

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human CD9 in direct ELISAs.
Source	Recombinant Monoclonal Rabbit IgG Clone # 2665FF
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	Chinese Hamster Ovary cell line, CHO-derived human CD9 Ser112-Ile195 Accession # P21926
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	HUVEC human umbilical vein endothelial cells and HCT-116 human colorectal carcinoma cell line
Simple Western	50 µg/mL	HUVEC human umbilical vein endothelial cell line whole cell lysate (WCL), Exosome Standards (HT-29 cell line) (NBP3-11685) and Exosome Standards (HCT-116 cell line) (NBP2-49854)

DATA

Western Blot

Detection of Human CD9 by Western Blot. Western Blot shows lysates of HUVEC human umbilical vein endothelial cells and HCT-116 human colorectal carcinoma cell line. PVDF membrane was probed with 2 µg/ml of Rabbit Anti-Human CD9 Monoclonal Antibody (Catalog # MAB11464) followed by HRP-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # HAF008). A specific band was detected for CD9 at approximately 23 kDa (as indicated). This experiment was conducted under reducing conditions and using Western Blot Buffer Group 1.

Simple Western

Detection of Human CD9 by Simple Western™. Simple Western shows lysates of HUVEC human umbilical vein endothelial cell line whole cell lysate (WCL), Exosome Standards (HT-29 cell line) (Catalog # NBP3-11685) and Exosome Standards (HCT-116 cell line) (Catalog # NBP2-49854), loaded at 0.5 mg/ml. A specific band was detected for CD9 at approximately 33 kDa (as indicated) using 50 µg/mL of Rabbit Anti-Human CD9 Monoclonal Antibody (Catalog # MAB11464) followed by 1:50 dilution of HRP-conjugated Anti-Rabbit IgG Secondary Antibody (Catalog # HAF008). This experiment was conducted under non-reducing conditions and using the 2-40kDa separation system.

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.
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

CD9, also known as Tspan29, is a 24-27 kDa cell surface protein belonging to the tetraspanin family (1). Common to other tetraspanins, CD9 is composed of four transmembrane domains, short N- and C-terminal cytoplasmic domains, and two extracellular loops. The larger extracellular loop, referred to as the LEL or EC2, contains highly conserved CCG and PXSC motifs (2, 3). The LEL mediates noncovalent protein-protein interactions, allowing tetraspanins to associate with each other as well as signaling molecules, structural proteins, and G-protein coupled receptors (4-6). Human CD9 is expressed in multiple cell and tissue types and has been identified in diverse biological roles due to its involvement in the formation of tetraspanin-enriched microdomains (TEMs). TEMs are associated with numerous processes ranging from cell adhesion and fusion, membrane trafficking, and endocytosis to leukocyte adherence and motility (4-7). These tetraspanin-enriched microdomains (TEMs) are associated with a wide range of functions from cell adhesion and fusion, membrane trafficking and endocytosis, and eukocyte adherence and motility. The LEL of human CD9 shares 77% and 84% amino acid sequence identity with mouse and rat CD9, respectively. CD9 can form homodimers or interact with other proteins including CD117, CD29, CD46, CD49c, CD81, CD315, Tspan4, TGF- α , and HBEGF (1, 4, 8-13). Increased expression of CD9 has been shown to enhance transmembrane TGF- α -induced EGFR stimulation (1), and injection of human CD9 mRNA into CD9 knock-out mouse oocytes restored sperm-egg fusion (14). CD9-LEL may also be involved in the inhibition of multinucleated giant cell formation (3) as well as possess anti-adhesive effects against bacteria trying to invade mammalian cells (6, 15). CD9 interacts with integrins to regulate cell adhesion and motility (16-18). CD9 has been implicated in platelet activation and aggregation (17, 19). It may act as the terminal signal of myelination in the peripheral nervous system and can regulate the formation of paranodal junctions (20). Also, it has been suggested CD9 plays an important role both in the self-antigen and recall antigen-induced T cell activation (21).

References:

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