

Product Datasheet

MDM2/HDM2 Antibody (SMP14) - BSA Free NB100-2736

Unit Size: 0.1 ml

Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.

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NB100-2736

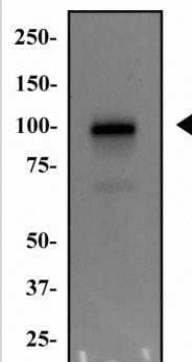
MDM2/HDM2 Antibody (SMP14) - BSA Free

Product Information	
Unit Size	0.1 ml
Concentration	1 mg/ml
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Monoclonal
Clone	SMP14
Preservative	0.02% Sodium Azide
Isotype	IgG1 Kappa
Purity	Protein G purified
Buffer	PBS
Product Description	
Host	Mouse
Gene ID	4193
Gene Symbol	MDM2
Species	Human, Mouse, Rat
Reactivity Notes	Rat reactivity reported in scientific literature (PMID: 15625077). Mouse reactivity reported in scientific literature (PMID: 25981963).
Specificity/Sensitivity	The SMP14 monoclonal antibody also recognises a peptide epitope around Thr-216 of murine MDM2 (when Thr-216 is unphosphorylated) see Zhang & Prives, 2001 (PMID: 11359766) for further details. SMP14 also cross reacts with some cytokeratins (6, 14 & 16). This is only a problem when working with certain epithelial cells and not fibroblasts.
Immunogen	Synthetic peptide C-SRPSTSSRRRAISE, corresponding to amino acids 154-167 of human MDM2/HDM2. [UniProt# Q00987]
Product Application Details	
Applications	Western Blot, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin, Immunoprecipitation
Recommended Dilutions	Western Blot 1:500, Immunohistochemistry 1:100-1:500, Immunoprecipitation 1:10-1:500, Immunohistochemistry-Paraffin 1:100-1:500, Immunohistochemistry-Frozen 1:200-1:500
Application Notes	This antibody has also been reported useful in IHC on resin tissue sections. Staining of IHC-P tissues requires boiling tissue sections in 10mM citrate buffer, pH 6.0, for 10-20 min followed by cooling at RT for 20 min.

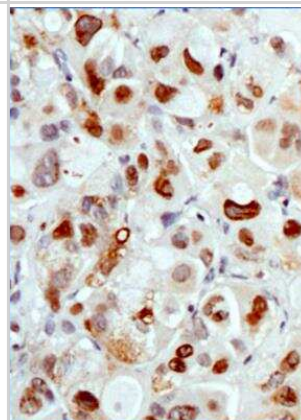


Images

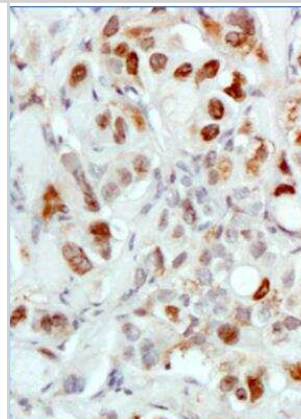
Western Blot: MDM2/HDM2 Antibody (SMP14) [NB100-2736] - Daudi whole cell protein was separated by SDS-PAGE on a 7.5% gel and transferred to PVDF membrane. The membrane was probed with anti-MDM2 antibody at 2 ug/ml and detected with an anti-mouse HRP secondary antibody using chemiluminescence.



Immunohistochemistry-Paraffin: MDM2/HDM2 Antibody (SMP14) [NB100-2736] - Analysis of tissue section of human breast cancer xenograft using MDM2/HDM2 antibody (clone SMP14) at 1:100 dilution. Several of the cancer cells developed a strong nuclear with weak cytoplasmic immunostaining of MDM2.



Immunohistochemistry-Paraffin: MDM2/HDM2 Antibody (SMP14) [NB100-2736] - Analysis of tissue section of human breast cancer xenograft using MDM2/HDM2 antibody (clone SMP14) at 1:100 dilution. Several of the cancer cells developed a strong nuclear with weak cytoplasmic immunostaining of MDM2.



Publications

Lee S, Jeong H, Hwang B et al. Helicobacter pylori promotes epithelial-to-mesenchymal transition by downregulating CK2B in gastric cancer cells *Biochimica et Biophysica Acta (BBA) - Molecular Basis of Disease* 2022-10-01 [PMID: 36404440]

Kiyga E, Adıguzel Z, Onay Ucar E Temozolomide increases heat shock proteins in extracellular vesicles released from glioblastoma cells *Molecular biology reports* 2022-06-25 [PMID: 35752701] (WB, Human)

Kaczorowski A, Tolstov Y, Falkenstein M et al. Rearranged ERG confers robustness to prostate cancer cells by subverting the function of p53 *Urol. Oncol.* 2020-07-13 [PMID: 32674955] (ICC/IF, Human)

Rodriguez-Lopez AM, Xenaki D, Eden TO et al MDM2 mediated nuclear exclusion of p53 attenuates EPE-induced apoptosis in neuroblastoma cells *Mol Pharmacol* 2001-01-01 [PMID: 11125034] (WB, Human)

Breton Y, Desrosiers V, Ouellet M et al. Expression of MDM2 in macrophages promotes the early post-entry steps of HIV-1 infection through inhibition of p53. *J. Virol.* 2019-01-23 [PMID: 30674627] (WB, Human)

Ramos GV, Cruz A, Silva WJ et al. Thyroid hormone upregulates MDM2 in rat type I fiber: implications for skeletal muscle mass regulation *Acta Physiol (Oxf)*. 2017-11-27 [PMID: 29178319] (Rat)

Joshi Y, Soria MG, Quadrato G et al. The MDM4/MDM2-p53-IGF1 axis controls axonal regeneration, sprouting and functional recovery after CNS injury. *Brain*. 2015-05-16 [PMID: 25981963] (WB, IHC-Fr, Mouse)

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Minsky N, Oren M . The RING domain of Mdm2 mediates histone ubiquitylation and transcriptional repression. *Mol Cell*. 2004-11-19 [PMID: 15546622] (IP, WB, Human)

Shieh SY, Ikeda M, Taya Y, Prives C. DNA damage-induced phosphorylation of p53 alleviates inhibition by MDM2. *Cell*. 1997-10-31 [PMID: 9363941] (WB, IP, Human)

Chen D, Zhang Z, Li M, Wang W et al. Ribosomal protein S7 as a novel modulator of p53-MDM2 interaction: binding to MDM2, stabilization of p53 protein, and activation of p53 function. *Oncogene*. 2007-08-02 [PMID: 17310983] (WB, Human)

Menendez S, Higgins M, Berkson RG et al. Nuclear export inhibitor leptomycin B induces the appearance of novel forms of human Mdm2 protein. *Br J Cancer*. 2003-02-24 [PMID: 12592381] (WB, Human)

More publications at <http://www.novusbio.com/NB100-2736>

Procedures

Western Blot Protocol for MDM2/HDM2 Antibody (NB100-2736)

Western Blot Protocol

1. Perform SDS-PAGE on samples to be analyzed, loading 10-25 ug of total protein per lane.
2. Transfer proteins to PVDF membrane according to the instructions provided by the manufacturer of the membrane and transfer apparatus.
3. Stain the membrane with Ponceau S (or similar product) to assess transfer success, and mark molecular weight standards where appropriate.
4. Rinse the blot TBS -0.05% Tween 20 (TBST).
5. Block the membrane in 5% Non-fat milk in TBST (blocking buffer) for at least 1 hour.
6. Wash the membrane in TBST three times for 10 minutes each.
7. Dilute primary antibody in blocking buffer and incubate overnight at 4C with gentle rocking.
8. Wash the membrane in TBST three times for 10 minutes each.
9. Incubate the membrane in diluted HRP conjugated secondary antibody in blocking buffer (as per manufacturer's instructions) for 1 hour at room temperature.
10. Wash the blot in TBST three times for 10 minutes each (this step can be repeated as required to reduce background).
11. Apply the detection reagent of choice in accordance with the manufacturer's instructions.

Immunohistochemistry-Paraffin Protocol for MDM2/HDM2 Antibody (NB100-2736)

Immunohistochemistry-Paraffin Embedded Sections

Antigen Unmasking:

Bring slides to a boil in 10 mM sodium citrate buffer (pH 6.0) then maintain at a sub-boiling temperature for 10 minutes. Cool slides on bench-top for 30 minutes (keep slides in the sodium citrate buffer at all times).

Staining:

1. Wash sections in deionized water three times for 5 minutes each.
2. Wash sections in PBS for 5 minutes.
3. Block each section with 100-400 ul blocking solution (1% BSA in PBS) for 1 hour at room temperature.
4. Remove blocking solution and add 100-400 ul diluted primary antibody. Incubate overnight at 4 C.
5. Remove antibody solution and wash sections in wash buffer three times for 5 minutes each.
6. Add 100-400 ul HRP polymer conjugated secondary antibody. Incubate 30 minutes at room temperature.
7. Wash sections three times in wash buffer for 5 minutes each.
8. Add 100-400 ul DAB substrate to each section and monitor staining closely.
9. As soon as the sections develop, immerse slides in deionized water.
10. Counterstain sections in hematoxylin.
11. Wash sections in deionized water two times for 5 minutes each.
12. Dehydrate sections.
13. Mount coverslips.





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Products Related to NB100-2736

HAF007	Goat anti-Mouse IgG Secondary Antibody [HRP]
NB720-B	Rabbit anti-Mouse IgG (H+L) Secondary Antibody [Biotin]
NBP1-43319-0.5mg	Mouse IgG1 Kappa Isotype Control (P3.6.2.8.1)
NB100-2736AF405	MDM2/HDM2 Antibody (SMP14) [Alexa Fluor® 405]

Limitations

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