In Vivo Carbon Fiber Electrode Thread (CFET) Implantation and Testing Procedures

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We use this protocol and it's working

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ABSTRACT

Procedures for implanting and testing CFET devices in rats are described.

1 All procedures involving animals were approved by the Committee on Animal Care at the Massachusetts Institute of Technology and were performed strictly following the U.S. National Research Council Guide for the Care and Use of Laboratory Animals. Long–Evans male rats were used for tissue retainment testing (n = 2) and dopamine recording (n = 1).

2 Rats were first anesthetized (1.5–2.0% isoflurane, 1 L/min oxygen) and were administered an analgesic (Meloxicam, 2 mg/kg) subcutaneously.

3 Rats were fixed in stereotactic frames (Stoelting, 51600, 51449) that carried vertical micromanipulators used to navigate to targeted brain areas.

4 A sterile field was created on the skin above the cranium.

5 The skin was incised and retracted to expose the cranium.

6 The CFET array was targeted to be placed in the right striatum (anteroposterior [AP] +0.5 mm, mediolateral [ML] +3 mm).

7 The reference Ag/AgCl electrode, made of an insulated silver wire (A-M Systems, 787000 and 786000) with an exposed 0.5–1 mm tip chlorinated in bleach overnight, was targeted to be placed in the contralateral hemisphere, in the cerebellum.

8 In addition, 5 – 10 bone screws (Stoelting, 51457) were placed on the circumference of the exposed calvarium in order to provide additional connections for electrical grounding as well as

to secure subsequent cement (for the tissue retainment tests).

9 A bare stainless-steel wire (A-M Systems, 792900) was wound around 2 or 3 screws to serve as an electronic ground connection.

10 The dura mater overlying the site calculated to be at the A-P level of striatum was removed with the bent tip of a 32G hypodermic stainless-steel needle.

11 Afterwards, the micromanipulator was manually driven at a rate of 0.1 – 0.3 mm/s to lower the maltose coated CFET tips from the starting position.

12 After the maltose coated CFET tips approached just above the brain surface, a small amount of saline was applied to dissolve ~ 1 mm length of the basal portions of the maltose coating of the CFET tips.

13 The non-maltose-coated portion of the CFETs was then lowered into the brain.

14 The penetration of CF into the brain was supported by the rigid maltose coating above the basal portion.

15 Afterwards, the process of saline dissolving and CFET array lowering was incrementally repeated until the array was lowered to a final depth of 4 – 6.5 mm dorsoventral (DV) relative to the cortical surface.

16 The implanted array was connected to the FSCV headstage for dopamine recording experiments.

17 For brain fixation and CFET sectioning experiments, the implant was secured to the skull by applying acrylic cement (Ortho-Jet, 0206) over the array and the skull. This was followed by protocols described in “Histology and Retention of Implanted Electrodes in Fixed Tissue”.

18 FSCV recording began once the array reached its targeted depth and continued for a period of 15 – 30 minutes until the background current had stabilized (i.e., < 1 nA change in a 30 s window for most of the functional channels).

19 3 mg/kg raclopride and 15 mg/kg cocaine were administered to the anesthetized rat intraperitoneally to induce dopamine transient signaling in the striatum.

20 Dopamine transients were observed five minutes after the injection of raclopride and cocaine. Recording continued for 2 hours after the drug administration.

21 Chemical selectivity to dopamine was confirmed by the presence of sharp current peaks at the known redox potentials for dopamine (~ –0.2 and 0.6 V) as well as by the selective action of the administered drugs acting at dopamine receptors and transporters.

22 The inserted depths were DV 6.5 mm for the functional recording experiments, and 5.5 mm and 4 mm DV for the tissue retainment studies.