abcam

Product datasheet

Anti-eIF5A antibody [EP526Y] ab32443

Recombinant RabMAb

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Overview

Product name Anti-elF5A antibody [EP526Y]

Description Rabbit monoclonal [EP526Y] to eIF5A

Host species Rahhit

Specificity This antibody shows low affinity in recognizing eIF5A2 recombinant protein.

Tested applications Suitable for: Flow Cyt (Intra), WB, IHC-P, ICC/IF Species reactivity Reacts with: Human, Recombinant fragment

Predicted to work with: Mouse, Rat, Drosophila melanogaster

Immunogen Synthetic peptide within Human eIF5A aa 1-100 (N terminal). The exact sequence is proprietary.

Database link: P63241

Positive control Jurkat and HeLa whole cell lysate (ab150035) and Adenocarcinoma of human uterus slides.

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. Upon delivery aliquot and store at -20°C. Avoid freeze / thaw cycles.

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 **EP526Y** Clone number

Isotype IgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab32443 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)		1/50 - 1/80. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB	★★★★☆ (1)	1/5000 - 1/10000. Predicted molecular weight: 18 kDa.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ICC/IF		1/500.

Target

Function

mRNA-binding protein involved in translation elongation. Has an important function at the level of mRNA turnover, probably acting downstream of decapping. Involved in actin dynamics and cell cycle progression, mRNA decay and probably in a pathway involved in stress response and maintenance of cell wall integrity. With syntenin SDCBP, functions as a regulator of p53/TP53 and p53/TP53-dependent apoptosis. Regulates also TNF-alpha-mediated apoptosis. Mediates effects of polyamines on neuronal process extension and survival. May play an important role in brain development and function, and in skeletal muscle stem cell differentiation. Also described as a cellular cofactor of human T-cell leukemia virus type I (HTLV-1) Rex protein and of human immunodeficiency virus type 1 (HIV-1) Rev protein, essential for mRNA export of retroviral transcripts.

Tissue specificity
Sequence similarities
Post-translational

Expressed in umbilical vein endothelial cells and several cancer cell lines (at protein level).

Belongs to the eIF-5A family.

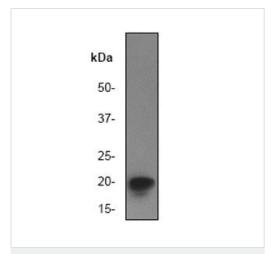
modifications

Cellular localization

eIF-5A seems to be the only eukaryotic protein to have an hypusine residue which is a post-translational modification of a lysine by the addition of a butylamino group (from spermidine).

Cytoplasm. Nucleus. Endoplasmic reticulum membrane. Nucleus > nuclear pore complex. Hypusine modification promotes the nuclear export and cytoplasmic localization and there was a dynamic shift in the localization from predominantly cytoplasmic to primarily nuclear under apoptotic inducing conditions.

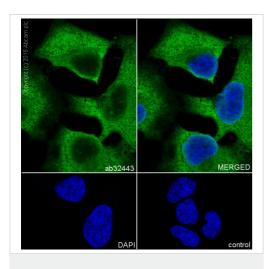
Images



Western blot - Anti-elF5A antibody [EP526Y] (ab32443)

Anti-elF5A antibody [EP526Y] (ab32443) at 1/10000 dilution + Jurkat cell lysate.

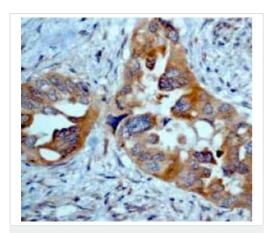
Predicted band size: 18 kDa **Observed band size:** 18 kDa



Immunocytochemistry/ Immunofluorescence - AntieIF5A antibody [EP526Y] (ab32443)

Immunocytochemistry/Immunofluorescence analysis of HeLa (human cervix adenocarcinoma) cells labelling eIF5A with purified ab32443 at 1/500. Cells were fixed with 4% Paraformaldehyde. An Alexa Fluor[®] 488-conjugated goat anti-rabbit IgG (ab150077) at 1/1000 dilution was used as the secondary antibody. Nuclei couterstained with DAPI (blue).

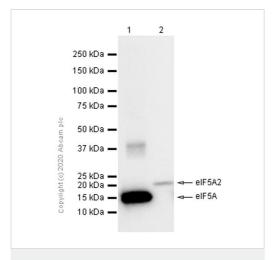
Secondary Only Control: PBS was used instead of the primary antibody as the negative control



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-elF5A antibody [EP526Y] (ab32443)

ab32443 at a 1:250 dilution staining elF5A in human adenocarcinoma of uterus using Immunohistochemistry, Paraffin Embedded Tissue.

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



Western blot - Anti-elF5A antibody [EP526Y] (ab32443)

All lanes : Anti-elF5A antibody [EP526Y] (ab32443) at 1/2000 dilution

Lane 1 : Recombinant Human eIF5A protein (<u>ab87457</u>)

Lane 2 : Recombinant Human eIF5A2 protein (<u>ab99140</u>)

Lysates/proteins at 0.01 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

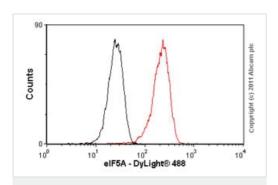
Developed using the ECL technique.

Predicted band size: 18 kDa **Observed band size:** 15,20 kDa

Exposure time: 20 seconds

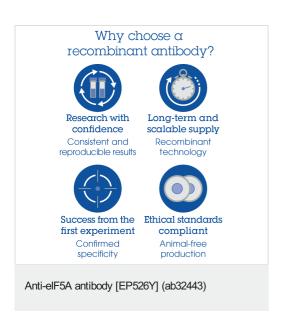
Blocking buffer: 5% NFDM/TBST

This antibody shows low affinity in recognizing eIF5A2 recombinant protein.



Flow Cytometry (Intracellular) - Anti-eIF5A antibody [EP526Y] (ab32443)

Overlay histogram showing Jurkat cells stained with ab32443 (red line). The cells were fixed with methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab32443, 1/50 dilution) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-rabbit lgG (H+L) (ab96899) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit monoclonal lgG (0.5µg/1x10⁶ cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a positive signal in Jurkat cells fixed with 4% paraformaldehyde (10 min)/permeabilized in 0.1% PBS-Tween used under the same conditions.



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