abcam

ab251540

Anti-HP1 gamma/CBX3 antibody [EPR19803] - BSA and Azide free

Recombinant | RabMAb | KO Validated

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	EPR19803
Purification technique	Affinity purification Protein A
Concentration	1.047 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

ICC/IF

Tested

Species	Mouse, Human
Dilution info	-

Notes	-

Expected

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

WB

Tested

Species	Mouse, Rat, Human
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Mouse, Human
Dilution info	-
Notes	-

Expected

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

IHC-P

Tested

Species	Mouse
Dilution info	-
Notes	This application is recommended for human and mouse only.

Species	Human
Dilution info	-
Notes	This application is recommended for human and mouse only.

Expected

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

Target data

See full target information CBX3

Function

Component of heterochromatin, which recognizes and binds histone H3 tails methylated at 'Lys-9', leading to epigenetic repression (PubMed:11242053). Also recognizes and binds histone H1.4 methylated at 'Lys-26' (H1.4K26me) (PubMed:16127177). Excluded from chromatin when histone H1.4 is Simultaneously methylated at Lys-26 (H1.4K26me) and phosphorylated at Ser-27 (H1.4S27Ph) (PubMed:16127177). Involved in the formation of functional kinetochore through interaction with MIS12 complex proteins (PubMed:15502821). Contributes to the conversion of local chromatin to a heterochromatin-like repressive state through H3 'Lys-9' trimethylation, mediates the recruitment of the methyltransferases SUV39H1 and/or SUV39H2 by the PER complex to the E-box elements of the circadian target genes such as PER2 itself or PER1 (By similarity). Mediates the recruitment of NIPBL to sites of DNA damage at double-strand breaks (DSBs) (PubMed:28167679).

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

ab251540 is the carrier-free version of ab213167.

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.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	The HP1 gamma protein also known as CBX3 is an essential component of the chromatin organization machinery. It has a molecular weight of approximately 22-25 kDa. HP1 gamma is expressed in a variety of tissues including brain liver and muscle indicating its broad functional involvement across different biological systems. It exhibits a high affinity for binding to histone H3 when it is methylated facilitating the formation of heterochromatin which is key to regulating gene expression and maintaining genome stability.
Biological function summary	HP1 gamma plays a significant role in gene silencing and chromatin structure maintenance. It forms part of a protein complex that includes chromatin remodeling enzymes and transcriptional repressors which help regulate access to genomic DNA. HP1 gamma's association with other chromatin-associated proteins helps modulate the transcriptional response to cellular stimuli ensuring appropriate gene expression patterns needed for normal cell function and development.
Pathways	HP1 gamma is involved in heterochromatin formation and maintenance pathways working alongside other proteins like SUV39H1 and HP1 alpha. Through these pathways HP1 gamma ensures the compact organization of repetitive DNA sequences and suppresses unnecessary transcription. It is also involved in the

	DNA damage response pathway playing a role in preserving genomic integrity by interacting with proteins such as BRCA1 and 53BP1 involved in DNA repair.
Associated diseases and disorders	HP1 gamma's dysregulation has been associated with cancer and neurological disorders. Overexpression or mutations in this protein can lead to aberrant gene silencing contributing to oncogenesis where cancer-related genes become improperly regulated. Additionally alterations in HP1 gamma function are linked to neurodegenerative diseases potentially through its interaction with proteins like MECP2 whereby changes in the chromatin environment influence neuronal gene expression and function. Therefore understanding HP1 gamma's role provides insights into potential therapeutic targets for these conditions.

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

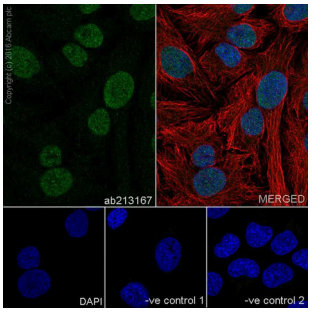
We do not recommend this combination. It is not covered by our product promise.

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.

11 product images



Immunocytochemistry/ Immunofluorescence - Anti-HP1 gamma/CBX3 antibody [EPR19803] - BSA and Azide free (ab251540)

HP1 gamma/CBX3 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-HP1 gamma/CBX3 antibody

This data was developed using ab213167, the same antibody clone in a different buffer formulation.

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling HP1 gamma/CBX3 with ab213167 at 1/100 dilution, followed by Goat Anti-Rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green).

Confocal image showing nuclear staining on HeLa cells.

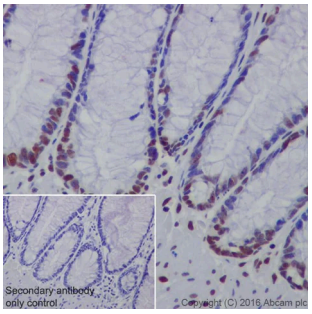
The nuclear counterstain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) at 1/1000 dilution and Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) preadsorbed (ab150120) at 1/1000 dilution (red).

The negative controls are as follows:-

-ve control 1: ab213167 at 1/100 dilution followed by ab150120 at 1/1000 dilution.

-ve control 2: ab7291 at 1/1000 dilution followed by ab150077 at 1/1000 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HP1 gamma/CBX3 antibody [EPR19803] - BSA and Azide free (ab251540)

HP1 gamma/CBX3 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-HP1 gamma/CBX3 antibody

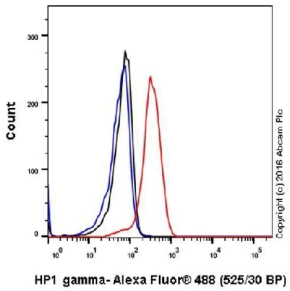
This data was developed using ab213167, the same antibody clone in a different buffer formulation.
Immunohistochemical analysis of paraffin-embedded Human colon tissue labeling HP1 gamma/CBX3 with ab213167 at 1/250 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Nucleus staining on epithelial cells of Human colon is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ab97051 at 1/500 dilution.

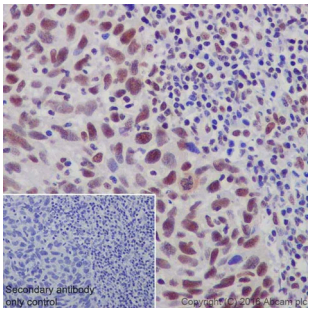
Perform heat mediated antigen retrieval with EDTA buffer pH 9 before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-HP1 gamma/CBX3 antibody [EPR19803] - BSA and Azide free (ab251540)

HP1 gamma/CBX3 Flow Cytometry (Intracellular) staining using rabbit Anti-HP1 gamma/CBX3 antibody

This data was developed using ab213167, the same antibody clone in a different buffer formulation.
Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling HP1 gamma/CBX3 with ab213167 at 1/60 dilution (red) compared with a Rabbit IgG, monoclonal-Isotype control (ab172730) (black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat anti rabbit IgG (Alexa Fluor® 488) at 1/2000 dilution was used as the secondary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HP1 gamma/CBX3 antibody [EPR19803] - BSA and Azide free (ab251540)

HP1 gamma/CBX3 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-HP1 gamma/CBX3 antibody

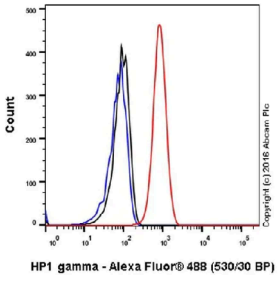
This data was developed using ab213167, the same antibody clone in a different buffer formulation.
Immunohistochemical analysis of paraffin-embedded Human cervix carcinoma tissue labeling HP1 gamma/CBX3 with ab213167 at 1/250 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Nucleus staining on cancer cells of Human cervix carcinoma is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ab97051 at 1/500 dilution.

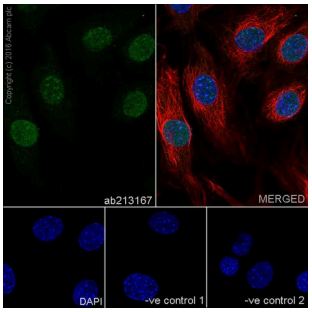
Perform heat mediated antigen retrieval with EDTA buffer pH 9 before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-HP1 gamma/CBX3 antibody [EPR19803] - BSA and Azide free (ab251540)

HP1 gamma/CBX3 Flow Cytometry (Intracellular) staining using rabbit Anti-HP1 gamma/CBX3 antibody

This data was developed using ab213167, the same antibody clone in a different buffer formulation.
Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed NIH/3T3 (Mouse embryonic fibroblast cell line) cells labeling HP1 gamma/CBX3 with ab213167 at 1/60 dilution (red) compared with a Rabbit IgG,monoclonal - Isotype control (ab172730) (black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat anti Rabbit IgG (Alexa Fluor® 488) at 1/2000 dilution was used as the secondary antibody.



Immunocytochemistry/ Immunofluorescence - Anti-HP1 gamma/CBX3 antibody [EPR19803] - BSA and Azide free (ab251540)

HP1 gamma/CBX3 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-HP1 gamma/CBX3 antibody

This data was developed using ab213167, the same antibody clone in a different buffer formulation.

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized NIH/3T3 (Mouse embryonic fibroblast cell line) cells labeling HP1 gamma/CBX3 with ab213167 at 1/100 dilution, followed by Goat Anti-Rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green).

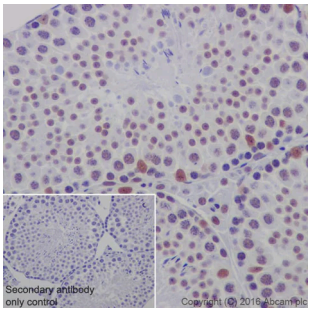
Confocal image showing nuclear staining on NIH/3T3 cells.

Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) at 1/1000 dilution and Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) preadsorbed (ab150120) at 1/1000 dilution (red).

The negative controls are as follows:-

-ve control 1: ab213167 at 1/100 dilution followed by ab150120 at 1/1000 dilution.

-ve control 2: ab7291 at 1/1000 dilution followed by ab150077 at 1/1000 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HP1 gamma/CBX3 antibody [EPR19803] - BSA and Azide free (ab251540)

HP1 gamma/CBX3 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-HP1 gamma/CBX3 antibody

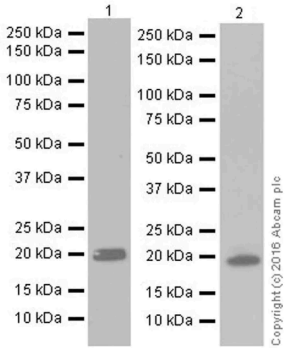
This data was developed using ab213167, the same antibody clone in a different buffer formulation.
Immunohistochemical analysis of paraffin-embedded Mouse testis tissue labeling HP1 gamma/CBX3 with ab213167 at 1/250 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Nucleus staining on Mouse testis is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ab97051 at 1/500 dilution.

Perform heat mediated antigen retrieval with EDTA buffer pH 9 before commencing with IHC staining protocol.



Western blot - Anti-HP1 gamma/CBX3 antibody [EPR19803] - BSA and Azide free (ab251540)

This data was developed using ab213167, the same antibody clone in a different buffer formulation.
Blocking and dilution buffer: 5% NFDM/TBST.

Exposure times: Lane 1: 3 minutes; Lane 2: 15 seconds.

All lanes:
Western blot - Anti-HP1 gamma/CBX3 antibody [EPR19803] (ab213167) at 1/1000 dilution

Lane 1:
HepG2 (Human liver hepatocellular carcinoma cell line) whole cell lysate at 20 µg

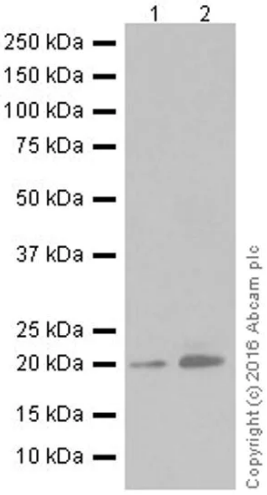
Lane 2:
HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate at 20 µg

Secondary

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

Predicted band size: 20 kDa

Observed band size: 21 kDa



Western blot - Anti-HP1 gamma/CBX3 antibody [EPR19803] - BSA and Azide free (ab251540)

This data was developed using ab213167, the same antibody clone in a different buffer formulation.
All lanes:
Western blot - Anti-HP1 gamma/CBX3 antibody [EPR19803] (ab213167) at 1/1000 dilution

Lane 1:
Human fetal brain lysate at 10 µg

Lane 2:
Human fetal kidney lysate at 10 µg

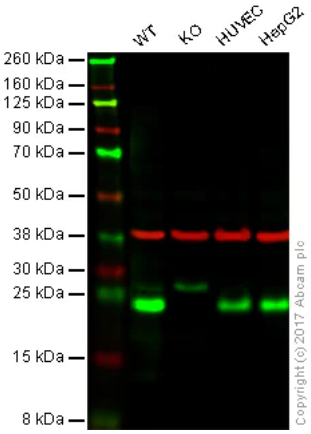
Secondary

All lanes:
Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/10000 dilution

Predicted band size: 20 kDa

Observed band size: 21 kDa

Exposure time: 3min



Western blot - Anti-HP1 gamma/CBX3 antibody [EPR19803] - BSA and Azide free (ab251540)

HP1 gamma/CBX3 Western blot staining using rabbit Anti-HP1 gamma/CBX3 antibody

This data was developed using ab213167, the same antibody clone in a different buffer formulation.
Lane 1: Wild type HAP1 whole cell lysate (20 µg)

Lane 2: CBX3 (KO) knockout HAP1 whole cell lysate (20 µg)

Lane 3: HUVEC whole cell lysate (20 µg)

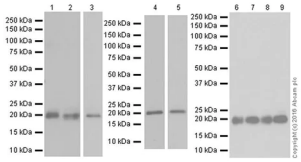
Lane 4: HepG2 whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - ab213167 observed at 23 kDa. Red - loading control, ab8245, observed at 37 kDa.

ab213167 was shown to recognize HP1 gamma/CBX3 when HP1 gamma/CBX3 knockout samples were used, along with additional cross-reactive bands. Wild-type and HP1 gamma/CBX3 knockout samples were subjected to SDS-PAGE. ab213167 and ab8245 (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 500 dilution and 1/10000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed ab216776 secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-HP1 gamma/CBX3 antibody [EPR19803] (ab213167)

Predicted band size: 20 kDa



Western blot - Anti-HP1 gamma/CBX3 antibody [EPR19803] - BSA and Azide free (ab251540)

This data was developed using ab213167, the same antibody clone in a different buffer formulation.
Blocking and dilution buffer: 5% NFDM/TBST.

Exposure time: Lanes 1 and 2: 3 minutes; Lane 3: 10 seconds; Lane 4: 30 seconds; Lane 5: 10 seconds; Lanes 6-9: 30 seconds.

All lanes:
Western blot - Anti-HP1 gamma/CBX3 antibody [EPR19803] (ab213167) at 1/1000 dilution

Lane 1:
Mouse brain lysate at 10 µg

Lane 2:
Mouse kidney lysate at 10 µg

Lane 3:
Mouse spleen lysate at 10 µg

Lane 4:
Rat brain lysate at 10 µg

Lane 5:
Rat spleen lysate at 10 µg

Lane 6:

C6 (Rat glial tumor cell line) whole cell lysate at 10 µg

Lane 7:
RAW 264.7 (Mouse macrophage cell line transformed with Abelson murine leukemia virus) whole cell lysate at 10 µg

Lane 8:
PC-12 (Rat adrenal gland pheochromocytoma cell line) whole cell lysate at 10 µg

Lane 9:
NIH/3T3 (Mouse embryonic fibroblast cell line) whole cell lysate at 10 µg

Secondary

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

Predicted band size: 20 kDa

Observed band size: 21 kDa

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