Anti-Histone H3 (acetyl K14) Antibody [JU43-26] - BSA and Azide free

HA750454

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, IP, SNAP-ChIP, CUT&Tag-seq
Molecular Wt:	Predicted band size: 15 kDa
Clone number:	JU43-26

Description: Core component of nucleosome. 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.

Immunogen: Synthetic peptide within Human Histone H3 aa 1-50 / 136 (acetyl K14) .

Positive control: Jurkat cell lysate, rat liver tissue lysate, mouse lung tissue lysate, Hela whole cell lysate, Hela treated with 1μM TSA for 18 hours whole cell lysate, F9, Hela, MCF-7, PC-3M, rat brain tissue, human kidney tissue, rat hippocampus tissue, human colon tissue, mouse testis tissue.

Subcellular location: Nucleus. Chromosome.

Database links: SwissProt: P68431 Human | P68433 Mouse | Q6LED0 Rat

Recommended Dilutions:

WB	1:500-1:2,000
IF-Cell	1:100
IF-Tissue	1:200
IHC-P	1:500
SNAP-ChIP	1:100
IP	1-2μg/sample

Storage Buffer: PBS (pH7.4).

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃ or -80℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

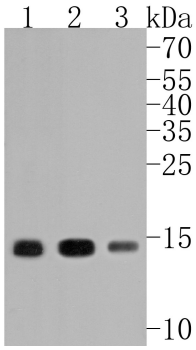

Fig1: Western blot analysis of Histone H3 (acetyl K14) on different lysates with Rabbit anti-Histone H3 (acetyl K14) antibody (HA750454) at 1/500 dilution.

Lane 1: Jurkat cell lysate(10 µg/Lane)
Lane 2: Rat liver tissue lysate
Lane 3: Mouse lung tissue lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 15 kDa
Observed band size: 15 kDa

Exposure time: 2 minutes;

15% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA750454) at 1/500 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 5,000 dilution was used for 1 hour at room temperature.

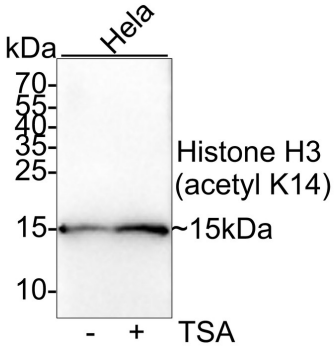

Fig2: Western blot analysis of Histone H3 (acetyl K14) on different lysates with Rabbit anti-Histone H3 (acetyl K14) antibody (HA750454) at 1/1,000 dilution.

Lane 1: Hela whole cell lysate
Lane 2: Hela treated with 1µM TSA for 18 hours whole cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 15 kDa
Observed band size: 15 kDa

Exposure time: 5 minutes;

10% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA750454) at 1/1,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:200,000 dilution was used for 1 hour at room temperature.

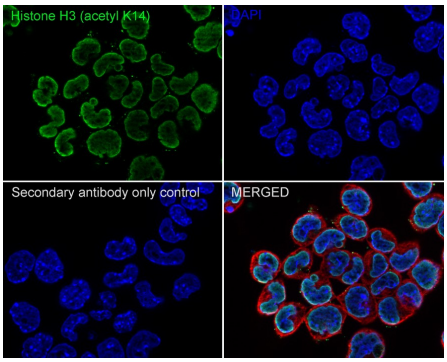

Fig3: Immunocytochemistry analysis of F9 cells labeling Histone H3 (acetyl K14) with Rabbit anti-Histone H3 (acetyl K14) antibody (HA750454) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-Histone H3 (acetyl K14) antibody (HA750454) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

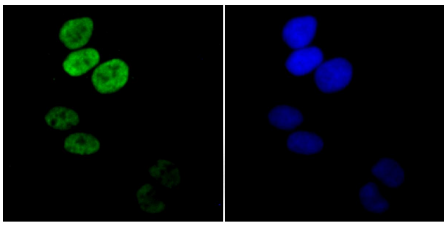

Fig4: Immunocytochemistry analysis of Hela cells labeling Histone H3 (acetyl K14) with Rabbit anti-Histone H3 (acetyl K14) antibody (HA750454) at 1/50 dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37 ℃, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-Histone H3 (acetyl K14) antibody (HA750454) at 1/50 dilution in 2% negative goat serum overnight at 4 ℃. Alexa Fluor®488 conjugate-Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. Nuclear DNA was labelled in blue with DAPI.

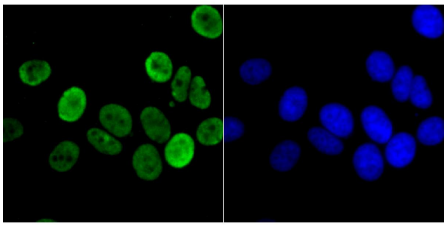

Fig5: Immunocytochemistry analysis of MCF-7 cells labeling Histone H3 (acetyl K14) with Rabbit anti-Histone H3 (acetyl K14) antibody (HA750454) at 1/50 dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37 ℃, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-Histone H3 (acetyl K14) antibody (HA750454) at 1/50 dilution in 2% negative goat serum overnight at 4 ℃. Alexa Fluor®488 conjugate-Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution.Nuclear DNA was labelled in blue with DAPI.

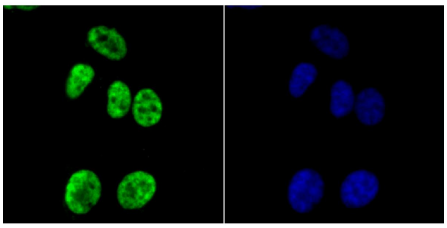

Fig6: Immunocytochemistry analysis of PC-3M cells labeling Histone H3 (acetyl K14) with Rabbit anti-Histone H3 (acetyl K14) antibody (HA750454) at 1/50 dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37 ℃, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-Histone H3 (acetyl K14) antibody (HA750454) at 1/50 dilution in 2% negative goat serum overnight at 4 ℃. Alexa Fluor®488 conjugate-Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution.Nuclear DNA was labelled in blue with DAPI.

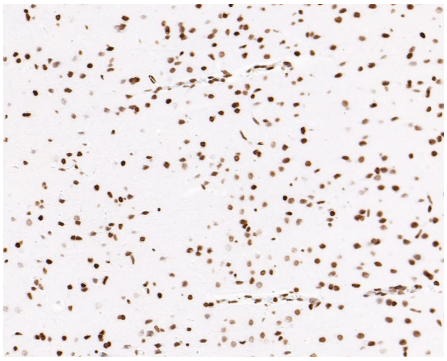

Fig7: Immunohistochemical analysis of paraffin-embedded rat brain tissue with Rabbit anti-Histone H3 (acetyl K14) antibody (HA750454) at 1/500 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750454) at 1/500 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

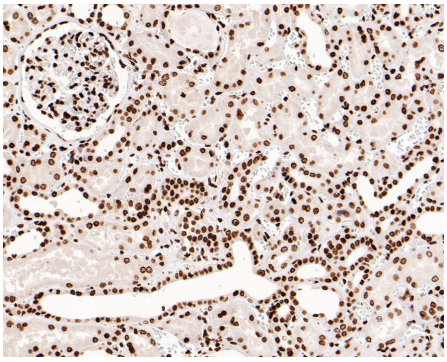

Fig8: Immunohistochemical analysis of paraffin-embedded human kidney tissue with Rabbit anti-Histone H3 (acetyl K14) antibody (HA750454) at 1/500 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750454) at 1/500 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

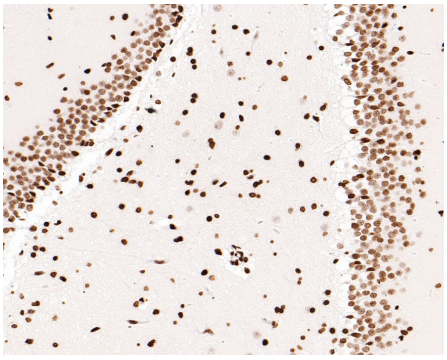

Fig9: Immunohistochemical analysis of paraffin-embedded rat hippocampus tissue with Rabbit anti-Histone H3 (acetyl K14) antibody (HA750454) at 1/500 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750454) at 1/500 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

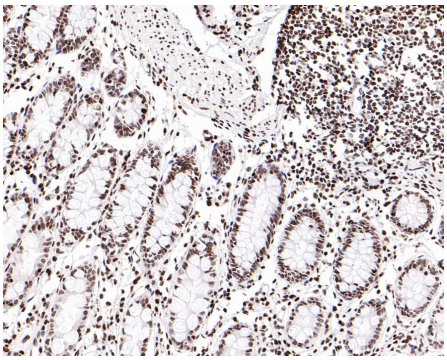

Fig10: Immunohistochemical analysis of paraffin-embedded human colon tissue with Rabbit anti-Histone H3 (acetyl K14) antibody (HA750454) at 1/500 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA750454) at 1/500 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

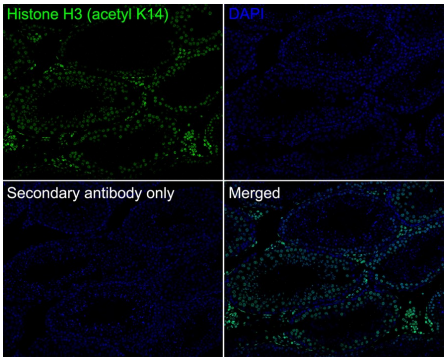

Fig11: Immunofluorescence analysis of paraffin-embedded mouse testis tissue labeling Histone H3 (acetyl K14) with Rabbit anti-Histone H3 (acetyl K14) antibody (HA750454) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA750454, green) at 1/200 dilution overnight at 4 ℃, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

Background References

1. Mandal P et al. H3 clipping activity of glutamate dehydrogenase is regulated by stefin B and chromatin structure. FEBS J 281:5292-308 (2014).
2. Anderson L et al. Histone deacetylase inhibition modulates histone acetylation at gene promoter regions and affects genome-wide gene transcription in Schistosoma mansoni. PLoS Negl Trop Dis 11:e0005539 (2017).