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Painting Neuropixels probes and other silicon probes for electrophysiological recordings

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[1](#) *Works for me* [dx.doi.org/10.17504/protocols.io.wxqffmw](https://doi.org/10.17504/protocols.io.wxqffmw)


ABSTRACT

This protocol is for consistently applying fluorescent dyes to Neuropixels probes and other types of silicon probes, which allows imaging and reconstruction of the electrode tracks in the brain in post hoc histology.

MATERIALS

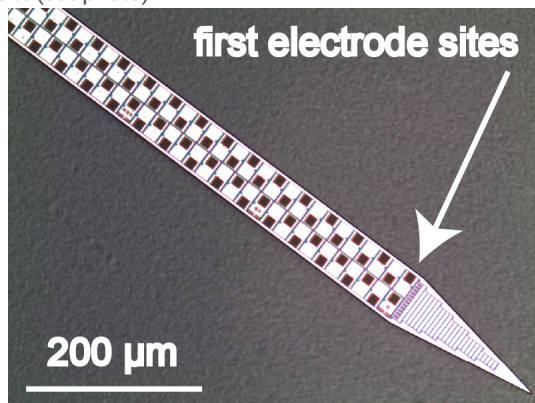
NAME ▾	CATALOG # ▾	VENDOR ▾
Microscope slides		
Coverslip		
CM-Dil	C7000	Thermo Fisher Scientific

MATERIALS TEXT

The CM-Dil should be stored in the freezer at -20°C. Before using, dissolve a 50 µg vial in 50 uL of pure ethanol. Tightly seal the vial after using to prevent the ethanol from evaporating. The remaining CM-Dil can be stored at room temperature until use. Protect from light.

- 1 Secure Neuropixels probes (shown) or other silicon probes onto a micromanipulator. The side with electrode sites should face up.
- 2 Secure a microscope slide onto a holder, and a clean coverslip at the end of the microscope slide. Place the coverslip attached to the microscope slide near to the Neuropixels probe (2-3 mm).
- 3 Using a micropipette, place 1 µL of CM-Dil solution close to the edge of the coverslip. To ensure a consistent labeling of the probe, make sure the pipette does not produce an air bubble in the drop of dye. The dye concentration should be 1 µg/µL CM-Dil in ethanol. The drop of dye will evaporate in ~30 s at room temperature, so it is important to complete step 4 quickly.

- 4 This step should be done under a microscope. Using the micromanipulator, dip the tip of the Neuropixels probe into the dye. Be careful not to get dye onto the electrode sites. For Neuropixels probes, the tip extends only about 150 μm from the first electrode site (see photo).



The electrodes sites should be visible under the

Neuropixels probe shank showing the electrodes sites and the tip.

microscope. The experimenter should be familiar with the setup to comfortably complete this step in considerably less than 30 s.

- 5 Wait until ethanol evaporates and dye dries onto the tip of the probe, around 30 seconds for ethanol-based CM-Dil.
- 6 Using the micromanipulator, slowly retract the probe away from the coverslip and dismount. One should be able to see dried pink dye on the tip of the probe. If not, repeat painting the probe.



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