

## **Paint-fill technique**

### Equipment

- 1) Manual manipulator right hand M3301R and a broad steel base for mounting manipulator, World Precision Instruments Tel. 941-371-1003
- 2) Hamilton 80601 710LT syringe 100ul
- 3) 19 gauge needle
- 4) Glass capillary tubing. Borosil 1.0x0.75 mm ID/fiber catalog # 30-30-0 from FHC Tel:207-666-8190
- 5) pipet puller
- 6) methylsalicylate, glycerol
- 7) 60mm glass petri dish with elastomer (follow instruction in Kit, add charcoal to darken the elastomer)
- 8) SV11 dissecting microscope with fiber optics
- 9) Clay Adams, Division of Becton Dickinson, New Jersey #7420 Intramedic polyethylene tubing ID .86mm.
- 10) 184 silicone elastomer kit from Dow Corning Corporation, Midland Mich. World Precision Science
- 11) Exterior Alkyd gloss white paint or White out fluid. The paint or White out is not soluble and is a suspension. If solution is clogging your injection needle, quick spin to remove the larger particles and use the supernatant.

### For 100 ml Bodian fix:

5ml glacial acetic acid

5ml formaldehyde (straight from the bottle, approx. 37% formaldehyde)

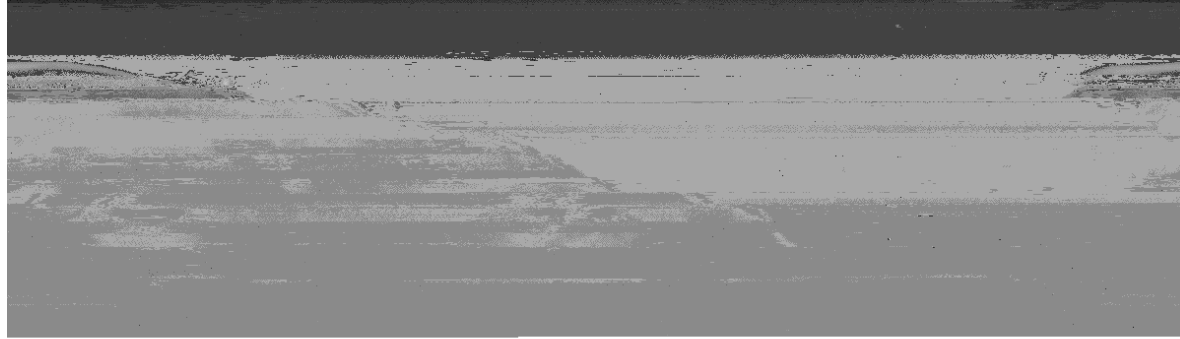
15 ml water

75 ml 100% ethanol

Tissue preparation

- 6) The specimen is placed under the microscope in a Petri dish with black elastomer filled with methyl salicylate. The cavity of the inner ear is visualized and injected (see ppt slides for orientation). For embryos up to E13.5, injection is approached laterally. Embryos older than E13.5 are hemisectioned and injections are approached medially through the cochlear duct or utricle area.
- 7) Depending on the size of specimens, the glass capillary tubing is cut accordingly right before injection.

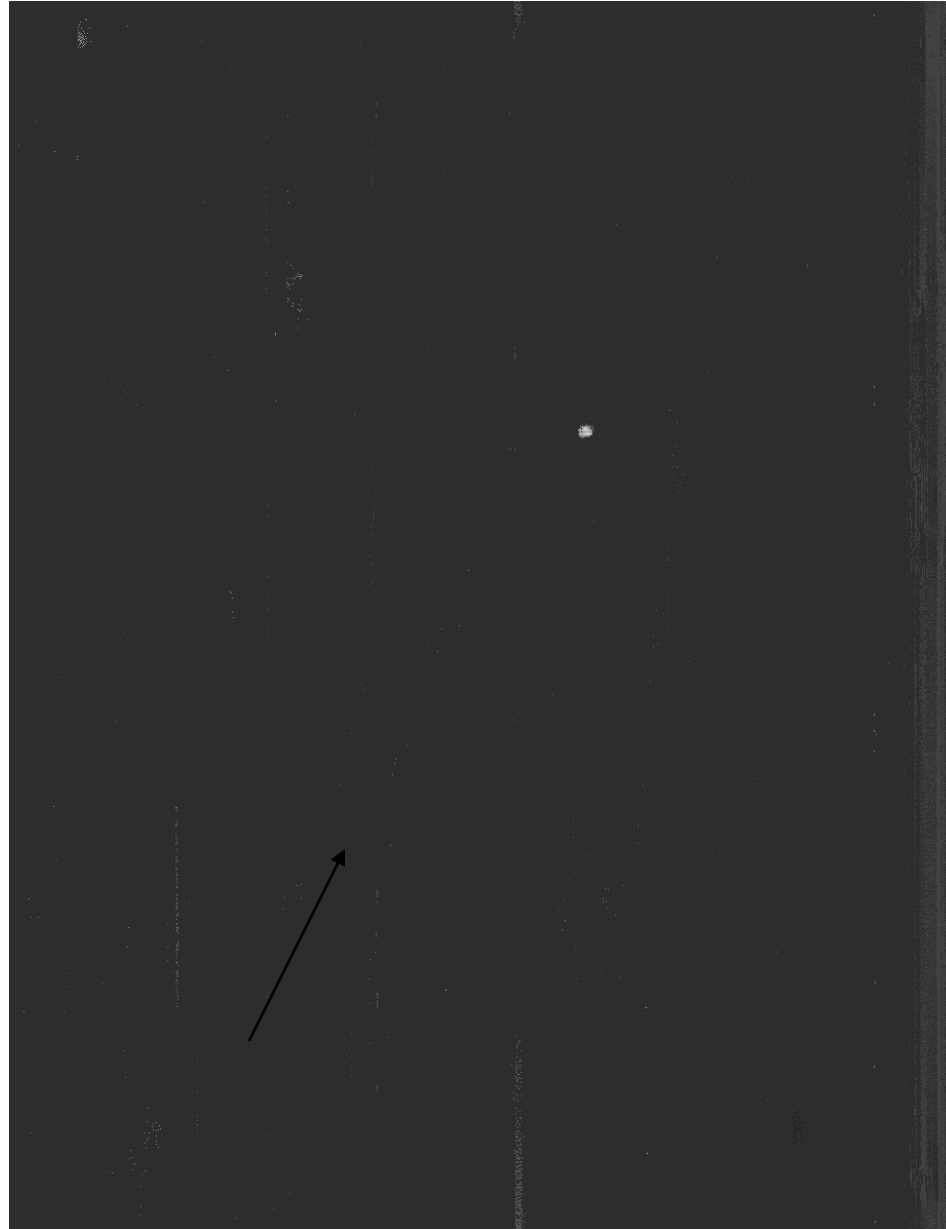
Hemisection the mouse head  
Do you see the ear?



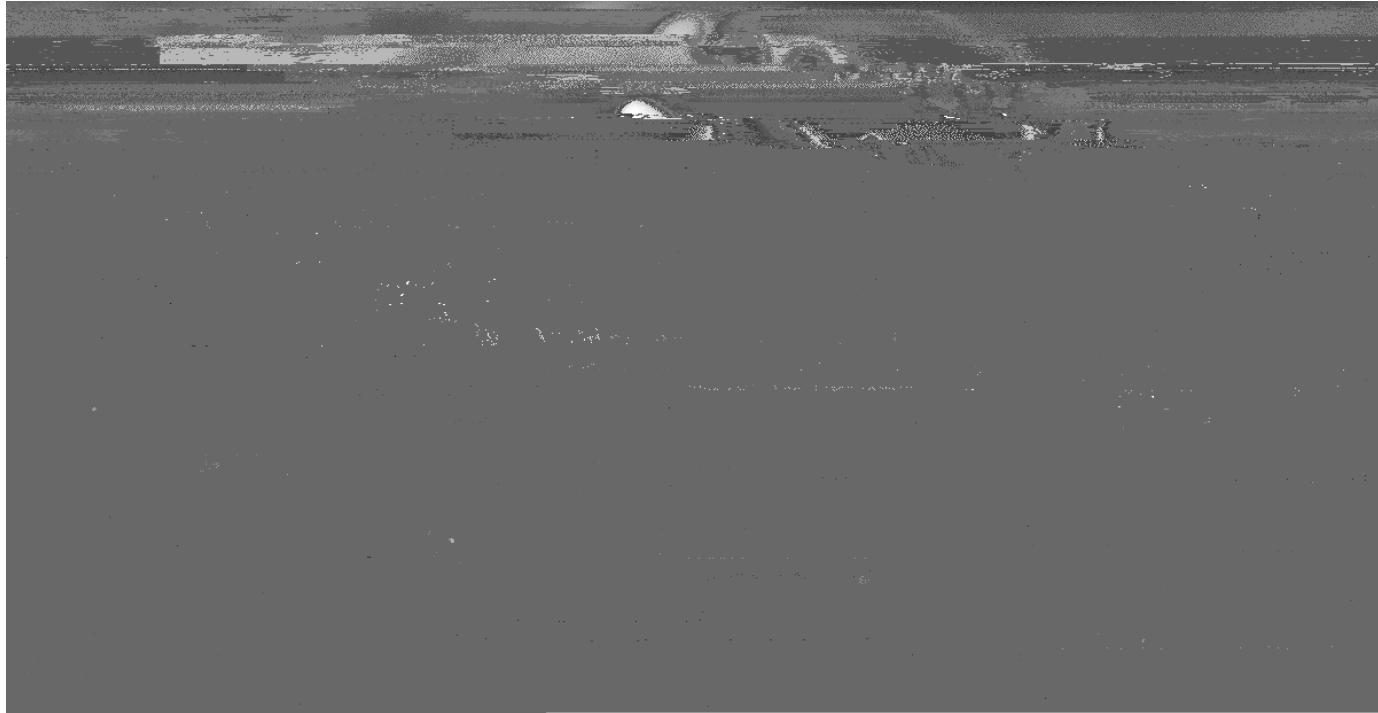
# Magnified view of the inner ear



Rotate specimen 90 degree clockwise from previous orientation  
Aim at the cochlear region with the arrow



# Injected specimen



For utricule injection, turn specimen 180 degree from the orientation in Slide 1. Aim for area by the arrow.

