ARTICLE
Reducing the Cost of Electrophysiology in the Teaching Laboratory

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Electrophysiology is a fundamental part of neuroscience and there are many published laboratory exercises suitable for undergraduates. However, the cost of equipping a lab is often a barrier to implementing these exercises. In this paper, we outline lab needs, suggest strategies for building a lab incrementally by adding equipment as budgets permit, and suggest specific areas for cost-cutting. We also point out instances in which it makes most sense to purchase or borrow research-grade equipment. A linked Google document lists specific items, prices, and purchase links.

Key words: electrophysiology, intracellular, extracellular, amplifier, micromanipulator, vibration isolation

This journal has published many laboratory exercises in electrophysiology, ranging, for example, from human electromyogram (EMG) and electroencephalogram (EEG) to *Drosophila* larval muscle, *Paramecium* membrane potential, plant action potentials, insect sensory systems, and crustacean neuromuscular systems (see selected exercises below the References section). Although the ongoing costs of an electrophysiology lab are relatively low, the initial setup cost is an insurmountable barrier for many institutions. Based on our experience teaching extra- and intracellular recording to students (Wyttenbach et al., 2014) and faculty (Johnson et al., 2014), we have come up with several recommendations for reducing both the setup and ongoing costs.

LAB NEEDS AND STRATEGIES

A basic research rig for extra- and intracellular recording would likely contain all of the items in Table 1, along with a shared programmable electrode puller. Even purchasing the least expensive versions of these items, such a rig could cost over $15,000, plus $4,000-9,000 for the shared electrode puller. Ideally, there would be enough rigs for 2-3 students per rig, so outfitting a teaching laboratory can be costly. Of course, these costs can be reduced if equipment is already available or can be borrowed. For example, microscopes and lights may be available from a general biology laboratory. If a nearby research lab has an electrode puller, electrodes can be pulled beforehand and brought to class. Depending on the lab exercises one wants to do, not all of this equipment is necessary: intracellular recording equipment is about ⅓ of the per-rig cost, while extracellular recording does not require the electrode puller. If the eventual goal is to do both intra- and extracellular recording, consider the following strategies.

Strategy 1, Start simply and add content

Start by building several basic rigs. On the strength of good course evaluations and promising student lab reports, request funding to improve those rigs and introduce new content. For example:

(1) EMGs, insect leg, and electric fish behavior do not need manipulators, vibration isolation, or data acquisition equipment. Minimal, low-cost amplification is needed; some signals are large enough to view directly on an oscilloscope or listen to with amplified speakers. Students can collect waveforms via the sound input of a computer. Table 2 shows items needed for such a minimal rig.

(2) Extracellular nerve recordings add an amplifier, suction electrode, and manipulator but require little or no vibration isolation. At this level, students can examine and pharmacologically manipulate motor, sensory, and rhythmic activity. The arthropod and mollusk preparations used in these exercises are inexpensive. Table 3 shows the additional items required for nerve recording.

(3) Finally, intracellular recordings let students analyze action potential shapes; examine and manipulate basic principles of synaptic transmission such as postsynaptic potentials, miniature endplate potentials, and synaptic plasticity; and pharmacologically manipulate ionic currents. Table 4 shows the additional items required for intracellular recording. The stimulator and isolation unit are not required for all exercises.

Strictly speaking, none of these exercises require a data-acquisition interface, but all would be considerably enhanced by it. Extracellular (AC-coupled) recordings can be acquired via the microphone input of a computer or tablet and viewed with free software. Oscilloscopes can be used; many campuses have unused ones in closets or they can be borrowed from a physics lab.

Strategy 2, Start small and add capacity

Start by building one full rig with high quality equipment (Table 1) and use it for demonstrations and individual projects. Gather evidence of success (student reports and evaluations) in order to request funding to make the experience available to more students. Which strategy works best will depend on the local budgetary and administrative environment. Some administrators prefer to grant large amounts of funding on a one-time basis, while others prefer to dole out smaller amounts on a more regular basis. Reiness (2012) suggests ways to persuade administrators to support neurophysiology teaching.

Programmable electrode puller
Dissecting microscope on a boom stand
Light source with fiber-optic light guides
Vibration-isolation table
Faraday cage
Micromanipulators on magnetic bases (two)
Extracellular and intracellular amplifiers
Stimulator and stimulus isolation unit
Data acquisition system and computer
Audio monitor
Cables and connectors
Dissection tools and prep dishes

Table 1. Full rig.

Pin, wire, or EMG electrodes
Audio monitor
Computer or tablet with microphone input
Extracellular amplifier (may not be needed)

Table 2. Minimal rig.

Microscope and lighting
Suction or pin electrodes
Extracellular amplifier
Coarse micromanipulator
Magnetic base
Preparation dish
Faraday cage or other shielding
Dissection tools and prep dishes

Table 3. Additions for nerve recording.

Intracellular electrodes and holders
Vibration isolation
Fine micromanipulator
Intracellular amplifier
Stimulator and isolation unit
Electrode puller (shared)

Table 4. Additions for intracellular recording.

EQUIPMENT

This section discusses major equipment in general terms. For specific recommendations with comparisons, prices, and links, see the Google document linked in the Resources section. That document also covers minor equipment such as cables and preparation dishes.

Vibration isolation

Vibration isolation requires a work surface with mass, isolation between work surface and substrate, and a stable substrate. Exercises using the minimal rig (Table 2) do not require vibration isolation. Even nerve recordings (Table 3 equipment) require little isolation. However, a work surface that can hold magnetic bases is highly desirable. On a stable bench, a thin sheet of steel will do. However, inexpensive vibration isolation can be obtained by placing a ⅜″ (10 mm) thick steel plate (24×36″, 600×900 mm) on four #10 rubber stoppers, tennis balls, or squash balls. In a basement lab far from vibration-producing machinery, this may also suffice for intracellular recordings. The only way to be sure is to try it. In less stable settings, an active vibration table is ideal, but they can cost over $3000. Even tabletop versions cost $1000-2000. As an alternative, a set of four passive inflatable isolators (~$100 apiece) under a thick steel plate can give sufficient vibration isolation for intracellular recording.

Electrode positioning

Nerve recording and intracellular recording require electrode positioning with micromanipulators mounted on magnetic bases (which in turn attach firmly to the steel plate mentioned above). Intracellular recording requires a three-axis manipulator with a fine-advance knob. Many teaching labs use the Märzhäuser M3301, Narishige M-3333, or Narishige MM-3 for intracellular recording. For extracellular recording, the fine-advance knob is not necessary, nor does the manipulator need to be as stable as the three cited above. The less expensive Kite (WPI brand) or Narishige M-3 will do, and Backyard Brains offers a very inexpensive plastic 3d-printed manipulator (described by Baden et al., 2015). There is also a variety of do-it-yourself designs using screws, micrometer heads, and springs to advance and retract the electrode (e.g., Krans et al., 2006).

For most applications, micromanipulators are mounted on magnetic bases via rods and clamps. These cost well over $100 when packaged with a manipulator but can be found very cheaply at machine-shop suppliers (e.g., Harbor Freight) or on Amazon; see the Google doc for specifics.

Optics

Microscopes (stereo with 6-40× head and 10× ocular, 10 cm working distance) can often be borrowed from general biology labs. A boom stand is ideal but not required. If the base gets in the way, many microscopes can be turned on their stands so that the base is behind the field of view (put a weight on it to keep the scope from toppling, a lead brick is ideal). Good used microscopes can be found on eBay; we often see Wild and Olympus models for sale. If purchasing new microscopes, look for ones with trinocular heads that can accept cameras. A variety of USB webcams can be fit into them, making it possible to demonstrate procedures or document behavior during recording. If a trinocular head is not available, many USB cameras fit in an eyepiece holder.

The lights provided with many student microscopes will not do. Most of them are AC powered and will bring noise into the recording. They are often rigidly attached to the microscope body or stand, with little ability to change the position. A light source with fiber-optic guide (preferably split into a Y-shape) is ideal but expensive. However, they are frequently available on eBay. As an alternative, look for LED microscope lights or mount a pair of LED flashlights on light-duty magnetic bases. With either LED or fiber-optic lights, you may need to turn off the lights during recording, due to electrical noise.

Electronics

Extracellular recording generally requires an amplifier designed for low input impedance and high gain. Look for a selectable gain with bandpass and notch filters. Insect leg recordings may need only 100×, crayfish nerve needs 1000×, and snail nerves often need 10,000×. A-M Systems and WPI sell amplifiers in the $1000-2500 range. If budget permits, these are good easy-to-use options. However, the circuitry is straightforward and there are many low-cost options for those willing to build, package, and debug circuits. Land et al. (2001) describe a general-purpose amplifier with 100× and 1000× settings and noise levels comparable to those of commercial units. Matsuzaka et al. (2012) describe a more complex circuit with a driven shield for noise reduction and separate bandpass filters for EMG and action potential recording. The parts for each of these cost below $50. Similarly, inexpensive do-it-yourself amplifiers are specialized for EMG (Crisp et al., 2016; Crisp, 2018) or EEG (Jain et al., 2011). For those who want pre-made solutions, Backyard Brains sells inexpensive SpikerBox amplifiers (Marzullo and Gage, 2012) in versions specialized for nerve (insect leg or earthworm) or human EMG, with fixed gain of 880×, priced from $100-250. In general, neither extra- nor intracellular recording in the teaching lab requires pre-amplification.

Intracellular amplifiers are specialized for high input impedance and low gain. We are not aware of any do-it-yourself intracellular amplifier designs. WPI and A-M Systems have comparable versions in the $1000-1500 range, while A-M Systems has a $2500 version with the added convenience of a digital readout for membrane potential and electrode resistance.

These days, most labs control stimulus timing by computer, but a stimulus isolation unit (SIU) is still needed. The SIU isolates the recording signal from much of the stimulus artifact by providing a separate ground path for the stimulus. Some DAQ systems (e.g., A-M Systems) provide an isolated human-safe output, but this is too weak to stimulate nerves through suction or pin electrodes. Commercial SIUs start around $1400, but Land et al. (2004) describe one that can be built for about $50. Unlike the commercial units, this one does not produce constant current, which is not necessary for student use.

Nerve stimulation through genetic engineering of neurons to contain light-sensitive ion channels (Sjulson et al., 2016) has opened up exciting possibilities for teaching (Pulver et al., 2011; Titlow et al., 2015; Pokala and Glater, 2018; Rose, 2018,). High quality, inexpensive LED circuits for optogenetic stimulation have been described (Pulver et al., 2011), or high intensity commercial LED flashlights can be used (Rose, 2018). There is no need for electrical stimulators and SIUs with optogenetic stimulation, and students are introduced to a state of the art technique in neuroscience. As with electrical stimulation, a computer would control stimulus timing.

Data acquisition

Ideally, each rig would have a data-acquisition (DAQ) system and computer for display and analysis. Together, these can be the most expensive part of a rig, costing several thousand dollars. Tables 2-4 do not list them. Strictly speaking, they are not necessary – research labs lacked them until the 1980s and still did more sophisticated recordings and analysis than would be expected in a teaching lab today. However, a good DAQ system adds so much to the learning experience so much that we suggest adding one as early as possible, even for the exercises requiring only minimal equipment (Table 2).

George (2006) describes several options. While his product reviews are dated, the design considerations he lists remain valid. For most teaching purposes, two channels of input, one channel of output, and a sampling rate of 10 kHz will suffice, although recording of pulse-type electric fish discharges requires rates up to 100 kHz. If an output channel (stimulus) is not needed, then one can use the sound input of a computer to record two channels at 44.1 kHz and save them to the hard drive for analysis (use free Audacity software or other low-cost audio software to record). However, sound-card data acquisition has the major limitation that it can only handle AC signals and thus cannot handle intracellular, slow EMG, or slow electro-retinogram (ERG) signals. Several free phone and tablet apps can give an oscilloscope display of sound input, again subject to this limitation; Backyard Brains has one intended for electrophysiology. Several labs and individuals have written their own DAQ software and made it available. We have used the free Spikehound software (Lott et al., 2009; no longer updated) and have heard that the inexpensive DataView software (Heitler, 2007; still actively updated) works well. Both are for Windows only and can use the microphone input or other DAQ hardware.

Many companies offer inexpensive USB-enabled DAQ boards. We have not tried them, but their specifications look good for neurophysiology. However, the software is basic and may require a lot of work to adapt for physiology. If budget permits, we recommend purchasing a commercial system with full-featured and well-supported software or, failing that, one of the custom solutions mentioned above.

Finally, some sort of audio monitor is helpful for all lab exercises. Students like the immediate feedback of hearing extracellular action potentials. While some DAQ systems support playback through computer speakers during recording, inexpensive cell phone speakers or computer speakers can be connected to the physiological amplifier with T-connectors and adaptors.

Data analysis

Spike sorting and rate calculation are built into the DAQ software discussed above. Users of the Crawdad manual (Wyttenbach et al., 2014) can paste waveform data into its analysis tools for simple window-discriminator peak-finding and instantaneous spike rate measurement. This may be helpful for those using audio input to collect waveforms. For more sophisticated spike sorting, Quiroga (2007) links to research labs that have made their software available.

In our classes, most students use spreadsheets such as Microsoft Excel (or free alternatives) for further analysis and graphing. This suffices for nearly all the lab exercises we do. More specialized analysis (e.g., shapes of action potential and postsynaptic potentials, quantal analysis of

miniature endplate potentials) can be done in MatLab or any programming language. This could be a motivating challenge for engineering and computer-science majors interested in neuroscience.

CONCLUSIONS

If funds are available, it makes sense to buy high-quality commercial solutions. Prioritize vibration isolation and good micromanipulators. These are durable items, and poor quality will make recording a frustrating experience. Next, consider options for data acquisition and analysis. If your teaching goals emphasize quantifying data, good DAQ software will greatly improve the quality of student work. If you cannot borrow good microscopes, make them the next priority, since they are a long-lasting investment (we still happily use Wild M5 microscopes from the 1970s). Finally, spend what is left on the best amplifiers you can afford. Economize on the rest, especially cables and other small, easily replaced items.

This article did not cover true do-it-yourself hardware. However, this is an exciting time for those who like to design and build custom equipment. As the cost and quality of 3D printing improve and platforms such as Arduino and Raspberry Pi have become established, designers have much to work with and will, we hope, share their designs widely.

RESOURCES

For specific product recommendations, comparisons, prices, and links, see the Google docs linked below (Figure 1). The editable one can be modified by anyone; comments on it are sent to the first author, who will reply whenever possible. In case the editable document becomes corrupted, the stable one, editable only by the author and periodically updated from the editable document, is available for reference.

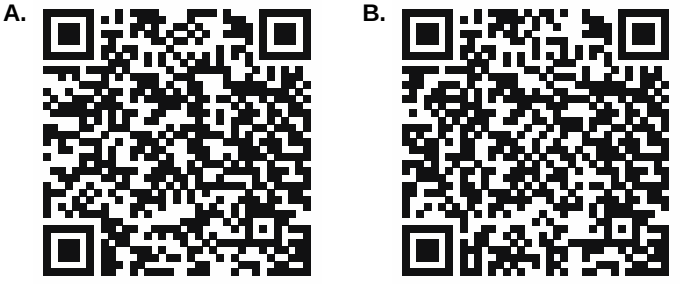

Figure 1. Google doc links. *A.* Editable (https://docs.google.com/document/d/1V6aLdTgNI50EHUrcHDQXZPcQKaKE9Mr3Tgf-gzaRSro). *B.* Stable (https://docs.google.com/document/d/1N0ADzuMRdyKLvUZ73qymol6uZIlL2Y0Axa48pbgErYg

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SELECTED LABORATORY EXERCISES

These are published in The Journal of Undergraduate Neuroscience Education; not cited but listed as a resource.

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