Product Datasheet

APE Antibody (13B8E5C2) - BSA Free
NB100-116

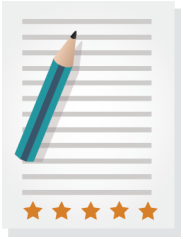
Unit Size: 0.1 mg

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

Reviews: 3 Publications: 105

Protocols, Publications, Related Products, Reviews, Research Tools and Images at:
www.novusbio.com/NB100-116

Updated 12/20/2023 v.20.1



NB100-116

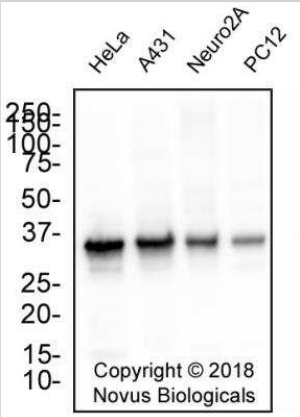
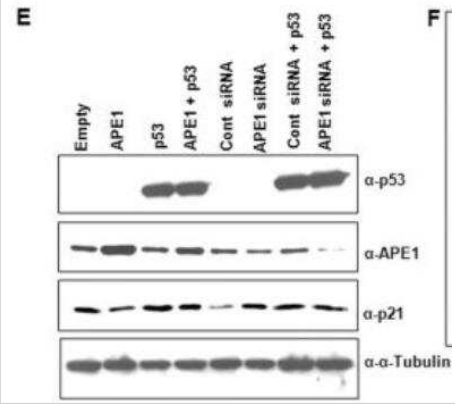
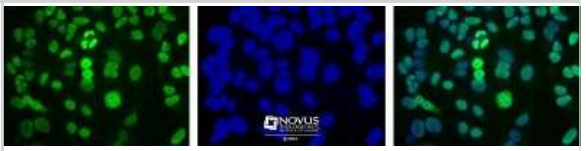
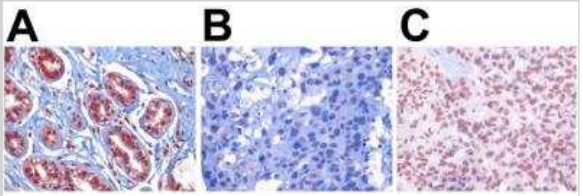
APE Antibody (13B8E5C2) - BSA Free

Product Information	
Unit Size	0.1 mg
Concentration	1.0 mg/ml
Storage	Store at 4C short term. Aliquot and store at -20C long term. Avoid freeze-thaw cycles.
Clonality	Monoclonal
Clone	13B8E5C2
Preservative	0.02% Sodium Azide
Isotype	IgG2b
Purity	Protein G purified
Buffer	PBS
Target Molecular Weight	37 kDa

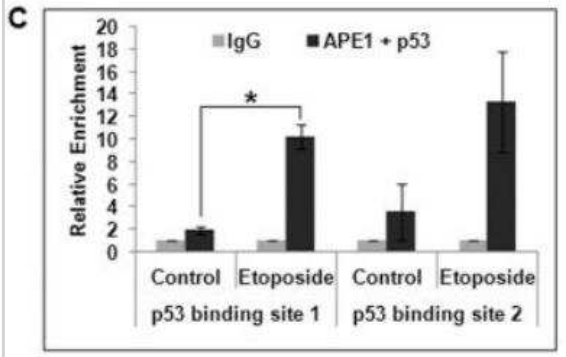
Product Description	
Host	Mouse
Gene ID	328
Gene Symbol	APEX1
Species	Human, Mouse, Rat, Primate
Immunogen	Purified human APE1 [Uniprot: P27695]

Product Application Details	
Applications	Western Blot, Simple Western, ELISA, Gel Super Shift Assays, Immunoblotting, Immunocytochemistry/ Immunofluorescence, Immunohistochemistry, Immunohistochemistry-Frozen, Immunohistochemistry-Paraffin, Immunoprecipitation, Proximity Ligation Assay, Chromatin Immunoprecipitation (ChIP), Knockdown Validated
Recommended Dilutions	Western Blot 1:100-1:2000, Simple Western 1:25, ELISA reported in scientific literature (PMID 21769563), Immunohistochemistry 1:100, Immunocytochemistry/ Immunofluorescence 1:50-1:200, Immunoprecipitation 1:10 - 1:500. Use reported in scientific literature (PMID 35286386), Immunohistochemistry-Paraffin 1:100, Immunohistochemistry-Frozen 1:10-1:500, Immunoblotting reported in scientific literature (PMID 27608656), Gel Super Shift Assays reported in scientific literature, Proximity Ligation Assay reported in scientific literature (PMID 27808278), Chromatin Immunoprecipitation (ChIP) 1:10-1:500, Knockdown Validated
Application Notes	In Western blot, 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. It can also be used on frozen and fixed-paraffin sections and cytospin preps. In IHC-P, staining was observed in the nucleus of a human breast cancer xenograft. Prior to immunostaining paraffin tissues, antigen retrieval with sodium citrate buffer (pH 6.0) is recommended. In Simple Western only 10 - 15 uL of the recommended dilution is used per data point. Separated by Size-Wes, Sally Sue/Peggy Sue.

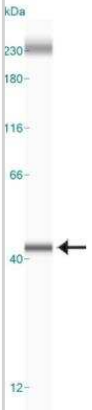
Images

Western Blot: APE Antibody (13B8E5C2) - BSA Free [NB100-116] - APE Antibody (13B8E5C2) [NB100-116] - Whole cell protein from human HeLa, A431, mouse Neuro2A and rat PC12 cells was separated on a 12% gel by SDS-PAGE, transferred to PVDF membrane and blocked in 5% non-fat milk in TBST. The membrane was probed with 2.0 ug/mL anti-APE-1 in block buffer and detected with an anti-mouse HRP secondary antibody using chemiluminescence.	
Knockdown Validated: APE Antibody (13B8E5C2) - BSA Free [NB100-116] - APE Antibody (13B8E5C2) [NB100-116] - Repression of p21 by APE1 in p53-null cells and effect of ectopic p53 in this repression. Representative Western analysis of p53, APE1, p21 and alpha-Tubulin levels in the same HCT116p53null cells. Image collected and cropped by CiteAb from the following publication (https://dx.plos.org/10.1371/journal.pone.0068467) licensed under a CC-BY license.	
Immunocytochemistry/Immunofluorescence: APE Antibody (13B8E5C2) - BSA Free [NB100-116] - APE Antibody (13B8E5C2) [NB100-116] - HeLa cells stained NB100-116 (Green) detected with DyLight Fluor 488 conjugated anti-rabbit IgG secondary antibody. Nuclei are counterstained with Hoechst 33258 (Blue).	
Immunohistochemistry: APE Antibody (13B8E5C2) - BSA Free [NB100-116] - APE Antibody (13B8E5C2) [NB100-116] - Immunohistochemical staining of APE1. A. Nuclear staining in the luminal epithelium of normal breast ducts and lobules. B. Low, and C. high nuclear APE1 expression in invasive breast cancer. (magnification x400). Image collected and cropped by CiteAb from the following publication (//dx.plos.org/10.1371/journal.pone.0099528) licensed under a CC-BY license.	

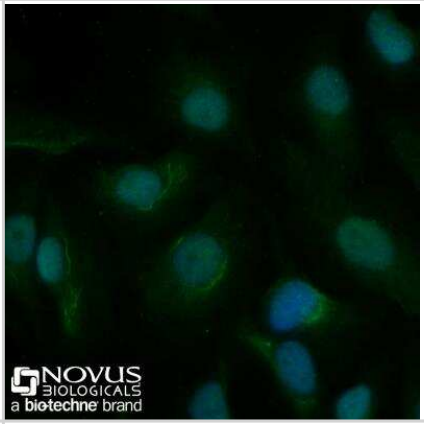
Chromatin Immunoprecipitation (ChIP): APE Antibody (13B8E5C2) - BSA Free [NB100-116] - APE Antibody (13B8E5C2) [NB100-116] - Association of p53 and APE1 on p53-binding sites in p21 promoter. Re-ChIP analysis (first IP with alpha-APE1 and the second IP with alpha-p53 antibody) showing simultaneous recruitment of APE1 and p53 in control vs. EPE treated cells; *: p value <0.05 (n=2) calculated based on APE1/p53 enriched DNA from control vs. etoposide treated cells. Image collected and cropped by CiteAb from the following publication (https://dx.plos.org/10.1371/journal.pone.0068467) licensed under a CC-BY license.



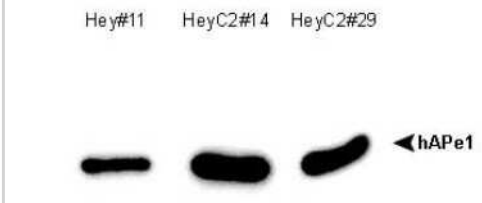
Simple Western: APE Antibody (13B8E5C2) - BSA Free [NB100-116] - APE Antibody (13B8E5C2) [NB100-116] - Image shows a specific band for APE1 in 0.1 mg/mL of HeLa lysate. This experiment was performed under reducing conditions using the 12-230kDa separation system. * Non-specific interaction with the 230 kDa Simple Western standard may be seen with this antibody.

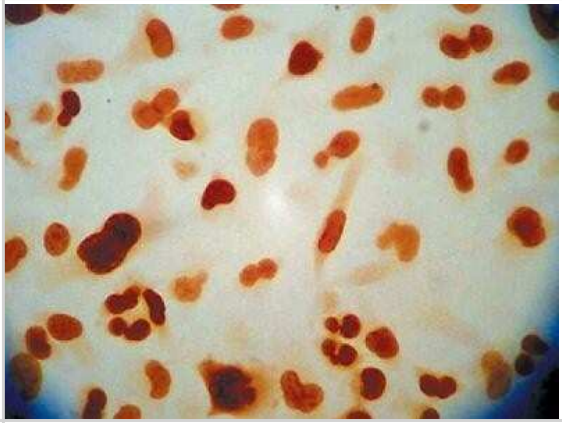
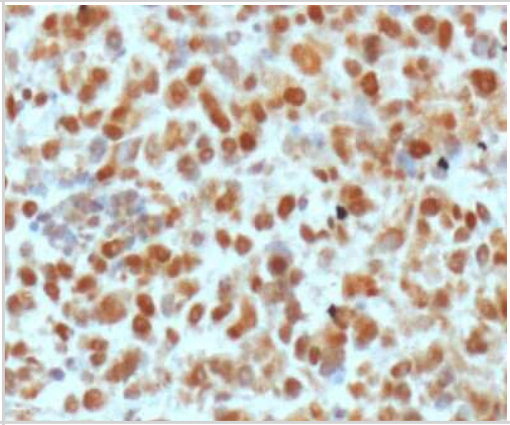
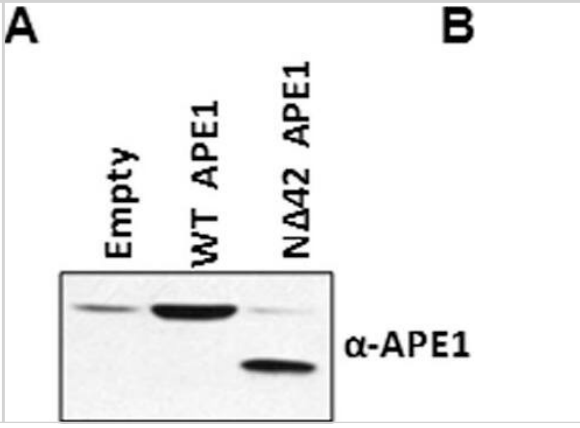


Immunocytochemistry/Immunofluorescence: APE Antibody (13B8E5C2) - BSA Free [NB100-116] - APE Antibody (13B8E5C2) [NB100-116] - HeLa cells were fixed for 10 minutes using 10% formalin and then permeabilized for 5 minutes using 1X PBS + 0.05% Triton X-100. The cells were incubated with anti-APE (13B8E5C2) at 5 ug/mL overnight at 4C and detected with an anti-mouse DyLight 488 (Green) at a 1:500 dilution. Nuclei were counterstained with DAPI (Blue). Cells were imaged using a 40X objective.

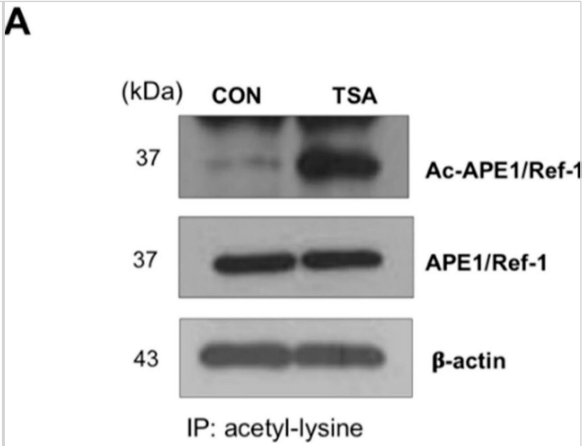


Western Blot: APE Antibody (13B8E5C2) - BSA Free [NB100-116] - APE Antibody (13B8E5C2) [NB100-116] - Ovarian Cancer cell lines.

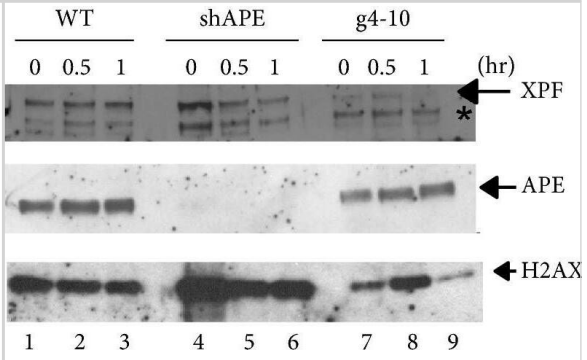


Immunocytochemistry/Immunofluorescence: APE Antibody (13B8E5C2) - BSA Free [NB100-116] - APE Antibody (13B8E5C2) [NB100-116] - Immunocytochemical detection of APE-ref-1 in breast cancer cell line MDA MB 231.	
Immunohistochemistry-Paraffin: APE Antibody (13B8E5C2) - BSA Free [NB100-116] - APE Antibody (13B8E5C2) [NB100-116] - APE1 antibody was tested in human breast cancer xenograft using DAB with hematoxylin counterstain.	
TARS expression co-localized with VEGF and in leukocytes. Tissues were stained by IHC as in Figure 2. Shown are 40x images of serial sections stained with (A,B) No primary Ab as a negative control (C,D) TARS or (E,F) VEGF. Bottom panels show examples of TARS staining in infiltrating leukocytes. Bar = 50 µm. See Additional File 3 for supporting images. Image collected and cropped by CiteAb from the following open publication (https://bmccancer.biomedcentral.com/articles/10.1186/1471-2407-14-620), licensed under a CC-BY license. Not internally tested by Novus Biologicals.	

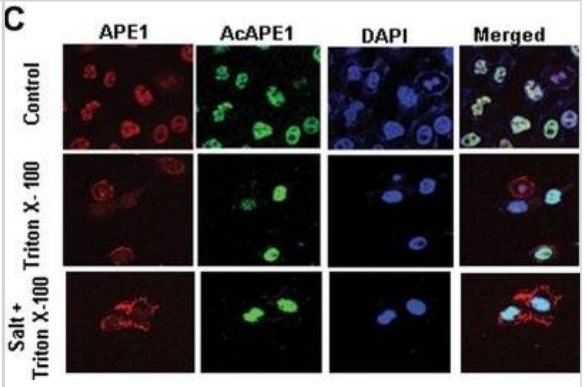
G-TPP activity is conserved in primary fibroblasts(A, C) Fibroblasts were treated with 15 μM G-TPP for the indicated time points. Cells were harvested and western blots were probed with antibodies against (A) PINK1, pS65-Ub and total Ub or (C) autophagy adapter proteins. GAPDH and Vinculin served as loading control. G-TPP treatment led to PINK1 stabilization and pS65-Ub induction in primary skin fibroblasts. p62 levels were induced upon G-TPP treatment, while other adapters seemed decreased. (B, D) Human fibroblasts were treated with 15 μM G-TPP for 16 h and fixed and stained with antibodies against (B) pS65-Ub (green) or (D) the autophagy adapters NBR1, NDP52, p62, OPTN and TAX1BP1 (green). Mitochondria were stained with antibodies against TOM20 (red), nuclei were visualized with Hoechst (blue). Scale bars indicate 10 μM. A magnified image of the boxed region, the fluorescence profile along the arrow and the Pearson's correlation coefficient of adapter protein and mitochondrial stainingare shown to the right. Shown is the mean ± SEM of at least five randomly selected images (unpaired, two-sided t-test, ***p < 0.0005). Image collected and cropped by CiteAb from the following open publication (https://www.oncotarget.com/lookup/doi/10.18632/oncotarget.22287), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



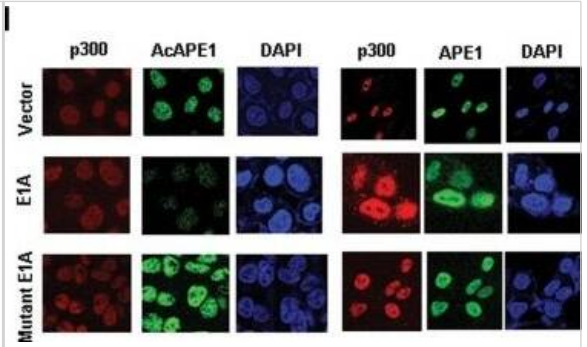
The levels of glial fibrillary acidic protein (GFAP) and S100β as astrocytic protein marker proteins in the hippocampal area. (A) Western blot images using GFAP, tubulin beta□3, and β□actin antibodies to total protein lysate in hippocampal area. The statistical results of internal standardized GFAP/β□tubulin ratio by β□actin levels relative to young adult wild□type C57BL/6J. White and black bars indicate the wild□type C57BL/6J and Tet□mev□1 mice, respectively. Data are expressed as mean ± SD; *P < 0.05; **P < 0.01; n = >12 in each group. (B) Western blot images using S100β, tubulin beta□3, and β□actin antibodies to total protein lysate in hippocampal area. The statistical results of internal standardized S100β/β□tubulin ratio by β□actin levels relative to young adult wild□type C57BL/6J. White and black bars indicate the wild□type C57BL/6J and Tet□mev□1 mice, respectively. Data are expressed as mean ± SD; *P < 0.05; **P < 0.01; n = >12 in each group. (C) Micrographs of immunohistochemical analysis on paraffined hippocampal tissue sections using GFAP and S100β antibody. Brown cells indicate GFAP□stained astrocytes. Scale bar = 100 μm. Image collected and cropped by CiteAb from the following open publication (https://pubmed.ncbi.nlm.nih.gov/27623715), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



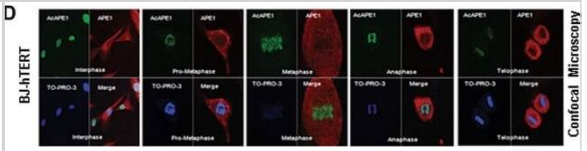
EBNA1 promotes the upregulation of oxidative DNA damage repair pathways in EBV converted cell lines and EBV positive BLs. a Representative western blots illustrating the expression of MTH1, OGG1, and MUTYH in pairs of EBV-negative and -positive cell lines. GAPDH was used as loading control. b Densitometry quantification of the specific bands. The intensity of the specific band in EBV positive cells relative the EBV-negative parental is shown. Mean ± SE of four independent experiments. c Representative western blots illustrating the expression of EBNA1, MTH1, OGG1, and MUTYH in the Mutu cell lines. d Densitometry quantification of expression in the EBV positive cell lines relative to the EBV-negative Mutu-30. Mean ± SE of four independent experiments Image collected and cropped by CiteAb from the following open publication (https://pubmed.ncbi.nlm.nih.gov/31511648), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



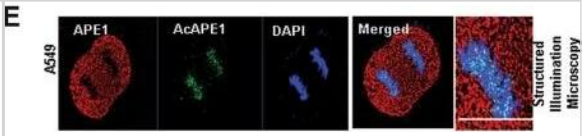
Western blot images (A) and quantifications (B–G) of cell lysates from C2C12 myotubes with control or UCHL1 siRNA knockdown for perilipin 2 (B), perilipin 5 (C), CD36 (D), HSL (E), MAGL (F), and SDHB (G). n = 4 per group. Image collected and cropped by CiteAb from the following open publication (https://pubmed.ncbi.nlm.nih.gov/35464088), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



BDNF lowered the mechanical withdrawal threshold further and promoted activation of astrocytes and microglia, and enhanced the p38/JNK pathway to aggravate the release of IL-1β and TNF-α in the SDH of CYP-induced cystitis. a BDNF treated every other day after CYP injection could further lower the mechanical withdrawal threshold and suppress the retrieval of mechanical threshold when compared with the CYP + PBS group. After the exogenous BDNF injection, Western blots showing the expression of b BDNF, c TrkB, dp-TrkB, e Iba1, f GFAP, gp-p38, hp-JNK, i TNF-α, and j IL-1β were all further upregulated when compared with the CYP + PBS group. Data of mechanical withdrawal threshold were analyzed using a two-way analysis of variance (ANOVA) followed by the Sidak's multiple comparisons test. All data were calculated as mean ± SEM (n = 10 per group). **p < 0.01, ***p < 0.001 vs. the control group. #p < 0.05, ##p < 0.01, ###p < 0.001 vs. the CYP + rBDNF group. $p < 0.05, $$p < 0.01, $$$p < 0.001 vs. the CYP + PBS group Image collected and cropped by CiteAb from the following open publication (https://pubmed.ncbi.nlm.nih.gov/31931832), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Primary human grade IV glioblastoma xenotransplant.(A) Dynamic PET image (coronal) of advanced tumor in the right hemisphere of the brain with [18F]PBR06 uptake confined primarily to the tumor. Arrows indicate tumor and infiltration into left hemisphere. (B) Time-activity curves of injected [18F]PBR06 in tumor (green) and contralateral brain (blue) [30]. (C) Correlative TSPO immunohistochemistry, Tumor + White Matter Tract (40X). Image collected and cropped by CiteAb from the following open publication (https://dx.plos.org/10.1371/journal.pone.0141659), licensed under a CC-BY license. Not internally tested by Novus Biologicals.



Publications

Chen YH, Kuo YY, You YQ et al. Endonuclease VIII-like 1 deficiency potentiates nigrostriatal dopaminergic neuron degeneration in a male mouse model of Parkinson's disease Journal of neurochemistry 2023-02-24 [PMID: 36840377]

Champion J Targeting the REF1/STAT3 axis to treat tuberous sclerosis Thesis 2023-01-01 (WB, Human)

Details:
1:1000 WB dilution

Ito M, Ducasa GM, Molina JD et al. ABCA1 deficiency contributes to podocyte pyroptosis priming via the APE1/IRF1 axis in diabetic kidney disease Scientific reports 2023-06-14 [PMID: 37316538] (WB, Mouse)

Details:
1:500 dilution, fresh sections used

Chieffi Baccari G, Falvo S, Di Fiore MM et al. High-fat diet affects autophagy and mitochondrial compartment in rat Harderian gland Journal of experimental zoology. Part A, Ecological and integrative physiology 2022-08-04 [PMID: 35927786]

Latino D, Chieffi Baccari G, Di Fiore MM et al. Autophagy and mitochondrial damage in the testis of high-fat diet fed rats General and comparative endocrinology 2022-08-13 [PMID: 35973585] (WB, Rat)

Details:
Dilution used for WB 1:1500

Rios-Covian D, Butcher LD, Ablack AL et al. A novel hypomorphic Apex1 mouse model implicates apurinic-apyrimidinic endonuclease 1 in oxidative DNA damage repair in gastric epithelial cells Antioxidants & redox signaling 2022-06-25 [PMID: 35754343] (WB)

Pramanik S, Chen Y, Song H et al. The human AP-endonuclease 1 (APE1) is a DNA G-quadruplex structure binding protein and regulates KRAS expression in pancreatic ductal adenocarcinoma cells Nucleic acids research 2022-03-14 [PMID: 35286386] (IP, WB, ICC/IF, Human)

Cun, Y, Dai, N Et al. APE1/Ref-1 enhances DNA binding activity of mutant p53 in a redox-dependent manner. Oncol Rep 2014-02-01 [PMID: 24297337] (IF/IHC, WB, Mouse)

Song H, Xi S, Chen Y Et Al. Histone chaperone FACT complex inhibitor CBL0137 interferes with DNA damage repair and enhances sensitivity of medulloblastoma to chemotherapy and radiation Cancer letters 2021-07-14 [PMID: 34271103] (IP, WB, ICC/IF)

Shen J, Varshney D, Simeone A et al. Promoter G-quadruplex folding precedes transcription and is controlled by chromatin Genome biology 2021-05-07 [PMID: 33962653] (WB, Human)

Gampala S, Shah F, Zhang C et al. Exploring transcriptional regulators Ref-1 and STAT3 as therapeutic targets in malignant peripheral nerve sheath tumours British journal of cancer 2021-03-03 [PMID: 33658640] (IHC-P, WB)

Bacolla A, Sengupta S, Ye Z et al. Heritable pattern of oxidized DNA base repair coincides with pre-targeting of repair complexes to open chromatin Nucleic acids research 2020-12-09 [PMID: 33300026] (ICC/IF, Human)

Details:
HCT116 cells

More publications at http://www.novusbio.com/NB100-116

Procedures

Western Blot Protocol for APE1 Antibody (NB100-116)

Western Blot
1. Gels, Whatman paper, and membranes are soaked in electroblotting buffer (25 mM Tris-HCl; 193 mM glycine; 20% methanol) for 15 minutes prior to transferring
2. Proteins separated on SDS-polyacrylamide gels are transferred onto 0.22 micron nitrocellulose sheets by electroblotting in a Transblot BioRad transfer apparatus in 25 mM Tris, 192 mM Glycine, 20% Methanol at 150 mA (70 V). The transfer is carried out for 1 hour at 4 degrees C.
3. Following protein transfer, the filter is blocked with Blotto [1X TBST (10X TBST = 1.5 M NaCl; 100 mM Tris-HCl, pH 8.0; 0.5% Tween 20; 2% NP-40; 0.2% SDS); 5% Carnation dried milk; 0.02% sodium azide] for 1 hour at room temperature on a rotator.
4. Dilute NB 100-116 (anti-APE/ref-1) in Blotto and incubate with the filter, at 4 degrees C overnight, on a rotator.
5. Wash filter 3 times in 1X TBST (50 mM Tris-HCl, 150 mM NaCl, 0.1% Tween 20, pH 7.5) for 10 minutes at 4 degrees C. Secondary antibody (peroxidase conjugated anti-mouse) is incubated with the blot for 30 minutes at room temperature. Cross-reacting proteins are detected using the Chemiluminescence Western Blotting Kit from Boehringer-Mannheim.
NOTE:
HeLa whole cell extracts (NB800-PC1) were used as 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. Pre-block the cells for 30 min. with 10% goat serum in TBS (200 ul).
6. Aspirate blocking solution and add NB 100-116 (anti-APE/ref-1), dilution 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 secondary antibody (anti-mouse 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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Products Related to NB100-116

NB800-PC1	HeLa Whole Cell Lysate
NBP1-49581	APE1 Redox Inhibitor
HAF007	Goat anti-Mouse IgG Secondary Antibody [HRP]
NB720-B	Rabbit anti-Mouse IgG (H+L) Secondary Antibody [Biotin]
NBP2-27231	Mouse IgG2b Isotype Control (MPC-11)

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.

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