

Video Article

# Application of Light-cured Dental Adhesive Resin for Mounting Electrodes or Microdialysis Probes in Chronic Experiments

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#### **Abstract**

In chronic recording experiments, self-curing dental acrylic resins have been used as a mounting base of electrodes or microdialysis-probes. Since these acrylics do not bond to the bone, screws have been used as anchors. However, in small experimental animals like finches or mouse, their craniums are very fragile and can not successfully hold the anchors. In this report, we propose a new application of light-curing dental resins for mounting base of electrodes or microdialysis probes in chronic experiments. This material allows direct bonding to the cranium. Therefore, anchor screws are not required and surgical field can be reduced considerably. Past experiences show that the bonding effect maintains more than 2 months. Conventional resin's window of time when the materials are pliable and workable is a few minutes. However, the window of working time for these dental adhesives is significantly wider and adjustable.

### Video Link

The video component of this article can be found at https://www.jove.com/video/249/

# **Protocol**

#### Step 1. Preparation of probe implantation

- 1. Anesthesia (not shown in the video)
  - Anesthetize each bird with an interperitoneal injection of a ketamine (12.5 mg/kg body weight) and xylazine (25 mg/kg) mixture to produce a steady level of surgical anesthesia.
- 2. Fix a head to a stereotaxic frame
  - Use Xylocaine<sup>(TM)</sup> jelly to reduce local pains of ear bars' pressure. The head of bird is fixed in a stereotaxic apparatus consisting of ear bars and a beak holder that holds the head at a  $45^{\circ}$  angle. During the stereotaxic surgery for implantation, warm the body of bird by surgical pad with thermal control (temperature =  $34\sim36^{\circ}$ C).
- 3. Disinfect surgical area
  - Disinfect the surgical field with isodine (TM) prior to the surgery.

# Step 2. Stereotaxic positioning

- 1. Reference point alignment
  - Align dummy probe (glass micropipette) to the Lambda (Y-point) and mark Y-point with the dye-filled pipette. Remaining Dextran-TMR of the last tracer study was used in the video.
- 2. Incision of the skin
  - Remove skin and expose the surface of the cranium. Then, clean the surface of the cranium with cotton swabs.
- 3. Positioning of dummy cannula (in horizontal direction)
  - Move the dummy cannula to the target site (A-P axis and Lateral coordination)
- 4. Small craniotomy and incision of dura mater.
  - Use a fine forceps for craniotomy and a fine injection needle (30 G) for incisions of the dura mater. Then, clean the surface of cranium.
- 5. Final positioning (implantation) of dummy cannula
  - Locate the tip of the guide cannula at 0.30 mm above the microdialysis probe-intended target region. Finally, clean the surface of cranium again, because a clean and dry surface is required for tough bonding.

# Step 3. Pretreatment of the cranial surface

(The similar procedure for "direct filling restoration" of the dental product shall be employed.)

1. Primer treatment

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- 1. Mix PRIMAR liquid A with B on a dish.
- 2. Apply the mixture on the surface of the cranium with a small brush.
- 3. Leave for 30 seconds.
- 4. Dry with mild air stream. (Do not wash.)

#### 2. Application of bonding agent

- 1. Apply the bonding agent on the surface of the cranium with a small brush.
- 2. Apply a gentle air stream. (Do not wash.)
- 3. Light-cure for 20 seconds.

#### Step 4. Mounting composite resin

- 1. Mount resin with plain plugger
  - If a Plain plugger is not available, a micro-spatula can be used instead.
- 2. Light curing
- 3. Incremental mounting
  - Incremental mounting is recommended to ensuring complete reaction of the resin.
  - For incremental mounting, apply bonding agent before every mounting.
- 4. Mold the composite resin and light-curing for final hardening.
- 5. End of the surgery

Clean the surgical area, apply antibiotic (Gentamicin ointment), and remove the animal from the stereotaxic flame. Then, keep the animal in a thermo-controlled chamber (temperature = 33-35° C) for post-anesthetic recovery.

As shown in the video, no abnormal behavior was observed in any of the animals on the next day after the surgery.

#### **Discussion**

Compared to the application of conventional self curing acrylic resins, the introduced technique has several important advantages:

#### 1. Direct bonding:

Self curing acrylic can be attached to the bone for a while but it does not adhere to the bone. Therefore, anchorage screws are required to fix a mounting base (See Figure 2A). In contrast, light curing resin can bond to the bone directly so screws are not required (Figure 2B). Therefore, size of mounting base can be reduced considerably. In addition, light curing resins also adheres to some metals, if you used "metal primer" with it. This feature of bonding to variable materials allows us a wide range of application.

Figure 2 shows an implantation using conventional self curing acrylics (in 2A) and an implantation using light curing adhesive resins (in 2B). In 2A, the anchor screw is used to fix the mounting base. In 2B, size of mounting base can be reduced considerably due to the fact that light curing resin adhere to the cranium and such screws are not

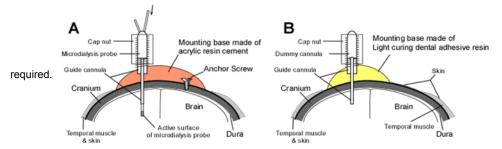


Figure 2 Please click here to view a larger version of this figure.

#### 2. Easy to handle:

Conventional resin's processing time window is relatively short, within few minutes. It has a high fluidity and it is not easy to handle at first. However, once chemical reaction started, reaction is accelerated by exothermal heat and resins become harder in short time. The processing time windows for light curing resin are significantly wider and adjustable. Basically, resins keep constant elasticity from the beginning to the moment when it is light cured.

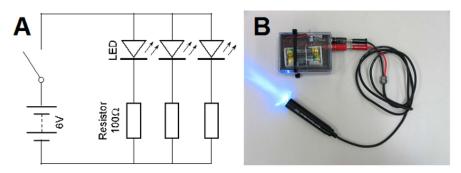
# 3. Reaction does not generate heat:

Curing reaction of self-curing resin is exothermal. When the mounting base is thick, this heat accumulates and may damage the tissue. Reaction of light curing resin does not generate heat and the material is biocompatible.

#### 4. Hand made LED light:

Cost of the material and special equipment like light unit can be a problem. In Figure 3, we introduce a hand made LED light unit which does not cost much. This hand-made light curing unit may be used as substitutes for expensive commercial unit.

Figure 3A shows a circuit diagram of light unit. Figure 3B shows three LEDs glued parallel to each other, and inserted into the holder of a white board marker.



High-power LEDs (e.g. OSUB5111A, OptoSupply, Hong Kong) are used. (Dominant wavelength = 470nm, Viewing angle = 15 degree)

Figure 3 - Circuit diagram (in A) and photograph (in B) of a hand-made light unit. Please click here to view a larger version of this figure.

This technique is applicable to chronic implantations of electrodes or microdialysis probe in other experimental animals such as fish, mouse, rabbit, cat and monkey.

## **Disclosures**

Experiments were undertaken in accordance with the guidelines for animal experimentation of the brain science institute, RIKEN (RIKEN-BSI) and the institute's animal ethics committee approved them.

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#### References

- 1. Adell A and Artigas F. In vivo Brain microdialysis: Principles and applications. In: Eds. Boulton A, Baker G and Bateson A. Neuromethods, Vol.32: In vivo Neuromethods. Totowa: Humana Press Inc, 1998: 325-57
- 2. Kendrick K. Microdialysis in large unrestrained animals: neuroendocrine and behavioral studies of acetylcholine, amino acid, monoamine and neuropeptide release in the sheep. In: Eds. Robinson T and Justice J. Techniques in the behavioral and neural sciences. Vol.7: Microdialysis in the Neuroscience. Amsterdam: Elsevier. 1991: 327-48
- 3. Oakley B and Schafer R. Experimental Neurobiology: A laboratory Manual. Ann Arbor: University of Michigan Press, 1978
- 4. Okumura T, Yamashita Y, Okanoya K and Tani J Function of the sensori-motor nucleus NIf in generation of complex syntactical song in the Bengalese Finch I. A Biological study. Soc. Neurosci Abst. 33. 2007, in press.
- 5. Vanderwolf C and Leung L. The relation of brain electrical activity to behavior. In: Eds. Boulton A, Baker G and Bateson A. Neruomethods, Vol.32: In vivo Neuromethods. Totowa: Humana Press Inc, 1998: 325-57
- 6. Yamashita Y, Okumura T, Okanoya K and Tani J. Function of the sensori-motor nucleus NIf in generation of complex syntactical song in the Bengalese finch. Proceeding of the 2nd International Symposium on Mobiligence. 2007: 101-104.