

HLDA10 VALIDATION FILE FOR CD369_Clec7A

More than two mAbs that have the same expression pattern on primary cells

Binding to transfectants

MOLECULE NAME: **C-type lectin domain family 7 member A**

ALTERNATIVE NAMES: Beta-glucan receptor, C-type lectin superfamily member 12, Dendritic cell-associated C-type lectin 1, DC-associated C-type lectin 1, Dectin-1, Clec7A/Dectin-1/BGR

GENE FAMILY:

PROTEIN: Single-pass type II membrane protein

FUNCTION: Lectin that functions as pattern receptor specific for beta-1,3-linked and beta-1,6-linked glucans, (cell wall constituents from pathogenic bacteria and fungi). Necessary for the TLR2-mediated inflammatory response and TLR2-mediated activation of NF-kappa-B. Enhances cytokine production in macrophages and dendritic cells. Mediates production of reactive oxygen species in the cell. Mediates phagocytosis of *C.albicans* conidia. Binds T-cells in a way that does not involve their surface glycans and plays a role in T-cell activation. Stimulates T-cell proliferation

EXPRESSION Raji, Daudi, NIH-hDectin-1, Monocytes, PMNs, DC, B cells

ANTIBODY INFORMATION

10-01

Antibody Name GE2
Specificity Human Dectin-1/ β -glucan receptor/Clec7A
Antibody Species Mouse
Ig Isotype IgG1
Immunogen NIH3T3- human BGR-A/Dectin-1a
Epitope Recognised
Submitter Gordon Brown
References (1)

10-35

Antibody Name Clone 259931, FAB1859P
Specificity Blood monocytes
Antibody Species mouse
Ig Isotype IgG2b
Immunogen NS0 with human Dectin-1/CLEC7A (Thr66-Met201)
Epitope Recognised
Submitter R&D
References (2)

10-79

Antibody Name	15 E 2
Specificity	
Antibody Species	Mouse
Ig Isotype	IgG2a. K
Immunogen	Ecto domain with C-term Fc fusion, RIBLS
Epitope Recognised	
Submitter	BioLegend
References	(3)

CONFIRMATION OF SPECIFICITY

	10-01	10-35	10-79
Expression on transfected cells (Figure 1)	+	+	+
Expression on cell line	weak	weak	weak
Expression on normal primary cell	Monocytes, Myeloid DC but not pDC CD11b+ thymic DC	Monocytes Myeloid DC but not pDC CD11b+ thymic DC	Monocytes Myeloid DC but not pDC CD11b+ thymic DC
Expression on in vitro derived cells	Weak on MoDC Downregulated in culture	On MoDC Downregulated in culture	On MoDC Downregulated in culture

CELL LINE EXPRESSION

	Cell lines	10-01	10-35	10-79
Burkitt lymphoma B cell lines	Raji	-	-/+	+
	Daudi		+	+
T cell leukemia	Jurkat			
Myeloid Leukemia	HEL	+/-	-	
	HL-60	+/-	-	+/-
	NB4	-	-	
	Thp-1	-	-	+
	U-937	-	-	
Hodgkins derived lines	KM-H2	+	-	

Figure 1: Transient transfection of CHO-K1 with Clec7A antibodies.

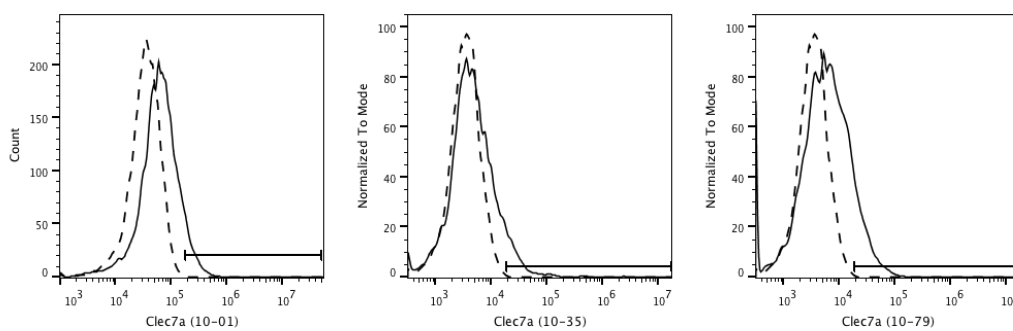


Figure 2: Binding of each antibody with fresh blood DC. Solid line = test antibody, dashed line = isotype control.

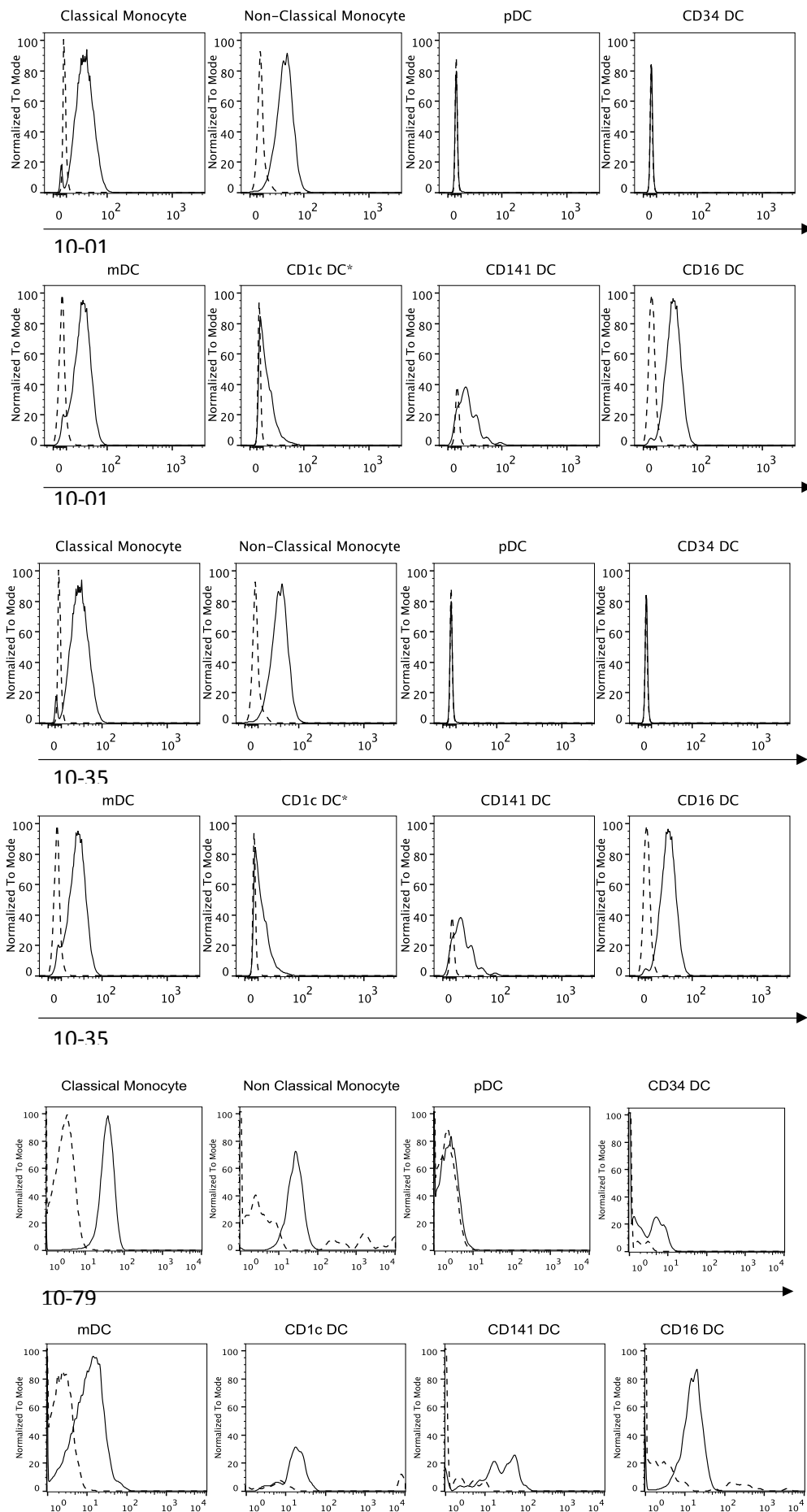
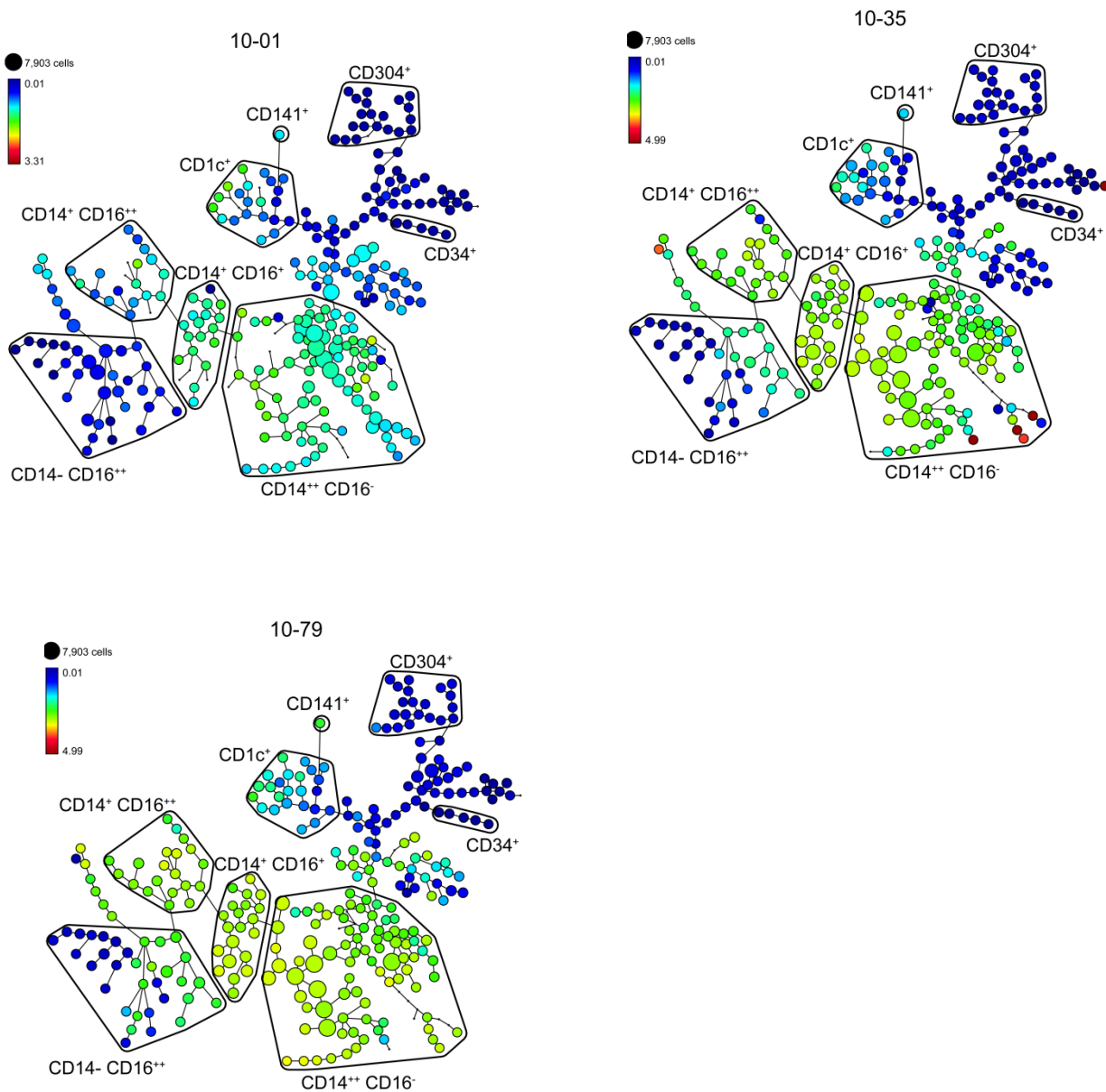


Figure 3. Clustering analysis using SPADE to demonstrate expression of Clec7A antibodies on fresh blood DC.



PUBLICATIONS USING THESE MaB

1. Willment JA, Marshall AS, Reid DM, Williams DL, Wong SY, Gordon S, et al. The human beta-glucan receptor is widely expressed and functionally equivalent to murine Dectin-1 on primary cells. *European journal of immunology*. 2005;35(5):1539-47. Epub 2005/04/09.
2. Heyl KA, Klassert TE, Heinrich A, Muller MM, Klaile E, Dienemann H, et al. Dectin-1 is expressed in human lung and mediates the proinflammatory immune response to nontypeable *Haemophilus influenzae*. *mBio*. 2014;5(5):e01492-14. Epub 2014/08/28.
3. Ni L, Gayet I, Zurawski S, Duluc D, Flamar AL, Li XH, et al. Concomitant activation and antigen uptake via human dectin-1 results in potent antigen-specific CD8+ T cell responses. *J Immunol*. 2010;185(6):3504-13. Epub 2010/08/24.