

Product datasheet

Anti-Mutant p53 antibody [Y5] ab32049

Recombinant RabMAb

★★★★★ 4 Abreviews 19 References 11 Images

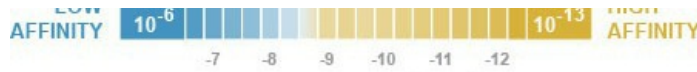
Overview

Product name	Anti-Mutant p53 antibody [Y5]
Description	Rabbit monoclonal [Y5] to Mutant p53
Host species	Rabbit
Specificity	ab32049 recognises human mutant forms of p53 but not human p53 wild type. ab32049 showed a negative signal on wildtype p53 cell lines (HepG2, A549, MCF-7) and a positive signal on mutant p53 cell lines (T47-D, Raji, A431) in WB.
Tested applications	Suitable for: ICC/IF, WB, IHC-P, Flow Cyt, IP
Species reactivity	Reacts with: Human
Immunogen	Synthetic peptide within Human Mutant p53 aa 1-100 (N terminal). The exact sequence is proprietary. Database link: P04637 (Peptide available as ab204287)
Positive control	A431 cell lysate and human skin cancer
General notes	A trial size is available to purchase for this antibody. Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information. Our RabMAb [®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents This product is a recombinant rabbit monoclonal antibody.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
Dissociation constant (K_D)	K _D = 2.02 x 10 ⁻¹⁰ M





[Learn more about K_D](#)

Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 49% PBS, 50% Glycerol, 0.05% BSA
Purity	IgG fraction
Clonality	Monoclonal
Clone number	Y5
Isotype	IgG

Applications

Our [Abpromise guarantee](#) covers the use of **ab32049** in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
ICC/IF		1/100.
WB	★★★★★	1/1000 - 1/5000. Detects a band of approximately 53 kDa (predicted molecular weight: 44 kDa).
IHC-P		1/50.
Flow Cyt		Use at an assay dependent concentration.
IP		1/20.

Target

Function

Acts as a tumor suppressor in many tumor types; induces growth arrest or apoptosis depending on the physiological circumstances and cell type. Involved in cell cycle regulation as a trans-activator that acts to negatively regulate cell division by controlling a set of genes required for this process. One of the activated genes is an inhibitor of cyclin-dependent kinases. Apoptosis induction seems to be mediated either by stimulation of BAX and FAS antigen expression, or by repression of Bcl-2 expression. In cooperation with mitochondrial PPIF is involved in activating oxidative stress-induced necrosis; the function is largely independent of transcription. Induces the transcription of long intergenic non-coding RNA p21 (lincRNA-p21) and lincRNA-Mkln1. LincRNA-p21 participates in TP53-dependent transcriptional repression leading to apoptosis and seem to have to effect on cell-cycle regulation. Implicated in Notch signaling cross-over. Prevents CDK7 kinase activity when associated to CAK complex in response to DNA damage, thus stopping cell cycle progression. Isoform 2 enhances the transactivation activity of isoform 1 from some but not all TP53-inducible promoters. Isoform 4 suppresses transactivation activity and impairs growth suppression mediated by isoform 1. Isoform 7 inhibits isoform 1-mediated apoptosis.

Tissue specificity

Ubiquitous. Isoforms are expressed in a wide range of normal tissues but in a tissue-dependent manner. Isoform 2 is expressed in most normal tissues but is not detected in brain, lung, prostate, muscle, fetal brain, spinal cord and fetal liver. Isoform 3 is expressed in most normal

tissues but is not detected in lung, spleen, testis, fetal brain, spinal cord and fetal liver. Isoform 7 is expressed in most normal tissues but is not detected in prostate, uterus, skeletal muscle and breast. Isoform 8 is detected only in colon, bone marrow, testis, fetal brain and intestine. Isoform 9 is expressed in most normal tissues but is not detected in brain, heart, lung, fetal liver, salivary gland, breast or intestine.

Involvement in disease

TP53 is found in increased amounts in a wide variety of transformed cells. TP53 is frequently mutated or inactivated in about 60% of cancers. TP53 defects are found in Barrett metaplasia a condition in which the normally stratified squamous epithelium of the lower esophagus is replaced by a metaplastic columnar epithelium. The condition develops as a complication in approximately 10% of patients with chronic gastroesophageal reflux disease and predisposes to the development of esophageal adenocarcinoma.

Esophageal cancer

Li-Fraumeni syndrome

Squamous cell carcinoma of the head and neck

Lung cancer

Choroid plexus papilloma

Adrenocortical carcinoma

Basal cell carcinoma 7

Sequence similarities

Belongs to the p53 family.

Domain

The nuclear export signal acts as a transcriptional repression domain. The TAD1 and TADII motifs (residues 17 to 25 and 48 to 56) correspond both to 9aaTAD motifs which are transactivation domains present in a large number of yeast and animal transcription factors.

Post-translational modifications

Acetylated. Acetylation of Lys-382 by CREBBP enhances transcriptional activity. Deacetylation of Lys-382 by SIRT1 impairs its ability to induce proapoptotic program and modulate cell senescence.

Phosphorylation on Ser residues mediates transcriptional activation. Phosphorylated by HIPK1 (By similarity). Phosphorylation at Ser-9 by HIPK4 increases repression activity on BIRC5 promoter. Phosphorylated on Thr-18 by VRK1. Phosphorylated on Ser-20 by CHEK2 in response to DNA damage, which prevents ubiquitination by MDM2. Phosphorylated on Ser-20 by PLK3 in response to reactive oxygen species (ROS), promoting p53/TP53-mediated apoptosis. Phosphorylated on Thr-55 by TAF1, which promotes MDM2-mediated degradation. Phosphorylated on Ser-33 by CDK7 in a CAK complex in response to DNA damage. Phosphorylated on Ser-46 by HIPK2 upon UV irradiation. Phosphorylation on Ser-46 is required for acetylation by CREBBP. Phosphorylated on Ser-392 following UV but not gamma irradiation. Phosphorylated on Ser-15 upon ultraviolet irradiation; which is enhanced by interaction with BANP. Phosphorylated by NUA1 at Ser-15 and Ser-392; was initially thought to be mediated by STK11/LKB1 but it was later shown that it is indirect and that STK11/LKB1-dependent phosphorylation is probably mediated by downstream NUA1 (PubMed:21317932). It is unclear whether AMP directly mediates phosphorylation at Ser-15. Phosphorylated on Thr-18 by isoform 1 and isoform 2 of VRK2. Phosphorylation on Thr-18 by isoform 2 of VRK2 results in a reduction in ubiquitination by MDM2 and an increase in acetylation by EP300. Stabilized by CDK5-mediated phosphorylation in response to genotoxic and oxidative stresses at Ser-15, Ser-33 and Ser-46, leading to accumulation of p53/TP53, particularly in the nucleus, thus inducing the transactivation of p53/TP53 target genes. Phosphorylated by DYRK2 at Ser-46 in response to genotoxic stress. Phosphorylated at Ser-315 and Ser-392 by CDK2 in response to DNA-damage.

Dephosphorylated by PP2A-PPP2R5C holoenzyme at Thr-55. SV40 small T antigen inhibits the dephosphorylation by the AC form of PP2A.

May be O-glycosylated in the C-terminal basic region. Studied in EB-1 cell line.

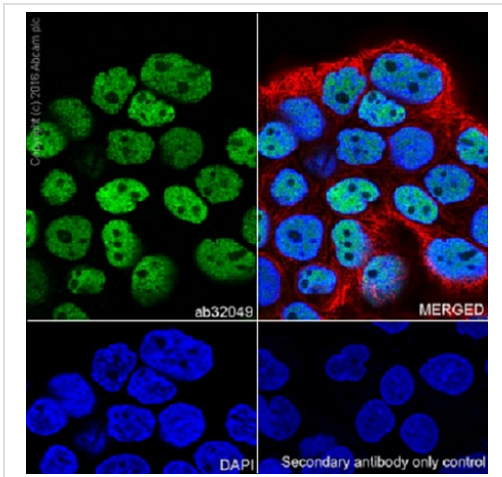
Ubiquitinated by MDM2 and SYVN1, which leads to proteasomal degradation. Ubiquitinated by RFW3, which works in cooperation with MDM2 and may catalyze the formation of short

polyubiquitin chains on p53/TP53 that are not targeted to the proteasome. Ubiquitinated by MKRN1 at Lys-291 and Lys-292, which leads to proteasomal degradation. Deubiquitinated by USP10, leading to its stabilization. Ubiquitinated by TRIM24, which leads to proteasomal degradation. Ubiquitination by TOPORS induces degradation. Deubiquitination by USP7, leading to stabilization. Isoform 4 is monoubiquitinated in an MDM2-independent manner. Monomethylated at Lys-372 by SETD7, leading to stabilization and increased transcriptional activation. Monomethylated at Lys-370 by SMYD2, leading to decreased DNA-binding activity and subsequent transcriptional regulation activity. Lys-372 monomethylation prevents interaction with SMYD2 and subsequent monomethylation at Lys-370. Dimethylated at Lys-373 by EHMT1 and EHMT2. Monomethylated at Lys-382 by SETD8, promoting interaction with L3MBTL1 and leading to repress transcriptional activity. Dimethylation at Lys-370 and Lys-382 diminishes p53 ubiquitination, through stabilizing association with the methyl reader PHF20. Demethylation of dimethylated Lys-370 by KDM1A prevents interaction with TP53BP1 and represses TP53-mediated transcriptional activation. Sumoylated with SUMO1. Sumoylated at Lys-386 by UBC9.

Cellular localization

Cytoplasm; Nucleus. Cytoplasm. Localized in both nucleus and cytoplasm in most cells. In some cells, forms foci in the nucleus that are different from nucleoli; Nucleus. Cytoplasm. Localized in the nucleus in most cells but found in the cytoplasm in some cells; Nucleus. Cytoplasm. Localized mainly in the nucleus with minor staining in the cytoplasm; Nucleus. Cytoplasm. Predominantly nuclear but localizes to the cytoplasm when expressed with isoform 4; Nucleus. Cytoplasm. Predominantly nuclear but translocates to the cytoplasm following cell stress and Cytoplasm. Nucleus. Nucleus > PML body. Endoplasmic reticulum. Mitochondrion matrix. Interaction with BANP promotes nuclear localization. Recruited into PML bodies together with CHEK2. Translocates to mitochondria upon oxidative stress.

Images

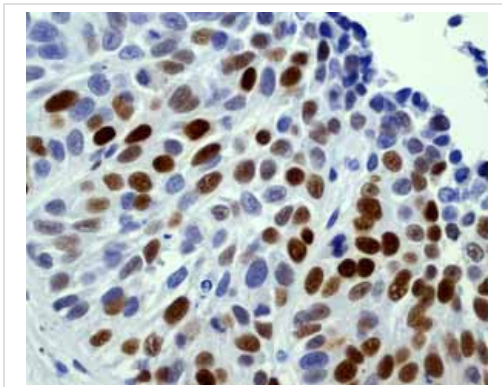


Immunocytochemistry/ Immunofluorescence - Anti-Mutant p53 antibody [Y5] (ab32049)

Confocal image showing nuclear staining on A431 cell line.

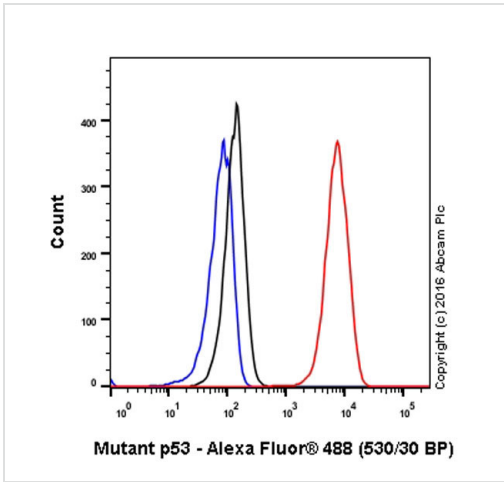
Immunocytochemistry/Immunofluorescence analysis of A431(Human epidermoid carcinoma cell line) labelling Mutant p53 with ab32049 at 1/100. Cells were fixed with 4% Paraformaldehyde (20 minutes) and permeabilized with 0.1% Triton X-100 (5 minutes). ab150077, Alexa Fluor[®] 488-conjugated goat anti-rabbit IgG (1/1000) was used as the secondary antibody. Cells were counterstained with ab195889, anti- alpha tubulin antibody (1/200) using an Alexa Fluor[®] 594-conjugated microtubule marker as the secondary. Nuclei were counterstained with DAPI (blue).

Secondary Only Control: PBS was used instead of the primary antibody as the negative control.



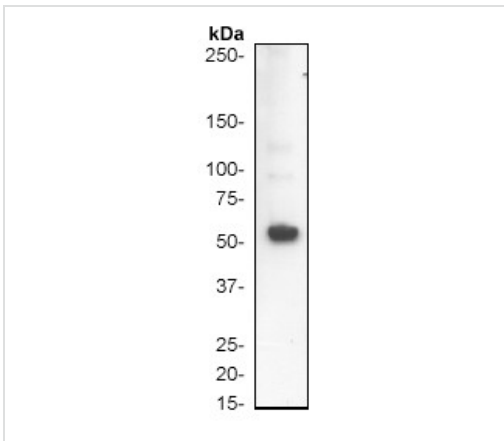
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Mutant p53 antibody [Y5] (ab32049)

ab32049 showing positive staining in Urinary bladder carcinoma tissue.



Flow Cytometry - Anti-Mutant p53 antibody [Y5] (ab32049)

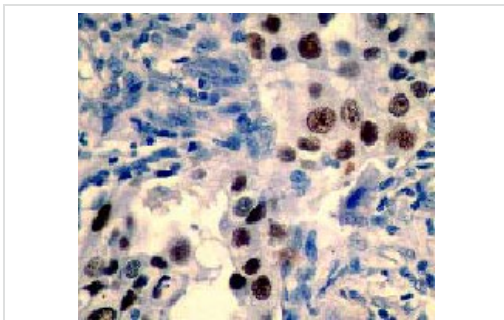
Flow Cytometry analysis of A431 (human epidermoid carcinoma) cells labeling Mutant p53 with unpurified ab32049 at 1/20 dilution (10ug/ml) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit IgG (Alexa Fluor® 488) (1/2000) was used as the secondary antibody. Rabbit monoclonal IgG (Black) was used as the isotype control, cells without incubation with primary antibody and secondary antibody (Blue) was used as the unlabeled control.



Western blot - Anti-Mutant p53 antibody [Y5] (ab32049)

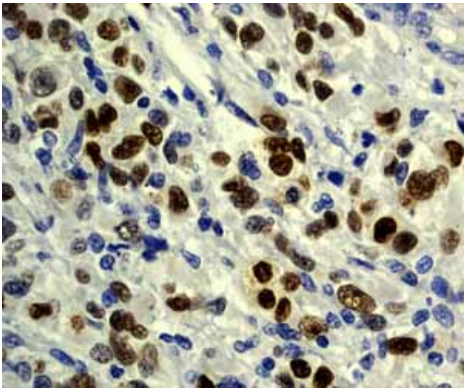
Anti-Mutant p53 antibody [Y5] (ab32049) at 1/500 dilution + A431 cell lysate

Predicted band size: 44 kDa
Observed band size: 50 kDa



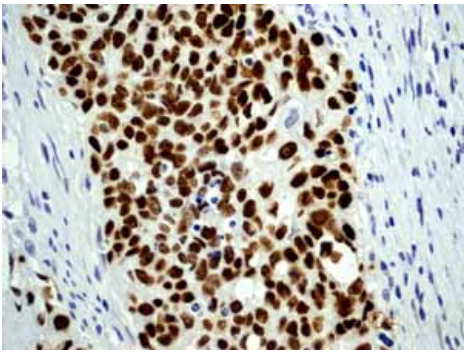
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Mutant p53 antibody [Y5] (ab32049)

Immunohistochemistry (Paraffin-embedded sections) using ab32049 at a dilution of 1/50 and human skin cancer



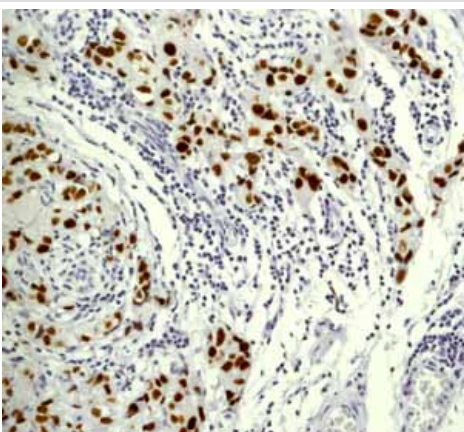
ab32049 showing positive staining in Glioma tissue.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Mutant p53 antibody [Y5] (ab32049)



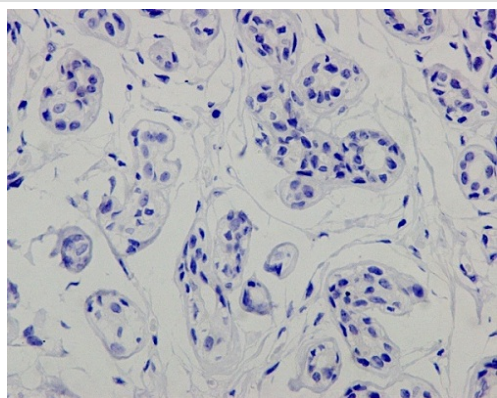
ab32049 showing positive staining in Gastric adenocarcinoma tissue.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Mutant p53 antibody [Y5] (ab32049)



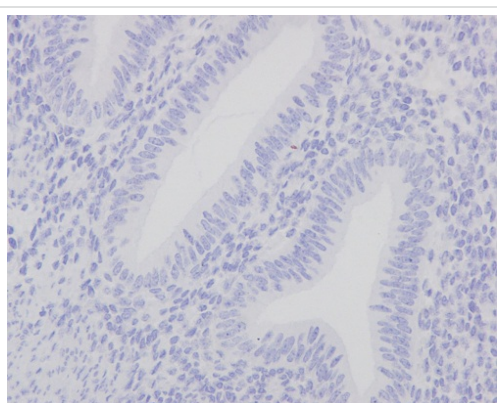
ab32049 showing positive staining in Breast carcinoma tissue.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Mutant p53 antibody [Y5] (ab32049)



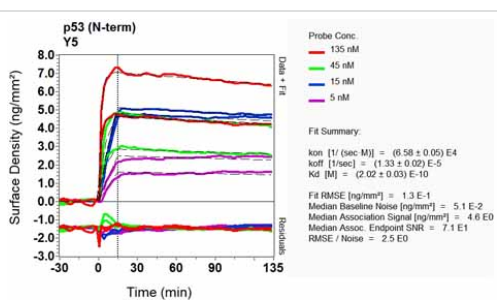
Immunohistochemical analysis of paraffin embedded normal Human breast tissue (negative control) labeling p53 with ab32049.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Mutant p53 antibody [Y5] (ab32049)



Immunohistochemical analysis of paraffin embedded normal Human uterus tissue (negative control) labeling p53 with ab32049.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Mutant p53 antibody [Y5] (ab32049)



Equilibrium disassociation constant (K_D)

Learn more about K_D

[Click here to learn more about \$K_D\$](#)

Other - Anti-Mutant p53 antibody [Y5] (ab32049)

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