

Materials List for:

Dissection, Culture, and Analysis of *Xenopus laevis* Embryonic Retinal Tissue

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Materials

Name	Company	Catalog Number	Comments
For Dissections and Culturing			
BD Falcon Easy Grip Tissue Culture Dishes, 35 mm	Fisher	08-772A	
Disposable Polystyrene Petri Dishes, 60 x 15 mm	Fisher	0875713A	
35 mm Nunclon Surface Petri Dishes (with Airvent)	Fisher	12-565-91	
Dumont Fine Forceps (Dumostar #55)	Fisher	NC9341917	
Cellattice Micro-ruled plastic coverslip, 25 mm	Fisher	50-313-17	
Ethyl-m-Aminobenzoate Methanesulfonate Salt (MS-222)	MP Biomedicals, LLC	103106	
Gentamicin Sulfate	Enzo Life Sciences	380-003-G025	
Gibco Trypsin (1:250) Powder	Life Technologies	27250-018	
Collagenase B from <i>Clostridium histolyticum</i>	Roche	11088831001	
Penicillin-Streptomycin	Gibco	15140-122	
Nile Blue Sulfate (optional)	Pfaltz Bauer	N05550	
500 ml Vacuum Filter/Storage Bottle System, 0.22 µm Pore 33.2 cm ² CN Membrane	Corning	430758	
For Calcium Imaging			
Fluo-4 AM 1 mM Solution in DMSO, Cell Permanent	Life Technologies	F-14217	
Pluronic F-127 10% Solution in Water	Life Technologies	P-6866	
LSM 510 Confocal Microscope System	Zeiss	Model Discontinued	
Blocking Reagent	Roche	11096176001	
For Fluorescent <i>in situ</i> hybridization (FISH)			
Anti-Digoxigenin-POD, Fab Fragments	Roche	11207733910	
Anti-DNP-HRP Antibody	Perkin-Elmer	NEL747A	
Cy3 NHS ester	GE Healthcare	PA13101	
NHS-Fluorescein	Thermo Scientific	46409	
Formamide, Deionized	Amresco	0606-950ML	
Torula RNA, Type IX	Sigma-Aldrich	R3629	

Heparin Sodium Salt, from Porcine Intestinal Mucosa	Sigma-Aldrich	H3393-250KU	
CHAPS	Sigma-Aldrich	C3023	

Table 2. Specific reagents and equipment.

Solution	Reference	Contents
Cell Culture Medium	Chang and Spitzer , 2009 ⁵⁴	116 mM NaCl 0.67 mM KCl 2 mM CaCl ₂ 1.31 mM MgSO ₄ 4.6 mM Tris 1 % (v:v) Penicillin/Streptomycin Adjust pH to 7.8 with HCl. Filter sterilize by passing through a 0.22 µm CN filter. Store at 4 °C.
Calcium-Magnesium Free Medium (CMF)	Gu <i>et al.</i> , 1994 ⁴² ; Gu and Spitzer, 1995 ⁵⁵	116 mM NaCl 0.67 mM KCl 4.6 mM Tris 0.4 mM EDTA 1 % (v:v) Penicillin/Streptomycin Adjust pH to 7.8 with HCl. Filter sterilize by passing through a 0.22 µm CN filter. Store at 4 °C.
Maleic Acid Buffer (MAB)	Sive <i>et al.</i> , 2000 ⁵³	100 mM maleic acid 150 mM NaCl (pH 7.5).
Marc's Modified Ringers (MMR), 10X	Sive <i>et al.</i> , 2000 ⁵³	100 mM NaCl mM KCl 1 mM MgSO ₄ 2 mM CaCl ₂ 5 mM HEPES pH adjusted to 7.4 with NaOH 0.1X MMR also contains 50 mg/ml gentamicin sulfate and pH is adjusted to 7.4 with NaOH.
MEMFA Solution, 10X	Sive <i>et al.</i> , 2000 ⁵³	0.1 M MOPS (Ph 7.4) 2 mM EGTA 1 mM MgSO ₄ 3.7% formaldehyde A 10X solution, without formaldehyde, can be stored at 4 °C. Formaldehyde is added fresh (1/10 volume of a standard 37% stock).
<i>In situ</i> Hybridization Buffer	Sive <i>et al.</i> , 2000 ⁵³	50% Formamide 5X SSC 1 mg/ml Torula RNA 100 mg/ml Heparin 1X Denhart's Solution 0.1% Tween 20 0.1% CHAPS 10 mM EDTA.

Solutions. *Gentamicin, an antibiotic, is used in our MMR solutions while penicillin and streptomycin are used in our culture media.