

Anti-CTCF Antibody [JM10-61] - BSA and Azide free

HA750394



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat, Zebrafish
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, IP, FC, CUT&Tag-seq, ChIP
Molecular Wt:	Predicted band size: 83 kDa
Clone number:	JM10-61

Description: CTCF belongs to the zinc finger transcription factor family, and it recognizes unusually long and remarkably divergent DNA target sequences to influence expression of many various genes. The DNA-binding domain of CTCF is composed of 11 Zn fingers including 10 that are of C2H2 class, and 1 that is of C2HC class, and they are highly conserved between vertebrate species. CTCF functions as a repressor of the c-myc gene and as a regulator of lysozyme gene expression. In addition, CTCF associates with the essential activator domain in the promotor region of the amyloid beta-protein precursor (APP) gene to activate transcription of APP. Expression of CTCF up-regulates APP expression and thereby, enhances synapse formations between primary neurons during development. CTCF is ubiquitously expressed and localized to the nucleus. During terminal differentiation, CTCF is negatively regulated by differential phosphorylation and also by decreases in CTCF mRNA and protein expression.

Immunogen: Synthetic peptide within Human CTCF aa 685-727 / 727.

Positive control: HeLa cell lysate, 293T cell lysate, NIH/3T3 cell lysate, C6 cell lysate, PC-12 cell lysate, HeLa, human liver tissue, human kidney tissue, rat colon tissue, rat kidney tissue.

Subcellular location: Nucleoplasm, Chromosome, centromere.

Database links: SwissProt: P49711 Human | Q61164 Mouse | Q9R1D1 Rat

Storage Buffer: 1*PBS (pH7.4).

Storage Instruction: Store at 2-8℃. Avoid freeze.

Purity: Protein A affinity purified.

Hangzhou Huaan Biotechnology Co., Ltd.

Orders: 0086-571-88062880

Technical: 0086-571-89986345

Service mail: support@huabio.cn

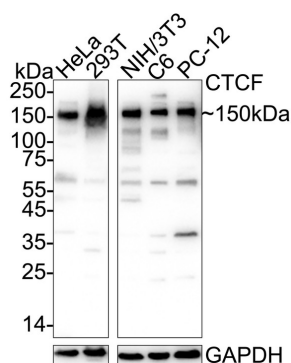
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Applications: WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Images

Fig1: This data was developed using ET1703-90, the same antibody clone in a different buffer formulation.

Western blot analysis of CTCF on different lysates with Rabbit anti-CTCF antibody (ET1703-90) at 1/1,000 dilution.



Lane 1: HeLa cell lysate

Lane 2: 293T cell lysate

Lane 3: NIH/3T3 cell lysate

Lane 4: C6 cell lysate

Lane 5: PC-12 cell lysate

Lysates/proteins at 20 µg/Lane.

Predicted band size: 83 kDa

Observed band size: 150 kDa

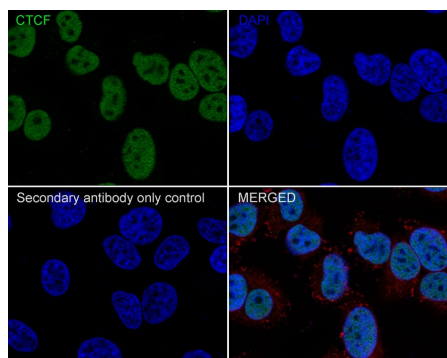
Exposure time: 4 seconds; ECL: K1801;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (ET1703-90) at 1/1,000 dilution was used in 5% NFDM/TBST at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

Fig2: This data was developed using ET1703-90, the same antibody clone in a different buffer formulation.

Immunocytochemistry analysis of HeLa cells labeling CTCF with Rabbit anti-CTCF antibody (ET1703-90) at 1/200 dilution.



Cells were fixed in 4% paraformaldehyde for 10 minutes at 37 °C, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-CTCF antibody (ET1703-90) at 1/200 dilution in 2% negative goat serum overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/200 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

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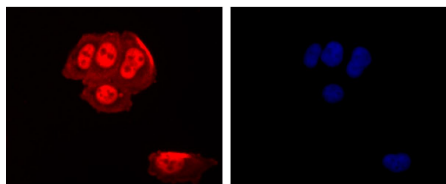


Fig3: This data was developed using ET1703-90, the same antibody clone in a different buffer formulation.

ICC staining of CTCF in MCF-7 cells (red). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1703-90, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®594 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

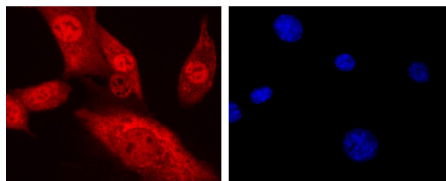


Fig4: This data was developed using ET1703-90, the same antibody clone in a different buffer formulation.

ICC staining of CTCF in NIH/3T3 cells (red). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (ET1703-90, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®594 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

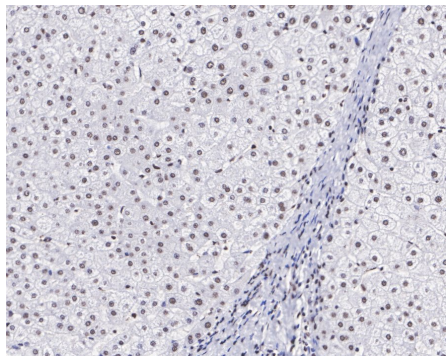


Fig5: This data was developed using ET1703-90, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded human liver tissue with Rabbit anti-CTCF antibody (ET1703-90) at 1/2,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1703-90) at 1/2,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

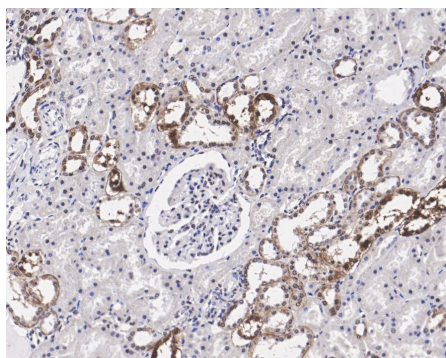


Fig6: This data was developed using ET1703-90, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded human kidney tissue with Rabbit anti-CTCF antibody (ET1703-90) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1703-90) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the

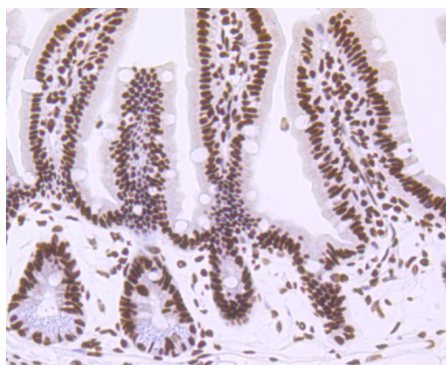


Fig7: This data was developed using ET1703-90, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded mouse colon tissue using anti-CTCF antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1703-90, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

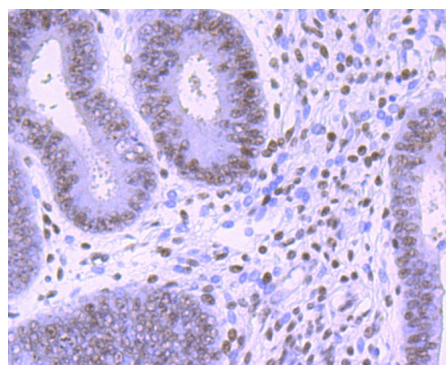


Fig8: This data was developed using ET1703-90, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded human endometrium tissue using anti-CTCF antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 8.0-8.4) for 20 minutes. The tissues were blocked in 5% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1703-90, 1/50) for 30 minutes at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

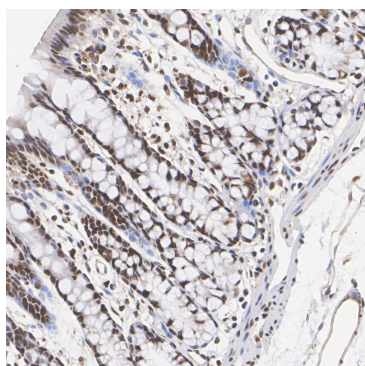


Fig9: This data was developed using ET1703-90, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded rat colon tissue with Rabbit anti-CTCF antibody (ET1703-90) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1703-90) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

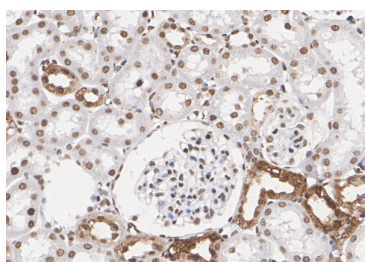


Fig10: This data was developed using ET1703-90, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded rat kidney tissue with Rabbit anti-CTCF antibody (ET1703-90) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1703-90) at 1/1,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

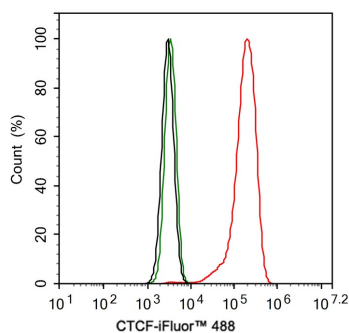


Fig11: This data was developed using ET1703-90, the same antibody clone in a different buffer formulation.

Flow cytometric analysis of HeLa cells labeling CTCF.

Cells were fixed and permeabilized. Then stained with the primary antibody (ET1703-90, 1ug/ml) (red) compared with Rabbit IgG Isotype Control (green). After incubation of the primary antibody at +4℃ for an hour, the cells were stained with a iFluor™ 488 conjugate-Goat anti-Rabbit IgG Secondary antibody (HA1121) at 1/1,000 dilution for 30 minutes at +4℃. Unlabelled sample was used as a control (cells without incubation with primary antibody; black).

Note: All products are “FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE”.

Background References

1. Harr JC et al. Directed targeting of chromatin to the nuclear lamina is mediated by chromatin state and A-type lamins. *J Cell Biol* 208:33-52 (2015).
2. Shi F et al. The expression of Pax6 variants is subject to posttranscriptional regulation in the developing mouse eyelid. *PLoS One* 8:e53919 (2013).

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