abcam

Product datasheet

Anti-ELK1 antibody [E277] ab32106

KO VALIDATED Recombinant RabMAb

46 References 7 Images

Overview

Product name	Anti-ELK1 antibody [E277]
Description	Rabbit monoclonal [E277] to ELK1
Host species	Rabbit
Specificity	This antibody does not cross react with other ETS family members
Tested applications	**Suitable for:** Flow Cyt (Intra), WB, IHC-P, ICC/IF
Species reactivity	**Reacts with:** Human **Predicted to work with:** Mouse, Rat
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Epitope	ab32106 reacts with an epitope located in the C terminal region of ELK1.
Positive control	IHC-P: Human breast carcinoma ICC/IF: A549 cells WB: HeLa, MCF7 and HAP1 cell lysates. Flow Cyt (intra): HeLa cells.
General notes	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 production For more information **see here**. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

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. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 49% PBS, 50% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified

Clonality	Monoclonal
Clone number	E277
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab32106 in the following tested applications.

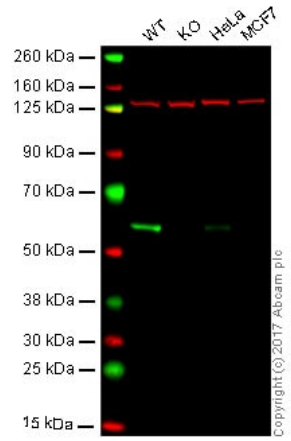
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		1/20. **ab172730** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
WB		1/500. Detects a band of approximately 62 kDa (predicted molecular weight: 45 kDa).
IHC-P		1/50. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ICC/IF		1/500.

Target

Function	Stimulates transcription. Binds to purine-rich DNA sequences. Can form a ternary complex with the serum response factor and the ETS and SRF motifs of the fos serum response element.
Tissue specificity	Lung and testis.
Sequence similarities	Belongs to the ETS family. Contains 1 ETS DNA-binding domain.
Post-translational modifications	Sumoylation represses transcriptional activator activity as it results in recruitment of HDAC2 to target gene promoters which leads to decreased histone acetylation and reduced transactivator activity. It also regulates nuclear retention. On mitogenic stimulation, phosphorylated on C-terminal serine and threonine residues by MAPK1. Ser-383 and Ser-389 are the preferred sites for MAPK1. In vitro, phosphorylation by MAPK1 potentiates ternary complex formation with the serum responses factors, SRE and SRF. Phosphorylation leads to loss of sumoylation and restores transcriptional activator activity.
Cellular localization	Nucleus.

Images

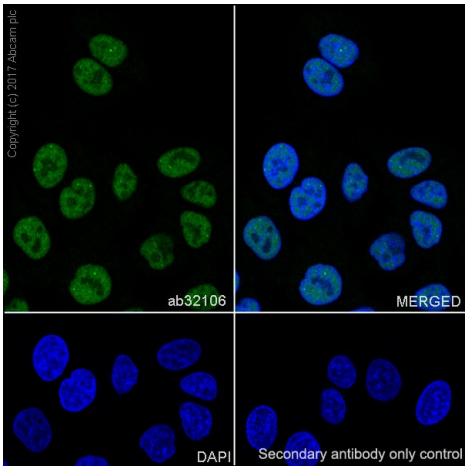


Western blot - Anti-ELK1 antibody [E277] (ab32106)

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)

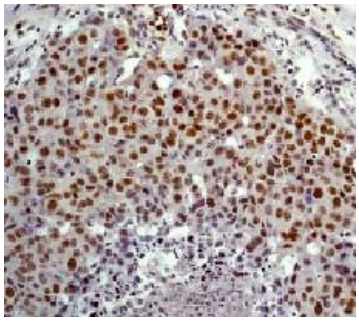
Lanes 1 - 4: Merged signal (red and green). Green - ab32106 observed at 55 kDa. Red - loading control, **ab18058**, observed at 130 kDa.

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.



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

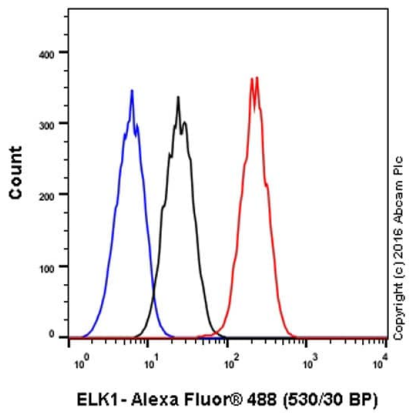
Immunocytochemistry/ Immunofluorescence analysis of A549 (human lung carcinoma epithelial cell) cells labeling ELK1 with purified ab32106 at 1/500 dilution (3.8 µg/mL). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**)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



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

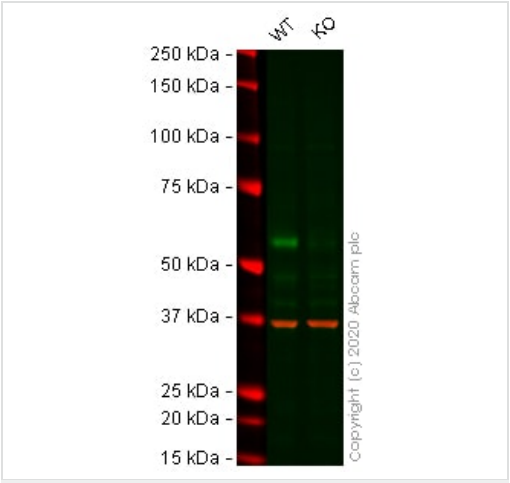
Immunohistochemical analysis of paraffin embedded human breast carcinoma using ab32106 at a dilution of 1/50

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



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

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 Fluor® 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)

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

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : ELK1 knockout HeLa cell lysate

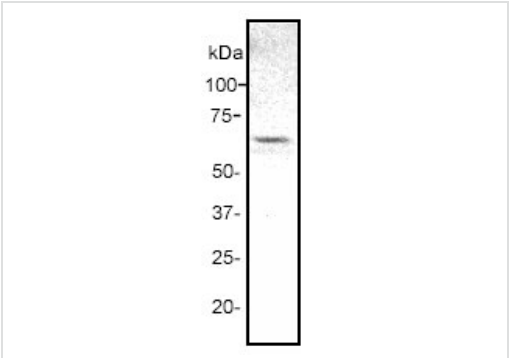
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 45 kDa
Observed band size: 55 kDa

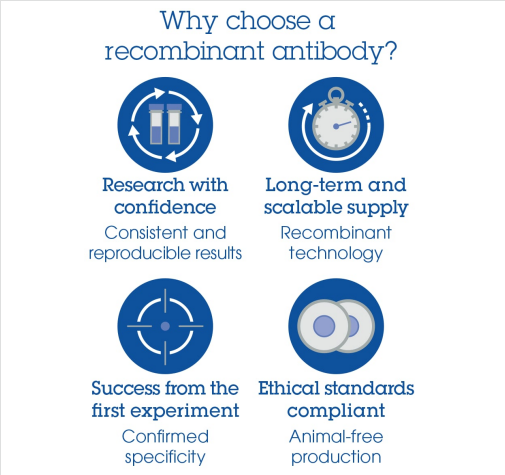
Lanes 1- 2: Merged signal (red and green). Green - ab32106 observed at 55 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

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.



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

Anti-ELK1 antibody [E277] (ab32106) at 1/500 dilution
Predicted band size: 45 kDa
Observed band size: 62 kDa



Anti-ELK1 antibody [E277] (ab32106)

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