

Materials List for

# Process Evaluation and Kinetics of Recombinant Chitin Deacetylase Expression in *E. coli* Rosetta pLysS Cells Using a Statistical Technique

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jove.com/video/64590

## Materials

Name	Company	Catalog Number	Comments
<b>Kits</b>			
Acetate assay kit	Megazyme, Ireland	K-ACETAK	The protocol has been slightly modified and optimized to perform the assay in 96 well plate
Glucose estimation kit	Agappe diagnosis Ltd., India	12018013	The protocol has been slightly modified and optimized to perform the assay in 96 well plate
<b>Chemicals</b>			
Acetic acid	Hi-media, India	AS001	Used for preparing SDS-PAGE staining and destaining solution
Acrylamide	Hi-media, India	MB068	Used for preparing SDS-PAGE gel
Ammonium persulphate	Hi-media, India	MB003	Used for preparing SDS-PAGE gel
Bis-acrylamide	Hi-media, India	MB005	Used for preparing SDS-PAGE gel
Coomassie brilliant blue G-250	Hi-media, India	MB092	Used for preparing SDS-PAGE staining and destaining solution
Coomassie brilliant blue R-250	Hi-media, India	MB153	Used for preparing Bradford's assay
Ethylene glycol chitosan	Sigma-aldrich, USA	E1502	Used to prepare Ethylene glycol chitin and Ethylene glycol chitin was used as substrate for enzymatic reaction
D-glucose	Hi-media, India	MB037	Used as an media component.
Imidazole	Hi-media, India	GRM1864	Used in lysis buffer
Lactose	Hi-media, India	GRM017	Used as an media component.
Methanol	Finar, India	30930LC250	Used for preparing SDS-PAGE staining and destaining solution
Sodium chloride (NaCl)	Hi-media, India	MB023	Used in lysis buffer
Phosphoric acid	Hi-media, India	MB157	Used for preparing Bradford's assay
sodium dodecyl sulfate (SDS)	Hi-media, India	GRM6218	Used for preparing SDS-PAGE gel

Sodium phosphate dibasic anhydrous	Hi-media, India	MB024	Used to prepare TB salt for media and buffer for enzymatic reaction.
Sodium phosphate monobasic anhydrous	Hi-media, India	GRM3964	Used to prepare TB salt for media and buffer for enzymatic reaction.
Tetramethylethylenediamine (TEMED)	Hi-media, India	MB026	Used for preparing SDS-PAGE gel
Tris base	Hi-media, India	MB029	Used for preparing SDS-PAGE gel
Tryptone	Hi-media, India	RM7707	Used as a media component.
Yeast extract	Hi-media, India	RM027	Used as a media component.
<b>Equipment</b>			
Alphamager HP gel documentation unit	ProteinSimple, USA	92-13823-00	Used to capture SDS-PAGE photographs
Benchtop mixer	Eppendorf, Germany	9.776 660	Used to keep for enzymatic reaction with 2 mL adaptor
Bioincubator shaker	Trishul instruments, India	13410622	Used to incubate bacterial culture at different temperature and RPM
Biospectrophotometer	Eppendorf, Germany	6135000009	Used to take all spectroscopic readings
Cooling centrifuge	Eppendorf, Germany	5805000017	Used to centrifuge culture, lysate and all other centrifuging protocols
Dry bath	Labnet International, USA	S81522039	Used to denature protein sample for SDS-PAGE
Micropipettes	Eppendorf, Germany	3123000900	Used throughout the protocol for volume measurements
Rocker shaker	Trishul instruments, India	11770719	Used to shake SDS-PAGE gel for staining and destaining
SDS-PAGE unit	Bio-Rad, USA	1658001FC	Used to cast and run SDS-PAGE gel
Ultra sonicator	Sonics & Materials, Inc., USA	VCX 130	Used to lyse the bacterial cell by ultra sonication
Weighing balance	Sartorius, Germany	BSA124 S	Used to measure weight throughout the protocol
<b>Devices</b>			
Nanosep Centrifugal Devices with Omega Membrane (3 kDa)	PALL life sciences, USA	OD003C33	Used to separate enzyme after substrate treatment
<b>Softwares</b>			
MINITAB	Version	Developed at	
MINITAB	17.0 (Trial version)	The Pennsylvania State University	Used to design the experimental model and analyse the data
ImageJ	1.53o	National Institutes of Health (NIH)	Used to analyse the expression level using SDS-PAGE image
<b>Plasmid</b>			
pET22b (+) DNA—Novagen	Merck- Millipore, USA	69744	Stored at - 20 °C
<b>Cells</b>			
E. coli Rosetta pLysS—Novagen	Merck- Millipore, USA	70956	Maintained in Luria–Bertani (LB) broth containing 25% glycerol at - 80 °C