Product Datasheet

APE Antibody NB100-101SS

Unit Size: 0.025 ml

Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.

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NB100-101SS

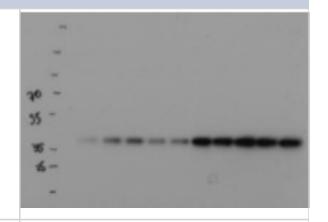
APE Antibody

AFE Antibody	
Product Information	
Unit Size	0.025 ml
Concentration	0.5 mg/ml
Storage	Aliquot and store at -20C or -80C. Avoid freeze-thaw cycles.
Clonality	Polyclonal
Preservative	0.05% Sodium Azide
Purity	Immunogen affinity purified
Buffer	Tris-glycine, 150 mM NaCl
Target Molecular Weight	37 kDa
Product Description	
Host	Rabbit
Gene ID	328
Gene Symbol	APEX1
Species	Human, Mouse, Rat, Primate, Rabbit
Species Reactivity	Human, mouse, primate and rat. Rabbit reactivity reported in the scientific literature (PMID: 15276530).
Immunogen	Affinity purified human APE1 [UniProt# P27695]
Notes	SH-SY5Y Lysate (nuclear extract) image in western blot provided via veririfed customer review.
Product Application Details	
Applications	Western Blot, Simple Western, Blocking/Neutralizing, Chromatin Immunoprecipitation, Immunocytochemistry/Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin, Immunoprecipitation
Recommended Dilutions	Blocking/Neutralizing 50-200 molar excess, Chromatin Immunoprecipitation 1:10 -1:500, Immunocytochemistry/Immunofluorescence 1:50-1:200, Immunohistochemistry 1:100, Immunohistochemistry-Frozen 1:100, Immunohistochemistry-Paraffin 1:100, Immunoprecipitation 7 ug/ml, Western Blot 1:1000, Simple Western 1:12.5
Application Notes	This APE1 antibody is useful for Blocking/Neutralizing, Chromatin Immunoprecipitation, Immunocytochemistry/Immunofluorescence, Immunohistochemistry, Immunoprecipitation and Western Blot. In WB this antibody detects a single band at 37 kDa. In IHC it can be competitively inhibited from recognizing the APE1 antigen in tissues using APE1 protein. This antibody can be used on frozen sections, fixed-paraffin sections and cytospin preps. NB100-101 can also be used following the apoptosis (TUNEL) procedure with the Boehringer-Mannheim TUNEL assay kit. Antibody staining should be performed AFTER the TUNEL assay. NB100-101 can inhibit the repair activity of APE1 protein. In Simple Western only 10-15 uL of the recommended dilution is used per data point.

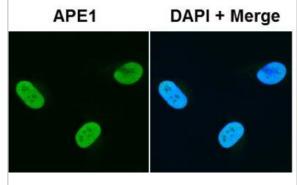


Images

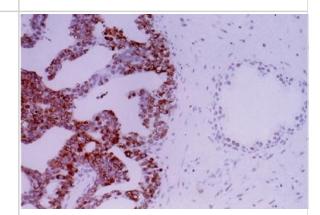
Western Blot: APE Antibody [NB100-101] - analysis of APE in human melanoma cell lysate using anti-APE antibody. Image from verified customer review.



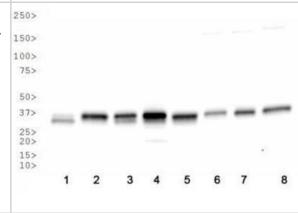
Immunocytochemistry/Immunofluorescence: APE1 Antibody [NB100-101] - IF on HeLa cells. Image from verified customer review.



Immunohistochemistry: APE1 Antibody [NB100-101] - Immunohistochemical staining of APE-ref-1 in prostate cancer.



Western Blot: APE1 Antibody [NB100-101] - Analysis of APE1 in cell lysates: 1. HeLa, 2. Ntera2, 3. A431, 4. HepG2, 5. MCF7, 6. NIH 3T3, 7. PC12, and 8. Cos 7.





Western Blot: APE1 Antibody [NB100-101] - SH-SY5Y (nuclear extract) tested at 1:1000 dilution. Image provided by verified customer review. 130 ---Immunocytochemistry/Immunofluorescence: APE1 Antibody [NB100-101] - Detection of APE1 (Green) in HepG2 cells using NB100-101. Nuclei (Blue) are counterstained with Hoechst 33258. Simple Western: APE Antibody [NB100-101] - Simple Western lane view shows a specific band for APE1 in 0.1 mg/ml of HeLa lysate. This experiment was performed under reducing conditions using the 12-230 kDa separation system.



Publications

Qutob H. DNA damage response and anti-apoptotic proteins expression in NPMc+ mutated cells in acute myeloid leukaemia Thesis. 2015 Jan 23 (ICC/IF, IP, Human)

Das D, Preet R, Mohapatra P et al. 5-Fluorouracil mediated anti-cancer activity in colon cancer cells is through the induction of Adenomatous Polyposis Coli: Implication of the long-patch base excision repair pathway. DnA Repair (Amst.). 2014 Nov 20 [PMID: 25460919] (WB, Human)

Abdel-Fatah TM, Perry C, Arora A et al. Is there a role for base excision repair in estrogen driven breast cancers? Antioxid. Redox Signal. 2014 Aug 11 [PMID: 25111287] (IHC-P, Human)

Details:

Rabbit polyclonal anti-APE1 antibody used for IHC on tissue microarray (TMAs) at a dilution of 1:500, incubated for 1 hour. Citrate buffer (pH6.0) was used as an antigen retrieval reagent (Table 3).

Liu GD, Xia L, Zhu JW et al. Genistein Alleviates Radiation-Induced Pneumonitis by Depressing Ape1/Ref-1 Expression to Down-regulate Inflammatory Cytokines. Cell Biochem. Biophys. 3/23/2014 [PMID: 24659138] (WB, Mouse)

Alagoz M, Wells OS, El-Khamisy SF. TDP1 deficiency sensitizes human cells to base damage via distinct topoisomerase I and PARP mechanisms with potential applications for cancer therapy. Nucleic Acids Res 2013 Dec 13 [PMID: 24335147] (WB, Human)

Abdel-Fatah TM, Perry C, Moseley P et al. Clinicopathological significance of human apurinic/apyrimidinic endonuclease 1 (APE1) expression in oestrogen-receptor-positive breast cancer. Breast Cancer Res Treat 2014 Jan 1 [PMID: 24381055] (IHC-P, Human)

Das D, Preet R, Mohapatra P et al. 1,3-Bis(2-chloroethyl)-1-nitrosourea enhances the inhibitory effect of Resveratrol on 5-fluorouracil sensitive/resistant colon cancer cells. World J Gastroenterol. 2013 Nov 14 [PMID: 24259968] (WB, Human)

Chu TH, Guo A, Wu W. Down-regulation of apurinic/apyrimidinic endonuclease 1 (APE1) in spinal motor neurons under oxidative stress. Neuropathol Appl Neurobiol 2013 Jul 1 [PMID: 23808792] (WB, IHC, Rat, Mouse)

Kim HL, Kim SU, Seo YR. A novel role for Gadd45alpha in base excision repair: Modulation of APE1 activity by the direct interaction of Gadd45alpha with PCNA. Biochem Biophys Res Commun 2013 Feb 25 [PMID: 23485469] (WB, Human)

Cardoso AA, Jiang Y, Luo M et al. APE1/Ref-1 Regulates STAT3 Transcriptional Activity and APE1/Ref-1-STAT3 Dual-Targeting Effectively Inhibits Pancreatic Cancer Cell Survival PLoS One 2012 [PMID: 23094050] (WB, Human)

Sultana R, McNeill DR, Abbotts R et al. Synthetic lethal targeting of DNA double-strand break repair deficient cells by human apurinic/apyrimidinic endonuclease inhibitors Int J Cancer 2012 Nov 15 [PMID: 22377908] (WB, Human)

Shaked H, Hofseth LJ, Chumanevich A et al. Chronic epithelial NF-kB activation accelerates APC loss and intestinal tumor initiation through iNOS up-regulation Proc Natl Acad Sci U S A 2012 Aug 14 [PMID: 22893683] (IHC, Mouse)

More publications at http://www.novusbio.com/NB100-101



Procedures

Western Blot Protocol for APE1 Antibody (NB100-101)

Western Blot

- 1. Proteins are separated on 4-20% Tris-glycine (SDS/PAGE) gels at 125V for 1.5 hours.
- 2. Proteins are transferred to 0.45 mm nitrocellulose sheets by electroblotting in a Novex XCell II transfer apparatus, using Novex Transfer Buffer. Gels, Whatman paper, and NC membranes are wet in electroblotting buffer prior to transferring. The transfer is carried out for 1.5 hours at 25V.
- 3. Following the protein transfer, the filters are blocked with 3% BSA in PBS for 1 hour at room temperature, gently shaking.
- 4. The filters are then washed 3x5 minutes in washing buffer [0.5% BSA and 0.01% Tween-20 in PBS] at room temperature, gently shaking.
- 5. NB100-101 (APE/ref-1 primary antibody) is diluted 1:1,000 in dilution buffer [0.5% BSA in PBS] and incubated with the filters for 1 hour at room temperature, gently shaking.
- 6. The filters are then washed 3x5 minutes in washing buffer at room temperature, gently shaking.
- 7. Secondary antibody [HRP conjugated goat anti-rabbit, BioRad] is diluted 1:10,000 in dilution buffer and incubated with the filters for 1 hour at room temperature, gently shaking.
- 8. The filters are then washed 3x5 minutes in washing buffer at room temperature, gently shaking. Cross-reacting proteins are detected using the Amplified Opti-4CN Western Blotting kit (BAR and streptavidin-HRP incubations and colormetric detection) from BioRad.

NOTE: HeLa whole cell extracts (NB800-PC1) were used a s a positive control for this antibody.

Immunohistochemistry/Immunocytochemistry

The description that follows is for cultured cells but can be used for cytospins.

- 1. Split cells into 3.5 cm culture dishes for growth.
- 2. Wash cells with 5 ml PBS.
- 3. Fix cells with approx. 3 ml Histochoice (Amresco) for 30 min. (Cryostat tissues for 45 min.) or use 10% formalin for 30 minutes.
- 4. Rinse cells with 5 ml TBS, wipe plate dry leaving a small circle of buffer and cells. Mark with red pencil.
- 5. Preblock the cells for 30 min. with 10% goat serum in TBS (200 ul).
- 6. Aspirate blocking solution and add NB100-101 (APE/ref-1 primary antibody) at a dilution of 1:100 in 10% goat serum.
- 7. Incubate in humidified chamber for 3 hours. (Overnight for tissue at 4 degrees C).
- 8. Incubate the cells with 1:100 diluted 20 antibody (anti-rabbit IgG made in goat) in 10% goat serum and TBS for 1 hour in humidified chamber.
- 9. Wash 2 times with 5 ml TBS for 5 min. each.
- 10. Block with ABC solution for 30 min.
- 11. Wash 2 times with 5 ml TBS for 5 min. each.
- 12. Incubate with DAB solution until signal develops. Place into dH2O. Add coverslip with Aqua-mount. TBS: 50 mM Tris, 150 mM NaCl, pH 7.5 ABC and DAB solutions: Vector Laboratories





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Limitations

This product is for research use only and is not approved for use in humans or in clinical diagnosis. Primary Antibodies are guaranteed for 1 year from date of receipt.

For more information on our guarantee, please visit www.novusbio.com/guarantee.

