abcam

Product datasheet

Anti-DNA PKcs antibody [EPR392] - BSA and Azide free ab174573

KO VALIDATED | Recombinant | RabMAb

4 Images

Overview

Product name	Anti-DNA PKcs antibody [EPR392] - BSA and Azide free
Description	Rabbit monoclonal [EPR392] to DNA PKcs - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** WB, ICC/IF **Unsuitable for:** Flow Cyt,IHC-P or IP
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide within Human DNA PKcs aa 50-150. The exact sequence is proprietary. Database link: P78527
Positive control	WB: K562, Molt4, MCF7, SH-SY5Y, 293T and PC3 cell lysates. ICC/IF: K-562 cells
General notes	Ab174573 is the carrier-free version of ab133516. This format is designed for use in antibody labeling, including fluorochromes, metal isotopes, oligonucleotides, enzymes. Our carrier-free formats are supplied in a buffer free of BSA, sodium azide and glycerol for higher conjugation efficiency. Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold. ab174573 is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm. *Maxpar® is a trademark of Fluidigm Canada Inc.* Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents. **We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.** Reproducibility is key to advancing scientific discovery and accelerating scientists' next breakthrough.

Abcam is leading the way with our range of recombinant antibodies, knockout-validated antibodies and knockout cell lines, all of which support improved reproducibility.

We are also planning to innovate the way in which we present recommended applications and species on our product datasheets, so that only applications & species that have been tested in our own labs, our suppliers or by selected trusted collaborators are covered by our Abpromise™ guarantee.

In preparation for this, we have started to update the applications & species that this product is Abpromise guaranteed for.

We are also updating the applications & species that this product has been "predicted to work with," however this information is not covered by our Abpromise guarantee.

Applications & species from publications and Abreviews that have not been tested in our own labs or in those of our suppliers are not covered by the Abpromise guarantee.

Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, as well as customer reviews and Q&As.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR392
Isotype	IgG

Applications

Our Abpromise guarantee covers the use of **ab174573** in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		Use at an assay dependent concentration. Detects a band of approximately 460 kDa (predicted molecular weight: 469 kDa).
ICC/IF		Use at an assay dependent concentration.

Application notes	Is unsuitable for Flow Cyt,IHC-P or IP.

Target

Function	Serine/threonine-protein kinase that acts as a molecular sensor for DNA damage. Involved in

DNA nonhomologous end joining (NHEJ) required for double-strand break (DSB) repair and V(D)J recombination. Must be bound to DNA to express its catalytic properties. Promotes processing of hairpin DNA structures in V(D)J recombination by activation of the hairpin endonuclease artemis (DCLRE1C). The assembly of the DNA-PK complex at DNA ends is also required for the NHEJ ligation step. Required to protect and align broken ends of DNA. May also act as a scaffold protein to aid the localization of DNA repair proteins to the site of damage. Found at the ends of chromosomes, suggesting a further role in the maintenance of telomeric stability and the prevention of chromosomal end fusion. Also involved in modulation of transcription. Recognizes the substrate consensus sequence [ST]-Q. Phosphorylates 'Ser-139' of histone variant H2AX/H2AFX, thereby regulating DNA damage response mechanism. Phosphorylates DCLRE1C, c-Abl/ABL1, histone H1, HSPCA, c-jun/JUN, p53/TP53, PARP1, POU2F1, DHX9, SRF, XRCC1, XRCC1, XRCC4, XRCC5, XRCC6, WRN, MYC and RFA2. Can phosphorylate C1D not only in the presence of linear DNA but also in the presence of supercoiled DNA. Ability to phosphorylate p53/TP53 in the presence of supercoiled DNA is dependent on C1D.

Sequence similarities

Belongs to the PI3/PI4-kinase family.
Contains 1 FAT domain.
Contains 1 FATC domain.
Contains 2 HEAT repeats.
Contains 1 PI3K/PI4K domain.
Contains 3 TPR repeats.

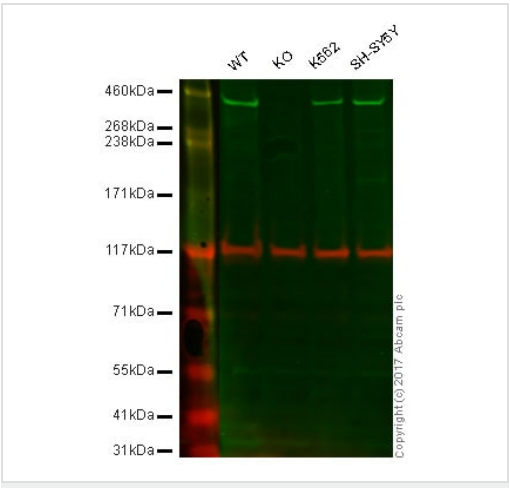
Post-translational modifications

Phosphorylated upon DNA damage, probably by ATM or ATR. Autophosphorylated on Thr-2609, Thr-2638 and Thr-2647. Thr-2609 is a DNA damage-inducible phosphorylation site (inducible with ionizing radiation, IR). Autophosphorylation induces a conformational change that leads to remodeling of the DNA-PK complex, requisite for efficient end processing and DNA repair. S-nitrosylated by GAPDH.

Cellular localization

Nucleus.

Images



Western blot - Anti-DNA PKcs antibody [EPR392] - BSA and Azide free (ab174573)

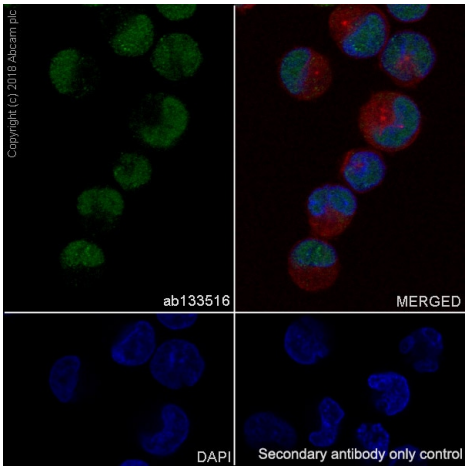
This WB data was generated using the same anti-DNA PKcs antibody clone, EPR392, in a different buffer formulation (cat# ab133516).

Lane 1: Wild type HAP1 whole cell lysate (40 µg)
Lane 2: DNA PKcs knockout HAP1 whole cell lysate (40 µg)
Lane 3: K562 whole cell lysate (20 µg)
Lane 4: SHSY5Y whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - ab133516 observed at 460 kDa. Red - loading control, ab18058, observed at 130 kDa.

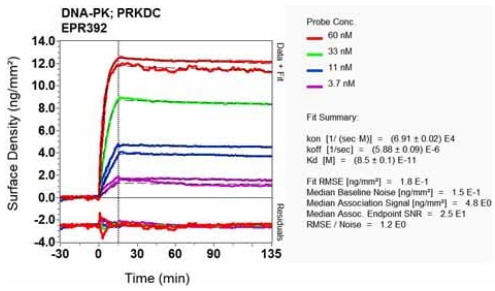
ab133516 was shown to specifically react with DNA PKcs when DNA PKcs knockout samples were used. Wild-type and DNA PKcs knockout samples were subjected to SDS-PAGE. Ab133516 and ab18058 (Mouse anti Vinculin loading control) were incubated overnight at 4°C at 1000 dilution and 1/10000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L

(IRDye® 800CW) preabsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed ab216776 secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-DNA PKcs antibody [EPR392] - BSA and Azide free (ab174573)

Immunocytochemistry/ Immunofluorescence analysis of K-562 (Human chronic myelogenous leukemia lymphoblast) cells labeling DNA PKcs with purified ab133516 at 1:200 dilution (9.3 µg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 µg/ml). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) was used as the secondary antibody at 1:1000 (2 µg/ml) dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



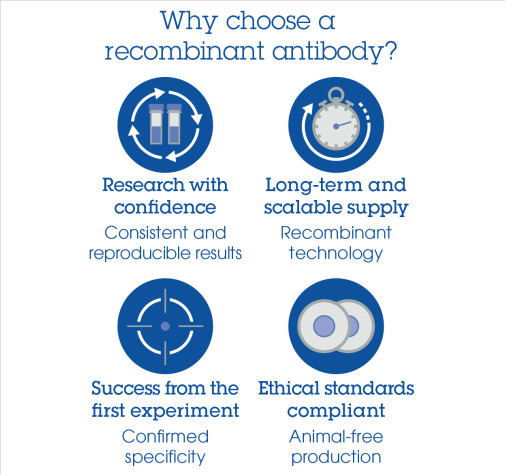
OI-RD Scanning - Anti-DNA PKcs antibody [EPR392] - BSA and Azide free (ab174573)

This kD data was generated using the same anti-DNA PKcs antibody clone, EPR392, in a different buffer formulation (cat# ab133516).

Equilibrium disassociation constant (K_D)

Learn more about K_D

Click here to learn more about K_D



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Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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