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Product datasheet

Anti-CREB antibody [LB9] ab178322

KO VALIDATED

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Overview

Product name	Anti-CREB antibody [LB9]
Description	Mouse monoclonal [LB9] to CREB
Host species	Mouse
Tested applications	**Suitable for:** ICC/IF, WB, IP, IHC-P
Species reactivity	**Reacts with:** Mouse, Rat, Human, African green monkey **Predicted to work with:** Non human primates
Immunogen	Recombinant full length protein corresponding to Human CREB aa 1-341. Sequence: MTMESGAENQ QSGDAAVTEA ENQQMTVQAQ PQIATLAQVS MPAAHATSSA PTVTLVQLPN GQTVQVHGVI QAAQPSVIQS PQVQTVQSSC KDLKRLFSGT QISTIAESED SQESVDSVTD SQKRREILSR RPSYRKILND LSSDAPGVPR IEEEKSEEET SAPAITTVTV PTPIYQTSSG QYIAITQGGA IQLANNGTDG VQGLQTLTMT NAAATQPGTT ILQYAQTTDG QQILVPSNQV VVQAASGDVQ TYQIRTAPTS TIAPGVVMAS SPALPTQPAE EAARKREVRL MKNREAAREC RRKKKEYVKC LENRVAVLEN QNKTLIEELK ALKDLYCHKS D Database link: **P16220** Run BLAST with Run BLAST with
Positive control	HeLa cells; Human bladder and rectal cancer tissues; Human tonsil tissue; Human Jurkat, NIH/3T3, C2C12, MDA-MB-231, T98G, HeLa, 293T, HepG2, A431, A549, K562, MCF7 and U2OS whole cell lysates; Mouse MEF, 3T3L1 and C2C12 whole cell lyastes; Rat NRK whole cell lysate; Monkey COS7 whole cell lysate.
General notes	The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

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	Preservative: 0.05% Sodium azide Constituents: 0.1% BSA, 99% PBS
Purity	Protein A purified
Clonality	Monoclonal
Clone number	LB9
Isotype	IgG1

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab178322 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
ICC/IF		1/500.
WB	★★★★☆ (1)	1/500. Detects a band of approximately 43 kDa (predicted molecular weight: 36 kDa).
IP		Use at an assay dependent concentration. Suggested use 2 µg.
IHC-P		1/200. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

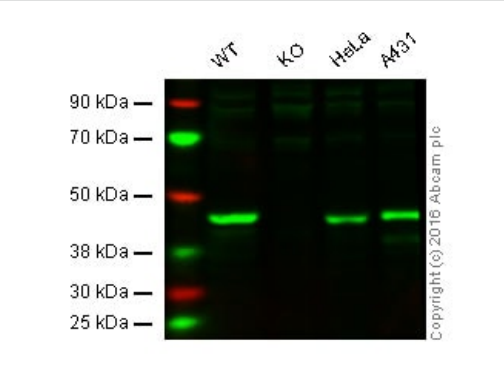
Target

Function This protein binds the cAMP response element (CRE), a sequence present in many viral and cellular promoters. CREB stimulates transcription on binding to the CRE. Transcription activation is enhanced by the TORC coactivators which act independently of Ser-133 phosphorylation. Implicated in synchronization of circadian rhythmicity.

Involvement in disease Defects in CREB1 may be a cause of angiomatoid fibrous histiocytoma (AFH) [MIM:612160]. A distinct variant of malignant fibrous histiocytoma that typically occurs in children and adolescents and is manifest by nodular subcutaneous growth. Characteristic microscopic features include lobulated sheets of histiocyte-like cells intimately associated with areas of hemorrhage and cystic pseudovascular spaces, as well as a striking cuffing of inflammatory cells, mimicking a lymph node metastasis. Note=A chromosomal aberration involving CREB1 is found in a patient with angiomatoid fibrous histiocytoma. Translocation t(2;22)(q33;q12) with CREB1 generates a EWSR1/CREB1 fusion gene that is most common genetic abnormality in this tumor type.

Sequence similarities	Belongs to the bZIP family. Contains 1 bZIP domain. Contains 1 KID (kinase-inducible) domain.
Post-translational modifications	Stimulated by phosphorylation. Phosphorylation of both Ser-133 and Ser-142 in the SCN regulates the activity of CREB and participates in circadian rhythm generation. Phosphorylation of Ser-133 allows CREBBP binding (By similarity). Phosphorylated upon DNA damage, probably by ATM or ATR. Sumoylated by SUMO1. Sumoylation on Lys-304, but not on Lys-285, is required for nuclear localization of this protein. Sumoylation is enhanced under hypoxia, promoting nuclear localization and stabilization.
Cellular localization	Nucleus.

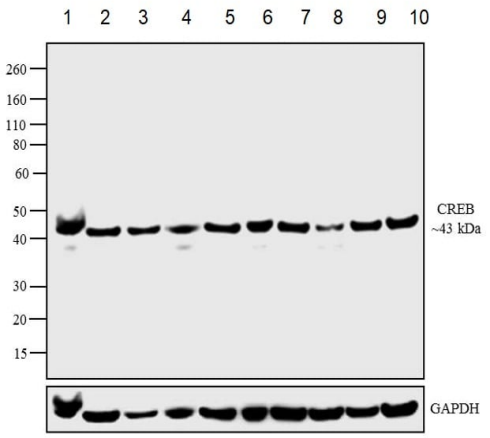
Images



Western blot - Anti-CREB antibody [LB9] (ab178322)

Lane 1: Wild-type HAP1 cell lysate (20 µg)
Lane 2: CREB knockout HAP1 cell lysate (20 µg)
Lane 3: HeLa cell lysate (20 µg)
Lane 4: A431 cell lysate (20 µg)
Lanes 1 - 4: Merged signal (red and green). Green - ab178322 observed at 44 kDa. Red - loading control, **ab18058**, observed at 124 kDa.

ab178322 was shown to recognize CREB when CREB knockout samples were used, along with additional cross-reactive bands. Wild-type and CREB knockout samples were subjected to SDS-PAGE. Ab178322 and **ab18058** (loading control to Vinculin) were diluted at 1/250 and 1/10,000 dilution respectively and incubated overnight at 4C. Blots were developed with IRDye® 800CW Goat anti-Rabbit IgG (H + L) and IRDye® 680 Goat anti-Mouse IgG (H + L) secondary antibodies at 1/10,000 dilution for 1 hour at room temperature before imaging.



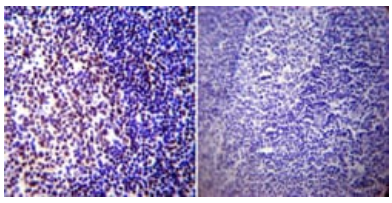
Western blot - Anti-CREB antibody [LB9] (ab178322)

All lanes : Anti-CREB antibody [LB9] (ab178322) at 1/500 dilution

Lane 1 : Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate
Lane 2 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysate
Lane 3 : K-562 (Human chronic myelogenous leukemia lymphoblast cell line) whole cell lysate
Lane 4 : Hep G2 (Human liver hepatocellular carcinoma cell line) whole cell lysate
Lane 5 : NIH/3T3 (Mouse embryonic fibroblast cell line) whole cell lysate
Lane 6 : C2C12 whole cell lysate
Lane 7 : U-2 OS (Human bone osteosarcoma epithelial cell line) whole cell lysate
Lane 8 : A549 (Human lung carcinoma cell line) whole cell lysate
Lane 9 : MDA-MB-231 (Human breast adenocarcinoma cell line) whole cell lysate
Lane 10 : T98G whole cell lysate

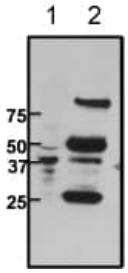
Lysates/proteins at 30 µg per lane.

Predicted band size: 36 kDa
Observed band size: 43 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CREB antibody [LB9] (ab178322)

Immunohistochemical analysis of deparaffinized Human tonsil tissue labeling CREB with ab178322 at 1/100 dilution followed by DAB staining.



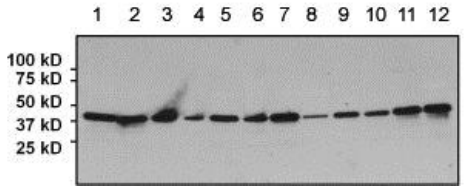
Immunoprecipitation - Anti-CREB antibody [LB9] (ab178322)

Immunoprecipitation analysis of CREB was performed on untreated 293T cells. The antigen:antbody complex was formed by binding 500µg whole cell lysate with 2µg ab178322 and captured on 50µl Protein A/G Plus Agarose.

Lane 1: 293T lysate

Lane 2: IP of 293T lysate

WB detection used ab178322 at 1/1000 followed with a goat anti-mouse-HRP secondary antibody at 1/20,000.



Western blot - Anti-CREB antibody [LB9] (ab178322)

All lanes : Anti-CREB antibody [LB9] (ab178322) at 1/500 dilution

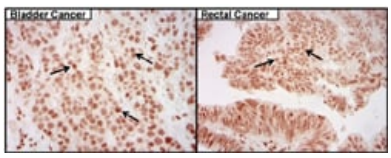
Lane 1 : HeLa whole cell lysate
Lane 2 : 293T whole cell lysate
Lane 3 : A431 whole cell lysate
Lane 4 : A549 whole cell lysate
Lane 5 : MEF whole cell lysate
Lane 6 : MCF7 whole cell lysate
Lane 7 : U2OS whole cell lysate
Lane 8 : K562 whole cell lysate
Lane 9 : COS7 whole cell lysate
Lane 10 : 3T3L1 whole cell lysate
Lane 11 : C2C12 whole cell lysate
Lane 12 : NRK whole cell lysate

Lysates/proteins at 25 µg per lane.

Secondary
All lanes : goat anti-mouse-HRP at 1/20000 dilution

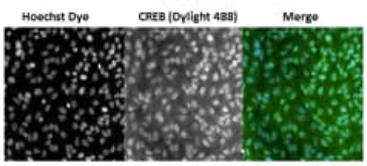
Developed using the ECL technique.

Predicted band size: 36 kDa



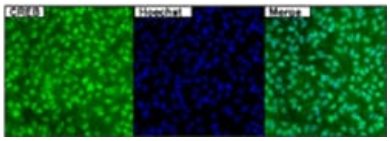
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-CREB antibody [LB9] (ab178322)

Immunohistochemical analysis of deparaffinized Human bladder cancer (left) and rectal cancer (right) tissues labeling CREB with ab178322 at 1/200 dilution followed by DAB staining.



Immunocytochemistry/ Immunofluorescence - Anti-CREB antibody [LB9] (ab178322)

Immunofluorescent analysis of untreated HeLa cells (formalin-fixed, 0.1% Triton X-100 permeabilized) labeling CREB with ab178322 at 1/400 dilution (green) followed with DyLight 488 goat anti-mouse IgG secondary antibody at 1/400 dilution. Nuclei (blue) were stained with Hoechst 33342 dye.



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