Anti-CEBP alpha Antibody [PSH20-43] - BSA and Azide free

HA751759

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IHC-P, IF-Cell, FC, IP
Molecular Wt:	Predicted band size: 38 kDa
Clone number:	PSH20-43

Description: The transcription factor C/EBP α (CCAAT-enhancer binding protein) is a heat-stable, sequence-specific DNA-binding protein that binds avidly to several different cis-regulatory DNA sequences commonly associated with viral and cellular genes transcribed by RNA polymerase II. C/EBP α regulates gene expression in a variety of tissues including liver, adipose, lung and intestine. C/EBP α is a basic region/leucine zipper transcription factor selectively expressed during the differentiation of liver, adipose tissue, blood cells and the endocrine pancreas. C/EBP α uses a bipartite structural motif to bind DNA and appears to function exclusively in terminally differentiated, growth-arrested cells. In the liver, C/EBP α is a transactivator of several genes, which are regulated by growth hormone. Growth hormone enhances not only the levels of C/EBP α mRNA and protein, but also the DNA binding activity of C/EBP α. C/EBP α functions as an important transcription factor that regulates different genes, including prolactin gene expression.

Positive control: THP-1 cell lysate, HepG2 cell lysate, LNCaP cell lysate, U-937 cell lysate, THP-1, human lung tissue, mouse liver tissue, mouse spleen tissue, rat liver tissue, rat lung tissue, rat spleen tissue, rat colon tissue.

Subcellular location: Nucleus, nucleolus.

Database links: SwissProt: P49715 Human | P53566 Mouse | P05554 Rat

Recommended Dilutions:

WB	1:5,000
IHC-P	1:200-1:1,000
IF-Cell	1:1,000
FC	1:1,000
IP	1-2μg/sample

Storage Buffer: PBS (pH7.4).

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

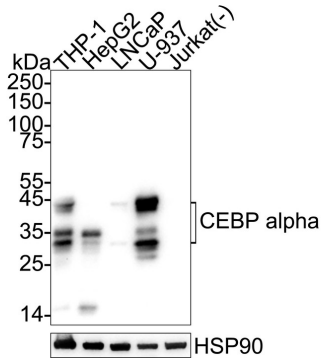

Fig1: Western blot analysis of CEBP alpha on different lysates with Rabbit anti-CEBP alpha antibody (HA751759) at 1/5,000 dilution.

Lane 1: THP-1 cell lysate (20 µg/Lane)
Lane 2: HepG2 cell lysate (20 µg/Lane)
Lane 3: LNCaP cell lysate (20 µg/Lane)
Lane 4: U-937 cell lysate (20 µg/Lane)
Lane 5: Jurkat cell lysate (negative) (20 µg/Lane)

Predicted band size: 38 kDa
Observed band size: 30-42 kDa
Exposure time: 20 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 (HA751759) at 1/5,000 dilution was used in primary antibody dilution (K1803) at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

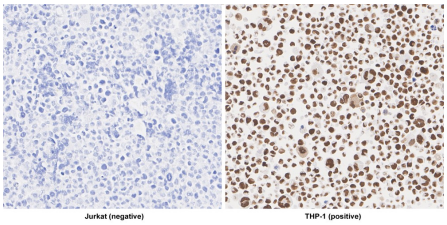

Fig2: Immunohistochemical analysis of paraffin-embedded Jurkat (left, negative) and THP-1 (right, positive) cells with Rabbit anti-CEBP alpha antibody (HA751759) at 1/1,000 dilution.

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 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA751759) 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.

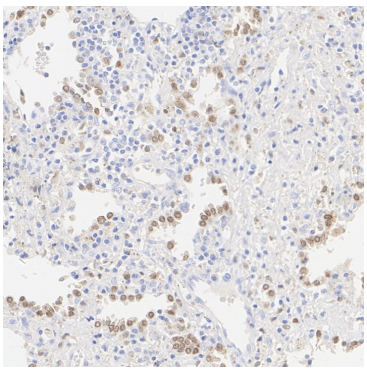

Fig3: Immunohistochemical analysis of paraffin-embedded human lung tissue with Rabbit anti-CEBP alpha antibody (HA751759) at 1/200 dilution.

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 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA751759) 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.

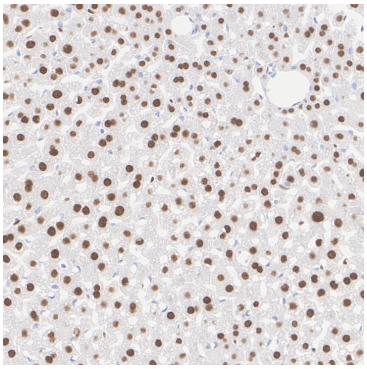

Fig4: Immunohistochemical analysis of paraffin-embedded mouse liver tissue with Rabbit anti-CEBP alpha antibody (HA751759) at 1/1,000 dilution.

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 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA751759) 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.

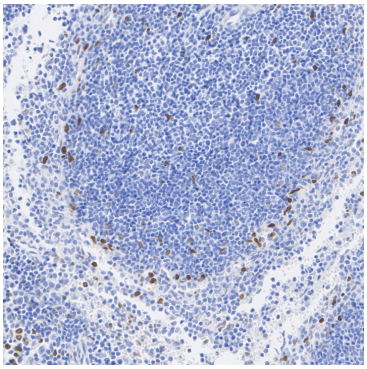

Fig5: Immunohistochemical analysis of paraffin-embedded mouse spleen tissue with Rabbit anti-CEBP alpha antibody (HA751759) at 1/1,000 dilution.

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 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA751759) 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.

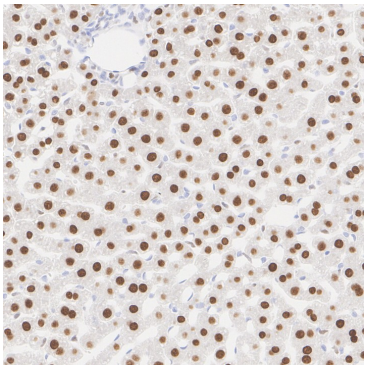

Fig6: Immunohistochemical analysis of paraffin-embedded rat liver tissue with Rabbit anti-CEBP alpha antibody (HA751759) at 1/1,000 dilution.

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 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA751759) 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.

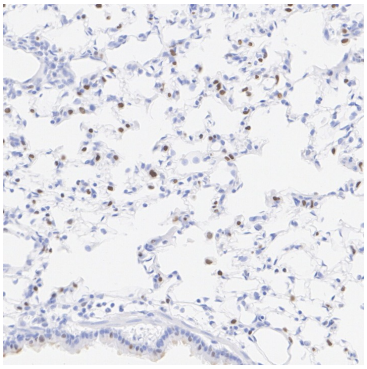

Fig7: Immunohistochemical analysis of paraffin-embedded rat lung tissue with Rabbit anti-CEBP alpha antibody (HA751759) at 1/1,000 dilution.

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 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA751759) 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.

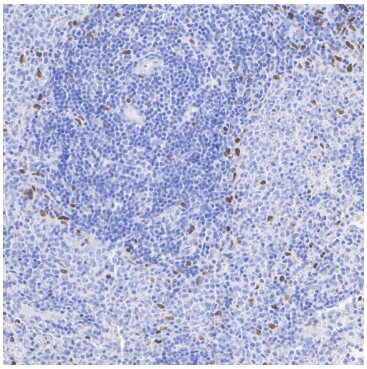

Fig8: Immunohistochemical analysis of paraffin-embedded rat spleen tissue with Rabbit anti-CEBP alpha antibody (HA751759) at 1/1,000 dilution.

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 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA751759) 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.

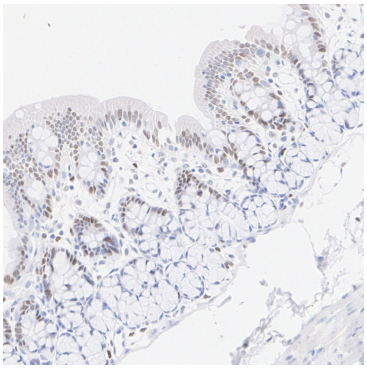

Fig9: Immunohistochemical analysis of paraffin-embedded rat colon tissue with Rabbit anti-CEBP alpha antibody (HA751759) at 1/200 dilution.

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 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA751759) 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.

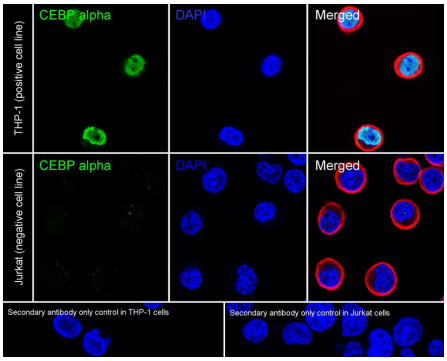

Fig10: Immunocytochemistry analysis of THP-1 (positive) and Jurkat (negative) labeling CEBP alpha with Rabbit anti-CEBP alpha antibody (HA751759) at 1/1,000 dilution.

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-CEBP alpha antibody (HA751759) at 1/1,000 dilution in 1% BSA in PBST overnight at 4 ℃. 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℃. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

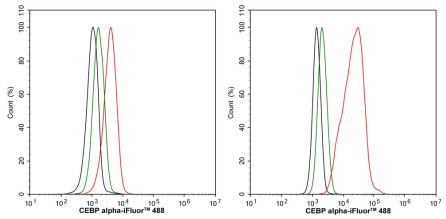

Fig11: Flow cytometric analysis of Jurkat (left, negative) and THP-1 (right, positive) cells labeling CEBP alpha.

Cells were fixed and permeabilized. Then stained with the primary antibody (HA751759, 1/1,000) (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).

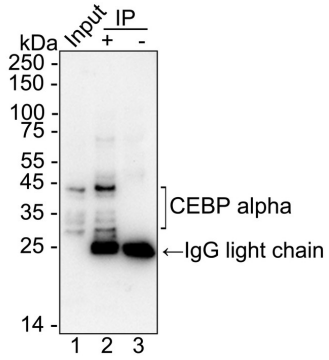

Fig12: CEBP alpha was immunoprecipitated from 0.2 mg LNCaP cell lysate with HA751759 at 2 µg/10 µl beads. Western blot was performed from the immunoprecipitate using HA751759 at 1/5,000 dilution. HRP Conjugated Anti-Rabbit IgG for IP Nano-secondary antibody at 1/5,000 dilution was used for 1 hour at room temperature.

Lane 1: LNCaP cell lysate (input)
Lane 2: HA751759 IP in LNCaP cell lysate
Lane 3: Rabbit IgG instead of HA751759 in LNCaP cell lysate

Blocking/Dilution buffer: primary antibody dilution (K1803)
Exposure time: 3 minutes; ECL: K1801

Background References

1. Christou-Kent M et al. CEBPA phase separation links transcriptional activity and 3D chromatin hubs. Cell Rep. 2023 Aug
2. Georgi JA et al. Prognostic impact of CEBPA mutational subgroups in adult AML. Leukemia. 2024 Feb