Anti-Phospho-p53 (S15) Antibody [PSH01-98] - BSA and Azide free

HA750772



Species reactivity: Human

Applications: WB, IF-Cell

Molecular Wt: Predicted band size: 53 kDa

Clone number: PSH01-98

Description: p53, a DNA-binding, oligomerization domain- and transcription activation domain-containing

tumor suppressor, upregulates growth arrest and apoptosis-related genes in response to stress signals, thereby influencing programmed cell death, cell differentiation, and cell cycle control mechanisms. p53 localizes to the nucleus, yet can be chaperoned to the cytoplasm by the negative regulator, MDM2. MDM2 is an E3 ubiquitin ligase that is upregulated in the presence of active p53, where it poly-ubiquitinates p53 for proteasome targeting. p53 fluctuates between latent and active DNA-binding conformations and is differentially activated through posttranslational modifications, including phosphorylation and acetylation. Mutations in the DNA-binding domain (DBD) of p53, amino acids 110-286, can compromise energetically-favorable association with cis elements and are implicated in several human

cancers.

Immunogen: Synthetic phospho-peptide corresponding to residues surrounding Ser15 of Human p53 aa

1-50 / 393.

Positive control: HT-29 treated with 0.5µM Doxorubicin for 24 hours cell lysate, HT-29 treated with 0.5µM

Doxorubicin for 24 hours cell.

Subcellular location: Cytoplasm, Nucleus, Nucleus, PML body, Endoplasmic reticulum, Mitochondrion matrix,

Cytoplasm, cytoskeleton, microtubule organizing center, centrosome.

Database links: SwissProt: P04637 Human

Recommended Dilutions:

WB 1:1,000 IF-Cell 1:10,000

Storage Buffer: PBS (pH7.4).

Storage Instruction: Store at +4 °C after thawing. Aliquot store at -20 °C. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

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Images

kDa 250-150-150-100-100-50-37-25-20-15-10-GAPDH - + + Doxorubicin - - + λpp **Fig1:** Western blot analysis of Phospho-p53 (S15) on different lysates with Rabbit anti-Phospho-p53 (S15) antibody (HA750772) at 1/1,000 dilution.

Lane 1: HT-29 cell lysate

Lane 2: HT-29 treated with 0.5 μ M Doxorubicin for 24 hours cell

lysate

Lane 3: HT-29 treated with 0.5 μM Doxorubicin for 24 hours, then

treated with λpp for 1 hour cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 53 kDa Observed band size: 53 kDa

Exposure time: 1 minute 46 seconds;

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 (HA750772) 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 1/50,000 dilution was used for 1 hour at room temperature.

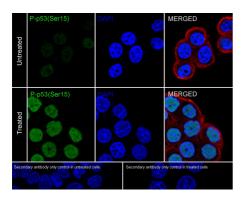


Fig2: Immunocytochemistry analysis of HT-29 treated with or without 0.5μM Doxorubicin for 24 hours cells labeling Phosphop53 (S15) with Rabbit anti-Phospho-p53 (S15) antibody (HA750772) at 1/10,000 dilution.

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-Phospho-p53 (S15) antibody (HA750772) at 1/10,000 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 $+4^{\circ}$ C. Goat Anti-Mouse IgG H&L (iFluor 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

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

Background References

- 1. Hu J et al. Targeting mutant p53 for cancer therapy: direct and indirect strategies. J Hematol Oncol. 2021 Sep
- 2. Hassin O et al. Drugging p53 in cancer: one protein, many targets. Nat Rev Drug Discov. 2023 Feb