

Establishing 3D Endometrial Organoids from the Mouse Uterus

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Materials

Name	Company	Catalog Number	Comments
Organoid Media Formulation			
Name	Company	Catalog Number	Final concentration
Corning Matrigel Growth Factor Reduced (GFR) Basement Membrane Matrix, *LDEV-free	Corning	354230	100%
Trypsin from Bovine Pancreas	Sigma Aldrich	T1426-1G	1%
Advanced DMEM/F12	Life Technologies	12634010	1X
N2 supplement	Life Technologies	17502048	1X
B-27™ Supplement (50X), minus vitamin A	Life Technologies	12587010	1X
Primocin	Invivogen	ant-pm-1	100 µg/mL
N-Acetyl-L-cysteine	Sigma Aldrich	A9165-5G	1.25 mM
L-glutamine	Life Technologies	25030024	2 mM
Nicotinamide	Sigma Aldrich	N0636-100G	10 nM
ALK-4, -5, -7 inhibitor, A83-01	Tocris	2939	500 nM
Recombinant human EGF	Peprtech	AF-100-15	50 ng/mL
Recombinant human Noggin	Peprtech	120-10C	100 ng/mL
Recombinant human Rspodin-1	Peprtech	120-38	500 ng/mL
Recombinant human FGF-10	Peprtech	100-26	100 ng/mL
Recombinant human HGF	Peprtech	100-39	50 ng/mL
WNT3a	R&D systems	5036-WN	200 ng/mL
Other supplies and reagents			
Name	Company	Catalog Number	Final concentration
Collagenase from Clostridium histolyticum	Sigma Aldrich	C0130-1G	5 mg/mL
Deoxyribonuclease I from bovine pancreas	Sigma Aldrich	DN25-100MG	2 mg/mL
DPBS, no calcium, no magnesium	ThermoFisher	14190-250	1X
HBSS, no calcium, no magnesium	ThermoFisher	14170112	1X

Falcon Polystyrene Microplates (24-Well)	Fisher Scientific	#08-772-51	
Falcon Polystyrene Microplates (12-Well)	Fisher Scientific	#0877229	
Falcon Cell Strainers, 40 µm	Fisher Scientific	#08-771-1	
Direct-zol RNA MiniPrep (50 µg)	Genesee Scientific	11-331	
Trizol reagent	Invitrogen	15596026	
DMEM/F-12, HEPES, no phenol red	ThermoFisher	11039021	
Fetal Bovine Serum, Charcoal stripped	Sigma Aldrich	F6765-500ML	2%
Estradiol (E2)	Sigma Aldrich	E1024-1G	10 nM
Formaldehyde 16% in aqueous solution, EM Grade	VWR	15710	4%
Epredia Cassette 1 Slotted Tissue Cassettes	Fisher Scientific	1000961	
Epredia Stainless-Steel Embedding Base Molds	Fisher Scientific	64-010-15	
Ethanol, 200 proof (100%)	Fisher Scientific	22-032-601	
Histoclear	Fisher Scientific	50-899-90147	
PermOUNT Mounting Medium	Fisher Scientific	50-277-97	
Epredia Nylon Biopsy Bags	Fisher Scientific	6774010	
HistoGel Specimen Processing Gel	VWR	83009-992	
Hematoxylin solution Premium	VWR	95057-844	
Eosin Y (yellowish) solution Premium	VWR	95057-848	
TBS Buffer, 20X, pH 7.4	GenDEPORT	T8054	1X
TBST (10X), pH 7.4	GenDEPORT	T8056	1X
Citric acid	Sigma Aldrich	C0759-1KG	
Sodium citrate tribasic dihydrate	Sigma Aldrich	S4641-500G	
Tween20	Fisher Scientific	BP337-500	
Bovine Serum Albumin (BSA)	Sigma Aldrich	A2153-100G	3%
DAPI Solution (1 mg/mL)	ThermoFisher	62248	1:1000 dilution
VECTASHIELD Antifade Mounting Medium	Vector Labs	H-1000-10	
Clear Nail Polish	Fisher Scientific	NC1849418	
Fisherbrand Superfrost Plus Microscope Slides	Fisher Scientific	22037246	
VWR Micro Cover Glasses	VWR	48393-106	
SuperScript VILO Master Mix	ThermoFisher	11755050	
SYBR Green PCR Master Mix	ThermoFisher	4364346	
Krt8 Antibody (TROMA-I)	DSHB	TROMA-I	1:50 dilution
Vimentin Antibody	Cell Signaling	5741S	1:200 dilution
Donkey anti-Rat IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor 594	ThermoFisher	A-21209	1:250 dilution
Donkey anti-Rabbit IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor 488	ThermoFisher	A-21206	1:250 dilution
ZEISS Stemi 508 Stereo Microscope	ZEISS		
ZEISS Axio Vert.A1 Inverted Routine Microscope with digital camera	ZEISS		
Primer Sequence	Forward (5'-3')	Reverse (5'-3')	—
Lipocalin 2 (Lcn2)	GCAGGTGGTACGTTGTGGG	CTCTTGAGCTCATAGATGGTGC	

Lactoferrin (Ltf)	TGAGGCCCTTGGACTCTGT	ACCCACTTTTCTCATCTCGTTC	
Progesterone (Pgr)	CCCACAGGAGTTTGTCAAGCTC	TAACTTCAGACATCATTCCGG	
Glyceraldehyde 3 phosphate dehydrogenase (Gapdh)	CAATGTGTCCGTCGTGGATCT	GCCTGCTTCACCACCTTCTT	