NEUROBIOLOGY LABORATORY

MCB 160L

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Notice to Students:

Read Exercise 1 before coming to first class meeting!

COURSE ORGANIZATION

Introduction

This course has several objectives. One is to introduce you to a variety of techniques used by neurobiologists to gain an understanding of how nervous systems work. Another is to develop exercises that improve your understanding of key principles in neuroscience. A third is to give you some idea of the skills that must be developed to be able to do neurobiological research. A fourth is to give you some idea of what it is like to grapple with neurobiological problems and seek experimental evidence to test hypotheses of how things work. A fifth is to provide you with enough familiarity with some aspects of neurobiological research that you can engage in independent honors research in faculty laboratories. Lastly, we hope this course will help you decide whether you are suited for a career in neurobiological research.

Neurobiology is a very diverse field. The nervous system is studied by an incredible variety of techniques in electrophysiology, biochemistry, anatomy, cell biology, molecular biology, genetics, behavioral analysis, biophysics, psychophysics, theoretical modeling, etc. No one is conversant with all these techniques, even those who are experts at some of them. There is no time to even expose you to all of these techniques. Rather, this course will select a few examples, from electrophysiology, cell biology, psychophysics, computer modeling, molecular biology, genetics, and anatomy. Experiments will be done on invertebrates, simple vertebrates, and humans, and will cover molecular channel properties, neuronal cell properties, organ and system properties, and behavior. Some techniques, like neurochemistry and molecular neurobiology, have been omitted because the techniques are essentially identical to those used in other areas of biochemistry. They will be used, however, in the preparation of specimens for study, and their roles in the exercises will be explained.

Other areas, like ultrastructural anatomy and magnetic resonance imaging, have been omitted because the techniques are simply too difficult to even develop a feel for in a few weeks, or because the equipment is prohibitively expensive (individual instruments costing over $1,000,000). Hopefully, the experiments chosen will give some idea of the breadth of techniques used and problems studied in neurobiology. If not, the instructor is always open to suggestions for alternative exercises.

The exercises in this course are stylistically quite diverse. But few of them are easy. You will need to learn to use new, complex, and unfamiliar equipment in short order. You will be developing skills such as fine dissection that normally take months to perfect. In each exercise we try to suggest more experiments than can normally be completed in the time available. The idea is to assure that the brightest students, the hardest workers, and those with the most luck, are still challenged and never bored!

Each exercise in this manual comes from a different source. Some were adapted and expanded from published undergraduate neurobiology laboratory manuals. Others were simplified from graduate laboratory courses taught elsewhere in the U.S. and Europe. Some were developed from scratch right here at Berkeley. All undergo continuing evolution and (we hope) improvement. Student input has had important effects on the exercises in the past. If you have specific criticisms and explicit suggestions for improvement (e.g., if you can correct errors or propose clarifications), your input will be most valued and will be taken very seriously.

You will gain the most from this course if you read the laboratory manual pertaining to each week's work before coming to class. The Graduate Student Instructor leading your laboratory section tends to get very annoyed when asked questions that are answered clearly in the syllabus. However, s/he will gladly answer questions about things that are not clear in the manual. The key to getting quality help from the GSI is to show that you have done your homework. This applies even to the very first exercise. Therefore, we ask that you read Exercise 1 before the first class meeting.

This course is meant to accompany, or follow, a basic lecture course in neurobiology. Completion of such a course is not, however, absolutely necessary, so long as you have read some sort of general introduction to the basic concepts and language of neuroscience (see Supplementary Readings). A lower division course in biology is, however, absolutely essential. You will also find the normal lower division courses in physics, chemistry, and mathematics to be very useful. An introductory course in (bio)physical chemistry would also be valuable.

Reports

The grade in this course will be based on two types of laboratory reports and on periodic exams. For exercises 1, 5, 6, 8, 10, and 11, you will hand in a short report. This report will normally be a 1- to 4-page form in which you fill in the blanks. You will be asked to answer questions about your experiments, and to recount or draw your results. This report may be due at the end of the exercise, or in some cases, it is to be submitted at the next class meeting (one-half week later).

For exercises 2, 3, 4, 7, 9 and 12, you will hand in a long report. This is a free-form report in which you describe your experimental results and answer questions posed in the laboratory syllabus. Do not write a scientific paper; omit introductions, methods, and discussion. Just state what you did and what you saw, and include all your data (usually in the form of photographs or Xerox copies of your partner's photographs). Brevity, completeness, and evidence that you understood what you did are rewarded with high grades. This report is due one week after completing the laboratory exercise; bring it to class the following week.

Please note that all reports are to be written up individually. You will of course collaborate with your lab partners in collecting, analyzing, and trying to understand the data. All members of a lab group will normally submit copies of the same results in their reports. But the final report, whether long or short, is to be written individually by each student. The report represents your work, your effort, your understanding, your ability to present your results and discuss them intelligently, and your answers to the questions posed in the exercises in your own words. The section instructors are quite adept at identifying plagiarism, and no credit will be given to duplicate reports or sections with very similar wording.

If you must miss an exercise or an exam due to a need to be out of town, you must make prior arrangements with the GSI to make up the exercise. If you miss an exercise due to illness or other unanticipated emergency, you must explain the problem to your GSI as soon as possible to arrange for completing the work. The GSI has the right to insist on written proof on official letterhead of your reason for absence. A report not handed in, or an exam missed without prior arrangement or legitimate proof of excuse, earns zero points.

Grades

Grades will be determined by total number of points earned as follows:

Exams:	2 X 50 = 100 points
Long Reports:	4 X 20 = 80 points
Crayfish, Oocyte Reports:	2 X 25 = 50 points
Short Reports:	6 X 10 = 60 points
	Total = 290 points

Laboratory Section 1 (Spring, 2014) TuTh 12-4 PM Control No. 58409

Week	Date	Exercise	Description	Report	Instructor
1	Tu Jan 21	1	Use of electrophysiological equipment	↓	Zucker
	Th 23		Use of electrophysiological equipment	Short	Zucker
2	Tu 28	2	Nerve excitability (frog sciatic)	↓	Zucker
	Th 30		Nerve excitability (frog sciatic)	Long	Zucker
3	Tu Feb 4	3	Voltage clamp computer simulations	↓	Zucker
	Th 6		Voltage clamp computer simulations	Long	Zucker
4	Tu 11	4	Intracellular recording 1: Dissection and recording	↓	Zucker
	Th 13		2: Effect of potassium and seeing EJPs	↓	Zucker
5	Tu 18		3: Correlated nerve and muscle recordings	↓	Zucker
	Th 20		4: Patterned nerve stimulation, EJPs and IJPs	↓	Zucker
6	Tu 25		5: Neuromodulatory control	Long	Zucker
	Th 27	5	EEG & human evoked potentials	Short	Zucker
7	Tu Mar 4	6a & 6b	Mechanoreceptors; Human sensory & motor systems	Short	Zucker
	Th 6		MIDTERM EXAM		Zucker
8	Tu 11	7a	Expression of transmitter-activated ion channels	↓	Kramer
	Th 13		Expression of transmitter-activated ion channels	↓	Kramer
9	Tu 18	7b	Expression of voltage-gated ion channels	↓	Kramer
	Th 20		Expression of voltage-gated ion channels	Long	Kramer
--	Tu 25		SPRING RECESS		
	Th 27		SPRING RECESS		
10	Tu Apr 1	8	Brain slice: long-term potentiation	↓	Zucker
	Th 3		Brain slice: epileptic focus	Short	Zucker
11	Tu 8	9	Neuron outgrowth: image processing	↓	Henteleff
	Th 10		Neuron outgrowth: image processing	Long	Henteleff
12	Tu 15	10	Neuron cytoskeleton: immunocytochemistry	↓	Larue
	Th 17		Neuron cytoskeleton: immunocytochemistry	Short	Larue
13	Tu 22	11	Mammalian neuroanatomy	Short	Larue
	Th 24	12	Neurogenetics 1: mutations & behavior	↓	Garriga
14	Tu 29	12	Neurogenetics 2: RNAi and development	↓	Garriga
	Th May 1		Neurogenetics 2: RNAi and development	Long	Garriga
(15)	W May 14		FINAL EXAM – Location TBD		Zucker
	7:00-10:00 PM		Exam Group 12		

Laboratory Section 2 (Spring, 2014) WF 12-4 PM   Control. No. 58412

Week	Date	Exercise	Description	Report	Instructor
1	W Jan 22	1	Use of electrophysiological equipment	↓	Zucker
	F 24		Use of electrophysiological equipment	Short	Zucker
2	W 29	2	Nerve excitability (frog sciatic)	↓	Zucker
	F 31		Nerve excitability (frog sciatic)	Long	Zucker
3	W Feb 5	3	Voltage clamp computer simulations	↓	Zucker
	F 7		Voltage clamp computer simulations	Long	Zucker
4	W 12	4	Intracellular recording 1: Dissection and recording	↓	Zucker
	F 14		2: Effect of potassium and seeing EJPs	↓	Zucker
5	W 19		3: Correlated nerve and muscle recordings	↓	Zucker
	F 21		4: Patterned nerve stimulation, EJPs and IJPs	↓	Zucker
6	W 26		5: Neuromodulatory control	Long	Zucker
	F 28	5	EEG & human evoked potentials	Short	Zucker
7	W Mar 5	6a & 6b	Mechanoreceptors; Human sensory & motor systems	Short	Zucker
	F 7		MIDTERM EXAM		Zucker
8	W 12	7a	Expression of transmitter-activated ion channels	↓	Kramer
	F 14		Expression of transmitter-activated ion channels	↓	Kramer
9	W 19	7b	Expression of voltage-gated ion channels	↓	Kramer
	F 21		Expression of voltage-gated ion channels	Long	Kramer
--	W 26		SPRING RECESS		
	F 28		SPRING RECESS		
10	W Apr 2	8	Brain slice: long-term potentiation	↓	Zucker
	F 4		Brain slice: epileptic focus	Short	Zucker
11	W 9	9	Neuron outgrowth: image processing	↓	Henteleff
	F 11		Neuron outgrowth: image processing	Long	Henteleff
12	W 16	10	Neuron cytoskeleton: immunocytochemistry	↓	Larue
	F 18		Neuron cytoskeleton: immunocytochemistry	Short	Larue
13	W 23	11	Mammalian neuroanatomy	Short	Larue
	F 25		Neurogenetics 1: mutations & behavior	↓	Garriga
14	W 30	12	Neurogenetics 2: RNAi and development	↓	Garriga
	F May 2		Neurogenetics 2: RNAi and development	Long	Garriga
(15)	W May 14		FINAL EXAM – Location TBD		Zucker
	7:00-10:00 PM		Exam Group 12		

Lecture Schedule

Wednesdays, 3-4 PM, 125 Li Ka Shing

Jan. 27	Nerve excitability (RZ)
Feb. 3	Voltage clamp simulations (RZ)
Feb. 10	Crayfish synapse: intracellular recording (RZ)
Feb. 17	PRESIDENTS' DAY HOLIDAY
Feb. 24	EEG (RZ)
Mar. 3	Mechanoreceptors; human sensory & motor (RZ)
Mar. 10	Expressing transmitter receptors (RK)
Mar. 17	Expressing ion channels (RK)
Mar. 24	SPRING RECESS
Mar. 31	Brain Slice (RZ)
Apr. 7	Neuron Outgrowth & Image Processing (RI)
Apr. 14	Neuron Cytoskeleton & Immunocytochemistry (DL)
Apr. 21	Neurogenetics 1: mutations & behavior (GG)
Apr. 28	Neurogenetics 2: RNAi and development (GG)

Faculty: RZ: Robert Zucker; RK: Richard Kramer; RI: Richard Ikegami (GG lab); DL: David Larue; GG: Gian Garriga

Supplementary Readings for MCB 160L

(on reserve in the Bioscience Library)

These selected readings have been taken from the following sources:

Animal Physiology by Roger Eckert and David Randall, W.H. Freeman and Co., S.F., 1978 (Pam 1)

Journal of Experimental Biology, Vol. 43, 1965 (Pam 7)

Fundamentals of Sensory Physiology by Robert F. Schmidt, Springer-Verlag, N.Y., 1978 (Pam 2, 4)

Textbook of Medical Physiology by Arthur C. Guyton, W.B. Saunders Co., Philadelphia, 1976 (Pam 2,3,4)

Introduction to Nervous Systems by Theodore H. Bullock, W.H. Freeman and Co., S.F., 1977 (Pam 6)

Frog Neurobiology, a Handbook. ed. by R Llinás and W. Precht, 1976 (Pam 4)

Unpublished, by Peter Getting (Pam 7)

Cold Spring Harbor Symposia on Quantitative Biology, Vol. 55, 1990 (Pam 8)

Nature Methods, Vol. 2, 2005 (Pam 9)

Differentiation, Vol. 72, 2004 (Pam 10)

Nature, Vol. 418, 2002 and Vol. 433, 2004 (Pam 10)

Exercise 2
- Propagation of Nerve Impulses — Pamphlet 1

Exercise 4
- A. Electrophysiological Recording (What They Normally Don't but Should Tell You)
- B. Reflex Control of Abdominal Flexor Muscles in the Crayfish — Pamphlet 7

Exercise 5
- Nonspike Signaling: Electric Fields of Units and Assemblies — Pamphlet 6

Exercise 6
1. A. Physiology of Vision (selections)
   B. Autonomic Control of Accommodation and Pupillary Aperature
   C. Color Vision — Pamphlet 2
2. A. Physical Principles of Optics
   B. The Optics of the Eye
   C. The Ophthalmoscope — Pamphlet 3
3. A. Somato-Visceral Sensibility (selections)
   B. Physiology of Hearing (selections)
   C. Hearing Abnormalities
   D. Physiology of the Sense of Equilibrium
   E. Vestibular Mechanism for Stabilizing the

Eyes and for Nystagmus
F. Clinical tests for Integrity of Vestibular Function
G. Physiology of Taste (selections) Pamphlet 4

4. Cutaneous Mechanoreceptors Pamphlet 5
(Pamphlets 2, 3, and 4 are for Exercise 6B; pamphlet 5 is for Exercise 6A)

Exercise 7
Molecular Studies of Voltage-Gated Potassium Channels Pamphlet 8

Exercise 10
Fluorescence Microscopy Pamphlet 9

Exercise 12
A. RNA Interference
B. High-Throughput RNAi in Caenorhabditis elegans
C. Unlocking the Potential of the Human Genome with RNA Interference Pamphlet 10

In addition to the selections from books and journals collected as pamphlets, the following books may be useful for the exercises indicated, and have been placed on reserve for this course in the Bioscience Library:

A very basic introduction to neuroscience, that can be read in one evening

Neurophysiology: A Primer by C. F. Stevens. John Wiley & Sons. Chapters 1-9.

Exercise 1

Physical Techniques in Biological Research by W. L. Nastuk, Vol. V, Electro-physiological Methods Part A, Academic Press, 1964. See especially chapters 1, 2, 3, 5, 7, 8, & 9.

Exercise 2

Physiology and Biophysics by T. Ruch and H. D. Patton, Vol 4, Excitable Tissues and Reflex Control of Muscle, W. B. Saunders Co., 20th ed., 1982. See especially chapter 4.

The Physiology of Excitable Cells by D. J. Aidley, Cambridge Univ. Press, 3rd ed., 1989. See especially chapter 4.

Exercise 3

Ionic Channels of Excitable Membranes by B. Hille, Sinauer Asso., 2nd Edn., 1992. See especially chapter 2.

Exercise 5

Human Brainwaves: the Psychological Significance of the Electroencephalogram by J. Empson, Stockton Press, 1986.

Exercise 9

Video Microscopy: The Fundamentals by Shinya Inoue and Kenneth R. Spring, Plenum Press, 1997.