

Anti-HP1 alpha Antibody [SR44-03]

ET1602-8



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, IP
Molecular Wt:	Predicted band size: 22 kDa
Clone number:	SR44-03

Description: The family of Heterochromatin Protein 1 (HP1) ("Chromobox Homolog", CBX) are highly conserved adapter proteins, which have important functions in the cell nucleus. These functions include gene repression by heterochromatin formation, transcriptional activation, regulation of binding of cohesion complexes to centromere, sequestration of genes to nuclear periphery, transcriptional arrest, maintenance of heterochromatin integrity, gene repression at single nucleosome level and gene repression by heterochromatinization of euchromatin. Three different paralogs of HP1 are found in mammals, HP1 α , HP1 β and HP1 γ .

Immunogen: Recombinant protein within Human HP1 alpha aa 101-191 / 191.

Positive control: HeLa cell lysate, MCF7 cell lysate, NIH/3T3 cell lysate, RAW264.7 cell lysate, C6 cell lysate, PC-12 cell lysate, mouse brain tissue lysate, HeLa, NIH/3T3, human tonsil tissue, human breast tissue, rat uterus tissue, human uterus tissue, mouse uterus tissue.

Subcellular location: Nucleus, Chromosome

Database links: SwissProt: P45973 Human | Q61686 Mouse
Entrez Gene: 300266 Rat

Recommended Dilutions:

WB	1:500-1:2,000
IF-Cell	1:2,000
IF-Tissue	1:50-1:200
IHC-P	1:50-1:200
IP	Use at an assay dependent concentration.

Storage Buffer: 1*TBS (pH7.4), 0.05% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Shipped at 4°C. Store at +4°C short term (1-2 weeks). Store at -20°C long term.

Purity: Protein A affinity purified.

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Orders: 0086-571-88062880

Technical: 0086-571-89986345

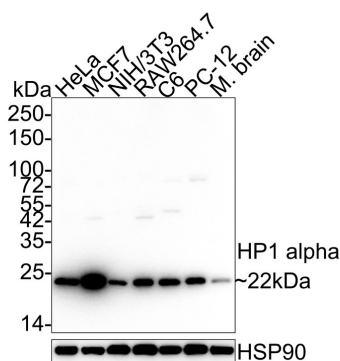
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Applications: WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Images

Fig1: Western blot analysis of HP1 alpha on different lysates with Rabbit anti-HP1 alpha antibody (ET1602-8) at 1/1,000 dilution.



Lane 1: HeLa cell lysate (10 µg/Lane)
 Lane 2: MCF7 cell lysate (10 µg/Lane)
 Lane 3: NIH/3T3 cell lysate (10 µg/Lane)
 Lane 4: RAW264.7 cell lysate (10 µg/Lane)
 Lane 5: C6 cell lysate (10 µg/Lane)
 Lane 6: PC-12 cell lysate (10 µg/Lane)
 Lane 7: Mouse brain tissue lysate (20 µg/Lane)

Predicted band size: 22 kDa

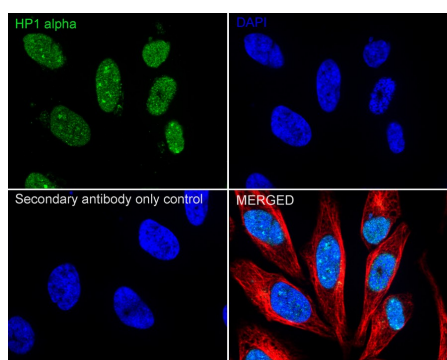
Observed band size: 22 kDa

Exposure time: 24 seconds;

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 (ET1602-8) 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: Immunocytochemistry analysis of HeLa cells labeling HP1 alpha with Rabbit anti-HP1 alpha antibody (ET1602-8) at 1/2,000 dilution.



Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-HP1 alpha antibody (ET1602-8) at 1/2,000 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (HA601187, red) was stained at 1/100 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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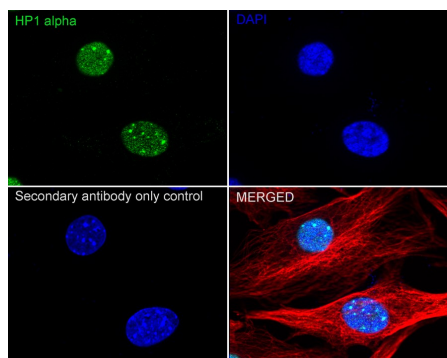
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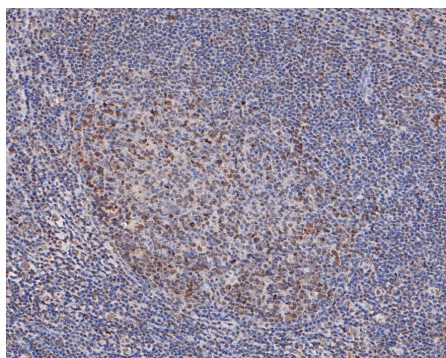
Fig3: Immunocytochemistry analysis of NIH/3T3 cells labeling HP1 alpha with Rabbit anti-HP1 alpha antibody (ET1602-8) at 1/2,000 dilution.



Cells were fixed in 4% paraformaldehyde for 15 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 15 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Rabbit anti-HP1 alpha antibody (ET1602-8) at 1/2,000 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

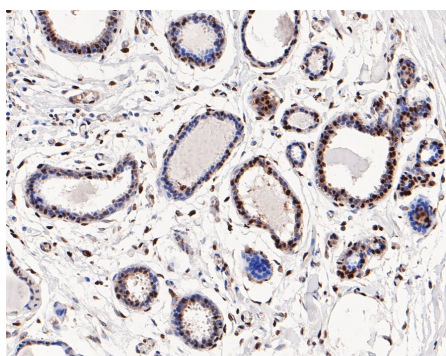
Beta tubulin (HA601187, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

Fig4: Immunohistochemical analysis of paraffin-embedded human tonsil tissue with Rabbit anti-HP1 alpha antibody (ET1602-8) at 1/50 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 (ET1602-8) at 1/50 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.

Fig5: Immunohistochemical analysis of paraffin-embedded human breast tissue with Rabbit anti-HP1 alpha antibody (ET1602-8) at 1/200 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 (ET1602-8) at 1/200 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.

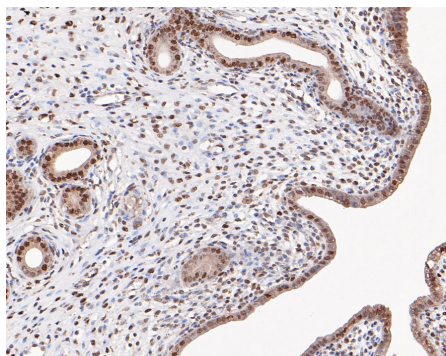


Fig6: Immunohistochemical analysis of paraffin-embedded rat uterus tissue with Rabbit anti-HP1 alpha antibody (ET1602-8) at 1/200 dilution.

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 (ET1602-8) at 1/200 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.

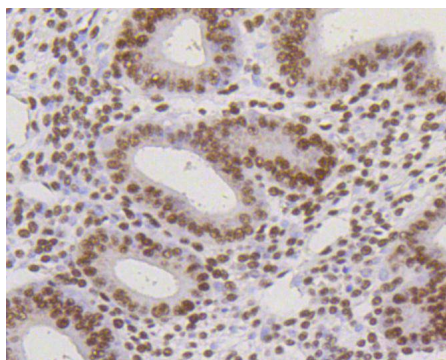


Fig7: Immunohistochemical analysis of paraffin-embedded human uterus tissue using anti-HP1 alpha antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1602-8, 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.

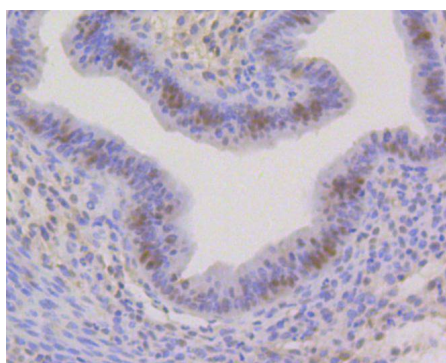


Fig8: Immunohistochemical analysis of paraffin-embedded mouse uterus tissue using anti-HP1 alpha antibody. The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 30 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (ET1602-8, 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.

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

Background References

1. "Initial characterization of the human central proteome." Burkard T.R., Planyavsky M., Kaupe I., Breitwieser F.P., Buerckstuemmer T., Bennett K.L., Superti-Furga G., Colinge J. BMC Syst. Biol. 5:17-17(2011).
2. "Lamin A rod domain mutants target heterochromatin protein 1alpha and beta for proteasomal degradation by activation of F-box protein, FBXW10." Chaturvedi P., Parnaik V.K. PLoS ONE 5:E10620-E10620(2010).

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