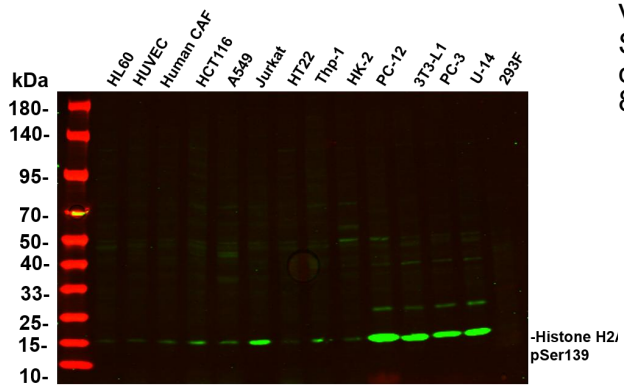
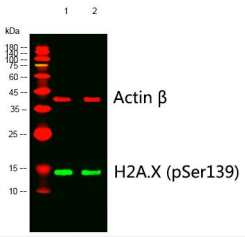
Histone H2A.X (Phospho Ser139) Rabbit mAb

Catalog No	YP-rAb-18473
Isotype	IgG
Reactivity	Human,Mouse,Rat,Hamster
Applications	WB,IHC,IF,IP,ELISA
Gene Name	H2AFX
Protein Name	Histone H2A.x
Purification Process	Protein A
Specificity	This antibody detects endogenous levels of Histone H2A.X only when phosphorylated at Ser139.This antibody does not recognize phosphorylated at other sites.
Formulation	PBS, 50% glycerol, 0.05% Proclin 300, 0.05%BSA
Source	Monoclonal, Rabbit,IgG
Dilution	IHC 1:2000-1:8000; WB 1:1000-1:5000; IF 1:200-1:1000; ELISA 1:5000-1:20000; IP 1:50-1:200, Note: For IHC, we suggest antigen retrieval with TE buffer pH 9.0
Concentration	0.5 mg/ml
Purity	≥90%
Storage Stability	-15° C to -25° C/1 year(Do not lower than -25° C)
Synonyms	H2AFX ; H2AX ; Histone H2A.x ; H2a/x, γ -H2AX,
Observed Band	15kD
Calculated Molecular Weight	15kD
Cell Pathway	Nucleus
Tissue Specificity	Lung,Placenta,
Function	developmental stage:Synthesized in G1 as well as in S-phase.,Domain:The [ST]-Q motif constitutes a recognition sequence for kinases from the PI3/PI4-kinase family.,Function:Variant histone H2A which replaces conventional H2A in a subset of nucleosomes. Nucleosomes wrap and compact DNA into chromatin, limiting DNA accessibility to the cellular machineries which require DNA as a template. Histones thereby play a central role in transcription regulation, DNA repair, DNA replication and chromosomal stability. DNA accessibility is regulated via a complex set of post-translational modifications of histones, also called histone code, and nucleosome remodeling. Required for checkpoint-mediated arrest of cell cycle progression in response to low doses of ionizing radiation and for efficient repair of DNA double strand breaks (DSBs) specifically when modified by C-terminal

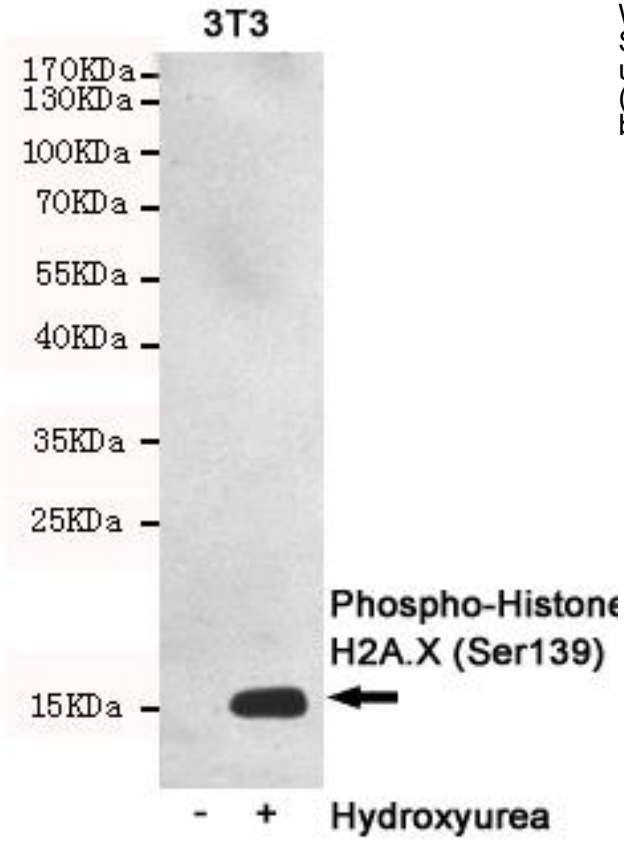
	phosphorylation.,PTM:Monoubiquitination of Lys-120 (H2AXK119ub) by RING1 and RNF2/RING2 complex gives a specific tag for epigenetic transcriptional repression. Following DNA double-strand breaks (DSBs), it is ubiquitinated through 'Lys-63' linkage of ubiquitin moieties by the E2 ligase UBE2N and the E3 ligases RNF8 and RNF168, leading to the recruitment of repair proteins to sites of DNA damage. Monoubiquitination and ionizing radiation-induced 'Lys-63'-linked ubiquitination are distinct events.,PTM:Phosphorylated on Ser-140 (to form gamma-H2AFX or H2AX139ph) in response to DNA double strand breaks (DSBs) generated by exogenous genotoxic agents and by stalled replication forks, and may also occur during meiotic recombination events and immunoglobulin class switching in lymphocytes. Phosphorylation can extend up to several thousand nucleosomes from the actual site of the DSB and may mark the surrounding chromatin for recruitment of proteins required for DNA damage signaling and repair. Widespread phosphorylation may also serve to amplify the damage signal or aid repair of persistent lesions. Phosphorylation of Ser-140 (H2AX139ph) in response to ionizing radiation is mediated by both ATM and PRKDC while defects in DNA replication induce Ser-140 phosphorylation (H2AX139ph) subsequent to activation of ATR and PRKDC. Dephosphorylation of Ser-140 by PP2A is required for DNA DSB repair. In meiosis, Ser-140 phosphorylation (H2AX139ph) may occur at synaptonemal complexes during leptotene as an ATM-dependent response to the formation of programmed DSBs by SPO11. Ser-140 phosphorylation (H2AX139ph) may subsequently occurs at unsynapsed regions of both autosomes and the XY bivalent during zygotene, downstream of ATR and BRCA1 activation. Ser-140 phosphorylation (H2AX139ph) may also be required for transcriptional repression of unsynapsed chromatin and meiotic sex chromosome inactivation (MSCI), whereby the X and Y chromosomes condense in pachytene to form the heterochromatic XY-body. During immunoglobulin class switch recombination in lymphocytes, Ser-140 phosphorylation (H2AX139ph) may occur at sites of DNA-recombination subsequent to activation of the activation-induced cytidine deaminase AICDA. Phosphorylation at Tyr-143 (H2AXY142ph) by BAZ1B/WSTF determines the relative recruitment of either DNA repair or pro-apoptotic factors. Phosphorylation at Tyr-143 (H2AXY142ph) favors the recruitment of APBB1/FE65 and pro-apoptosis factors such as MAPK8/JNK1, triggering apoptosis. In contrast, dephosphorylation of Tyr-143 by EYA proteins (EYA1, EYA2, EYA3 or EYA4) favors the recruitment of MDC1-containing DNA repair complexes to the tail of phosphorylated Ser-140 (H2AX139ph).,similarity:Belongs to the histone H2A family.,subunit:The nucleosome is a histone octamer containing two molecules each of H2A, H2B, H3 and H4 assembled in one H3-H4 heterotetramer and two H2A-H2B heterodimers. The octamer wraps approximately 147 bp of DNA. Interacts with numerous proteins required for DNA damage signaling and repair when phosphorylated on Ser-140. These include MDC1, TP53BP1, BRCA1 and the MRN complex, composed of MRE11A, RAD50, and NBN. Interaction with the MRN complex is mediated at least in part by NBN. Also interacts with DHX9/NDHII when phosphorylated on Ser-140.,
Background	Histones are basic nuclear proteins that are responsible for the nucleosome structure of the chromosomal fiber in eukaryotes. Two molecules of each of the four core histones (H2A, H2B, H3, and H4) form an octamer, around which approximately 146 bp of DNA is wrapped in repeating units, called nucleosomes. The linker histone, H1, interacts with linker DNA between nucleosomes and functions in the compaction of chromatin into higher order structures. This gene encodes a replication-independent histone that is a member of the histone H2A family, and generates two transcripts through the use of the conserved stem-loop termination motif, and the polyA addition motif. [provided by RefSeq, Oct 2015],
matters needing attention	Avoid repeated freezing and thawing!
Usage suggestions	This product can be used in immunological reaction related experiments. For more information, please consult technical personnel.



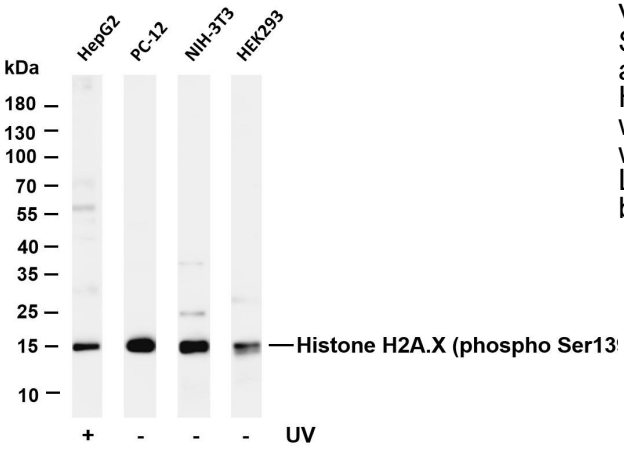
Various whole cell lysates were separated by 4-20% SDS-PAGE, and the primary antibody was used at 4℃, over night with a 1:2500 dilution . The Dylight 800-conjugated Goat anti-Rabbit antibody



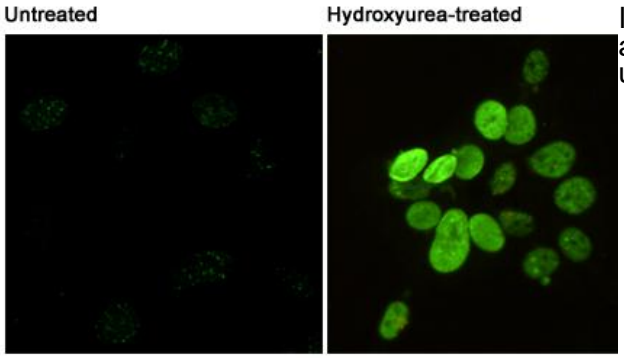
Western blot analysis of lysates from 1) 4T1, 2) 293 cells, (Green) primary antibody was diluted at 1:1000, 4° over night, Dylight 800 secondary antibodywas diluted at 1:10000, 37° 1hour. (Red) Actin β Monoclonal Antibody(5B7) antibody was diluted at 1:5000 as loading control, 4° over night,Dylight 680 secondary antibodywas diluted at 1:10000, 37° 1hour.



Western blot detection of Phosphorylation of H2A.X at Serine 139 in 3T3 or Hydroxyurea-treated 3T3 cell lysates using Phospho-Histone H2A.X (Ser139) mouse mAb (1:2000 diluted).Predicted band size:15KDa.Observed band size:15KDa.



Various whole cell lysates were separated by 4-20% SDS-PAGE, and the membrane was blotted with anti-Histone H2A.X (phospho Ser139) antibody. The HRP-conjugated Goat anti-Rabbit IgG(H + L) antibody was used to detect the antibody. Lane 1: HepG2 treated with UV for 30 minutes Lane 2: PC-12 Lane 3: NIH-3T3 Lane 4: HEK293 Predicted band size: 15kDa Observed band size: 15kDa



Immunofluorescent analysis of Phosphorylation of H2A.X at Serine 139 in 3T3 or Hydroxyurea-treated 3T3 cells using Phospho-Histone H2A.X