

Microfluidic Co-Culture Models for Dissecting the Immune Response in in vitro Tumor Microenvironments

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Materials

Name	Company	Catalog Number	Comments
Cell culture materials			
50 mL tubes	Corning-Sigma Aldrich, St. Louis, MO	CLS430828	centrifuge tubes
5-aza-2'-deoxycytidine DAC	Millipore-Sigma; St. Louis, MO	A3656	DNA-hypomethylating agent
6-well plates	Corning-Sigma Aldrich, St. Louis, MO	CLS3506	culture dishes
75 cm ² cell culture treated flask	Corning, New York, NY	430641U	culture flasks
A365M	American Type Culture Collection (ATCC), Manassas, VA	CVCL_B222	human melanoma cell line
Doxorubicin hydrochloride	Millipore-Sigma; St. Louis, MO	D1515	anthracycline antibiotic
Dulbecco's Modified Eagle Medium DMEM	EuroClone Spa, Milan, Italy	ECM0728L	Culture medium for SK-MEL-28 cells
Dulbecco's Phosphate Buffer Saline w/o Calcium w/o Magnesium	EuroClone Spa, Milan, Italy	ECB4004L	saline buffer solution
Fetal Bovine Serum	EuroClone Spa, Milan, Italy	ECS0180L	ancillary for cell culture
Ficoll	GE-Healthcare	17-1440-02	separation of mononuclear cells from human blood.
hemocytometer	Neubauer		Cell counter
Heparinized vials	Thermo Fisher Scientific Inc., Waltham, MA		Vials for venous blood collection
interferon alpha-2b	Millipore-Sigma; St. Louis, MO	SRP4595	recombinant human cytokine
L-Glutamine 100X	EuroClone Spa, Milan, Italy	ECB3000D	ancillary for cell culture
Liquid nitrogen			
Lympholyte cell separation media	Cedarlane Labs, Burlington, Canada		Separation of lymphocytes by density gradient centrifugation
Lymphoprep	Axis-Shield PoC AS, Oslo, Norway		
Matrigel	Corning, New York, NY	354230	growth factor reduced basement membrane matrix
MDA-MB-231	American Type Culture Collection (ATCC), Manassas, VA	HTB-26	human breast cancer cell line

Penicillin/ Streptomycin 100X	EuroClone Spa, Milan, Italy	ECB3001D	ancillary for cell culture
Pipet aid	Drummond Scientific Co., Broomall, PA	4-000-201	Liquid handling
PKH26 Red Fluorescent cell linker	Millipore-Sigma; St. Louis, MO	PKH26GL	red fluorescent cell dye
PKH67 Green fluorescent cell linker	Millipore-Sigma; St. Louis, MO	PKH67GL	green fluorescent cell dye
RPMI-1640	EuroClone Spa, Milan, Italy	ECM2001L	Culture medium for MDA-MB-231 cells
serological pipettes (2 mL, 5 mL, 10 mL, 25 mL, 50 mL)	Corning- Millipore-Sigma; St. Louis, MO	CLS4486; CLS4487; CLS4488; CLS4489; CLS4490	Liquid handling
sterile tips (1-10 µL, 10-20 µL, 20-200 µL, 1000 µL)	EuroClone Spa, Milan, Italy	ECTD00010; ECTD00020; ECTD00200; ECTD01005	tips for micropipette
Timer			
Trypan Blue solution	Thermo Fisher Scientific Inc., Waltham, MA	15250061	cell stain to assess cell viability
Trypsin	EuroClone Spa, Milan, Italy	ECM0920D	dissociation reagent for adherent cells
Cell culture equipment			
EVOS-FL fluorescence microscope	Thermo Fisher Scientific Inc., Waltham, MA		Fluorescent microscope for living cells
Humified cell culture incubator	Thermo Fisher Scientific Inc., Waltham, MA	311 Forma Direct Heat COIncubator; TC 230	Incubation of cell cultures at 37 °C, 5% CO ₂
Juli Microscope	Nanoentek		
Laboratory refrigerator (4 °C)	FDM		
Laboratory Safety Cabinet (Class II)	Steril VBH 72 MP		Laminar flow hood
Optical microscope	Zeiss		
Refrigerable centrifuge	Beckman Coulter		
Thermostatic bath			
Microfabrication materials			
3-Aminopropyl)triethoxysilane (Aptes)	Sigma Aldrich	A3648	silanizing agent for bonding PDMS to plastic coverslip
Chromium quartz masks / 4"x4", HRC / No AZ	MB W&A, Germany		optical masks for photolithography
Glass coverslip, D 263 M Schott glass, (170 ± 5 µm)	Ibidi, Germany	10812	
Hydrogen Peroxide solution 30%	Carlo Erba Reagents	412081	reagents for piranha solution
Methyl isobutyl ketone	Carlo Erba Reagents	461945	PMMA e-beam resist developer
Microscope Glass Slides (Pack of 50 slides) 76.2 mm x 25.4 mm	Sail Brand	7101	substrates for bonding chips
Miltex Biopsy Punch with Plunger, ID 1.0mm	Tedpella		dermal biopsy punches for chip reservoirs
PMMA 950 kDa	Allresist, Germany	AR-P. 679.04	Positive electronic resists for patterning optical masks
Polymer untreated coverslips	Ibidi, Germany	10813	substrates for bonding chips
Prime CZ-Si Wafer, 4", (100), Boron Doped	Gambetti Xenologia Srl, Italy	30255	
Propan-2-ol	Carlo Erba Reagents	415238	
Propylene glycol monomethyl ether acetate (PGMEA)	Sigma Aldrich	484431-4L	SU-8 resists developer
SU-8 3005	Micro resist technology, Germany	C1.02.003-0001	Negative Photoresists
SU-8 3050	Micro resist technology, Germany	C1.02.003-0005	Negative Photoresists
Suite of Biopunch, ID 4.0 mm, 6.0 mm, 8.0 mm	Tedpella	15111-40, 15111-60, 15111-80	dermal biopsy punches for chip reservoirs

Sulfuric acid 96%	Carlo Erba Reagents	410381	reagents for piranha solution
SYLGARD 184 Silicone Elastomer Kit	Dowsil, Dow Corning	11-3184-01	Silicone Elastomer (PDMS)
Trimethylchlorosilane (TMCS)	Sigma Aldrich	92360-100ML	silanizing agent for SU-8 patterned masters
Microfabrication equipment			
100 kV e-beam lithography	Raith-Vistec EBPG 5HR		
hotplate			
Optical lithography system	EV-420 double-face contact mask-aligner		
Reactive Ion Etching system	Oxford plasmalab 80 plus system		
Vacuum dessicator			