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Anti-APE1 APEX1 Antibody Picoband™ (monoclonal, 5C11)

Catalog # ABO14336

Product Information

Application	WB, IHC, ICC, FC
Primary Accession	P27695
Host	Mouse
Isotype	Mouse IgG2b
Reactivity	Human
Clonality	Monoclonal
Format	Lyophilized
Description	Anti-APE1 APEX1 Antibody Picoband™ (monoclonal, 5C11) . Tested in Flow Cytometry, IHC, ICC, WB applications. This antibody reacts with Human.
Reconstitution	Add 0.2ml of distilled water will yield a concentration of 500ug/ml.

Additional Information

Gene ID	328
Other Names	DNA repair nuclease/redox regulator APEX1, 3.1.11.2, 3.1.21.-, APEX nuclease, APEN, Apurinic-apyrimidinic endonuclease 1, AP endonuclease 1, APE-1, DNA-(apurinic or apyrimidinic site) endonuclease, Redox factor-1, REF-1, DNA repair nuclease/redox regulator APEX1, mitochondrial, APEX1, APE, APE1, APEX, APX, HAP1, REF1
Calculated MW	35555
Application Details	Western blot, 0.1-0.5 □g/ml Immunohistochemistry (Paraffin-embedded Section), 0.5-1 □g/ml Immunohistochemistry (Frozen Section), 0.5-1 □g/ml Immunocytochemistry, 0.5-1 □g/ml Flow Cytometry, 1-3 □g/1x10^6 cells
Subcellular Localization	Nucleus. Nucleus, nucleolus. Nucleus speckle. Endoplasmic reticulum. Cytoplasm. Detected in the cytoplasm of B-cells stimulated to switch (By similarity). Colocalized with SIRT1 in the nucleus. Colocalized with YBX1 in nuclear speckles after genotoxic stress. Together with OGG1 is recruited to nuclear speckles in UVA-irradiated cells. Colocalized with nucleolin and NPM1 in the nucleolus. Its nucleolar localization is cell cycle dependent and requires active rRNA transcription. Colocalized with calreticulin in the endoplasmic reticulum. Translocation from the nucleus to the cytoplasm is stimulated in presence of nitric oxide (NO) and function in a CRM1-dependent manner, possibly as a consequence of demasking a nuclear export signal (amino acid position 64-80). S-nitrosylation at Cys-93 and Cys-310 regulates its

	nuclear-cytosolic shuttling. Ubiquitinated form is localized predominantly in the cytoplasm..
Source	Eukaryota
Contents	Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg NaN3.
Clone Names	Clone: 5C11
Immunogen	E.coli-derived human APE1 recombinant protein (Position: P2-L318). Human APE1 shares 94% and 93% amino acid (aa) sequence identity with mouse and rat APE1, respectively.
Cross Reactivity	No cross-reactivity with other proteins.
Storage	Store at -20˚C for one year from date of receipt. After reconstitution, at 4˚C for one month. It can also be aliquotted and stored frozen at -20˚C for six months. Avoid repeated freeze-thaw cycles.

Protein Information

Name	APEX1
Synonyms	APE, APE1, APEX, APX, HAP1, REF1
Function	Multifunctional protein that plays a central role in the cellular response to oxidative stress. The two major activities of APEX1 are DNA repair and redox regulation of transcriptional factors (PubMed:11118054, PubMed:11452037, PubMed:15831793, PubMed:18439621, PubMed:18579163, PubMed:21762700, PubMed:24079850, PubMed:8355688, PubMed:9108029, PubMed:9560228). Functions as an apurinic/apyrimidinic (AP) endodeoxyribonuclease in the base excision repair (BER) pathway of DNA lesions induced by oxidative and alkylating agents. Initiates repair of AP sites in DNA by catalyzing hydrolytic incision of the phosphodiester backbone immediately adjacent to the damage, generating a single-strand break with 5'-deoxyribose phosphate and 3'-hydroxyl ends. Also incises at AP sites in the DNA strand of DNA/RNA hybrids, single-stranded DNA regions of R-loop structures, and single-stranded RNA molecules (PubMed:15380100, PubMed:16617147, PubMed:18439621, PubMed:19123919, PubMed:19188445, PubMed:19934257, PubMed:20699270, PubMed:21762700, PubMed:24079850, PubMed:8932375, PubMed:8995436, PubMed:9804799). Operates at switch sites of immunoglobulin (Ig) constant regions where it mediates Ig isotype class switch recombination. Processes AP sites induced by successive action of AICDA and UNG. Generates staggered nicks in opposite DNA strands resulting in the formation of double-strand DNA breaks that are finally resolved via non-homologous end joining repair pathway (By similarity). Has 3'-5' exodeoxyribonuclease activity on mismatched deoxyribonucleotides at the 3' termini of nicked or gapped DNA molecules during short-patch BER (PubMed:11832948, PubMed:1719477). Possesses DNA 3' phosphodiesterase activity capable of removing lesions (such as phosphoglycolate and 8- oxoguanine) blocking the 3' side of DNA strand breaks (PubMed:15831793, PubMed:7516064). Also acts as an endoribonuclease involved in the control of single-stranded RNA metabolism. Plays a role in regulating MYC mRNA turnover by preferentially cleaving in between UA and CA dinucleotides of the MYC coding region determinant (CRD). In association with NMD1, plays a role in the rRNA quality control process during cell cycle progression (PubMed:19188445, PubMed:19401441, PubMed:21762700). Acts as a loading factor for POLB onto non-incised AP sites in DNA and stimulates the 5'-terminal deoxyribose 5'-phosphate (dRp)

excision activity of POLB (PubMed:9207062). Exerts reversible nuclear redox activity to regulate DNA binding affinity and transcriptional activity of transcriptional factors by controlling the redox status of their DNA-binding domain, such as the FOS/JUN AP-1 complex after exposure to IR (PubMed:10023679, PubMed:11118054, PubMed:11452037, PubMed:18579163, PubMed:8355688, PubMed:9108029). Involved in calcium-dependent down-regulation of parathyroid hormone (PTH) expression by binding to negative calcium response elements (nCaREs). Together with HNRNPL or the dimer XRCC5/XRCC6, associates with nCaRE, acting as an activator of transcriptional repression (PubMed:11809897, PubMed:14633989, PubMed:8621488). May also play a role in the epigenetic regulation of gene expression by participating in DNA demethylation (PubMed:21496894). Stimulates the YBX1-mediated MDR1 promoter activity, when acetylated at Lys-6 and Lys-7, leading to drug resistance (PubMed:18809583). Plays a role in protection from granzyme-mediated cellular repair leading to cell death (PubMed:18179823). Binds DNA and RNA. Associates, together with YBX1, on the MDR1 promoter. Together with NPM1, associates with rRNA (PubMed:19188445, PubMed:19401441, PubMed:20699270).

Cellular Location

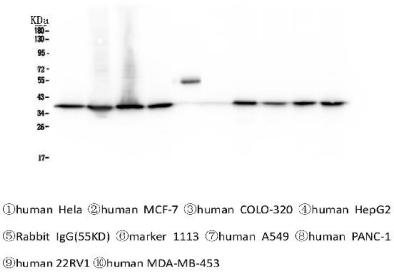
Nucleus {ECO:0000255|PROSITE-ProRule:PRU00764}. Nucleus, nucleolus. Nucleus speckle. Endoplasmic reticulum. Cytoplasm Note=Detected in the cytoplasm of B-cells stimulated to switch (By similarity). Colocalized with SIRT1 in the nucleus. Colocalized with YBX1 in nuclear speckles after genotoxic stress. Together with OGG1 is recruited to nuclear speckles in UVA-irradiated cells. Colocalized with nucleolin and NPM1 in the nucleolus. Its nucleolar localization is cell cycle dependent and requires active rRNA transcription. Colocalized with calreticulin in the endoplasmic reticulum. Translocation from the nucleus to the cytoplasm is stimulated in presence of nitric oxide (NO) and function in a CRM1-dependent manner, possibly as a consequence of demasking a nuclear export signal (amino acid position 64-80). S- nitrosylation at Cys-93 and Cys-310 regulates its nuclear-cytosolic shuttling. Ubiquitinated form is localized predominantly in the cytoplasm.

Background

APEX1, also called apurinic endonuclease (APE), is a DNA repair enzyme having apurinic/apyrimidinic (AP) endonuclease, 3-prime, 5-prime-exonuclease, DNA 3-prime repair diesterase, and DNA 3-prime-phosphatase activities. The human APEX1 gene consists of 5 exons spanning 2.64 kb and exists as a single copy in the haploid genome. Using in situ hybridization, the APEX1 gene is mapped to 14q11.2-q12. The predicted APEX1 protein, which contained probable nuclear transport signals, was identified as a member of a family of DNA repair enzymes found in lower organisms. The abundance of the large form of APEX1 was increased in leiomyoma extracts relative to myometrial tissue extracts, and the large form was dominant in cell lines derived from leiomyosarcomas. The exonuclease activity of nuclear APEX1 can remove the anti-HIV nucleoside analogs AZT and D4T from the 3-prime terminus of a nick more efficiently than can cytosolic exonucleases.

Images

Figure 1. Western blot analysis of APE1 using anti-APE1 antibody (M00627).
Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.
Lane 1: human Hela whole cell lysates,
Lane 2: human MCF-7 whole cell lysates,



①human Hela ②human MCF-7 ③human COLO-320 ④human HepG2
⑤Rabbit IgG(55KD) ⑥marker 1113 ⑦human A549 ⑧human PANC-1
⑨human 22RV1 ⑩human MDA-MB-453

Lane 3: human COLO-320 whole cell lysates,
Lane 4: human HepG2 whole cell lysates,
Lane 5: Rabbit IgG,
Lane 6: Marker 1113,
Lane 7: human A549 whole cell lysates ,
Lane 8: human PANC-1 whole cell lysates,
Lane 9: human 22RV1 whole cell lysates,
Lane 10: human MDA-MB-453 whole cell lysates.
After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-APE1 antigen affinity purified monoclonal antibody (Catalog # M00627) at 0.5 µg/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-mouse IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1001) with Tanon 5200 system.

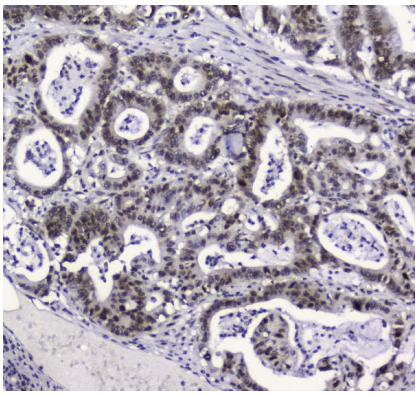


Figure 2. IHC analysis of APE1 using anti-APE1 antibody (M00627).
APE1 was detected in paraffin-embedded section of human intestinal cancer tissue. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 µg/ml mouse anti-APE1 Antibody (M00627) overnight at 4°C. Biotinylated goat anti-mouse IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Strepavidin-Biotin-Complex (SABC)(Catalog # SA1021) with DAB as the chromogen.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.