Anti-S100 alpha 6 Antibody [JF0976]

ET1702-28

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, IP, FC
Molecular Wt:	Predicted band size: 10 kDa
Clone number:	JF0976

Description: Calcyclin, also known as Prolactin receptor-associated protein (PRA), growth factor-inducible protein 2A9, S-100 calcium-binding protein A6 (S-100A6) or MLN 4, is a homodimeric member of the S-100 calcium-binding protein family whose expression is upregulated in proliferating and differentiating cells. Calcyclin is inducible by growth factors and overexpressed in acute myeloid leukemias. It is expressed in a cell-specific manner in subpopulations of neurons and astrocytes and in epithelial cells and fibroblasts. Calcyclin is a specific target of S-100B protein in vivo. The binding of Calcyclin to S-100B is stabilized by S-100B-bound calcium and zinc. Calcyclin associates with both Annexin XI and CacyBP (calcyclin-binding protein). It functions to activate several processes along the calcium signal transduction pathway including the regulation of cell growth, proliferation, secretion and exocytosis.

Immunogen: Recombinant full length protein of Human S100 alpha 6 aa 1-90 / 90.

Positive control: HeLa cell lysate, A549 cell lysate, Neuro-2a cell lysate, C6 cell lysate, mouse lung tissue lysate, C6, human kidney tissue, human stomach carcinoma tissue, human colon carcinoma tissue, mouse brain tissue, mouse kidney tissue, rat kidney tissue.

Subcellular location: Nucleus envelope, Cytoplasm, Cell membrane.

Database links: SwissProt: P06703 Human | P14069 Mouse | P05964 Rat

Recommended Dilutions:

WB	1:2,000-1:5,000
IF-Cell	1:100-1:500
IF-Tissue	1:100-1:500
IHC-P	1:50-1:1,000
FC	1:50-1:100
IP	Use at an assay dependent concentration.

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

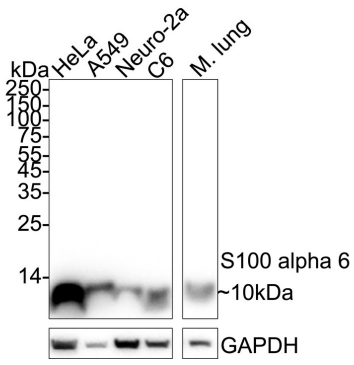

Fig1: Western blot analysis of S100 alpha 6 on different lysates with Rabbit anti-S100 alpha 6 antibody (ET1702-28) at 1/2,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: A549 cell lysate
Lane 3: Neuro-2a cell lysate
Lane 4: C6 cell lysate
Lane 5: Mouse lung tissue lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 10 kDa
Observed band size: 10 kDa

Exposure time: 10 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 (ET1702-28) 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.

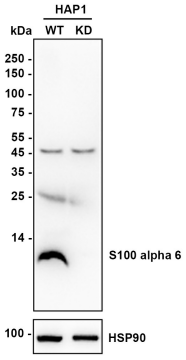

Fig2: Western blot analysis of S100 alpha 6 on different lysates with Rabbit anti-S100 alpha 6 antibody (ET1702-28) at 1/2,000 dilution.

Lane 1: HAP1-parental cell lysate
Lane 2: HAP1-S100 alpha 6 KD cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 10 kDa
Observed band size: 10 kDa

Exposure time: 1 minute; 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 (ET1702-28) at 1/2,000 dilution was used in primary antibody dilution (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.

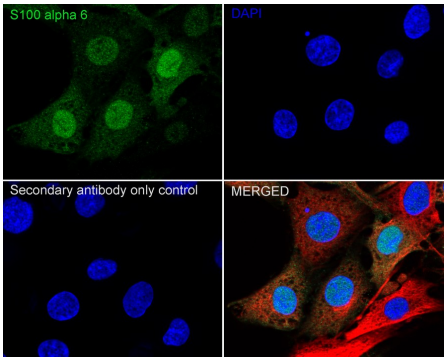

Fig3: Immunocytochemistry analysis of C6 cells labeling S100 alpha 6 with Rabbit anti-S100 alpha 6 antibody (ET1702-28) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 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-S100 alpha 6 antibody (ET1702-28) 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 (HA601187, 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.

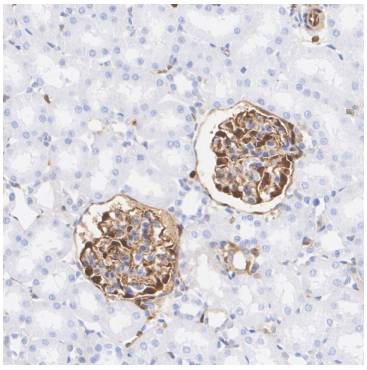

Fig4: Immunohistochemical analysis of paraffin-embedded rat kidney tissue with Rabbit anti-S100 alpha 6 antibody (ET1702-28) at 1/1,000 dilution.

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 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1702-28) at 1/1,000 dilution for 1 hour 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.

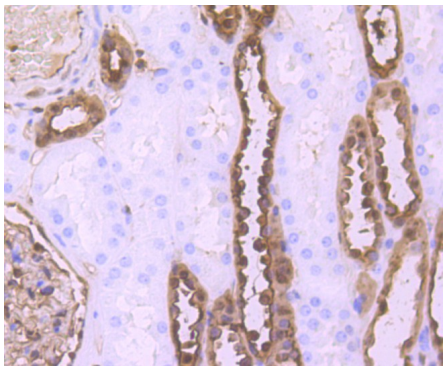

Fig5: Immunohistochemical analysis of paraffin-embedded human kidney tissue using anti-S100 alpha 6 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1702-28, 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.

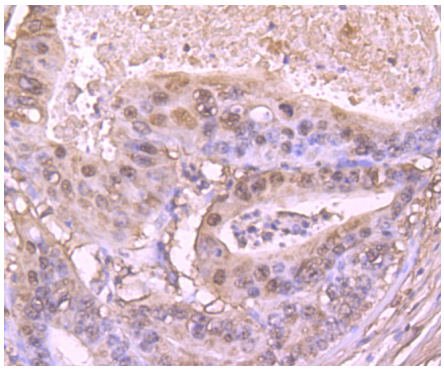

Fig6: Immunohistochemical analysis of paraffin-embedded human stomach carcinoma tissue using anti-S100 alpha 6 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1702-28, 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.

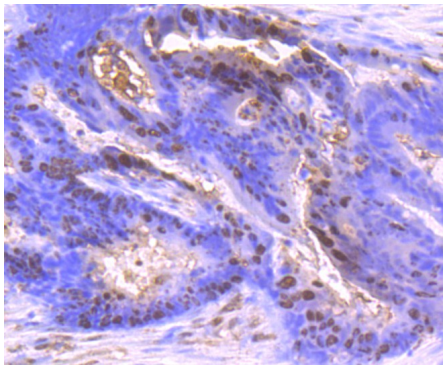

Fig7: Immunohistochemical analysis of paraffin-embedded human colon carcinoma tissue using anti-S100 alpha 6 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1702-28, 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.

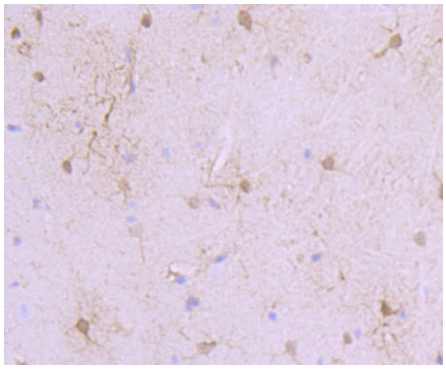

Fig8: Immunohistochemical analysis of paraffin-embedded mouse brain tissue using anti-S100 alpha 6 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1702-28, 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.

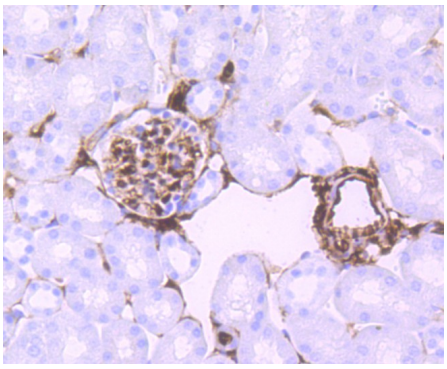

Fig9: Immunohistochemical analysis of paraffin-embedded mouse kidney tissue using anti-S100 alpha 6 antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1702-28, 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.

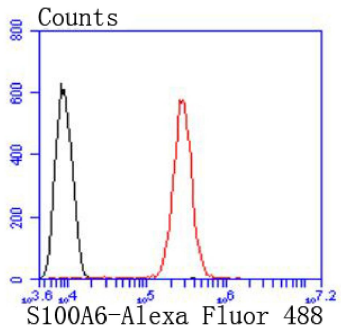

Fig10: Flow cytometric analysis of S100 alpha 6 was done on SH-SY5Y cells. The cells were fixed, permeabilized and stained with the primary antibody (ET1702-28, 1/50) (red). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor 488-conjugated Goat anti-Rabbit IgG Secondary antibody at 1/1000 dilution for 30 minutes.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. Ralhan R et al. Immunohistochemical Subcellular Localization of Protein Biomarkers Distinguishes Benign from Malignant Thyroid Nodules: Potential for Fine-Needle Aspiration Biopsy Clinical Application. Thyroid 25:1224-34 (2015).
2. Alves RM et al. iTRAQ-based quantitative proteomic analysis of submandibular glands from rats with STZ-induced hyperglycemia. J Biochem 153:209-20 (2013).