

Date: _____

Time point: _____

of ears: _____

Immuno protocol:

- Fix 1 hour in 4% paraformaldehyde in PBS.
- Rinse 3x in PBS, 5 minutes each rinse.
- Permeabilize tissue in 1% Triton X-100 in PBS for 10 minutes.
- Rinse 3x in PBS, 5 minutes each rinse.
- Block in 10% NGS in PBS for 10 minutes.
- Rinse 3x in PBS for 5 minutes.

- Primary Antibody in PBS: Myo6 Polyclonal #1548 at 1:200 for 2h@RT
- Rinse 3x in PBS, 5 minutes each rinse.
- Secondary antibody in PBS: GAR #233 @ 1:1000 * for 2h@RT.
- Rinse 3x in PBS, 5 minutes each rinse.

Optional:

- Counterstain: Phalloidin 1:200 in PBS _____ for 45 min@ RT .
- Rinse 3x in PBS, 5 minutes each rinse.

- Nuclear label: _____ for _____.
- Rinse 3x in PBS, 5 minutes each rinse.
- Mount tissue in anti-fade on slide, coverslip and seal

*We use Invitrogen (Molecular Probes) AlexaFluor 488 (green, #A11001) or 568 (red, #A11004) secondary antibodies.