Anti-APE1 Antibody [12H2-R]

HA601212

Product Type:	Recombinant Mouse monoclonal IgG1, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IHC-P
Molecular Wt:	Predicted band size: 36 kDa
Clone number:	12H2-R

Description: The role of transcription factors in the regulation of gene expression is well established. Although the activity of these factors can be regulated by phosphorylation, evidence has indicated regulation of DNA binding mediated by changes in reduction-oxidation (redox) status. Mutational analysis has identified a single conserved cysteine residue mapping within the DNA binding domains of Fos and Jun. Chemical oxidation or modification of this cysteine residue inhibits the DNA binding activity of Fos and Jun. A similar mode of regulation has been recently proposed for other nuclear transcription factors. Oxidation is reversible by these compounds or by a cellular redox/DNA repair protein identified originally as Ref-1 (redox factor 1). Ref-1 is identical to a previously characterized DNA repair enzyme designated HAP1, APE or APEX.

Immunogen: Recombinant protein within Human APE1 aa 20-318/318.

Positive control: HeLa cell lysate, LNCaP cell lysate, HEK-293 cell lysate, HL-60 cell lysate, A431 cell lysate, NIH/3T3 cell lysate, C6 cell lysate, rat testis tissue lysate, A549, HeLa, human colon carcinoma tissue, human kidney tissue, mouse liver tissue, rat colon tissue.

Subcellular location: Nucleus. Endoplasmic reticulum, Cytoplasm.

Database links: SwissProt: P27695 Human | P28352 Mouse | P43138 Rat

Recommended Dilutions:

WB	1:1,000
IF-Cell	1:100
IHC-P	1:200-1:1,000

Storage Buffer: PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.

Purity: Protein A affinity purified.

Images

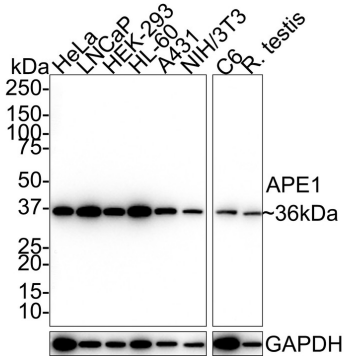

Fig1: Western blot analysis of APE1 on different lysates with Mouse anti-APE1 antibody (HA601212) at 1/1,000 dilution.

Lane 1: HeLa cell lysate (20 µg/Lane)
Lane 2: LNCaP cell lysate (20 µg/Lane)
Lane 3: HEK-293 cell lysate (20 µg/Lane)
Lane 4: HL-60 cell lysate (20 µg/Lane)
Lane 5: A431 cell lysate (20 µg/Lane)
Lane 6: NIH/3T3 cell lysate (20 µg/Lane)
Lane 7: C6 cell lysate (20 µg/Lane)
Lane 8: Rat testis tissue lysate (40 µg/Lane)

Predicted band size: 36 kDa
Observed band size: 36 kDa

Exposure time: 3 minutes;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA601212) at 1/1,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

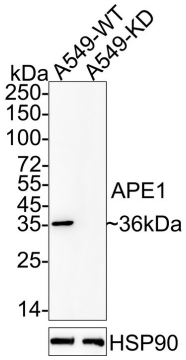

Fig2: Western blot analysis of APE1 on different lysates with Mouse anti-APE1 antibody (HA601212) at 1/5,000 dilution.

Lane 1: A549-WT cell lysate
Lane 2: A549-KD APE1 cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 36 kDa
Observed band size: 36 kDa

Exposure time: 3 minutes; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA601212) at 1/5,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

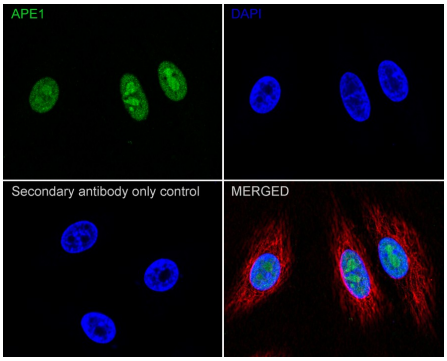

Fig3: Immunocytochemistry analysis of A549 cells labeling APE1 with Mouse anti-APE1 antibody (HA601212) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Mouse anti-APE1 antibody (HA601212) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI. beta Tubulin (ET1602-4, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) were used as the secondary antibody at 1/1,000 dilution.

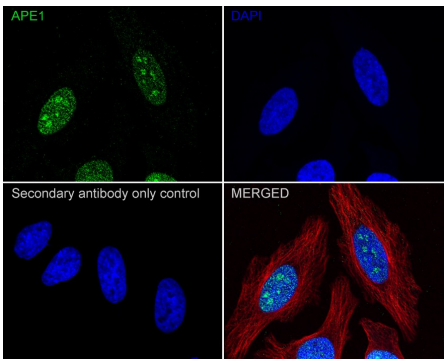

Fig4: Immunocytochemistry analysis of HeLa cells labeling APE1 with Mouse anti-APE1 antibody (HA601212) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Mouse anti-APE1 antibody (HA601212) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI. beta Tubulin (ET1602-4, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) were used as the secondary antibody at 1/1,000 dilution.

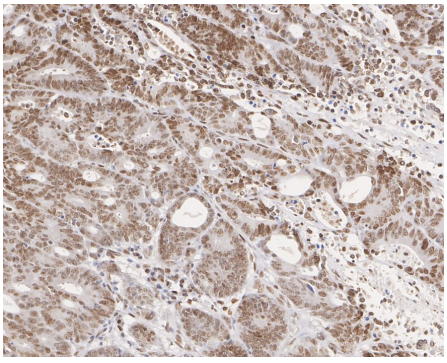

Fig5: Immunohistochemical analysis of paraffin-embedded human colon carcinoma tissue with Mouse anti-APE1 antibody (HA601212) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA601212) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

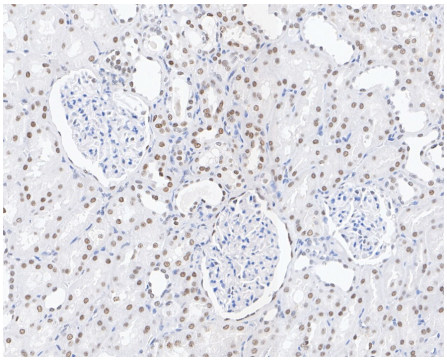

Fig6: Immunohistochemical analysis of paraffin-embedded human kidney tissue with Mouse anti-APE1 antibody (HA601212) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA601212) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

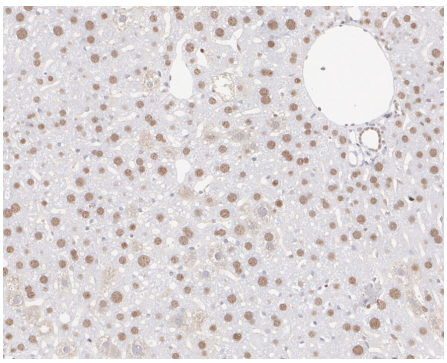

Fig7: Immunohistochemical analysis of paraffin-embedded mouse liver tissue with Mouse anti-APE1 antibody (HA601212) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA601212) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

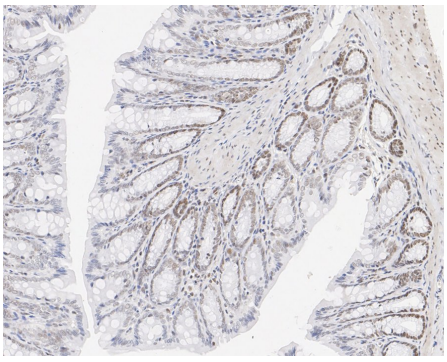

Fig8: Immunohistochemical analysis of paraffin-embedded rat colon tissue with Mouse anti-APE1 antibody (HA601212) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA601212) at 1/200 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

Background References

1. Robson C N et al. Isolation of cDNA clones encoding a human apurinic/apyrimidinic endonuclease that corrects DNA repair and mutagenesis defects in E. coli xth (exonuclease III) mutants. Nucleic Acids Res 19:5519-5523 (1991).
2. Fan Z et al. Cleaving the oxidative repair protein Ape1 enhances cell death mediated by granzyme A. Nat Immunol 4:145-153 (2003).