

Materials List for:

A Yeast 2-Hybrid Screen in Batch to Compare Protein Interactions

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Materials

Name	Company	Catalog Number	Comments
Illumina HiSeq 4000	Illumina		deep sequencing platform
Monoclonal anti-HA antibodies	Biolegend	901514	Primary Antibody to detect expression of HA in pGal4AD constructs
Polyclonal anti-myc antibodies	QED Biosciences Inc	18826	Primary Antibody to detect expression of MYC in pTEF-GBD constructs
Narl	New England BioLabs	R0191S	
EcoRI-HF	New England BioLabs	R3101S	
BamHI-HF	New England BioLabs	R3236S	
XhoI	New England BioLabs	R0146S	
Polyethylene Glycol 3350, powder	J.T. Baker	U2211-08	
Salmon Sperm DNA	Trevigen, Inc sold by Fisher Scientific	50-948-286	carrier DNA for yeast transformation section 3.2.1.
Kanamycin Monosulfate	Research Products International	K22000	
LE Agarose	GeneMate	E-3120-500	used for making DNA agarose gels
Sodium Chloride	Research Products International	S23025	
Tryptone	Research Products International	T60060	
D-Sorbitol	Research Products International	S23080	
Lithium Acetate Dihydrate	MP Biomedicals	155256	
Calcium Chloride	ThermoFisher	C79	
EDTA Sodium Salt	Research Products International	E57020	
Yeast Extract Powder	Research Products International	Y20020	
Yeast Nitrogen Base (ammonium sulfate) w/o amino acids	Research Products International	Y20040	
CSM-Trp-Leu+40ADE	Formedium	DCS0789	
CSM-Trp-Leu-His+40ADE	Formedium	DCS1169	
CSM-Leu-Met	Formedium	DCS0549	
CSM-Trp-Met	Bio 101, Inc	4520-922	
L-Methionine	Formedium	DOC0168	
Adenine	Research Products International	A11500	
D-(+)-Glucose	Research Products International	G32045	
Bacto Agar	BD	214010	used for making media plates in section 1
Peptone	Research Products International	P20240	
3-amino-1,2,4 Triazole	Sigma	A8056	
2-Mercaptoethanol (BME)	Sigma-Aldrich	M6250	

Zymolyase 100T	USBiological	Z1004	
Potassium phosphate dibasic	Sigma	P8281	
Phenol:Chloroform:IAA	Ambion	AM9732	
Ammonium Acetate	Sigma-Aldrich	238074	
Ethanol	Decon Laboratories, INC	2716	
RNAse A	ThermoFisher	EN0531	
Urea	Research Products International	U20200	
SDS	Research Products International	L22010	
glycerol	Sigma Aldrich	G5516	
Tris-HCl	Gibco	15506-017	
bromophenol blue	Amresco	449	
Gibson Assembly Cloning Kit	New England Biolabs	E5510S	Rapid assembly method for cloning of plasmids in section 2
NEBNext High-Fidelity 2x PCR Master Mix	New England Biolabs	M0541S	Used for amplification of products for Gibson Assembly in Section 2.3 as well as sample preparation for DEEPN deep sequencing in section 6.2.1
Ethidium Bromide	Amresco	0492-5G	
QIAquick PCR purification kit	Qiagen	28104	Used for purification of pcr products in section 6.2.3
Qiaquick DNA Gel Extraction Kit	Qiagen	28704	Used for purification of digested pTEF-GBD in section 2.1
KAPA Hyper Prep kit	KAPA Biosystems	KK8500	preparation kit for deep sequencing
Codon optimization	http://www.jcat.de		
Codon optimization	https://www.idtdna.com/CodonOpt		
gBlocks	Integrated DNA Technologies		DNA fragments used for cloning in Section 2.2
Strings	Thermofisher		DNA fragments used for cloning in Section 2.2
GenCatch Plasmid DNA mini-prep Kit	EPOCH Life Sciences		Used to prepare quantities of DNA in Section 2.3
Covaris E220	Covaris		high performance ultra-sonicator in section 7
oligo nucelotide 5'- CGGTCTT CAATTTCTCAAGTTTCAG -3'	Integrated DNA Technologies or Thermofisher		used for pcr amplification and sequencing 5' insert pTEF-GBD during plasmid construction
oligo nucelotide 5'-GAGTAACG ACATTCCCAGTTGTTC-3'	Integrated DNA Technologies or Thermofisher		used for pcr amplification and sequencing 5' insert pTEF-GBD during plasmid construction
oligo nucelotide 5'-CACCGTAT TTCTGCCACCTCTTCC-3'	Integrated DNA Technologies or Thermofisher		used for pcr amplification and sequencing 3' insert pTEF-GBD during plasmid construction
oligo nucelotide 5'-GCAACCGC ACTATTTGGAGCGCTG-3'	Integrated DNA Technologies or Thermofisher		used for pcr amplification and sequencing 3' insert pTEF-GBD during plasmid construction
oligonucleotide 5'-GTTCCGATG CCTCTGCGAGTG-3'	Integrated DNA Technologies or Thermofisher		5' Pimer used for insert amplification of pGAL4AD
oligonucleotide 5'-GCACATGCT AGCGTCAAATACC-3'	Integrated DNA Technologies or Thermofisher		3' Pimer used for insert amplification of pGAL4AD
oligonucleotide 5'-ACCCAAGCA GTGGTATCAACG-3'	Integrated DNA Technologies or Thermofisher		5' Pimer used for insert amplification of pGADT7

oligonucleotide 5'- TATTTAGA AGTGTCAACAACGTA -3'	Integrated DNA Technologies or ThermoFisher		3' Primer used for insert amplification of pGADT7
PJ69-4A MatA yeast strain	http://depts.washington.edu/yeastrc/ James P, Halladay J, Craig EA: Genomic Libraries and a host strain designed for highly efficient two-hybrid selection in yeast. GENETICS 1996 144:1425-1436		MATA leu2-3,112 ura3-52 trp1-901 his3-200 gal4D, gal80D, GAL-ADE2 lys2::GAL1-HIS3 met2::GAL7
pTEF-GBD	Dr. Robert Piper Lab		Gal4-DNA binding domain expression plasmid
pGal4AD (pPL6343)	Dr. Robert Piper Lab		Gal4-activation domain expression plasmid
100 mm petri dishes	Kord-Vallmark sold by VWR	2900	
125 mL PYREX Erlenmeyer flask	Fisher Scientific	S63270	
250 mL PYREX Erlenmeyer flask	Fisher Scientific	S63271	
1,000 mL PYREX Erlenmeyer flask	Fisher Scientific	S63274	
2,000 mL PYREX Erlenmeyer flask	Fisher Scientific	S63275	
20 X 150 mm Disposable Culture Tube	ThermoFisher	14-961-33	
pipet-aid	Drummond	4-000-100	
5 mL Serological Pipette	Denville	P7127	
10 mL Serological Pipette	Denville	P7128	
25 mL Serological Pipette	Denville	P7129	
1,000 mL PYREX Griffin Beaker	Fisher Scientific	02-540P	
1,000 mL PYREX Reusable Media Storage Bottle	Fisher Scientific	06-414-1D	
1,000 mL graduated cylinder	Fisher Scientific	08-572-6G	
SpectraMax 190	Molecular Devices		used to measure the Optical Density of cells
NanoDrop 2000	Thermo Scientific	ND-2000	Spectrophotometer used to quantify amount of DNA
Electronic UV transilluminator	Ultra Lum	MEB 20	used to visualize DNA in an Ethidium Bromide agarose gel
P1000 Gilson PIPETMAN	Fisher Scientific	F123602G	
P200 Gilson PIPETMAN	Fisher Scientific	F123601G	
P20 Gilson PIPETMAN	Fisher Scientific	F123600G	
P10 Gilson PIPETMAN	Fisher Scientific	F144802G	
1250 µL Low Retention Pipette Tips	GeneMate	P-1236-1250	
200 µL Low Retention Pipette Tips	VWR	10017-044	
10 µL XL Low Retention Pipette Tips	VWR	10017-042	
50 mL conical tube	VWR	490001-627	
15 mL conical tube	VWR	490001-621	
cell scraper	Denville Scientific	TC9310	
1.5 mL Microcentrifuge tubes	USA Scientific	1615-5500	
HCl	Fluka Analytical	318949-1L	
NaOH	J.T. Baker	5674-02	
Wooden applicators	Solon Care	55900	
Eppendorf microcentrifuge 5424	Fisher Scientific	05-400-005	microcentrifuge
Sorvall ST16R	Thermo Fisher Scientific	75004381	benchtop centrifuge

Amersham ECL Rabbit IgG, HRP-linked whole Ab (from donkey)	GE Healthcare	NA934-1ML	Secondary Antibody
Amersham ECL Mouse IgG, HRP-linked whole Ab (from sheep)	GE Healthcare	NA931-1ML	Secondary Antibody
SuperSignal West Pico Chemiluminescent Substrate	Thermo Fisher Scientific	34080	ECL detection solution
Isotemp Incubator	Thermo Fisher Scientific		Incubator
Mutitron 2	INFORS HT		Shaking incubator
Isotemp Digital-Control Water Bath Model 205	Fisher Scientific		water bath
Y2H mouse cDNA library in Y187 (pan tissue)	Clontech	630482	commercially available cDNA Library
Y2H mouse cDNA library in Y187 (mouse brain)	Clontech	630488	commercially available cDNA Library
pGADT7 AD Vector	Clontech	630442	commercially available AD Vector housing many cDNA libraries
pGBKT7 DNA-BD Vector	Clontech	630443	commercially available DNA-BD Vector
Biolase DNA Polymerase	Bioline	BIO-21042	DNA polymerase used for section 2.4
GeneMate GCL-60 Thermal Cycler	BioExpress	P-6050-60	pcr machine
TempAssure 0.5 mL PCR tubes	USA Scientific	1405-8100	