

Materials List for:

Lymphocyte Isolation from Human Skin for Phenotypic Analysis and *Ex Vivo* Cell Culture

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Materials

Name	Company	Catalog Number	Comments
Disposable Biopsy Punch	Kai Europe	BP-40F	4 mm
Disposable sterile scalpels	Dalhausen	1100000510	
gentleMACS C tube	Miltenyi Biotech	130-093-237	Blue-capped, used as the dissociation tube.
gentleMACS Dissociator	Miltenyi Biotech	130-093-235	Automated tissue dissociator. Using the "program spleen_01" for dissociation of the skin biopsy.
Cell strainer	BD	352350	70 µm nylon
96-well U-bottom plate	Greiner Bio-One	650180	
RPMI 1640	Life Technologies	22409-015	
Sodium pyruvate	Life Technologies	11360-039	
GlutaMAX	Life Technologies	35050-061	
Penicillin/Streptomycin	Life Technologies	15140-122	
Human Pooled Serum (HPS)	in house prepared		
Collagenase I	Sigma-Aldrich	C2674	Type 1-A, suitable for cell culture
DNase I	Calbiochem	260913	
PBS	B Braun	3623140	
BSA	Sigma-Aldrich	A4503-500G	
Fixable Viability Dye (FVD) APC-eFluo780	eBioscience	65-0865-18	Stain dead cells prior to cell fixation; dilute with PBS at 1:1,000
Fixation/Permeabilization Concentrate	eBioscience	00-5123-43	
Fixation/Permeabilization Diluent	eBioscience	00-5223-56	
Permeabilization Buffer (10x)	eBioscience	00-8333-56	
BV421 Mouse anti-human CD45	BD	563879	Clone: HI30; dilution factor 1:50
FITC Mouse anti-human CD14	Dako	T0844	Clone: TUK4; dilution factor 1:50
PE Mouse anti-human CD56	Dako	R7127	Clone: MOC-1; dilution factor 1:50
ECD Mouse anti-human CD3	Beckman - Coulter	A07748	Clone: UCHT1; dilution factor 1:50 for surface staining; dilution factor 1:25 for intracellular staining.
PC5.5 Mouse anti-human CD4	Beckman - Coulter	B16491	Clone: 13B8.2; dilution factor 1:200
PeCy7 Mouse anti-human CD11c	Beckman - Coulter	A80249	Clone: BU15; dilution factor 1:50
APC Mouse anti-human CD1c	Miltenyi Biotech	130-090-903	Clone: AD5-8E7; dilution factor 1:10
APC-AlexFluo700 Mouse anti-human CD8	Beckman - Coulter	A66332	Clone: B9.11; dilution factor 1:400

APC-AlexFluo750 Mouse anti-human CD19	Beckman - Coulter	A94681	Clone: J3-119; dilution factor 1:50
PeCy7 Mouse anti-human CD25	eBioscience	AD5-8E7	Clone: BC96; dilution factor 1:50
PE Rat anti-human CLA	Miltenyi Biotech	130-091-635	clone: HECA-452; dilution factor 1:
eFluo450 Rat anti-human Foxp3	eBioscience	48-4776-42	Clone: PCH101; intracellular staining; dilution factor 1:50
AlexFluo488 Mouse anti-human IL-17A	eBioscience	53-7179-42	Clone: eBio64DEC17; intracellular staining; dilution factor 1:50
PeCy7 Mouse anti-human IFNg	eBioscience	25-7319-41	Clone: 4S.B3; intracellular staining; dilution factor 1:400
Flow-Count Fluorospheres	Beckman - Coulter	7547053	Counting beads, for flow cytometry