abcam

Product datasheet

Anti-p53 antibody [G59-12] ab308609





8 图像

概述

产**品名称** Anti-p53抗体[G59-12]

宿主 Mouse

经测试应用 适用于: WB, IHC-P, Flow Cyt (Intra), IP

种属反应性 与反应: Human

不与反应: Mouse, Rat

免疫原 Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

阳性对照 WB: Wild-type HAP1 and HEK-293 whole cell lysate. IHC-P: Human endometrial cancer tissue.

Wild-type HAP1 cell pellet. Flow Cyt (Intra): Parental HAP1 cells. IP: HEK-293 whole cell lysate.

常规说明 This antibody clone is manufactured by Abcam. If you require a custom buffer formulation or

conjugation for your experiments, please contact **orders@abcam.com**.

This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility

- Improved sensitivity and specificity

- Long-term security of supply

- Animal-free production

For more information see here.

性能

形式 Liquid

存放说明 Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

存储溶液 pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 40% Glycerol (glycerin, glycerine), 0.05% BSA, 59% PBS

纯**度** Protein A purified

 克隆
 单克隆

 克隆编号
 G59-12

1

同种型 lgG1

应用

The Abpromise guarantee

Abpromise™承诺保证使用ab308609于以下的经测试应用

"应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB		1/1000. Predicted molecular weight: 43 kDa.
IHC-P		1/100.
Flow Cyt (Intra)		1/1000.
IP		1/30.

靶标

功能

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 transactivator 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. Implicated in Notch signaling cross-over. 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.

组织特异性

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.

疾病相关

Note=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.

Defects in TP53 are a cause of esophageal cancer (ESCR) [MIM:133239].

Defects in TP53 are a cause of Li-Fraumeni syndrome (LFS) [MIM:151623]. LFS is an autosomal dominant familial cancer syndrome that in its classic form is defined by the existence of a proband affected by a sarcoma before 45 years with a first degree relative affected by any tumor before 45 years and another first degree relative with any tumor before 45 years or a sarcoma at any age. Other clinical definitions for LFS have been proposed (PubMed:8118819 and PubMed:8718514) and called Li-Fraumeni like syndrome (LFL). In these families affected relatives develop a diverse

set of malignancies at unusually early ages. Four types of cancers account for 80% of tumors occurring in TP53 germline mutation carriers: breast cancers, soft tissue and bone sarcomas, brain tumors (astrocytomas) and adrenocortical carcinomas. Less frequent tumors include choroid plexus carcinoma or papilloma before the age of 15, rhabdomyosarcoma before the age of 5, leukemia, Wilms tumor, malignant phyllodes tumor, colorectal and gastric cancers. Defects in TP53 are involved in head and neck squamous cell carcinomas (HNSCC) [MIM:275355]; also known as squamous cell carcinoma of the head and neck. Defects in TP53 are a cause of lung cancer (LNCR) [MIM:211980].

Defects in TP53 are a cause of choroid plexus papilloma (CPLPA) [MIM:260500]. Choroid plexus papilloma is a slow-growing benign tumor of the choroid plexus that often invades the leptomeninges. In children it is usually in a lateral ventricle but in adults it is more often in the fourth ventricle. Hydrocephalus is common, either from obstruction or from tumor secretion of cerebrospinal fluid. If it undergoes malignant transformation it is called a choroid plexus carcinoma. Primary choroid plexus tumors are rare and usually occur in early childhood. Defects in TP53 are a cause of adrenocortical carcinoma (ADCC) [MIM:202300]. ADCC is a rare childhood tumor of the adrenal cortex. It occurs with increased frequency in patients with the Beckwith-Wiedemann syndrome and is a component tumor in Li-Fraumeni syndrome.

Belongs to the p53 family.

The nuclear export signal acts as a transcriptional repression domain. The TADI 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.

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 Thr-55 by TAF1, which promotes MDM2-mediated degradation. 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 upon DNA damage, probably by ATM or ATR. Phosphorylated on Ser-15 upon ultraviolet irradiation; which is enhanced by interaction with BANP.

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 RFWD3, 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. Demethylation of dimethylated Lys-370 by KDM1A prevents interaction with TP53BP1 and represses TP53-mediated transcriptional activation.

序列相似性 结构域

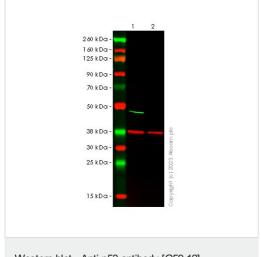
翻译后修饰

细胞定位

Sumoylated by SUMO1.

Cytoplasm; Cytoplasm. Nucleus. Nucleus > PML body. Endoplasmic reticulum. Interaction with BANP promotes nuclear localization. Recruited into PML bodies together with CHEK2; 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 and Nucleus. Cytoplasm. Predominantly nuclear but translocates to the cytoplasm following cell stress.

图片



Western blot - Anti-p53 antibody [G59-12] (ab308609)

All lanes : Anti-p53 antibody [G59-12] (ab308609) at 1/1000 dilution

Lane 1 : Wild-type HAP1 whole cell lysate 20 μg **Lane 2 :** p53 knockout HAP1 whole cell lysate 20 μg

Secondary

All lanes : Goat anti-Mouse IgG H&L (IRDye® 800CW) preadsorbed (<u>ab216772</u>) at 1/20000 dilution

Predicted band size: 43 kDa **Observed band size:** 53 kDa

Blocking and diluting buffer and concentration: Intercept® (TBS) Blocking Buffer diluted with an equal volume of TBS.

Lysates at 20 µg per lane.

The samples were run on a Bis-Tris gel under reducing conditions.

Western blot: Anti-p53 antibody (ab308609) staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody (ab181602) loading control staining at 1/2000 dilution, shown in red.

In Western blot, ab308609 was shown to bind specifically to p53. Target of interest was observed at 53 kDa in wild-type HAP1 cell lysates (lane 1) with no signal observed at this size in p53 knockout cell lysates (lane 2). To generate this image, samples were first run on an SDS-PAGE gel then transferred onto an immobilon-FL PVDF membrane. Membranes were blocked in a fluorescent western blot (TBS-based) blocking solution before incubation with primary antibodies overnight at 4°C. Blots were washed in TBS-T, incubated with secondary antibodies Goat Anti-Mouse IgG H&L

(IRDye® 800CW) and Goat Anti-Rabbit lgG H&L (IRDye® 680RD) (ab216777) at 1/20000 dilution for 1 h at room temperature, washed again then imaged.

1 2

250 kDa —
150 kDa —
100 kDa —
75 kDa —
37 kDa —
37 kDa —
25 kDa —
20 kDa —
15 kDa —
10 kDa —

15 kDa —

10 kDa —

1 to kDa —

Vinculin
ab129002

Western blot - Anti-p53 antibody [G59-12] (ab308609)

All lanes : Anti-p53 antibody [G59-12] (ab308609) at 1/1000 dilution

Lane 1 : HEK-293 (human embryonic kidney epithelial cell) whole cell lysate 20 μg

Lane 2 : Saos-2 (human osteosarcoma epithelial cell) whole cell lysate 20 µg

Secondary

All lanes : Peroxidase-Conjugated Goat anti-Mouse IgG (H+L) at 1/10000 dilution

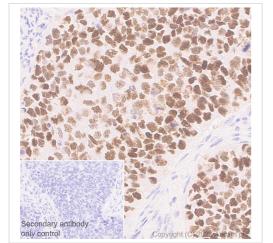
Predicted band size: 43 kDa **Observed band size:** 53 kDa

Blocking and diluting buffer and concentration: 5% NFDM/TBST.

Negative control: Saos-2 (PMID: 17202229; PMID: 9786925).

In Western blot, anti- Vinculin antibody (<u>ab129002</u>) loading control staining at 1/10000 dilution.

Exposure time: 180 seconds.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-p53 antibody [G59-12] (ab308609)

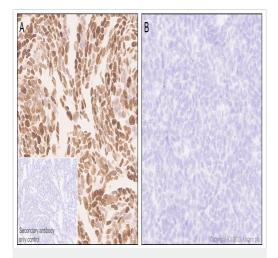
Immunohistochemical analysis of paraffin-embedded human endometrial cancer tissue labeling p53 with ab308609 at 1/100 (10.28 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Mainly nuclear staining on human endometrial cancer.

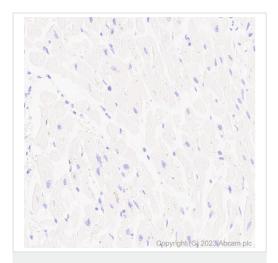
The section was incubated with ab308609 for 30 mins at room temperature, followed by anti-mouse IgG1 antibody (ab125913) for 8 mins during the LeicaDS9800 kit staining procedure. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-p53 antibody [G59-12] (ab308609)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-p53 antibody [G59-12] (ab308609)

Immunohistochemical analysis of paraffin-embedded cells labeling p53 with ab308609 at 1/100 (10.28 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Mainly nuclear staining on (A) wild-type HAP1 cell pellet, no staining on (B) p53 knockout HAP1 cell pellet.

The section was incubated with ab308609 for 30 mins at room temperature, followed by anti-mouse IgG1 antibody (ab125913) for 8 mins during the LeicaDS9800 kit staining procedure. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

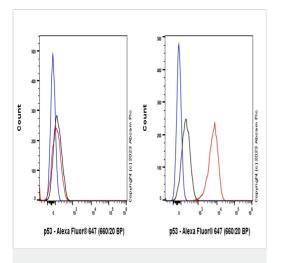
Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.

Immunohistochemical analysis of paraffin-embedded human cardiac muscle tissue labeling p53 with ab308609 at 1/100 (10.28 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection).

Negative control: No staining on human cardiac muscle.

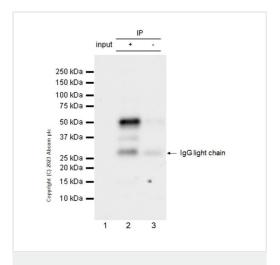
The section was incubated with ab308609 for 30 mins at room temperature, followed by anti-mouse IgG1 antibody (ab125913) for 8 mins during the LeicaDS9800 kit staining procedure. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.



Flow Cytometry (Intracellular) - Anti-p53 antibody [G59-12] (ab308609)

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized p53 KO HAP1 (human p53 knockout chronic myelogenous leukemia near-haploid cell, Left) / Parental HAP1 (Right) cells labelling p53 with ab308609 at 1/1000 dilution (0.1 ug) (Red) compared with a Mouse monoclonal lgG (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat Anti-Mouse lgG (Alexa Fluor® 647, ab150119) at 1/5000 dilution was used as the secondary antibody.



Immunoprecipitation - Anti-p53 antibody [G59-12] (ab308609)

p53 was immunoprecipitated from 0.35 mg HEK-293 (human embryonic kidney epithelial cell) whole cell lysate with ab308609 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab308609 at 1/1000 dilution. mouse IgG for IP (HRP) (ab131368) was used at 1/5000 dilution.

Lane 1: HEK-293 whole cell lysate

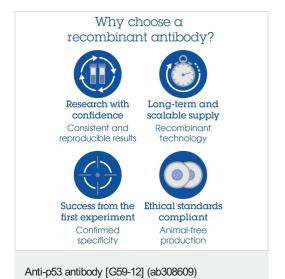
Lane 2: ab308609 IP in HEK-293 whole cell lysate

Lane 3: Mouse IgG1 monoclonal isotype control (ab18443) instead of ab308609 in HEK-293 whole cell lysate

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 23 seconds.

The binding pattern observed is consistent with what has been described in the literature (PMID: 31045216; PMID: 16131611).



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