

Datasheet for ABIN967511

anti-MDM2 antibody (AA 154-167)



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Overview

Quantity:	0.1 mg
Target:	MDM2
Binding Specificity:	AA 154-167
Reactivity:	Human, Mouse, Rat
Host:	Mouse
Clonality:	Monoclonal
Conjugate:	This MDM2 antibody is un-conjugated
Application:	Western Blotting (WB), Immunoprecipitation (IP), Immunohistochemistry (Frozen Sections) (IHC (fro)), Immunohistochemistry (Formalin-fixed Sections) (IHC (f)), BioImaging (BI)

Product Details

Brand:	BD Pharmingen™
Immunogen:	Human MDM2 aa. 154-167
Clone:	SMP14
Isotype:	IgG1
Cross-Reactivity:	Mouse (MURINE), Rat (RATTUS)
Characteristics:	<ol style="list-style-type: none"> 1. Since applications vary, each investigator should titrate the reagent to obtain optimal results. 2. This antibody has been developed and certified for the bioimaging application. However, a routine bioimaging test is not performed on every lot. Researchers are encouraged to titrate the reagent for optimal performance. 3. Triton is a trademark of the Dow Chemical Company.

Product Details

4. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
5. Please refer to us for technical protocols.

Purification:	The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.
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Target Details

Target:	MDM2
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Alternative Name:	MDM2 (MDM2 Products)
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Background:	<p>MDM2, originally described as a gene product of mouse double minute chromosomes, is a nuclear oncoprotein that can inhibit the action of certain tumor suppressor proteins. For example, MDM2 binds to the acidic activation domain (residues 2042) of the p53 tumor suppressor protein, and the p53-MDM2 complex down regulates the transcriptional activity of p53. p53 plays a role in the normal cell cycle by activating the transcription of genes that cause arrest in G1. The expression of MDM2 is itself, induced by p53 and may be a way for p53 to self-regulate its activity during the normal cell cycle. However, overexpression of MDM2 results in the loss of p53-regulated growth control and consequently, deregulated cell proliferation.</p> <p>MDM2 also binds to the Retinoblastoma tumor suppressor protein (Rb) and inhibits its growth regulatory function. MDM2 can directly augment proliferation by binding to two transcription factors E2F1 and DP1, and stimulating the activity of the S-phase inducing E2F1/DP1 heterodimer. MDM2 migrates at a reduced molecular weight of ~95 kDa.</p> <p>The SMP14 clone has been reported to recognize human, mouse and rat MDM2 while exhibiting a slight cross-reactivity with cytokeratins 6, 14 and 16 in some experimental systems. In the immunoprecipitation application, SMP14 has been reported to precipitate MDM2 and p53-MDM2 complexes. MCF7 human breast carcinoma cells (ATCC HTB-22) and NIH/3T3 mouse fibroblasts (ATCC CRL-1658) are suggested as western blot and immunoprecipitation positive controls. SMP14 has been reported to be useful for the immunohistochemical staining of acetone-fixed, frozen sections and of formalin-fixed, paraffin-embedded tissue sections. In addition to a nuclear staining of MDM2, cytoplasmic staining may also be observed which is likely to be attributable to the slight crossreactivity of the SMP14 clone with cytokeratins.</p>
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Molecular Weight:	95 kDa
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Pathways:	p53 Signaling , PI3K-Akt Signaling , Cell Division Cycle , Fc-epsilon Receptor Signaling Pathway , EGFR Signaling Pathway , Neurotrophin Signaling Pathway , Autophagy , Ubiquitin Proteasome
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Application Details

Application Notes:	<p>Bioimaging</p> <ol style="list-style-type: none">1. Seed the cells in appropriate culture medium at ~10,000 cells per well in an 96-well Imaging Plate and culture overnight.2. Remove the culture medium from the wells, and fix the cells by adding 100 myl of Fixation Buffer to each well. Incubate for 10 minutes at room temperature (RT).3. Remove the fixative from the wells, and permeabilize the cells using either 90% methanol, or Triton™ X-100: a. Add 100 myl of -20°C 90% methanol to each well and incubate for 5 minutes at RT. OR b. Add 100 myl of 0.1% Triton™ X-100 to each well and incubate for 5 minutes at RT.4. Remove the permeabilization buffer, and wash the wells twice with 100 myl of 1× PBS.5. Remove the PBS, and block the cells by adding 100 myl of to each well. Incubate for 30 minutes at RT.6. Remove the blocking buffer and add 50 myl of the optimally titrated primary antibody (diluted in Stain Buffer) to each well, and incubate for 1 hour at RT.7. Remove the primary antibody, and wash the wells three times with 100 myl of 1× PBS.8. Remove the PBS, and add the second step reagent at its optimally titrated concentration in 50 myl to each well, and incubate in the dark for 1 hour at RT.9. Remove the second step reagent, and wash the wells three times with 100 myl of 1× PBS.10. Remove the PBS, and counter-stain the nuclei by adding 200 myl per well of 2 myg/ml Hoechst 33342 in 1× PBS to each well at least 15 minutes before imaging.11. View and analyze the cells on an appropriate imaging instrument.
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Comment:	Related Products: ABIN967389, ABIN968585
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Restrictions:	For Research Use only
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Handling

Format:	Liquid
Concentration:	0.5 mg/mL
Buffer:	Aqueous buffered solution containing ≤0.09 % sodium azide.
Preservative:	Sodium azide
Precaution of Use:	This product contains Sodium azide: a POISONOUS AND HAZARDOUS SUBSTANCE which should be handled by trained staff only.

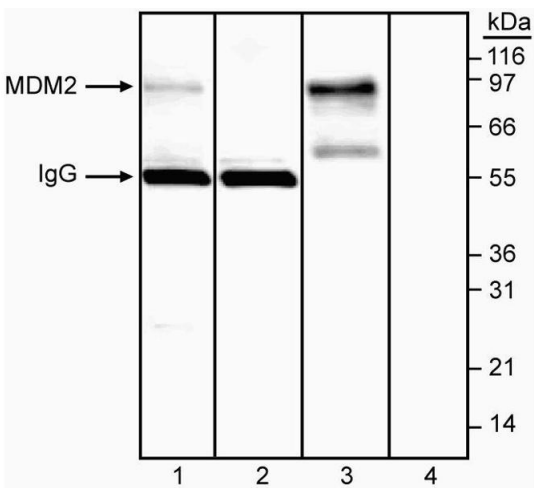
Handling

Storage:	4 °C
Storage Comment:	Store undiluted at 4°C.

Publications

Product cited in:	Martin, Trouche, Hagemeier, Sørensen, La Thangue, Kouzarides: "Stimulation of E2F1/DP1 transcriptional activity by MDM2 oncoprotein." in: Nature , Vol. 375, Issue 6533, pp. 691-4, (1995) (PubMed).
	Picksley, Vojtesek, Sparks, Lane: "Immunochemical analysis of the interaction of p53 with MDM2;--fine mapping of the MDM2 binding site on p53 using synthetic peptides." in: Oncogene , Vol. 9, Issue 9, pp. 2523-9, (1994) (PubMed).

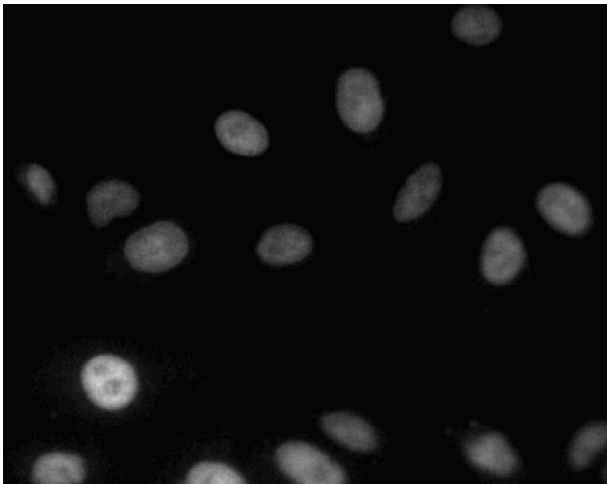
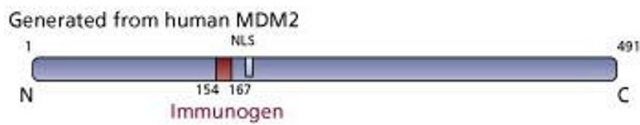
Images



Western Blotting

Image 1. Immunoprecipitation/western blot analysis of MDM2. A MCF7 human breast carcinoma cell lysate was used for immunoprecipitation at 2 ug antibody/1 million cells (lanes 1-2) and western blotting at 1 µg/ml (lane 1-4) with the anti-MDM2 antibody (clone SMP14), MCF7 cells (lanes 1 and 3) and isotype control (lanes 2 and 4). MDM2 is identified as a band of ~95 kDa, as well as a putative MDM2 breakdown product at ~62 kDa. The 55 kDa band represents the IgG heavy chain used for immunoprecipitation.

Image 2.



Immunofluorescence

Image 3. Immunofluorescent staining of A549 (ATCC CCL-185) cells. Cells were seeded in a 96 well imaging plate at ~10,000 cells per well. After overnight incubation, cells were stained using the alcohol perm protocol and the anti-MDM2 antibody. The second step reagent was FITC goat anti mouse Ig. The image was taken on a BD Pathway™ 855 Bioimager using a 20x objective. This antibody also stained HeLa (ATCC CCL-2) and U-2 OS (ATCC HTB-96) cells and can be used with either perm protocol.

Please check the [product details page](#) for more images. Overall 4 images are available for ABIN967511.