

Materials List for

# Magnetic Fluorescent Bead-Based Dual-Reporter Flow Analysis of PDL1-Vaxx Peptide Vaccine-Induced Antibody Blockade of the PD-1/PD-L1 Interaction

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## Materials

Name	Company	Catalog Number	Comments
<b>Buffers</b>			
Activation Buffer: 0.1 M NaH <sub>2</sub> PO <sub>4</sub> , pH 6.2	Millipore/Sigma	S3139	
Assay/Wash Buffer: PBS-TBN (1x PBS, pH 7.4 + 0.1% BSA + 0.05 % (v/v) Tween-20; 0.05% NaN <sub>3</sub> )	Millipore/Sigma	P3563 (PBS+Tween20), A7888 (Bovine serum albumin), S8032 (sodium azide)	
Coupling Buffer: 50 mM 2-morpholinoethanesulfonic acid (MES), pH 5.0	MilliporeSigma	M2933	
<b>Coupling Reagents</b>			
1-Ethyl-3-[3-dimethylaminopropyl]carbodiimide hydrochloride (EDC)	ThermoFisher Scientific (Waltham, MA)	77149	
xMAP Antibody Coupling Kit (if desired), includes:	Luminex Corp. (Austin, TX)	40-50016	
EDC, 10 mg			
sNHS solution, 250 μL			
Activation/Coupling Buffer: 0.1 M 2-morpholinoethanesulfonic acid (MES), pH 6.0			
Wash Buffer: 1x PBS, pH 7.4 + 0.1% BSA + 0.05 % (v/v) Tween-20; 0.05% NaN <sub>3</sub> (PBS-TBN)			
Sulfo-NHS (N-hydroxysulfosuccinimide)	ThermoFisher Scientific (Waltham, MA)	24510	
<b>Instrumentation and Ancillary Lab Supplies</b>			
xMAP INTELLIFLEX (dual-reporter instrument)	Luminex Corp. (Austin, TX)	INTELLIFLEX-DRSE-RUO	

Low protein-binding round bottom 96-well plate	ThermoFisher Scientific (Waltham, MA)	07-200-761	
Luminex Magnetic Plate Separator (or comparable)	Luminex Corp. (Austin, TX)	CN-0269-01	
Luminex Magnetic Tube Separator (or comparable)	Luminex Corp. (Austin, TX)	CN-0288-01	
MagPlex Microspheres (magnetic, fluorescent, 6.5- $\mu$ m-diameter beads)	Luminex Corp. (Austin, TX)	MC-1**** (varies by bead label)	
Protein LoBind microcentrifuge tubes	ThermoFisher Scientific (Waltham, MA)	022431081	
<b>Peptides, Antibodies, &amp; Detection Reagents</b>			
Atezolizumab (humanized anti-PDL1 IgG1 monoclonal antibody), positive control	Genentech/Roche (San Francisco, CA)	n/a (prescription medications)	
Biotinylated recombinant human PDL1	Sino Biological (Wayne, PA)	10084-H49H-B	
Brilliant Violet 421-conjugated donkey anti-human IgG	Jackson ImmunoResearch Laboratories Inc. (Westgrove, PA)	709-675-149	
Brilliant Violet 421-conjugated donkey anti-rabbit IgG	Jackson ImmunoResearch Laboratories Inc. (Westgrove, PA)	711-675-152	
Recombinant human PD-1 (poly-histidine tagged)	Acro Biosystems (Newark, DE)	PD1-H5256	
Streptavidin-conjugated R-phycoerythrin (SAPE)	Agilent (Santa Clara, CA)	PJRS34-1	
Trastuzumab (Herceptin, humanized anti-HER2 monoclonal antibody), negative control	Genentech/Roche (San Francisco, CA)	n/a (prescription medications)	