

# **Product datasheet**

# Alexa Fluor® 555 Anti-gamma H2A.X (phospho S139) antibody [EP854(2)Y] ab206900

Recombinant RabMAb

★★★★★ <u>2 Abreviews</u> 2 Images

Overview			
Product name	Alexa Fluor® 555 Anti-gamma H2A.X (phospho S139) antibody [EP854(2)Y]		
Description	Alexa Fluor® 555 Rabbit monoclonal [EP854(2)Y] to gamma H2A.X (phospho S139)		
Host species	Rabbit		
Conjugation	Alexa Fluor® 555. Ex: 555nm, Em: 565nm		
Tested applications	Suitable for: ICC/IF		
Species reactivity	Reacts with: Human		
	Predicted to work with: Mouse, Rat, Sheep 🛛 🔺		
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.		
Positive control	ICC/IF: Jurkat cells.		
General notes	Our RabMAb <sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to <b>RabMAb<sup>®</sup> patents</b> .		
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## Properties

Form

Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Stable for 12 months at -20°C. Store In the Dark.
Storage buffer	pH: 7.40 Preservative: 0.02% Sodium azide Constituents: 1% BSA, 30% Glycerol (glycerin, glycerine), PBS
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EP854(2)Y
lsotype	lgG

# Applications

The Abpromise guarantee Our <u>Abpromise guarantee</u> covers the use of ab206900 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
ICC/IF		1/100.

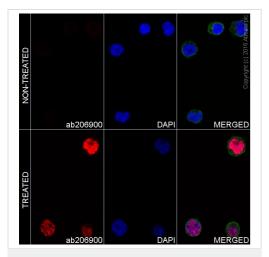
Target	
Function	Variant histone H2A which replaces conventional H2A in a subset of nucleosomes. Nucleosomes wrap and compact DNA into chromatin, limiting DNA accessibility to the cellular machineries which require DNA as a template. Histones thereby play a central role in transcription regulation, DNA 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.
Sequence similarities	Belongs to the histone H2A family.
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.
Post-translational modifications	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 proapoptosis 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 MDC1containing DNA repair complexes to the tail of phosphorylated Ser-140 (H2AX139ph). Monoubiguitination 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. Monoubiguitination and ionizing radiation-induced 'Lys-63'-linked ubiguitination are distinct events.

#### **Cellular localization**

Nucleus. Chromosome.

#### Images



Immunocytochemistry/ Immunofluorescence - Alexa Fluor® 555 Anti-gamma H2A.X (phospho S139) antibody [EP854(2)Y] (ab206900) ab206900 staining Histone H2A.X (phospho S139) in Jurkat cells. The cells were incubated with 25µM Etoposide for 5 hours (Treated) or solvent-only for control purposes (Non-treated). Cells were fixed with 4% formaldehyde (10min) and then blocked in 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab206900 at 1/100 dilution (shown in pseudo colour red) and <u>ab195887</u> Mouse monoclonal [DM1A] to alpha Tubulin - Microtubule Marker (Alexa Fluor<sup>®</sup> 488) at 2µg/ml (shown in green) overnight at +4°C. Nuclear DNA was labelled in blue with DAPI.



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