

Materials List for:

In Vitro Synthesis of Modified mRNA for Induction of Protein Expression in Human Cells

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Materials

Name	Company	Catalog Number	Comments
Cell culture			
DMEM, high glucose	PAA	E15-009	
FBS	Life Technologies	10500	
Penicillin/Streptomycin	PAA	P11-010	100x
L-glutamine	PAA	M11-004	200 mM
DPBS without calcium and magnesium	PAA	E15-002	
0.04% Trypsin / 0.03% EDTA	Promocell	C-41020	
TNS	Promocell	C-41120	Trypsin Neutralizing Solution, 0.05% trypsin inhibitor in 0.1% BSA
HEK-293 cells	ATCC	CRL-1573	
Consumables			
Tissue culture plates, 12-well	Corning	3512	
Cell culture flask (75 cm ²)	Corning	430641	
DNase-and RNase-free 1.5 ml sterile microcentrifuge tubes	Eppendorf	0030 121.589	Safe-Lock, Biopur
15 ml conical tubes	greiner bio-one	188271	
PCR clean and sterile epT.I.P.S. dualfilter pipette tips	Eppendorf	10 µl: 022491202; 100 µl: 022491237; 1,000 µl: 022491253	
Cryovial	greiner bio-one	122279-128	Cryo.s
14 ml polypropylene round bottom tube for bacterial culture	BD Falcon	352059	
Plasmid amplification and purification			
pcDNA 3.3_eGFP Plasmid	Addgene	26822	
One Shot Top10 chemically component Escherichia coli	Invitrogen	C4040-10	
Sterile water (Ampuwa)	Fresenius Kabi	1636071	
LB medium (Luria/Miller)	Carl Roth	X968.1	Dissolve 25 g L ⁻¹ in sterile water.
LB agar (Luria/Miller)	Carl Roth	X969.1	Dissolve 40 g L ⁻¹ in sterile water.
Ampicillin Ready Made Solution	Sigma Aldrich	A5354	100 mg/ml
Glycerol	Sigma Aldrich	G2025	
QIAprep Spin Miniprep Kit	Qiagen	27104	
mRNA production			
HotStar HiFidelity Polymerase Kit	Qiagen	202602	
QIAquick PCR Purification Kit	Qiagen	28106	

MEGAscript T7 Kit	Life Technologies	AM1334	
5-Methylcytidine-5'-triphosphate	Trilink	N1014	5-Methyl-CTP
Pseudouridine-5'-triphosphate	Trilink	N1019	Pseudo-UTP
3'-O-Me-m ⁷ G(5')ppp(5')G RNA cap structure analog	New England Biolabs	S1411L	
RiboLock RNase Inhibitor	Thermo Scientific	EO0381	40 U/μl
TURBO DNase	Life Technologies	AM1334	2 U/μl (from MEGAscript T7 Kit)
Antarctic phosphatase	New England Biolabs	MO289S	
RNeasy mini kit	Qiagen	74104	
RNaseZap solution	Life Technologies	AM9780	
Transfection			
Lipofectamine 2000 Transfection Reagent	Invitrogen	11668-019	Cationic lipid transfection reagent
Opti-MEM I Reduced Serum Media	Invitrogen	11058-021	Improved Minimal Essential Medium (MEM) that allows a reduction of Fetal Bovine Serum (FBS) supplementation
Gel electrophoresis			
Agarose	Sigma-Aldrich	A9539	
Gelred Nucleic Acid Gel Stain	Biotium	41003	10,000x in water
peqGOLD Range Mix DNA-Ladder	Peqlab	25-2210	
Flow cytometry analyses			
CellFIX (1x)	BD Biosciences	340181	10x concentrate
Primer for insert amplification and poly (T) tail PCR			
Forward Primer (HPLC-grade) 10 μM	Ella Biotech		5'-TTGGACCCTCGTAC AGAAGCTAATACG-3'
Reverse Primer (HPLC-grade) 10 μM	Ella Biotech		5'-T ₁₂₀ -CTTCCTACT CAGGCTTTATTCAA AGACCA-3'
Equipment			
Cell incubator	Binder		CO ₂ (5%) and O ₂ (20%)
CASY cell counter	Schärfe System		
Sterile workbench	BDK Luft-und Reinraumtechnik GmbH		
Bacterial incubator	Incutec		
Water bath			
ScanDrop spectrophotometer	Analytic Jena		
PCR thermocycler	Eppendorf		
Microcentrifuge	Eppendorf		
Vortex	peqlab		
Thermomixer	Eppendorf		
Gel apparatus for electrophoresis	Bio-Rad		
Gel documentation system	Bio-Rad		
FACScan System	BD Biosciences		
Fluorescence microscope	Nikon		
Phase-contrast microscope	Zeiss		