

Anti-Histone H3 Antibody [A11-D7]

M1309-1



Product Type:	Mouse monoclonal IgG2b, primary antibodies
Species reactivity:	Human, Mouse, Rat, Zebrafish
Applications:	WB, IF-Cell, IHC-P, IF-Tissue
Molecular Wt:	Predicted band size: 15 kDa
Clone number:	A11-D7

Description: The nucleosome, made up of DNA wound around eight core histone proteins (two each of H2A, H2B, H3, and H4), is the primary building block of chromatin. 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 N-terminal human Histone H3.

Positive control: HeLa cell lysate, A549 cell lysate, HT-29 cell lysate, HEK-293 cell lysate, C2C12 cell lysate, L-929 cell lysate, C6 cell lysate, zebrafish tissue lysates, HepG2 cell lysate, A431 cell lysate, MCF7 cell lysate, NIH/3T3 cell lysate, PC-12 cell lysate, Hela, F9, human skin tissue, human liver tissue, human testis tissue, mouse brain tissue, mouse testis tissue, rat testis tissue.

Subcellular location: Nucleus.

Database links: SwissProt: P68431 Human | P84243 Human | Q16695 Human | Q6NXT2 Human | Q71D13 Human | P68433 Mouse | P84228 Mouse | Q6LED0 Rat

Recommended Dilutions:

WB	1:5,000-1:10,000
IF-Cell	1:200-1:500
IHC-P	1:1,000
IF-Tissue	1:200

Storage Buffer: 1*PBS (pH7.4), 0.2% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

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

Purity: Protein A affinity purified.

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Orders:0086-571-88062880

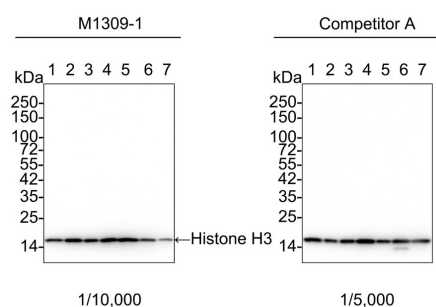
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Images

Fig1: Western blot analysis of Histone H3 on different lysates with Mouse anti-Histone H3 antibody (M1309-1) at 1/10,000 dilution and competitor's antibody at 1/5,000 dilution.



Lane 1: HeLa cell lysate
Lane 2: A549 cell lysate
Lane 3: HT-29 cell lysate
Lane 4: HEK-293 cell lysate
Lane 5: C2C12 cell lysate
Lane 6: L-929 cell lysate
Lane 7: C6 cell lysate

Lysates/proteins at 10 µg/Lane.

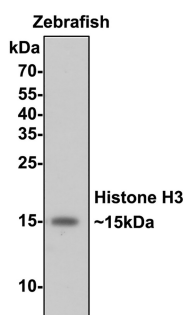
Predicted band size: 15 kDa
Observed band size: 15 kDa

Exposure time: 18 seconds;

4-20% 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 (M1309-1) at 1/10,000 dilution and competitor's antibody at 1/5,000 dilution were used in 5% NFDM/TBST at 4°C overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

Fig2: Western blot analysis of Histone H3 on zebrafish tissue lysates with Mouse anti-Histone H3 antibody (M1309-1) at 1/500 dilution.



Lysates/proteins at 10 µg/Lane.

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

Exposure time: 1 minute;

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 (M1309-1) at 1/500 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-mouse IgG - HRP Secondary Antibody (HA1006) at 1:150,000 dilution was used for 1 hour at room temperature.

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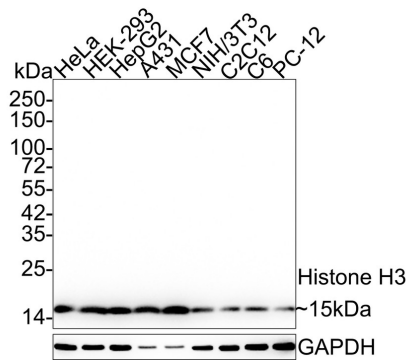
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Fig3: Western blot analysis of Histone H3 on different lysates with Mouse anti-Histone H3 antibody (M1309-1) at 1/5,000 dilution.



Lane 1: HeLa cell lysate (15 µg/Lane)
 Lane 2: HEK-293 cell lysate (15 µg/Lane)
 Lane 3: HepG2 cell lysate (15 µg/Lane)
 Lane 4: A431 cell lysate (15 µg/Lane)
 Lane 5: MCF7 cell lysate (15 µg/Lane)
 Lane 6: NIH/3T3 cell lysate (15 µg/Lane)
 Lane 7: C2C12 cell lysate (15 µg/Lane)
 Lane 8: C6 cell lysate (15 µg/Lane)
 Lane 9: PC-12 cell lysate (15 µg/Lane)

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

Exposure time: 17 seconds;

4-20% 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 (M1309-1) at 1/5,000 dilution was used in 5% NFDM/TBST at 4°C overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1:50,000 dilution was used for 1 hour at room temperature.

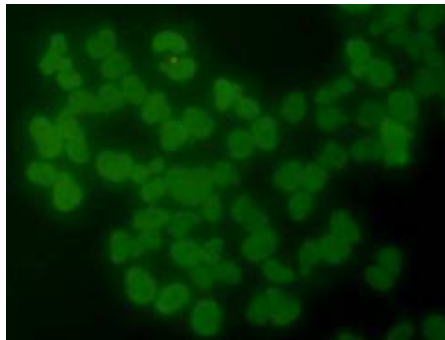


Fig4: ICC staining of Histone H3 in HeLa cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 10% negative goat serum for 15 minutes at room temperature. Cells were probed with the primary antibody (M1309-1, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 conjugate-Goat anti-Mouse IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

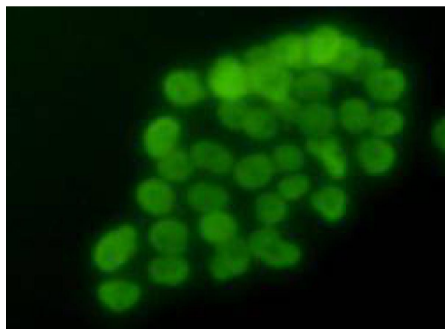


Fig5: ICC staining of Histone H3 in F9 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 10% negative goat serum for 15 minutes at room temperature. Cells were probed with the primary antibody (M1309-1, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 conjugate-Goat anti-Mouse IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

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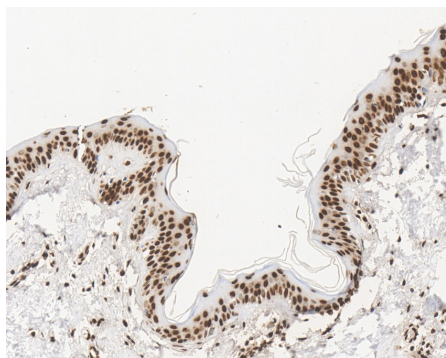


Fig6: Immunohistochemical analysis of paraffin-embedded human skin tissue with Mouse anti-Histone H3 antibody (M1309-1) at 1/1,000 dilution.

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 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (M1309-1) at 1/1,000 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.

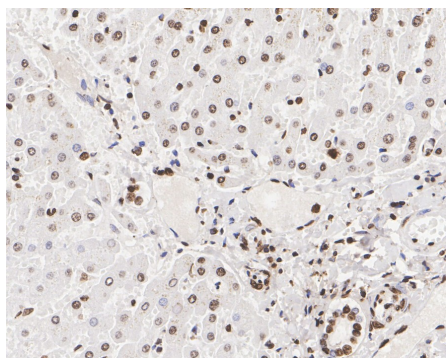


Fig7: Immunohistochemical analysis of paraffin-embedded human liver tissue with Mouse anti-Histone H3 antibody (M1309-1) at 1/1,000 dilution.

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 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (M1309-1) at 1/1,000 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.

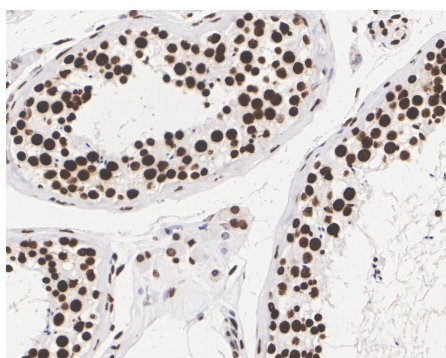


Fig8: Immunohistochemical analysis of paraffin-embedded human testis tissue with Mouse anti-Histone H3 antibody (M1309-1) at 1/1,000 dilution.

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 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (M1309-1) at 1/1,000 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.

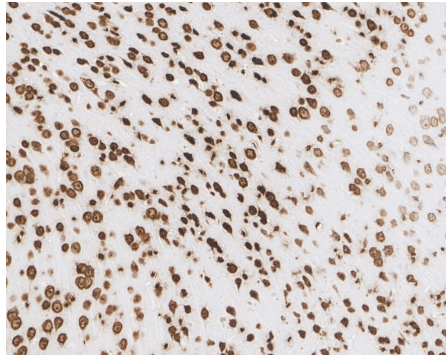


Fig9: Immunohistochemical analysis of paraffin-embedded mouse brain tissue with Mouse anti-Histone H3 antibody (M1309-1) at 1/1,000 dilution.

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 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (M1309-1) at 1/1,000 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.

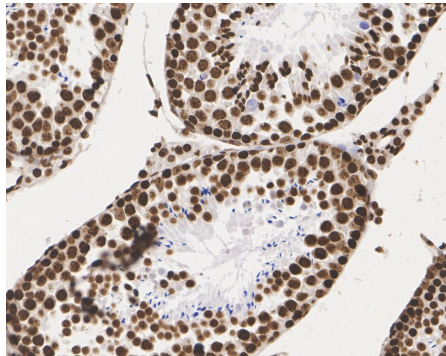


Fig10: Immunohistochemical analysis of paraffin-embedded mouse testis tissue with Mouse anti-Histone H3 antibody (M1309-1) at 1/1,000 dilution.

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 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (M1309-1) at 1/1,000 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.

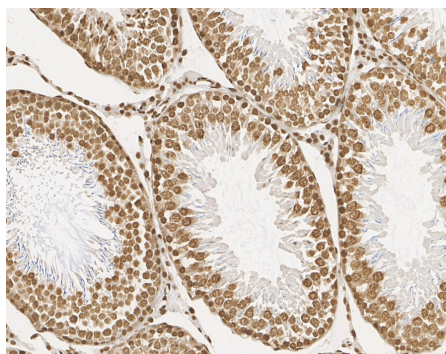


Fig11: Immunohistochemical analysis of paraffin-embedded rat testis tissue with Mouse anti-Histone H3 antibody (M1309-1) at 1/1,000 dilution.

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 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (M1309-1) at 1/1,000 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.

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. Flanagan J.F., Mi L.-Z., Chruszcz M., Cymborowski M., Clines K.L., Kim Y., Minor W., Rastinejad F., Khorasanizadeh S."Double chromodomains cooperate to recognize the methylated histone H3 tail."Nature 438:1181-1185(2005)
2. "Arginine methylation of the histone H3 tail impedes effector binding."Iberg A.N., Espejo A., Cheng D., Kim D., Michaud-Levesque J., Richard S., Bedford M.T.J. Biol. Chem. 283:3006-3010(2008)

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