abcam

Product datasheet

Anti-NXF1 antibody [EPR8009] ab129160



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Overview

Product name Anti-NXF1 antibody [EPR8009]

Description Rabbit monoclonal [EPR8009] to NXF1

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), WB, IP, ICC/IF

Species reactivity Reacts with: Human

Immunogen Synthetic peptide within Human NXF1. The exact sequence is proprietary.

Positive control WB: HeLa, 293T, K-562 and HepG2 lysates. ICC/IF: HeLa cells. Flow Cyt (intra): HepG2 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**.

Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with

these species. Please contact us for more information.

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

Clonality Monoclonal Clone number **EPR8009**

Isotype ΙgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab129160 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.
WB	****(1)	1/1000 - 1/10000. Detects a band of approximately 70 kDa (predicted molecular weight: 70 kDa).
IP	*****(1)	1/10 - 1/100.
ICC/IF		1/200 - 1/500.

Target

Function

Involved in the nuclear export of mRNA species bearing retroviral constitutive transport elements (CTE) and in the export of mRNA from the nucleus to the cytoplasm. The NXF1-NXT1 heterodimer is involved in the export of HSP70 mRNA in conjunction with THOC4 and THOC5.

Tissue specificity

Expressed ubiquitously.

Sequence similarities

Belongs to the NXF family.

Contains 4 LRR (leucine-rich) repeats.

Contains 1 NTF2 domain.

Contains 1 RRM (RNA recognition motif) domain.

Contains 1 TAP-C domain.

Domain

The minimal CTE binding domain consists of an RNP-type RNA binding domain (RBD) and $\,$

leucine-rich repeats.

The nucleoporin binding domain consists of a NTF2 domain (also called NTF2-like domain) and a TAP-C domain (also called UBA-like domain). It has 2 nucleoporin-FG-repeats binding sites (one in the NTF2 and the other in the TAP-C domain) which contribute to nucleoporin association and act synergistically to export cellular mRNAs.

The NTF2 domain is functional only in the presence of NXT1 and is essential for the export of mRNA from the nucleus.

The TAP-C domain mediates direct interactions with nucleoporin-FG-repeats and is necessary and sufficient for localization of NXF1 to the nuclear rim. The conserved loop 594-NWD-596 of the TAP-C domain has a critical role in the interaction with nucleoporins.

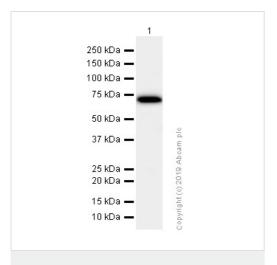
The leucine-rich repeats are essential for the export of mRNA from the nucleus.

The RNA-binding domain is a non-canonical RNP-type domain.

Cellular localization

Nucleus > nucleoplasm. Nucleus speckle. Cytoplasm. Localized predominantly in the nucleoplasm and at both the nucleoplasmic and cytoplasmic faces of the nuclear pore complex. Shuttles between the nucleus and the cytoplasm. Travels to the cytoplasm as part of the exon junction complex (EJC) bound to mRNA.

Images



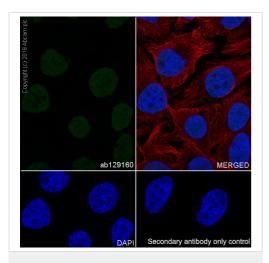
Western blot - Anti-NXF1 antibody [EPR8009] (ab129160)

Anti-NXF1 antibody [EPR8009] (ab129160) at 1/2000 dilution (Purified) + K-562 (Human chronic myelogenous leukemia lymphoblast) whole cell lysates at 15 µg

Secondary

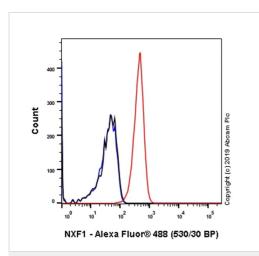
Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/2000 dilution

Predicted band size: 70 kDa Observed band size: 70 kDa



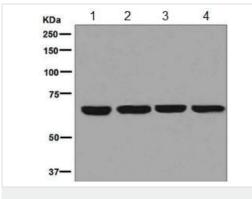
Immunocytochemistry/ Immunofluorescence - Anti-NXF1 antibody [EPR8009] (ab129160)

Immunocytochemistry/ Immunofluorescence analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling NXF1 with purified ab129160 at 1/200 dilution (0.62 μ g/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1/200 (2.5 μ g/ml). Goat anti rabbit lgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1/1000 (2 μ g/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Flow Cytometry (Intracellular) - Anti-NXF1 antibody [EPR8009] (ab129160)

Intracellular Flow Cytometry analysis of HepG2 (Human hepatocellular carcinoma epithelial cell) cells labeling NXF1 with purified ab129160 at 1/20 dilution (5 μ g/ml) (Red). Cells were fixed with 4% Paraformaldehyde and permeabilised with 90% Methanol. A Goat anti rabbit lgG (Alexa Fluor® 488, **ab150077**) secondary antibody was used at 1/2000. Isotype control - Rabbit monoclonal lgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Western blot - Anti-NXF1 antibody [EPR8009] (ab129160)

All lanes : Anti-NXF1 antibody [EPR8009] (ab129160) at 1/1000 dilution

Lane 1 : HeLa cell lysate Lane 2 : 293T cell lysate

Lane 3: K562 cell lysate

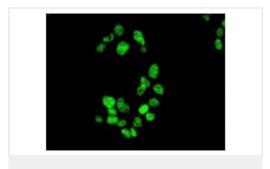
Lane 4: HepG2 cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

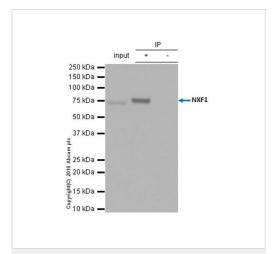
All lanes: Goat anti-rabbit HRP at 1/2000 dilution

Predicted band size: 70 kDa



Immunocytochemistry/ Immunofluorescence - Anti-NXF1 antibody [EPR8009] (ab129160)

ab129160 at 1/250 dilution staining NXF1 in HepG2 cells by immunofluorescence.



Immunoprecipitation - Anti-NXF1 antibody [EPR8009] (ab129160)

ab129160 at 1/20 immunoprecipitating NXF1 in HepG2 (human hepatocellular carcinoma) whole cell lysate observed at 70 KDa (Lanes 1 and 2).

Lane 1 (input): HepG2 whole cell lysate, 10µg

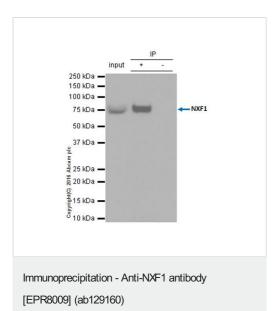
Lane 2 (+): ab129160 + HepG2 whole cell lysate

Lane 3 (-): Rabbit monoclonal lgG ($\underline{ab172730})$ instead of ab129160 in

HepG2 whole cell lysate

For western blotting, ab129160 at 1/500 dilution and <u>ab131366</u> VeriBlot for IP (HRP) at 1/1000 were used.

Blocking and Diluting buffer and concentration: 5% NFDM/TBST.



ab129160 at 1/20 immunoprecipitating NXF1 in HeLa (human cervix adenocarcinoma) whole cell lysate observed at 70 KDa (Lanes 1 and 2).

Lane 1 (input): HeLa whole cell lysate, 10µg

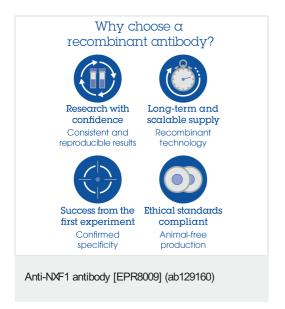
Lane 2 (+): ab129160 + HeLa whole cell lysate

Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab129160 in

HeLa whole cell lysate

For western blotting, ab129160 at 1/500 dilution and <u>ab131366</u> VeriBlot for IP (HRP) at 1/1000 were used.

Blocking and Diluting buffer and concentration: 5% NFDM/TBST.



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