Materials List for:

Artificial RNA Polymerase II Elongation Complexes for Dissecting Cotranscriptional RNA Processing Events

Melvin Noe Gonzalez¹, Joan W. Conaway^{1,2}, Ronald C. Conaway^{1,2}

Correspondence to: Ronald C. Conaway at RCC@stowers.org

URL: https://www.jove.com/video/59497

DOI: doi:10.3791/59497

Materials

Name	Company	Catalog Number	Comments
[α- ³² P] UTP (3000 Ci/mmol), 1 mCi	Perkin Elmer	NEG007H001MC	For radiolabeling RNA
2x RNA loading dye	New England Biolabs	B0363S	Highly recommended. For preparing RNA during gel loading
40% Bis:Acrylamide solution	Biorad	1610144	
Bovine Serum Albumin (20 mg/mL)	New England Biolabs	B9000S	
Cdk7/Cyclin H/MAT1 (CAK complex) Protein, active, 10 µg	Millipore Sigma	14-476	Used to phosphorylate Pol II CTD
DNA oligonucleotides	IDT		See Table 8 for purity specifications
Dynabeads MyOne Streptavidin C1	Life Technologies Invitrogen	65001	We have also used Dynabeads M-280 streptavidin without problem but prefer MyOne Streptavidin beads because they sediment more slowly
GlycoBlue Coprecipitant (15 mg/mL)	Life Technologies Invitrogen	AM9516	Highly recommended. Dyes nucleic-acid pellet blue making reactions much easier to handle
Hoefer SE600 standard dual cooled vertical electrophoresis system with 1 mm spacers and 15 well comb	Hoefer	SE600-15-1.5	
MaXtract high density tubes (1.5 mL)	Qiagen	129046	Highly recommended. Contains a high density gel that forms a stable barrier between aqueous and organic phases; improves RNA yields during extractions
Proteinase K solution (20 mg/mL)	Life Technologies Invitrogen	25530049	
RNA oligonucleotides	Trilink		See Table 8 for more details
Yeast Inorganic Pyrophosphatase (100 units/mL)	New England Biolabs	NEBM2403S	Required only during capping reactions

¹Stowers Institute for Medical Research

²Department of Biochemistry and Molecular Biology, Kansas University Medical Center