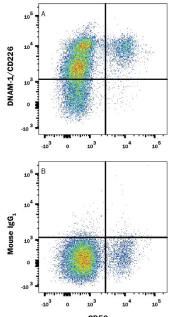
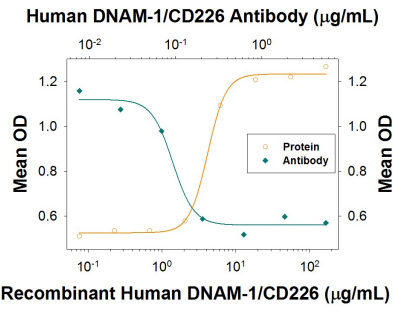


DESCRIPTION	
Species Reactivity	Human
Specificity	Detects human DNAM-1/CD226 in direct ELISAs and Western blots.
Source	Monoclonal Mouse IgG ₁ Clone # 102511
Purification	Protein A or G purified from ascites
Immunogen	Mouse myeloma cell line NS0-derived recombinant human DNAM-1/CD226 Glu19-Asn247 (predicted) Accession # Q15762
Endotoxin Level	<0.10 EU per 1 µg of the antibody by the LAL method.
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

APPLICATIONS		
Please Note: Optimal dilutions should be determined by each laboratory for each application. <i>General Protocols</i> are available in the <i>Technical Information</i> section on our website.		
	Recommended Concentration	Sample
Western Blot	1 µg/mL	Recombinant Human DNAM-1/CD226 Fc Chimera (Catalog # 666-DN) under non-reducing conditions only
Flow Cytometry	0.25 µg/10 ⁶ cells	See Below
CyTOF-ready	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	
Neutralization	Measured by its ability to neutralize DNAM-1/CD226-mediated adhesion of the COLO 205 human colorectal adenocarcinoma cell line. Cosman, D. <i>et al.</i> (1997) <i>Immunity</i> 7:273. The Neutralization Dose (ND ₅₀) is typically 0.1-0.6 µg/mL in the presence of 20 µg/mL Recombinant Human DNAM-1/CD226 Fc Chimera.	

DATA	
<p>Flow Cytometry</p>  <p>Detection of DNAM-1/CD226 in Human PBMCs by Flow Cytometry. Human peripheral blood mononuclear cells (PBMCs) gated on CD3⁺ cells were stained with Mouse Anti-Human NCAM-1/CD56 APC-conjugated Monoclonal Antibody (Catalog # FAB2408A) and either (A) Mouse Anti-Human DNAM-1/CD226 Monoclonal Antibody (Catalog # MAB666) or (B) Mouse IgG₁ Isotype Control (Catalog # MAB002) followed by Phycoerythrin-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # F0102B).</p>	<p>Neutralization</p>  <p>Cell Adhesion Mediated by DNAM-1/CD226 and Neutralization by Human DNAM-1/CD226 Antibody. Recombinant Human DNAM-1/CD226 Fc Chimera (Catalog # 666-DN), immobilized onto a microplate, supports the adhesion of the COLO 205 human colorectal adenocarcinoma cell line in a dose-dependent manner (orange line). Adhesion elicited by Recombinant Human DNAM-1/CD226 Fc Chimera (20 µg/mL) is neutralized (green line) by increasing concentrations of Mouse Anti-Human DNAM-1/CD226 Monoclonal Antibody (Catalog # MAB666). The ND₅₀ is typically 0.1-0.6 µg/mL.</p>

PREPARATION AND STORAGE	
Reconstitution	Reconstitute at 0.5 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	<p>Use a manual defrost freezer and avoid repeated freeze-thaw cycles.</p> <ul style="list-style-type: none"> • 12 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

DNAX accessory molecule-1 (DNAM-1), also known as CD226, is a 65 kDa type I transmembrane glycoprotein in the immunoglobulin superfamily (1). Mature human DNAM-1 contains a 236 amino acid (aa) extracellular domain (ECD) with two Ig-like C2-set domains and a 61 aa cytoplasmic region that contains motifs for binding PDZ domains and band 4.1 family proteins (1, 2). Within the ECD, human DNAM-1 shares 50% and 52% aa sequence identity with mouse and rat DNAM-1, respectively. DNAM-1 is expressed on multiple lymphoid and myeloid cell types and interacts with CD155/PVR and Nectin-2/CD112 (3, 4). Ligation of DNAM-1 promotes the activation of NK cells, CD8⁺ T cells, and mast cells (2-6), dendritic cell maturation, megakaryocyte and activated platelet adhesion to vascular endothelial cells, and monocyte extravasation; it inhibits the formation of osteoclasts (7-10). Platelet-endothelium interactions mediated by DNAM-1 enable the metastasis of tumor cells to the lung (11). In activated, but not in resting NK, T, and mast cells, the *cis* association of DNAM-1 with CD18 contributes to the tyrosine and serine phosphorylation of DNAM-1 during activation (6, 9, 12-14).

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