

Materials List for:

Methods to Classify Cytoplasmic Foci as Mammalian Stress Granules

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Materials

Name	Company	Catalog Number	Comments
U-2 OS	ATCC	ATCC®HTB-96™	Commonly written "U2OS" Culture at 37 °C under 5% CO ₂ in 10% FBS/DMEM
Dulbecco's Modified Eagle's Medium (DMEM)	Corning	10-013-CV	Contains 4.5 g/L glucose, pyruvate, and phenol red Supplemented with 10% FBS, 10 mM HEPES, and penicillin/ streptomycin Prewarm prior drug(s) dilution
Fetal Bovine Serum (FBS)	Sigma	F2442-500ml	Used to supplement media
HEPES (1 M)	Thermo Fisher Scientific	15630-080	Used to supplement media
penicilline streptomycine	Sigma	P0781-100ml	Used to supplement media
24-well plate	Costar	3524	
lab tissues (kimwipes)	KIMTECH SCIENCE	34120	
Coverglass for Growth Cover Glasses	Fisher Scientific	12-545-82	Autoclaved before use Keep sterile in the tissue culture hood
Sodium (meta)arsenite ≥90% (SA)	Sigma	S7400-100G	Dissolve in water to 1 M concentration, then dilute to 100 μM as a working stock
Vinorelbine (VRB)	TSZ CHEM	RS055	Dissolve in water to 10 mM
Puromycin	Sigma	P9620	Used to assess translation level (ribopuromylation) -Used to assess SG connection with active translation Dilute in water to 10 mg/mL
Emetine dihydrochloride hydrate	Sigma	E2375	Used in combination with puromycin to assess general translation level Used to assess SG connection with active translation -Dilute in water to 10 mg/mL
Cycloheximide (CHX)	Sigma	C4859	Used to assess SG connection with active translation Dilute in water to 10 mg/mL
Phosphate Buffered Saline (PBS)	Lonza / VWR	95042-486	Wash buffer for immunofluorescence
Paraformaldehyde reagent grade, crystalline (PFA)	Sigma	P6148-500G	Make a 4% solution in hot PBS in fume hood, stir until dissolved Aliquots can be stored at - 20 °C for several months

			Hazardous, use ventilation Discard in special waste
Methanol, ACS	BDH / VWR	BDH1135-4LP	Prechill to -20°C before use Discard in special waste
Normal Horse Serum (NHS)	Thermo Fisher Scientific	31874	Dilute to 5% in PBS Add sodium azide for storage at 4°C Blocking solution for immunofluorescence
Ethanol, Pure, 200 Proof (100%),USP, KOPTEC	Decon Labs / VWR	89125-188	Dilute to 70% with water
Fisherbrand Superfrost Plus Microscope Slides	Fisherbrand	12-550-15	
mouse anti G3BP1 antibody (TT-Y)	Santa Cruz	sc-81940	Store at 4 °C 1/100 dilution
rabbit anti eIF4G antibody (H-300)	Santa Cruz	sc-11373	Store at 4 °C 1/250 dilution
goat anti eIF3η (N-20) antibody	Santa Cruz	sc-16377	eIF3η is also known as eIF3b Store at 4 °C 1/250 dilution
mouse anti puromycin 12D10 antibody	Millipore	MABE343	Store at 4 °C 1/1000 dilution
Cy2 AffiniPure Donkey Anti-Mouse IgG (H+L)	Jackson ImmunoResearch	715-225-150	Reconstitute in water per manufacturer's instructions then store at 4°C 1/250 dilution
Cy3 AffiniPure Donkey Anti-Rabbit IgG (H+L)	Jackson ImmunoResearch	711-165-152	Reconstitute in water as per manufacturer's instructions then store at 4°C 1/2500 dilution
Cy5 AffiniPure Donkey Anti-Goat IgG (H+L)	Jackson ImmunoResearch	705-175-147	Reconstitute in water as per manufacturer's instructions then store at 4°C 1/250 dilution
Hoechst 33258 solution	Sigma	94403-1ML	Incubate with secondary antibodies Stock solution 0.5 mg/mL in dH2O, protect from light -Store at 4 °C
Cy3 Streptavidin	Jackson ImmunoResearch	016-160-084	Reconstitute in water per manufacturer's instructions, then store at 4°C 1/250 dilution
oligo-(dT) _{40x} probe biotinilated	Integrated DNA Technologies (IDT)	/	Reconstitute in water to 100 ng/ mL, aliquot and store at -20°C Dilute in hybridation buffer 1/50 prior to use Custom order
hybridation box	/	/	Make a moisture chamber with any storing slide box by adding humidified paper on the bottom - prewarm the box prior use
20x SSC buffer	Thermo fisher Ambion	AM9763	Dilute to 2X SSC using RNase Free water (DEPC treated) Store at RT Wash buffer for FISH
PerfectHybPlus Hybridization Buffer	Sigma	H7033-50ml	Block and probe incubation buffer for FISH
slide mounting media	home made	/	Mix 5 g of "cold-soluble" poly(vinyl alcohol) in 20 mL of PBS

			Mix by sonication, followed by stirring O/N at RT Add 5 mL of glycerol and 0.2 mL of 20 % sodium azide, and stir for 16 h at RT Centrifugation at 20,000 g for 20 min, discard large pellet Aliquot viscous liquid, long-term storage at -20°C, 1 week at 4°C
Parafilm "M"	Sigma	P7793-1EA	
Poly(vinyl alcohol)	Sigma	P-8136-250G	Reagent to make vinol
Glycerol	Sigma	G5516-100ML	Reagent to make vinol
sodium azide	Fisher Scientific	S2002-5G	Preservative agent for blocking solution and vinol Make a 20 % dilution in water