

Materials List for:

Combining X-Ray Crystallography with Small Angle X-Ray Scattering to Model Unstructured Regions of Nsa1 from *S. Cerevisiae*

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Materials

Name	Company	Catalog Number	Comments
Molecular Cloning of Nsa1			
pMBP2 parallel vector	Sheffield et al, Protein Expression and Purification 15, 34-39 (1999)		We used a modified version of pMBP2 which included an N-terminal His-tag (pHMBP)
<i>S. cerevisiae</i> genomic DNA	ATCC	204508D-5	
Primers for cloning Nsa1			
SC_Nsa1_FLFw	IDT		CGC CAA AGG CCT ATGAGGTTACTAGTCAGCTGTGT GGATAG
SC_Nsa1_FLRv	IDT		AATGCAGCGGCCGCTCAAATTTT GCTTTTCTTACTGGCTTTAGAAGC AGC
SC_Nsa1_DeltaCFw	IDT		GGGCGCCATGGGATCCATGAGG TTACTAGTCAGCTGTGTGG
SC_Nsa1_DeltaCRv	IDT		GATTGCGAAAGCGGCCGCTTAAAC CTTCCTTTTGTCTCCC
Recombinant Protein Production and Purification of Nsa1			
<i>Escherichia coli</i> BL21 (DE3) Star Cells	Invitrogen	C601003	
pMBP- NSA1 and various truncations	Lo et al., 2017		
Selenomethionine	Molecular Dimensions	MD12-503B	
IPTG, Dioxane-Free	Promega	V3953	
EDTA Free Protease Inhibitor Cocktail	Sigma-Aldrich	4693159001	
Sodium Chloride	Caledon Laboratory Chemicals	7560-1-80	
Magnesium Chloride hexahydrate	Sigma-Aldrich	M2670	
Tris Buffer, 1 M pH7.5	KD Medical	RGF-3340	
Glycerol	Invitrogen	15514-029	
beta-mercaptoethanol	Sigma	M6250	
1M Imidazole, pH 8.0	Teknova	I6980-06	
Talon Affinity Resin	Clontech	635503	
Amicon Ultra 15 mL Centrifugal Filter (MWCO 10K)	Millipore	UFC901024	
HiLoad 16/600 Superdex 200 Prep Grade Gel Filtration Column	GE-Healthcare	28989335	

TEV Protease	Prepared by NIEHS Protein Expression Core	Expression plasmid provided by NCI (Tropea et al. Methods Mol Biology, 2009)	
4-15% Mini-PROTEAN TGX Precast Protein Gels	BioRad	456-8056	
Crystallization, Proteolytic Screening			
Crystal Screen	Hampton Research	HR2-110	
Crystal Screen 2	Hampton Research	HR2-112	
Salt Rx	Hampton Research	HR2-136	
Index Screen	Hampton Research	HR2-144	
PEG/Ion Screen	Hampton Research	HR2-139	
JCSG+	Molecular Dimensions	MD1-37	
Wizard Precipitant Synergy	Molecular Dimensions	MD15-PS-T	
Swissci 96-well 3-drop UVP sitting drop plates	TTP Labtech	4150-05823	
3inch Wide Crystal Clear Sealing Tape	Hampton Research	HR4-506	
Proti-Ace Kit	Hampton Research	HR2-429	
PEG 1500	Molecular Dimensions	MD2-100-6	
PEG 400	Molecular Dimensions	MD2-100-3	
HEPES/sodium hydroxide pH 7.5	Molecular Dimensions	MD2-011-	
Sodium Citrate tribasic	Molecular Dimensions	MD2-100-127	
22 mm x 0.22 mm Siliconized Coverslips	Hampton Research	HR3-231	
24 Well Plates with sealant (VDX Plate with Sealant)	Hampton Research	HR3-172	
18 mM Mounted Nylon Loops (0.05 mm to 0.5 mM)	Hampton Research	HR4-945, HR4-947, HR4-970, HR4-971	
Seed Bead Kit	Hampton Research	HR2-320	
Magnetic Crystal Caps	Hampton Research	HR4-779	
Magnetic Cryo Wand	Hampton Research	HR4-729	
Cryogenic Foam Dewar	Hampton Research	HR4-673	
Crystal Puck System	MiTeGen	M-CP-111-021	
Full Skirt 96 well Clear Plate	VWR	10011-228	
AxyMat Sealing Mat	VWR	10011-130	
Equipment			
UVEX-m	JAN Scientific, Inc.		
Nanodrop Lite Spectrophotometer	Thermo-Fisher		
Mosquito Robot	TTP Labtech		
Software/Websites			
HKL2000	Otwinoski and Minor, 1997		
Phenix	Adams et al., 2010		
Coot	Emsley et al., 2010		
ATSAS	Petoukhov et al., 2012		https://www.embl-hamburg.de/biosaxs/atsas-online/
Scatter	Rambo and Tainer, 2013		
Pymol	The PyMOL Molecular Graphics System, Version 1.8 Schrödinger, LLC.		

BUNCH	Petoukhov and Svergun, 2005		
CRY SOL	Svergun et al, 1995		
PRIMUS	Konarev et al, 2003		
EOM	Tria et al, 2015		