Hyperdrive Assembly Instructions
CENL updated/modified, 2/2015
Check server for updates

Obtain a disassembled hyperdrive and check that it is complete. Obtain access to the hyperdrive building tools.

Write drive core number here:_____.

When you know it, name of the rat:_____ and date of surgery_____.

The instructions below are in strict sequential order. Check off each step when done. You will be working close to the limit of human performance: eye-hand coordination, hand tremor, cleanliness, organization, patience… know yourself and take breaks when necessary! In principle, you can build a drive in one weekend (2-3 days). In practice, a week should suffice. Try not to 'drag' the building process, the longer you take the higher the risks of errors.

1) Make sure that all of the listed parts and tools are available. Obtain, or prepare 20 tetrodes (30cm long total wire length)

___ Collet Nut ___Collet ferrule ___ Post ___ PCB Nut ___ Core ___ Legs (14)
___ Bundle ___ Post/Anchor screws ___ Shroud screws ___ Tetrode cover (optional)
___Shroud ___ Dust Cap/Cover ___ Tetrode cap ___ PC Board

2) Inspect the hyperdrive parts.

___ A) Use a sharpened length of 23 G cannula (not a needle) to ream out each drive cannula hole in the core (outer circle) until travel is absolutely unimpeded.

B) Under the microscope, check the following for each leg:
- ___ All parts of the leg are clear of residue (make sure that 30 G cannula will slide through the drive cannula).
- ___ Bottom of drive cannula is smooth.
- ___ Axes of bearing and drive cannula are parallel to one another and both parts are secure in carrier assembly.
- ___ Notches on drive cannula can be seen and are deep.

C) Use tetrode turning tool to carefully install each leg of the hyperdrive. Check that the notches are facing inward (towards the center of the drive). While installing, check the following:
- ___ Bearing accepts tool and rotates smoothly on drive guide screw. If not, replace the leg.
- ___ Leg travels freely all the way up/down screw, while drive screw remains locked into core.

___ D) If a leg does not pass the above tests, remove and replace it and repeat steps A-D. Label the replaced leg with the drive #, date and a clear description of the problem, and return it to its original container.

___ E) Remove each leg while supporting it to avoid twisting or bending the drive cannula.

___ F) Test the fit of all parts (except bundle) that are to be inserted into or onto the hyperdrive during actual assembly. This includes 1) the post assembly: post, post-screw, PC board, PCB nut and cap. 2) Shroud and shroud screws. 3) Collet ferrule and collet nut.

3) Load the guide cannula bundle. (See Appendix: Bundle Manufacturing)

- Note: In some cases, a refurbished drive may still contain a functioning cannula bundle. If so, simply proceed to step #4
- Ream the cannula bundle with a 0.005/0.006 piece of music wire, even if it has already been built for you. Inspect the edges and make sure they are not sharp and/or damaged
- Cut 14 pieces of 0.005 music wire, 10 cm long, with a pair of wire cutters (not scissors)

___ A) Place the part of the bundle with the heat shrink tubing melted on it into the collet (check orientation - notched end of collet points toward bottom/brain-end of bundle) so that 5 mm of the base part of

the bundle protrude. Clamp the collet onto the heat shrink using padded pliers. Use sharp blade, if necessary, to cut off any heat shrink tubing that extends above the collet. You might have to use Teflon tape to ensure a snug fit. After you make sure you have a good fit, take the ferrule out until step J.

___ B) Use one of the pieces of music wire cut above. Bend over the tip at one end by about 90 deg (use coarse tweezers).

___ C) Align the soldered end of the guide cannula bundle with the bottom of the core.

___ D) Following the bundle loading diagram, thread the unbent end of the music wire through a guide cannula, starting at the soldered end of the bundle, and then through the appropriate drive cannula hole in the core. This may be easiest to do if you place the bundle soldered end up above the upside down core and thread the wire down while watching under a microscope. Make sure the music wires *do not cross each other*, so that the cannulae splay out nicely.

Bundle Loading Diagram (see appendix)

___ E) Bend over the end of the music wire protruding from the core to prevent it from sliding out (90 deg). The bent end should be less than 5mm.

___ F) Similarly, thread pieces of music wire through the thirteen remaining guide cannulae (back to B)

___ G) After you have done all 14 cannulae, gently push or tap the music wire pieces until their bent ends are flush with the bottom of the bundle.

___ H) Carefully push the bundle up through the bottom of the core so that the ends of the guide cannulae rest near the drive cannula holes. Ensure that the bundle is not twisting.

___ J) Holding the bundle upright with the soldered end resting on the hard table surface, push down on the core (hold the white part, not the rods) so that the bundle cannulae slide up through the drive cannula holes in the core. Reposition the ferrule and push the bundle into the core until it seats firmly. You may do step M here.

___ K) Apply silicone grease to the flat surface on the inside of the collet nut (not on the threads) or on the notch part of the collet ferrule, and screw the collet nut firmly onto the core, periodically checking that the collet is seating correctly (not tilting) in the nut and that the bundle is not twisting in the core as you tighten. If the bundle begins to twist, immediately stop and reverse your motion; take care to avoid twisting the bundle as you finish mounting the collet nut.

___ L) Confirm that 5 mm of the bundle protrude from the bottom of the core (if not, remove nut and adjust collet on bundle). These 5 mm are necessary to ensure proper application of celastic during surgery.

___ M) With the wire cutters, cut the bent parts of all music wires, and remove them from each guide cannula. If the wire does not slide out of any cannula or exits any cannula with a lot of curvature, replace the bundle and return to step A. You may do this step after J, if you prefer.

___ N) Guide cannulae should protrude above core ~1-2 mm. If they protrude too much, it may be difficult to load the legs without bending the bundle.

___ O) From the top of the drive, thread a sharpened, unbent piece of 0.006" music wire through each guide cannula. It might help using sand paper to smooth the tip of the wire to ensure better entry into the cannulae (it is a tight fit). If it doesn't move all the way through or is very curved when it exits from any cannulae, then there is a problem with the bundle; repair or replace it and return to step A.

4) Re-inspect and reload legs. Do <u>NOT</u> use force when loading legs. If force is needed, something is wrong.

___ A) Under the microscope, check each leg for the following:

___ Drive cannula is smooth at the bottom.

___ Bearing accepts tool and rotates smoothly in carrier.

___ All parts of leg are clear of residue – ream with sharpened 30g cannula.

___ Axes of bearing and drive cannula are parallel to one another and both parts are secure in carrier

___The notches on the drive cannula can be seen and are deep.

___ B) Load each leg and test its integrity:

___ Carefully load leg, being sure not to bend or push guide cannula into center of drive. (It is often helpful to insert a piece of music wire to help align the drive cannula with the guide cannula). This step should be done while watching the bundle from the top of the core under the microscope, to ensure it doesn't bend.

___ Make sure that drive screw and cannula are parallel.

___ Under the microscope, check for backlash; the cannula and carrier should move up and down smoothly, with no delay as the bearing is turned.

___ C) Replace any leg that fails integrity testing and repeat steps A & B. Label the replaced leg with the drive #, date and a clear description of the problem, and return it to its original container.

___ D) Once all of the legs have been loaded and tested, screw one of the legs down to the bottom position (~1 mm from core). Leave the other legs about 2/3 of the way up.

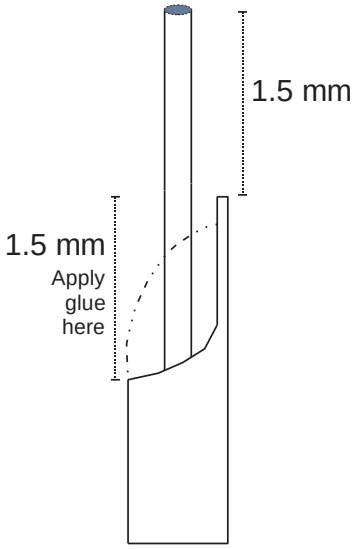
___ E) Check the bundle through the center of the core to make sure that none of the cannulae have been bent. If any have, remove and discard the bundle (save the collet ferrule and nut) and start again at step 3.

5) Insert wire connectors into drive core. (Usually 5: the number needed will depend on the number of stimulation or other wires to be connected to the board of the hyperdrive. Two are mandatory for the ground wires. If these connectors are already in the core, just make sure they are clean and that a 30G cannula can be inserted, and go to 6). If you need to replace one of them, do the following steps:

___ A) Cut, smooth and ream out the ends of the appropriate number of 13 mm-long pieces of 23 G stainless steel tubing.

___ B) Crimp each tubing piece at the center and insert and push it into one of the inner circle holes in the drive core until both ends are slightly recessed in the hole. They should fit very tightly. (Choose inner circle holes that will be just below the desired connector board holes when the board is installed, i.e; 5, 6 &7). Use the round needle nose pliers for crimping.

6) Cut and load and glue silica tubing.



___ A) Using a piece of .005" music wire, measure the distance from the top of the cannula of the leg that was left at its lowest position to 3-4mm (for CA1, for VTA make this 4-6 mm) beyond the bottom of the bundle (the total should be 33-37mm). Enter length here: _____mm.

___ B) Wash your hands and clean off your work space with EtOH – dust and oil at this step can slow you down considerably.

___ C) Screw the legs ¾ of the way down so all are at the same level.

___ D) Use a #11 scalpel blade to cut 14 lengths of tubing. To get a clean cut, gently roll the silica tubing back and forth under the blade until it snaps. It is helpful to put the pieces in a weighing boat so none of them get lost.

___ E) Use the microscope to perform the next steps F-K.

___ F) Load the silica into each cannula from the bottom of the core and verify that it comes out of the top of the leg cannula approximately 1.0mm (including notch). Or visually: the silica should stick out of the leg by a bit less than the length of the notch (see figure). You can adjust how much the silica protrude by pushing it flush with the bottom of the bundle and moving the leg up/down (the silica should stay in place).

___ G) Secure the drive into a holder and glue the silica tubing pieces to the leg cannulae near the very top of the cannulae using thick superglue. Do not use thin glue. Put the glue at the base of the notch on the leg. Keep the drive near <u>horizontal</u> to avoid that the glue runs down the leg to the bundle creating a blockage that will bend the bundle. Also make sure no glue is put on the rotating nut of the leg, or the threads of the rod guide. Do 3 legs at a time, then wait 15 mins, and turn the drive to do the next 3 legs.

Tricks:

- Put a drop of glue on a piece of aluminum foil, then use a 30 G needle to deposit the glue.
- Alternatively (***works better***) use a 31G Insulin syringe. Put a small drop of glue inside, then replace the plunger and push small drops as needed.

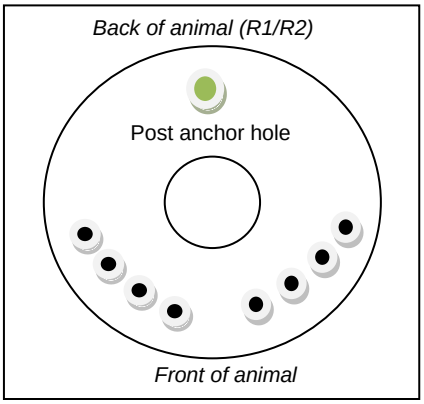
___ H) Repeat the application of superglue to ensure a secure connection between the metal and silica.
___ I) (Optional) Once the superglue has set, apply a drop of clear nail polish using the same technique as with the glue. This also provides a secure seal of silica to metal.
___ J) Keep the drive upright and allow silica/legs to dry overnight before proceeding.

_____ **End Day 1** _____

___ K) Gently move the legs up-down to make sure they are not glued to the bundle by accident (look at the tip of the drive, see if the canullae move when you turn)
___ L) Pass a dummy tetrode through each silica tube to ensure it goes through smoothly and easily, without curving as it exits. You can also use a 0.002 actuator wire. Use black plastic-tipped tweezers to push tetrode/wire through.

***** *From this point on, never screw legs all the way down -- leave at least 1mm between bearing and core.*
***** *Keep the drive in a holder*

7) Insert grounds (2), EMG/EEG and stimulation wires if any.



___ A) Cut one 2.5 cm long piece of 0.0055" diameter teflon coated stainless steel wire for each ground wire to be connected and two such pieces of 0.0055" wire for each stimulation electrode..
___ B) Strip both ends of each wire back 2-3 mm (use scalpel).
___ C) Cut (with diamond wheel) and smooth a 7 mm long piece of 30 G cannula for each wire. You may need to use a 30G needle to clear the end of the cannula to make it hollow.
___ D) Crimp a prepared piece of 30 G cannula onto one end of each wire and slightly bend the tubing near the middle-distal end (~2-3 mm from tip). See picture in the 'ground screw' appendix.
___ E) Push the bent 30 G cannula pieces into the appropriate wire connectors in the drive core so that the wires extend above the core. The cannula can stick out about 2 mm. Label the diagram (G1,G2 for the grounds, etc…)
___ F) Record the EIB holes used for each EEG/EMG channel:

E1:_____ E2:_____
E3:_____ E4:_____

8) Load tetrodes (see also Appendix: Tetrode Manufacture).

___ A) You should have about 18-20 tetrodes ready (see Appendix II).
___ B) Under the microscope at high magnification, using plastic black forceps only if necessary (use clean fingers) and taking great care not to bend it, feed a tetrode into each silica cannulae. Mounting the drive so that the leg you're loading it into is aligned with your hand movement may facilitate tetrode loading. If the tetrode appears bent or curved after being loaded, replace it. You will want to load the tetrode far enough into the silica so that about 2/3 of the "loop" extends above the leg. In general, when manipulating tetrodes, hold them by the loop, not by the tip (that part will go in the brain).

9) Install post and Electrode Interface Board (connector board).

___ A) Screw each leg down to bottom position (~1 mm from core), watching carefully to ensure that none of the cannulae in the bundle bend (this is best done under the scope). If any one of the legs seems to threaten to bend its bundle cannula, remove the tetrode and silica (by soaking the leg in acetone) and replace them following steps 4, 6 and 7.
___ B) Make sure that the connector board is clean. Double check all the holes in the board, make sure they are unobstructed. Do not use sharp metal objects (e.g. no needles) to clean the holes. (Optional) use the impedance tester (attach syringe needles to leads) to ensure it is free of cross connections (wash your hands before handling the board).
___C) Screw the post screw partly into place – it's easier to get it started before you insert the post.
___D) Insert the post into the core and lock it into place by tightening down the post screw. Do not overtighten or you will damage the post.
___E) Inspect the board holes. Make sure they are free of leftover wires.
___F) Set the board on the post so that the numbers on the board line up with those on the core (e.g. R1/R2 should be on top of R1 and R2). Insert and **tighten down the PCB nut**. Make sure it is tight, but not too tight, as you will glue it down later on.

10) Connect Ground, EEG/LFP/EMG and Stimulation Wires to Board.

___ A) Bend the stainless steel wires so that they run along the post and secure the stripped ends into the appropriate holes in the connector board by tightly pushing gold pins into the holes alongside the wires. Use the 'large gold pins' (if this does not work, use the 'small pins'). Using the special pin crimping tool, secure the gold pin into the board and cut off excess wire at the top of each hole. Be careful, pins tend to want to jump!
___B) (Optional) Test connections using the impedance meter. Touch leads from the board end down to the corresponding 23G on the bottom of the core.

11) Connect tetrodes to board. Use Dumont #5 forceps, and the dissecting scope at about 1 Mag.

___ A) Thread all four electrode wires of tetrodes R1 & R2 through the appropriate holes in the connector board so that the tips just project from the top of the hole.
___ B) Insert a ('large') gold pin into each hole and push it in tightly, holding and adjusting the wires to ensure that the tips of the wires do not get pushed back out of the hole.
___ C) Take the pin crimping tool and gently squeeze the gold pin into the board. BE CAREFUL NOT TO JERK THE PLIERS AND RIP OUT THE WIRE! Make sure the tool is exactly aligned with the board not to chip it.
___ D) Repeat steps A, B & C for all the other tetrodes, connecting each individual electrode wire into a separate hole. Thread the 4 wires first (one pair at a time). Use the forceps to pinch the large gold pins (hollow center), and place 4 of them gently in the holes (tap gently to immobilize them). Use the pin crimping tool to secure them in place.
___ E) Ensure all connections are secure and unbroken.
___ F) Use a 20cm length of suture to pull and tie the tetrode wires back toward the connector board post. To do so, create a large loop with the string with a double twist. Place the loop around and under the board, and gently pull. The diameter of the suture loop should be a little bigger than half that of the PC board. Put a small drop of super glue on the knot. Cut the excess suture string.
___ G) Ensure each tetrode has enough slack - watch as you move its leg up and down to its limits to ensure that the wires will never be pulled too tight between connector board and drive cannulae.
___ H) With all legs pulled up turn one leg at the lowest position, cut the tetrode to an even length so that a minimum of 9 mm protrude from the bundle (assuming hippocampal dorsal CA1 recordings. You may adjust for other areas. This cut isn't the final cut yet).
___ I) Repeat H for all tetrodes.
___ J) Cut off excess wire above pins with the mini scissors.

12) Pretest tetrode connections (optional).

___ A) Turn on the impedance tester and check the battery by pushing down the top (battery/test) switch to "test". The needle should read at least 4 on the scale. If below 4.0, replace batteries. Clip one input lead from the impedance tester to the base of the testing well, and clip a syringe needle (25G or more) to the other.
___ B) Use resistors to ensure that the impedance tester is calibrated properly in the MΩ range. Clear the work area of all unnecessary tools. Clean.
___ C) With the drive in the holder, extend tetrodes approximately 3 full turns out of bundle so that all protrude an equal length.
___ D) Fill the well with saline solution.
___ E) Carefully attach drive to the mount ring above the testing well and lower the drive until all tetrodes are in the solution. Danger – it is easy to ruin the drive now with one false move!
___ F) Have meter set in test mode and range on 5M. Touch needle to each channel on the hyperdrive connector board and watch for the scale to drop from open circuit to approximately 2-5 MΩ
If there is no drop you may have a bad channel. To be sure, after you are done, use a regular voltmeter and re-test the ones you suspect are disconnected. If this test is also negative you can try 1) to push the pins down a bit more or 2) recut the tip of the tetrode. If after this you still do not see a reasonable impedance, replace the tetrode.
___ G) Check off channels as they pass the test (Ground: test between connector and cannula at bottom of drive. Should be low):

	TT1	**TT2**	**TT3**	**TT4**	**TT5**	**TT6**	**TT7**	**TT8**	**TT9**	**TT10**	**TT11**	**TT12**	**R1**	**R2**	**GROUND**
1															
2															
3															
4															

___ H) Return the drive to the drive holder. Let tetrodes dry overnight
___ I) (Optional) Use the impedance tester to test for short circuits between all neighboring and "next-door-down" channels, including references. Any short circuits (< 4.0 MΩ) must be repaired. This can often be accomplished by separating wires that are touching one another as they exit the holes in the connector board.

13) Glue tetrodes into drive.

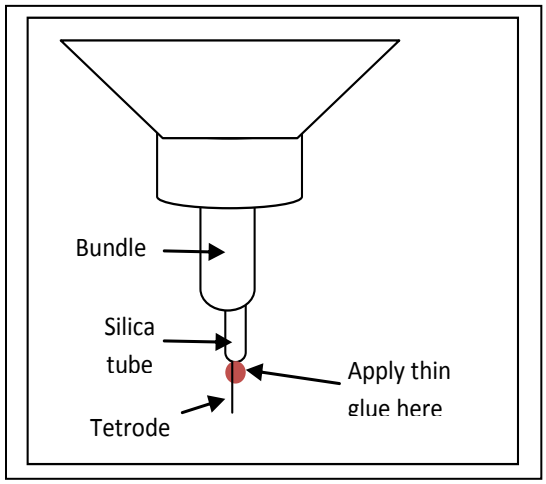
___ A) With all tetrodes lowered, inspect the way the tetrode exists the silica tubing on top of the leg cannula. Try and gently bend the cannula inward (towards the post) about 90 deg.
___ B) With the drive in drive holder, glue each tetrode into place at the top (near PC board) of the silica/tetrode joint using **thick** superglue. To apply glue, use tricks in step 6G. Let dry overnight. Be careful not to put glue on the leg screw or rods nearby (rehearse without glue, make sure you have the right angle). If this is too hard, raise the leg until the top is more accessible.

___ R1 ___R2 ___TT1 ___TT2 ___TT3 ___TT4 ___TT5 ___TT6
___ TT7 ___TT8 ___TT9 ___TT10 ___TT11 ___TT12

_____ **End Day 2** _____

___ C) Retract all tetrodes. If they are very long, you can cut them, but no closer than 4 mm from the bundle

___ D) Place the drive/holder horizontally. One by one, extend the tetrode to its full extent. Place a drop of **thin** superglue on aluminum foil (or cover-cap of a centrifuge tube, or see tricks in 6G). Use a piece of 0.005' music wire (or less preferably, 25 G needle) to apply glue at the junction between the silica and the tetrode (see figure). The drop of glue at the tip of the wire (or syringe) should be barely visible to the naked eye (but visible under the scope). The glue should flow within the silica tube by capillary force. Let dry for at least 5 mins.



Note: **This is a dangerous step**: make sure not to touch the tetrode (or it will bend), and make sure not to put too big of a drop (or the tetrode will not retract). Don't use a syringe to dispense the glue here, drops may be too big. A mistake here will make you lose 2 days' worth of work per tetrode damaged!

Note: The glue should be very liquid. Replace the drop on the aluminum foil if it thickens (typically, after 2-3 tetrodes). Get a twisted piece of kimwipe ready in case you put too much glue and need to suck it out.

___ E) When the glue is dry, inspect under the microscope the smoothness of each tetrode's travel as the leg is turned up and down. Replace any tetrodes that have a jerky movement. Retract tetrode. Repeat B-C for all tetrodes and references.

___ F) (Optional) Attach the tetrode cover to the connector board so that the turning tool will reach the leg bearings easily.

14) Finalize the drive assembly

___ A) Apply a small drop of Loctite/regular-superglue with 25G needle at the bottom of the PCB nut; Make sure there is no glue on the sides of the threads (this is where the screw of the cap will go). **Tighten down the PCB nut slightly with the PCB nut tool**. Do this quickly: the loctite will harden and screwing after that point can ruin the PCB nut.

___ B) Retract the tetrodes/legs to their upper most position

___ C) Attach the shroud and dust cover to the drive

15) Make final cut of tetrodes.

___ A) Secure hyperdrive horizontally.

___ B) One-by-one, advance tetrodes all the way (until nut is ~1 mm from core). Under the microscope, cut each tetrode to the correct length. Use sharp portion of 'final cut' tetrode scissors (You may need to experiment with a few locations on the scissors with a dummy tetrode first. You should barely be able to feel the tetrode being cut. If you can feel the scissor blades cutting the tetrode, then the blade is not sharp enough, and it is pinching the wires). Inspect the cut after it is made. If the tetrode is bent or if the edge looks splayed or somehow "not clean", recut it. Retract the tetrode, and be careful not to break the silica, or bump into the PC-board.

___ R1 ___R2 ___TT1 ___TT2 ___TT3 ___TT4 ___TT5 ___TT6
___ TT7 ___TT8 ___TT9 ___TT10 ___TT11 ___TT12

16) Measure dead turns

Under the dissecting scope, for each tetrode, assess the number of turns (in 1/8) required to change the direction of movement of the tetrode.

R1 _____up _____down	R2 _____up _____down	TT1 _____up _____down
TT2 _____up _____down	TT3 _____up _____down	TT4 _____up _____down
TT5 _____up _____down	TT6 _____up _____down	TT7 _____up _____down
TT8 _____up _____down	TT9 _____up _____down	TT10_____up _____down
TT11_____up _____down	TT12 _____up _____down	

End Day 3

17) Gold plate tetrodes (no more than one or two <u>days before</u> the surgery). This step takes ~1:30 h.

___ A) Test the batteries of the impedance meter. Calibrate impedance meter (use 2 resistors, 500k and 2.0 MΩ). Connect the positive pole of the stimulator to the EXT positive pole of the impedance meter, to the ground pin of the well. After turning the stimulator on, make sure the LED on top of the red (positive) pole is on (use 'polarity select to switch). Organize your work space so that your hand does not come close to the hyperdrive when manipulating the knobs.

___ B) Fill testing well with fresh gold solution. There are 2 bottles: One stock and one 'in use'. Use the latter unless it is dark (in which case, discard and replace with stock solution). Use one of the 'test' needles (with a gold pin at the tip) and pinch it with the alligator clip. This needle can go in the black connector pins (a 25 G needle will work too).

___ C) With all tetrodes retracted, place the drive in the holder, and secure it with the L-shaped screw. Adjust the height of the holder (the bundle should <u>not</u> touch the solution).

___ D) Lower one tetrode at a time (start with R1 and R2). Make sure to bring the entire drive up (metal knob) before lowering or retracting a tetrode.

1. With the impedance meter switch to 'Test', touch the pin hole on the corresponding black connector on the board. Measure the impedance (should be between 1 and 4 MΩ), and write it down (table below). If it reads greater than 5, make sure the tetrode is indeed touching the water. If so, just enter '>5' in the table.
2. Then switch the impedance meter to 'Ext', and on the stimulator, pass 2.5 µA current through the electrode channel for 1-2 sec by pressing the DC/Test toggle switch to momentary. You should hear a low-pitch tone.
3. Stop injecting, and switch the impedance meter to 'Test' again. The electrode impedance should have dropped from its initial value to 0.5-1.0 MΩ.
4. Repeat the current injection if the impedance is not between 0.5-1.0 MΩ. If you hear a high-pitch tone, there is an open circuit (bad contact, or tetrode not in the water…). If you hear a low-pitch tone but the impedance does not drop, the circuit is closed but has a high impedance. If the impedance does not change after multiple attempts at lowering it, try and increase the current a bit (3.0 or 3.5 µA).
5. **TRICKS:** There are a number of tricks that may help, including moving tetrode in and out of the gold solution, passing current in opposite direction, redoing gold pin connection and re-cutting tetrode. Some of these may result in a sudden drastic reduction of impedance; if impedance drops below 200 KΩ, tetrode should be recut and re-plated. Repeat D for each channel of each tetrode.

Record Impedances:

Initial (before passing any current)

	TT1	TT2	TT3	TT4	TT5	TT6	TT7	TT8	TT9	TT10	TT11	TT12	R1	R2	GROUND
1															
2															XXXX
3															
4															

Final

	TT1	TT2	TT3	TT4	TT5	TT6	TT7	TT8	TT9	TT10	TT11	TT12	R1	R2	GROUND
1															
2															XXXX
3															
4															

___ E) (optional) Check connections of stimulus and ground (for ground, check that all grounds connector pins are connected to each other, and to the bottom of the drive). You can use a regular volt-meter for this.

___F) Turn **OFF** the impedance meter and stimulator. With all tetrodes retracted, raise the drive up to make space above the well. Use pipette to empty the gold-plating solution (put back in the 'in use' bottle, we recycle it 2-5 times). Carefully place the rinse-well on top of the gold-plating well, fill with 70% ethanol. Extend all tetrodes half-way. Lower the drive and then soak the bundle, and extended tetrodes in 70% ethanol for 10 minutes.

___G) Raise the drive and **carefully** absorb excess fluids with the rolled tip of the corner of a KIM-wipe. Let dry for 20 mins. Retract tetrodes.

___H) Under the dissecting scope, adjust each tetrode so that it is flush with the bundle, then turn an additional **1/2 turn up**. *** Be careful not to pull tetrodes up too high as it may place too much tension on them and the suture string or break the silica on the board. Enter the orientation of each tetrode (you will need this in LabBook later)

TT1	TT2	TT3	TT4	TT5	TT6	TT7	TT8	TT9	TT10	TT11	TT12	R1	R2

___I) Reposition the tetrode cap, wrap cap with coban, insert the anchor screw, and set the drive aside until implantation.

___J) Cleanup the gold plating station. The gold solution is re-usable, so put it back in the vile. Use double distilled water and washout the well thoroughly. Blot extra water with Kim-Wipe.

___K) Weigh the drive. Record the total hyperdrive weight here: _____ g.

18) Final preparation (on implantation day, before starting with surgery).

___ A) Make sure you have the right holder/screw for the stereotaxic apparatus.
___ B) Put the drive upside-down, and using a 30G needle/syringe, put a small drop of pure mineral oil on the tip of the bundle. The oil should be absorbed within. Keep drive upside-down for 10 minutes.
___ C) Give the drive to the surgeon. Update this sheet with the name of the rat and surgery date (Page 1)

YOU ARE DONE!

Additional Suggestions and Comments

1. The most important factor in how well you can see what you are doing under the microscope is illumination. Make sure you have lots of light in your field of view under the scope.
2. Work in a clean area. This will actually help your drive come out cleaner and ensure it runs smoothly. It also saves time and supplies from becoming wasted.
3. Follow the checklist and ask others for advice.

Drive Notes

Appendix I

Tetrode Making Instructions

1. Equipment
   - Working microwire.
   - Automated tetrode spinner: (Neuralynx)
   - Alligator Clip style holder for tetrode ends (Alligator clip w/ heat shrink padding and magnet glued to the bottom side).
   - Heat Gun with J-clip attachment (Ace Digital)
   - Tetrode Scissors (NOT the final cut scissors).
   - Antistatic Storage Box
   - A pair of good, fine forceps

2. Instructions

A. Preparation: Check the working spool of tetrode wire (in Tupperware). Carefully transfer 640 cm (i.e. ~21ft, i.e. ~56 spool-turns) of wire from the stock spool to the working spool. Put the stock spool away.
B. Set-up the workspace (clean, comfortable…). Set the spinner on 'manual'.
C. Take the wire out of the Tupperware box extremely carefully. DO NOT touch the wire (hold it by the plastic supports on the end of the spool). Gently grab the free end of the wire and unroll the. Cut one length of microwire approximately **31.5 cm** long. If wire is mangled or looks damaged in any way, discard and replace it. If at any time there is a bend in the wire or tetrode, do not use it for loading a hyperdrive.
D. Bring two ends of the wire together making sure that the ends are even. Gently run wire through fingers in order to sick them together so they appear to be one wire with a small loop at the bottom.
E. Snip the wire in the middle of the loop and again fold the two ends together to form another loop in the wire. Again make sure that all four loose ends of the wire are even and together. Use the alligator clip to clip the four ends together. Try to clip 5mm or less from the bottom as clipping to high will make a short tetrode. If any gaps between the adjacent wires appear, unclip, smooth the wires, and re-clip. If any bends or kinks appear, discard and start over.
F. Loop the wire over the suspension bar. Lower the assembly onto the spinner.
G. The clip should not touch the spinner, but should be close to it – approximately ½ cm above the surface. Turn the spinner on and clear the counter window. Begin turning in the clockwise (forward/right) direction and continue for **70 turns**. Twisting should be a smooth action. If the twisting becomes jerky or wild, the magnet is too high from spinner- adjust it. When you read 70 right turn, stop the spinner. Now unspin for **35 turns** in the counter clockwise (left) direction.
H. Ensure the J-clip attachment is on the heat gun and set it on 5 (about 950 degF). Position the heat gun next to the Tetrode such that the walls of the J-clip are evenly close to the wire. Move the heat gun along the length of the tetrode wires for **5 cycles at 2 cm/sec** (Up and down is considered 1 cycle). Heat the entire length of the tetrode, excluding the loop around the suspension bar. Also, do not lower the heat gun to where the clip begins. The goal is to fuse together the insulation between the wires without shorting them together. DO NOT put the heat gun directly in contact with the tetrode, as it will cut or burn the wires. The individual wires will need to be separated from the loop end later so do not heat it too high. Be sure to move the gun continuously to prevent over heating.
I. Turn off gun and set aside.
J. Holding the tetrode by the clip to support it, gently slide the tetrode off the suspension bar. A good tetrode will be somewhat rigid and should not bend. Holding the clip horizontally over a flat surface, carefully cut the tetrode off just above the clip.
K. Make sure the finished tetrode is straight (handle it by the loop end only to avoid bending the twisted end). It should be somewhat flexible, yet strong (a little stiff) due to twisting. A tetrode needs to be approximately 7.5 (+/- .5) cm in length from the top of the loop to the other end. If the loop looks to be an unusual size, then discard (or save as a dummy, in the dummy/practice box).
L. Repeat steps B through J for each tetrode (make about 18-20 tetrodes).
M. Place in Antistatic storage box with date and number of tetrodes. Keep clean and dry at room temperature. Do not store more than 25 tetrodes in one box. Make sure not to leave the alligator clip on the spinner when you are done… it demagnetizes.

Appendix II

Making Bundles

This procedure produces two 27.5 mm long, 14 cannulae bundles.
Safety: Always wear goggles when using the drill and diamond bit.

Supplies

1. 14 cannulae, stainless steel: 30 G (Thin wall)
2. Heat-shrink tubing: 1/16" dia.
3. Scissors and blade
4. Caliper (mm divisions)
5. Heat gun
6. Drill (preferably table-mounted)
7. Diamond wheel bit (use the one labeled 'in use')
8. Soldering iron and soldering holder (2 aligator clips)
9. Solder: DIA 020, CORE 66, alloy SN63PB37, Kester brand flux 331
10. Stainless steel soldering flux (use a plastic pipette and put ~1ml in a weight boat)
11. Distilled water
12. Petri dish, small cup or dish to contain distilled water
13. Safety goggles
14. Fine tip marker

Procedure

1. Cut heat-shrink tubing (use blade): three 10 mm pieces, two 6 mm pieces

2. Hold 14 cannulae together in a bundle. Slide one 10 mm piece of heat-shrink tubing over one end of the bundle. When the ends of all 14 cannulae are flush, apply heat to shrink tubing. This piece of tubing will hold the cannulae bundle together throughout fabrication. It will be removed later.

   NOTE: The cannulae should naturally move into the optimal space-saving configuration shown in figure 1; if not, remove heat shrink and repeat this step.

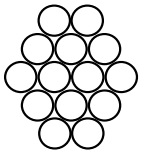


Figure 1 Cross-section of cannulae
3-4-4-3 bundle configuration as viewed from end

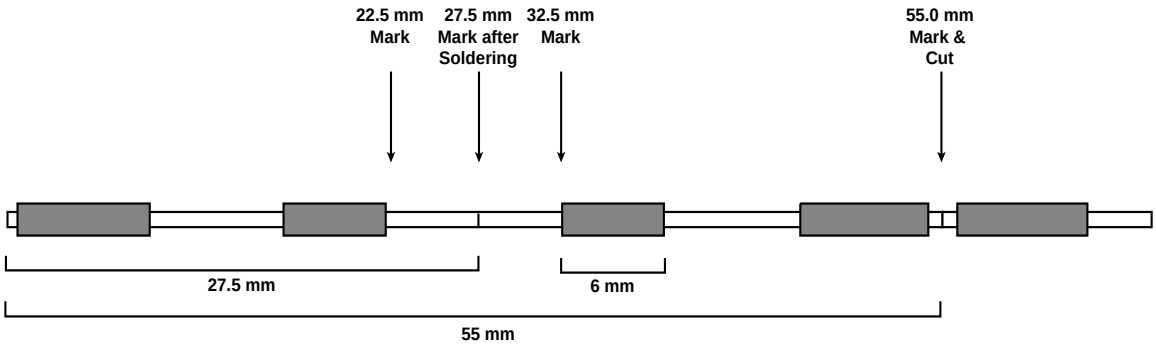


Figure 2 Marks for heat-shrink and cutting

3. Using the calipers and fine tip marker, mark 22.5 mm and 32.5 mm from the heat-shrink bound end of the bundle, as shown in figure 2.

4. Align each piece of 6 mm heat-shrink tubing with the appropriate mark, as shown in figure 2. Attach each piece independently by applying heat.

   NOTE: Use a clean tipped iron. First, partially shrink the piece of tubing to allow for adjustments in alignment. Once aligned satisfactorily, apply more heat to shrink further. Applying too much heat, however, can melt the tubing.

5. Using the calipers and fine tip marker, mark 55 mm from the heat-shrink bound end of the bundle.

6. Attach the two remaining 10 mm pieces of heat-shrink tubing on either side of this line, allowing a 1-2 mm gap between them with the line still visible, as shown in figure 2.

7. **Safety goggles!** Use the diamond wheel bit to cut the bundle at the 55 mm mark. Clamp the dremel, attach the diamond wheel. Set the dremel to about 5.

   NOTE: The length of the cannulae may force the cut to be slightly angled. If ends look uneven, sand gently with the flat surface of the diamond wheel bit. Unfortunately, sanding often clogs the cannulae with metal shavings. When sanding, hold bundle in a fixed position (do not turn) in order to minimize this clogging.

8. Set soldering iron at maximum. Solder the middle 10 mm segment of the bundle, between the two 6 mm heat shrink pieces. Leave a small space (1 mm) between the solder and each piece of heat shrink in order to prevent melting the heat-shrink with the soldering iron, as shown in figure 3. Apply an even coat of solder on all sides of the bundle.

   NOTE: To improve the solder's adherence to the stainless steel cannulae, apply a generous drop of flux on the bundle surface before soldering. You can also use a Q-tip, dip the cotton in the flux, then apply the flux directly on the solder as you are soldering (it will make a pchhhhhit sound!). Alternate flux and solder throughout soldering process. Also, keep the soldering iron tip clean by removing excess solder on a damp sponge. Clamp the bundle in the alligator clip stand while soldering keeps both hands free in order to better apply the flux and solder. Be careful with the flux because it contains highly corrosive acid. Use water to wash hands, work surface, and any tools that contact the flux.

9. Rinse soldered area with water to remove flux. Rub with emery cloth to remove black spots. Check that solder has evenly covered the middle segment without any gaps, bubbles, or divots. Continue soldering if necessary.

10. Using the calipers and fine tip marker, mark 27.5 mm from the heat-shrink bound end of the bundle. This should be the exact center of the bundle.

11. Use the diamond wheel bit to cut the bundle at the 27.5 mm mark. If ends look uneven, sand gently with the flat surface of the wheel bit.

12. Clear the tip of the bundle (solder side) with a 30G needle, under the dissecting scope.

13. Place the two finished bundles in a small dish of distilled water and allow them to soak for 10 min to 1 hour. Then transfer to the vial 'bundles'.

Notes: To cut individual cannula or the bundle, you may use a piece of wood (2 by 4). Use transparent scotch tape to hold the cannulae/bundle, and cut through the tape. Practice a bit to make sure the cut is nicely vertical.

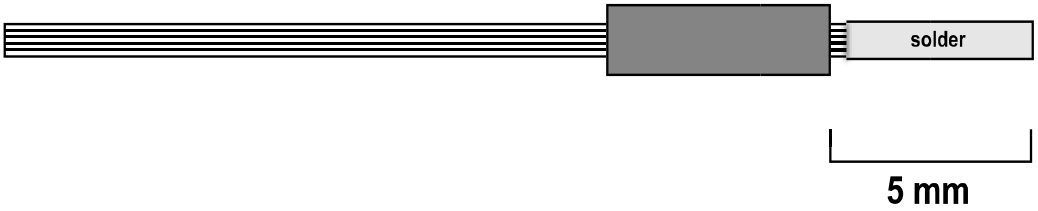
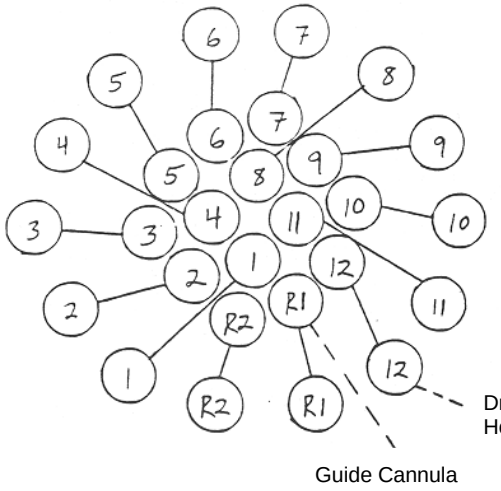
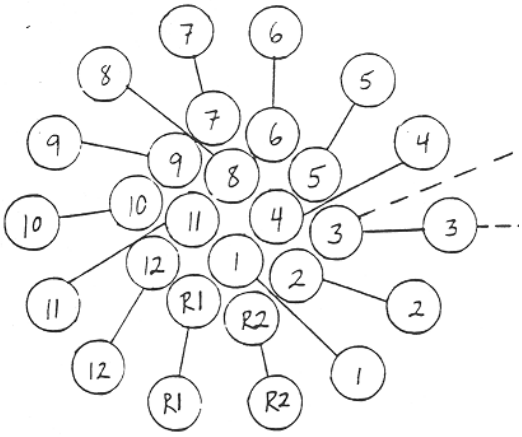


Figure 3 Single finished bundle after soldering and cutting

Bundle Loading Diagram

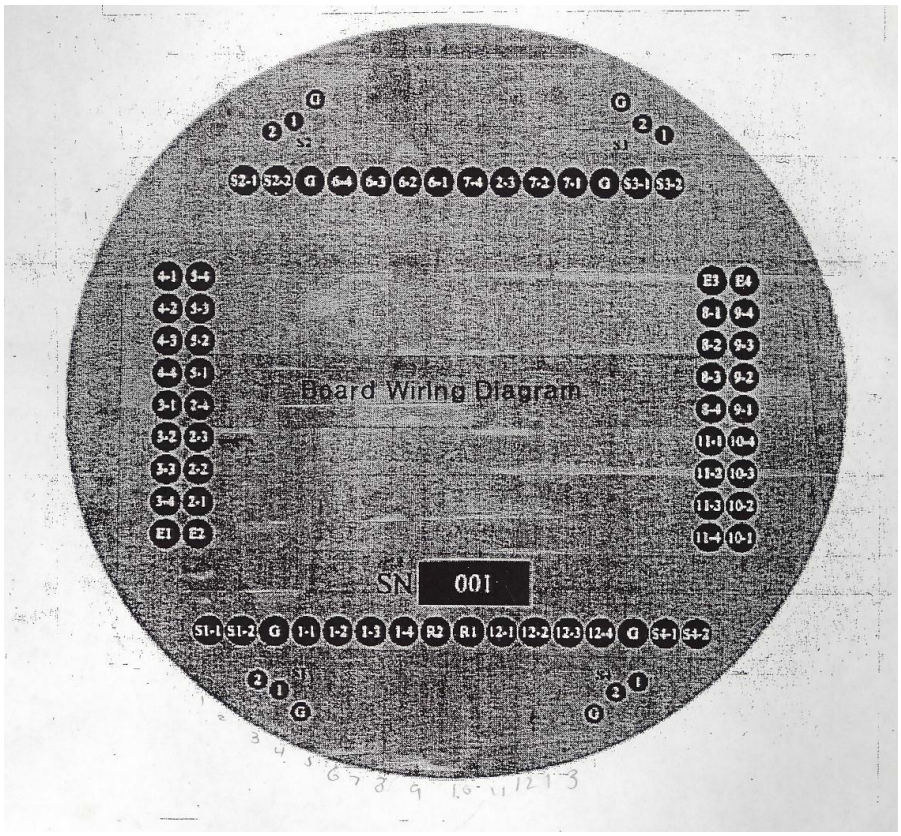


Top view of bundle loading pattern



Bottom view of bundle loading pattern

Connectors pin assignments



Tools/Parts	**Quantity**
70% Ethanol	A/R
Anchor Screw (Stainless Steel, 0-80 x 1/4" Socket Head; Small Parts)	1
Antistatic Fluid	A/R
Bone Wax (Sterile)	A/R
Bundle (see Appendix II) 30G tubing from Small Parts, Inc. (.012 inch O.D., .006 inch I.D.).	1
Bundle Loading Tool	1
Cactus Needles, Dried	A/R
Collet Ferrule (Kopf)	1
Collet Nut (Kopf)	1
Connector Board (Tetrode Interface Board Assembly; Neuralynx)	1
Core (Assembly; Kopf)	1
Crimping Tool (a blunted wire cutter works well)	1
Distilled Water	A/R
Drill with Diamond Wheel Disk Bit (Handheld drill best)	1
Dust Cover (Assembly; Kopf)	1
Forceps (Dumont #5; Fine Science Tools)	2+
Gold Plating Solution (Sifco Selective Plating)	A/R
Heat Shrink Tubing, 1/16" diam.	A/R
Heat Shrink Tubing, 3/32" diam.	A/R
Hex Wrench (0.05")	1
Hot Air Gun (HL 1802 E manufactured by Steinel recommended)	1
Hyperdrive Holder (Kopf) with Adjustable Support Armature	1
Impedance Tester (manufactured by BAK)	1
Impedance Testing Facility (Adjustable-height drive holder over fluid well with protruding wire)	1
Leg (Drive Linkage Assembly; Kopf)	14
Needle Holder (Fine Science Tools)	1
PC Board Nut (Kopf)	1
PCB Nut Tool (Assembly; optional; Kopf)	1
Pliers (pad with folded tissue)	1
Post (Assembly; Kopf)	1
Post Screw (Stainless Steel, 0-80 x 1/4" Socket Head; Small Parts)	1
Razor Blade (single edged) or Scalpel Blade	1
Resistors, Set of (<1 - 5 MΩ range)	1
Scissors, Fine Surgical (Fine Science Tools)	1
Screwdriver, Small Flathead	1
Shroud (Kopf)	1
Shroud Screw (Stainless Steel, 0-80 x 1/8" Flat Head; Small Parts)	2
Silica Tubing (125 OD/ 065 ID) www.Polymicro.com (TSP065125)	A/R
Silicone (or Teflon) Grease (Medical Grade; Fine Science Tools)	A/R
Spatula (small with flat spade-shaped end)	1
Stainless Steel Tubing, 23 G (Small Parts)	A/R
Stainless Steel Tubing, 30 G (Small Parts)	A/R
Steel Music Wire, 0.005" Diameter (Small Parts)	A/R
Steel Music Wire, 0.006" Diameter (Muller Music Center)	A/R
Stereoscopic Microscope with Bright Light Source	1
Stimulus Isolator (manufactured by World Precision Instruments)	1
Superglue, Extra Thick (Maxi-Cure Cyanoacrylate; HobbyTown USA 3700 E. Speedway, Tucson, AZ)	A/R
Superglue, Super Thin (Insta-Cure Cyanoacrylate; HobbyTown USA 3700 E. Speedway, Tucson, AZ)	A/R
Suture	A/R
Syringe Needles (23 guage)	2
Teflon Coated Stainless Steel Wire, 0.0045" Coated Diameter	A/R
Tetrode Cap (Kopf)	1
Tetrode Cover (Kopf)	1
Tetrode Scissors (Tungsten Carbide-edged Surgical Scissors; Fine Science Tools)	1
Tetrode Turning Tool (Drive Tool Assembly; Kopf)	1
Tetrodes (see Appendix II: Tetrode Manufacture) Kanthal Palm Coast (KPC# PF228/591) RO-800	14+
Vernier Caliper (mm divisions)	1

Ground screws instructions

Equipment:

- Stainless steel, Teflon coated: 0.003" bare, 0.0055 coated (AM-systems)
- Connectors: use 30G tubing, 5-7mm long. Use a 30G needle to clear out one of the extremities.
- Scalpel blade (#11)
- Solder: '44' Rosin. core alloy SN 63pb37. Dia 0.031, core 66
- Flux: liquid flux, Use a 30G needle and a 1ml syringe to dispense (need very little)
- Ground screws
- Black rubber mat (to cut), forceps.
- 1 Alligator clip (smooth kind, copper, with shrink wrap) and some bee wax or clay.
- Work under the dissecting scope. Bring the soldering iron near the scope.

Procedure:

A. Cut wire ~5.0 cm length using a scalpel blade.
B. Strip off ~3mm of Teflon on both ends using the scalpel blade on the black rubber mat. Lay wire flat, then gently rotate the wire with the blade to mark a circle. Then slide the Teflon off the tip. Don't push too hard, or you will weaken the wire and risk to break it later.
C. Place a screw in an alligator clip (between shrink wrap, horizontal). Use bee wax or clay to stick the clips on the table.
D. Take one end of the wire, and wrap it at least once around the screw, just under the head. The curved Dumont Forceps 7 are good for this.
E. Place a drop of flux just under the screw head.
F. Use the solder (very clean iron tip) and make a drop just below the screw head. Very little solder is needed. Try to make it as small as possible (Fig 1).
G. Slide the other end of the wire in the 30G connector. The Teflon should barely fit (make it go in > 1mm). Crimp the connector over the bare end (make sure the crimping is on the bare wire, not the Teflon). Using a needle-nose plier and the crimping tool, make a small bend at the tip of the connector (Fig 2). Don't crimp, just bend.
H. Use fine sand paper (i.e. P100) to polish the tip of the connector, to ensure a good fit. Try and see how the connector fits at the bottom of the hyperdrive.
I. After all of them are done, tug gently to make sure everything holds. Use a voltmeter to make sure there is a connection.
J. Store for later use.

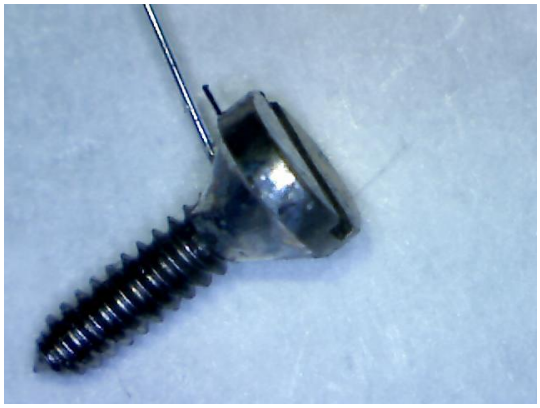


Fig1: Screw, after soldering

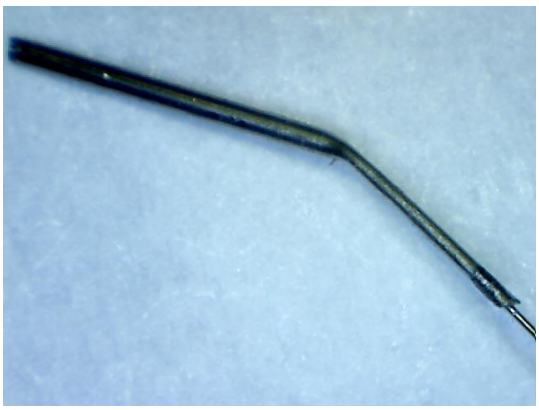


Fig 2: Connector after crimping and bending

EMG - LFP instructions

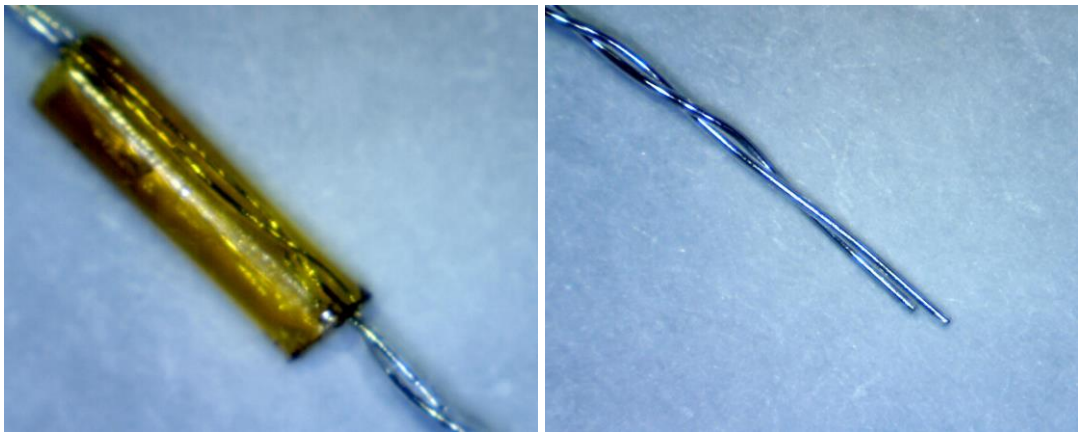
In some cases, you might want to add independent channels (up to 4) for additional LFP and/or EMG data. Your drive has to be configured for these extra channels (i.e. the connector cannulae at the bottom of the drive should be connected to the appropriate Ex pins on the EIB board (step 7).

EMG:

- Cut a 7 cm long piece of EEG wire (0.003” bare, 0.0055” coated).
- With a sharp scalpel blade strip off ~3mm of Teflon on both ends.
- Obtain a 5-7mm long piece of 30G cannula (as in ground screws), and crimp it to one end.
- Check that the 30G cannula fits the hole at the bottom of the drive. Bend and polish if necessary.

LFP:

- Cut a 5 cm long piece of EEG wire (as above).
- Cut a 5 mm piece of polymide tubing (wide enough so that the wire fits inside).
- With a sharp scalpel blade strip off ~3mm of Teflon on one end.
- Obtain a 5-7mm long piece of 30G cannula (as in ground screws), and crimp it to the bare end.
- Fit the coated end of the wire in the polymide tube and let it stick out (5 + X + 2) mm (where X is the desired depth of the LFP)
- Apply thick glue to the polymide/wire junction (the glue should go in by capillary action)
- Let it dry overnight.
- With a sharp blade cut 2 mm from the tip (i.e. the tip should be at least (X+5) mm).
- Check that the 30G cannula fits the hole at the bottom of the drive. Bend and polish if necessary.
- Note: when implanting, the piece of polymide tubing will be submerged in acrylic.



LFP wires. **Left**: Polymide sleeve, glued to the wire. **Right**: Staggered dual LFP wires (about 0.5 mm).