Anti-14-3-3 gamma Antibody [SD20-65]

ET1612-9

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IHC-P, FC
Molecular Wt:	Predicted band size: 28 kDa
Clone number:	SD20-65

Description: 14-3-3 proteins regulate many cellular processes relevant to cancer biology, notably apoptosis, mitogenic signaling and cell-cycle checkpoints. Seven isoforms comprise this family of signaling intermediates, denoted 14-3-3 b, g, e, z, h, q and s. 14-3-3 proteins form dimers that present two binding sites for ligand proteins, thereby bringing together two proteins that may not otherwise associate. These ligands largely share a 14-3-3 consensus binding motif and exhibit serine/threonine phosphorylation. 14-3-3 proteins function in broad regulation of these ligand proteins; by cytoplasmic sequestration, occupation of interaction domains and import/export sequences, prevention of degradation, activation/repression of enzymatic activity, and facilitation of protein modification. Loss of expression contributes to a vast array of pathogenic cellular activities.

Immunogen: Synthetic peptide within Human 14-3-3 gamma aa 131-180 / 247.

Positive control: 293T cell lysate, K562 cell lysate, A431 cell lysate, Hela cell lysate, K562, human breast cancer tissue, human lung cancer tissue, human ovary cancer tissue.

Subcellular location: Cytoplasm.

Database links: SwissProt: P61981 Human | P61982 Mouse | P61983 Rat

Recommended Dilutions:

WB	1:1,000-1:2,000
IHC-P	1:200-1:1,000
FC	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

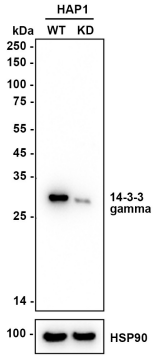

Fig1: Western blot analysis of 14-3-3 gamma on different lysates with Rabbit anti-14-3-3 gamma antibody (ET1612-9) at 1/2,000 dilution.

Lane 1: HAP1-parental cell lysate
Lane 2: HAP1-14-3-3 gamma KD cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 28 kDa
Observed band size: 28 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 (ET1612-9) 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.

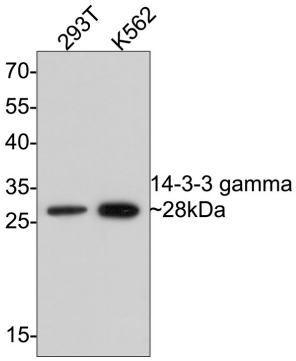

Fig2: Western blot analysis of 14-3-3 gamma on different lysates with Rabbit anti-14-3-3 gamma antibody (ET1612-9) at 1/1,000 dilution.

Lane 1: 293T cell lysate
Lane 2: K562 cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 28 kDa
Observed band size: 28 kDa

Exposure time: 2 minutes;

12% 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 (ET1612-9) at 1/1,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:300,000 dilution was used for 1 hour at room temperature.

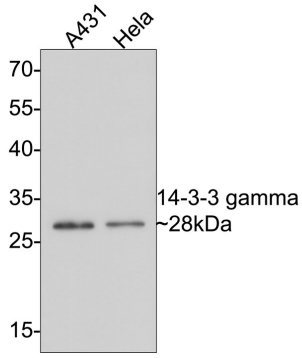

Fig3: Western blot analysis of 14-3-3 gamma on different lysates with Rabbit anti-14-3-3 gamma antibody (ET1612-9) at 1/500 dilution.

Lane 1: A431 cell lysate
Lane 2: Hela cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 28 kDa
Observed band size: 28 kDa

Exposure time: 2 minutes;

12% 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 (ET1612-9) at 1/500 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:300,000 dilution was used for 1 hour at room temperature.

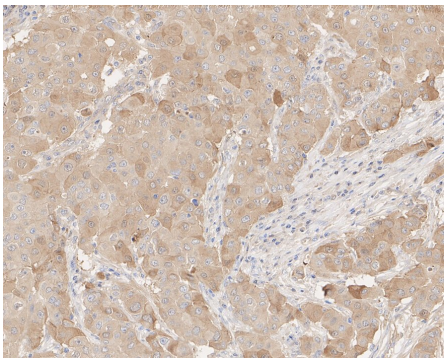

Fig4: Immunohistochemical analysis of paraffin-embedded human breast cancer tissue with Rabbit anti-14-3-3 gamma antibody (ET1612-9) at 1/1,000 dilution.

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 (ET1612-9) 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.

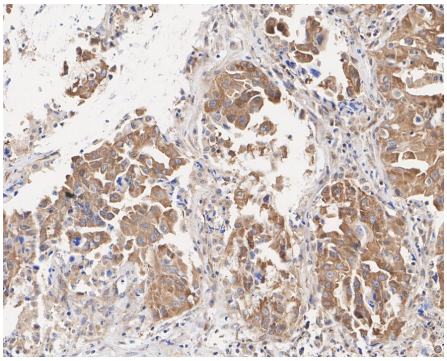

Fig5: Immunohistochemical analysis of paraffin-embedded human lung cancer tissue with Rabbit anti-14-3-3 gamma antibody (ET1612-9) at 1/1,000 dilution.

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 (ET1612-9) 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.

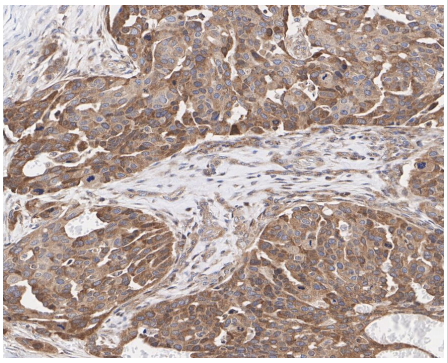

Fig6: Immunohistochemical analysis of paraffin-embedded human ovary cancer tissue with Rabbit anti-14-3-3 gamma antibody (ET1612-9) at 1/200 dilution.

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 (ET1612-9) at 1/200 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.

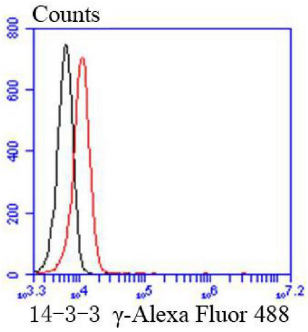

Fig7: Flow cytometric analysis of 14-3-3 gamma was done on K562 cells. The cells were fixed, permeabilized and stained with the primary antibody (ET1612-9, 1/50) (red). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor®488 conjugate-Goat anti-Rabbit IgG Secondary antibody at 1/1,000 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. Scheibner, KA. et al. 2012. MiR-27a functions as a tumor suppressor in acute leukemia by regulating 14-3-3θ. PLoS ONE. 7: e50895.
2. Song, Y. et al. 2012. Expression of 14-3-3γ in patients with breast cancer: correlation with clinicopathological features and prognosis. Cancer Epidemiol. 36: 533-536.