

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human JAM/VE-JAM in direct ELISAs
Source	Monoclonal Mouse IgG _{2A} Clone # 988905
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	Mouse myeloma cell line NS0-derived recombinant human JAM-B/VE-JAM Phe29-Asn236 Accession # P57087
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. *General Protocols* are available in the *Technical Information* section on our website.

	Recommended Concentration	Sample
Flow Cytometry	0.25 µg/10 ⁶ cells	See Below
Neutralization	Measured by its ability to neutralize JAM-B/VE-JAM-mediated adhesion of the J45.01 human acute lymphoblastic leukemia T lymphocyte cell line. Fong, S. <i>et al.</i> (2002) <i>J. Immunol.</i> 168:1618. The Neutralization Dose (ND ₅₀) is typically 35-210 ng/mL in the presence of 0.2 µg/mL Recombinant Human JAM-B/VE-JAM Fc Chimera.	

DATA

<p>Flow Cytometry</p> <p>Detection of JAM-B/VE-JAM in HEK293 Human Cell Line Transfected with Human JAM-B/VE-JAM and eGFP by Flow Cytometry. HEK293 human embryonic kidney cell line transfected with (A) human JAM-B/VE-JAM or (B) irrelevant transfectants and eGFP was stained with Mouse Anti-Human JAM-B/VE-JAM Monoclonal Antibody (Catalog # MAB10744) followed by APC-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # F0101B). Quadrant markers were set based on control antibody staining (Catalog # MAB003). View our protocol for Staining Membrane-associated Proteins.</p>	<p>Neutralization</p> <p>Human JAM-B/VE-JAM Antibody (µg/mL)</p> <p>Cell Adhesion Mediated by JAM-B/VE-JAM and Neutralization by Human JAM-B/VE-JAM Antibody. Recombinant Human JAM-B/VE-JAM Fc Chimera (Catalog # 1074-VJ), immobilized onto a microplate supports the adhesion of the J45.01 human acute lymphoblastic leukemia T lymphocyte cell line in a dose-dependent manner (orange line), as measured by Calcein AM. Adhesion elicited by Recombinant Human JAM-B/VE-JAM Fc Chimera (0.2 µg/mL) is neutralized (green line) by increasing concentrations of Mouse Anti-Human JAM-B/VE-JAM Monoclonal Antibody (Catalog # MAB10744). The ND₅₀ is typically 35-210 ng/mL.</p> <p>Recombinant Human JAM-B/VE-JAM (µg/mL)</p>
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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	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <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

The family of junctional adhesion molecules (JAM), comprising at least three members, are type I transmembrane receptors belonging to the immunoglobulin (Ig) superfamily (1, 2). These proteins are localized in the tight junctions between endothelial cells or epithelial cells. Some family members are also found on blood leukocytes and platelets. JAM-B, alternatively named vascular endothelial JAM (VE-JAM), is expressed prominently on high endothelial venules of lymphoid organs where it is localized to the intercellular boundaries of high endothelial cells. It is also expressed on the endothelium of a variety of non-lymphoid organs, especially the heart and placenta (3, 5). Human JAM-B cDNA predicts a 298 amino acid (aa) precursor protein with a putative 28 aa signal peptide, a 209 aa extracellular region containing two Ig domains, a 23 aa transmembrane domain and a 38 aa cytoplasmic domain containing a PDZ-binding motif and a PKC phosphorylation site. Human JAM-B shares approximately 79% aa sequence homology with its mouse homologue. It also shares approximately 35% aa sequence homology with human JAM-A or JAM-C. JAM-B exhibits homotypic interactions, as well as heterotypic interactions with JAM-C, but not JAM-A (4, 5, 7). It is also a ligand for the Integrin $\alpha_4\beta_1$. However, the JAM-B/ $\alpha_4\beta_1$ interaction is facilitated only after prior adhesion of JAM-B to JAM-C (6). Through its heterotypic interactions with JAM-C, JAM-B is an adhesive ligand for T, NK, and dendritic cells, and may play a role in regulating leukocyte transmigration (5).

The nomenclature used for the JAM family proteins is confusing. VE-JAM has been referred in the literature variously as JAM-B or JAM-3. Until further clarification, R&D Systems has adopted the nomenclature where both mouse and human VE-JAM are referred to as JAM-B.

References:

1. Chavakis, T. *et al.* (2003) *Thromb. Haemost.* **89**:13.
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3. Palmeri, A. *et al.* (2000) *J. Biol. Chem.* **275**:19139.
4. Cunningham, S. *et al.* (2000) *J. Biol. Chem.* **275**:34750.
5. Liang, T. *et al.* (2002) *J. Immunol.* **168**:1618.
6. Cunningham, A. *et al.* (2002) *J Biol. Chem.* **277**:27589.
7. Arrate, M. *et al.* (2001) *J. Biol. Chem.* **276**:45826.