

Materials List for:

CRISPR-Cas9-Mediated Genome Editing in the Filamentous Ascomycete *Huntiella omanensis*

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Materials

Name	Company	Catalog Number	Comments
EcoRI-HF	New England Biolabs, Ipswich, USA	R3101S	
EnGen Spy Cas9 NLS protein	New England Biolabs, Ipswich, USA	M0646T	Used to assemble the RNP
Eppendorf 5810 R centrifuge	Eppendorf, Hamberg, Germany		
FastStart Taq DNA Polymerase	Sigma, St Louis, USA	12032902001	Standard DNA polymerase
GeneJET Gel Extraction Kit	ThermoFisher Scientific, Waltham, USA	K0691	
HindIII-HF	New England Biolabs, Ipswich, USA	R3104S	
HiScribe TM T7 Quick High Yield RNA synthesis kit	New England Biolabs, Ipswich, USA	E2050S	
Hygromycin B from <i>Streptomyces hygroscopicus</i>	Sigma, St Louis, USA	10843555001	
Infors HT Ecotron Shaking Incubator	Infors AG, Bottmingen, Switzerland		
LongAmp Taq DNA Polymerase	New England Biolabs, Ipswich, USA	M0323S	Long-range, high-fidelity DNA polymerase
Malt extract agar, 2% (MEA)			20 g ME and 20 g agar in 1 l ddH ₂ O
Malt extract	Sigma, St Louis, USA	70167-500G	
Agar	Sigma, St Louis, USA	A5306	
Malt Extract broth, 1% (MEB)	Sigma, St Louis, USA	70167-500G	2 g ME in 200 ml ddH ₂ O
Malt Extract broth, 2% (MEB)	Sigma, St Louis, USA	70167-500G	4 g ME in 200 ml ddH ₂ O
Miracloth	Merck Millipore, New Jersey, USA	475855	
Nylon membrane (positively charged)	Sigma, St Louis, USA	11209299001	
Osmotic control medium (OCM)			0.3% yeast extract, 20% sucrose, 0.3% casein hydrolysate
Casein Hydrolysate	Sigma, St Louis, USA	22090	
Sucrose	Sigma, St Louis, USA	84097	
Yeast extract	Sigma, St Louis, USA	Y1625	
Osmotic control medium (OCM) agar			Osmotic control medium (OCM) + 1% agar
Agar	Sigma, St Louis, USA	A5306	
PCR DIG Labeling Mix	Sigma, St Louis, USA	11585550910	

Phusion High-Fidelity DNA Polymerase	ThermoFisher Scientific, Waltham, USA	F-530XL	High fidelity DNA polymerase
Plasmid pcb1004	N/A	N/A	From: Carroll et al., 1994
Presynthesized sgRNA	Inqaba Biotec, Pretoria, South Africa		Ordered as an synthesized dsDNA with specified sequence
Proteinase K	Sigma, St Louis, USA	P2308	
PTC Solution			30% polyethylene glycol 8000 in STC buffer from above
Polyethylene glycol 8000	Sigma, St Louis, USA	1546605	
RNase A	ThermoFisher Scientific, Waltham, USA	12091021	
RNAfold Webserver	Institute for Theoretical Chemistry, University of Vienna	N/A	http://rna.tbi.univie.ac.at/cgi-bin/RNAWebSuite/RNAfold.cgi
RNAstructure	Mathews Lab	N/A	https://rna.urmc.rochester.edu/RNAstructureWeb/Servers/Predict1/Predict1.html
Sorbitol, 1 M	Sigma, St Louis, USA	1617000	182.17g sorbitol in 1 l ddH2O
STC Buffer			20% sucrose, 50 mM Tris-HCl pH 8.00 and 50 mM CaCl ₂
Calcium chloride	Sigma, St Louis, USA	429759	
Tris-HCl pH 8.00	Sigma, St Louis, USA	10812846001	
Sucrose	Sigma, St Louis, USA	84097	
<i>Trichoderma harzianum</i> lysing enzymes	Sigma, St Louis, USA	L1412	
Zeiss Axioskop 2 Plus Ergonomic Trinocular Microscope	Zeiss, Oberkochen, Germany		