HLDA10 VALIDATION FILE FOR CD367 DCIR ClecSF6

MOLECULE NAME: CLECSF6

ALTERNATIVE NAMES: C-type lectin DDB27, C-type lectin superfamily member 6, Dendritic cell

immunoreceptor (DCIR), Lectin-like immunoreceptor, C-type lectin domain

family 4 member A; CLEC4A; CLECSF6; HDCGC13P; LLIR

GENE FAMILY C-type lectin family

PROTEIN Single pass type II transmembrane protein

FUNCTION May be involved in regulating immune reactivity. May play a role in modulating dendritic cells (DC) differentiation and/or maturation. May be involved via its ITIM motif (immunoreceptor tyrosine-based inhibitory motifs) in the inhibition of B-cell-receptor-mediated calcium mobilization and protein tyrosine phosphorylation.

EXPRESSION Expressed in dendritic cells, myeloid cells, B-cells and HL-60 cells (at protein level). TNF alpha, IL-1 alpha, and LPS, down-regulated expression at the surface of neutrophils (at protein level). Expressed preferentially in hematopoietic tissues. Expressed in peripheral blood leukocytes, neutrophils, moderate quantities in spleen, lymph node, and bone marrow, and at very low levels in thymus. Expressed in Ag-presenting cells (DC, monocytes, macrophages and B-cells), as well as on granulocytes. Expression was decreased in DC by signals inducing its maturation (e.g. CD40 ligand, LPS, and TNF alpha).

10-13

Antibody Name: FAB1748P, 216110 (PE conjugate)

Specificity: Human PB neutrophils

Antibody Species: Mouse Ig Isotype: IgG1

Immunogen:

Epitope Recognised:

Submitter: R&D Systems

References:

10-71

Antibody Name: 111F8.04

Specificity:

Antibody Species: Mouse Ig Isotype: IgG1

Immunogen: Human DCIR-Ig fusion protein

Epitope Recognised:

Submitter: Dendritics, Australian Biosearch

References: (1)

10-72

Antibody Name: 9E8

Specificity: Human
Antibody Species: Mouse
Ig Isotype: IgG1, k

Immunogen: Ectodomain of CLEC4A-human Fc fusion protein

Epitope Recognised:

Submitter: BioLegend

References: (2)

(3-5)

SPECIFICITY

Expression on	Validation of 10-13 and 10-72 monoclonal antibodies binding			
transfected CHO	transfected cells (Fig 1)			
Expression on cell line	Expression on Cell lines			
Expression on normal primary cell	Cluster analysis on primary monocytes and dendritic cells			
Expression on in vitro derived cells	Expressed by in vitro derived macrophages and weakly by MoDC			

CELL LINE EXPRESSION

	Cell lines	10-13	10-71	10-72
Burkitt lymphonma B cell	Raji	+	+	
lines				
	Daudi	+	+	
T cell leukemia	Jurkat			
Myeloid Leukemia	HL60	-		-
	NB4	-		
	U937	+/-	+/-	+/-
	THP-1	+/-		
	HEL	+	+	+
Hodgkins derived lines	KM-H2	-	-	-

Figure 1. Transfection of HEK OR CHO cells with DCIR cDNA

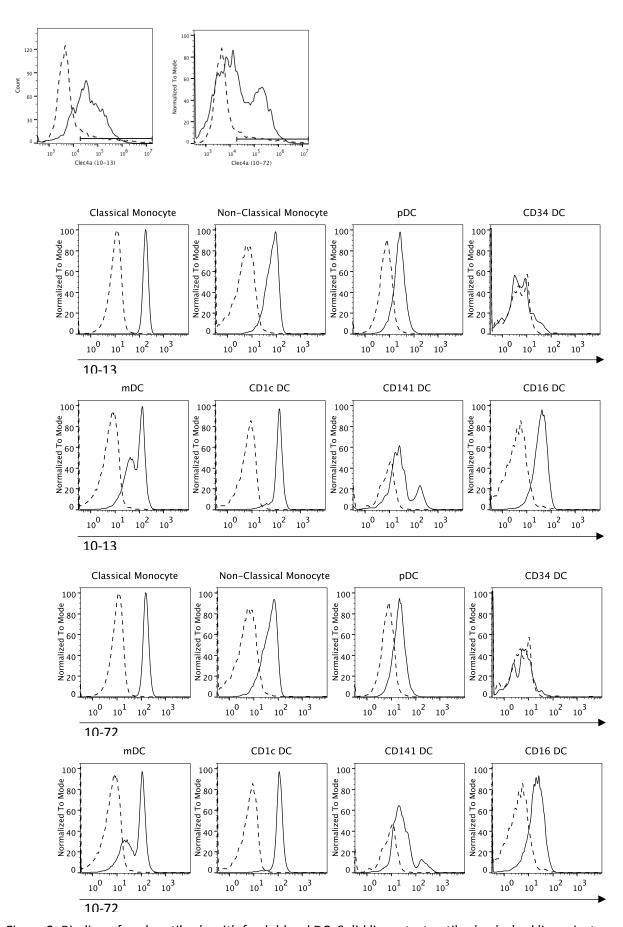


Figure 2: Binding of each antibody with fresh blood DC. Solid line = test antibody, dashed line = isotype control.

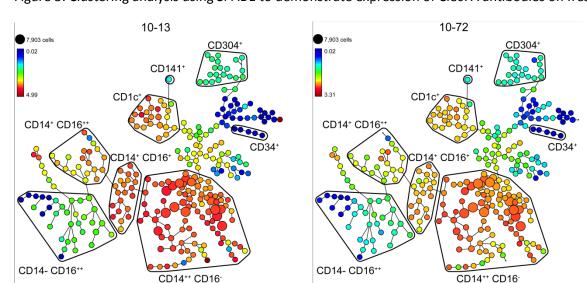


Figure 3. Clustering analysis using SPADE to demonstrate expression of Clec7A antibodies on fresh blood DC.

PUBLICATIONS USING ANTIBODIES

- 1. Lambert AA, Barabe F, Gilbert C, Tremblay MJ. DCIR-mediated enhancement of HIV-1 infection requires the ITIM-associated signal transduction pathway. Blood. 2011;117(24):6589-99. Epub 2011/05/04.
- 2. Klechevsky E, Flamar AL, Cao Y, Blanck JP, Liu M, O'Bar A, et al. Cross-priming CD8+ T cells by targeting antigens to human dendritic cells through DCIR. Blood. 2010;116(10):1685-97. Epub 2010/06/10.
- 3. Richard M, Veilleux P, Rouleau M, Paquin R, Beaulieu AD. The expression pattern of the ITIM-bearing lectin CLECSF6 in neutrophils suggests a key role in the control of inflammation. Journal of leukocyte biology. 2002;71(5):871-80. Epub 2002/05/08.
- 4. Huang X, Yuan Z, Chen G, Zhang M, Zhang W, Yu Y, et al. Cloning and characterization of a novel ITIM containing lectin-like immunoreceptor LLIR and its two transmembrane region deletion variants. Biochemical and biophysical research communications. 2001;281(1):131-40. Epub 2001/02/17.
- 5. Bates EE, Fournier N, Garcia E, Valladeau J, Durand I, Pin JJ, et al. APCs express DCIR, a novel C-type lectin surface receptor containing an immunoreceptor tyrosine-based inhibitory motif. J Immunol. 1999;163(4):1973-83. Epub 1999/08/10.