Anti-Phospho-p53 (S392) Antibody [SI17-04]

ET1606-24

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IHC-P, IP
Molecular Wt:	Predicted band size: 53 kDa
Clone number:	SI17-04

Description: p53 is a DNA-binding, oligomerization domain- and transcription activation domain-containing tumor suppressor that 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, an E3 ubiquitin ligase that is upregulated in the presence of active p53, where MDM2 polyubiquitinates p53 for proteasome targeting. p53 can assemble into tetramers in the absence of DNA, 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) (amino acids 110-286) of p53 can compromise energetically favorable association with cis elements and are implicated in several human cancers. Phosphorylation of p53 at residue Thr 155 is mediated by the COP9 signalosome (CSN) and targets p53 to ubiquitin-26S Proteasome-dependent degradation.

Immunogen: Synthetic phospho-peptide corresponding to residues surrounding Ser392 of Human p53 aa 344-393 / 393.

Positive control: 293 cell lysate, F9 cell lysate, A431 cell lysate, human stomach carcinoma tissue, mouse prostate tissue, A549 cells.

Subcellular location: Cytoplasm, Nucleus, Endoplasmic reticulum, Mitochondrion matrix.

Database links: SwissProt: P04637 Human | P02340 Mouse | P10361 Rat

Recommended Dilutions:

WB	1:1,000-1:5,000
IHC-P	1:50-1:200
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: Store at +4℃ after thawing. Aliquot store at -20℃ or -80℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

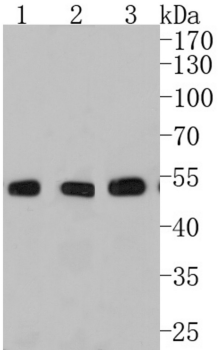

Fig1: Western blot analysis of Phospho-p53 (S392) on different cell lysates with Rabbit anti-Phospho-p53 (S392) antibody (ET1606-24) at 1:1,000 dilution.

Lane 1: 293 cell lysate
Lane 2: F9 cell lysate
Lane 3: A431 cell lysate

Lysates/proteins at 10 µg/Lane.

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

Exposure time: 30 Seconds;

10% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% BSA for 1 hour at room temperature. The primary antibody (ET1606-24) at 1:1,000 dilution was used in PBS 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.

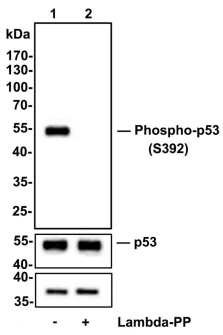

Fig2: Western blot analysis of Phospho-p53(S392) on 293 cell lysates.

Lane 1: 293 cells, whole cell lysate, 10ug/lane
Lane 2: 293 cells treated with 2.8ug/ul lambda-PP for 30 minutes, whole cell lysates, 10ug/lane

All lanes :
Anti-Phospho-p53(S392) antibody (ET1606-24) at 1:500 dilution. Anti-GAPDH antibody (ET1601-4) at 1:10,000 dilution. Goat Anti-Rabbit IgG H&L (HRP) (HA1001) at 1/200,000 dilution.

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

Blocking and diluting buffer: 5% BSA.

Exposure time: 10 seconds

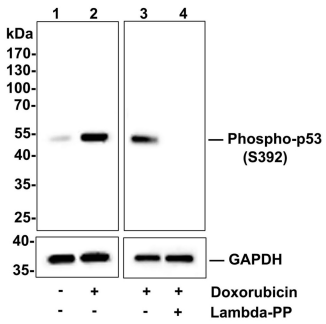

Fig3: Western blot analysis of Phospho-p53(S392) on A549 cell lysates.

Lane 1: A549 cells, whole cell lysate, 10ug/lane
Lane 2/3: A549 cells treated with 250nM Doxorubicin overnight, whole cell lysates, 10ug/lane
Lane 4: A549 cells treated with 250nM Doxorubicin overnight, then treated with 2.8ug/ul lambda-PP for 30 minutes, whole cell lysates, 10ug/lane

All lanes :
Anti-Phospho-p53(S392) antibody (ET1606-24) at 1:500 dilution. Anti-GAPDH antibody (ET1601-4) at 1:10,000 dilution. Goat Anti-Rabbit IgG H&L (HRP) (HA1001) at 1/200,000 dilution.

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

Blocking and diluting buffer: 5% BSA.

Exposure time: 5 minutes

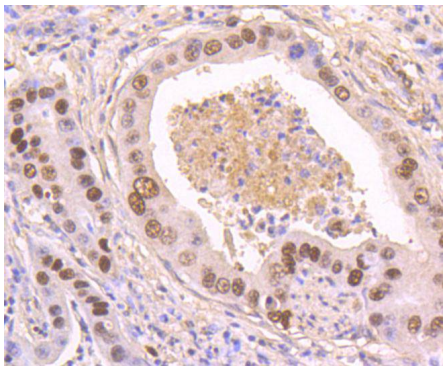

Fig4: Immunohistochemical analysis of paraffin-embedded human stomach carcinoma tissue using anti-Phospho-p53 (S392) antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1606-24, 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.

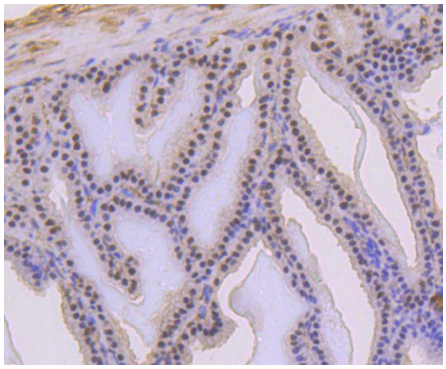

Fig5: Immunohistochemical analysis of paraffin-embedded mouse prostate tissue using anti-Phospho-p53 (S392) antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes.The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (ET1606-24, 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.

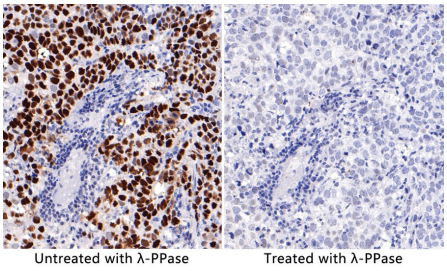

Fig6: Immunohistochemical analysis of paraffin-embedded Human gastric adenocarcinoma tissue untreaed and treated with λ-PPase with Rabbit anti-Phospho-p53 (S392) antibody (ET1606-24) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 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 (ET1606-24) 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.

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

Background References

1. Albert, TK. et al. 2016. The Establishment of a Hyperactive Structure Allows the Tumour Suppressor Protein p53 to Function through P-TEFb during Limited CDK9 Kinase Inhibition. PloS one. 11: e0146648.
2. Albert, TK. et al. 2014. Characterization of molecular and cellular functions of the cyclin-dependent kinase CDK9 using a novel specific inhibitor. British journal of pharmacology. 171: 55-68.