

Materials List for:

Use of Immunolabeling to Analyze Stable, Dynamic, and Nascent Microtubules in the Zebrafish Embryo

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URL: <https://www.jove.com/video/55792>

DOI: [doi:10.3791/55792](https://doi.org/10.3791/55792)

Materials

Name	Company	Catalog Number	Comments
Agarose			Used to treat petridishes. Prepare 1% agarose by heating a solution of 1 gram agarose per 100 ml 1X embryo medium in a microwave until polymerized.
Kpipes	Sigma	P7643	
NaCl	Sigma	S7653	
Tris-HCl	Sigma	T3253-500G	
KCl	Sigma	P9333-500G	
CaCl ₂ ·2H ₂ O	Sigma	C5080	
NP-40	American Bioanalyticals	AB01424	
EGTA	Sigma	E3889-25G	
MgCl ₂	Sigma	M2670-500G	
Bovine serum albumin (BSA)	Fisher	BP1605	
Triton-x	American Bioanalyticals	AB02025	
Anti-Fade mounting medium	Invitrogen	P10144	
Mouse anti-β-tubulin	Developmental studies Hybridoma Bank	E7	1/200
Rabbit anti-γ-tubulin	Genetex	GTX113286	1/500
Rabbit anti-α-tubulin	Genetex	GTX108784	1/1000*
Rabbit anti-detyrosinated-tubulin	Millipore	AB3201	1/200-1/1000* Titrate antibody with first use of new lot.
Rabbit anti-tyrosinated-tubulin	Millipore	ABT171	1/500
Mouse anti-centrin	Millipore	04-1624	1/1000
Goat 488 anti-rabbit	Thermofisher	A11008	1/500
Goat 594 anti-rabbit	Thermofisher	A11012	1/500
Goat 594 anti-mouse	Thermofisher	A11005	1/500
Goat 488 anti-mouse	Thermofisher	A11001	1/500
Vibratome	Vibratome	1500	
Forceps	World Precision Instruments	555227F	
100 mm petri dish	Cell treat	229693	
35 mm petri dish	Cell treat	229638	
50 ml falcon tube	Fisher	14-432-22	
Woven nylon mesh 70 um	Amazon.com	B0043D1SZG	

Micropipette	Gilson	F123602	
Glass pipette	Fisher	NC-999363-9	
Aquarium sealant	Amazon.com, by MarineLand	Silicone Sealer 1 oz (Tube)	
Ring stand	Fisher	14-675BO	
Microbore PTFE Tubing, 0.022"ID	Cole-Parmer	WU-06417-21	
Modeling clay	Amazon.com	Sargent Art 22-4000	Any wax or oil based non-toxic modeling clay will suffice
Clamp	Fisher	02-215-466	
60ml syringe	Fisher	14-820-11	
Embryo medium (E3)			34.8 g NaCl 1.6 g KCl 5.8 g CaCl ₂ ·2H ₂ O 9.78 g MgCl ₂ ·6H ₂ O To prepare a 60X stock, dissolve the ingredients in H ₂ O, to a final volume of 2 L. Adjust the pH to 7.2 with NaOH. Autoclave. To prepare 1X medium, dilute 16.5 mL of the 60X stock to 1 L.
Blocking Solution			50 ml TBS-NP-40 2.5 ml normal goat serum 1 g BSA 625 µl Triton-X
TBS-NP-40 (pH 7.6)			155 mM NaCl 10 mM Tris HCl 0.1% NP-40
2x MAB (pH6.4)			160 mM KPIPES 10 mM EGTA 2 mM MgCl ₂
Commercial 3-D Image processing Software	PerkinElmer	Volocity (V 6.2)	
Dry block heater	VWR	12621-108	Used as a hot plate to melt agarose in Protocol 1.
Dissecting Microscope	Leica	MZ12	
Confocal Microscope	Leica	SP5	
Flat embedding mold	emsdiasum.com	BEEM 70904-01	
Public domain image processing software	NIH	ImageJ (V 1.5)	
* Success varies by lot number			