

Anti-Phospho-Histone H3 (Ser10), clone 3H10, FITC Conjugate

Monoclonal Antibody

Cat. # 16-222

Lot # DAM1479581

pack size: 100 µg

Store at -20°C

FOR RESEARCH USE ONLY



Certificate of Analysis

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Applications	Species Cross-Reactivity	Antibody Isotype	Epitope/Region	Host Species	Molecular Weight	Accession #
WB, IC	H	IgG	Ser10	M	17 kDa	NP_003484

Background

Histone H3 is one of the five main histone proteins involved in the structure of chromatin in eukaryotic cells. Featuring a main globular domain and a long N-terminal tail, H3 is involved with the structure of the nucleosomes of the 'beads on a string' structure. The N-terminal tail of histone H3 protrudes from the globular nucleosome core and can undergo several different types of epigenetic modifications that influence cellular processes. These modifications include the covalent attachment of methyl or acetyl groups to lysine and arginine amino acids and the phosphorylation of serine or threonine.

Presentation

Purified mouse monoclonal IgG in buffer containing PBS with 0.05% sodium azide, pH7.1.

Specificity

Recognizes histone H3 phosphorylated at Ser10, MW 17 kDa.

Species Cross-reactivity

Human. Broad species cross-reactivity is expected.

Immunogen

A proprietary immunogen based on a peptide sequence containing phospho-serine corresponding to residue 10 of human histone H3. Clone 3H10.

Molecular Weight

17 kDa

Method of Purification

Protein G purified.

Storage and Handling

Stable for 1 year at -20°C from date of receipt.

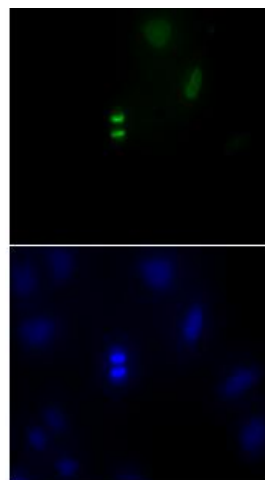
Handling Recommendations: Upon first thaw, and prior to removing the cap, centrifuge the vial and gently mix the solution. Aliquot into microcentrifuge tubes and store at -20°C. Avoid repeated freeze/thaw cycles, which may damage IgG and affect product performance.

Control

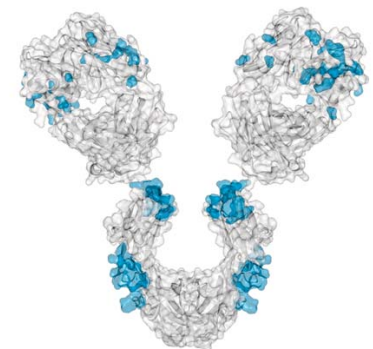
Mitotic HeLa cells (IF).

Quality Control Testing

Immunocytochemistry: Mitotic HeLa cells showed positive chromosome staining with 4µg/mL of this antibody.

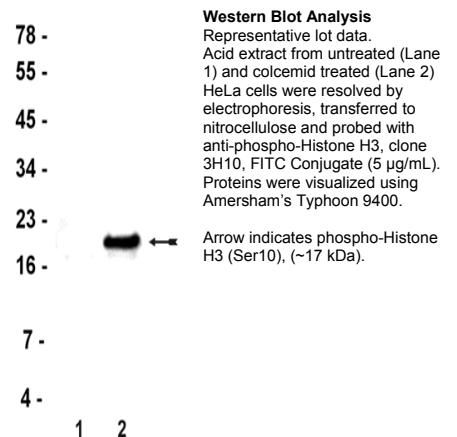


Immunocytochemistry
Representative lot data.
Mitotic HeLa cells were stained with 4 µg/mL of this antibody (green) and DAPI (blue).



Additional Research Applications

Western Blot Analysis: 0.5-5 µg/mL of this lot detected phosphorylated histone H3 in acid extracted proteins from mitotic HeLa cells (Catalog # 17-306) treated with colcemid.



APPLICATION LEGEND: WB Western Blotting IP Immunoprecipitation IC Immunocytochemistry IF Immunofluorescence
IH Immunohistochemistry (Tissue)

SPECIES LEGEND: H Human M Mouse R Rat Rb Rabbit WR Most Common Vertebrates

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PROTOCOL

Western Blot

1. Perform SDS-polyacrylamide gel electrophoresis (SDS-PAGE) on a cell lysate sample (cell lysis buffer: 50 mM Tris-HCl, pH 7.4; 1% NP-40; 0.25% sodium deoxycholate; 150 mM NaCl; 1 mM EDTA; 1 mM PMSF; 1 µg/mL each aprotinin, leupeptin, pepstatin; 1 mM Na3VO4; 1 mM NaF) and transfer the proteins to nitrocellulose.
2. Wet the blotted nitrocellulose in PBS for 5 minutes.
3. Block the blotted nitrocellulose in Odyssey® Blocking Buffer (Li-Cor, Catalog # 927-40000) for 1 hour at room temperature with constant agitation.
4. Incubate the nitrocellulose with **0.5-5 µg/mL of anti-phospho-Histone H3 (Ser10), clone 3H10, FITC Conjugate**, diluted in Odyssey® Blocking Buffer for 1 hour or longer with agitation at room temperature. Protect from light during incubation.
5. Wash the nitrocellulose 4 times for 5 minutes each at room temperature in PBS-0.05% Tween®-20 with agitation. Protect from light.
6. Rinse the nitrocellulose with PBS to remove residual Tween®-20. The membrane is now ready to scan.
7. Use detection method of choice (Li-Cor® Odyssey™ Infrared Imaging System or Amersham Biosciences Typhoon Imaging System).

Immunocytochemistry

1. Plate cells on coverslips in each well of a plate. Place the cells in a CO₂ incubator at 37°C for 24 hours.
2. Remove media and wash the cells with PBS by rinsing twice.
3. Add fixative (3.7% formaldehyde) in PBS for 20 minutes at room temperature. Wash two times with PBS for 5 minutes.
4. Permeabilize with 0.5% Triton X-100 for 2 minutes.
5. Wash the cells twice with PBS for 5 minutes.
6. Incubate the cells with 4 µg/mL of **anti-phospho-Histone H3 (Ser10), clone 3H10, FITC Conjugate** in PBS for 1 hour.
7. Wash the cells twice with PBS for 5 minutes.
8. Mount the coverslip to a slide and dry.
9. Examine the cells under a fluorescent microscope.

RELATED PRODUCTS (specific)

cat #	description
05-499	■ Anti-Histone H3
06-755	■ Anti-Histone H3
07-690	■ Anti-Histone H3, CT, pan
05-928	■ Anti-Histone H3, CT, pan, clone A3S
07-081	■ Anti-phospho (Ser10)-acetyl (Lys14)-Histone H3
07-554	■ Anti-phospho (Thr3)-monomethyl (Lys4) Histone H3
16-189	■ Anti-phospho-Histone H3 (Ser10), biotin conjugate
05-806	■ Anti-phospho-Histone H3 (Ser10), clone 3H10
16-218	■ Anti-Phospho-Histone H3 (Ser10), clone 3H10, Cy5 conjugate
05-817	■ Anti-phospho-Histone H3 (Ser10), clone MC463
05-598	■ Anti-phospho-Histone H3 (Ser10), clone RR002
14-411	■ Histone H3
12-357	■ Histone H3 Peptide
12-403	■ Histone H3 Peptide, biotin conjugate
12-404	■ Histone H3 Peptide, biotin conjugate, residues 21-44
14-494	■ Histone H3, human
14-494H	■ Histone H3, human
12-349	■ Goat Anti-Mouse IgG, HRP conjugate

RELATED PRODUCTS (non-specific)

cat #	description
IPVH00010	■ Immobilon-P 26.5 cm x 3.75 m Roll PVDF 0.45 µm
IPFL00010	■ Immobilon-FL 26.5 cm x 3.75 m Roll PVDF 0.45 µm
IPVH07850	■ Immobilon-P 7 x 8.4 cm PVDF 0.45 mm (sheet) 50/pk
ISEQ00010	■ Immobilon-P SQ 26.5 cm x 3.75 m 1 roll PVDF 0.2 µm
ISEQ07850	■ Immobilon-P 7 x 8.4 cm PVDF 0.2 mm (sheet) 50/pk
IPFL07810	■ Immobilon-FL 7 x 8.4 cm PVDF 0.45 mm (sheet) 10/pk
WBKLS0050	■ IMMOBILON WESTERN CHEMILUM HRP SUBSTRATE 50 mL
17-373SP	■ Spray & Glow™ ECL Western Blotting 40 mL
2060	■ Re-Blot Western Blot Recycling Kit
2500	■ Re-Blot Plus Western Blot Recycling Kit
B2080-175GM	■ Blot Quick Blocker Membrane Blocking Agent 175G
2170	■ CHEMIBLOCKER-1LT
20-200	■ IMMUNOBLOT BLOCKING REAGENT 20G
12-302	■ EGF-Stimulated A431 Cell Lysate
12-349	■ Goat Anti-Mouse IgG, HRP conjugate
12-110	■ Phosphotyrosine control (EGF-stim A431 cell lysate)

■ antibodies ■ Multiplex products ■ biotools ■ cell culture ■ enzymes ■ kits ■ proteins/peptides ■ siRNA/cDNA products

Please visit www.millipore.com for additional product information, test data and references

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