

DNA-PKcs monoclonal antibody

Catalog: MB66666

Host: Mouse

Reactivity: Human

BackGround:

DNA-dependent protein kinase (DNA-PK) is an important factor in the repair of double-stranded breaks in DNA. Cells lacking DNA-PK or in which DNA-PK is inhibited fail to show proper nonhomologous end-joining (NHEJ). DNA-PK is composed of two DNA-binding subunits (Ku70 and Ku86) and one 450 kDa catalytic subunit (DNA-PKcs). It is thought that a heterodimer of Ku70 and Ku86 binds to double-stranded DNA broken ends before DNA-PKcs binds and is activated. Activated DNA-PKcs is a serine/threonine kinase that has been shown to phosphorylate a number of proteins in vitro, including p53, transcription factors, RNA polymerase, and Ku70/Ku86. DNA-PKcs autophosphorylation at multiple sites, including Thr2609 and Ser2056, results in an inactivation of DNA-PK kinase activity and NHEJ ability. It has been demonstrated, however, that DNA-PK preferentially phosphorylates substrates before it autophosphorylates, suggesting that DNA-PK autophosphorylation may play a role in disassembly of the DNA repair machinery. Autophosphorylation at Thr2609 has also been shown to be required for DNA-PK-mediated double-strand break repair, and phosphorylated DNA-PK co-localizes with H2A.X and 53BP1 at sites of DNA damage. Phosphorylation at Ser2056 occurs in response to double-stranded DNA breaks and ATM activation.

Product:

Liquid in PBS containing 50% glycerol, 0.5% BSA and 0.02% sodium azide, pH 7.3.

Molecular Weight:

~ 450 kDa

Swiss-Prot:

P78527

Purification&Purity:

The antibody was purified by immunogen affinity chromatography.

Applications:

WB (1/500 - 1/1000), IHC (1/50 - 1/100), IF/ICC (1/50 - 1/100), IP (1/10 - 1/50)

Storage&Stability:

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

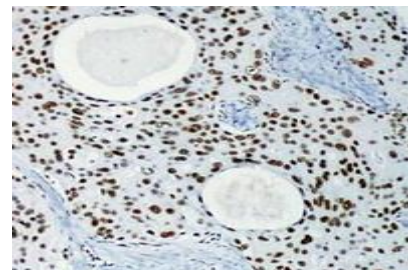
Specificity:

Recognizes endogenous levels of DNA-PKcs protein.

DATA:



Western blot analysis of DNA-PKcs expression in HeLa (A), K562 (B) whole cell lysates.



Immunohistochemical analysis of DNA-PKcs staining in human breast carcinoma formalin fixed paraffin embedded tissue section. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.35). The section was then incubated with the antibody at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

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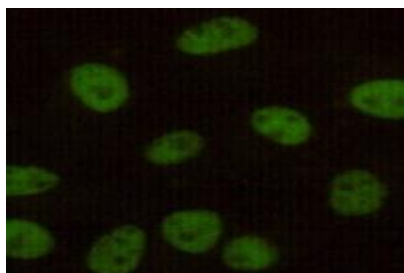
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Immunofluorescent analysis of DNA-PKcs staining in HeLa cells. For-

malin-fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 5-10 minutes and blocked with 3% BSA-PBS for 30 minutes at room temperature. Cells were probed with the primary antibody in 3% BSA-PBS and incubated overnight at 4 °C in a humidified chamber. Cells were washed with PBST and incubated with a AF488-conjugated secondary antibody (green) in PBS at room temperature in the dark.

Note:

For research use only, not for use in diagnostic procedure.

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