

Materials List for:

Using Sniper-Cas9 to Minimize Off-target Effects of CRISPR-Cas9 Without the Loss of On-target Activity Via Directed Evolution

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URL: <https://www.jove.com/video/59202>

DOI: [doi:10.3791/59202](https://doi.org/10.3791/59202)

Materials

Name	Company	Catalog Number	Comments
Alkaline Phosphatase, Calf Intestinal (CIP)	NEB	M0290L	
Ampicillin Sodium Salt	Fisher Chemical	BP1760-25	
Anhydrotetracycline hydrochloride	Sigma Aldrich	37919 or 94664	
Antibiotic Antimycotic solution	Welgene	LS203-01	Media component
BamHI	Enzynomics	R003L	
Chloramphenicol	Sigma Aldrich	R4408	
Diversify PCR Random Mutagenesis	Clontech	630703	Error-prone PCR kit
DNA-Shuffling Kit	Jena Bioscience	PP-103	
Dulbecco's Modified Eagle's Medium (DMEM)	Welgene	LM001-17	Culture media
Endura Electrocompetent Cells (DUO)	Lucigen	60242-2	<i>E. coli</i> electrocompetent cells for library preparation
Fetal Bovine Serum (FBS)	Welgene	S101-01	Media component
Gene Pulser II	Bio-Rad		<i>E. coli</i> electroporation equipment
GeneMorph II Random Mutagenesis Kit	Agilent	200550	Error-prone PCR kit
genomic DNA prep kit	GenAll	106-152	gDNA-prep kit
Glycerol	BioShop	GLY001.1	
GP/MP Cuvette, 0.1cm	Bio-Rad	BR165-2089	<i>E. coli</i> electroporation equipment
HEK293T cell	ATCC	CRL-11268	Human cell line
Kanamycin sulfate	Acros Organics	450810500	
L-(+)-Arabinose	Sigma Aldrich	A3256-25G	
LaboPass PCR Purification Kit	Cosmogenetech	CMR0112	
LaboPass Plasmid Mini	Cosmogenetech	CMP0112	Mini-prep kit
LB Agar Miller	Formedium	LMM0202	
LB Broth Miller	Formedium	LMM0102	
Lipofectamine	Invitrogen	11668019	Transfection reagent
NEBuilder HiFi DNA Assembly Master Mix	NEB	E2621L	Gibson assembly (isothermal <i>in vitro</i> recombination) mixture
Neon Transfection System	Thermo	MPK5000	RNP Electroporation equipment
Neon Transfection System 10 µL Kit	Thermo	MPK1096	RNP Electroporation equipment
NucleoBond Xtra Midi EF	Macherey-Nagel	740420.50	Midi-prep kit

Oligo Clean & Concentrator	Zymo Research	D4061	DNA purification kit that enables low-volume elution
Opti-MEM	Gibco	LS31985070	Transfection media
p11-lacY-wtx1	Addgene	#123945	
pgrg36	Addgene	#16666	<i>E. coli</i> mutator strain
Phusion High-Fidelity DNA Polymerase	NEB	M0530L	
Pyrophosphatase	NEB	M2403L	
Ribonucleotide Solution Set	NEB	N0450L	
RNase inhibitor	NEB	M0314L	
T7 RNA polymerase	NEB	M0251L	
Turbo Dnase	Ambion	AM2238	
XbaI	Enzynomics	R013L	
XL1-Red Competent Cells	Agilent	200129	