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Chronic lens implantation and inertial sensor holder placement

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Cristina Alcacer[1]

[1]Alcala University

Christine Weber-Schmidt
Allen Institute

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We use this protocol and it's working

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Abstract

This procedure is meant to take place 3 weeks post lesion and viral injection.

Guidelines

All procedures were done in aseptic conditions.

Materials

-Stereotaxic surgery set up required (this protocol uses Kopf instruments)
-Gradient index (GRIN) lens (diameter: 1 mm, length: 4 mm; Inscopix)
-Surgical Tools

Before start

Mice were kept in deep anesthesia using 1-3% isoflurane and oxygen (at 1L/min flow rate) and mouse body temperature was maintained at 37 degrees Celsius using a temperature controller (ATC1000, World Precision Instruments). Surgeries were performed on a stereotactic frame (David Kopf Instruments, Model 962LS) with a mouse adaptor (David Kopf Instruments, Model 923-B Mouse gas anesthesia heads holder).

Chronic lens implantation Surgery

1 Anesthetize mouse with isoflurane.

2 Remove hair/shave head if needed and transfer mouse to stereotaxic apparatus and secure the head and keep mouse anesthetized for length of procedure.

3 Disinfect surgical area of the mouse head with 70% ethanol and iodine.

4 Make a small incision in the skin to expose the skull.

5 Carefully remove any connective and muscle tissue from the exposed area.

6 Level the skull surface to be at less than 0.05mm by comparing the height of bregma and lambda, and also in medial-lateral directions.

7 Remove skull over implantation site to fit implant diameter of 1mm at coordinates: AP= + 0.5, ML= -2.3, DV= 2.3.

8 Carefully aspirate 1.8-2 mm of overlying cortical tissue with a 30-gauge blunt needle and try to minimize bleeding before lens insertion.

9 Place lens and secure to the skull using superglue and a self-curing adhesive resin cement (Super-Bond C&B Kit).

10 Place a small screw on the surface of the skull.

11 Add a layer of resin cement to surround the lens and cover the screw to increase the lens bond to the skull and this will help to minimize motion artifacts during imaging.

12 On top of the resin cement, add a mixture of black Ortho-Jet powder and liquid resin (Lang dental, USA) until the lens is covered all around.

13 Add tape to the lens surface for protection post-surgery.

14 Inject carprofen (5mg/kg; 10uL/10g body weight) as an analgesia. Then inject 0.6 mL sterile glucose-ringer acetate (subcutaneously) to prevent dehydration.

15 Allow animal to recover for one week.

Baseplate attachment & Focal Plane Adjustment

16 One week post GRIN lens implantation, anesthetize mouse with isoflurane for baseplate attachment and focal plane adjustment.

17 Transfer mouse to stereotaxic apparatus and secure the head and keep mouse anesthetized for length of procedure.

18 Remove the tape from the GRIN lens carefully as to not scratch the lens.

19 Attach the microendoscope baseplate to the microendoscope and align to the lens on the animal then adjust the scope to the best focal plane to observe neuronal structures and blood vessels when present.
Note: the microendoscope with the baseplate are held with an adapted holder for the microendoscope which is attached to the stereotaxe arm

20 One the best focus is found, begin to secure the baseplate to the head cap by adding first a layer of self curing adhesive resin cement.

21 Add a second layer of black cement to permanently secure the baseplate to the head cap.

22 Carefully remove the microscope and attach a baseplate cover to the baseplate.

23 Now cement a holder or small connector for the wireless IMU on the back of the baseplate.

24 Recover the animal from the procedure.

25 The imaging field of view is inspected and allowed to clear for several days prior to imaging and behavioral experiments. Note-mice can have a bad focal plane, movement artifacts, or lack cells of interest. In these cases, mice are excluded prior to experimental data collection.