

Preparation of Rodent Temporal Bones for Scanning Electron Microscopy

1. Fix tissue in 2.5% glutaraldehyde in 0.1M sodium cacodylate buffer pH 7.3 with 3mM CaCl₂ (2 - 3 hours minimum, but better if done for 24-48 hrs. can be done at room temp or 4C, ideally in a sealed container in a fume hood). If possible, perfusion through the round window and/or an open fenestra at the helicotrema)
2. Rinse in 0.1M cacodylate buffer (3 x 5 min, room temp)
3. Fix in 1% OsO₄ in 0.1M cacodylate. (30 – 60 min, room temp, in fume hood)
4. Rinse in cacodylate buffer (3

Supplies:

Glutaraldehyde solution (25%): 18426 from Ted Pella

Sodium cacodylate

CaCl₂

OsO₄; 4% aqueous solution – 18459 (10x2ml) from Ted Pella