

DESCRIPTION

Species Reactivity	Mouse
Specificity	Detects recombinant mouse CD31/PECAM1 protein in Direct ELISA.
Source	Monoclonal Rat IgG _{2A} Clone # 1094515
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	Mouse myeloma cell line, NS0-derived mouse CD31/PECAM-1 Glu18-Lys590 Accession # Q08481
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose.

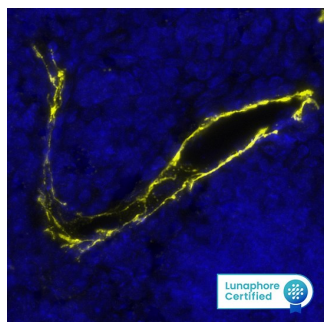
APPLICATIONS

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	2 µg/mL	bEnd.3 mouse endothelioma cell line
Multiplex Immunofluorescence	3 µg/mL	Perfusion fixed paraffin-embedded sections of mouse thymus
Immunohistochemistry	0.5-25 µg/mL	Perfusion fixed paraffin-embedded sections of mouse kidney, liver, and pancreas

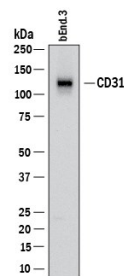
DATA

Multiplex Immunofluorescence



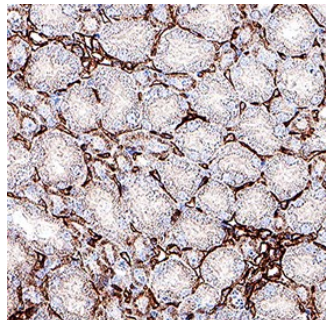
Detection of CD31 in Mouse Thymus via seqIF™ staining on COMET™ CD31 was detected in immersion fixed paraffin-embedded sections of Mouse Thymus using Rat Anti-Mouse CD31, Monoclonal Antibody (Catalog #MAB) at 3µg/mL at 37 ° Celsius for 4 minutes. Before incubation with the primary antibody, tissue underwent an all-in-one dewaxing and antigen retrieval preprocessing using PreTreatment Module (PT Module) and Dewax and HIER Buffer H (pH 9; Eprelia Catalog # TA-999-DHBH). Tissue was stained using the Alexa Fluor™ 647 Goat anti-Rat IgG Secondary Antibody at 1:200 at 37 ° Celsius for 2 minutes. (Yellow; Lunaphore Catalog # DR647RT) and counterstained with DAPI (blue; Lunaphore Catalog # DR100). Specific staining was localized to the membrane. Protocol available in [COMET™ Panel Builder](#).

Western Blot



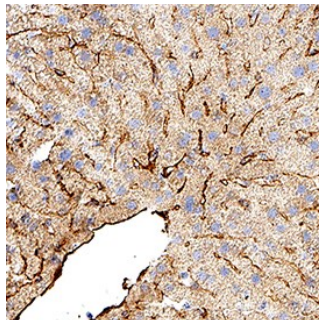
Detection of Mouse CD31/PECAM-1 by Western Blot. Western Blot shows lysates of bEnd.3 mouse endothelioma cell line. PVDF membrane was probed with 2 µg/ml of Rat Anti-Mouse CD31/PECAM-1 Monoclonal Antibody (Catalog # MAB11714) followed by HRP-conjugated Anti-Rat IgG Secondary Antibody (Catalog # HAF005). A specific band was detected for CD31/PECAM-1 at approximately 130 kDa (as indicated). This experiment was conducted under reducing conditions and using [Western Blot Buffer Group 1](#).

Immunohistochemistry



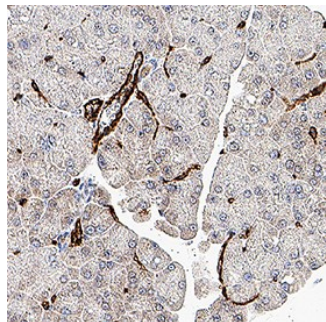
Detection of CD31/PECAM-1 in Mouse Kidney. CD31/PECAM-1 was detected in perfusion fixed paraffin-embedded sections of mouse kidney using Rat Anti-Mouse CD31/PECAM-1 Monoclonal Antibody (Catalog # MAB11714) at 0.5 µg/ml overnight at 4 °C. Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using VisUCyte Antigen Retrieval Reagent-Basic (Catalog # VCTS021). Tissue was stained using the HRP-conjugated Anti-Rat IgG Secondary Antibody (Catalog # HAF005) and counterstained with hematoxylin (blue). Specific staining was localized to the membrane of endothelial cells. View our protocol for [Chromogenic IHC Staining of Paraffin-embedded Tissue Sections](#).

Immunohistochemistry



Detection of CD31/PECAM-1 in Mouse Liver. CD31/PECAM-1 was detected in perfusion fixed paraffin-embedded sections of mouse liver using Rat Anti-Mouse CD31/PECAM-1 Monoclonal Antibody (Catalog # MAB11714) at 0.5 µg/ml overnight at 4 °C. Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using VisUCyte Antigen Retrieval Reagent-Basic (Catalog # VCTS021). Tissue was stained using the HRP-conjugated Anti-Rat IgG Secondary Antibody (Catalog # HAF005) and counterstained with hematoxylin (blue). Specific staining was localized to the membrane of endothelial cells. View our protocol for [Chromogenic IHC Staining of Paraffin-embedded Tissue Sections](#).

Immunohistochemistry



Detection of CD31/PECAM-1 in Mouse Pancreas. CD31/PECAM-1 was detected in perfusion fixed paraffin-embedded sections of mouse pancreas using Rat Anti-Mouse CD31/PECAM-1 Monoclonal Antibody (Catalog # MAB11714) at 0.5 µg/ml overnight at 4 °C. Before incubation with the primary antibody, tissue was subjected to heat-induced epitope retrieval using VisUCyte Antigen Retrieval Reagent-Basic (Catalog # VCTS021). Tissue was stained using the HRP-conjugated Anti-Rat IgG Secondary Antibody (Catalog # HAF005) and counterstained with hematoxylin (blue). Specific staining was localized to the membrane of endothelial cells. View our protocol for [Chromogenic IHC Staining of Paraffin-embedded Tissue Sections](#).

PREPARATION AND STORAGE

Reconstitution	Reconstitute lyophilized material at 0.2 mg/ml in sterile PBS. For liquid material, refer to CoA for concentration.
Shipping	Lyophilized product is shipped at ambient temperature. Liquid small pack size (-SP) is shipped with polar packs. Upon receipt, store immediately at the temperature recommended below.
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

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. CD31 is concentrated at cell-cell junctions and is required for transendothelial migration (TEM) (1-3). The extracellular domain (ECD) of CD31 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 CD31 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 CD31 is predominant. A form lacking the ITSM predominates in mouse (9). Mouse CD31 ECD shows 77%, 63%, 63%, 63% and 61% amino acid (aa) identity with rat, human, canine, porcine and bovine CD31, respectively. CD31 participates with other adhesion molecules in some functions, but is the critical molecule for TEM. Homotypic CD31 adhesion in trans, combined with cycling of CD31 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, CD31 is necessary for limiting thrombus formation (12) and promoting integrin-mediated clot retraction and platelet spreading (13), but mechanisms for these phenomena are unclear. CD31^{-/-} mice are deficient in chemokine-mediated chemotaxis (14).

References:

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