

Histone H2A.X (Phospho-S139) monoclonal antibody

Catalog: MB66691

Host: N

Mouse

Reactivity: Human, Mouse

BackGround:

Histone H2A.X is a variant histone that represents approximately 10% of the total H2A histone proteins in normal human fibroblasts. H2A.X is required for checkpoint-mediated cell cycle arrest and DNA repair following double-stranded DNA breaks. DNA damage, caused by ionizing radiation, UV-light, or radiomimetic agents, results in rapid phosphorylation of H2A.X at Ser139 by PI3K-like kinases, including ATM, ATR, and DNA-PK. Within minutes following DNA damage, H2A.X is phosphorylated at Ser139 at sites of DNA damage. This very early event in the DNA-damage response is required for recruitment of a multitude of DNA-damage response proteins, including MDC1, NBS1, RAD50, MRE11, 53BP1, and BRCA1. In addition to its role in DNA-damage repair, H2A.X is required for DNA fragmentation during apoptosis and is phosphorylated by various kinases in response to apoptotic signals. H2A.X is phosphorylated at Ser139 by DNA-PK in response to cell death receptor activation, c-Jun N-terminal Kinase (JNK1) in response to UV-A irradiation, and p38 MAPK in response to serum starvation. H2A.X is constitutively phosphorylated on Tyr142 in undamaged cells by WSTF (Williams-Beuren syndrome transcription factor). Upon DNA damage, and concurrent with phosphorylation of Ser139, Tyr142 is dephosphorylated at sites of DNA damage by recruited EYA1 and EYA3 phosphatases. While phosphorylation at Ser139 facilitates the recruitment of DNA repair proteins and apoptotic proteins to sites of DNA damage, phosphorylation at Tyr142 appears to determine which set of proteins are recruited. Phosphorylation of H2A.X at Tyr142 inhibits the recruitment of DNA repair proteins and promotes binding of pro-apoptotic factors such as JNK1. Mouse embryonic fibroblasts expressing only mutant H2A.X Y142F, which favors recruitment of DNA repair proteins over apoptotic

proteins, show a reduced apoptotic response to ionizing radiation. Thus, it appears that the balance of H2A.X Tyr142 phosphorylation and dephosphorylation provides a switch mechanism to determine cell fate after DNA damage.

Product:

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

Molecular Weight:

~ 15 kDa

Swiss-Prot:

P16104

Purification&Purity:

The antibody was purified by immunogen affinity chromatography.

Applications:

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

Storage&Stability:

Store at $4 \,^{\circ}{\rm C}$ short term. Aliquot and store at -20 $^{\circ}{\rm C}$ long term. Avoid freeze-thaw cycles.

Specificity:

Recognizes endogenous levels of Histone H2A.X (pS139) protein.

DATA:

130	~
95	
70	
55	
43	
33	
25	
17	-
10	

Western blot analysis of Histone H2A.X (pS139) expression in NIH3T3 treated with Hydroxyurea (A) whole cell lysates.

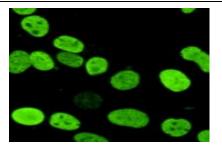
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PRODUCT DATA SHEET

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Immunofluorescent analysis of Histone H2A.X (pS139) staining in A549 cells. Formalin-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 $\,\,{}^\circ\!\!{\rm C}$ in a hidified 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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