

# Complementation of Splicing Activity by a Galectin-3 - U1 snRNP Complex on Beads

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## Materials

Name	Company	Catalog Number	Comments
anti-U1 snRNP	The Binding Site	Hu ENA-RNP #33471	human autoimmune serum specific for U1 snRNP
bottle top vacuum filter	Fisher Scientific	Corning 431153 (0.22 µm; PES 150 ml)	for filtering solutions containing Tris
centrifuge	International Equipment Company	IEC Model PR-6	for pelleting Sepharose beads in immunoprecipitation
diethylpyrocarbonate (DEPC)	Sigma-Aldrich	159220-5G	for treatment of water used in preparation of all solutions
dimethylpimelimidate (DMP)	Sigma-Aldrich	80490-5G	for cross-linking antibody to Sepharose beads
electrophoresis cell	BioRad Laboratories, Inc	Mini-Protean II	for SDS-PAGE separation of proteins
ethanolamine	Sigma-Aldrich	411000-100ml	for blocking after the cross-linking reaction
gel electrophoresis system	Hoefer, Inc	HSI SE 500 Series	for separating snRNAs by gel electrophoresis
gel slab dryer	BioRad	Model 224	for drying gel slabs for autoradiography
Hybond ECL membrane	GE Healthcare	RPN3032D (0.2 µm; 30 cm x 3 m)	for immunoblotting of proteins on membrane
microdialyzer (12 x 100 µl sample capacity)	Pierce	Microdialyzer System 100	for exchanging the buffer of nuclear extract
microdialyzer membranes (8K cutoff)	Pierce	66310	for exchanging the buffer of nuclear extract
non-fat dry milk	Spartan Stores	Spartan Instant Non-fat Dry Milk	
Protein A Sepharose CL-4B	Millipore-Sigma	GE 17-0780-01	for coupling antibody to beads
Proteinase K	Millipore-Sigma	P2308-5mg	for stopping the splicing reaction to isolate the RNAs
RNasin	Promega	N2111	for inhibiting ribonuclease activity
rocker/rotator	Lab Industries, Inc	Labquake Shaker 400-110	for mixing protein solutions in coupling reactions and in immunoprecipitation

Safety-Solve	Research Products International Corp.	No. 111177	scintillation counting cocktail for determination of radioactivity in splicing substrate
scintillation counter	Beckman Instruments	LS6000SC	scintillation counter for determination of radioactivity
speed vacuam concentrator	Savant	SVC 100H	for drying ethanol-precipitated RNA pellets
Transphor electrophoresis unit	Hoefer, Inc	Hoefer TE Series Transphor	for protein transfer from SDS-PAGE to blotting membrane