

Co-Culture *In Vitro* Systems to Reproduce the Cancer-Immunity Cycle

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Materials

Name	Company	Catalog Number	Comments
1.5 mL microtubes	Eppendorf	30120086	
100 kV e-beam lithography	Vistec		
100 mm Petri dishes	Greiner Bio One	664160	
12-well plates	Euroclone	ET3012	
15 and 50 mL tubes	Corning	352096; 352070	
40 µm cell strainer	Corning	CLS431750	
5 mL polystyrene tubes	Greiner Bio-One	120180	
70 µm cell strainer	Corning	CLS431751	
75 cm ² cell culture treated flask	Euroclone	ET7076	
Adsorbent wipes			
Allumin foil			
anti-mouse CD107a (LAMP-1) Antibody	Miltenyi Biotec	130-111-319	
anti-mouse CD25 (7D4) Antibody	Miltenyi Biotec	130-118-678	
anti-mouse CD3 (17A2) Antibody	BioLegend	100206	
Aptes	Sigma Aldrich	440140	
BD Cytofix/Cytoperm Plus Fixation/ Permeabilization Solution Kit with BD GolgiPlug	BD Biosciences	555028	
BD GolgiPlug Protein Transport Inhibitor (Containing Brefeldin A)	BD Biosciences	555029	
BD GolgiStop Protein Transport Inhibitor (Containing Monensin)	BD Biosciences	554724	
Bovine serum albumin (BSA)	US Biological, Salem	A1312	
CD11c Monoclonal Antibody (N418)	eBioscience	12-0114-81	
CD137 (4-1BB) Monoclonal Antibody (17B5)	eBioscience	17-1371-82	
CD3 Monoclonal Antibody (17A2)	eBioscience	25-0032-82	

CD44 Monoclonal Antibody (IM7)	eBioscience	11-0441-82	
CD45 Monoclonal Antibody (30-F11)	Invitrogen	MCD4528	
CD69 Monoclonal Antibody (H1.2F3)	eBioscience	48-0691-82	
CD8a Monoclonal Antibody (53-6.7)	eBioscience	11-0081-82	
CD8a Monoclonal Antibody (53-6.7)	eBioscience	17-0081-82	
CD95 (APO-1/Fas) Monoclonal Antibody (15A7)	eBioscience	53-0951-82	
Cell counting slides	Kova International	87144E	
Chromium quartz masks	MB W&A, Germany		
Click-iT Plus EdU Alexa Fluor 647 Flow Cytometry Assay Kit	Invitrogen	C10635	
CytoFLEX Flow Cytometer	Beckman Coulter		
Dead Cell Removal Kit	Miltenyi Biotec	130-090-101	
Dulbecco's Phosphate-Buffered Saline (D-PBS)	EuroClone	ECB4053L	
EDTA	Invitrogen	AM9260G	
Fetal bovine serum (FBS)	EuroClone	ECS0180L	
Flowjo v10.0.7	Flowjo, LLC		
Granzyme B Monoclonal Antibody (NGZB)	eBioscience	12-8898-82	
H ₂ O ₂	Sigma Aldrich		
H ₂ SO ₄	Sigma Aldrich		
hotplate			
Humified cell culture incubator (37°, 5% CO ₂)	Thermo Scientific		
Ice machine	Brema Ice Makers		
IFN gamma Monoclonal Antibody (XMG1.2)	eBioscience	11-7311-82	
Illustrator CC 2015	Adobe Systems Inc.		
ImageJ	National Institute of Health		
Incucyte 2022A Software	Sartorius		
Incucyte Cytotox Dye for Counting Dead Cells	Sartorius	4632	
Incucyte SX5 Live-Cell Analysis System	Sartorius		
JuLi Smart Fluorescent Live Cell Imaging Microscope	Bulldog Bio		
Laboratory bench			
Laboratory refrigerator (4°C)			
Laboratory Safety Cabinet (Class II)	Angelantoni		
L-glutamine 200 mM	EuroClone	ECB3004D	
LIVE/DEAD Fixable Aqua Dead Cell Stain Kit	Invitrogen	L34957	
LIVE/DEAD Fixable Near-IR Dead Cell Stain Kit	Invitrogen	L10119	
MACS columns	Miltenyi Biotec	130-042-201; 130-042-401	
MACS separators	Miltenyi Biotec	130-042-10; 130-042-302	
MCA205 mouse fibrosarcoma cell line	Sigma-Aldrich	SCC173	
Microbiologically controlled animal facility equipped with Class II safety cabine			

MicroCL 21R Microcentrifuge	Thermo Scientific	75002552	
Microsoft Excel	Microsoft, Redmond		
Mouse: C57BL/6J	The Jackson Laboratory	000664	
Naive CD8a+ T Cell Isolation Kit, mouse	Miltenyi Biotec	130-096-543	
Nikon ECLIPSE Ts2	Nikon Instruments Inc.		
NIS-Elements BR 5.30.0064-BIT	Nikon Instruments Inc.		
Optical lithography	EVG		
Penicillin G sodium salt and streptomycin sulfate	EuroClone	ECB3001D/1	
Pipet aid	Drummond Scientific Co., Broomall, PA	4-000-201	
Pipettes	Eppendorf		
PKH67 Fluorescent Cell Linker Kits	Sigma-Aldrich	PKH67GL-1KT	fluorescent cell linker kit
plastic coverslip	IBIDI	10812	
Propidium Iodide	Thermo Scientific	P1304MP	
Reactive Ion Etching system	Oxford plasmalab		
Roswell Park Memorial Institute 1640 (RPMI 1640)	EuroClone	ECB9006L	
serological pipettes (2 mL, 5 mL, 10 mL, 25 mL)	Corning- Millipore-Sigma; St. Louis, MO	CLS4486; CLS4487; CLS4488; CLS4489	
SL 16 Centrifuge Series	Thermo Scientific	75004031	
Sterile scalpels, surgical forceps, scissors and pliers			
Sterile tips (1–10 µL, 20–200 µL, 1000 µL)	EuroClone Spa, Milan, Italy	ECTD00010; ECTD00020; ECTD00200; ECTD01005	
SU-8 3000 series	MicroChem corp, Newton, (MA)		
Suite of dermal biopsy punches	Kai Medical, Tedpella		
Sylgard 184	Dowsil, Dow Corning	101697	
TCR beta Monoclonal Antibody (H57-597)	eBioscience	12-5961-82	
Thermostatic bath			
Timer			
TMCS	Sigma Aldrich	92360	
Trypan Blue Stain (0.4%)	Thermo Scientific	15250061	
Trypsin-EDTA w/ Phenol Red	EuroClone	ECM0920	
Vacuum dessicator			