

Materials List for:

In Vivo Augmentation of Gut-Homing Regulatory T Cell Induction

Hongzheng Bi^{1,2}, Samiksha Wasnik¹, David J. Baylink¹, Chenfan Liu^{1,3,4}, Xiaolei Tang^{1,3}

¹Division of Regenerative Medicine, Department of Medicine, Loma Linda University

²Zhengzhou University

³Department of Veterinary Biomedical Sciences, College of Veterinary Medicine, Long Island University

⁴Jinan Infectious Disease Hospital, Shandong University

Correspondence to: Xiaolei Tang at xiaolei.tang@liu.edu

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Materials

Name	Company	Catalog Number	Comments
10 mL syringes	ThermoFisher Scientific	Cat# 03-377-23	
100 mm x 20 mm culture dishes	Sigma-Aldrich	Cat# CLS430167	
12-well culture plates	ThermoFisher Scientific	Cat# 07-200-82	
150 mm x 25 mm culture dishes	Sigma-Aldrich	Cat# CLS430559	
25-hydroxycholecalciferol (25[OH]D)	Sigma-Aldrich	Cat# H4014	
293T cells	ATCC	CRL-3216	
2-mercaptoethanol	ThermoFisher Scientific	Cat#: 21985023	
6-well culture plates	ThermoFisher Scientific	Cat# 07-200-83	
ALDEFLUOR kit	Stemcell Technologies	Cat# 01700	
Anti-CYP27B1	Abcam	Cat# ab95047	
BD FACSAria II	BD Biosciences	N/A	
CaCl ₂	Sigma-Aldrich	Cat# C1016	
CM-10-D cell culture medium			DMEM medium containing 10% fetal bovine serum (FBS), 100 U/ml penicillin/streptomycin, 0.055 mM 2-mercaptoethanol (2-ME), 1 mM sodium pyruvate, 0.1 mM nonessential amino acid, and 2 mM L-glutamine.
CM-10-R cell culture medium			RPMI 1640 medium (no glutamine) containing 10% fetal bovine serum (FBS), 100 U/ml penicillin/streptomycin, 0.055 mM 2-mercaptoethanol (2-ME), 1 mM sodium pyruvate, 0.1 mM nonessential amino acid, and 2 mM L-glutamine.
CM-4-D cell culture medium			DMEM medium containing 4% fetal bovine serum (FBS), 100 U/ml penicillin/streptomycin, 0.055 mM 2-mercaptoethanol (2-ME), 1 mM sodium pyruvate, 0.1 mM nonessential amino acid, and 2 mM L-glutamine.
Corning bottle-top vacuum filters, 0.22 μm, 500 mL	Sigma-Aldrich	Cat# CLS430513	
Corning bottle-top vacuum filters, 0.45 μm, 500 mL	Sigma-Aldrich	Cat# CLS430514	
Dissecting scissor	ThermoFisher Scientific	Cat# 08-940	

DMEM medium	ThermoFisher Scientific	Cat# 11960044	
Fetal bovine serum	ThermoFisher Scientific	Cat# 16000044	
Forceps	ThermoFisher Scientific	Cat# 22-327379	
Gibco ACK lysing buffer	ThermoFisher Scientific	Cat# A1049201	
Glycerol	Sigma-Aldrich	Cat# G5516	
Goat anti-rabbit IgG	Abcam	Cat# ab205718	
HEPES	Millipore	Cat# 391340	
Lenti-CYP-ALDH	Custom-made	1.6-kb mouse CYP27B1 and ALDH1a2 cDNAs were amplified by PCR using a plasmid containing the CYP27B1 cDNA and a plasmid containing the ALDH1a2 cDNA respectively (GeneCopoeia). The amplified CYP27B1 cDNA fragment with a 5' KOZAK ribosome entry sequence was cloned into the pRRL-SIN.cPPT.PGKGFP.WPRE lentiviral vector (Addgene). The resulting construct was designated as lenti-CYP-GFP. The amplified ALDH1a2 cDNA fragment was cloned into the lenti-CYP-GFP to replace the GFP and was designated as lenti-CYP-ALDH. This bicistronic plasmid expresses CYP27B1 controlled by SFFV promoter and ALDH1a2 controlled by PGK promoter.	
L-glutamine	ThermoFisher Scientific	Cat#25030081	
Lipopolysaccharide	Sigma-Aldrich	Cat# L3755	
Murine GM-CSF	Peptotech	Cat# 315-03	
Murine IL-4	Peptotech	Cat# 214-14	
Na ₂ HPO ₄	Sigma-Aldrich	Cat# NIST2186II	
NaCl	Sigma-Aldrich	Cat# S9888	
Needles	ThermoFisher Scientific	Cat# 14-841-02	
Nonessential Amino Acids	ThermoFisher Scientific	Cat#: 11140076	
pCMVR8.74	Addgene	Plasmid# 22036	
Penicillin/Streptomycin	ThermoFisher Scientific	Cat#15140148	
Phosphate Balanced Solution (PBS)	ThermoFisher Scientific	Cat#: 20012027	
PMD2G	Addgene	Plasmid# 12259	
Polypropylene tube, 15 mL	ThermoFisher Scientific	Cat# AM12500	
Polypropylene tube, 50 mL	ThermoFisher Scientific	Cat# AM12502	
Protamine sulfate	Sigma-Aldrich	Cat# P3369	
Rabbit polyclonal IgG isotype control	Abcam	Cat# ab171870	
Radioimmunoassay for 1,25(OH) ₂ D measurement	Heartland Assays		
RPMI 1640 medium, no glutamine	ThermoFisher Scientific	Cat# 21870076	
Sodium pyruvate	ThermoFisher Scientific	Cat#: 11360070	
Sorvall Legend XTR Centrifuge	ThermoFisher Scientific	Cat# 75004521	
Sterile Cell strainers, 40 mm	ThermoFisher Scientific	Cat# 07-201-430	
Sterile storage bottles, 500 mL	ThermoFisher Scientific	Cat# CLS431432	