Mouse/Rat CD31/PECAM-1 Antibody

Antigen Affinity-purified Polyclonal Goat IgG
Catalog Number: AF3628

DESCRIPTION

Species Reactivity	Mouse/Rat
Specificity	Detects mouse CD31/PECAM-1 in direct ELISAs and Western blots. In direct ELISAs and Western blots, approximately 10% cross-reactivity with recombinant human CD31 and recombinant porcine CD31 is observed. Detects mouse CD31 and rat CD31 in flow cytometry.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	Mouse myeloma cell line NS0-derived recombinant mouse CD31/PECAM-1 Glu18-Lys590 Accession # Q08481
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.

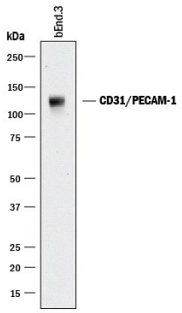
APPLICATIONS

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
Western Blot	0.5 µg/mL	See Below
Flow Cytometry	2.5 µg/10^6 cells	See Below
Immunohistochemistry	5-15 µg/mL	See Below
CyTOF-ready	Ready to be labeled using established conjugation methods. No BSA or other carrier proteins that could interfere with conjugation.	

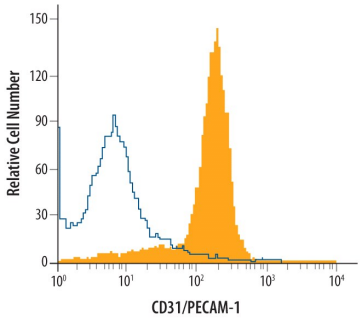
DATA

Western Blot



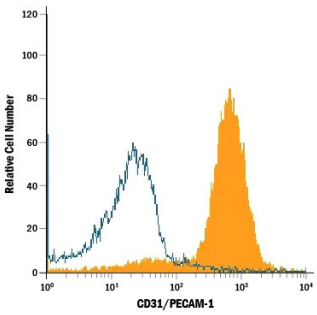
Detection of Mouse CD31/PECAM-1 by Western Blot. Western blot shows lysates of bEnd.3 mouse endothelioma cell line. PVDF membrane was probed with 0.5 µg/mL of Goat Anti-Mouse/Rat CD31/PECAM-1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3628) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF017). A specific band was detected for CD31/PECAM-1 at approximately 130 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Flow Cytometry



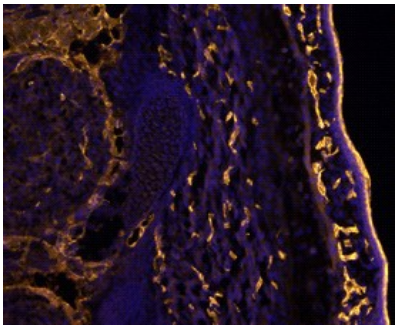
Detection of CD31/PECAM-1 in Mouse Splenocytes by Flow Cytometry. Mouse splenocytes were stained with Goat Anti-Mouse/Rat CD31/PECAM-1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3628, filled histogram) or control antibody (Catalog # AB-108-C, open histogram), followed by Allophycocyanin-conjugated Anti-Goat IgG Secondary Antibody (Catalog # F0108).

Flow Cytometry



Detection of CD31/PECAM-1 in Rat Splenocytes by Flow Cytometry. Rat splenocytes were stained with Goat Anti-Mouse/Rat CD31/PECAM-1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3628, filled histogram) or isotype control antibody (Catalog # AB-108-C, open histogram), followed by Allophycocyanin-conjugated Anti-Goat IgG Secondary Antibody (Catalog # F0108).

Immunohistochemistry



CD31/PECAM-1 in Mouse Embryo. CD31/PECAM-1 was detected in immersion fixed frozen sections of mouse embryo (E13.5) using Goat Anti-Mouse/Rat CD31/PECAM-1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3628) at 10 µg/mL overnight at 4 °C. Tissue was stained using the NorthernLights™ 557-conjugated Anti-Goat IgG Secondary Antibody (yellow; Catalog # NL001) and counterstained with DAPI (blue). Specific staining was localized to developing endothelium. View our protocol for Fluorescent IHC Staining of Frozen Tissue Sections.

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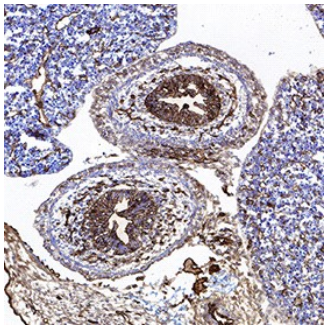
Immunohistochemistry



CD31/PECAM-1 in Mouse Embryo. CD31/PECAM-1 was detected in immersion fixed frozen sections of mouse embryo (14 d.p.c.) using Goat Anti-Mouse/Rat CD31/PECAM-1 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3628) at 10 µg/mL for 1 hour at room temperature followed by incubation with the Anti-Goat IgG VisUCyte™ HRP Polymer Antibody (Catalog # VC004). Tissue was stained using DAB (brown) and counterstained with hematoxylin (blue). Specific staining was localized to developing guts. View our protocol for IHC Staining with VisUCyte HRP Polymer Detection Reagents.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 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.** ● 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

PECAM-1 (Platelet-Endothelial Cell Adhesion Molecule-1), also known as CD31, is a 130 kDa type I transmembrane glycoprotein adhesion molecule in the immunoglobulin superfamily (1, 2). Expression is restricted to cells involved in circulation, especially endothelial cells, platelets, monocytes, neutrophils and lymphocyte subsets. PECAM-1 is concentrated at cell-cell junctions and is required for Transendothelial Migration (TEM) (1-3). The Extracellular Domain (ECD) of PECAM-1 has ten potential N-linked glycosylation sites and six C2-type Ig-like domains, the first of which is critical for adhesion and extravasation (3, 4). The cytoplasmic domain contains Immunoregulatory Tyrosine-based Inhibitory and Switch Motifs (ITIM, ITSM) that mediate both inhibition and activation via phosphotyrosine-mediated engagement of SH2-containing signaling molecules (1, 5). Metalloproteinase-mediated ectodomain shedding occurs during apoptosis (6) but increased serum PECAM-1 ectodomain in HIV and active multiple sclerosis occurs independent of apoptosis (7, 8). In humans, expression of six isoforms with exon deletions in the cytoplasmic domain is tissue- and stage-specific, but full-length PECAM-1 is predominant. A form lacking the ITSM predominates in mouse (9). Mouse PECAM-1 ECD shows 77%, 63%, 63%, 63%, and 61% amino acid (aa) identity with rat, human, canine, porcine, and bovine PECAM-1, respectively. PECAM-1 participates with other adhesion molecules in some functions, but is the critical molecule for TEM. Homotypic PECAM-1 adhesion in trans, combined with cycling of PECAM-1 to and from surface-connected endothelial cell vesicles, leads leukocytes across endothelial tight junctions (3, 10). Homotypic adhesion and signaling functions also strongly suppress mitochondria-dependent apoptosis (11). In platelets, PECAM-1 is necessary for limiting thrombus formation (12) and promoting integrin-mediated clot retraction and platelet spreading (13), but mechanisms for these phenomena are unclear. PECAM$^{-/-}$ mice are deficient in chemokine-mediated chemotaxis (14).

References:

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