

Product datasheet

Anti-HP1 gamma/CBX3 antibody [EPR19803] ab213167

KO VALIDATED Recombinant **RabMAb**

★★★★☆ [3 Abreviews](#) [3 References](#) [12 Images](#)

Overview

Product name	Anti-HP1 gamma/CBX3 antibody [EPR19803]
Description	Rabbit monoclonal [EPR19803] to HP1 gamma/CBX3
Host species	Rabbit
Tested applications	Suitable for: WB, Flow Cyt (Intra), IHC-P, ICC/IF
Species reactivity	Reacts with: Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: Human fetal brain and fetal kidney lysates; HepG2, HeLa, C6, RAW 264.7, PC-12 and NIH/3T3 whole cell lysates; Mouse brain, kidney and spleen lysates; Rat brain and spleen lysates. IHC-P: Human colon and cervix carcinoma tissues; Mouse testis tissue. ICC/IF: NIH/3T3 and HeLa cells. Flow Cyt (intra): HeLa and NIH/3T3 cells.
General notes	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none">- High batch-to-batch consistency and reproducibility- Improved sensitivity and specificity- Long-term security of supply- Animal-free production <p>For more information see here.</p> <p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents.</p>

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR19803

Isotype

IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab213167 in the following tested applications. The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB	★★★★★ (1)	1/1000. Detects a band of approximately 21 kDa (predicted molecular weight: 21 kDa).
Flow Cyt (Intra)		1/60.
IHC-P		1/250. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. This application is recommended for human and mouse only.
ICC/IF	★★★★★ (1)	1/100.

Target

Function

Seems to be involved in transcriptional silencing in heterochromatin-like complexes. Recognizes and binds histone H3 tails methylated at 'Lys-9', leading to epigenetic repression. May contribute to the association of the heterochromatin with the inner nuclear membrane through its interaction with lamin B receptor (LBR). Involved in the formation of functional kinetochore through interaction with MIS12 complex proteins.

Sequence similarities

Contains 2 chromo domains.

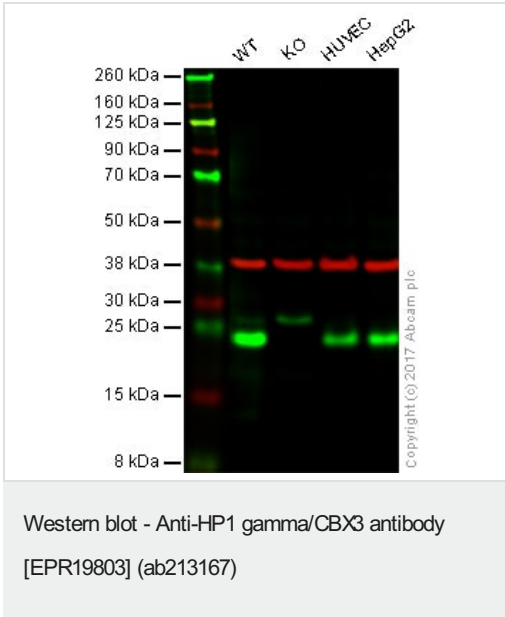
Post-translational modifications

Phosphorylated by PIM1. Phosphorylated during interphase and possibly hyper-phosphorylated during mitosis.

Cellular localization

Nucleus. Associates with euchromatin and is largely excluded from constitutive heterochromatin. May be associated with microtubules and mitotic poles during mitosis.

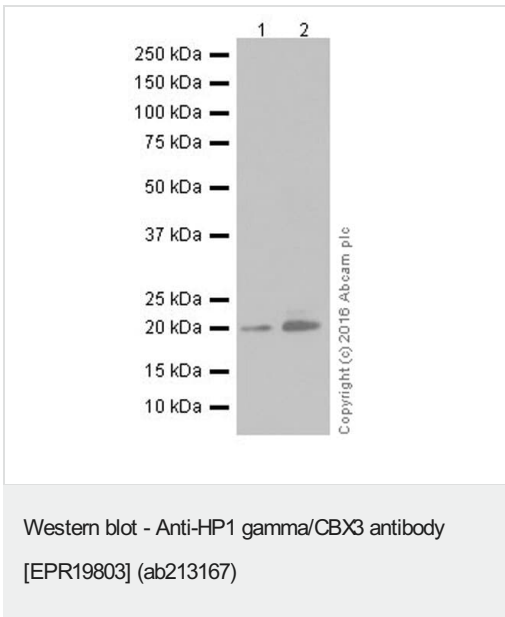
Images



Lanes:

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

- Lane 1 :** Human fetal brain lysate
- Lane 2 :** Human fetal kidney lysate

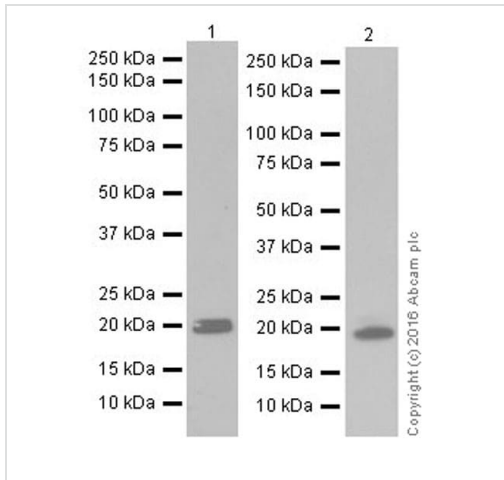
Lysates/proteins at 10 µg per lane.

Secondary

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

Predicted band size: 21 kDa
Observed band size: 21 kDa

Exposure time: 3 minutes



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

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

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

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

Lysates/proteins at 20 µg per lane.

Secondary

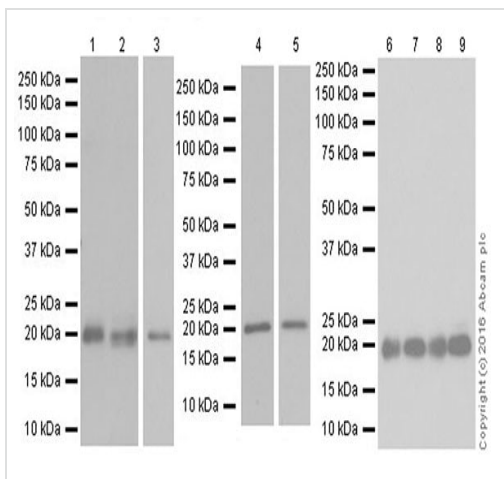
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 21 kDa

Observed band size: 21 kDa

Blocking/Dilution buffer: 5% NFDm/TBST.

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



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

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

Lane 1 : Mouse brain lysate

Lane 2 : Mouse kidney lysate

Lane 3 : Mouse spleen lysate

Lane 4 : Rat brain lysate

Lane 5 : Rat spleen lysate

Lane 6 : C6 (Rat glial tumor cell line) whole cell lysate

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

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

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

Lysates/proteins at 10 µg per lane.

Secondary

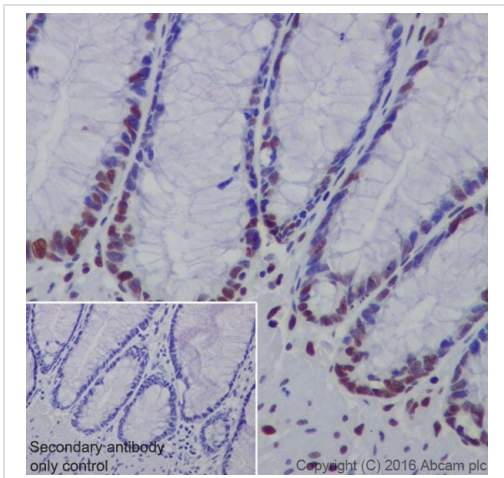
All lanes : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/100000 dilution

Predicted band size: 21 kDa

Observed band size: 21 kDa

Blocking/Dilution buffer: 5% NFDm/TBST.

Exposure time: Lane 1 and 2: 3 minutes; Lane 3: 10 seconds; Lane 4: 30 seconds; Lane 5: 10 seconds; Lane 6,7,8 and 9: 30 seconds.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HP1 gamma/CBX3 antibody [EPR19803] (ab213167)

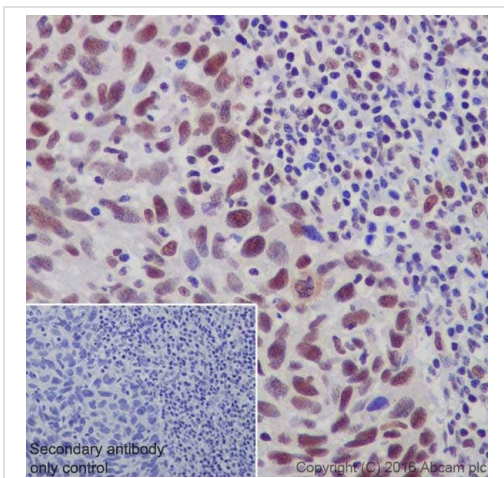
Immunohistochemical analysis of paraffin-embedded Human colon tissue labeling HP1 gamma/CBX3 with ab213167 at 1/250 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/500 dilution.

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.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HP1 gamma/CBX3 antibody [EPR19803] (ab213167)

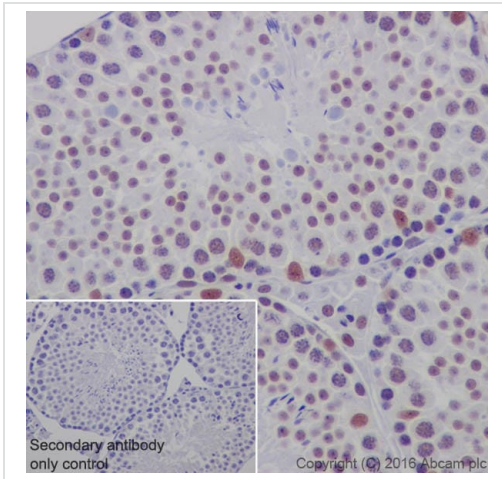
Immunohistochemical analysis of paraffin-embedded Human cervix carcinoma tissue labeling HP1 gamma/CBX3 with ab213167 at 1/250 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/500 dilution.

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.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HP1 gamma/CBX3 antibody [EPR19803] (ab213167)

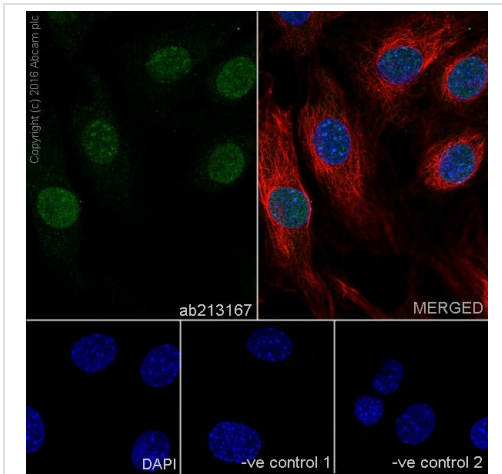
Immunohistochemical analysis of paraffin-embedded Mouse testis tissue labeling HP1 gamma/CBX3 with ab213167 at 1/250 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

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.



Immunocytochemistry/ Immunofluorescence - Anti-HP1 gamma/CBX3 antibody [EPR19803] (ab213167)

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).

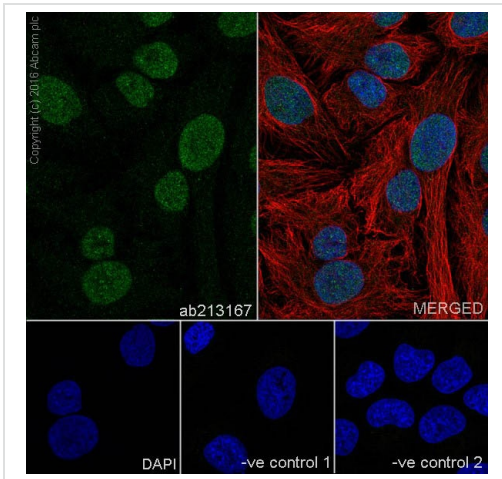
Confocal image showing nuclear staining on NIH/3T3 cells.

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.



Immunocytochemistry/ Immunofluorescence - Anti-HP1 gamma/CBX3 antibody [EPR19803] (ab213167)

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.

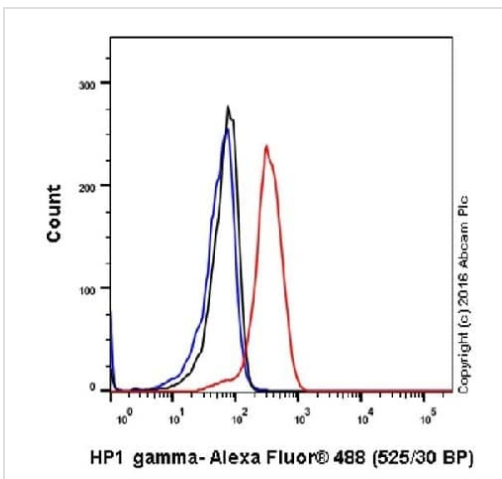
The nuclear counterstain is DAPI (blue).

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:-

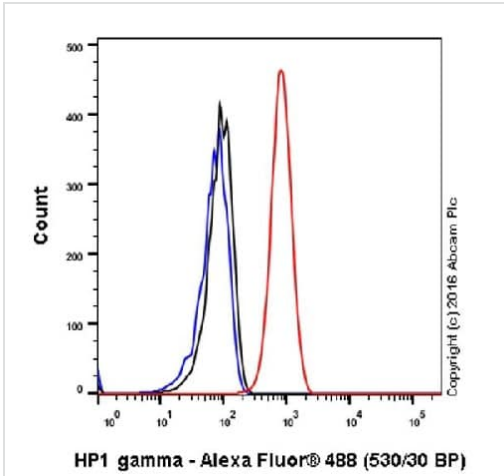
-ve control 1: ab213167 at 1/100 dilution followed by **ab150120** at 1/1000 dilution.

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



Flow Cytometry (Intracellular) - Anti-HP1 gamma/CBX3 antibody [EPR19803] (ab213167)





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.



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.

Flow Cytometry (Intracellular) - Anti-HP1 gamma/CBX3 antibody [EPR19803] (ab213167)

Why choose a recombinant antibody?

 <p>Research with confidence Consistent and reproducible results</p>	 <p>Long-term and scalable supply Recombinant technology</p>
 <p>Success from the first experiment Confirmed specificity</p>	 <p>Ethical standards compliant Animal-free production</p>

Anti-HP1 gamma/CBX3 antibody [EPR19803] (ab213167)

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