

Materials List for:

Identification of Cyclin-dependent Kinase 1 Specific Phosphorylation Sites by an *In Vitro* Kinase Assay

Heying Cui¹, Kyle M. Loftus¹, Crystal R. Noell¹, Sozanne R. Solmaz¹

¹Department of Chemistry, State University of New York at Binghamton

*These authors contributed equally

Correspondence to: Sozanne R. Solmaz at ssolmaz@binghamton.edu

URL: <https://www.jove.com/video/57674>

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Materials

Name	Company	Catalog Number	Comments
2800 mL baffled Fernbach flask	Corning	44232XL	
ampicillin	Gold Biotechnology	A-301-25	
ATP	Fisher Scientific	BP413-25	
benzamidinium hydrochloride	Millipore Sigma	B6506-25	
bottletop filter	Corning	431161	
Cdk1/cyclin B recombinant, human 20,000 U/mL	New England Biolabs	P6020	
Cdk1/cyclin B (alternate source)	EMD Millipore	14-450	
Cdk1/cyclin B (alternate source)	Invitrogen	PV3292	
Cdk1/cyclin B + 10x PK buffer	New England Biolabs	P6020	
CENP-F (residues 2987 – 3065) pGEX6P1 plasmid	Available upon request.		
centrifuge: Heraeus Multifuge X3R, cooled, with TX-1000 swing-out rotor	Thermo Scientific	10033-778	
centrifugal filter units: Amicon Ultra-15 centrifugal filter units, 3 kDa cutoff, Ultracel-PL membranes	EMD Millipore	UFC900324	
chloramphenicol	Gold Biotechnology	C-105-100	
D/L methionine	Agros Organics / Fisher	125650010	
desalting pipet tips: Zip tips	Millipore Sigma	ZTC18S008	
disposable chromatography columns, Econo-Pac 1.5 x 12 cm	Biorad	7321010	
dithiothreitol	Gold Biotechnology	DTT50	
<i>E. coli</i> Rosetta 2(DE3)pLysS strain	EMD Millipore	71403	
electrospray ionization Fourier transform ion	Bruker Amazon	Apex III	
cyclotron resonance mass spectrometer			
electrospray ionization ion trap mass spectrometer	Bruker Amazon	custom	
fixed angle rotor: Fiberlite F15-8x-50cy	Thermo Scientific	97040-276	
FPLC system: Äkta Pure FPLC	GE Healthcare	29032697	
Gel filtration column: Superdex 200 Increase 10/300 GL	GE Healthcare	28990944	
glutathione agarose	Pierce	16101	

glutathione, reduced	Millipore Sigma	G4251-50g	
incubation shaker: multitron shaker	Infors	I10102	
isopropyl β -D-1-thiogalactopyranoside	Gold Biotechnology	I2481C50	
kanamycin	Gold Biotechnology	K-120-25	
karyopherin α pet-28a pres plasmid	Available upon request.		
Luria Bertani medium	Fisher Scientific	BP1426-2	
microcentrifuge 5418R, refrigerated	Eppendorf	5401000013	
microtubes (0.5 mL)	Eppendorf	30121023	
microtubes (1.5 mL)	Eppendorf	30120086	
Nickel affinity gel: His-Select Nickel affinity gel	Millipore Sigma	P6611-100ml	
pGEX-6P-1 plasmid	Millipore Sigma	GE28-9546-48	
phenylmethylsulfonyl fluoride	Gold Biotechnology	P470-10	
PS protease: PreScission protease	GE Healthcare	27084301	
Phos-tag acrylamide	Wako Pure Chem. Ind.	304-93521	
reduced glutathione	Millipore Sigma	G4251-50g	
roundbottom centrifuge tubes (Oakridge tubes)	Fisher Scientific	055291D	
site-directed mutagenesis kit: QuikChange Lightning	Agilent	210518	
Site-Directed Mutagenesis Kit			
sonifier: Branson S-250D sonifier	Branson	15 338 125	
Spectra/Por 1RC dialysis membrane (6-8 kDa cutoff)	Spectrum Labs	08 670B	
swing out rotor TX-1000	Thermo Scientific	10033-778	