abcam

Product datasheet

Anti-MDM2 (phospho S166) antibody [EPR1450(2)] ab170880

Recombinant RabMAb

11 References 6 Images

Overview

Product name Anti-MDM2 (phospho S166) antibody [EPR1450(2)]

Description Rabbit monoclonal [EPR1450(2)] to MDM2 (phospho S166)

Host species Rabbit

Tested applications Suitable for: WB, IHC-P, Dot blot

Unsuitable for: Flow Cyt,ICC/IF or IP

Species reactivity Reacts with: Human

Predicted to work with: Rat
Does not react with: Mouse

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control MCF-7 cell lysates treated with IGF-1, Human gastric carcinoma tissue and Human placenta

tissue

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

- High batch-to-batch consistency and reproducibility

Improved sensitivity and specificityLong-term security of supplyAnimal-free production

For more information **see here**.

Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**[®] **patents**.

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 long

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: 9% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA, 50% Tissue culture

supernatant

Purity Tissue culture supernatant

Clonality Monoclonal
Clone number EPR1450(2)

Isotype IgG

Applications

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab170880 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
WB		1/50000 - 1/200000. Predicted molecular weight: 55 kDa.
IHC-P		1/50 - 1/100. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
Dot blot		1/1000.

Application notes

Is unsuitable for Flow Cyt,ICC/IF or IP.

Target

Function

E3 ubiquitin-protein ligase that mediates ubiquitination of p53/TP53, leading to its degradation by the proteasome. Inhibits p53/TP53- and p73/TP73-mediated cell cycle arrest and apoptosis by binding its transcriptional activation domain. Also acts as an ubiquitin ligase E3 toward itself and ARRB1. Permits the nuclear export of p53/TP53. Promotes proteasome-dependent ubiquitin-independent degradation of retinoblastoma RB1 protein. Inhibits DAXX-mediated apoptosis by inducing its ubiquitination and degradation. Component of the TRIM28/KAP1-MDM2-p53/TP53 complex involved in stabilizing p53/TP53. Also component of the TRIM28/KAP1-ERBB4-MDM2 complex which links growth factor and DNA damage response pathways.

Tissue specificity

Ubiquitous. Isoform Mdm2-A, isoform Mdm2-B, isoform Mdm2-C, isoform Mdm2-D, isoform Mdm2-E, isoform Mdm2-F and isoform Mdm2-G are observed in a range of cancers but absent in normal tissues.

Involvement in disease

Note=Seems to be amplified in certain tumors (including soft tissue sarcomas, osteosarcomas and gliomas). A higher frequency of splice variants lacking p53 binding domain sequences was found in late-stage and high-grade ovarian and bladder carcinomas. Four of the splice variants show loss of p53 binding.

Sequence similarities

Belongs to the MDM2/MDM4 family.
Contains 1 RanBP2-type zinc finger.
Contains 1 RING-type zinc finger.
Contains 1 SWIB domain.

Domain

Region I is sufficient for binding p53 and inhibiting its G1 arrest and apoptosis functions. It also binds p73 and E2F1. Region II contains most of a central acidic region required for interaction with ribosomal protein L5 and a putative C4-type zinc finger. The RING finger domain which coordinates two molecules of zinc interacts specifically with RNA whether or not zinc is present and mediates the heterooligomerization with MDM4. It is also essential for its ubiquitin ligase E3 activity toward p53 and itself.

Post-translational modifications

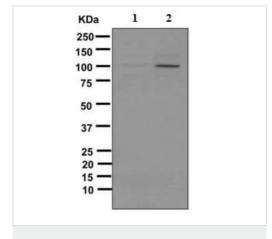
Phosphorylated in response to ionizing radiation in an ATM-dependent manner.

Auto-ubiquitinated; which leads to proteasomal degradation. Deubiquitinated by USP2 leads to its accumulation and increases deubiquitinilation and degradation of p53/TP53. Deubiquitinated by USP7; leading to stabilize it.

Cellular localization

Nucleus > nucleoplasm. Cytoplasm. Nucleus > nucleolus. Expressed predominantly in the nucleoplasm. Interaction with ARF(P14) results in the localization of both proteins to the nucleolus. The nucleolar localization signals in both ARF(P14) and MDM2 may be necessary to allow efficient nucleolar localization of both proteins. Colocalizes with RASSF1 isoform A in the nucleus.

Images



Western blot - Anti-MDM2 (phospho S166) antibody [EPR1450(2)] (ab170880) **All lanes :** Anti-MDM2 (phospho S166) antibody [EPR1450(2)] (ab170880) at 1/50000 dilution

Lane 1: MCF7 cell lysate

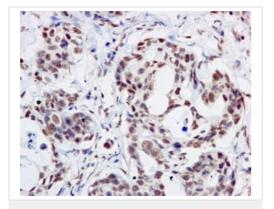
Lane 2: MCF7 cell lysate treated with IGF-1

Lysates/proteins at 10 µg per lane.

Secondary

All lanes: HRP labelled goat anti-rabbit lgG at 1/2000 dilution

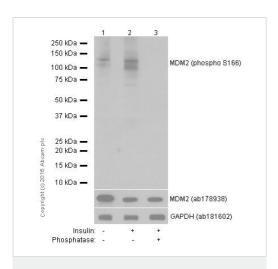
Predicted band size: 55 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MDM2 (phospho S166) antibody [EPR1450(2)] (ab170880)

Immunohistochemical analysis of paraffin-embedded human gastric carcinoma tissue labeling MDM2 (phospho S166) using ab170880 at a 1/50 dilution.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Western blot - Anti-MDM2 (phospho S166) antibody [EPR1450(2)] (ab170880)

All lanes : Anti-MDM2 (phospho S166) antibody [EPR1450(2)] (ab170880) at 1/100000 dilution

Lane 1: A549 (Human lung carcinoma epithelial cell) whole cell lysate

Lane 2: A549 (Human lung carcinoma epithelial cell) treated with insulin at 1ug/ml for 150 minutes. Whole cell lysate

Lane 3: A549 (Human lung carcinoma epithelial cell) treated with insulin at 1ug/ml for 150 minutes. Whole cell lysate. Then the membrane was incubated with phosphatase

Lysates/proteins at 15 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

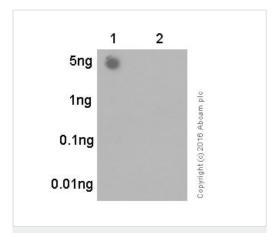
Predicted band size: 55 kDa **Observed band size:** 90-140 kDa

Exposure time: 5 seconds

Diluting and blocking buffer: 2% BSA/TBST

The molecular weight is the same with the one from this paper

PMID: 25392082

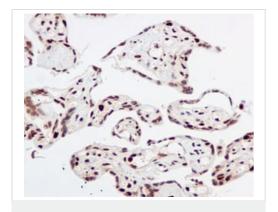


Dot Blot - Anti-MDM2 (phospho S166) antibody [EPR1450(2)] (ab170880)

Dot blot analysis of MDM2 (phospho S166) phospho peptide (Lane 1), MDM2 non-phospho peptide (Lane 2), labeling MDM2 (phospho S166) with ab170880 at a dilution of 1/1000. Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated (ab97051) was used as the secondary antibody at a dilution of 1/100000.

Blocking and dilution buffer: 5% NFDM/TBST.

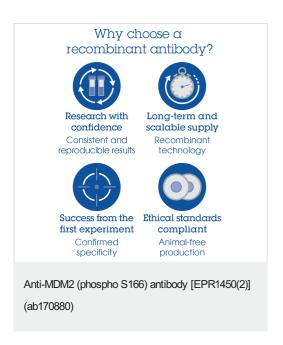
Exposure time: 3 minutes.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MDM2 (phospho S166) antibody [EPR1450(2)] (ab170880)

Immunohistochemical analysis of paraffin-embedded human placenta tissue labeling MDM2 (phospho S166) using ab170880 at a 1/50 dilution.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



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