

Materials List for:

Isolation of Stem-like Cells from 3-Dimensional Spheroid Cultures

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Materials

Name	Company	Catalog Number	Comments
0.05% Trypsin-EDTA	Gibco	25300-054	
1 mL tuberculin syringes	Bectin Dickinson	BD 309625	
1.5 mL microcentrifuge tubes, sterile			
100 mm culture dishes	Corning/Falcon	353003	
12-well culture plate	Corning/Falcon	353043	
15 mL centrifuge tubes	Corning/Falcon	352097	
22 x 22 mm coverslips, sq	Corning	284522	For MatTek 35 mm culture dish
24 x 50 mm coverslips	Corning	2975245	
26G x 1.5 inch hypodermic needle	Monoject	1188826112	
2N HCl			
35 mm culture dish with cover glass bottom	MatTek Corp	P35G-0-10-C	Glass bottom No. 0, uncoated, γ irradiated
40 μ m pore nylon cell strainer	Corning	352340	
5% CO ₂ culture incubator, 37 °C	Forma		
50 mL centrifuge tubes	Corning/Falcon	352098	
5mL Polystyrene Round-Bottom Tube with strainer snap cap	Corning	352235	35 μ m nylon mesh
6-well culture plates	Corning	353046	
8-well chamber slides	Millipore Sigma	PEZGS0816	
Aqueous mounting medium containing DAPI	Vector Laboratories	H-1200	A nuclear fluorescent dye
Biological safety cabinet, Level 2 certified			
BrdU (5-bromo-2'-deoxyuridine)	Sigma-Aldrich	B5002	1 mM stock solution in DMSO
Centrifuge for 1.5 mL microcentrifuge tubes	Eppendorf		
Centrifuge for 15 mL tubes	Beckman Coulter		Allegra 6
CFSE (carboxyfluorescein succinimidyl ester)	Thermo Fisher Scientific	C34554	5 mM stock solution in DMSO
cytochalasin D	Thermo Fisher Scientific	PHZ1063	
Dispase 1U/mL	StemCell Technologies	07923	
FACS CellSorter MoFlo XDP	Beckman Coulter	s	
Far-Red pro-dye	Thermo Fisher	C34564	5 mM stock solution in DMSO
Fetal Bovine Serum (FBS)			
Fibronectin	Sigma-Aldrich	F0895	For coating 100 mm culture dishes

Fluorescent microscope with color digital camera	Carl Zeiss		Axioskop 20 fluorescent microscope; color digital Axiocamera
Goat anti-Mouse IgG (H+L) Highly Cross-Adsorbed Secondary Antibody, Alexa Fluor 488	Thermo Fisher	A-11029	
HPrEC (Primary normal human prostate epithelial cells)	Lifeline Cell Technology	FC-0038	Pooled from 3 young (19-21yr od) disease-free organ donors; 1×10^5 cells/mL; stored in liquid nitrogen
ice bucket and ice			
Inverted microscope with digital camera			
Matrigel, low growth factor, phenol-red free	Corning	356239	
Methanol	Corning	A452-4	
Mouse anti-BrdU antibody	Cell Signaling	5292S	
Mouse IgG antibody (negative control)	Santa Cruz Biotechnology	sc-2025	
Normal goat serum	Vector Laboratories	S-1000	
Phosphate Buffered Saline (PBS), pH 7.4	Sigma-Aldrich	P5368-10PAK	
Pipettors and tips, various sizes			
PrEGM (ProstaLife Epithelial Cell Growth Medium)	Lifeline Cell Technology	LL-0041	
Propidium Iodide (PI)	R & D Systems	5135/10	10 μ g/mL PI in PBS stored at 4 °C in the dark
Serological pipets, various sizes			
Software for sphere counting and size measurements			
Software: 3D images using Imaris an imageing software with freeform drawing capabilities			
Triton X-100	Millipore Sigma	T8787	
Water bath, 37 °C			
z-stack images using a transmitted light inverted fluorescent confocal microscope			