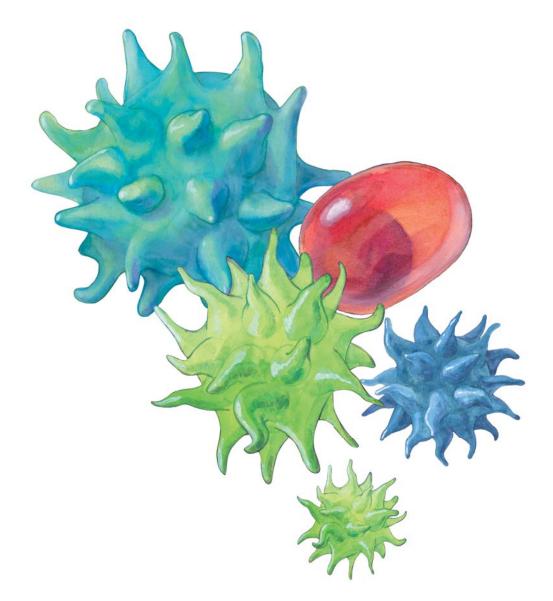
# Dendritic Cell Development & Subset Identification





# **Dendritic Cell Development**

Dendritic cells (DCs) are a heterogeneous population of immune cells that are critical mediators of innate and adaptive immunity. DC precursors develop in the bone marrow.<sup>1</sup> While plasmacytoid DCs complete development in the bone marrow, most DCs complete development in lymphoid and peripheral tissues.1 Knockout mouse models have identified several cytokines and transcription factors that are critical for DC development in vivo.<sup>2,3</sup> A limited number of cytokines and transcription factors, including Flt-3 Ligand, Ikaros, PU.1, and GFI-1, are essential during early DC development and their silencing results in global reductions in DC numbers.<sup>2,3</sup> Additional cytokines and transcriptional regulators are necessary during the late phases of DC differentiation.<sup>2,3</sup> Late developmental factors act to promote or maintain the differentiation of specific DC subsets.<sup>2,3</sup> The final stage in DC development occurs upon activation. Activation of human and mouse DCs can be detected by upregulation of MHC class II, B7-1/CD80, B7-2/CD86, CD40/TNFRSF5, or CD83 expression.<sup>4</sup> Understanding DC developmental pathways has facilitated the in vitro generation of DCs from bone marrow or blood-derived DC precursors using cytokine cocktails of Flt-3 Ligand, M-CSF, GM-CSF, and IL-4.<sup>5</sup>

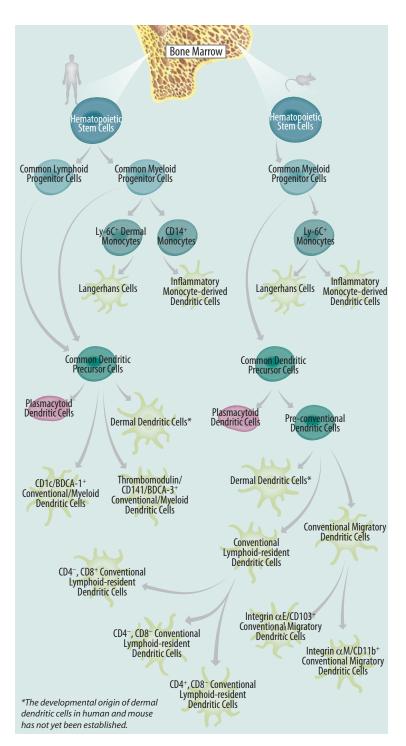
#### References

- 1. Liu, K. & M.C. Nussenzweig (2010) Immunol. Rev. 234:45.
- 2. Belz, G.T. & S.L. Nutt (2012) Nat. Rev. Immunol. 12:101.
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R&D Systems offers a wide range of proteins & antibodies for studying DC development. For a complete product listing, please visit our website at www.RnDSystems.com/DCDevelopment

#### Products for Dendritic Cell Development Research

		Proteins			Anti	bodies		
Molecule	Species		B/N	E	FC	ICC/IHC	IP	WB
B7-1/CD80	Human, Mouse	✓	✓	✓	✓	✓		✓
B7-2/CD86	Human	$\checkmark$	✓		✓	✓		✓
	Mouse	✓	✓		✓			✓
BATF3	Human							✓
CD40/TNFRSF5	Human	✓	✓		✓	<ul> <li>✓</li> </ul>		✓
	Mouse	✓		✓	✓	✓	✓	✓
CD83	Human	$\checkmark$			✓	✓		✓
	Mouse	✓	✓		~	✓		✓
CSL/Rbp-J	Human					✓		✓
Flt-3/Flk-2	Human, Mouse	$\checkmark$			✓	✓		✓
GFI-1	Human					✓		✓
GM-CSF R $\alpha$	Human	$\checkmark$			✓			✓
	Mouse	✓	✓		~	✓		
ID2	Human					✓		✓
Ikaros	Human				✓	✓		✓
IRF2	Human							✓
	Mouse							✓
IRF4	Human					<ul> <li>✓</li> </ul>		✓
	Mouse							✓
IRF8	Human					<ul> <li>✓</li> </ul>		✓
M-CSF R	Human	✓	✓	✓	✓	<ul> <li>✓</li> </ul>		✓
	Mouse	✓			✓	<ul> <li>Image: A start of the start of</li></ul>		<ul> <li>✓</li> </ul>
Notch-2	Human	✓			✓	<ul> <li>✓</li> </ul>		✓
	Mouse	✓			~			✓
PU.1/Spi-1	Human				✓	<ul> <li>✓</li> </ul>		✓
	Mouse							✓
RelB	Human					✓		✓
Spi-B	Human							✓
	Mouse				~	<ul> <li>✓</li> </ul>		



### Growth Factors for Dendritic Cell Research

Molecule	Species	Proteins	ELISAs	ELISpot/FluoroSpot Kits & Development Modules	Multiplex Array/ Assay Kits & Reagents	Antibodies
Flt-3 Ligand	Human	<ul> <li>✓</li> </ul>	~			✓
	Mouse	✓	~			✓
GM-CSF	Human	✓	<ul> <li>✓</li> </ul>	✓	<ul> <li>✓</li> </ul>	✓
	Mouse	✓	✓	$\checkmark$	<ul> <li>✓</li> </ul>	✓
IL-4	Human	✓	✓	✓	✓	✓
	Mouse	✓	✓	$\checkmark$	<ul> <li>✓</li> </ul>	✓
M-CSF	Human	✓	<ul> <li>✓</li> </ul>			✓
	Mouse	<ul> <li>✓</li> </ul>	~		✓	✓

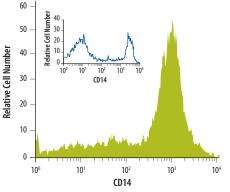
Application Key: B/N Blocking/Neutralization, E ELISA, FC Flow Cytometry, ICC/IHC Immunocytochemistry/Immunohistochemistry, IP Immunoprecipitation, WB Western blot

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# **Products for Dendritic Cell Differentiation & Characterization**

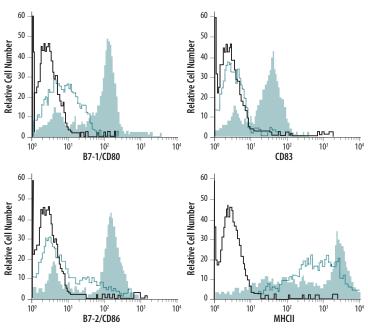
R&D Systems StemXVivo<sup>M</sup> Serum-free Dendritic Cell Base Media (Catalog # CCM003) is specially formulated and optimized for the culture and differentiation of human dendritic cells (DCs). This base media, supplemented with the appropriate cytokines and growth factors,\* allows monocyte-derived DCs (MoDCs) to be generated from a pure population of isolated CD14<sup>+</sup> monocytes (A and B). Using antibodies directed against specific cell surface markers, immature and lipopolysaccharide (LPS)-induced mature MoDCs can be characterized (C). MoDCs generated under these conditions are biologically active (D).

#### A. Isolation of Human Dendritic Cell (DC) Precursors



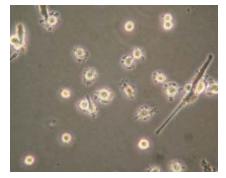
A. Isolation of CD14\* Monocytes using the MagCellect<sup>™</sup> Human CD14\* Cell Isolation Kit. Human CD14\* cells were isolated from ficoll separated peripheral blood mononuclear cells using the MagCellect Human CD14\* Cell Isolation Kit (Catalog # MAGH105). All viable cells were stained before (inset) and after (green) isolation using a PE-conjugated Mouse Anti-Human CD14 Monoclonal Antibody (Catalog # FAB3832P).

#### C. Phenotypic Analysis of Cultured Monocyte-derived Dendritic Cells (moDCs)



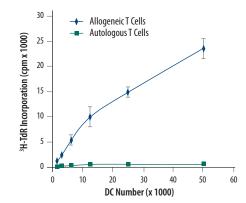
Phenotypic Analysis of Cultured Monocyte-derived Dendritic Cells Before and After LPS-induced Maturation. Immature monocyte-derived dendritic cells (open histograms-green line) were cultured for seven days in Human StemXVivo Serum-free Dendritic Cell Base Media (Catalog # CCM003) supplemented with Recombinant Human GM-CSF (Catalog # 215-GM), Recombinant Human IL-4 (Catalog # 204-IL), and gentamycin. Mature monocyte-derived dendritic cells (filled histograms), were grown under the same conditions for seven days and then induced with LPS for an additional 48 hours. Both sets of cells were subsequently assessed by flow cytometry using the indicated antibodies. Cells stained with isotype-matched controls are shown for comparison (open histograms-black line).

## B. Culture of DC Precursors in Growth Factor-supplemented Human StemXVivo Serum-free Dendritic Cell Base Media



Morphology of LPS-treated Monocyte-derived Dendritic Cells Cultured with Human StemXVivo Serum-free Dendritic Cell Base Media. CD14<sup>+</sup> monocytes, isolated from peripheral mononuclear cells using the MagCellect Human CD14<sup>+</sup> Cell Isolation Kit (Catalog # MAGH105), were cultured in Human StemXVivo Serum-free Dendritic Cell Base Media (Catalog # CCM003) in the presence of Recombinant Human GM-CSF (Catalog # 215-GM), Recombinant Human IL-4 (Catalog # 204-IL) and gentamicin for seven days. Lipopolysaccharide (LPS) was added to the cell suspension for an additional 48 hours to induce dendritic cell maturation. An image demonstrating the morphology of the resulting monocyte-derived dendritic cells is shown.

### D. Assessment of the Biological Activity of Mature LPS-treated moDCs



Mature Monocyte-derived Dendritic Cells Induce Proliferation of Allogeneic T Cells. CD14<sup>+</sup> monocytes were cultured for seven days in Human StemXVivo Serum-free Dendritic Cell Base Media (Catalog # CCM003) supplemented with Recombinant Human GM-CSF (Catalog # 215-GM), Recombinant Human IL-4 (Catalog # 204-IL) and gentamycin. The cells were subsequently treated with LPS for an additional 48 hours to induce dendritic cell maturation. Graded doses of these mature monocyte-derived dendritic cells were incubated with 1 x 10<sup>5</sup> autologous or allogeneic CD3<sup>+</sup> T cells for five days. <sup>3</sup>H-TdR was added to the culture for the final 18 hours and T cell proliferation was measured using a scintillation counter. Results are presented as the mean cpm obtained from three experiments.

To view a complete protocol describing the generation of monocyte-derived dendritic cells from CD14<sup>+</sup> monocytes, please visit our website at www.RnDSystems.com/ExVivoDCProtocol

\*StemXVivo Serum-free Dendritic Cell Base Media must be supplemented with cytokines and growth factors appropriate for the desired cell culture application. R&D Systems offers an extensive range of high quality cytokine- and growth factor-related products, including traditional and cell culture grade proteins, Animal-Free proteins, and GMP-grade proteins. For more information, please visit our website at www.RnDSystems.com/Proteins.

# For research use only. Not for use in diagnostic procedures.

# **Dendritic Cell Subsets & Markers**

Several phenotypic and functional DC subsets have been characterized including classical/conventional DCs (cDCs), Langerhans cells, inflammatory/ monocyte-derived DCs (moDCs), and plasmacytoid DCs (pDCs). While there is functional homology between mouse and human DCs, differences between mouse and human DC subset development and cell surface markers have complicated their characterization.<sup>1</sup> Most DCs have a characteristic morphology defined by long dendrite extensions as well as high levels of MHC class II expression. In contrast, pDCs have a rounded morphology similar to plasma cells and low MHC class II expression. DCs initiate innate immunity via detection of pathogens by pattern recognition receptors and secretion of pro-inflammatory or tolerogenic cytokines.<sup>2</sup> They also initiate adaptive immune activity or tolerance by acting as professional antigen-presenting cells (APCs).

All dendritic cells differentiate from a common myeloid progenitor and can be lymphoid-resident or reside in peripheral tissues and migrate to lymphoid organs, where they activate T cells.<sup>3</sup> High concentrations of DCs are present at areas of intense antigen exposure such as the skin, lung, and intestine.<sup>3</sup> As part of the first line of defense against exogenous pathogens, the skin houses a number of highly specialized DC subsets.<sup>4,5</sup> Langerhans cells are a unique subset of epidermal DCs and appear to function similarly in mice and humans. Dermal dendritic cell subsets also appear to function similarly in mice and humans, but their identifying markers differ significantly between the two species.<sup>6,7</sup> Two additional human and five additional mouse cDCs subsets have been identified that can be characterized by their expression of specific cell surface markers, transcription factors, and secreted factors. In contrast to cDCs, moDCs develop from monocytes at sites of inflammation and have been experimentally characterized in mice in vivo.8 Multiple moDC subsets arise from LY-6Chigh monocytes.9 Endogenous human moDCs that are pro-inflammatory in nature and likely correspond to mouse moDCs have been identified in peripheral blood.<sup>10</sup> pDCs are inefficient APCs and instead function in a primarily innate capacity through the detection of viral nucleic acids and the secretion of Type I IFNs, TNF- $\alpha$ , and IL-6.<sup>11</sup>

#### References

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- 11. Gilliet, M. et al. (2008) Nat. Rev. Immunol. 8:594.

#### Human CD1c/BDCA-1<sup>+</sup> Conventional/Myeloid Dendritic Cells

Cell Surface Markers	Lin <sup>-</sup> (CD3 <sup>-</sup> , CD14 <sup>+</sup> , CD19 <sup>+</sup> , CD20 <sup>+</sup> , CD56 <sup>+</sup> ), MHC class II <sup>+</sup> , CD1c/BDCA-1 <sup>+</sup> , Integrin $\alpha$ M/CD11b <sup>+</sup> , Integrin $\alpha$ X/CD11c <sup>+</sup> , SIRP $\alpha$ /CD172a <sup>+</sup>
Secreted Molecules	TNF-α, IL-6, IL-1β/IL-1F2

#### Human Thrombomodulin/CD141/BDCA-3<sup>+</sup> Conventional/Myeloid Dendritic Cells

Cell Surface Markers	Lin <sup>-</sup> (CD3 <sup>-</sup> , CD14 <sup>-</sup> , CD19 <sup>-</sup> , CD20 <sup>-</sup> , CD56 <sup>-</sup> ), MHC class II <sup>+</sup> , CD1a <sup>+</sup> , CLEC9a <sup>+</sup> , Integrin αX/CD11c <sup>+</sup> , Thrombomodulin/CD141/BDCA-3 <sup>+</sup> , XCR1 <sup>+</sup>
Secreted Molecules	IFN-β, IL-12

#### Mouse CD4<sup>-</sup>CD8 $\alpha^+$ Conventional Lymphoid-resident Dendritic Cells

Cell Surface Markers	MHC class II+, CD1d1+, CD4 <sup>+</sup> , CD8 $\alpha$ +, CLEC9a+, DEC-205/CD205+, Integrin $\alpha$ M/CD11b+, Integrin $\alpha$ X/CD11c+, Langerin/CD207+/-, XCR1+
Secreted Molecules	IFN-γ, ΙL-12

#### Mouse CD4+CD8 $\alpha$ <sup>-</sup> Conventional Lymphoid-resident Dendritic Cells

Cell Surface Markers	MHC class II+, CD4+, CD8 $\alpha$ <sup>-</sup> , CD172a/SIRP $\alpha$ <sup>+</sup> , Integrin $\alpha$ M/CD11b+, Integrin $\alpha$ X/CD11c+
Secreted Molecules	ΙFN-γ, ΙL-2, ΙL-6

#### Mouse CD4<sup>-</sup>CD8 $\alpha$ <sup>-</sup> Conventional Lymphoid-resident Dendritic Cells

Cell Surface Markers	MHC class II^+, CD4 <sup>+</sup> , CD8 <sup>+</sup> , CD172a/SIRP $\alpha^+$ , Integrin $\alpha$ M/CD11b <sup>+</sup> , Integrin $\alpha$ X/CD11c <sup>+</sup>
Secreted Molecules	IFN-γ, IL-2, IL-6

#### Mouse Integrin $\alpha$ E/CD103<sup>+</sup> Conventional Migratory Dendritic Cells

Cell Surface Markers	MHC class II+, CD1d1+, CD8 $\alpha^{+/\cdot}$ , CLEC9a^{+/\cdot}, DEC-205/CD205^{+/\cdot}, EpCAM/TROP1^{+/\cdot}, Integrin $\alpha$ E/CD103+, Integrin $\alpha$ X/CD11c+, Langerin/CD207^{+/\cdot}, XCR1^{+/\cdot}
Secreted Molecules	IL-12, IL-23

#### Mouse Integrin $\alpha$ M/CD11b<sup>+</sup> Conventional Migratory Dendritic Cells

Cell Surface Markers	MHC class II+, DEC-205/CD205+, Integrin $\alpha$ M/CD11b+, Integrin $\alpha$ X/CD11c+
Secreted Molecules	IL-6, IL-10

#### Human Dermal Dendritic Cells

CD14 <sup>+</sup> Cells Cell Surface Markers	MHC class II+, CD1c/BDCA1+, CD14+, CD45+, DC-SIGN/CD209+, Integrin $\alpha$ M/CD11b+, Integrin $\alpha$ X/CD11c+, Thrombomodulin/CD141/BDCA-3+ $^{\prime\prime}$
CD14 <sup>-</sup> Cells Cell Surface Markers	MHC class II+, CD1a+, CD1c/BDCA-1+, CD14 <sup>-</sup> , CD45+, Integrin $\alpha$ M/CD11b+, Integrin $\alpha$ X/CD11c+

#### Mouse Dermal Dendritic Cells

Langerin <sup>+</sup> Cells Cell Surface Markers	MHC class II+, EpCAM/Trop1+, Integrin $\alpha$ X/CD11c+, Integrin $\alpha$ E/CD103+, Integrin $\alpha$ M/CD11b <sup>low</sup> , Langerin/CD207+, SIRP $\alpha$ /CD172a+/-
Langerin <sup>-</sup> Cells Cell Surface Markers	MHC class II <sup>+</sup> , DEC-205/CD205 <sup>+</sup> , F4/80/EMR1 <sup>+</sup> , Integrin $\alpha$ X/CD11c <sup>+</sup> , Integrin $\alpha$ M/CD11b <sup>high</sup> , Langerin/CD207 <sup>-</sup>

### Human Langerhans Cells

Cell Surface Markers	MHC class II <sup>+</sup> , CD1a <sup>+</sup> , CD14 <sup>+</sup> , CD45 <sup>+</sup> , CD83 <sup>+</sup> , E-Cadherin <sup>+</sup> , EpCAM/TROP1 <sup>+</sup> , Integrin α.M/CD11b <sup>+/-</sup> ,
	Integrin $\alpha$ X/CD11c <sup>+</sup> , Langerin/CD207 <sup>+</sup>

#### **Mouse Langerhans Cells**

Cell Surface Markers	MHC class II+, CD45+, DEC-205/CD205+, E-Cadherin+, EpCAM/TROP1+, F4/80/EMR1+, Integrin $\alpha$ M/CD11b+,
	Integrin cxX/CD11c <sup>+</sup> , Langerin/CD207 <sup>+</sup>

#### Human Inflammatory/Monocyte-derived Dendritic Cells

	· · · · · ·	•	
	Cell Surface Markers	MHC class II <sup>+</sup> , CD14 <sup>-</sup> , Fcy RIII/CD16 <sup>+</sup> , Integrin $\alpha$ X/CD11c <sup>+</sup>	
	Secreted Molecules	IL-23, TNF-α	

#### Mouse Inflammatory/Monocyte-derived Dendritic Cells

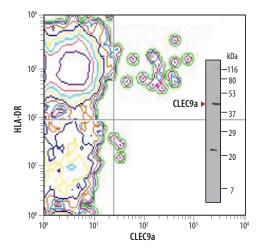
Cell Surface Markers	HC class II+, DC-SIGN/CD209+, Integrin $\alpha$ M/CD11b+, Integrin $\alpha$ X/CD11c+, Ly-6C+, MAC3+				
Secreted Molecules	IL-12, NO, TNF-α				

### Human Plasmacytoid Dendritic Cells

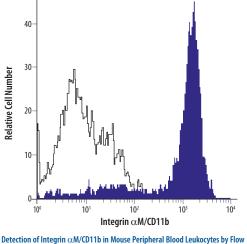
Cell Surface/Membrane-associated	B220/CD45R <sup>+</sup> , DLEC/CLEC4C/BDCA-2 <sup>+</sup> , IL-3 Rcx/CD123 <sup>+</sup> , Integrin cxX/CD11c <sup>low</sup> , Neuropilin-1/BDCA-4 <sup>+</sup> , TLR7 <sup>+</sup> ,
Markers; Transcription Factors	TLR9+; E2-2+, IRF7+, IRF8+, Spi-B+
Secreted Molecules	IFN-α, IFN-β, IL-6, TNF-α

#### Mouse Plasmacytoid Dendritic Cells

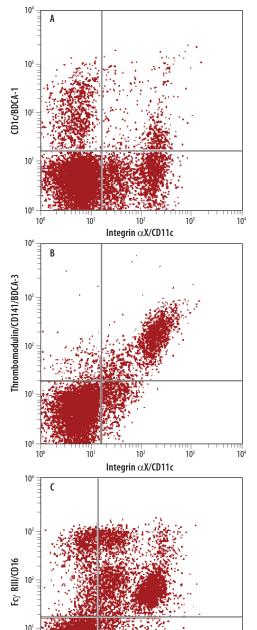
Cell Surface/Membrane-associated Markers; Transcription Factors	$B220/CD45R^+, \text{ Gr-1/Ly-6G^+, Integrin } \alpha X/CD11c^+, Ly-6C^+, PDCA-1/Bst2^+, Siglec-H^+, TLR7^+, TLR9^+; E2-2^+, IRF7^+, IRF8^+, Spi-B^+$
Secreted Molecules	IFN-α, IFN-β, IL-6, IL-12, TNF-α

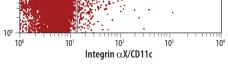


Detection of CLEC9a by Flow Cytometry and Western Blot. Whole human blood was stained with an APC-conjugated Mouse Anti-Human CLEC9a Monoclonal Antibody (Catalog # FAB6049A) and a PE-conjugated Mouse Anti-Human HLA-DR Monoclonal Antibody (Catalog # FAB4869P). The data shown is gated on monocytes based on forward and side scatter parameters. Quadrants were set based on staining with an APC-conjugated Mouse IgG, Isotype Control (Catalog # IC002A). CLEC9a was also detected by Western blot in Iysates from human CD14+ peripheral blood mononuclear cells (inset). The membrane was probed with a Sheep Anti-Human CLEC9a Antigen Affnity-purified Polyclonal Antibody (Catalog # AF6049) followed by an HRP-conjugated Donkey Anti-Sheep IgG Secondary Antibody (Catalog # HAF016).



Systemetry. Mouse peripheral blood leukocytes were stained with a PE-conjugated Rat Anti-Mouse Integrin  $\alpha$ CM/CD11b Monoclonal Antibody (Catalog # FAB1124P; filled histogram) or PE-conjugated Rat  $IgG_{28}$  Isotype Control (Catalog # ICO13P; open histogram).





Detection of Myeloid Dendritic Cells by Multi-Color Flow Cytometry. Human peripheral blood mononuclear cells were stained simultaneously with the indicated antibodies included in the Human Myeloid Dendritic Cell Multi-Color Flow Cytometry Kit (Catalog # FMC016). Dot plots show the relative populations of cells stained with A) APC-conjugated CD1c/BDCA-1 and Fluorescein-conjugated Integrin  $\alpha X/CD11c$ , or C) PerCP-conjugated Fc $\gamma$  RIII/CD16 and Fluorescein-conjugated Integgrin  $\alpha X/CD11c$ . Quadrants were set based on isotype controls. Note: In addition to the population of Integrin  $\alpha X/CD11c$ , Fc $\gamma$  RIII/CD16\* myeloid dendritic cells that are detected in part C, other populations of Fc $\gamma$  RIII/CD16\* cells present in peripheral blood including granulocytes and natural killer cells may also be detected.

To view additional information on DC subsets and a complete product listing, please visit www.RnDSystems.com/DCSubsets

# **Products for Dendritic Cell Subset Research**

# Dendritic Cell Surface Markers

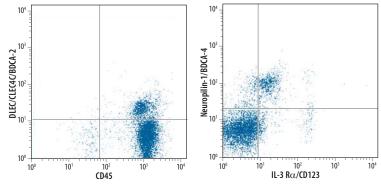
Denuntic Cen 2	Juniace	Indiricers	_				اد مرا تو م	: f	۸.		d:		
	Proteins Fluorochrome Conjugated Antibodies for A Flow Cytometry*						Antibodies for Other Applications						
Molecule	Species		APC	CFS/ FITC	PE	PerCP	Alexa	Fluor® / 700	B/N	E	ICC/ IHC	IP	WB
B220/CD45R	Mouse		~	✓	~	✓	✓				~	~	
BST2/Tetherin	Human				~		~						
CD1a	Human		~		~		~				~		
CD1c/BDCA-1	Human		~		~								✓
CD1d1	Mouse	~											~
CD3	Human										~	~	
	Mouse		~	✓	~	✓	~	~			~	~	
CD4	Human	~	~	✓	~	✓		~	~		✓		~
	Mouse		~	✓	~	✓	~	~			~	~	~
<b>CD8</b> α	Human		~	~	~	✓					~		
	Mouse		~	~	~	~	~				~	~	
CD14	Human	~	~	✓	~	✓		~	~	~	✓		~
	Mouse	~	~			~		~					~
CD19	Human		~	✓	~	✓		~					
	Mouse		~										
CD45	Human	✓	~		~						✓		
	Mouse	<ul> <li>✓</li> </ul>	~	~	~	~					~	~	~
CLEC9a	Human	<ul> <li>✓</li> </ul>	~		~		~	~	~		~	-	~
ceecsu	Mouse	✓	~		~	✓	~				✓		~
DC-SIGN/CD209	Human	✓	~	✓	~	~		~	~			-	✓
DEC-205/CD205	Human	-		~	-	~						-	<ul> <li>✓</li> </ul>
020203/00203	Mouse		~		~								<ul> <li>✓</li> </ul>
E-Cadherin	Human		· •		· •		~			~	<ul> <li>✓</li> </ul>		· ~
E-Caulielili	Mouse	· ·	•	<ul> <li>✓</li> </ul>	•		•			•	•		· ~
EnCAM/Tran1		· ·	• •	• •	• •					•	• •	-	• •
EpCAM/Trop1 F4/80/EMR1	Human Mouse	•	• •	•	• •	· ✓				• •			-
			▼ ✓		▼ ✓	▼ ✓				•		-	
Fcy RIII/CD16	Human	✓	▼ ✓	✓ ✓	✓ ✓	▼ ✓		<ul> <li>✓</li> </ul>	<ul> <li>✓</li> </ul>	✓			✓
CD 4/1 / C	Mouse	~	✓ ✓	✓ ✓	✓ ✓	✓ ✓		~	~	~	✓ ✓	<ul> <li>✓</li> </ul>	~
GR-1/Ly-6G	Mouse		✓ ✓	✓ ✓	✓ ✓	✓ ✓		<ul> <li>✓</li> </ul>			~	~	_
HLA-DR	Human	✓	•	v	✓ ✓	▼ ✓		•	✓		<ul> <li>✓</li> </ul>		<b>√</b>
IL-3 Rα/CD123	Human	✓ ✓			✓ ✓	✓ ✓			~		~		✓ ✓
	Mouse	~				~						-	
Integrin $\alpha$ E/CD103	Mouse		✓ ✓		✓ ✓		<ul> <li>✓</li> </ul>						✓ ✓
Integrin $\alpha$ M/	Human		<ul> <li>✓</li> </ul>		<ul> <li>✓</li> </ul>	<ul> <li>✓</li> </ul>	✓				<ul> <li>✓</li> </ul>		✓
CD11b	Mouse		✓	<ul> <li>✓</li> </ul>	✓	✓		<ul> <li>✓</li> </ul>			✓	✓	
Integrin \alpha X/CD11c	Human	✓	✓	✓	✓	✓		✓				✓	✓
	Mouse		✓									_	✓
Langerin/CD207	Human	✓	✓	✓	✓	✓					✓		✓
MHC class II (I-A/I-E)	Mouse		~	~									
MS4A1/CD20	Human		✓	✓	✓								
NCAM-1/CD56	Human	✓	✓		✓					✓	✓		✓
	Mouse	✓											✓
Neuropilin-1/	Human	✓	~	<b>~</b>	✓	✓		✓	✓		✓		✓
BDCA-4	Mouse	✓	✓		✓		✓		✓		✓		✓
Siglec-H	Mouse		✓		✓		✓						
SIRPa/CD172a	Human	~	~	✓	✓	✓							~
	Mouse	~											~
Thrombomodulin/	Human	~	~	~	~					✓	✓	~	~
CD141/BDCA-3	Mouse	~	~	~	✓						✓	~	~
TLR7	Human			~	~	✓							~
	Mouse												~
TLR9	Human						~						
XCR1	Human			~	~			~			~	1	~

# **Dendritic Cell Secreted Factors**

Molecule	Species	Proteins	ELISAs	ELISpot/FluoroSpot Kits & Development Modules	Multiplex Array/ Assay Kits & Reagents	Antibodies
IFN-a	Human	✓	✓			✓
	Mouse	✓	✓			✓
IFN-β	Human	✓	✓			✓
	Mouse	✓	✓			✓
IFN-γ	Human	✓	✓	✓	✓	✓
	Mouse	✓	✓	✓	✓	✓
IL-1β/IL-1F2	Human	✓	✓	✓	✓	✓
	Mouse	✓	✓		✓	✓
IL-6	Human	✓	✓	✓	✓	✓
	Mouse	✓	✓	✓	✓	✓
IL-10	Human	✓	✓	✓	✓	✓
	Mouse	✓	✓	✓	✓	✓
IL-12	Human	✓	✓	✓	✓	✓
	Mouse	✓	✓	✓	✓	✓
IL-12/IL-23 p40	Human	✓	✓	✓		✓
	Mouse	✓	✓	✓		✓
IL-23	Human	✓	✓	✓	✓	✓
	Mouse	✓	✓		✓	✓
TNF-α	Human	✓	✓	✓	✓	✓
	Mouse	✓	✓	✓	✓	✓

# Multi-Color Flow Cytometry Kits for Dendritic Cell Research

Kits	Kit Antibodies			
Human Myeloid Dendritic Cells	CD1c/BDCA-1-APC, Thrombomodulin/CD141/ BDCA-3-PE, Integrin &X/CD11c-Fluorescein, Fcy RIII/ CD16-PerCP			
Human Plasmacytoid Dendritic Cells	CD45-APC, DLEC/CLEC4C/BDCA-2-PE, IL-3 R/ CD123-PerCP, Neuropilin-1/BDCA-4-Fluorescein			



**Detection of Plasmacytoid Dendritic Cells by Multi-Color Flow Cytometry.** Plasmacytoid dendritic cells, enriched from human peripheral blood mononuclear cells, were stained with the indicated antibodies provided in the Human Plasmacytoid Dendritic Cell Multi-color Flow Cytometry Kit (Catalog # FMC017). Dot plots show the relative populations of cells co-stained with APC-conjugated Mouse Anti-Human CD45 and PE-conjugated Goat Anti-Human DLEC/CLEC4C/BDCA-2 or with PerCP-conjugated Mouse Anti-Human Neuropilin-1/BDCA-4. Quadrants were set based on isotype controls.

R&D Systems offers additional products for other areas of dendritic cell research and for multiple species. For more information and an up-to-date product listing, please visit our website at www.RnDSystems.com/DendriticCells

\*Biotinylated and unlabeled forms are also available for most antibodies.

Application Key: B/N Blocking/Neutralization, E ELISA, ICC/IHC Immunocytochemistry/Immunohistochemistry, IP Immunoprecipitation, WB Western blot Fluorochrome Key: APC Allophycocyanin, CFS/FITC Carboxyfluorescein/Fluorescein isothiocyanate, PE Phycoerythrin, PerCP Peridinin-chlorophyll-protein complex