

Anti-DNA PKcs pT 2609 (RABBIT) Antibody

DNA PKcs phosphoT2609 Antibody Catalog # ASR5260

Specification

Anti-DNA PKcs pT 2609 (RABBIT) Antibody - Product Information

Host Conjugate Target Species Reactivity Clonality Application Application Note	Rabbit Unconjugated Human Polyclonal WB, E, IP, I, LCI Anti-DNA PKcs pT2609 antibody has been tested for use in ELISA, western blot, and IP. Specific conditions for reactivity should be optimized by the end user. Expect a band approximately 460 kDa in size corresponding to DNA PKcs by western blotting in the appropriate cell lysate or extract. Alternate splice variants have been described for this protein. Best western blotting results are seen when IP is performed prior to detection. This antibody detects an inducible signal at the correct height after DNA damage. However, the antibody may also detect an IR-inducible signal in cells lacking DNA PKcs (MO59J cells) at the same size as DNA PKcs if western blotting is performed directly. We believe that this additional band is 53BP1 that runs at the same size as DNA PKcs and is also phosphorylated in an IR-dependent manner. Similar results were seen with another phospho DNA PKcs
Dhusiaal Chata	antibody indicating that this result is general to antibodies to this phospho site, rather than specific to this antibody.
Buffer	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Immunogen	Rabbit Anti-DNA PKcs pT2609 affinity purified antibody was prepared from whole rabbit serum produced by repeated immunizations with a synthetic peptide corresponding to amino acids surrounding Thr 2609 of human DNA PKcs
Preservative	0.01% (w/v) Sodium Azide

Anti-DNA PKcs pT 2609 (RABBIT) Antibody - Additional Information



Gene ID 5591

Other Names 5591

Purity

DNA PKcs pT2609 antibody is directed against the phosphorylated form of human DNA PKcs at the pT2609 residue. The product was affinity purified from monospecific antiserum by immunoaffinity purification. Antiserum was first purified against the phosphorylated form of the immunizing peptide. The resultant affinity purified antibody was then cross-adsorbed against the non-phosphorylated form of the immunizing peptide. This phospho-specific polyclonal antibody reacts with phosphorylated pT2609 of human DNA PKcs. Reactivity with non-phosphorylated human DNA PKcs is minimal. A BLAST analysis was used to suggest reactivity with this protein from human and chimpanzee based on 100% homology for the immunogen sequence. However, cross-reactivity is expected with mouse, rat, dog, chicken and horse DNA PKcs based on a high degree of homology to the immunogen sequence. Cross-reactivity with DNA PKcs homologues from other sources has not been determined.

Storage Condition

Store vial at -20° C prior to opening. Aliquot contents and freeze at -20° C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use.

Precautions Note

This product is for research use only and is not intended for therapeutic or diagnostic applications.

Anti-DNA PKcs pT 2609 (RABBIT) Antibody - Protein Information

Name PRKDC

Synonyms HYRC, HYRC1

Function

Serine/threonine-protein kinase that acts as a molecular sensor for DNA damage (PubMed:11955432, PubMed:12649176, PubMed:14734805, PubMed:33854234). Involved in DNA non-homologous end joining (NHEJ) required for double-strand break (DSB) repair and V(D)J recombination (PubMed:11955432, PubMed:12649176, PubMed:14734805, PubMed:33854234, PubMed:34352203). Must be bound to DNA to express its catalytic properties (PubMed:11955432). Promotes processing of hairpin DNA structures in V(D)J recombination by activation of the hairpin endonuclease artemis (DCLRE1C) (PubMed:11955432). Recruited by XRCC5 and XRCC6 to DNA ends and is required to (1) protect and align broken ends of DNA, thereby preventing their degradation, (2) and sequester the DSB for repair by NHEJ (PubMed:11955432, PubMed:12649176, PubMed:14734805, PubMed:<a href="http://www.uniprot.org/citations/15574326"



target=" blank">15574326, PubMed:33854234). Acts as a scaffold protein to aid the localization of DNA repair proteins to the site of damage (PubMed:11955432, PubMed:12649176, PubMed:14734805, PubMed:15574326). The assembly of the DNA-PK complex at DNA ends is also required for the NHEJ ligation step (PubMed:11955432, PubMed:12649176, PubMed:14734805, PubMed:15574326). Found at the ends of chromosomes, suggesting a further role in the maintenance of telomeric stability and the prevention of chromosomal end fusion (By similarity). Also involved in modulation of transcription (PubMed:11955432, PubMed:12649176, PubMed:14734805, PubMed:15574326). As part of the DNA-PK complex, involved in the early steps of ribosome assembly by promoting the processing of precursor rRNA into mature 18S rRNA in the small-subunit processome (PubMed:32103174). Binding to U3 small nucleolar RNA, recruits PRKDC and XRCC5/Ku86 to the small-subunit processome (PubMed:32103174). Recognizes the substrate consensus sequence [ST]-Q (PubMed:11955432, PubMed:12649176, PubMed:14734805, PubMed:15574326). Phosphorylates 'Ser-139' of histone variant H2AX, thereby regulating DNA damage response mechanism (PubMed:14627815, PubMed:16046194). Phosphorylates ASF1A, DCLRE1C, c-Abl/ABL1, histone H1, HSPCA, c-jun/JUN, p53/TP53, PARP1, POU2F1, DHX9, FH, SRF, NHEJ1/XLF, XRCC1, XRCC4, XRCC5, XRCC6, WRN, MYC and RFA2 (PubMed:10026262, PubMed:10467406, PubMed:11889123, PubMed:12509254, PubMed:14599745, PubMed:14612514, PubMed:14704337, PubMed:15177042, PubMed:1597196, PubMed:16397295, PubMed:18644470, PubMed:2247066, PubMed:2507541, PubMed:26237645, PubMed:26666690, PubMed:28712728, PubMed:29478807, PubMed:30247612, PubMed:8407951, PubMed:8464713, PubMed:9139719, PubMed:9362500). Can phosphorylate C1D not only in the presence of linear DNA but also in the presence of supercoiled DNA (PubMed:<a



href="http://www.uniprot.org/citations/9679063" target=" blank">9679063). Ability to phosphorylate p53/TP53 in the presence of supercoiled DNA is dependent on C1D (PubMed:9363941). Acts as a regulator of the phosphatidylinositol 3-kinase/protein kinase B signal transduction by mediating phosphorylation of 'Ser-473' of protein kinase B (PKB/AKT1, PKB/AKT2, PKB/AKT3), promoting their activation (PubMed: 15262962). Contributes to the determination of the circadian period length by antagonizing phosphorylation of CRY1 'Ser-588' and increasing CRY1 protein stability, most likely through an indirect mechanism (By similarity). Plays a role in the regulation of DNA virusmediated innate immune response by assembling into the HDP-RNP complex, a complex that serves as a platform for IRF3 phosphorylation and subsequent innate immune response activation through the cGAS-STING pathway (PubMed:28712728). Also regulates the cGAS-STING pathway by catalyzing phosphorylation of CGAS, thereby impairing CGAS oligomerization and activation (PubMed:33273464). Also regulates the cGAS-STING pathway by mediating phosphorylation of PARP1 (PubMed:35460603).

Cellular Location Nucleus. Nucleus, nucleolus. Cytoplasm, cytosol

Anti-DNA PKcs pT 2609 (RABBIT) Antibody - Protocols

Provided below are standard protocols that you may find useful for product applications.

- <u>Western Blot</u>
- Blocking Peptides
- Dot Blot
- Immunohistochemistry
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- <u>Cell Culture</u>

Anti-DNA PKcs pT 2609 (RABBIT) Antibody - Images



Western blot using Rockland's Affinity Purified anti-DNAPKcs antibody shows detection of a 460 kDa band corresponding to human DNAPKcs in various preparations. Lane 1: Fus1 untreated, Lane 2: Fus1 IR (20Gy, 4h), Lane 3: Fus1 DNAPK inhibitor + IR, Lane 4: MO59J (DNAPK-) untreated, Lane 5: MO59J IR, Lane 6: Fus1 untreated, Lane 7: Fus1 IR (20Gy, 4h), Lane 8: Fus1 DNAPK inhibitor + IR, Lane 9: MO59J untreated, Lane 10: MO59J IR. Lanes 1-5 are nuclear extract whereas lanes



6-10 are whole cell lysates. MO59J is a cell line that lacks DNA-PKcs. FUS1 is the matched cell line complemented with a chromosomal fragment containing the DNA-PKcs gene. Approximately 20 µg of lysate was run on SDS-PAGE and transferred onto nitrocellulose, followed by reaction with a 1:1,000 dilution of anti-DNAPKcs antibody. Detection occurred using a 1:5,000 dilution of HRP-labeled Goat anti-Rabbit IgG for 1 hour at room temperature. A chemiluminescence system was used for signal detection (Roche) using a 1 min exposure time.

Anti-DNA PKcs pT 2609 (RABBIT) Antibody - Background

DNA dependent Protein Kinase (also called DNAPK, DNPK1, HYRC1, Protein Kinase DNA Activated Catalytic Polypeptide, XRCC7 and P460) consists of the 460 kDa DNA PKcs and a heterodimeric regulatory complex comprised of p70 Ku and p80 Ku (Ku autoantigen). DNA PKcs is a nuclear protein serine/threonine kinase present in a wide variety of eukaryotic species. DNA PKcs phosphorylates transcription factors, Sp1, Oct-1, p53 and SV40 large T antigen. DNA PKcs is involved in repairing double stranded DNA breaks. At the onset of apoptosis, DNA PKcs is rapidly inactivated by cleavage of the catalytic subunit into smaller polypeptides. Proteolysis of DNA PKcs is inhibited by the cysteine protease inhibitors iodoacetamide and N-ethylmaleimide. Alternative splicing can occur for this protein to produce at least two isoforms. Rabbit Anti-DNA PKcs pT2609 Antibody is useful for researchers interested in DNA damage.