# Materials List for:

**In Vivo Calcium Imaging of Lateral-line Hair Cells in Larval Zebrafish**

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## Materials

<table>
<thead>
<tr>
<th>Name</th>
<th>Company</th>
<th>Catalog Number</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Section 1</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>α-Bungarotoxin</td>
<td>R&amp;D Systems</td>
<td>2133</td>
<td>For paralyzing larvae prior to imaging</td>
</tr>
<tr>
<td>Phenol red Solution 0.5%</td>
<td>Sigma-Aldrich</td>
<td>P0290</td>
<td>For visualization of α-bungarotoxin solution during heart injection</td>
</tr>
<tr>
<td>Ethyl 3-aminobenzoate methanesulfonate (MESAB, MS-222, tricaine)</td>
<td>Sigma-Aldrich</td>
<td>A5040</td>
<td>For anesthetizing larvae</td>
</tr>
<tr>
<td><strong>Section 2</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Imaging chamber</td>
<td>Siskiyou</td>
<td>PC-R</td>
<td>Platform to mount larvae for imaging</td>
</tr>
<tr>
<td>No. 1.5 Coverslips square, 22&quot;x22 mm</td>
<td>VWR</td>
<td>48366-227</td>
<td>To seal imaging chamber</td>
</tr>
<tr>
<td>High vacuum silicone grease</td>
<td>Fischer Scientific</td>
<td>14-635-5D</td>
<td>For affixing of coverslip to imaging chamber</td>
</tr>
<tr>
<td>Silicone encapsulant clear 0.5 g kit</td>
<td>Ellsworth Adhesives</td>
<td>Dow Corning Sylgard, 184 SIL ELAST KIT 0.5KG</td>
<td>To fill imaging chamber to create a surface to pin fish</td>
</tr>
<tr>
<td>Oven</td>
<td>Techne</td>
<td>HB-1D</td>
<td>For drying silicone encapsulant</td>
</tr>
<tr>
<td>Stereomicroscope</td>
<td>Carl Zeiss Microscopy</td>
<td>Stem 2000 with transmitted light illumination</td>
<td>For illuminating wire, forceps and scissors to make pins</td>
</tr>
<tr>
<td>Fine forceps</td>
<td>Fine Science Tools</td>
<td>Dumont #5 (0.05 x 0.02 mm) Item No. 11295-10</td>
<td>For making pins</td>
</tr>
<tr>
<td>Fine scissors</td>
<td>Cole-Palmer</td>
<td>5.5&quot;, EW-10818-00</td>
<td>For cutting tungsten wire to make pins</td>
</tr>
<tr>
<td>Tungsten wire, 0.035 mm</td>
<td>Goodfellow</td>
<td>W005131</td>
<td>For head pins to immobilize larvae</td>
</tr>
<tr>
<td>Tungsten wire, 0.025 mm</td>
<td>ThermoFischer Scientific</td>
<td>AA10405-H4</td>
<td>For tail pins to immobilize larvae</td>
</tr>
<tr>
<td><strong>Section 3</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Micropipette guller</td>
<td>Sutter Instrument Company</td>
<td>P-97</td>
<td>For pulling capillary glass for fluid-jet and heart injection needles</td>
</tr>
<tr>
<td>Borosilicate glass capillaries w/ filament</td>
<td>Sutter Instrument Company</td>
<td>BF 150-86-10</td>
<td>Glass to be pulled into α-bungarotoxin injection needles for heart injection</td>
</tr>
<tr>
<td>Borosilicate glass capillaries w/o filament</td>
<td>Sutter Instrument Company</td>
<td>B 150-86-10</td>
<td>Glass to be pulled into fluid-jet needles to stimulate hair cells</td>
</tr>
<tr>
<td>Pipette polisher</td>
<td>Narishige</td>
<td>MF-830 microforge</td>
<td>To polish fluid-jet pipette tips to smooth jagged breaks</td>
</tr>
<tr>
<td>Ceramic tile</td>
<td>Sutter Instrument Company</td>
<td>NC9569052</td>
<td>For scoring and evening breaking fluid-jet needles</td>
</tr>
<tr>
<td><strong>Sections 4 &amp; 5</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Fine forceps</td>
<td>Fine Science Tools</td>
<td>Dumont #5 (0.05 x 0.02 mm) Item No. 11295-10</td>
<td>For pinning larvae</td>
</tr>
<tr>
<td>------------------</td>
<td>----------------------------</td>
<td>--------------------------------------------</td>
<td>------------------</td>
</tr>
<tr>
<td>Gel loading tips</td>
<td>Eppendorf</td>
<td>5242956003</td>
<td>For backfilling heart injection needles</td>
</tr>
<tr>
<td>Stereomicroscope</td>
<td>Carl Zeiss Microscopy</td>
<td>Stem 2000 with transmitted light illumination</td>
<td>For illuminating larvae during pinning and heart injection</td>
</tr>
<tr>
<td>Glass capillary/needle holder</td>
<td>WPI</td>
<td>MPH315</td>
<td>To hold fluid-jet needles</td>
</tr>
<tr>
<td>Manual micromanipulator</td>
<td>Narishige</td>
<td>M-152</td>
<td>For holding and positioning of needle holder to inject α-bungarotoxin</td>
</tr>
<tr>
<td>Magnetic stand</td>
<td>Narishige</td>
<td>GJ-1</td>
<td>For holding manual micromanipulator for α-bungarotoxin injection</td>
</tr>
<tr>
<td>Pressure injector</td>
<td>Eppendorf</td>
<td>Femtojet 4x</td>
<td>To deliver α-bungarotoxin</td>
</tr>
</tbody>
</table>

**Sections 6, 7 & 8**

<table>
<thead>
<tr>
<th>Confocal microscope</th>
<th>Bruker</th>
<th>Swept field/Opterra confocal microscope</th>
<th>Fixed, upright microscope with DIC optics and 488 nm laser with appropriate filters</th>
</tr>
</thead>
<tbody>
<tr>
<td>Microscope software</td>
<td>Bruker</td>
<td>Prairieview 5.3</td>
<td>To coordinate and control the microscope, lasers, stage, piezo-z, cameras and fluid jet</td>
</tr>
<tr>
<td>10x air objective</td>
<td>Nikon</td>
<td>MRH00101</td>
<td>Low magnification for positioning of larvae and fluid jet</td>
</tr>
<tr>
<td>60x water objective</td>
<td>Nikon</td>
<td>MRF07620</td>
<td>Water immersion objective with high NA (1.0) and adequate working distance (2.0 mm)</td>
</tr>
<tr>
<td>Piezo-Z objective scanner with controller/driver</td>
<td>Physik Intruments instruments/Bruker</td>
<td>01144210/UM-Z-PZ</td>
<td>High-speed z-stack acquisition with 0.025μm accuracy</td>
</tr>
<tr>
<td>EMCCD camera</td>
<td>QImaging</td>
<td>Rolera EM-C2 EMCCD camera</td>
<td>Camera with small pixel size that can acquire up to 100 frames per s</td>
</tr>
<tr>
<td>Circular chamber adapter</td>
<td>Siskiyou</td>
<td>PC-A</td>
<td>For holding and rotating imaging chamber on microscope stage</td>
</tr>
<tr>
<td>Motorized Z-deck stage</td>
<td>Prior Scientific</td>
<td>ZDN12MP</td>
<td>Microscope stage that can hold and move sample and micromanipulator with fluid-jet needle together</td>
</tr>
<tr>
<td>Z-deck stage insert adaptor</td>
<td>NIH Machine shop</td>
<td>custom</td>
<td>To fit the circular chamber adaptor onto the z-deck stage</td>
</tr>
<tr>
<td>Fluid-jet apparatus</td>
<td>ALA scientific instruments</td>
<td>HSPC-1 High-speed pressure Clamp with PV-PUMP</td>
<td>For controlling and delivering the fluid-jet stimulus</td>
</tr>
<tr>
<td>Masterflex Peroxide-cured silicone tubing (1 feet)</td>
<td>Cole- Palmer</td>
<td>Masterflex L/S 13, 96400-13</td>
<td>For connecting the fluid-jet needle holder to pressure pump</td>
</tr>
<tr>
<td>Motorized micromanipulator</td>
<td>Sutter Instrument Company</td>
<td>MP-225</td>
<td>For holding and positioning of needle holder for fluid jet</td>
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<tr>
<td>Micromanipulator controller</td>
<td>Sutter Instrument Company</td>
<td>MPC-200</td>
<td>For controlling fluid-jet needle manipulator</td>
</tr>
<tr>
<td>Gel loading tips</td>
<td>Eppendorf</td>
<td>5242956003</td>
<td>For backfilling fluid-jet needles</td>
</tr>
<tr>
<td>Glass capillary/needle holder</td>
<td>WPI</td>
<td>MPH315</td>
<td>To hold fluid-jet needles</td>
</tr>
<tr>
<td>PSI manometer</td>
<td>Sper Scientific</td>
<td>840081</td>
<td>For measuring pressure clamp output</td>
</tr>
</tbody>
</table>

**Section 9**

<p>| BAPTA, Tetrapotassium Salt, cell impermeant | ThermoFischer Scientific | B1204 | To cleave tips links, uncouple MET channels and block all evoked GCaMP6s signals |</p>
<table>
<thead>
<tr>
<th>Material</th>
<th>Vendor</th>
<th>Code</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Isradipine</td>
<td>Sigma-Aldrich</td>
<td>I6658</td>
<td>To block signals dependent on the L-type calcium channels (CaV1.3) at the presynapse</td>
</tr>
<tr>
<td>DMSO</td>
<td>Sigma-Aldrich</td>
<td>D8418</td>
<td>Solvent for pharmacological compounds</td>
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</tbody>
</table>

**Section 10**

<table>
<thead>
<tr>
<th>Software</th>
<th>Vendor</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prism7</td>
<td>Graphpad</td>
<td>Software to plot GCaMP6 intensity changes</td>
</tr>
<tr>
<td>FIJI</td>
<td>Schindelin, et., al.</td>
<td><a href="https://fiji.sc/">https://fiji.sc/</a></td>
</tr>
<tr>
<td>Times Series Analyzer V3 Plugin</td>
<td>Balaji J 2007, Dept. of Neurobiology, UCLA</td>
<td><a href="https://imagej.nih.gov/ij/plugins/time-series.html">https://imagej.nih.gov/ij/plugins/time-series.html</a></td>
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