

Cell dissociation from *N. furzeri* embryos

1. Directly squash Nfur embryos with tweezers. Remove chorion and yolk. Also, remove YSL as much as possible.
 2. Transfer embryos to a new plate of PBS on ice, or directly collect and transfer all embryos into 1.5ml Eppendorf tubes. Spin down embryos by benchtop minicentrifuge, 1min.
 3. Wash with PBS x 2
 4. Remove supernatant. Add 250 ul 0.25% Trypsin which is pre-warmed at room temp.
 5. Pipetting cells and incubate at 26 – 28°C for 3 min
 6. Repeat steps 5-6, twice
 7. Centrifuge 500g, 3 min
 8. Remove supernatant. Add CO2 independent medium
- Trypsin treatment for only one cycle seems to leave many neural tubes intact.
 - Notice that cells might not completely form a solid pellet at the tube bottom; might stick along the tube wall.

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