

# An experiential electrophysiological course for biomedical engineers



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## ABSTRACT

An interdisciplinary course at the senior/graduate level has been developed and implemented to provide engineers with electrophysiological training. The contemporary engineering curricula generally consist of a significant amount of theory and mathematics. For biomedical engineering students this often results in limited hands-on experiences with live tissue samples and biological experimental techniques. In the Biomedical Engineering Program at the University of Rhode Island, this issue is addressed to some extent by implementing an experiential electrophysiology laboratory. The standard curriculum includes fundamental biology courses including Mammalian Physiology and Human Anatomy; however the underlying electrophysiological relationship is not fully obviated. The two-semester project course establishes this relationship and provides laboratory skills in dissection, instrumentation and physiological measurements. Additionally, the course serves as an electrical engineering design elective by incorporating electrical engineering applications with neuroscience. Two experiments were chosen to elucidate electrophysiological principals as well as providing a physiological template to construct an electrical model: 1) microelectrode recording of neuronal action potentials from the cerebral ganglia of the pond snail (*Lymnaea stagnalis*), 2) measuring of contractile forces and action potentials in odontophore protractor muscles of the American channeled whelk (*Busycon canaliculatum*) by use of a sucrose gap methodology. A circuit has been designed and constructed that emulates the action potential output of the neuron RPD1, located in the right parietal ganglion, of *Lymnaea stagnalis*. This laboratory has proven to be an effective way to provide undergraduate biomedical engineering students with invaluable live tissue experimentation skills in neuroscience and electrophysiology, while maintaining electrical engineering design applications.

## INTRODUCTION

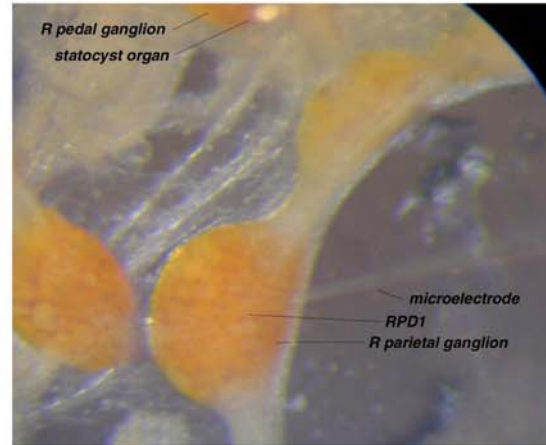
BIOMEDICAL engineering education may come from within a traditional engineering curriculum. This is the case at the Electrical Engineering Department of the University of Rhode Island. The program focuses on electrical engineering as it pertains to biomedical applications. A need exists to integrate the traditional engineering training with the intricacies of biological systems. Engineers are better prepared to solve problems when they have a first-hand understanding of what the problem is and its cause. For medical education during the last decade, the pedagogical style has also shifted from factual teaching towards contextual, or problem-based, learning [1]. To this end, we have developed a laboratory course to address these issues and give the biomedical engineering students the insight and research skills necessary to understand and help solve biological and physiological problems.

## METHODS

### Action Potential Recording Using Microelectrode Methodology

In 1952, Hodgkin and Huxley published a series of four papers describing the inward and outward currents of Na<sup>+</sup> and K<sup>+</sup> through the cell membrane [2]. These papers were the result of years of experimentation with the squid giant axon and the voltage clamp. In order to measure the influx of sodium ions and the outward flow of potassium ions, the membrane potential must be held fixed, or clamped, at a certain value. Therefore, any change in current, i.e. the flow of ions, which underlies an action potential, can be detected.

The neurophysiologic experiment conducted at our self-constructed lab at the University of Rhode Island involves the cerebral ganglia of the pond snail, *Lymnaea stagnalis*. Although the ganglia are very small, on the order of 200 to 400µm, the individual neurons are comparatively large. In fact, they are large enough to record the action potential with a pulled glass pipette microelectrode. However, removing the ganglia intact requires terrific skill and a great deal of practice since the ganglia are completely obstructed, positioned inferior to the buccal muscle. Once the buccal muscle is moved, the ganglia are exposed and the process of meticulously separating the ganglia from the surrounding tissue begins [3].



*Lymnaea Stagnalis* Cerebral Ganglia Highlighting the Right Parietal Ganglion and RPD1.

### Membrane Potential and Contractility Measurements Using Double Sucrose Gap Methodology

The American channeled whelk, *Busycon canaliculatum*, has a proboscis that varies from 4 to 8 cm in length. The muscles that control the radula, the teeth-like eating mechanism, are the odontophore protractors and retractors. The protractors are primarily used for re-positioning, while the retractors perform the power stroke in the scraping movement of eating. As such, the protractor muscles are singular and smaller in nature, and this makes threading through the double rubber membrane of the sucrose gap set up easier (figure 2).

Preparation of the tissue sample begins with breaking the shell of the *Busycon* to expose the snail body. Beneath the mantle and slightly lateral to the midline, the proboscis is located. This tube-like structure, covered by a protective sheath, is then separated from the body by removing it at the base. Once the proboscis is removed, careful dissection of the protective sheath reveals the musculature associated with the movements of the radula and the odontophore cartilage. Next, the protractor muscles are removed [4]. Successful recording of contractility from the force transducers (figure 2) requires proper tension in the attaching filament. Muscle contraction is achieved by introducing KCl to one side.

## RESULTS

Since the electrophysiological experiments themselves have been well documented by those much more qualified than biomedical engineers, we decided to stick to what we know best: Active electrical circuits and circuit modeling.

The neuron emulator (figure 3) performs real-time, interactive simulation of a neuron in terms of the following: 1) electrode and membrane resistance and capacitance for both current input and voltage output ports (figure 4), 2) membrane resting potential when input current is zero, 3) firing of action potentials above a depolarization current threshold, 4) adaptation of the spiking response (figure 5), 5) hyper-polarization following each spike, and 6) response to voltage clamping.

The neuron emulator was presented to the University of Rhode Island Intellectual Property Committee (IPC) and has subsequently been provided protection under U.S. provisional patent laws. The device will be made here at the University of Rhode Island, where further *Lymnaea Stagnalis* experimentation is providing the inspiration for improved modeling and interactive controls.

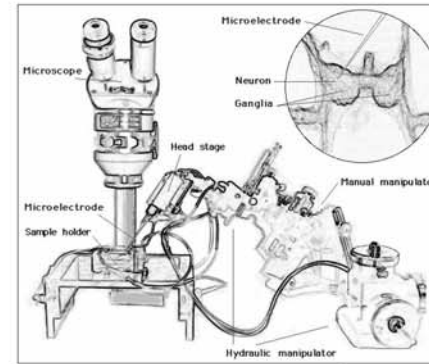


Figure 1. Microelectrode setup for neuron action potential recording. Inset shows the ganglia of *Lymnaea stagnalis*. (Mag. 500X)



Figure 3. The Neuron Emulator

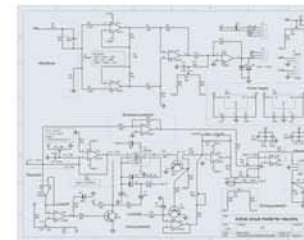


Figure 4. Schematic showing current model and potential model.

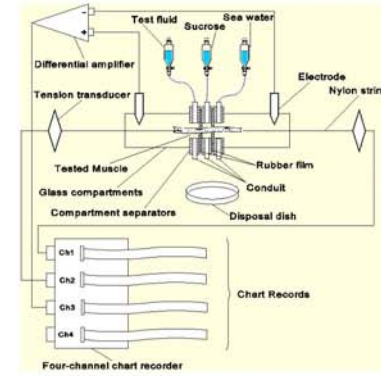


Figure 2. Diagram of double sucrose gap methodology.

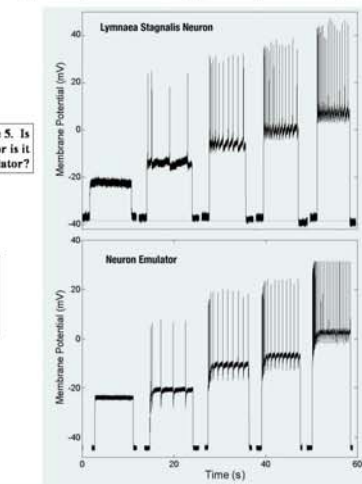


Figure 5. Is it live or is it Emulator?

## DISCUSSION

The experiments developed in this neurophysiology course have provided invaluable experimental training for the biomedical engineering students. Despite the specificity of the experiments, they exemplify advanced research in neuroscience and are technically challenging to motivate the students. As stated, a further goal of this course was to incorporate electrical engineering design skills. This was successfully accomplished with the design and manufacture of the Neuron Emulator, currently under the protection of U.S. Provisional Patent.

## REFERENCES

- [1] Epstein RJ. Learning from the problems of problem-based learning. *BMC Education* 4: 1-7, 2004.
- [2] Hodgkin AL, Huxley AF. A quantitative description of membrane current and its application to conduction and excitation in nerve. *J. Physiol* 463: 391-407, 1952.
- [3] Walker RJ. Intracellular Microelectrode Recording from the Brain of Helix. In: Experiments in Physiology and Biochemistry, ed. by Kerkut GA. New York: Academic Press, 1968.
- [4] Harrington, Lesley et al. Voltage Clamp of Cardiac Muscle in a Double Sucrose Gap. *Biophysical Journal* 13: 626-647, 1973.