

## Anti-MDM2 pS185 (RABBIT) Antibody

Mdm2 phospho S185 Antibody Catalog # ASR5220

## Specification

## Anti-MDM2 pS185 (RABBIT) Antibody - Product Information

Host Conjugate Target Species Reactivity Clonality Application Application Note	Rabbit Unconjugated Mouse Human, Mouse Polyclonal WB, E, IP, I, LCI This affinity purified antibody has been tested for use in ELISA and by western blot. Specific conditions for reactivity should be optimized by the end user. Expect bands approximately 102 kDa in size corresponding to phosphorylated MDM2 protein by western blotting in the appropriate cell lysate or extract.
Physical State	Liquid (sterile filtered)
Buffer	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Immunogen	This affinity purified antibody was prepared from whole rabbit serum produced by repeated immunizations with a synthetic peptide corresponding to an internal region near aa 175-200 of mouse MDM2.
Preservative	0.01% (w/v) Sodium Azide

# Anti-MDM2 pS185 (RABBIT) Antibody - Additional Information

Gene ID 17246

Other Names 17246

#### **Purity**

This affinity-purified antibody is directed against the phosphorylated form of mouse MDM2 protein at the pS185 residue. The product was affinity purified from monospecific antiserum by immunoaffinity purification. Antiserum was first purified against the phosphorylated form of the immunizing peptide. The resultant affinity purified antibody was then cross-adsorbed against the non-phosphorylated form of the immunizing peptide. Reactivity occurs against Mouse MDM2 pS185 protein and the antibody is specific for the phosphorylated form of the protein. Reactivity with non-phosphorylated mouse MDM2 is minimal by ELISA and western blot. A BLAST analysis was used to suggest minimal cross reactivity with MDM2 homologues from other sources.

#### Storage Condition

Store vial at -20° C prior to opening. Aliquot contents and freeze at -20° C or below for extended



storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use.

### **Precautions Note**

This product is for research use only and is not intended for therapeutic or diagnostic applications.

## Anti-MDM2 pS185 (RABBIT) Antibody - Protein Information

## Name Mdm2

### Function

E3 ubiquitin-protein ligase that mediates ubiquitination of p53/TP53, leading to its degradation by the proteasome (PubMed:<a href="http://www.uniprot.org/citations/15195100" target="\_blank">15195100</a>, PubMed:<a href="http://www.uniprot.org/citations/21804542" target="\_blank">21804542</a>). Inhibits p53/TP53- and p73/TP73- mediated cell cycle arrest and apoptosis by binding its transcriptional activation domain (By similarity). Also acts as a ubiquitin ligase E3 toward itself, ARRB1 and ARBB2 (PubMed:<a href="http://www.uniprot.org/citations/11588219" target=" blank">11588219</a>). Permits the

nuclear export of p53/TP53 (By similarity). Promotes proteasome-dependent ubiquitin-independent degradation of retinoblastoma RB1 protein (By similarity). Inhibits DAXX-mediated apoptosis by inducing its ubiquitination and degradation (By similarity). Component of the TRIM28/KAP1-MDM2-p53/TP53 complex involved in stabilizing p53/TP53 (By similarity). Also a component of the TRIM28/KAP1-ERBB4-MDM2 complex which links growth factor and DNA damage response pathways (By similarity). Mediates ubiquitination and subsequent proteasome degradation of DYRK2 in nucleus (By similarity). Ubiquitinates IGF1R and SNAI1 and promotes them to proteasomal degradation (By similarity). Ubiquitinates DCX, leading to DCX degradation and reduction of the dendritic spine density of olfactory bulb granule cells (PubMed:<a href="http://www.uniprot.org/citations/25088421" target=" blank">25088421</a>). Ubiquitinates DLG4, leading to proteasomal degradation of DLG4 which is required for AMPA receptor endocytosis (PubMed: <a href="http://www.uniprot.org/citations/14642282" target=" blank">14642282</a>). Negatively regulates NDUFS1, leading to decreased mitochondrial respiration, marked oxidative stress, and commitment to the mitochondrial pathway of apoptosis (PubMed:<a href="http://www.uniprot.org/citations/30879903" target=" blank">30879903</a>). Binds NDUFS1 leading to its cytosolic retention rather than mitochondrial localization resulting in decreased supercomplex assembly (interactions between complex I and complex III), decreased complex I activity, ROS production, and apoptosis (PubMed:<a href="http://www.uniprot.org/citations/30879903" target=" blank">30879903</a>).

#### **Cellular Location**

Nucleus, nucleoplasm. Cytoplasm. Nucleus, nucleolus. Nucleus {ECO:0000250|UniProtKB:Q00987} Note=Colocalizes with RASSF1 isoform A in the nucleus (By similarity) 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 {ECO:0000250|UniProtKB:Q00987}

#### **Tissue Location**

Ubiquitously expressed at low-level throughout embryo development and in adult tissues. MDM2-p90 is much more abundant than MDM2-p76 in testis, brain, heart, and kidney, but in the thymus, spleen, and intestine, the levels of the MDM2 proteins are roughly equivalent.

## Anti-MDM2 pS185 (RABBIT) Antibody - Protocols



Provided below are standard protocols that you may find useful for product applications.

- <u>Western Blot</u>
- <u>Blocking Peptides</u>
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- <u>Cell Culture</u>

# Anti-MDM2 pS185 (RABBIT) Antibody - Images



Affinity Purified Anti-MDM2 pS185 (Rabbit) is shown to detect a 102 kDa band (arrow) corresponding to phosphorylated mouse MDM2 present in a 293T whole cell lysate. Cells were serum-starved for 24 hours prior to harvest. Approximately 20  $\mu$ g of lysate was loaded per lane for SDS-PAGE. Untreated cells are shown in lane 1, whereas cells in lane 2 were treated with IGF-1 (100 ng/ml) for 20 min prior to harvest. Follow reaction of antibody with a 1:2000 dilution of HRP Goat-a-Rabbit IgG for visualization.

# Anti-MDM2 pS185 (RABBIT) Antibody - Background

MDM2 is a nuclear phosphoprotein with an apparent molecular mass of 90 kD that forms a complex with the p53 tumor suppressor protein. Human MDM2 was identified as a homologous product of the 'murine double minute 2' gene (mdm2). The MDM2 gene enhances the tumorigenic potential of cells when it is overexpressed and encodes a putative transcription factor. Forming a tight complex with the p53 gene, the MDM2 oncogene can inhibit p53-mediated transactivation. MDM2 binds to p53 and amplification of MDM2 in sarcomas leads to escape from p53-regulated growth control. This mechanism of tumorigenesis parallels that for virus-induced tumors in which viral oncogene products bind to and functionally inactivate p53. Overexpression of the MDM2 oncogene was found in leukemias. Inactivation of tumor suppressor genes leads to deregulated cell proliferation and is a key factor in human tumorigenesis. MDM2 interacts physically and functionally with the retinoblastoma (RB) protein and can inhibit its growth regulatory capacity. Both RB and p53 can be subjected to negative regulation by the product of a single cellular protooncogene. The interference of binding to p53 prevents the interaction of MDM2 and its regulation of the transcriptional activity of p53 in vivo. Direct association of p53 with the cellular protein MDM2 results in ubiquitination and subsequent degradation of p53. MDM2-p53 complexes were preferentially found in S/G2M phases of the cell cycle. The MDM2 gene is alternatively spliced, producing 5 additional splice variant transcripts from the full length MDM2 gene. Four out of five of these alternatively spliced forms (MDM2a-MDMd) are missing substantial portions of the p53 binding domain and retain the acidic domain and the zinc-finger domains. The fifth and smallest transcript (MDM2e) retains the largest spliced region encoding the p53 binding domain; however, it lacks the nuclear localization signal,



the acidic domain and zinc-finger domains. The alternatively spliced transcripts tend to be expressed in tumorigenic tissue, whereas the full-length MDM2 transcript is expressed in normal tissue. MDM2 is found in the nucleus and cytoplasm, however, it is expressed predominantly in the nucleoplasm. Interaction with ARF (P14) results in the localization of both proteins to the nucleus. The nucleolar localization signals in both ARF and MDM2 may be necessary to allow efficient nucleolar localization of both proteins.