

Materials List for:

Study of the DNA Damage Checkpoint using *Xenopus* Egg Extracts

Jeremy Willis¹, Darla DeStephanis¹, Yogin Patel¹, Vrushab Gowda¹, Shan Yan¹

¹Department of Biology, University of North Carolina at Charlotte

*These authors contributed equally

Correspondence to: Shan Yan at Shan.Yan@uncc.edu

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Materials

Name	Company	Catalog Number	Comments
Reagents			
Anti-Chk1 P-S344 antibody	Cell Signaling	2348L	
Anti-Chk1 antibody	Santa Cruz	SC7898	
Aprotinin	MP Biomedicals	0219115880	
Cycloheximide	Sigma	C7698-5G	
Cytochalasin B	EMD	250233	
Dithiothreitol (DTT)	VWR	JTF780-2	
hCG	Sigma	CG10-10VL	
L-Cysteine	Sigma	C7352-1KG	
Leupeptin	VWR	97063-922	
Methyl methanesulfonate (MMS)	Sigma	129925-5G	
Nocodazole	Sigma	M1404-2MG	
PMSG	Calbiochem	367222	
Sample buffer	Sigma	S3401	
Tautomycin	Wako Chemicals USA	209-12041	
Equipment			
Bucket for egg laying	Rubbermaid commercial products	6308	
CL2 IEC centrifuge with swinging bucket rotor	Thermo Scientific	004260F	
HB6 swinging bucket rotor	Thermo Scientific	11860	
Sorvall RC6 plus superspeed centrifuge	Thermo Scientific	46910	
UV crosslinker	UVP	95-0174-01	
Solutions			
1x MMR			100 mM NaCl, 2 mM KCl, 0.5 mM MgSO ₄ , 2.5 mM CaCl ₂ , 5 mM HEPES, adjust pH to 7.8 with 10 M NaOH
Aprotinin/Leupeptin stock			10 mg/ml each in water. Store 20 µl aliquots at -80 °C.
Buffer X			0.2 M sucrose, 80 mM KCl, 15 mM NaCl, 5 mM MgCl ₂ , 1 mM EDTA, 10 mM HEPES, adjust pH to 7.5 by HCl
Cycloheximide stock			10 mg/ml in water. Store 1 ml aliquots at -20 °C.
Cytochalasin B stock			5 mg/ml in DMSO. Store 20 µl aliquots at -20 °C.

Dithiothreitol (DTT) stock			1 M in water. Store 1 ml aliquots at -20 °C.
ELB			0.25 M sucrose, 1 mM DTT, 50 µg/ml cycloheximide, 2.5 mM MgCl ₂ , 50 mM KCl, 10 mM HEPES, pH7.7
Nocodazole stock			10 mg/ml in DMSO. Store 5 µl aliquots at -80 °C.
Energy Mixture			375 mM creatine phosphate, 50 mM ATP, and 25 mM MgCl ₂ . Aliquots are saved at -80 °C.
Nuclear dye solution			0.4 µg/ml H-chst 33258, 25% glycerol (v/v), in 1x PBS
Tautomycin stock			100 µM in DMSO. Store 10 µl aliquots at -80 °C.