abcam

ab32106

Anti-ELK1 antibody [E277]

Recombinant | RabMAb | Advanced Validation | KO Validated | 20ul selling size

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 50% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	E277
Purification technique	Affinity purification Protein A
Specificity	This antibody does not cross react with other ETS family members
Epitope	ab32106 reacts with an epitope located in the C terminal region of ELK1.
Concentration	0.244 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

ChIC/CUT&RUN-seq

Tested

Species	Human

Dilution info	-
Notes	-

Predicted

Species	Mouse, Rat
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human
Dilution info	1/50
Notes	Perform heat-mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Predicted

Species	Mouse, Rat
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	1/500
Notes	-

Predicted

Species	Mouse, Rat
Dilution info	-

Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/500
Notes	-

Predicted

Species	Mouse, Rat
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/20
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Predicted

Species	Mouse, Rat
Dilution info	-
Notes	-

Target data

See full target information ELK1

Function	Transcription factor that binds to purine-rich DNA sequences (PubMed:10799319, PubMed:7889942). Forms a ternary complex with SRF and the ETS and SRF motifs of the serum response element (SRE) on the promoter region of immediate early

genes such as FOS and IER2 (PubMed:1630903). Induces target gene transcription upon JNK and MAPK-signaling pathways stimulation (PubMed:7889942).

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

Notes

Patented technology
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?
This product is a recombinant monoclonal antibody, which offers several advantages including:

- High batch-to-batch consistency and reproducibility
- Improved sensitivity and specificity
- Long-term security of supply
- Animal-free batch production

For more information, read more on recombinant antibodies.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	ELK1 also known as ELK-1 or ETS-domain protein ELK1 is a transcription factor involved in regulating gene expression. It is a member of the ETS family of transcription factors and has a molecular mass of approximately 54 kDa. ELK1 is mainly expressed in the nucleus of cells and plays a significant role in various tissues by interacting with other proteins. It has sequence-specific DNA-binding ability which allows it to influence the transcription of target genes.

Biological function summary	ELK1 functions in the regulation of gene expression related to cellular proliferation and differentiation. It often forms a ternary complex with serum response factor (SRF) on serum response elements (SREs) to modulate the transcriptional responses of genes important for cell growth. The activation of ELK1 is phosphorylation-dependent primarily by mitogen-activated protein kinases (MAPKs) which further facilitates its role as a transcription regulator. This phosphorylation increases the transcriptional activity enabling ELK1 to actively participate in cellular responses to external signals.
Pathways	ELK1 participates prominently in the MAPK/ERK signaling pathway and the JNK signaling pathway. Through these pathways ELK1 gets activated influencing the transcription of immediate early genes responsible for cell cycle progression and apoptosis. ELK1 interacts with other proteins in these pathways such as MEK and ERK leading to a coordinated response to growth signals. These interactions underline its importance in signal transduction processes and cellular response mechanisms.
Associated diseases and disorders	ELK1 has associations with certain cancers and neurodegenerative disorders. In particular its dysregulation has been noted in the context of breast cancer where ELK1 is implicated in abnormal cell proliferation. Similarly in neurodegenerative diseases like Alzheimer's ELK1's altered activity levels can affect neuron function and survival. Other proteins such as amyloid precursor protein in Alzheimer's or estrogen receptor in breast cancer connect with ELK1 influencing disease progression through complex signaling networks.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

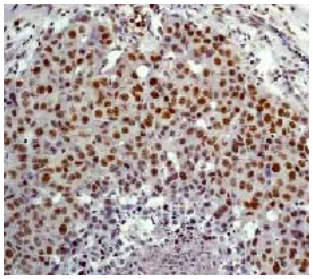
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:

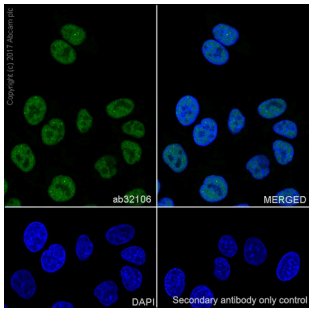
9 product images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ELK1 antibody [E277] (ab32106)

ELK1 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-ELK1 antibody

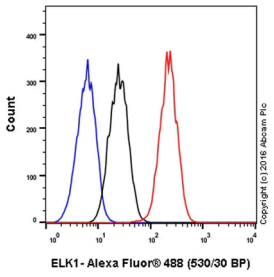
Immunohistochemical analysis of paraffin embedded human breast carcinoma using ab32106 at a dilution of 1/50
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-ELK1 antibody [E277] (ab32106)

ELK1 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-ELK1 antibody

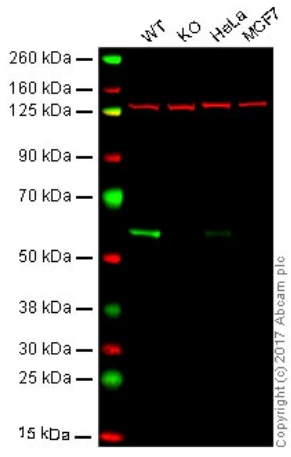
Immunocytochemistry/ Immunofluorescence analysis of A549 (human lung carcinoma epithelial cell) cells labeling ELK1 with purified ab32106 at 1/500 dilution (3.8 µg/mL). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) was used as the secondary antibody at 1/1000 (2 µg/mL) dilution. DAPI (blue) was used as nuclear counterstain.



Flow Cytometry (Intracellular) - Anti-ELK1 antibody [E277] (ab32106)

ELK1 Flow Cytometry (Intracellular) staining using rabbit Anti-ELK1 antibody

Intracellular Flow Cytometry analysis of HeLa (human cervix adenocarcinoma) cells labeling ELK1 with unpurified ab32106 at 1/20 dilution (10ug/mL) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit IgG (Alexa Fluorr® 488) at 1/2000 dilution was used as the secondary antibody. Rabbit monoclonal IgG (Black) was used as the isotype control, cells without incubation with primary antibody and secondary antibody (Blue) were used as the unlabeled control.



Western blot - Anti-ELK1 antibody [E277] (ab32106)

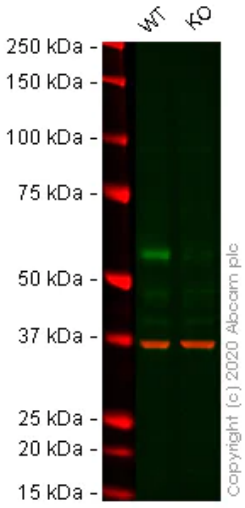
ELK1 Western blot staining using rabbit Anti-ELK1 antibody

Lane 1: Wild-type HAP1 whole cell lysate (20 µg)
Lane 2: ELK1 knockout HAP1 whole cell lysate (20 µg)
Lane 3: HeLa positive whole cell lysate (21 µg)
Lane 4: MCF7 negative whole cell lysate (20 µg)
Lanes 1 - 4: Merged signal (red and green). Green - ab32106 observed at 55 kDa. Red - loading control, ab18058, observed at 130 kDa.

ab32106 was shown to specifically react with ELK1 in wild-type HAP1 cells as signal was lost in ELK1 knockout cells. Wild-type and ELK1 knockout samples were subjected to SDS-PAGE. ab32106

and ab18058 (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/500 dilution and 1/20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed ab216776 secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.
All lanes:
Western blot - Anti-ELK1 antibody [E277] (ab32106)

Predicted band size: 45 kDa



Western blot - Anti-ELK1 antibody [E277] (ab32106)

ELK1 Western blot staining using rabbit Anti-ELK1 antibody

Lanes 1- 2: Merged signal (red and green). Green - ab32106 observed at 55 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (ab8245) observed at 37 kDa.
ab32106 was shown to react with ELK1 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line ab261764 (knockout cell lysate ab256904) was used. Wild-type HeLa and ELK1 knockout HeLa cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab32106 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) overnight at 4°C at a 1 in 500 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye®800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye®680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-ELK1 antibody [E277] (ab32106) at 1/500 dilution

Lane 1:
Wild-type HeLa cell lysate at 20 µg

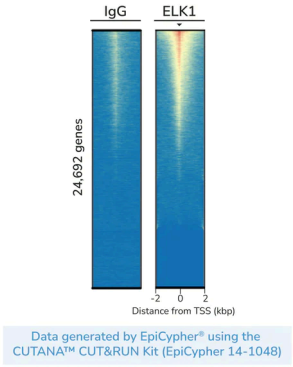
Lane 2:
ELK1 knockout HeLa cell lysate at 20 µg

Lane 2:
Western blot - Human ELK1 knockout HeLa cell line (ab261764)

Performed under reducing conditions.

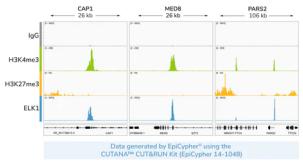
Predicted band size: 45 kDa

Observed band size: 55 kDa



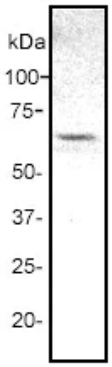
ChIC/CUT&RUN sequencing - Anti-ELK1 antibody [E277] (ab32106)

CUT&RUN profiling with ELK1 antibody demonstrates robust genome-wide enrichment in cells. Heatmaps of genome-wide signal flanking annotated transcription start sites (TSSs, +/- 2 kbp) display CUT&RUN data generated using the CUTANA™ CUT&RUN Kit (EpiCypher 14-1048) with ELK1 antibody (Abcam ab32106, 0.5 µg). 500,000 HeLa cells were used per reaction. IgG antibody was included as a negative control to assess non-specific background. Libraries were prepared using the CUTANA™ CUT&RUN Library Prep Kit (EpiCypher 14-1001). Sequencing was performed with paired-end 50 bp reads, and data were processed on CUTANA™ Cloud (cloud.epicypher.com) by alignment to the hg38 genome. Heatmaps were generated using ChAsE (Younesy et al., *Bioinformatics* 2016; PMID 27378294). Row-linked data are ranked by intensity relative to ELK1, with red indicating high localized enrichment and blue denoting background.



ChIC/CUT&RUN sequencing - Anti-ELK1 antibody [E277] (ab32106)

CUT&RUN profiling with ELK1 antibody reveals the expected genomic enrichment pattern in cells. Representative genome browser tracks show CUT&RUN data generated using the CUTANA™ CUT&RUN Kit (EpiCypher 14-1048) with ELK1 antibody (Abcam ab32106, 0.5 µg). 500,000 HeLa cells were used per reaction. IgG, H3K4me3, and H3K27me3 antibodies were included as controls to assess non-specific background, active promoters, and repressed chromatin, respectively. Libraries were prepared using the CUTANA™ CUT&RUN Library Prep Kit (EpiCypher 14-1001). Sequencing was performed with paired-end 50 bp reads, and data were processed on CUTANA™ Cloud (cloud.epicypher.com) by alignment to the hg38 genome. Images were generated using Integrative Genomics Viewer (IGV, Broad Institute).



Western blot - Anti-ELK1 antibody [E277] (ab32106)

All lanes:
Western blot - Anti-ELK1 antibody [E277] (ab32106) at 1/500 dilution

Predicted band size: 45 kDa

Observed band size: 62 kDa

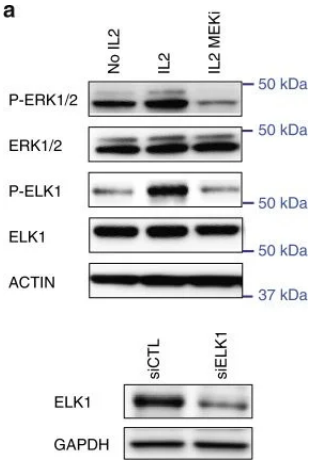


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Western blot - Anti-ELK1 antibody [E277] (ab32106)

ELK1 Western blot staining using rabbit Anti-ELK1 antibody

Western Blotting using Anti-ELK1 antibody [E277], ab32106. Publication image from Caron, G. et al., 2017, Nat Commun, 29129929. Legend direct from paper.

ELK1 is a mediator of IL-2 signalling and binds within BACH2 super-enhancer. a Upper panel: MAP kinase activity analysed by phospho-specific immunoblotting in D2-activated B cells, starved and stimulated or not for 5 min with IL-2 and MEK inhibitor (MEKi) or DMSO; IL-2 signal triggers ERK and ELK1 phosphorylation (P-ERK1/2, P-ELK1) which is blocked with MEKi treatment. β-ACTIN was used as loading control. One representative of two experiments is shown. Lower panel: Protein expression levels of total ELK1 analysed by immunoblotting 2 days after siELK1 electroporation. GAPDH was used as loading control. b Effect of ELK1 inhibition on gene expression levels analysed by QRT-PCR in D3 B cells primed (IL2) or not (No IL2) with IL-2 for 24 h and electroporated at D1 with siELK1 or siCTL. BACH2, IRF8 and MYD88 expression levels in CFSElo-cell sorted compartments were normalised to expression levels in IL-2 primed control-B cells. Means ± s.e.m. of three independent experiments. c Effect of siELK1 on the ability to differentiate into plasmablasts (CD38hi) in condition that favours plasma cell generation (IL-2 priming). Data from six independent experiments are presented relative to CD38hi cell number generated by IL-2-primed and siCTL-electroporated cells at D1 (fixed to 100%). Means ± s.e.m. d Human BACH2 locus including the position of the ELK1 consensus binding site (MATINSPECTOR prediction) in intron 1, within an enhancer region (FANTOM5 prediction) and the positions detected by QPCR for the ChIP assays within the proximal promoter and intron 1 identified by dark boxes. Numbers indicate positions relative to the TSS (+1). Ex1 means Exon 1. e ChIP assay of the in vivo binding of ELK1 in IL-2-primed D3 B cells. DNA immunoprecipitated by total ELK1 antibody or immunoglobulin G (IgG CTL) was amplified by real-time PCR using primers flanking the putative ELK1 binding site in BACH2 intron 1 (predicted BACH2 enhancer position), known ELK1 binding regions in MCL1, MYD88 and FOS genes, and negative control primers as a reference (negative site). Means ± s.e.m. of four independent experiments. f ChIP analysis of histone modifications and p300 binding in IL-2-primed D3 B cells. Immunoprecipitated DNA was amplified by QPCR using primers in the BACH2 proximal promoter (BACH2 promoter) and predicted enhancer (BACH2 enhancer). Silent locus (negative site) and GAPDH specific positive control primers were used.

Data are representative of two experiments (average and standard deviation of binding events per 1000 cells) with IgG control ChIP values (IgG CTL) shown separately. For experiments shown in b, c, e, *p < 0.05 **p < 0.01 ***p < 0.005, two-tailed unpaired Student’ s t-test

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