

Materials List for:

# Cycloheximide Chase Analysis of Protein Degradation in *Saccharomyces cerevisiae*

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## Materials

Name	Company	Catalog Number	Comments
Desired yeast strains, plasmids, standard medium and buffer components			
Disposable borosilicate glass tubes	Fisher Scientific	14-961-32	Available from a variety of manufacturers
Temperature-regulated incubator (e.g. Heratherm Incubator Model IMH180)	Dot Scientific	51028068	Available from a variety of manufacturers
New Brunswick Interchangeable Drum for 18 mm tubes (tube roller)	New Brunswick	M1053-0450	A tube roller is recommended to maintain overnight starter cultures of yeast cells in suspension. Alternatively, if a tube roller is unavailable, a platform shaker in a temperature-controlled incubator may be used for overnight starter cultures. A platform shaker or tube roller may be used to maintain larger cultures in suspension.
New Brunswick TC-7 Roller Drum 120 V 50/60 H	New Brunswick	M1053-4004	For use with tube roller
SmartSpec Plus Spectrophotometer	Bio-Rad	170-2525	Available from a variety of manufacturers
Centrifuge 5430	Eppendorf	5427 000.216	Rotor that is sold with unit holds 1.5 and 2.0 ml microcentrifuge tubes. Rotor may be swapped for one that holds 15 and 50 ml conical tubes
Fixed-Angle Rotor F-35-6-30 with Lid and Adapters for Centrifuge Model 5430/R, 15/50 ml Conical Tubes, 6-Place	Eppendorf	F-35-6-30	
15-ml screen printed screw cap tube 17 x 20 mm conical, polypropylene	Sarstedt	62.554.205	Available from a variety of manufacturers
1.5-ml flex-tube, PCR clean, natural microcentrifuge tubes	Eppendorf	22364120	Available from a variety of manufacturers
Analog Dri-Bath Heaters	Fisher Scientific	1172011AQ	It is recommended that two heaters are available (one for incubating cells during cycloheximide treatment and one for boiling lysates to denature proteins). Alternatively, 30 °C water bath may be used for incubation of cells in the presence of cycloheximide. Boiling water bath with hot plate

			may alternatively be used to denature proteins.
Heating block for 12 x 15 ml conical tubes	Fisher Scientific	11-473-70	For use with Dri-Bath Heater during incubation of cells in the presence of cycloheximide.
Heating block for 20 x 1.5 ml conical tubes	Fisher Scientific	11-718-9Q	For use with Dri-Bath Heater to boil lysates for protein denaturation.
SDS-PAGE running and transfer apparatuses, power supplies, and imaging equipment or darkrooms for SDS-PAGE and transfer to membrane			Will vary by lab and application
Western blot imaging system (e.g. Li-Cor Odyssey CLx scanner and Image Studio Software)	Li-Cor	9140-01	Will vary by lab and application
EMD Millipore Immobilon PVDF Transfer Membranes	Fisher Scientific	IPFL00010	Will vary by lab and application
Primary antibodies (e.g. Phosphoglycerate Kinase (Pgk1) Monoclonal antibody, mouse (clone 22C5D8))	Life Technologies	459250	Will vary by lab and application
Secondary antibodies (e.g. Alexa-Fluor 680 Rabbit Anti-Mouse IgG (H + L))	Life Technologies	A-21065	Will vary by lab and application