Head-Mounting Transmitter Surgical Implantation Protocol for Recording EEG and EMG

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Disclaimer: This is a draft protocol, as the full procedure has not yet been tested with our system. The portion of the protocol covering HMT attachment and EEG recording has been verified. However, the EMG electrode implantation method-specifically for placement in the neck-as not yet been validated with our system.

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1. Introduction

Disclaimer: These protocols serve as a general guide. Surgical and animal welfare requirements vary across institutions. Always consult your institution's veterinary staff to ensure compliance with local guidelines.

Warning: Bare, unwrapped HMTs are vulnerable to static electricity. Do not carry bare HMTs around while wearing rubber shoes, wool sweaters, or any other static-generating item of clothing.

Purpose: The purpose of this protocol is to detail the procedure for attaching the Head Mounting Transmitter to the head of a mouse or rat to record EEG and EMG.

Materials

- Electrode Interface Fixture (EIF)
- Head-Mounting Transmitter (<u>HMT</u>)
- Anesthetic setup (e.g., 3-4% isoflurane, ketamine, eye lubricant, antibiotics)
- Sterile surgical tools (suture kit, forceps, scissors, scalpel, burr drill)
- Pointed syringe needle (use ~23G for thinner C leads, ~20G for thicker B leads)
- Dental cement
- Vetbond biocompatible cyanoacrylate adhesive
- Hydrogen peroxide (6% v/v diluted)
- Kwik-Cast biocompatible silicone sealant
- Ethanol (70% or 100%)
- Stereotaxic apparatus
- Sterile marker
- Medical tape, clear plumber's tape, white Teflon tape
- Wooden applicators
- Cotton applicators (Q-tips)
- All necessary electrode materials

Related Protocols

- <u>Cleaning and Storage</u> of telemetry devices
- X-Electrode Crimp Contact Tutorial (Video)
- J-Electrode Implantation (Visual Guide)
- Silicone Removal from Leads
- HMT Implantation, Kate Hills
- <u>Telemetry System Set-Up</u>

2. Device Preparation

- 1. Ensure the EIF and electrodes are pre-assembled.
- 2. Prepare the EMG lead and electrode:
 - Ensure the EMG lead, which will be soldered to the EIF, has 3-4 mm of exposed coil wire at its terminus. You can strip additional silicone from the lead if necessary (see our <u>lead stripping video</u> for guidance).
 - Confirm that the exposed coil wire can pass through the selected syringe needle.
- 3. Sterilize all tools and materials prior to use. You can soak devices in 70% or 100% ethanol for 10 minutes, but do not exceed 60 minutes.
- 4. Calibrate and check stereotaxic apparatus.

3. Animal Preparation and Anesthesia

- 1. Weigh the animal.
- 2. Anesthetize the animal according to protocol provided by your institutions guidelines. Perform toe-pinch method to ensure proper anesthesia depth. Administer pain medications (consult your institutional guidelines for guidance on pain medication protocols).
- 3. Place the animal on a heating pad or temperature-controlled surgical platform.
- 4. Place the animal in a stereotaxic stand, connect to anesthetic, and apply eye lubricant.
- 5. Disinfect the surgical site.

4. Head Incision and Exposure



Step 4: head incision and scalp exposure.

1. Using forceps and scissors, lift the scalp and remove a patch of skin to expose the top of the skull. This is where you will mount the EIF and insert the EEG electrodes.

2. For EMG recordings:

- Extend the head incision a few millimeters beyond the skull to expose the trapezius muscles on the neck. Do not make a second incision-simply extend the original incision.
- Expose both trapezius muscles, as the electrode will be implanted through both.
- 3. Apply Vetbond to the skin edges to prevent bleeding and to secure the skin in place. Avoid getting Vetbond in the eyes and on the exposed skull as you will be cleaning it shortly.

5. Skull Cleaning

- 1. Clean the skull surface thoroughly using a bone scrapper to remove the connective tissue.
- 2. Optional: apply diluted hydrogen peroxide (6% v/v) to the skull and immediately dry it. This highlights any remaining connective tissue on the surface of the skull that can be removed using a bone scrapper.

6. Burr Hole Mapping and Drilling



Step 6: Burr hole mapping.

- 1. Glue the skin of the scalp in place with Vetbond to prevent bleeding and keep skin retracted for surgery.
- 2. Zero the stereotaxic stand using bregma.
- 3. Using your stereotaxic apparatus, locate the desired burr hole coordinates, and mark them on the skull with a sterilized marker.
- 4. Carefully drill burr holes at marked positions. Stop until there is a thin layer of bone covering the dura. Use a sterile syringe needle or tweezers to pock through to the dura.
- 5. If using set screws to anchor electrodes, confirm their fit in the burr holes.
- 6. Dab excess fluid with sterile cotton.



Step 6: Testing the fit of the set screws.

7. EIF Connector Placement



Step 7: EIF lowered to just above the scalp in order to measure and mark required lead lengths.

Note: The EIFs are not reusable once implanted. The HMTs, however, are reusable.

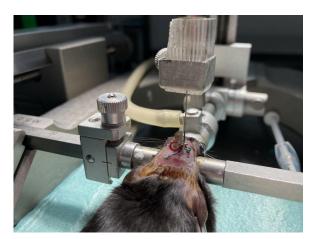
- 1. Secure the EIF in the stereotaxic arm.
- 2. Align the EIF:
 - Anterior-posterior axis straight
 - Pins facing left (antenna should point backward)
 - For EMG recording: Ensure the EMG lead is in the most posterior postion on the EIF, to reduce the distance it has to travel to reach the neck muscles.
- 3. Lower the connector such that the bottom of the connector just touches the skull.
- 4. Measure, mark, and trim silicone leads.
- 5. Remove the EIF from holder if needed, to adjust leads. Cut and strip the ends of the leads as the <u>electrode implantation protocol describes</u>.
- 6. Remount the EIF to the previous positions (i.e., touching the skull) and apply a small amount of dental cement under the connector. This is to secure the EIF in place on the skull. Precaution must be taken to avoid the flow of the detal cement into the burr holes.

8. EEG Electrode Placement



Step 8: Surface electrodes and accompanying set screws implanted, depth electrode not yet implanted.

- 1. Insert electrodes into burr holes-surface electrodes first, then depth electrodes. For more information on inserting our different types of electrodes, see our <u>Electrode Implantation Protocol</u>.
- 2. Control bleeding with cotton applicators.
- 3. Wrap excess leads around the EIF to reduce protrusion.



Step 8: All EEG electrodes implanted and in place. Vetbond coating over set screws.

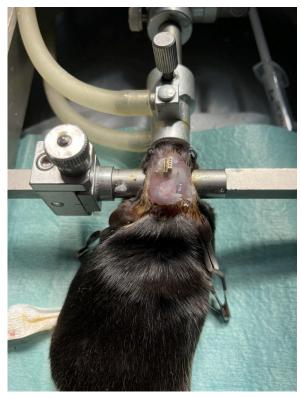
9. EMG Electrode Placement

- 1. Identify the top of the trapezius muscles at the base of the skull and locate the midline between them.
- 2. Use a pointed syringe needle to create a ~2 mm horizontal tunnel through the trapezius muscles, try to go through both muscles. Begin the insertion on the side of the neck opposite the EMG lead, so that the hub (base) of the syringe needle is on the side of the neck furthest from the lead connection on the EIF, also known as the "exit" of the tunnel. This orientation allows you to remove the needle later by gripping the hub while holding the wire in place.
- 3. The needle does not need to go more then a few mm deep into the muscle. It must go deep enough that it can not easily be ripped out of the skin. The needle should create a "tunnel", meaning it should have both and entrance and an exit into the muscle.
- 4. Control bleeding with sterile cotton if needed.
- 5. Leave the needle in place.
- 6. With the syringe still in the muscle, insert the stripped coiled wire (e.g., lead with <u>A-Coil terminus</u>) of your EMG lead through end of the needle. Thread the wire through until it exits from the other end of the muscle tunnel, leaving 1-2mm exposed and on top of the muscle.

10. Securing the EMG Electrode

- 1. Ensure 1-2 mm of the bare wire protrudes from the muscle on the exit side before removing the needle.
- 2. Carefully remove the needle while keeping the wire in position. It is helpful to have the wire extend past the end of the needle a small amount, so you can grip it with tweezers while removing the needle.
- 3. Once the needle is removed, use tweezers to attach the OSI-provided wire cap to the exposed end of the wire at the exit of the tunnel. The wire cap should cover all of the exposed coiled wire that sits on the muscle. The purpose of the cap is to insulate the exposed electrode wire, reducing the risk of artifacts in the EMG signal.
- 4. Optional: Use a small amount of Kwik-Cast sealant to secure the cap. Be careful not to allow any sealant to drip into the muscle tunnel.
- 5. Place a few sutures over the capped wire, at the exit of the tunnel, to anchor it in the muscle and hold the cap in place on the exposed wire.
- 6. Suture the subcutaneous lead at the muscle entry point to prevent movement of the implanted wire.
- 7. Optional: Apply a small amount of dental cement to the portion of the lead before it enters the muscle to reduce movement under the skin. Avoid excess cement, which may restrict neck mobility.

11. Securing EIF Implantation



Step 11: EIF and EEG electrodes implanted and attached, then covered with dental cement.

Note: Do not close the animal up by suturing the skin back together on the head, rather, we reccommend creating a head fixture out of dental cement to secure the EIF and cover the electrodes. You will not attach the HMT until the animal recovers from surgery. This means the EIF will be sitting exposed on the animals head for a few days and needs to be covered in some way to protect from debris and damage.

1. EMG electrode and lead:

- Ensure the electrode is secure and all adhesives are fully cured.
- Verify there is some slack in the EMG lead to accommodate head and neck movement during recovery.
- Close the incision over the EMG site by suturing the skin up to the base of the skull. While you can use dental cement to secure the EMG electrode implantation, dental cement on the neck can restrict the animals movement.
- 2. Apply dental cement to secure electrodes and connector base.
- 3. Cover exposed leads and skull surface-avoid covering the connector top.
- 4. Allow cement to fully cure and trim excess.

5. Cover the EIF top with tape/parafilm and optionally Kwik-Cast to protect it from debris and dislodging while the animal recovers and the HMT is not yet connected.

12. Surgical Recovery

- Allow several days of recovery in a qiet, clean environment.
- Monitor for pain, distress, weight loss, or infection.
- Consult your vet for protocols on antibiotic treatments.
- If the dental cement cap detaches or electrodes become exposed, consult a vet immediately-euthanasia may be required.

13. HMT Attachment



Step 13: The HMT fully attached to a mouse head, with a Kwik-Cast sealant.

- 1. Discharge you body of any electric static before handling the bare HMT, if possible.
- 2. Prepare the HMT:
 - Confirm the HMTs connection to the telemetry system.
 - Fold the HMT in half as shown here.
 - Wrap the HMT in 2-3 layers of teflon tape, do not cover the connector.
 - Wrap the HMT in one layer of transparent tape and one layer of clear plumbers tape.
- 3. Briefly re-anesthetize the animal.
- 4. Attach the HMT to the EIF connector.
- 5. Secure the connection with Kwik-Cast.
- 6. Confirm electrical contact and alignment.
- 7. Monitor for scratching or attempts to dislodge the device.

14. HMT Replacement

Note: Replacement can be done with or without anesthesia. Some users prefer anesthesia to reduce stress.

- 1. Carefully remove Kwik-Cast from the old HMT.
- 2. Gently disconnect it from the EIF.
- 3. Attach the partner HMT (identical configuration and channel) to the EIF.
- 4. Secure the new HMT with Kwik-Cast
- 5. Return the animal to housing monitor its recovery.
- 6. Unwrap the original HMT and remove the battery by gently pushing it out with a wooden applicator.

15. Battery Replacement

Note: Only insert the new battery into the HMT right before surgery, otherwise it can damage the HMT and drain the battery.

- 1. Load a new battery just before use.
- 2. Clean the HMT before use:
 - Scrub with hot water and toothbrush
 - Soak briefly in ethanol make sure to thoroughly clean it.
 - Dry with compressed air (20 psi) if available
- 3. Wear sterile gloves when loading the new battery.
- 4. Wrap the HMT with Teflon and transparent tape.

Notes and Best Practices

- Maintain sterility of all tools and materials.
- Use minimal cement beneath the connector.
- Avoid getting Vetbond on fur or unintended skin areas.
- Carefully document electrode coordinates and placements.
- Do not co-house animals with HMTs; they may remove each other's transmitters.
- EMG electrode and lead:
 - Ensure the electrode is secure and all adhesives are fully cured.
 - Verify there is some slack in the EMG lead to accommodate head and neck movement during recovery.
 - Close the incision over the EMG site by suturing the skin up to the base of the skull. While you can use dental cement to secure the EMG electrode implantation, dental cement on the neck can restrict the animals movement.