



INSTRUCTIONS FOR NERVE CUFF ELECTRODES

STERILIZATION

Nerve cuffs are made entirely of autoclavable materials – silicone rubber, Teflon*, and stainless steel. They can be steam autoclaved without special precautions. If gas sterilization (EtO) is preferred, be sure to pack nerve cuffs in a gas-permeable bag and allow adequate out-gassing time (at least 48 hours) to be sure all toxic gases have been desorbed from the silicone rubber. For preparations in which aseptic technique is not essential, rinsing in 70% ethanol, followed by air-drying is an effective cleaning technique to remove dust. Surgical sterilization solutions are best avoided because they are difficult to remove completely from the interstices of the electrode wires.

SURGICAL HANDLING

The nerve should be completely freed from surrounding tissue attachments for as long a length as possible. Avoid cutting nerve branches, as these usually contain essential feeding blood vessels; include them in the tube if necessary. Make sure there will be some slack in the nerve at either end, and that it will neither be kinked by an abrupt change of direction near the cuff end, nor stretched by attached connective tissue. Pass one of the suture organizer tabs and sutures under the nerve, and position the cuff parallel to the nerve, with the opening facing up just under it. Fix one set of sutures in place (e.g. clamping organizer tab to stable structure) and open the cuff by pulling evenly on all the opposite sutures simultaneously (using the opposite organizer tab). The nerve should slip in easily. In some tight access situations, it may not be possible to open the entire cuff evenly. In this situation, the nerve can be started into one end and the tube zipped open by an inserted glass probe, sliding the nerve in as you proceed to the opposite end. The compression on the nerve from the edges of the silicone tube is usually not severe enough to cause direct fiber damage, but will temporarily occlude blood flow, so be ready to act quickly.

SURGICAL FIXATION

The sutures attached to the cuff are arranged to allow easy, secure closure of the slit by tying them snugly across the opening. The suture organizer keeps them from tangling, so cut each end free as it is needed. A simple square knot should be sufficient. Take care not to pull too tightly as the tube may be crushed, along with the nerve. A snug, even closure is essential for tri-polar recordings, as any leak greatly degrades the common mode rejection for extrinsic sources of noise. Even though the cuff appears to lie snugly closed without sutures, connective tissue will eventually penetrate and separate the seam. Of course for acute recording, this is

not a problem and you may want to save the sutures for handles in future use. If the cuff lies freely in a mobile space (e.g. between crossing muscles), it is advisable to secure it to some relatively stable fascial plane which will move with the nerve. A fixation suture wrapped around the cuff or around the side arm tube containing the leads will provide strain relief to prevent traction on the leads from being transmitted to the nerve. Be sure to leave plenty of slack in the leads and to direct them so as not to apply pressure on any overlying skin incisions. Consider the mechanical dynamics between nerve and adjacent tissues carefully before using fixation techniques. They may cause the very traction that you seek to avoid.

TESTING

If you have any doubts about the integrity of the electrical leads or their insulation, the best test is a “bubble test” done *in vitro* in a bowl of saline. Use a low-voltage DC battery (6-9 volts) to apply *the negative voltage* to each lead in succession as the cuff lies immersed in the saline. The positive side of the battery should go to a large surface area ground (the outside of a stainless steel bowl works well). When contact is made, you should see a stream of bubbles (hydrogen gas) coming from the connected electrode within the tube and from no other electrodes or external point along the tube or leads. The best test of an implanted nerve cuff is the contact impedance measured by a low-current AC (1 kHz) impedance meter. Measure both the impedance of each contact versus remote ground and the inter-electrode impedances for various contact pairs. The pair-wise impedance will be somewhat less than twice the individual contact impedance (a range of 2-5 kilohm is typical), and will tend to be somewhat larger for more distant pairs than for adjacent pairs. Individual contacts near the ends of the tube generally have slightly lower impedances vs. ground and vs. each other than do central contacts. Expect a fair amount of contact variability, though, between contacts and over time. Changes of a factor of two in either direction are not significant. This test is mostly for gross loss of continuity or insulation integrity, both of which are most likely to occur at the connection point rather than in the cuff itself. There is no simple electrical test for the viability of the nerve except the ability to evoke and/or record action potentials with the appropriate stimuli. If you don't have an AC impedance tester which is accurate in this range, use a sine wave generator with a large series resistance (1 megohm) and blocking capacitor (0.1 uF) to generate a constant current sine wave. If you start with 10 V p-p @ 1 kHz, the signal across the electrode will be 10 mV/kilohm.

CONNECTION OF LEADS

The stranded stainless steel wire which forms the contacts in the tube and the flexible leads was selected for its strength and biocompatibility. It is somewhat difficult to make good mechanical and electrical connections. The best technique is soldering with the use of special stainless steel fluxing liquid. Carefully strip the Teflon* insulation from the end by nicking and pulling it, taking care not to nick the wire strands. Dip the wire end in the acid flux or paint it on generously using a cotton swab. Allow a few seconds for it to take effect. Using a freshly

tinned, clean soldering iron (regular 60/40 rosin core solder), quickly dip the fluxed wire into the solder ball. You should hear a hiss as the flux evaporates, and the wire strands should be immediately drawn together by a solder bead, which will be firmly fixed to the wire after it cools. Cut off any stray ends that have not been well tinned. If you have any doubt about the adhesion of the solder, cut off at least 5 mm past the exposed portion and start again. Do not attempt to re-flux or re-solder the first site, as the rosin will coat the strands and make this ineffectual. Once the end is tinned, it can be soldered in the conventional manner to any pin or other termination. Be careful to protect any connection points from fluid leakage.

For simulating, it is common to select a bipolar pair of adjacent cuff contacts and use an isolated source of charge-balanced, biphasic stimulation, with the cathodal first stimulus applied to the contact closest to the direction in which propagation is desired. If you wish to record the stimulated volley from another set of electrodes in the same cuff, stimulus isolation, an intervening ground contact and a very fast recovery amplifier will probably be necessary to escape the stimulus artifact. Remember that the contacts are stainless steel and subject to corrosion with repeated stimulation, particularly if net DC is passed. Always use a series-blocking capacitor even when the stimulus is biphasic in shape to prevent any net DC current. Use stimulus waveforms as short in duration as possible to minimize electrode polarizations. Because of the confined and optimally oriented current flow, thresholds should be low and quite uniform for all of the fibers of a given caliber. To obtain stable differential thresholds for various populations of nerve fibers in the face of scar tissue formation around the nerve and contacts, it will be necessary to use accurate controlled current stimulation rather than voltage stimulation.

CONFIGURATION OF LEADS

The contacts are numbered sequentially from distal to proximal according to the handedness of the cuff. The right-hand (standard configuration) can be visualized by holding your right hand palm upward and imagining your thumb to represent the exit of the leads. The cuff then lies with the slit facing up (parallel to fingers) and the first lead comes from the contact closest to your fingertips (distal). This lead is identified by one knot and each subsequent contact proximally has one additional knot near its free end. For left-handed cuffs, the conventions are the same but the orientation is for the left hand held palm upward. For tri-polar recording, a triad of contacts evenly spaced with respect to each other is selected. No contact should be closer to an end than one diameter of the tubing. The two outer contacts are connected to the negative (inverting) input of a differential amplifier and the central contact is connected to the positive (non-inverting) input. The ground or reference of the amplifier must be connected to some other point in the preparation (a fourth cuff contact can be used, preferably at or near an end).

TROUBLE-SHOOTING

The most common electrical failures are poor solder joints or fluid leakage at the connection point to the leads, resulting in open circuits (noisy recordings) or degraded common mode rejection and spurious stimulation. The latter may be difficult to detect because of the normally low contact impedances. Look particularly for sudden impedance decreases.

The most common cuff failure is incomplete closure of the slit, resulting in degraded common mode rejection of external signal sources, such as EMG. The most common surgical/biological problem is nerve blockage. This can occur immediately at surgery as a result of stretching, in which case it may recover over a day or two, or from ischemia of the nerve, which will generally not show up for many hours and will not recover except by the slow process of regeneration distally. A more pernicious problem arises from constriction of the nerve by the inevitable pseudomesothelial encapsulation of the silicone rubber of the tube. This tends to occur at 7-10 days post-op, and can produce an abrupt complete blockage (particularly in large diameter fibers) in a nerve that has been quite functional since surgery. The recommended margin of the cuff inner diameter of 1.4 times the nerve bundle outer diameter is critical. While excessive over-sizing degrades the recorded signal or stimulation threshold only gradually, just slight under-sizing can result in this complete block. If block develops in a tube that is clearly not undersized, the most likely cause is kinking of the nerve at the ends, caused by poor surgical placement, inadequate dissection of connective tissue, or traction from the leads. If cuffs are reused, carefully examine the placement of each contact along the walls of the tubing after it has been removed and cleaned. Occasionally a loop of wire becomes bent away from the wall and will act as a guillotine on any nerve bundle implanted into such a cuff.

*Teflon is a registered trademark of DuPont