abcam

ab308248

Anti-Histone H3 (unmodified K27) antibody [EPR23553-22]

Recombinant | RabMAb | Advanced Validation | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogens	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EPR23553-22
Purification technique	Affinity purification Protein A
Concentration	0.416 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

WB

Tested

Species	Human
Dilution info	1/1000
Notes	-

Species	Mouse
Dilution info	1/1000
Notes	-

Species	Rat
Dilution info	1/1000
Notes	-

Species	Recombinant fragment - Human
Dilution info	1/1000
Notes	-

Not recommended

Species	Synthetic peptide
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human
Dilution info	1/4000
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Species	Mouse
Dilution info	1/4000
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Species	Rat
Dilution info	1/4000
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Not recommended

Species	Recombinant fragment - Human, Synthetic peptide
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/200
Notes	-

Species	Mouse
Dilution info	1/200
Notes	-

Expected

Species	Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Not recommended

Species	Recombinant fragment - Human, Synthetic peptide
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/500

Notes	-

Species	Mouse
Dilution info	1/500
Notes	-

Expected

Species	Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Not recommended

Species	Recombinant fragment - Human, Synthetic peptide
Dilution info	-
Notes	-

IP

Not recommended

Species	Human, Mouse, Rat, Recombinant fragment - Human, Synthetic peptide
Dilution info	-
Notes	-

ChIP-seq

Tested

Species	Human
Dilution info	4 µg/cells
Notes	-

Expected

Species	Mouse, Rat

Dilution info	Use at an assay dependent concentration.
Notes	-

Not recommended

Species	Recombinant fragment - Human, Synthetic peptide
Dilution info	-
Notes	-

PepArr

Tested

Species	Synthetic peptide
Dilution info	0.1 µg/mL
Notes	-

Expected

Species	Human, Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Not recommended

Species	Recombinant fragment - Human
Dilution info	-
Notes	-

ChIC/CUT&RUN-seq

Tested

Species	Human
Dilution info	5 µg
Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Not recommended

Species	Recombinant fragment - Human, Synthetic peptide
Dilution info	-
Notes	-

Target data

See full target information H3C1 unmod K27

Function	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.

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

Patented technology
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?
This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free batch production

For more information, read more on recombinant antibodies.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

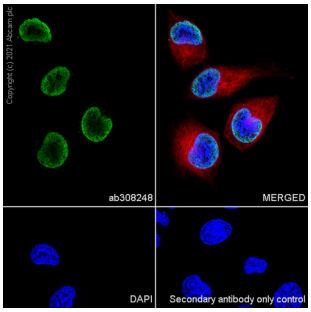
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

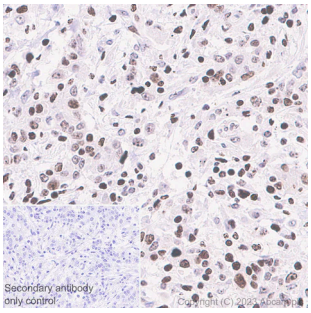
Full details and terms and conditions can be found here:
Terms & Conditions.

20 product images



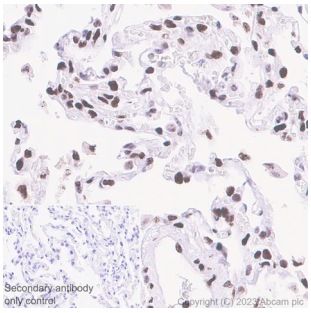
Immunocytochemistry/ Immunofluorescence - Anti-Histone H3 (unmodified K27) antibody [EPR23553-22] (ab308248)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized HeLa (human cervix adenocarcinoma epithelial cell) cells labelling Histone H3 (unmodified K27) with ab308248 at 1/200 (2.08 ug/ml) dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 2ug/ml dilution (Green). Confocal image showing nuclear staining in HeLa cells.Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8). ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 2.5ug/ml dilution (Red). The Nuclear counterstain was DAPI (Blue). Secondary antibody only control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 2ug/ml dilution.



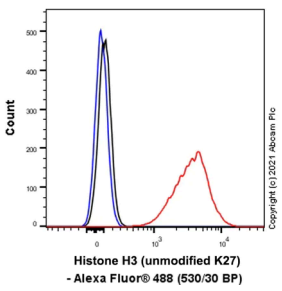
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Immunohistochemical analysis of paraffin-embedded Human gastric adenoc tissue labeling Histone H3 (unmodified K27) with ab308248 at 1/4000 (0.104 ug/ml) followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer). Nuclear staining on human gastric adenocarcinoma. The section was incubated with ab308248 at 4 ☒ overnight. Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer). Heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0)



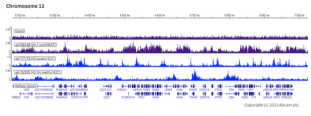
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H3 (unmodified K27) antibody [EPR23553-22] (ab308248)

Immunohistochemical analysis of paraffin-embedded Human lung tissue labeling Histone H3 (unmodified K27) with ab308248 at 1/4000 (0.104 ug/ml) followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer). Nuclear staining on human lung. The section was incubated with ab308248 at 4 ☒ overnight. Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer). Heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0)



Flow Cytometry (Intracellular) - Anti-Histone H3 (unmodified K27) antibody [EPR23553-22] (ab308248)

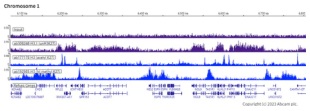
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ChIP-sequencing - Anti-Histone H3 (unmodified K27) antibody [EPR23553-22] (ab308248)

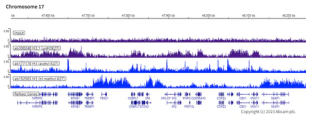
Chromatin was prepared from HeLa cells. Cells were fixed with 1% formaldehyde for 10 minutes. ChIP was performed with 10^7 cells and 4 µg of Histone H3 (unmodified K27) (ab308248). ChIP

DNA was sequenced on the Illumina NovaSeq6000 to a depth of 30 million reads. Line 1 shows the input as the negative control and line 2 shows results of H3 (unmodified K27) (ab308248). H3 acetyl K27 (ab177178) and H3 tri Methyl K27(ab192985) are shown as controls in lines 3 and 4 respectively. The reads using the unmodified H3K27 antibody (ab308248) do not align with the peaks from the experiment with antibodies to modifications (ab177178 and ab192985), which is as expected for this target.



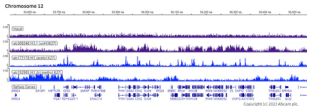
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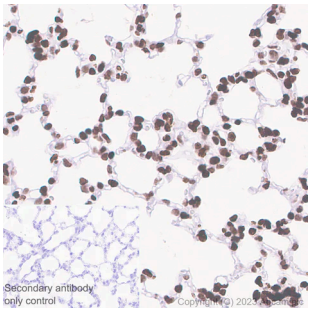
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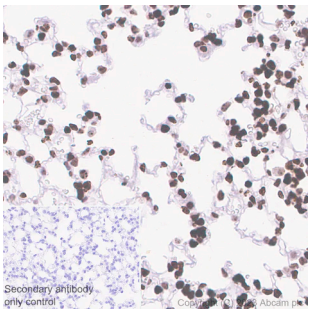
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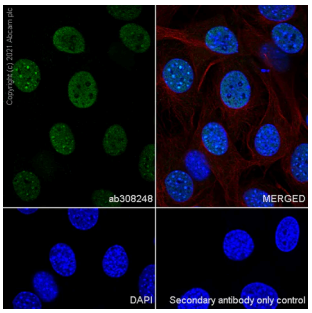
Immunohistochemical analysis of paraffin-embedded Rat lung tissue labeling Histone H3 (unmodified K27) with ab308248 at 1/4000 (0.104 ug/ml) followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer). Nuclear staining on rat lung. The section was incubated with ab308248 at 4 ☒ overnight. Counterstained with Hematoxylin. Secondary antibody only control:

Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer). Heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0)



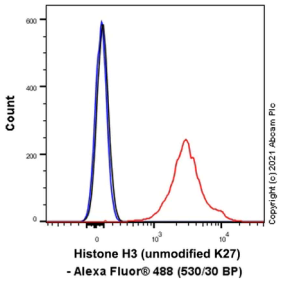
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Immunohistochemical analysis of paraffin-embedded Mouse lung tissue labeling Histone H3 (unmodified K27) with ab308248 at 1/4000 (0.104 ug/ml) followed by a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer). Nuclear staining on mouse lung. The section was incubated with ab308248 at 4 ☒ overnight. Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use Goat Anti-Rabbit IgG H&L (HRP polymer). Heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0)



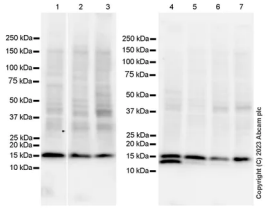
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Western blot - Anti-Histone H3 (unmodified K27) antibody [EPR23553-22] (ab308248)

Histone H3 (unmodified K27) Western blot staining using rabbit Anti-Histone H3 (unmodified K27) antibody

Blocking and diluting buffer and concentration: 1% BSA/TBST Lysates were freshly made and used for Western blotting immediately to minimize protein degradation. Exposure time: Lane 1-3: 26 seconds, lane 4-7: 15 seconds
All lanes:
Western blot - Anti-Histone H3 (unmodified K27) antibody [EPR23553-22] (ab308248) at 1/1000 dilution

Lane 1:
HeLa (human cervical adenocarcinoma epithelial cell) whole cell lysate at 20 µg

Lane 2:
NIH/3T3 (mouse embryonic fibroblast) whole cell lysate at 20 µg

Lane 3:
PC-12 (rat adrenal gland pheochromocytoma cell) whole cell lysate at 20 µg

Lane 4:
THP-1 (human monocytic leukemia monocyte) whole cell lysate at 20 µg

Lane 5:
Jurkat (human T cell leukemia T lymphocyte) whole cell lysate at 20 µg

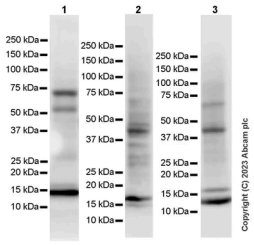
Lane 6:
RAW 264.7 (mouse Abelson murine leukemia virus-induced tumor macrophage) whole cell lysate at 20 µg

Lane 7:
2.4G2 (rat B cell lymphoma B lymphocyte) whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Observed band size: 15 kDa



Western blot - Anti-Histone H3 (unmodified K27) antibody [EPR23553-22] (ab308248)

Histone H3 (unmodified K27) Western blot staining using rabbit Anti-Histone H3 (unmodified K27) antibody

Blocking and diluting buffer and concentration: 1% BSA/TBST The lysates of lane2 and lane3 were freshly made and used for Western blotting immediately to minimize protein degradation.
Exposure time: Lane 1-2: 26 seconds, lane 3: 15 seconds
All lanes:
Western blot - Anti-Histone H3 (unmodified K27) antibody [EPR23553-22] (ab308248) at 1/1000 dilution

Lane 1:
Human spleen tissue lysate at 20 µg

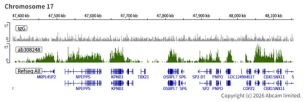
Lane 2:
Mouse spleen tissue lysate at 20 µg

Lane 3:
Rat spleen tissue lysate at 20 µg

Secondary

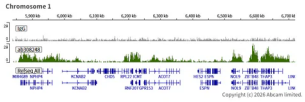
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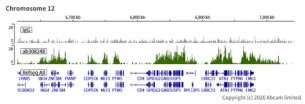
ChIC/CUT&RUN sequencing - Anti-Histone H3 (unmodified K27) antibody [EPR23553-22] (ab308248)

ChIC/CUT&RUN was performed using a pAG-MNase at a final concentration of 700 ng/mL, 2.5 x 10^5 HeLa (human cervical adenocarcinoma epithelial cell) cells and 5 µg of ab308248 [EPR23553-22]. The resulting DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 10 million reads. The negative IgG control ab172730 is also shown. The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.



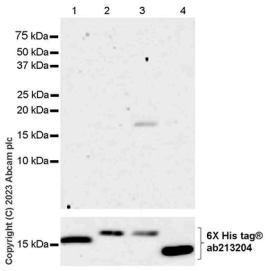
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Western blot - Anti-Histone H3 (unmodified K27) antibody [EPR23553-22] (ab308248)

Histone H3 (unmodified K27) Western blot staining using rabbit Anti-Histone H3 (unmodified K27) antibody

Blocking and diluting buffer and concentration: 5% NFDM/TBST.
This antibody does not cross-react with human H2A, H2B and H4C1.

This blot was developed using a high sensitivity ECL substrate. The high-sensitivity ECL substrate used allows for the detection of proteins in the mid-femtogram range.

In Western blot, anti-His antibody (ab213204) staining at 1/5000 dilution.

All lanes:
Western blot - Anti-Histone H3 (unmodified K27) antibody [EPR23553-22] (ab308248) at 1/1000 dilution

Lane 1:
His tagged Human H2A recombinant protein at 100 ng

Lane 2:
His tagged Human H2B recombinant protein at 100 ng

Lane 3:
His tagged Human H3C1 recombinant protein at 50 ng

Lane 4:
His tagged Human H4C1 recombinant protein at 200 ng

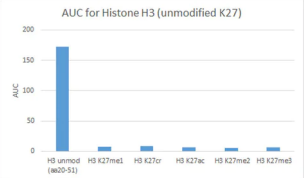
Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Developed using the ECL technique.

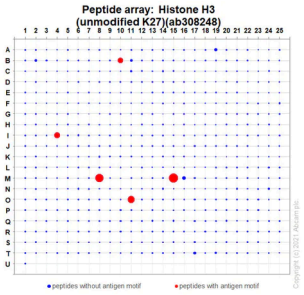
Observed band size: 15 kDa

Exposure time: 180s



Peptide Array - Anti-Histone H3 (unmodified K27) antibody [EPR23553-22] (ab308248)

Peptide array analysis of ab308248 at (0.1ug/ml) followed by a Goat Anti-Rabbit IgG, (H+L), Fluo 647nm conjugated at 1:50,000 dilution. The affinity of ab308248 to the un-modified Histone H3 K27 sequence and peptides modified at K27 from our peptide array are shown here as a histogram. AUC stands for Area under Curve.



Peptide Array - Anti-Histone H3 (unmodified K27) antibody [EPR23553-22] (ab308248)

Peptide array analysis of ab308248 at (0.1ug/ml) followed by a Goat Anti-Rabbit IgG, (H+L), Fluo 647nm conjugated at 1:50,000 dilution. ab308248 has been tested in Peptide Array against 501 different modified and unmodified histone peptides; each peptide is printed on the array at six concentrations (each in triplicate). Circle area represents affinity between the antibody and a peptide: all antigen-containing peptides are displayed as red circles, all other peptides as blue circles. The affinity is calculated as the area under the curve when antibody binding values are plotted against the corresponding peptide concentration. Each circle area is normalized to the peptide with the strongest affinity. The complete dataset, including full list of all peptides and information on the position of each peptide in the diagram, is available under the Product Protocol section.

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.