

Anti-CTCF antibody [EPR18253] - ChIP Grade ab188408

重组 RabMAb

8 References **18 图像**

概述

| | |
|--------------|---|
| 产品名称 | Anti-CTCF抗体[EPR18253] - ChIP Grade |
| 描述 | 兔单克隆抗体[EPR18253] to CTCF - ChIP Grade |
| 宿主 | Rabbit |
| 经测试应用 | 适用于: ChIP-sequencing, IHC-P, ChIP, ICC/IF, WB, ChIC/CUT&RUN-seq |
| 种属反应性 | 与反应: Mouse, Rat, Human |
| 免疫原 | Synthetic peptide. This information is proprietary to Abcam and/or its suppliers. |
| 阳性对照 | WB: HeLa, LLC, RAW 264.7, HEK-293, HepG2, MCF7, C6, PC-12 and NIH/3T3 whole cell lysates; human fetal brain, fetal heart, fetal kidney and fetal spleen lysates; mouse brain, heart, kidney and spleen lysates; rat brain, heart, kidney and spleen lysates. IHC-P: Human endometrium, mouse liver and rat stomach tissues. ICC/IF: HeLa and NIH/3T3 cells. ChIP: Chromatin prepared from HeLa cells. ChIP-seq: Chromatin prepared from HeLa and Mouse Embryonic Fibroblasts cells. ChIC/CUT&RUN-Seq: HeLa and F9 cells. |
| 常规说明 | <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> |

性能

| | |
|-------------|--|
| 形式 | Liquid |
| 存放说明 | 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. |
| 存储溶液 | <p>pH: 7.2</p> <p>Preservative: 0.01% Sodium azide</p> <p>Constituents: 59% PBS, 0.05% BSA, 40% Glycerol (glycerin, glycerine)</p> |
| 纯度 | Protein A purified |

| | |
|------|----------|
| 克隆 | 单克隆 |
| 克隆编号 | EPR18253 |
| 同种型 | IgG |

应用

The Abpromise guarantee **Abpromise™**承诺保证使用ab188408于以下的经测试应用

“应用说明”部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

| 应用 | Ab评论 | 说明 |
|------------------|------|--|
| ChIP-sequencing | | Use 4-8µg for 10 ⁷ cells. |
| IHC-P | | 1/4000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. |
| ChIP | | Use 2 µg for 25 µg of chromatin. |
| ICC/IF | | 1/2000. |
| WB | | 1/5000. Detects a band of approximately 140,130, 97, 80, 73, 70, 55 kDa (predicted molecular weight: 83, 46 kDa). |
| ChIC/CUT&RUN-seq | | Use at an assay dependent concentration. 5 µg |

靶标

功能

Chromatin binding factor that binds to DNA sequence specific sites. Involved in transcriptional regulation by binding to chromatin insulators and preventing interaction between promoter and nearby enhancers and silencers. Acts as transcriptional repressor binding to promoters of vertebrate MYC gene and BAG1 gene. Also binds to the PLK and PIM1 promoters. Acts as a transcriptional activator of APP. Regulates APOA1/C3/A4/A5 gene cluster and controls MHC class II gene expression. Plays an essential role in oocyte and preimplantation embryo development by activating or repressing transcription. Seems to act as tumor suppressor. Plays a critical role in the epigenetic regulation. Participates to the allele-specific gene expression at the imprinted IGF2/H19 gene locus. On the maternal allele, binding within the H19 imprinting control region (ICR) mediates maternally inherited higher-order chromatin conformation to restrict enhancer access to IGF2. Plays a critical role in gene silencing over considerable distances in the genome. Preferentially interacts with unmethylated DNA, preventing spreading of CpG methylation and maintaining methylation-free zones. Inversely, binding to target sites is prevented by CpG methylation. Plays an important role in chromatin remodeling. Can dimerize when it is bound to different DNA sequences, mediating long-range chromatin looping. Mediates interchromosomal association between IGF2/H19 and WSB1/NF1 and may direct distant DNA segments to a common transcription factory. Causes local loss of histone acetylation and gain of histone methylation in the beta-globin locus, without affecting transcription. When bound to chromatin, it provides an anchor point for nucleosomes positioning. Seems to be essential for homologous X-chromosome pairing. May participate with Tsix in establishing a regulatable epigenetic switch for X chromosome inactivation. May play a role in preventing the propagation of stable methylation at the escape genes from X- inactivation. Involved in sister chromatid cohesion.

Associates with both centromeres and chromosomal arms during metaphase and required for cohesin localization to CTCF sites. Regulates asynchronous replication of IGF2/H19.

组织特异性

Ubiquitous. Absent in primary spermatocytes.

序列相似性

Belongs to the CTCF zinc-finger protein family.

Contains 11 C2H2-type zinc fingers.

结构域

The 11 zinc fingers are highly conserved among vertebrates, exhibiting almost identical amino acid sequences. Different subsets or combination of individual zinc fingers gives the ability to CTCF to recognize multiple DNA target sites.

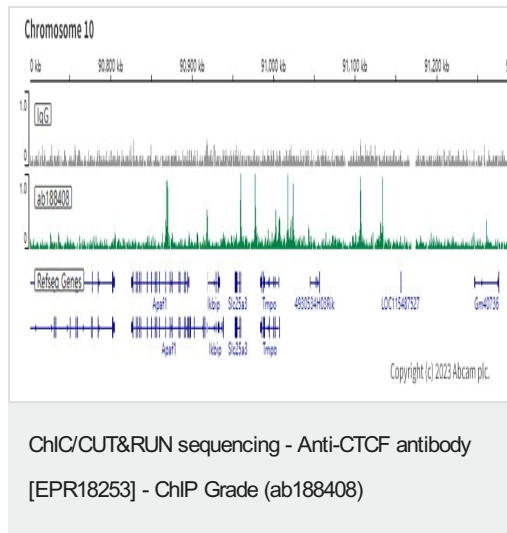
翻译后修饰

Sumoylated on Lys-74 and Lys-689; sumoylation of CTCF contributes to the repressive function of CTCF on the MYC P2 promoter.

细胞定位

Nucleus > nucleoplasm. Chromosome. Chromosome > centromere. May translocate to the nucleolus upon cell differentiation. Associates with both centromeres and chromosomal arms during metaphase. Associates with the H19 ICR in mitotic chromosomes. May be preferentially excluded from heterochromatin during interphase.

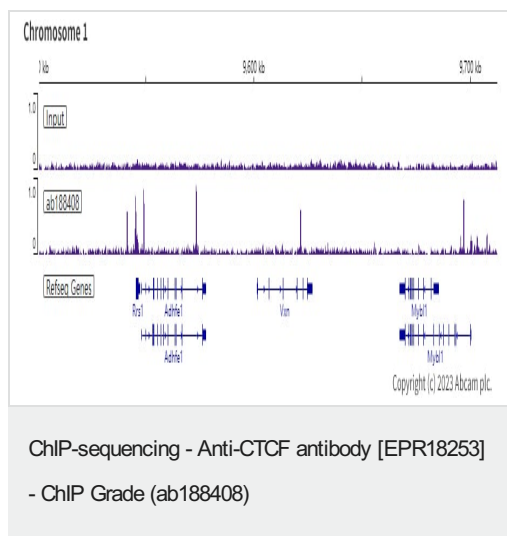
图片



ChIC/CUT&RUN was performed using a pAG-MNase at a final concentration of 700 ng/mL, 2.5×10^5 F9 (Mouse embryonic testicular cancer cell line) cells and 5 μ g of ab188408 [EPR18253]. 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.

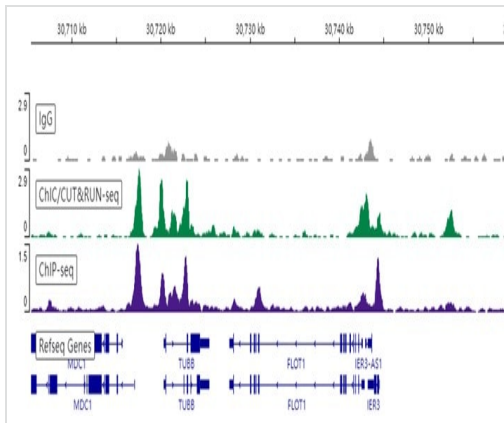
Additional screenshots of mapped reads can be downloaded [here](#).

The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.



Chromatin was prepared from Mouse Embryonic Fibroblast cells. Cells were fixed with 1% formaldehyde for 10 minutes. ChIP was performed with 10^7 cells and 8 μ g of [Anti-CTCF antibody] (ab188408). ChIP DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 30 million reads. The Input control is also shown.

Additional screenshots of mapped reads can be downloaded [here](#).



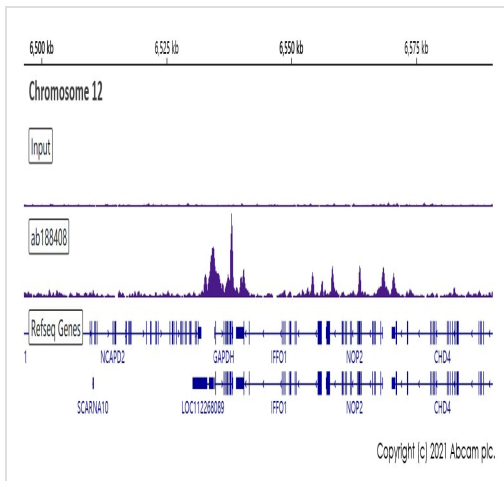
ChIC/CUT&RUN sequencing - Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408)

ChIC/CUT&RUN was performed using a pAG-MNase at a final concentration of 700 ng/mL, 2×10^5 HeLa (Human cervix adenocarcinoma epithelial cell line) cells and 5 μ g of ab188408 [EPR18253]. 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 ChIP data was conducted on chromatin prepared from HeLa cells. Cells were fixed with 1% formaldehyde for 10 minutes. ChIP was performed with 10^7 HeLa cells and 8 μ g of ab188408. ChIP DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 30 million reads.

Additional screenshots of mapped reads can be downloaded [here](#).

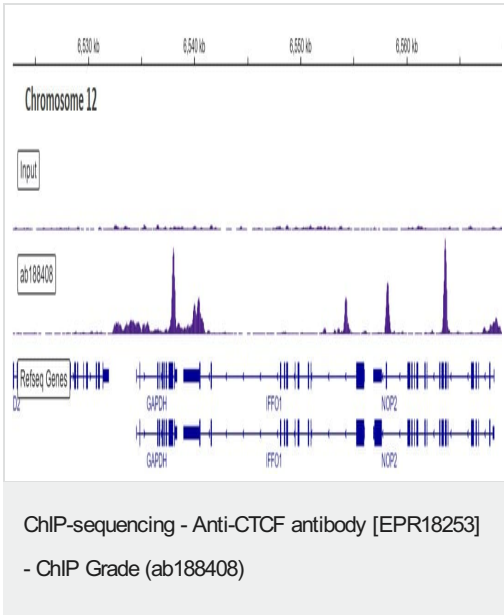
The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.



ChIP-sequencing - Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408)

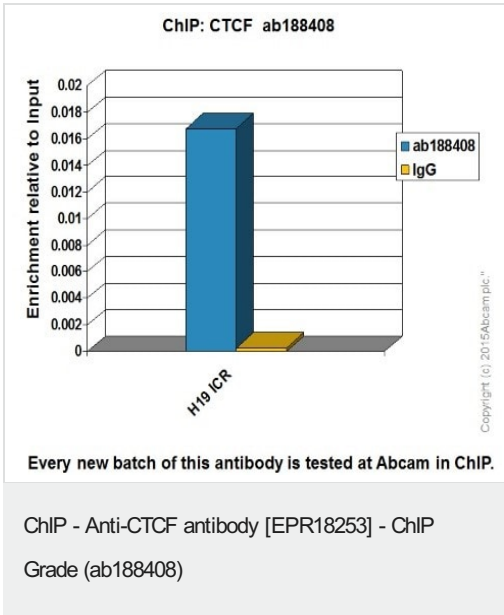
Chromatin was prepared from HeLa cells. Cells were fixed with 1% formaldehyde for 10 minutes. ChIP was performed with 30 μ g of chromatin and 4 μ g of ab188408. ChIP DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 30 million reads. ChIP-Seq validation performed with ChIP-Kit Transcription Factors ChIP-Seq (**ab270813**).

Additional screenshots of mapped reads can be downloaded [here](#).

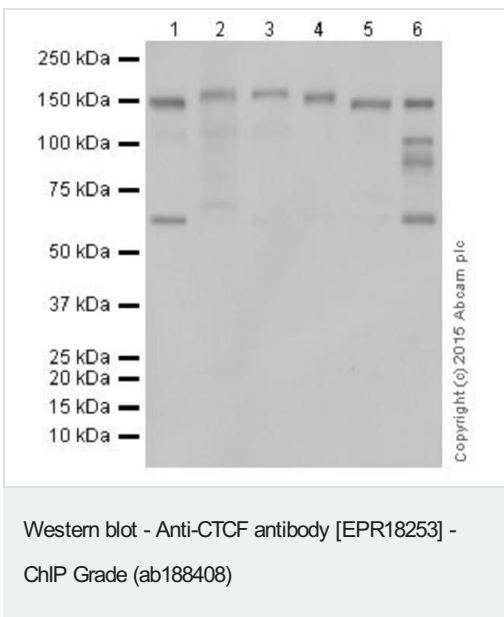


Chromatin was prepared from HeLa cells. Cells were fixed with 1% formaldehyde for 10 minutes. ChIP was performed with 30 µg of chromatin and 4 µg of Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408). ChIP DNA was sequenced on the Illumina NextSeq 500 to a depth of 30 million reads. ChIP-Seq validation performed by Active Motif, Carlsbad, CA.

Additional screenshots of mapped reads can be downloaded [here](#).



Chromatin was prepared from HeLa (Human epithelial cells from cervix adenocarcinoma) cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10 minutes. The ChIP was performed with 25µg of chromatin, 2µg of ab188408 (blue), and 20µl of Anti Rabbit IgG sepharose beads. 2µg of rabbit normal IgG was added to the beads control (yellow). The immunoprecipitated DNA was quantified by real time PCR (Sybr green approach).



All lanes : Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408) at 1/5000 dilution

Lane 1 : HeLa (Human epithelial cells from cervix adenocarcinoma) whole cell lysate

Lane 2 : LLC (Mouse lung carcinoma) whole cell lysate

Lane 3 : RAW 264.7 (Mouse macrophage cells transformed with Abelson murine leukemia virus) whole cell lysate

Lane 4 : HEK-293 (Human epithelial cells from embryonic kidney) whole cell lysate

Lane 5 : HepG2 (Human liver hepatocellular carcinoma) whole cell lysate

Lane 6 : MCF7 (Human breast adenocarcinoma cell line) 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

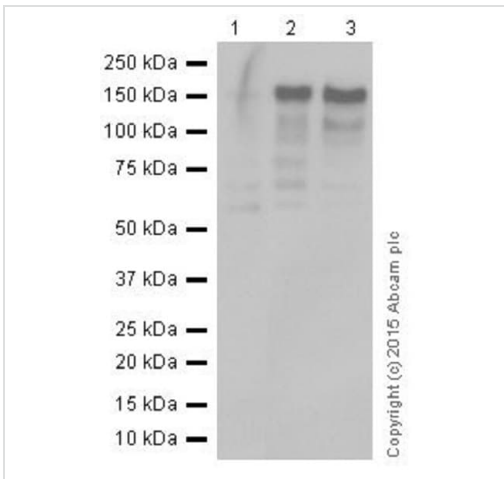
Predicted band size: 83, 46 kDa

Exposure time: 1 second

Blocking/Dilution buffer: 5% NFDm/TBST.

Observed MW: 140, 97, 80, 73, 70, 55 kDa.

The multiples bands observed in the WB have been reported in literature (PMID: 12878173, PMID: 8246978).



Western blot - Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408)

All lanes : Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408) at 1/1000 dilution

Lane 1 : C6 (Rat glial tumor cells) whole cell lysate

Lane 2 : PC-12 (Rat adrenal gland pheochromocytoma) whole cell lysate

Lane 3 : NIH/3T3 (Mouse embryonic fibroblast cells) 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

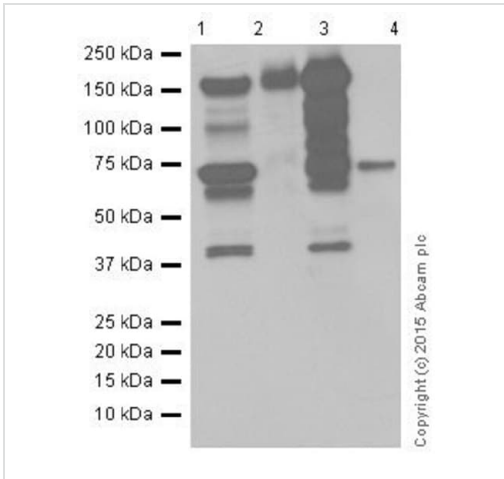
Predicted band size: 83, 46 kDa

Exposure time: 1 second

Blocking/Dilution buffer: 5% NFDm/TBST.

Observed MW: 140, 130, 97, 80, 73, 70, 55 kDa.

The multiples bands observed in the WB have been reported in literature (PMID: 12878173, PMID: 8246978).



Western blot - Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408)

All lanes : Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408) at 1/1000 dilution

Lane 1 : Human fetal brain lysate

Lane 2 : Human fetal heart lysate

Lane 3 : Human fetal kidney lysate

Lane 4 : Human fetal spleen lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Anti-Rabbit IgG (HRP), specific to the non-reduced form of IgG at 1/100000 dilution

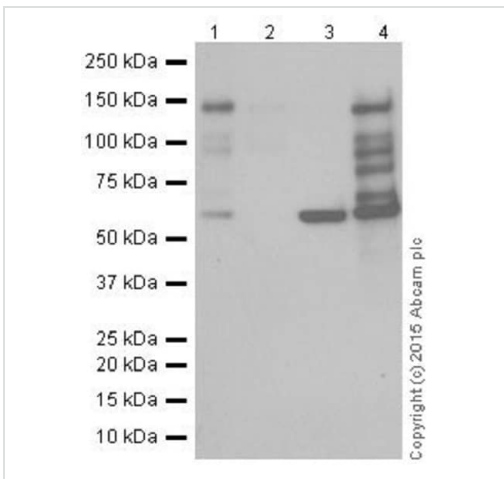
Predicted band size: 83, 46 kDa

Exposure time: 15 seconds

Blocking/Dilution buffer: 5% NFDm/TBST.

Observed MW: 140, 130, 97, 80, 73, 70, 55 kDa.

The multiples bands observed in the WB have been reported in literature (PMID: 12878173, PMID: 8246978).



Western blot - Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408)

All lanes : Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408) at 1/1000 dilution

Lane 1 : Mouse brain lysate

Lane 2 : Mouse heart lysate

Lane 3 : Mouse kidney lysate

Lane 4 : Mouse spleen lysate

Lysates/proteins at 10 µg per lane.

Secondary

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

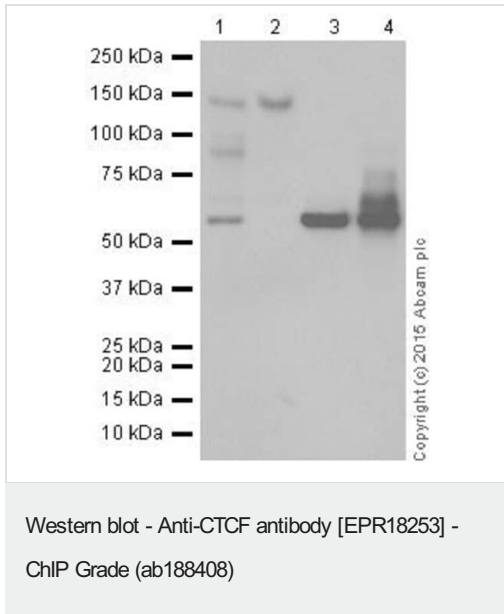
Predicted band size: 83, 46 kDa

Exposure time: 15 seconds

Blocking/Dilution buffer: 5% NFDm/TBST.

Observed MW: 140, 130, 97, 80, 73, 70, 55 kDa.

The multiples bands observed in the WB have been reported in literature (PMID: 12878173, PMID: 8246978).



All lanes : Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408) at 1/1000 dilution

Lane 1 : Rat brain lysate

Lane 2 : Rat heart lysate

Lane 3 : Rat kidney lysate

Lane 4 : Rat spleen lysate

Lysates/proteins at 10 µg per lane.

Secondary

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

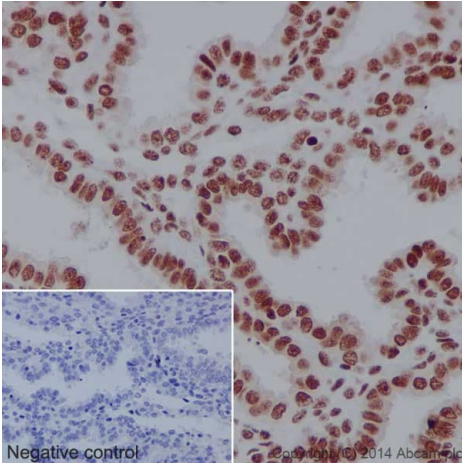
Predicted band size: 83, 46 kDa

Exposure time: 2 seconds

Blocking/Dilution buffer: 5% NFDm/TBST.

Observed MW 140, 130, 97, 80, 73, 70, 55 kDa.

The multiples bands observed in the WB have been reported in literature (PMID: 12878173, PMID: 8246978).

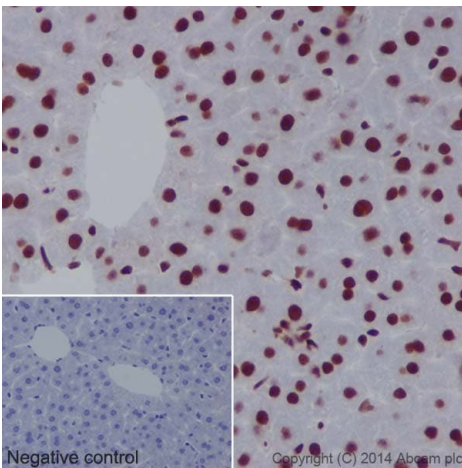


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408)

Immunohistochemical analysis of paraffin-embedded human endometrium tissue labeling CTCF with ab188408 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Nuclear staining on human endometrium is observed. Counter stained with Hematoxylin.

Negative control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

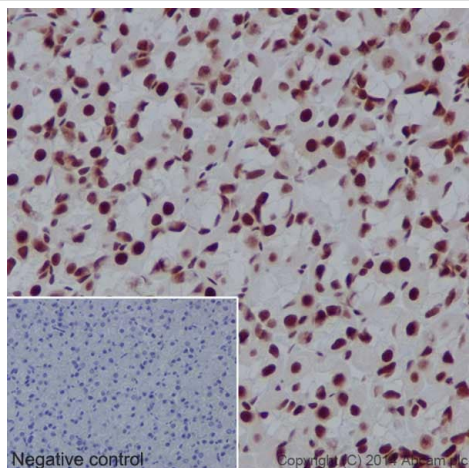


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408)

Immunohistochemical analysis of paraffin-embedded mouse liver tissue labeling CTCF with ab188408 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Nuclear staining on hepatocytes of mouse liver is observed. Counter stained with Hematoxylin.

Negative control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

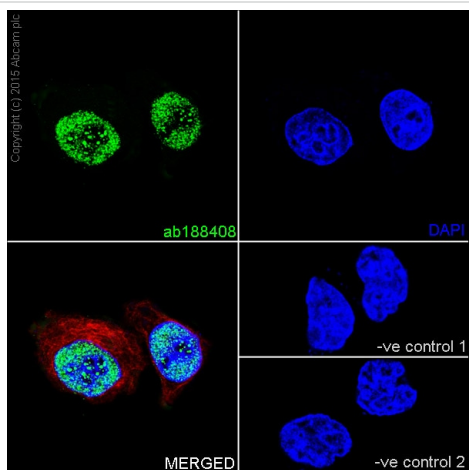


Immunohistochemical analysis of paraffin-embedded rat stomach tissue labeling CTCF with ab188408 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/500 dilution. Nuclear staining on the epithelium cells of rat stomach is observed. Counter stained with Hematoxylin.

Negative control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408)



Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cells from cervix adenocarcinoma) cells labeling CTCF with ab188408 at 1/2000 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 cell line. The nuclear counter stain is DAPI (blue).

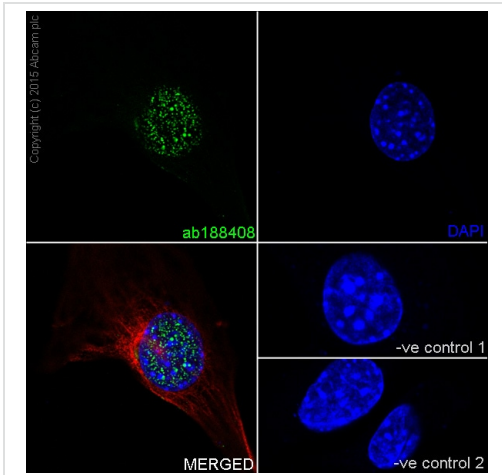
Tubulin is detected with Anti-alpha Tubulin mouse MAb ([ab7291](#)) at 1/1000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor[®] 594) ([ab150120](#)) secondary antibody at 1/1000 dilution (red).

The negative controls are as follows:

-ve control 1: ab188408 at 1/2000 dilution followed by Goat Anti-Mouse IgG H&L (Alexa Fluor[®] 594) ([ab150120](#)) secondary antibody at 1/1000 dilution.

-ve control 2: [ab7291](#) Anti-alpha Tubulin mouse MAb ([ab7291](#)) at 1/1000 dilution followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) ([ab150077](#)) secondary antibody at 1/1000 dilution.

Immunocytochemistry/ Immunofluorescence - Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408)



Immunocytochemistry/ Immunofluorescence - Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408)

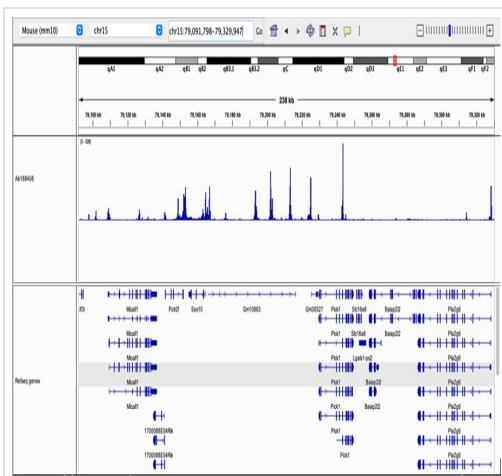
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized NIH/3T3 (Mouse embryonic fibroblast cells) cells labeling CTCF with ab188408 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution (green). Confocal image showing nuclear staining on NIH/3T3 cell line. The nuclear counter stain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin mouse MAb (**ab7291**) at 1/1000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) (**ab150120**) secondary antibody at 1/1000 dilution (red).

The negative controls are as follows:

-ve control 1: ab188408 at 1/2000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) (**ab150120**) secondary antibody at 1/1000 dilution.

-ve control 2: Anti-alpha Tubulin mouse MAb (**ab7291**) at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/1000 dilution.



CUT&Tag sequencing - Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408)


This experiment and image is courtesy of Dr Marek Bartosovic, Gonçalo Castelo-Branco Group, Karolinska Institutet.

CUT&Tag-seq was performed using 200,000 Oli-neu (Oligodendrocyte progenitor) cells. Cells were permeabilized with 0.05% Digitonin and 0.01% NP-40 for 3 minutes. A 1:100 dilution of Recombinant Anti-CTCF antibody [EPR18253] - ChIP Grade (ab188408) was used, along with a Guinea pig anti-rabbit Secondary. DNA was seq using Illumina NovaSeq S Prime to a depth of 24 million reads.

This image is courtesy of Dr Marek Bartosovic, Gonçalo Castelo-Branco Group, Karolinska Institutet.

The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.

Why choose a recombinant antibody?



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

Anti-CTCF antibody [EPR18253] - ChIP Grade
(ab188408)

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