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Protocol status: Working We use this protocol and it's working

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Ouragel Application for Acute Electrophysiology Recordings

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ABSTRACT

This protocol describes the process of coating an implant with duragel in preparation for in vivo electrophysiology experiments. Duragel seals and protects the surface of the brain while also being soft enough for silicon probes to penetrate through, allowing recordings to take place without disturbing or damaging the brain. It is possible to perform acute recordings for up to two weeks after the duragel layer has been added.

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MATERIALS

PROTOCOL integer ID: 81475

Anesthesia and related:

■ Soflurane 1-3% Patterson Veterinary Catalog #07-893-1389

Tool Kit:

Equipment	
Iris forceps	NAME
Surgical tools	TYPE
Fine Science Tools	BRAND
11064-07	SKU

Consumable supplies:

- Duragel Ellsworth Adhesives Catalog #DOW DOWSIL™ 3-4680
- Alcohol prep pad VWR International Catalog #15648-907
- Pressn Seal Food Wrap Amazon Catalog #B001RHUP4Q
- Luer-Lock Syringe, 1ml (VWR BD-309628 or equivalent)
- 20G x 1 needle (VWR BD305176 or equivalent)
- Small weigh boats
- Double Cotton Tipped Applicator VWR International Catalog #89133-810
- KimWipes Fischer Scientific
- Sugi mounted sterile absorption spears (Fine Science Tools 18105-01)

Equipment:

Equipment

Model 1900 Stereotaxic Alignment Instrument

NAME

Surgical equipment

TYPE

Kopf

BRAND

Model 1900

SKU



Equipment

Leica M80 Stereo Microscope

NAME

Surgical equipment

TYPE

Leica

BRAND

M80

SKU

Equipment

Isoflurane with oxygen delivery system

NAME

Surgical equipment

TYPE

Patterson

BRAND

78914722

SKU

Equipment	
Isoflurane induction chamber	NAME
Surgical equipment	TYPE
Patterson	BRAND
78933385	SKU

Equipment	
Small Animal Temperature Controller	NAME
Surgical equipment	TYPE
CWE	BRAND
08-13000	SKU

- Fiber optic illuminator
- Ear bar Headframe clamp (0155-100-00 or equivalent)

Safety:

- Non-Sterile Gloves
- Disposable lab coat
- Face mask; 0.6 micron filter (optional)
- Biohazard sharps disposal container
- Biohazard waste disposal container

BEFORE START INSTRUCTIONS

Wear appropriate PPE including gloves, lab coat and optional face mask. Have biohazard waste and sharps disposal accessible.

Setup 1 Draw up the white and blue duragel components into separate 1 mL Luer lock syringes. Attach 20G needles 3m Note The duragel components should be dispensed into separate containers when the duragel first arrives. This makes it easier to draw up the two components separately without cross-contamination! 2 Turn on microscope lights and heating pad. 10s 3 Wrap heating pad in press 'n' seal. 30s 4 Anesthetize the mouse (see section Prepare the anesthesia system and anesthetize the mouse (all 3m procedures) in protocol General Setup and Takedown Procedures for Rodent Neurosurgery). 5 Transfer mouse to stereotax. 2m 6 If the mouse does not currently have a well and well cap, attach a well to the headframe at this time. 15m

Procedure 7 Clean the surface of the implant and the ground wire with an alcohol wipe. 8

1m

Using forceps, peel away the elastomer covering the implant.

10s

8.1 If the holes in the implant fill with CSF, gently dab it away with a Sugi or Kimwipe.

9 Using forceps, insert the ground wire into the designated grounding hole in the implant. 1m

Note

The ground wire should touch the surface of the brain, but should not penetrate or compress the brain.



Elastomer removed and ground wire inserted into grounding hole.

10 Prepare the duragel mixture. 5m

10.1 Add the white and blue components to a clean weigh boat, using a ratio of 35:50 white to blue. 1m

Note

This ratio can be achieved by counting drops as you dispense the components from the syringes. More blue will ensure the mixture cures a little tougher, and will not turn sticky in a way that can damage recording electrodes.

10.2 Use the broken end of a cotton swab to mix the two components until the mixture is uniform in 1m color.

11 Apply the duragel to the surface of the implant.

5m

Note

Depending on the implant, you may wish to tilt the mouse's head to ensure even coverage. For instance, for the dual hemisphere implant, turn the mouse's head 10 degrees to the right prior to application.

- Apply a few drops at a time with the broken end of a cotton swab (you can use the same swab you used to mix the components). Do not apply directly to the holes in the implant as this can cause bubbles. Try to apply near the holes and let the mixture flow into the holes.
- 11.2 If bubbles do form in the holes, try to push them out with the cotton swab.
- 11.3 Continue applying until all parts of the implant are covered with a layer of duragel at least 1 mm thick.





Implant covered with layer of duragel.

- 12 Wait to allow the duragel to cure. You can test this by checking the unused mixture in the weigh boat: tilt t 10m boat to see if the mixture is still flowing. If it's not, the duragel is cured enough. This usually takes around ten minutes.
- 13 Replace well cap and return mouse to its cage.

1m

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Note

Wait at least 24 hours before performing electrophysiological recordings, to allow sufficient time for the duragel to cure completely. Otherwise, there is a risk that the duragel will stick to the probes used for recording.

3m **Take Down** 14 10s Dispose of the syringes containing the duragel components in the sharps container. 15 Dispose of the elastomer implant cover and any other consumable that came in contact with the mouse in 30s the biohazard waste container. 16 10s Turn off oxygen and isoflurane flow. 17 10s Turn off heating pad and microscope lights. 18 2m Clean and disinfect stereotax, surfaces, and induction chamber with 70% ethanol.