

Materials List for:

Preparing Fresh Retinal Slices from Adult Zebrafish for *Ex Vivo* Imaging Experiments

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Materials

Name	Company	Catalog Number	Comments
zebrafish	Univeristy of Washington South Lake Union Aquatics Facility		stocks maintained in-house as stable transgenic lines
petroleum jelly	Fisher Scientific	19-090-843	for petroleum jelly syringe
3-mL slip tip syringe	Fisher Scientific	14-823-436	for petroleum jelly syringe
20g 3.8 cm slip tip needle	Fisher Scientific	14-826-5B	for petroleum jelly syringe
plain 7 cm X 2.5 cm microscope slide	Fisher Scientific	12-550-A3	for eyecup dissection, slicing chamber
Seche Vite clear nail polish	Amazon	B00150LT40	for slicing chamber
18 mm X 18 mm #1 glass coverslips	Fisher Scientific	12-542A	for imaging ladders
unflavored dental wax	Amazon	B01K8WNL5A	for imaging ladders
double edge razor blades	Stoelting	51427	for tissue slicing
tissue slicer with digital micrometer	Stoelting	51415	for tissue slicing
filter paper - white gridded mixed cellulose, 13 mm diameter, 0.45 µm pore size	EMD Millipore	HAWG01300	filter paper for mounting retinas
10 cm petri dish	Fisher Scientific	FB0875712	for fish euthanasia, dissection, imaging ladder assembly
15 cm plain-tipped wood applicator stick	Fisher Scientific	23-400-112	for wire eye loop tool
30g (0.25 mm diameter) tungsten wire	Fisher Scientific	AA10408G6	for wire eye loop tool
D-glucose	Sigma Aldrich	G8270	component of supplement stock solution
sodium L-lactate	Sigma Aldrich	L7022	component of supplement stock solution
sodium pyruvate	Sigma Aldrich	P2256	component of supplement stock solution
L-glutamine	Sigma Aldrich	G3126	component of supplement stock solution
L-glutathione, reduced	Sigma Aldrich	G4251	component of supplement stock solution
L-ascorbic acid	Sigma Aldrich	A5960	component of supplement stock solution
NaCl	Sigma Aldrich	S7653	component of Ringer's solution
KCl	Sigma Aldrich	P9333	component of Ringer's solution
CaCl ₂ · 2H ₂ O	Sigma Aldrich	C3881	component of Ringer's solution
NaH ₂ PO ₄	Sigma Aldrich	S8282	component of Ringer's solution

MgCl ₂ · 6H ₂ O	Sigma Aldrich	M0250	component of Ringer's solution
HEPES	Sigma Aldrich	H3375	component of Ringer's solution
Tris base	Fisher Scientific	BP152	component of Na ⁺ -free Ringer's solution
6 N HCl	Fisher Scientific	02-003-063	component of Na ⁺ -free Ringer's solution
KH ₂ PO ₄	Sigma Aldrich	P5655	component of Na ⁺ -free Ringer's solution
50 mL conical centrifuge tube	Denville Scientific	C1062-P	container for Ringer's solution
Vannas scissors - 8 cm, angled 5 mm blades	World Precision Instruments	501790	micro-scissors for eyecup dissection
Swiss tweezers - #5, 11 cm, straight, 0.06 X 0.07 mm tips	World Precision Instruments	504510	fine forceps for eyecup dissection and slice manipulation
single edge razor blades	Fisher Scientific	12-640	for eyecup dissection and trimming filter paper
EMD Millipore filter forceps	Fisher Scientific	XX6200006P	flat forceps for handling wet filter paper
C ¹² 558/568 BODIPY	Fisher Scientific	D3835	stains live cell nuclei; incubate 5 µg/mL for 15 min at room temperature
propidium iodide (PI)	Fisher Scientific	P3566	stains dead cell nuclei; incubate 5 µg/mL for 20 min at room temperature
Hoechst 33342	Fisher Scientific	62249	stains live cell nuclei; incubate 5 µg/mL for 20 min at room temperature
Tetramethylrhodamine, methyl ester (TMRM)	Fisher Scientific	T668	stains functional, negatively-charged mitochondria; incubate 1 nM for 30 min at room temperature
tissue perfusion chamber	Cell MicroControls	BT-1-18/BT-1-18BV [-SY]	imaging chamber for injection or perfusion
2-(N-(7-Nitrobenz-2-oxa-1,3-diazol-4-yl)Amino)-2-Deoxyglucose (NBDG)	Fisher Scientific	N13195	fluorescent glucose analog administered orally to zebrafish 30 min prior to euthanasia
Olympus laser scanning confocal microscope	Olympus	FV1000	confocal microscope for visualizing fluorescence of slices at single-cell resolution
Carbonyl cyanide 3-chlorophenylhydrazone (CCCP)	Sigma Aldrich	C2759	experimental reagent which ablates mitochondrial respiration; treat slices to a final concentration of 1 µM
miniature aspirator positioner	Cell MicroControls	FL-1	for perfusion
perfusion manifold, gas bubbler manifold, flow valve, 60cc syringe holder	Warner Instruments	various	for perfusion