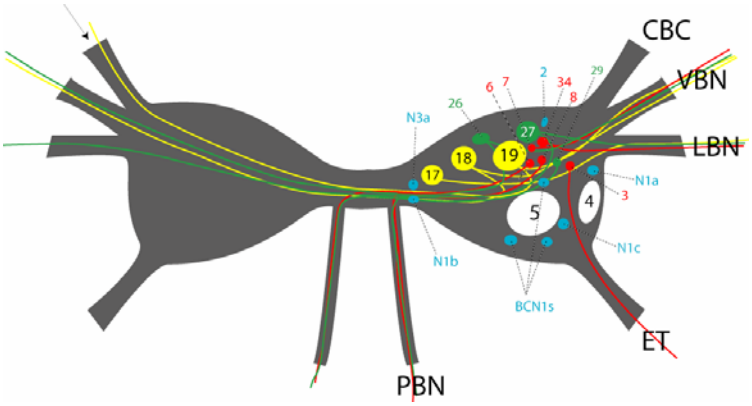
| 4 | Feb 1 | *Analysis and Discussion*: review synaptic plasticity, discuss paper.<br>*Lab:* Intracellular recording (III). Continue to flex your intracellular prowess, while thinking about how to identify individual cells and hit them repeatedly. Apply neurotransmitters or antagonists to help narrow down the possibilities.<br>*Reading:* Nusbaum and Marder (1987). *J. Neuroscience.* 9(5):1591-1607. (An example of how identified neurons get to be identified in the first place.) |
| 5 | Feb 8 | *Analysis and Discussion:* Show results from previous week, discuss paper<br>*Lab:* extracellular recording (I). Learn to make and wield an extracellular electrode to record from or stimulate a nerve.<br>*Reading:* Quinlan and Murphy (1991). *J. Neurophys.* 66:1264-1271. |
| 6 | Feb 15 | *Analysis and Discussion:* show results from previous week, discuss paper<br>*Lab:* simultaneous intracellular and extracellular recording (I). Extracellular recordings to place the intracellular recording in the context of the buccal rhythm.<br>*Reading:* Quinlan, Gregory, Murphy (1995) *J.Neurophys.* 73:945-956. (Neuron B2, the interneuron mediating phase 2). |
| 7 | Feb 23 | *Analysis and Discussion:* show previous week's recordings and fills, discuss paper<br>*Lab:* intracellular and extracellular recording (I!). Try to target same neuron is in Week 6 and again fill it with fluourescent dye. Add a stimulating electrode to activate fibers in incoming nerve trunks.<br>*Reading:* Achee and Zoran (1997). *J. Exp. Biol.* 200:1537-1548. (How one neuromodulator can have different effects on two cells, due to different signal transduction mechanisms.) |
| 8 | March 1 | *Analysis and Discussion:* MIDTERM EXAM.<br>*Lab:* Work in groups of two to formulate research proposals.<br>*Reading:* Quinlan, Arnett, Murphy (1997). *J. Neurophys.* 78:812. |
| | SPRING BREAK | |
| 9 | March 15 | *Analysis and Discussion:* Discuss papers from weeks 7 and 8; present and discuss research proposals, and meet individually with instructor to get final approval.<br>*Lab:* continue recording with suction electrode and one recording electrode.<br>*Reading:* Quinlan and Murphy (1997) *J. Neurophys.* 75:561-574. |
| 10 | March 22 | *Analysis and Discussion:* show results from previous week, discuss paper.<br>*Lab:* Begin work on independent research proposal. |
| 11 | March 29 | *Analysis and Discussion:* Analysis on previous week's recordings.<br>*Lab:* Continue working on research proposal |
| 12 | April 5 | *Analysis and Discussion:* Analysis on previous week's recordings.<br>*Lab:* Continue working on research proposal |
| 13 | April 12 | *Analysis and Discussion:* Analysis on previous week's recordings.<br>*Lab:* Continue working on research proposal |
| 14 | April 19 | Present results of research projects. Written report due April 26th. |




*Heliosoma* buccal ganglia, located on the dorsal surface of the buccal mass. Neurons that can be easily identified based on size, position and electrophysiological profile are B17, B18, B19, B5, and B4.

OBJECTIVES and projects:

a. *Intracellular recording: weeks 2-4*
Learn to record from individual neurons, and strategies for identifying them.

b. *Extracellular recording: week 5*
Learn to make and use a suction electrode to record from or stimulate nerves

c. *Combined intra- and -extracellular recording*
Use the extracellular electrode to help place the intracellular recording in the context of the three-phase buccal rhythm

d. *Other possible projects:*

i. Intracellular recording from salivary gland cells
ii. Dye fills to trace the processes of a cell
iii. Neuromodulation 1: effects of exogenous neuromodulators
iv. Neuromodulation 2: buccal ganglia with and without the ring ganglia
v. Neuromodulation 3: stimulating incoming nerve trunks
vi. Recording from 2 neurons at once