abcam

ab272942

Anti-Histone H3.3 (mutated G34V) antibody [EPR23520-5] - ChIP Grade - BSA and Azide free

Recombinant | RabMAb

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR23520-5
Purification technique	Affinity purification Protein A
Concentration	1.015 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Human
Dilution info	-

Notes	-

Not recommended

Species	Synthetic peptide, Mouse, Rat
Dilution info	-
Notes	-

ChIP

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Synthetic peptide, Mouse, Rat
Dilution info	-
Notes	-

Dot

Tested

Species	Synthetic peptide
Dilution info	-
Notes	-

Expected

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

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

WB

Expected

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

Not recommended

Species	Synthetic peptide, Mouse, Rat
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Synthetic peptide, Mouse, Rat
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Synthetic peptide, Mouse, Rat
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human
Dilution info	-
Notes	-

Not recommended

Species	Mouse, Rat, Synthetic peptide
Dilution info	-
Notes	-

Target data

See full target information H3-3A G34V

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	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

ab272942 is the carrier-free version of ab254401.

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.

Conjugation ready

Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with 1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

Compatibility

This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

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.

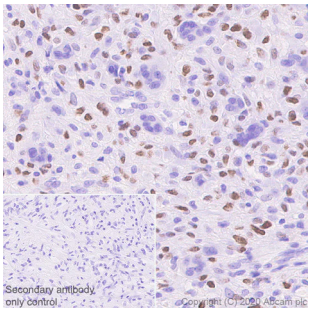
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

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.

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7 product images



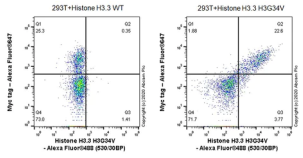
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H3.3 (mutated G34V) antibody [EPR23520-5] - ChIP Grade - BSA and Azide free (ab272942)

Histone H3.3 (mutated G34V) Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Histone H3.3 (mutated G34V) antibody

Immunohistochemical analysis of paraffin-embedded human giant cell tumor of bone tissue labeling Histone H3.3(mutated G34 V) with ab254401 at 1/1000 dilution (0.542 µg/ml) dilution followed by ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Positive staining on human giant cell tumor of bone. (PMID: 29241742). The section was incubated with ab254401 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.
Secondary antibody only control: Secondary antibody is ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.

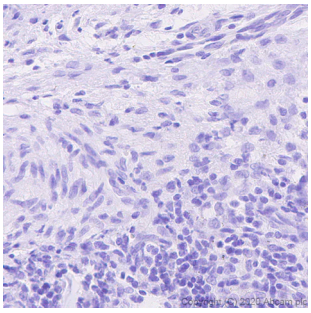
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab254401).



Flow Cytometry (Intracellular) - Anti-Histone H3.3 (mutated G34V) antibody [EPR23520-5] - ChIP Grade - BSA and Azide free (ab272942)

Histone H3.3 (mutated G34V) Flow Cytometry (Intracellular) staining using rabbit Anti-Histone H3.3 (mutated G34V) antibody

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol-permeabilized 293T (Human embryonic kidney epithelial cell) transfected with myc tagged Histone H3.3 WT construct (Left panel) and myc-tagged Histone H3.3 H3G34V construct (Right panel) cells labelling Histone H3.3 (mutated G34 V) with ab254401 at 1/500 dilution (0.1µg). A Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab254401).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H3.3 (mutated G34V) antibody [EPR23520-5] - ChIP Grade - BSA and Azide free (ab272942)

Histone H3.3 (mutated G34V) Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Histone H3.3 (mutated G34V) antibody

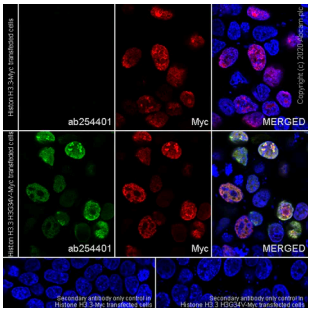
Immunohistochemical analysis of paraffin-embedded human chondroblastoma tissue labeling Histone H3.3(mutated G34 V) with ab254401 at 1/1000 dilution (0.542 µg/ml) dilution followed by ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).
Negative control: No staining on human chondroblastoma (PMID: 29241742).

The section was incubated with ab254401 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab254401).

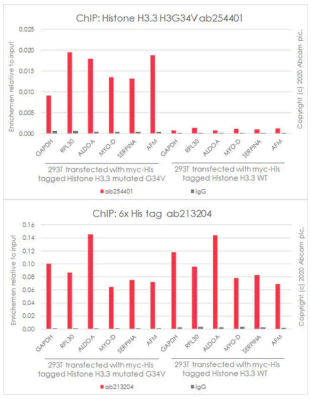


Immunocytochemistry/ Immunofluorescence - Anti-Histone H3.3 (mutated G34V) antibody [EPR23520-5] - ChIP Grade - BSA and Azide free (ab272942)

Histone H3.3 (mutated G34V) Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Histone H3.3 (mutated G34V) antibody

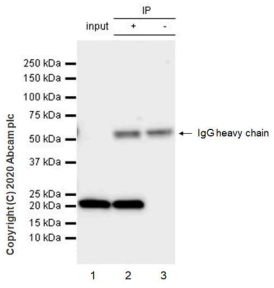
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized 293T cells labelling Histone H3.3(mutated G34 V) with ab254401 at 1/1000 dilution, followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) antibody at 1/1000 2 µg/ml dilution (Green). Confocal image showing nuclear staining in 293T cells transfected with Histone H3.3 H3G34V-Myc plasmid, while no staining in 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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab254401).



ChIP - Anti-Histone H3.3 (mutated G34V) antibody [EPR23520-5] - ChIP Grade - BSA and Azide free (ab272942)

Chromatin was prepared from 293T transfected with myc-His tagged Histone H3.3 mutated G34V and Histone H3.3 WT cells according to the Abcam Dual-X-ChIP protocol. Cells were fixed with then formaldehyde for 10min.
The ChIP was performed with 25 µg of chromatin, 2 µg of ab254401 (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.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab254401).



Immunoprecipitation - Anti-Histone H3.3 (mutated G34V) antibody [EPR23520-5] - ChIP Grade - BSA and Azide free (ab272942)

Histone H3.3(mutated G34 V) was immunoprecipitated from 0.35 mg HEK-293T (human embryonic kidney) transfected with Histone H3.3 G34V expression vector containing a myc-His-tag® whole cell lysate 10 µg with ab254401 at 1/30 dilution (2µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab254401 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP)(ab131366) was used at 1/5000 dilution.
Lane 1: HEK-293T transfected with Histone H3.3 G34V expression vector containing a myc-His-tag® whole cell lysate 10 µg

Lane 2: ab254401 IP in HEK-293T transfected with Histone H3.3 G34V expression vector containing a myc-His-tag® whole cell lysate

Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab254401 in HEK-293T transfected with Histone H3.3 G34V expression vector containing a myc-His-tag® whole cell lysate

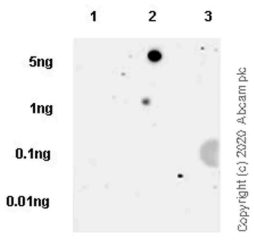
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 32 seconds.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab254401).

All lanes:
Immunoprecipitation - Anti-Histone H3.3 (mutated G34V) antibody [EPR23520-5] - ChIP Grade (ab254401)

Predicted band size: 15 kDa



Dot Blot - Anti-Histone H3.3 (mutated G34V) antibody [EPR23520-5] - ChIP Grade - BSA and Azide free (ab272942)

Histone H3.3 (mutated G34V) Dot Blot staining using rabbit Anti-Histone H3.3 (mutated G34V) antibody

Dot blot analysis of Histone H3.3 (mutated G34 V) labeled with ab254401 at 1/1000 dilution.
Lane 1: Histone H3.3 H3G34V peptide (aa28-40).
Lane 2: Histone H3.3 H3G34V peptide (aa26-38).
Lane 3: Histone H3.3 WT peptide (aa26-40).

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.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab254401).

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