Anti-Syntenin Antibody [JE40-72]

ET7109-14

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, FC, IP
Molecular Wt:	Predicted band size: 32 kDa
Clone number:	JE40-72

Description: Syntenin-1 (also known as syntenin, syndecan binding protein, melanoma differentiation-associated protein 9 or proTGF-alpha cytoplasmic domain-interacting protein 18) is a protein that binds to the cytoplasmic domains of the syndecans in yeast 2-hybrid screens and other assays. Syntenin-1 contains a tandem repeat of PDZ domains that reacts with the FYA (phe-tyr-ala) carboxy-terminal amino acid sequence of the syndecans. It may function as an adaptor that couples syndecans to cytoskeletal proteins or cytosolic downstream signal-effectors. Syntenin-1 colocalizes and interacts specifically with immature, intracellular forms of proTGF-alpha. It is a human gamma interferon responsive protein. Syntenin-1 contains PSD-95/Discs large/zO-1 (PDZ) domains and associates with the cytoplasmic tail of the IL-5Ralpha. It directly associates with the transcription factor Sox-4. The PDZ proteins PICK1, GRIP, ABP and syntenin-1 bind multiple glutamate receptor subtypes.

Immunogen: Recombinant protein within Human Syntenin aa 1-118 / 298.

Positive control: HeLa cell lysate, A549 cell lysate, 293T cell lysate, HeLa, human tonsil tissue, human colon tissue.

Subcellular location: Cytoplasm. Cytoskeleton. Membrane. Nucleus. Secreted.

Database links: SwissProt: O00560 Human

Recommended Dilutions:

WB	1:2,000-1:5,000
IF-Cell	1:50-1:200
IF-Tissue	1:50-1:200
IHC-P	1:50-1:200
FC	1:1,000
IP	1:10-1:50

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.

Purity: Protein A affinity purified.

Images

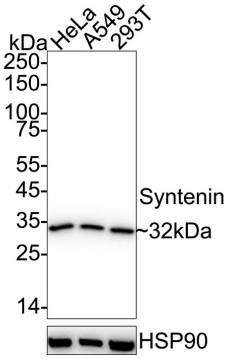

Fig1: Western blot analysis of Syntenin on different lysates with Rabbit anti-Syntenin antibody (ET7109-14) at 1/2,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: A549 cell lysate
Lane 3: 293T cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 32 kDa
Observed band size: 32 kDa

Exposure time: 5 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET7109-14) at 1/2,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

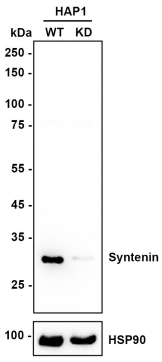

Fig2: Western blot analysis of Syntenin on different lysates with Rabbit anti-Syntenin antibody (ET7109-14) at 1/5,000 dilution.

Lane 1: HAP1-parental cell lysate
Lane 2: HAP1-Syntenin KD cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 32 kDa
Observed band size: 32 kDa

Exposure time: 30 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET7109-14) at 1/5,000 dilution was used in K1803 at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

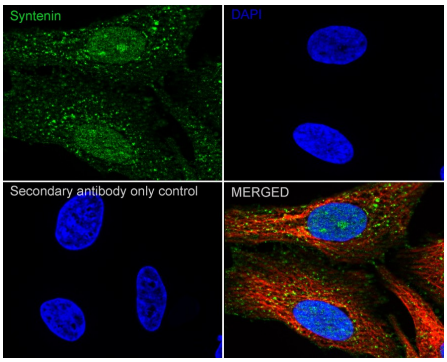

Fig3: Immunocytochemistry analysis of HeLa cells labeling Syntenin with Rabbit anti-Syntenin antibody (ET7109-14) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-Syntenin antibody (ET7109-14) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

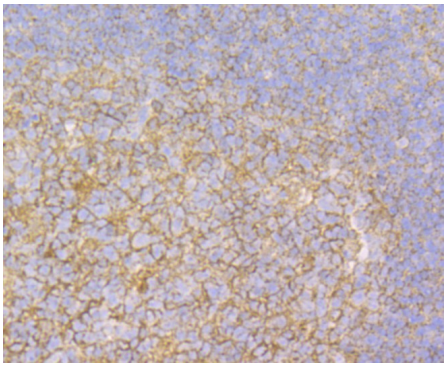

Fig4: Immunohistochemical analysis of paraffin-embedded human tonsil tissue using anti-Syntenin antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes.The tissues were blocked in 1% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET7109-14, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

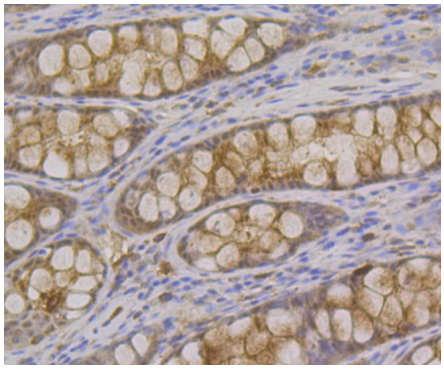

Fig5: Immunohistochemical analysis of paraffin-embedded human colon tissue using anti-Syntenin antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes.The tissues were blocked in 1% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET7109-14, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

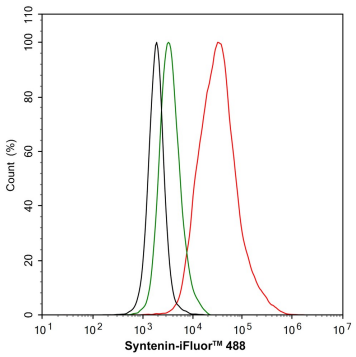

Fig6: Flow cytometric analysis of HeLa cells labeling Syntenin.

Cells were fixed and permeabilized. Then stained with the primary antibody (ET7109-14, 1/1,000) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4℃ for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. Fernandez-Larrea J et al. A role for a PDZ protein in the early secretory pathway for the targeting of proTGF-alpha to the cell surface. Mol Cell 3:423-433 (1999).
2. Zimmermann P et al. Characterization of syntenin, a syndecan-binding PDZ protein, as a component of cell adhesion sites and microfilaments. Mol Biol Cell 12:339-350 (2001).