TECHNICAL NOTE
Construction of a Simple Suction Electrode for Extracellular Recording and Stimulation

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Principles of signal transmission in nervous systems are commonly demonstrated in the undergraduate neuroscience laboratory through extracellular recording of nerve and muscle action potentials. Here we describe the construction of a simple suction electrode that we use routinely in our laboratory classes for nerve recording and stimulation. The electrode parts are relatively inexpensive, easily available from established scientific and electronic distributors and local hardware stores, and the electrode is resilient to student handling. Our undergraduate students use this electrode design for high resolution, extracellular recordings of action potentials from crayfish motor and sensory nerves and insect muscle, and for stimulation of crustacean and insect motor nerves.

Key words: extracellular recording, suction electrode, action potential, crayfish, teaching lab

Student laboratory exercises in undergraduate neurophysiology labs routinely use the extracellular recording technique to teach principles of cellular and systems neuroscience. Exercises using different extracellular recording configurations to examine a variety of bioelectric phenomena such as action potential conduction, stimulus response functions and adaptation of sensory organs, the organization of motor activity, and electrocardiograms have been described in the neuroscience teaching literature (for example, see: Wyttenbach et al., 1999; Olivo, 2003; Berlind, 2005; Dawson and Robertson, 2005; Friedman, 2005; Gray and Robertson, 2005; Silver, 2005; Trimmer, 2005; Krans et al., 2006; Ramos et al., 2007). Extracellular electrophysiological recording has simpler technical requirements than intracellular recording, and thus is attractive to use for undergraduate lab exercises. Extracellular recording usually does not require fine micromanipulators, microelectrode pullers, or acquisition systems capable of recording DC voltage shifts. In addition, extracellular AC recordings can be acquired directly through the sound port of a lap or desktop computer for convenient and cost-effective display and analysis (Lott et al., 2007).

We use suction electrodes for recording nerve and muscle activity in our undergraduate lab classes and in neuroscience demonstrations and workshops for both students and teachers at the high school and college levels. Our college students also stimulate motor nerves with these electrodes to examine dynamic properties of synaptic transmission (Wyttenbach et al., 1999). We previously described a suction electrode design for student recording that used a plastic syringe barrel to house suction tubing and input cables to an AC amplifier, and used ultra micro gel loading tips to interface with motor or sensory nerves (Wyttenbach et al., 1999; Land et al., 2001). The major advantage of this electrode was that the tip did not break easily, and it could be quickly replaced when damaged. The small diameter of the gel loading tip allowed good recordings from preparations described in the Crawdad neurophysiology lab manual (see examples in Wyttenbach et al., 1999 and Land et al., 2001). The main disadvantage was that the electrode had several glued connections which weakened with student use, allowing air to disrupt the vacuum seal between the electrode and nerve. This often reduced the signal to noise ratio, adding variability to the quality of student recordings.

The electrode construction we describe here consistently improves the resolution of recordings, is durable, and is also simple to build. It uses a plastic rod to house the amplifier input cables, a pin jack to connect the active recording lead to an intracellular electrode holder with a side suction port, and a glass capillary tip, broken to the appropriate size, to interface with nerves. Since suction is applied directly to the electrode holder, it is easier to avoid and fix vacuum leaks, and thus maintain a good seal between the electrode tip and a nerve.

We introduced this suction electrode to faculty at the IFEL (Introduction to FUN Electrophysiological Workshops) workshop held at Bowdoin College, July 27-30, 2006. This workshop was sponsored by NSF (DUE-0231019), the Faculty for Undergraduate Neuroscience (FUN), Edvotek, and ADInstruments (Mead et al., 2007). The faculty workshop participants each made and used electrodes of this design for their workshop recordings. They thought it could be useful for their neurophysiology laboratory teaching, and encouraged us to provide a description of its construction for the neuroscience teaching community.

MATERIALS AND METHODS

Electrode Construction

The suction electrode consists of the following parts: Input cables for an AC amplifier, plastic tube for housing input cables and a pin jack, an intracellular electrode holder (silver wire style with side port), tygon tubing, a 3-way stopcock, syringe, silver wire, and a pulled capillary glass tip (Fig. 1). Table 1 lists the individual electrode parts and their sources. Construction of a single electrode takes

about one hour the first time. A supplemental instruction video for the electrode construction can be found at: http://www.bowdoin.edu/faculty/s/shauptma/videos.shtml.

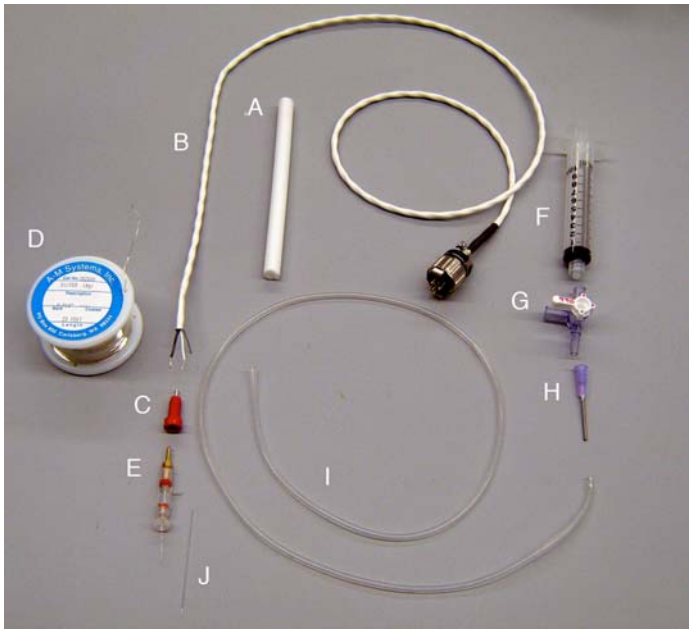

Figure 1. Suction electrode parts. *A.* Electrode handle. *B.* Input cable. *C.* Pin jack. *D.* Silver wire spool. *E.* Microelectrode holder. *F.* 10 ml Disposable syringe. *G.* 3-way stopcock. *H.* Blunted syringe needle. *I.* Tygon tubing. *J.* Glass microelectrode.

Electrode handle and input leads

A 15 cm piece of 3/8" X ¼" PEX tube (sold in home improvement stores as: "faucet riser," "lavatory riser," "sink riser" or "toilet riser") cut from a longer commercial piece with a small handsaw or a utility knife serves as the main body, or handle, of the suction electrode. Prepare the handle for the input cable by first making a hole 2 cm from one end of the tube (the distal end) with a soldering iron. The hole should be wide enough to pull the reference wire of the differential amplifier through with forceps (Fig. 2).

We use a 5 pin input cable that is specific for an A-M Systems differential amplifier. The amplifier input connector will vary depending on your type of AC amplifier. For example, pin, banana or BNC connectors can be added to a two-conductor, shielded audio cable of appropriate length to interface with the amplifier. The A-M Systems cable has three wires: a driven ground, and two insulated differential leads. The A-M amplifier still requires an external ground connected to a system ground point. Our directions apply to this cable, but they can be easily adapted to other input cables.

Remove about 5 cm of outside insulation and shielding from the distal, free end of the A-M Systems cable. Trim the shielding wire back to the insulation edge and separate the two differential lead wires. Strip about a centimeter of insulation from the ends of the white (positive lead) and black wire (differential reference lead for the A-M amplifier) wires, or enough to solder them to the pin jack or the bare, silver wire (see below). Push the input cable through the electrode handle so the black and white wires extend about 3 cm out the distal end. Before soldering, dip the ends of both in phosphoric acid to ensure a good solder joint (For tips on soldering, see http://www.elexp.com/t_solder.htm). Pull the black wire (the external reference wire) back through the hole in the handle with forceps and solder the bare end to a 15 cm length of 0.01 inch diameter silver wire (Fig. 2). Solder the white wire to the pin jack (Fig. 2), and insert the pin jack into the handle, screwing it in if necessary, by rotating the cable along with the pin jack and pulling the cable from the other end of the handle. If the pin jack fits too loosely, add a drop of Superglue to the pin jack base, and twist it into the handle to spread the Superglue.

The electrode handle we use fits into our student manipulators (model M3333 from Narishige, MM33 from Fine Science Tools and the Kite model from World Precision Instruments), but its diameter may be too large for some micromanipulators. Alternative handles are listed in Table 1, but it may be necessary to make the internal diameter of these a little larger for the pin jack to fit. This can be done by scraping the inside of the tube with the blade of a small screwdriver.

To finish the electrode handle, fill the reference wire hole in the handle with hot glue. Push the cable down just a little, not tight, into the proximal end of the handle, and fill this end with glue too. The glue is a strain relief for the input cable and reference wire.

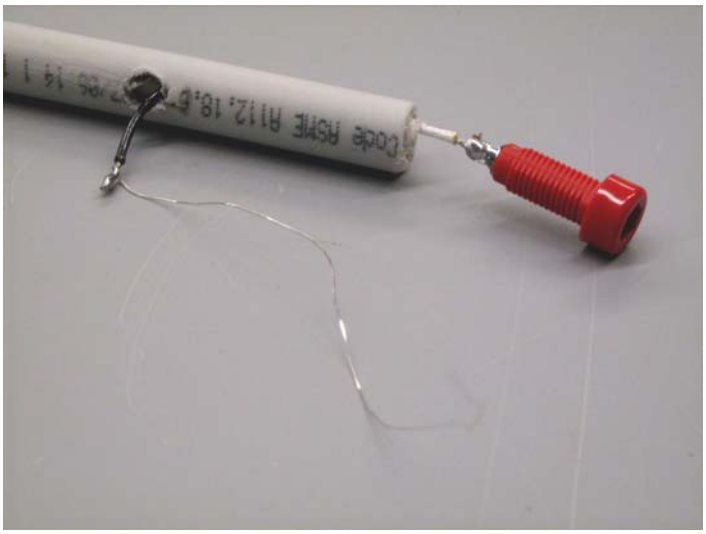

Figure 2. Electrode handle showing 5 mm hole for external reference wire soldered to silver wire lead, and pin jack soldered to the internal (white) amplifier.

Suction apparatus

The suction components of the electrode are shown in Figure 3. Cut a 25 cm length of the 1/16th inch ID, 1/8th inch OD Tygon tubing. Using a triangular file, score one side of a 16 gauge syringe needle a centimeter or so above the tip. Using two pliers, one on either side of the score, snap off the tip of the needle and discard it in a sharps container. Use the file to smooth the edges of the tip. Insert the needle into one end of the tubing. Fit the needle into the 3-way valve, and connect a 10 ml syringe

to the other side of the valve. Connect the free end of the tubing to the side vent of the microelectrode electrode holder. Insert the pin of the electrode holder into the pin jack on the electrode handle.

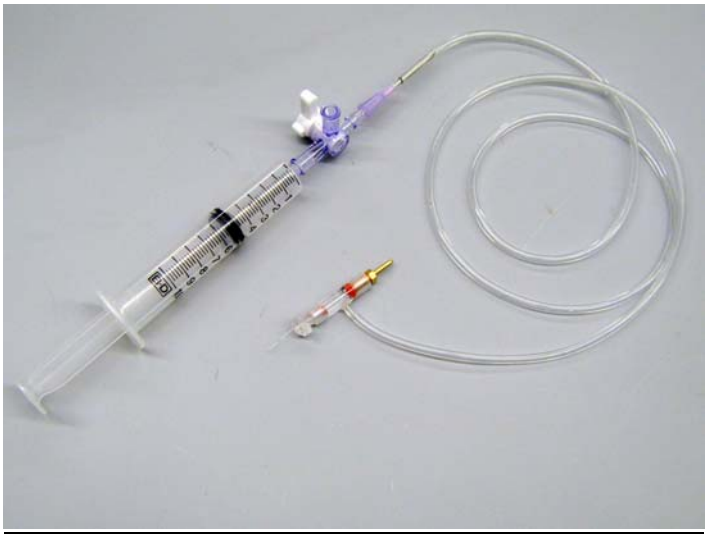

Figure 3. Assembled suction components of the electrode. See Fig. 1 legend for parts identification.

Recording tip

Prepare some glass capillary tubes as if they were to be used as intracellular electrodes. If an electrode puller is not available, pull a piece of capillary electrode glass in half over the flame of a Bunsen burner or alcohol lamp. We use 1 mm outside diameter glass with or without an internal filament, but any size glass that fits into your microelectrode holder will do. Glass needles with short shanks clog less frequently than those with long, drawn out shanks. Prepare a tip opening of the appropriate size, a little larger than the diameter of the nerve, by using dissecting forceps to break off the tip. This can be done more finely by scoring the glass with a diamond tipped "pencil" under a microscope and then breaking the tip at the score. We found that breaking the electrode tip when it is close to the nerve allows us to better judge the correct tip size. The jagged edges of the broken tip can be smoothed by fire polishing it over an alcohol lamp, but this will reduce the size of the tip opening, and may close it completely. Suction will be less effective with very small tip sizes. Fire polishing recording tips for crayfish motor and sensory nerves is not necessary, but it can improve the electrode seal. We have found fire polishing helpful for insect muscle recordings (Johnson et al., 2006).

To complete the electrode, insert the glass tip in the microelectrode holder by loosening the cap of the electrode holder, and sliding the electrode glass into the holder, with the holder's recording wire inside the capillary tube. Make sure the capillary tube is pushed past the gasket in the holder, and tighten the holder's cap gently to ensure good suction. A finished suction electrode is shown in Figure 4.

Testing and using the suction electrode

First, check the electrical continuity of the input cable with ohmmeter leads attached to the appropriate wires exiting either side of the input cable. With the continuity confirmed, put the suction electrode into a micromanipulator and position the tip near the nerve. Use the manipulator controls to touch the nerve with the tip of the electrode. Make sure the 3-way stopcock is correctly configured for suction through the electrode tip, and pull the nerve into the electrode with the syringe. It is best to suck up a loop of the nerve, but it is also possible to suck the end of a cut nerve into the electrode, as done when recording from the crayfish muscle receptor organ (Wyttenbach et al., 1999). However, be sure to suck in more than the cut end of a nerve, because the damaged nerve ending may be dead. The electrode tip can also be sealed against the side of a large, less flexible nerve (*en passant* recording), such as the ventral nerve cord of an insect or crustacean. Once the nerve is positioned for recording, turn the off position of the stopcock towards the tubing to hold the vacuum.

The quality of the recording depends on how well the nerve fits into or against the electrode. If this connection is loose because of a large electrode tip, the signal to noise ratio of the recording will be poor, and the nerve may slip out of the electrode. Replace the tip with a smaller one.

The signal to noise ratio can sometimes be improved by gently pushing the electrode tip and nerve against other tissue, like muscle, thus creating more electrical resistance between the active lead and the reference wires. If the tip is too small, it may not be possible to generate enough suction to hold the nerve tight; break the tip wider with dissecting forceps. Pulling too tightly on the suction can stretch and damage a nerve.

Spontaneous tonic motor activity from nerve 3 of a crayfish abdominal ganglion was recorded using this suction electrode connected to an A-M systems Model 1700 Differential AC amplifier (gain = 1000X). The amplifier output was sent to an ADInstruments Powerlab box for signal display and analysis. Detailed background, all procedures, and recording set-up for this preparation are found in Wyttenbach et al., Lab 2 (1999).

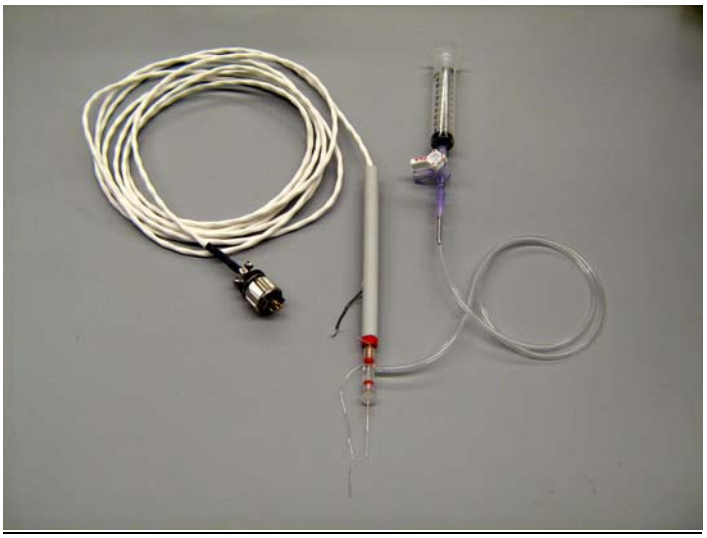

Figure 4. Completed suction electrode. See Fig. 1 legend for parts identification.

Price	Company	Contact Information	Description
Electrode handle			
$2- $3 ft	Hardware stores	Home Depot, Lowes	PEX tubing, 3/8”X1/4”, sometimes sold as “sink riser”
$2 - $10	McMaster-Carr	www.mcmaster.com	Narrower OD alternatives : Butyrate hollow rod: 5/16”x3/16” PETG hollow rod: 5/16”x3/16” Garolite hollow rod: 5/16”x3/16”
Pin jack			
$1.50	Newark Electronics	www.newark.com	Insulated deluxe tip jacks, various colors
Tubing			
$8/50 ft	Cole-Parmer	www.coleparmer.com	Tygon tubing, 1/16” ID, 1/8” OD
Electrode holder			
$45	Warner Instruments	www.warneronline.com	ESW-M10P electrode holder w/side vent
3-way stopcock			
$20/pack of 10	Cole-Parmer	www.coleparmer.com	Luer connection, male slip
Syringe			
$24.50/box	Cole-Parmer	www.coleparmer.com	10 ml disposable syringe, tip
Syringe needle			
$22.75/pack of 100	Cole-Parmer	www.coleparmer.com	B-D disposable syringe, 16 gauge, 1 ½”
Cable			
$30/3-ft cable	A-M Systems	www.a-msystems.com	Prepared for A-M systems interface (4 come with the amplifier)
$10/50 ft	Radio Shack	radioshack.com	24-gauge audio cable (2 conductor, shielded) For BNC or Pin connectors
Silver Wire			
$41/spool	A-M Systems, Inc.	www.a-msystems.com	.010” , not coated
Capillary glass			
$22/package of 250	A-M Systems, Inc.	www.a-msystems.com	Thin-walled, 1 mm X.75mm, with filament, 4”
Total cost for one electrode without connector cable ~ $50.00			

Table 1. Parts, description and source listing for the suction electrode described in the text. Prices are in US dollars at the time of publication.

RESULTS AND DISCUSSION

This motor activity shown in the extracellular recording example of Figure 5 from nerve 3 of the crayfish abdominal ganglia contributes to postural control of the intact crayfish tail (Kennedy and Takeda, 1965). This nerve contains only six motor axons, which fire spontaneously or after reflex activation (Wyttenbach et al., 1999). The axonal diameters of these motor axons are different enough for students to record extracellular AP amplitude classes that correspond to motor axons of specific diameters (Wyttenbach et al., 1999). With our earlier suction electrode design students typically observed only four distinct amplitude classes. Examples of our earlier “best” recordings are found in Land et al. (2001) and Wyttenbach et al. (1999). These did not have the resolution to routinely define the six different AP amplitude classes that are seen in Figure 5. We point out that the recording of Figure 5 was made by a high-school student in a recent Bowdoin College summer program, which highlights the new electrode’s ease of use. Our students also use this suction electrode to record locomotor activity from house fly muscles and sensory responses from crustacean muscle receptor organs, and to stimulate fly and crayfish motor nerves to examine synaptic transmission (Wyttenbach et al., 1999; Johnson et al., 2002, 2006). In our experience at both Cornell and Bowdoin, this electrode produces consistently higher resolution recordings, and is less prone to the air leaks that can compromise suction that occur in our earlier design.

Impatience and carelessness by students will lead to frequently broken glass electrode tips, and require the

instructor to remove broken glass from the electrode holder. We have experienced this in our classes. One of us (RHB) has had student experience with both our suction electrode designs, and he has certainly broken his share of glass electrode tips. Despite that, he still prefers our new design because of the higher resolution recordings possible, and the relative ease of changing tips or adjusting them for different nerve sizes. An alternative we have not tried is to attach a short length of small diameter tubing to a blunt capillary tube in the electrode holder. This would allow a flexible contact with the nerve or muscle (see Miller, 1979 and Yoshida, 2001). In summary, extracellular glass electrodes are not student-proof, but we feel that their advantages strongly outweigh any increased time and effort required to replace a broken glass electrode or clean out the electrode holder.

Figure 5. Spontaneous motor activity from nerve 3 of a crayfish abdominal ganglion recorded with the suction electrode described here. The numbered action potentials (APs) indicate six different AP amplitude classes, each from an axon of a different diameter. Number 1 is an AP from the smallest axon and number 6 is from the largest axon.

Our goal here was to provide a useful construction guide for faculty or students to make a suction electrode that was durable, easy to use, achieved high quality extracellular recordings from motor and sensory nerves, and efficiently stimulated nerves. There is nothing especially novel about this particular suction electrode design. It is a modification of various other designs for nerve recording and stimulation that are currently used for research and teaching (see Miller 1979, and references in Land et al., 2001 for a variety of suction electrode designs for different purposes). There is a commercial suction electrode available from A-M Systems, Inc. (www.a-msystems.com), that is suitable for the types of extracellular recording we described here. Its cost at $62.50 is comparable to our design, considering that an electrode holder costs around $45 (Table 1, note that the same electrode holder can also be used for intracellular recording exercises). We found that the A-M electrode was less stable on the student manipulators than our design because their amplifier connecting cable attaches to a BNC port on the handle itself.

Our new electrode design can be used for nerve recording or stimulation in both invertebrate and vertebrate preparations. In addition, our undergraduates and high school students have used this electrode to record field potentials from electric organ discharges by simply putting the electrode in an aquarium containing mormyrid fish (Wyttenbach et al., 1999). We present this construction guide for a suction electrode as part of a long-term project to increase inexpensive technical resources available to neuroscience teaching faculty (see also Land et al., 2001, 2004; Krans et al., 2006).

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