abcam

ab272691

Anti-Histone H3.3 (mutated G34W) antibody [EPR23581-39] - ChIP Grade

Anti-Histone H3.3 (mutated G34W) antibody [EPR23581-39] - ChIP Grade (ab272691) is a rabbit monoclonal antibody detecting Histone H3.3 in **Western Blot, Flow Cytometry (Intra), IP, IHC-P, ICC/IF, ChIP, Dot Blot**. Suitable for **Human**.

- Biophysical QC for unrivalled batch-batch consistency

Recombinant | RabMAb | 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
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EPR23581-39
Purification technique	Affinity purification Protein A
Concentration	0.5 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

ChIP

Tested

Species	Human
Dilution info	2 µg for 25 µg chromatin
Notes	-

Not recommended

Species	Synthetic peptide, Transfected cell lysate, Transfected cell line
Dilution info	-
Notes	-

Dot

Tested

Species	Synthetic peptide
Dilution info	1/1000
Notes	-

Expected

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

Not recommended

Species	Transfected cell lysate, Transfected cell line
Dilution info	-
Notes	-

WB

Tested

Species	Transfected cell lysate
Dilution info	1/1000

Notes	-

Expected

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

Not recommended

Species	Synthetic peptide, Transfected cell line
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/100
Notes	-

Not recommended

Species	Synthetic peptide, Transfected cell lysate, Transfected cell line
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Transfected cell line
Dilution info	1/50
Notes	-

Expected

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

Not recommended

Species	Synthetic peptide, Transfected cell lysate
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human
Dilution info	1/250
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Not recommended

Species	Synthetic peptide, Transfected cell lysate, Transfected cell line
Dilution info	-
Notes	-

Target data

See full target information H3-3A G34W

Function	Variant histone H3 which replaces conventional H3 in a wide range of nucleosomes in active genes. Constitutes the predominant form of histone H3 in non-dividing cells and is incorporated into chromatin independently of DNA synthesis. Deposited at sites of nucleosomal displacement throughout transcribed genes, suggesting that it represents an epigenetic imprint of transcriptionally active chromatin. 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

What is this antibody validated in?
Anti-Histone H3.3 (mutated G34W) antibody [EPR23581-39] - ChIP Grade (ab272691) is a rabbit recombinant monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Intra), Immunoprecipitation (IP), Immunohistochemistry (IHC-P), Immunocytochemistry/immunofluorescence (ICC/IF), ChIP, Dot Blot in Human samples.

What is the molecular weight of Histone H3.3?
Anti-Histone H3.3 (mutated G34W) [EPR23581-39] - ChIP Grade (ab272691) specifically detects a band for Histone H3.3 (UniProt: P84243) at a molecular weight of 15kDa.

Other related products
We have a range of other formats of antibody clone [EPR23581-39] also available for your convenience: ab272691, Carrier free - ab272700, PE - ab318402, APC - ab318505, Alexa Fluor® 488 - ab318608, Alexa Fluor® 647 - ab318711, Alexa Fluor® 594 - ab318814, Alexa Fluor® 555 - ab318914, Alexa Fluor® 750 - ab321540

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.

Trial sizes available!
Test your antibody or perform pre-screening before committing to a larger quantity. Sold in 20µl. Discover our selection of trial-size 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

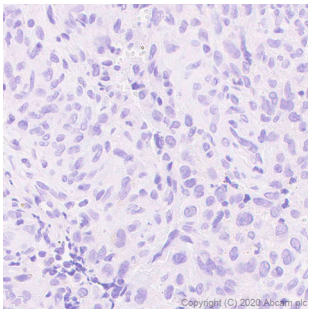
We do not recommend this combination. It is not covered by our product promise.

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.

7 product images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H3.3 (mutated G34W) antibody [EPR23581-39] - ChIP Grade (ab272691)

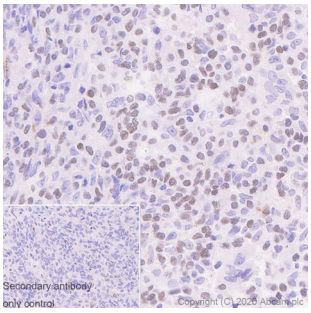
Histone H3.3 (mutated G34W) Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Histone H3.3 (mutated G34W) antibody

Immunohistochemical analysis of paraffin-embedded human chondroblastoma tissue labeling Histone H3.3(mutated G34 W) with ab272691 at 1/250 dilution followed by ready to use Goat Anti-Rabbit IgG H&L (HRP).
Negative control: No staining in human chondroblastoma (PMID: 29757500).

Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is ready to use Goat Anti-Rabbit IgG H&L (HRP).

Heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0).

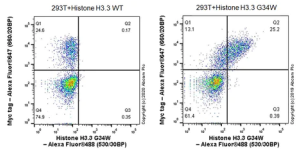


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H3.3 (mutated G34W) antibody [EPR23581-39] - ChIP Grade (ab272691)

Histone H3.3 (mutated G34W) Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Histone H3.3 (mutated G34W) antibody

Immunohistochemical analysis of paraffin-embedded human giant cell tumor of bone tissue labeling Histone H3.3(mutated G34 W) with ab272691 at 1/250 dilution followed by ready to use Goat Anti-Rabbit IgG H&L (HRP). Nuclear staining in human giant cell tumor of bone (PMID: 29757500). Counterstained with Hematoxylin.
Secondary antibody only control: Secondary antibody is ready to use Goat Anti-Rabbit IgG H&L (HRP).

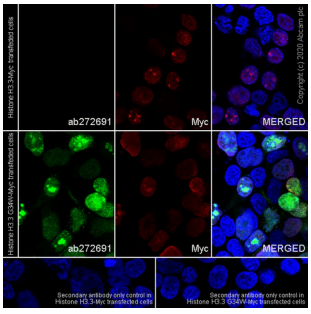
Heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0).



Flow Cytometry (Intracellular) - Anti-Histone H3.3 (mutated G34W) antibody [EPR23581-39] - ChIP Grade (ab272691)

Histone H3.3 (mutated G34W) Flow Cytometry (Intracellular) staining using rabbit Anti-Histone H3.3 (mutated G34W) antibody

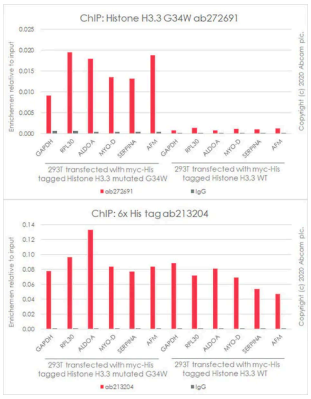
Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed 90% methanol-permeabilized HEK-293T (Human embryonic kidney epithelial cell) transfected with myc tagged Histone H3.3 WT construct (Left panel) and myc-tagged Histone H3.3 G34W construct (Right panel) cells labelling Histone H3.3 (mutated G34 W) with ab272691 at 1/50 dilution (1µg). A Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody.



Immunocytochemistry/ Immunofluorescence - Anti-Histone H3.3 (mutated G34W) antibody [EPR23581-39] - ChIP Grade (ab272691)

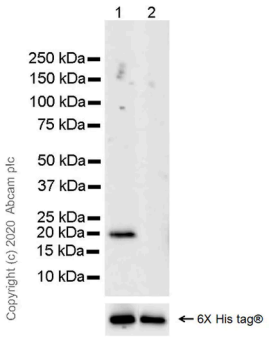
Histone H3.3 (mutated G34W) Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Histone H3.3 (mutated G34W) antibody

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HEK-293T cells labelling Histone H3.3(mutated G34 W) with ab272691 at 1/100 dilution, followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) antibody at 1/1000 2 µg/ml dilution (Green). Confocal image showing nuclear staining in HEK-293T cells transfected with Histone H3.3 G34W-Myc plasmid, while no staining in HEK-293T cells transfected with H3.3 WT -Myc plasmid. Myc-Tag Mouse mAb (Alexa Fluor® 647) was used to counterstain tubulin at 1/200 dilution (Red). The nuclear counterstain was DAPI (Blue).
Secondary antibody only control: Secondary antibody is ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 2 µg/ml dilution.



ChIP - Anti-Histone H3.3 (mutated G34W) antibody [EPR23581-39] - ChIP Grade (ab272691)

Chromatin was prepared from HEK-293T transfected with myc-His tagged Histone H3.3 mutated G34W and Histone H3.3 WT cells according to the Abcam Dual-X-ChIP protocol. Cells were fixed with formaldehyde for 10min.
The ChIP was performed with 25 µg of chromatin, 2 µg of ab272691 (red), or 2 µg of rabbit normal IgG ab172730 (gray) and 20 µl of Protein A/G sepharose beads. The immunoprecipitated DNA was quantified by real time PCR (Taqman approach for active and inactive loci, Sybr green approach for heterochromatic loci).
Primers and probes are located in the first kb of the transcribed region.



Western blot - Anti-Histone H3.3 (mutated G34W) antibody [EPR23581-39] - ChIP Grade (ab272691)

Histone H3.3 (mutated G34W) Western blot staining using rabbit Anti-Histone H3.3 (mutated G34W) antibody

Blocking and diluting buffer and concentration: 5% NFDM/TBST.
Exposure time: 3 minutes.

All lanes:
Western blot - Anti-Histone H3.3 (mutated G34W) antibody [EPR23581-39] - ChIP Grade (ab272691) at 1/1000 dilution

Lane 1:
HEK-293T (human embryonic kidney) transfected with Histone H3.3 G34W expression vector containing a myc-His-tag®, whole cell lysate at 40 µg

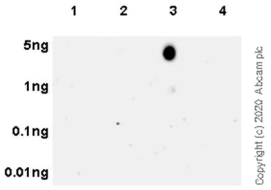
Lane 2:
HEK-293T transfected with Histone H3.3 (WT) expression vector containing a myc-His-tag®, whole cell lysate at 40 µg

Secondary

All lanes:
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (ab97051) at 1/50000 dilution

Predicted band size: 15 kDa

Observed band size: 20 kDa



Dot Blot - Anti-Histone H3.3 (mutated G34W) antibody [EPR23581-39] - ChIP Grade (ab272691)

Histone H3.3 (mutated G34W) Dot Blot staining using rabbit Anti-Histone H3.3 (mutated G34W) antibody

Dot blot analysis of Histone H3.3 (mutated G34 W) labeled with ab272691 at 1/1000 dilution.
Lane 1: Histone H3.3 H3G34W peptide (aa28-37).
Lane 2: Histone H3.3 H3G34W peptide (aa33-43).
Lane 3: Histone H3.3 H3G34W peptide (aa28-43).
Lane 4: Histone H3.3 WT peptide (aa28-43).

Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution was used as secondary antibody.

Blocking and dilution buffer: 5% NFDM/TBST.

Exposure time: 3 minutes.

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