

Establishing Human Lung Organoids and Proximal Differentiation to Generate Mature Airway Organoids

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Materials

Name	Company	Catalog Number	Comments
Reagents for lung organoid culture			
Advanced DMEM/F12	Invitrogen	12634010	-
A8301	Tocris	2939	500nM
B27 supplement	Invitrogen	17504-044	1x
Cultrex Reduced Growth Factor Basement Membrane Matrix, Type 2 (BME 2)	Trevigen	3533-010-0	70-80%
FGF-10	Peptotech	100-26	20 ng/mL
FGF-7	Peptotech	100-19	5 ng/mL
GlutaMAX (glutamine)	Invitrogen	35050061	1x
HEPES 1M	Invitrogen	15630-056	10 mM
Heregulin β -1	Peptotech	100-03	5 nM
N-Acetylcysteine	Sigma-Aldrich	A9165	1.25 mM
Nicotinamide	Sigma-Aldrich	N0636	10 mM
Noggin (conditional medium)	home made	-	10x
Penicillin-Streptomycin (10,000 U/ mL)	Invitrogen	15140-122	1x
Primocin	Invivogen	ant-pm-1	100 μ g/mL
Rspondin1 (conditional medium)	home made	-	10x
SB202190	Sigma-Aldrich	S7067	1 μ M
Y-27632	Tocris	1254	5 μ M
Proximal differentiation medium			
DAPT	Tocris	2634	10 μ M
Heparin Solution	StemCell Technology	7980	4 μ g/mL
Hydrocortisone Stock Solution	StemCell Technology	7925	1 μ M
PneumaCult-ALI 10X Supplement			air liquid interface supplement
PneumaCult-ALI Basal Medium	StemCell Technology	05001	air liquid interface basal medium

PneumaCult-ALI Maintenance Supplement			air liquid interface maintenance supplement
Y-27632	Tocris	1254	10 μ M
Equipment			
Biological safety cabinet	Baker	1-800-992-2537	
Carl Zeiss LSM 780 or 800	Zeiss		confocal microscope
CO2 Incubator	Thermo Fisher Scientific	42093483	
Stereo-microscope	Olympus Corporation	CKX31SF	
Centrifuge	Eppendorf	5418BG040397	
Serological pipettor	Eppendorf		
Micropipette	Eppendorf		
ZEN black or ZEN blue software	Zeiss		analysis software
Consumables			
12mm Trans-well	StemCell Technology	#38023	
12-well cell culture plate	Cellstar	665970	
15- and 50 ml conical tubes	Thermo Fisher Scientific	L6BF5Z8118	
24-well cell culture plate	Cellstar	662160	
6.5mm Trans-well	StemCell Technology	#38024	
Medical Syringe Filter Unit, 0.22 μ m	Sigma-Aldrich	SLGPR33RB	
Microfuge tubes	Eppendorf		
Micropipette tips	Thermo Fisher Scientific	TFLR140-200-Q21190531	
Pasteur pipette glass	Thermo Fisher Scientific	22-378893	
Serological pipettes(5ml, 10ml, 25ml)	Thermo Fisher Scientific	BA08003, 08004, 08005	
Antibodies			
Goat Anti-Mouse Alexa Fluor 594	Invitrogen	A11005	
Goat Anti-Mouse, Alexa Fluor 488	Invitrogen	A11001	
Goat Anti-Rabbit Alexa Fluor 488	Invitrogen	A11034	
Goat Anti-Rabbit Alexa Fluor 594	Invitrogen	A11037	
Goat Anti-Rat Alexa Fluor 594	Invitrogen	A11007	
Mouse Anti-Cytokeratin 5	Abcam	ab128190	
Mouse Anti-FOX J1	Invitrogen	14-9965-82	
Mouse Anti-Mucin 5AC	Abcam	ab3649	
Mouse Anti- β -tubulin 4	Sigma	T7941	
Rabbit Anti-p63	Abcam	ab124762	
Rat Anti-Uteroglobin/CC-10	R&D Systems	MAB4218-SP	
Other reagent			
TrypLE Select Enzyme (10X)	Thermo Fisher Scientific	A1217701	dissociation enzyme