abcam

Product datasheet

Anti-IGF2BP1/IMP1 antibody [EPR26408-18] ab290736

Recombinant RobMAb

1 References 13 Images

Overview

Product name Anti-IGF2BP1/IMP1 antibody [EPR26408-18]

Description Rabbit monoclonal [EPR26408-18] to IGF2BP1/IMP1

Host species Rabbit

Specificity Please note that this antibody does not react with Rat species for WB application.

This antibody has no cross-reactivity with mouse IGF2BP2 or IGF2BP3.

Tested applications Suitable for: Flow Cyt (Intra), WB, IHC-P, mIHC, ICC/IF

Unsuitable for: IP

Species reactivity Reacts with: Mouse, Rat, Human

Immunogen Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Mouse testis, liver, kidney tissue lysate. Caco-2, NIH/3T3, U-2 OS, 293T whole cell lysates.

IHC-P: Human, mouse, rat testis. ICC/IF: NIH/3T3 cells. Flow Cyt (Intra): U-2 OS cells. mIHC-P:

Human testis tissue.

General notesThis 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 $\underline{\text{see here}}$.

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 long

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

Purity Protein A purified

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Clonality Monoclonal

Clone number EPR26408-18

Isotype IgG

Applications

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab290736 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)		1/500.
WB		1/1000. Detects a band of approximately 65 kDa (predicted molecular weight: 63 kDa).
IHC-P		1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
mIHC		1/500.
ICC/IF		1/50.

Application notes

Is unsuitable for IP.

Target

Function

RNA-binding factor that affects mRNA nuclear export, localization, stability and translation. Component of the CRD-mediated complex that promotes MYC mRNA stabilization. Regulates mRNA stability during the integrated cellular stress response (ISR) in stress granules (SGs). Stabilizes the BTRC/FBW1A mRNA from degradation by disrupting miRNA-dependent interaction with AGO2. Identified in a HCV IRES-mediated translation complex, that enhances translation at the Hepatitis C virus (HCV) RNA-replicon via the internal ribosome entry site (IRES), but does not affect 5'cap-dependent translation. Acts as a HIV-1 retrovirus restriction factor that reduces HIV-1 assembly by inhibiting viral RNA packaging, assembly and processing of HIV-1 GAG protein on cellular membranes. Binds to mRNAs in stress granules (SGs). Binds to the stem-loop IV of the 5'-UTR and to the variable region and the poly(U-C) motif of the 3'-UTR of the HCV RNA-replicon. Binds to the 5'-UTR of the insulin-like growth factor 2 (IGF2) mRNA and regulates its subcellular localization and translation. Binds both to the coding region mRNA stability determinant (CRD) and to AU-rich sequences in the 3'-UTR of the MYC and CD44 mRNAs and stabilizes these mRNAs. Binds to the fourth and fifth exons of the oncofetal H19 and neuron-specific TAU mRNAs and regulates their localizations. Binds to the adenine-rich autoregulatory sequence (ARS) 5'-UTR of the PABPC1 mRNA and is involved in its translational repression. The RNA-binding activity to ARS is stimulated by PABPC1. Binds to the coding sequence region of BTRC/FBW1A mRNA and mediates stabilization of BTRC/FBW1A and MYC mRNAs in response to beta-catenin signaling. Binding to RNA employs a cooperative, sequential mechanism of homo- or heterodimerisation. Also involved in growth or survival of lung-cancer cells. Protects the MYC and MDR-1 mRNAs from cleavage by a endoribonuclease, thus prolonging their stabilities (By similarity). Binds to the 3'-UTR axonal localization signal (ALS) of

TAU mRNA (By similarity). Binds to a conserved 54-nucleotide element in the 3'-UTR of the beta actin mRNA known as the 'zipcode' (By similarity). Promotes translocation of the beta-actin mRNA to dendrites (By similarity). May act as a regulator of mRNA transport to activated synapses in response to synaptic activity.

Expressed in fetal liver, fetal lung, fetal kidney, fetal thymus, fetal placenta, fetal follicles of ovary, gonocytes of testis, oocytes, spermatogonia and semen (at protein level). Expressed in testicular and lung cancer (at protein level). Expressed in kidney, prostate, trachea, testis and lung cancer.

Belongs to the RRM IMP/VICKZ family.

Contains 4 KH domains.

Contains 2 RRM (RNA recognition motif) domains.

The third and fourth KH domains encompass the protein dimerization motif and are necessary and sufficient for RNA binding. The four KH domains are important for granule formation and SGs targeting. Contains two nuclear export signals, situated within the second and fourth KH domains. The four KH domains are important to suppress HIV-1 infectivity.

Phosphorylated. Phosphorylation may influence mRNA translation.

Nucleus. Cytoplasm. Cell projection > lamellipodium. Cell projection > dendrite. Cell projection > dendritic spine. Localized in cytoplasmic mRNP granules containing untranslated mRNAs.

Targeted to stress granules (SGs), but not processing bodies (PBs), during cellular stress.

Colocalizes with G3BP1 and TIAL1 in SGs. Colocalizes with HIV-1 GAG at the cell edges. Found in lamellipodia of the leading edge, in the perinuclear region, and beneath the plasma membrane. The subcytoplasmic localization is cell specific and regulated by cell contact and growth.

Colocalized with H19 RNA at lamellipodia. Colocalized with CD44 mRNA in RNP granules.

Nuclear export is mediated by XPO1/CRM1. In motile cells, is transported towards the leading edge into the cortical region of the lamellipodia where it is connected to microfilaments (By similarity). Present in the form of granules and into F-actin-rich protrusion of dendrites, spines and subsynaptic sites (By similarity). Colocalizes with beta-actin mRNA in dendrites and spines (By similarity).

Tissue specificity

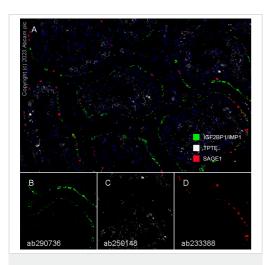
Sequence similarities

Domain

Post-translational modifications

Cellular localization

Images



Multiplex immunohistochemistry - Anti-IGF2BP1/IMP1 antibody [EPR26408-18] (ab290736)

Fluorescence multiplex immunohistochemical analysis of paraffinembedded Human testis tissue.

Neuronal depolarization by KCI induces its rapid efflux from the cell body into dendrites.

Panel A: Merged staining of anti-IGF2BP1/IMP1 (green; Opal™690), anti-TPTE (gray; Opal™520) and anti-SAGE1 (red; Opal™570) on human testis.

Panel B: Anti-IGF2BP1/IMP1 stained on cytoplasm of spermatogonia.

Panel C: Anti-TPTE stained on spermatocytes.

Panel D: Anti-SAGE1 stained on nucleus of spermatogonia.

The section was incubated in three rounds of staining: in the order of ab290736, **ab250148**, and **ab233388** for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an

Opal[™] 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins. Counterstained with DAPI.

B C D

Multiplex immunohistochemistry - Anti-IGF2BP1/IMP1 antibody [EPR26408-18] (ab290736)

ab290736

Fluorescence multiplex immunohistochemical analysis of paraffinembedded Human testis tissue.

Panel A: Merged staining of anti-AKAP4 (magenta; Opal™690), anti-TPTE (green; Opal™520) and anti-IGF2BP1/IMP1 (red; Opal™570) on human testis.

Panel B: Anti-IGF2BP1/IMP1 stained on spermatogonia.

Panel C: Anti-TPTE stained on spermatocytes.

Panel D: Anti-AKAP4 stained on spermatids.

The section was incubated in three rounds of staining: in the order of <u>ab238887</u>, <u>ab250148</u>, and ab290736 for 30 mins at room temperature. Each round was followed by a separate fluorescent tyramide signal amplification system. The immunostaining was performed on a Leica Biosystems BOND® RX instrument with an Opal[™] 4-color kit. Image acquisition was performed with Leica SP8 confocal microscope. Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 20 mins. Counterstained with DAPI.

1 2 3 4

250 kDa150 kDa100 kDa75 kDa50 kDa37 kDa
25 kDa225 kDa220 kDa27 kDa15 kDa10 kDa15 kDa10 kDa27 kDa28 kDa29 kDa27 kDa28 kDa29 kDa27 kDa27 kDa28 kDa29 kDa20 kDa-

Western blot - Anti-IGF2BP1/IMP1 antibody [EPR26408-18] (ab290736)

All lanes : Anti-IGF2BP1/IMP1 antibody [EPR26408-18] (ab290736) at 1/1000 dilution

Lane 1 : Caco-2 (human colorectal adenocarcinoma epithelial cell) whole cell lysate.

Lane 2 : U-2 OS (human bone osteosarcoma epithelial cell) whole cell lysate.

Lane 3: 293T (human embryonic kidney epithelial cell) whole cell lysate.

Lane 4: NIH/3T3 (mouse embryonic fibroblast) whole cell lysate.

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 63 kDa Observed band size: 65 kDa Exposure time: 3 minutes

Blocking / Diluent buffer and concentration: 5% NFDM/TBST

Exposure time:

Lane 1-3: 10 seconds

Lane 4: 26 seconds

Negative control: U-2 OS (PMID: 26917013)

All lanes : Anti-IGF2BP1/IMP1 antibody [EPR26408-18] (ab290736) at 1/1000 dilution

Lane 1 : MYC/DDK-tagged mouse IGF2BP1 full-length recombinant protein 10 ng

Lane 2: MYC/DDK- tagged mouse IGF2BP2 full-length recombinant protein 10 ng

Lane 3 : His-tagged mouse IGF2BP3 full-length recombinant protein 10 ng

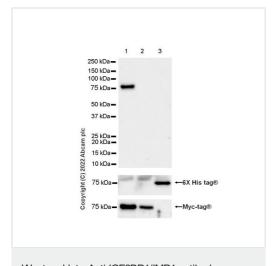
Secondary

All lanes : Goat Anti-Rabbit $\lg G \ H\&L \ (HRP) \ (\underline{ab97051})$ at 1/100000 dilution

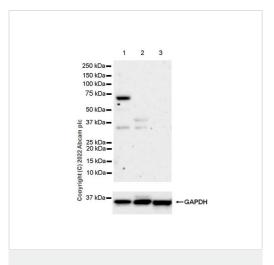
Predicted band size: 63 kDa

Exposure time: 48 seconds

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



Western blot - Anti-IGF2BP1/IMP1 antibody [EPR26408-18] (ab290736)



Western blot - Anti-IGF2BP1/IMP1 antibody [EPR26408-18] (ab290736) **All lanes :** Anti-IGF2BP1/IMP1 antibody [EPR26408-18] (ab290736) at 1/1000 dilution

Lane 1: Mouse testis tissue lysate.

Lane 2 : Mouse liver tissue lysate.

Lane 3: Mouse kidney tissue lysate.

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 63 kDa **Observed band size:** 65 kDa

Exposure time: 3 minutes

Blocking / Dilution buffer and concentration: 5% NFDM/TBST

The expression profile observed is consistent with that described in the literature (PMID: 26917013).

Negative control: mouse liver, mouse kidney (PMID: 26917013). Two unidentified bands around 37 kDa were observed.

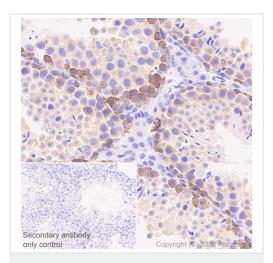
Secondary antibody

Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-IGF2BP1/IMP1 antibody [EPR26408-18] (ab290736)

Immunohistochemical analysis of paraffin-embedded Human testis tissue labelling IGF2BP1/IMP1 with ab290736 at 1/2000 (0.284 ug/ml) followed by a ready to use LeicaDS9800 (BONDTM Polymer Refine Detection). Positive staining on human testis (PMID:28333300) . The section was incubated with ab290736 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND[®] RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (BONDTM Polymer Refine Detection).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

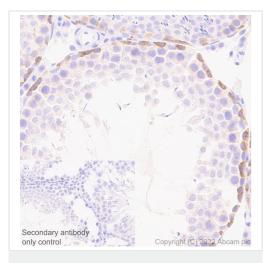


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-IGF2BP1/IMP1 antibody [EPR26408-18] (ab290736)

Immunohistochemical analysis of paraffin-embedded Mouse testis tissue labelling IGF2BP1/IMP1 with ab290736 at 1/2000 (0.284 ug/ml) followed by a ready to use LeicaDS9800 (BONDTM Polymer Refine Detection). Positive staining on mouse testis. The section was incubated with ab290736 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND[®] RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (BONDTM Polymer Refine Detection).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins

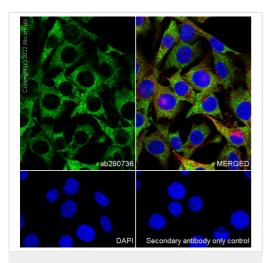


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-IGF2BP1/IMP1 antibody [EPR26408-18] (ab290736)

Immunohistochemical analysis of paraffin-embedded Rat testis tissue labelling IGF2BP1/IMP1 with ab290736 at 1/2000 (0.284 ug/ml) followed by a ready to use LeicaDS9800 (BONDTM Polymer Refine Detection). Positive staining on rat testis. The section was incubated with ab290736 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND[®] RX instrument Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (BONDTM Polymer Refine Detection) was used.

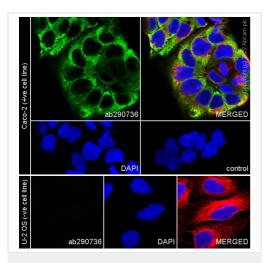
Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins



Immunocytochemistry/ Immunofluorescence - Anti-IGF2BP1/IMP1 antibody [EPR26408-18] (ab290736)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeilized NIH/3T3 (mouse embryonic fibroblast) cells lebelling IGF2BP1/IMP1 with ab290736 at 1/50 (11.34 ug/ml) dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) preadsorbed antibody at 1/1000 (2 ug/ml) dilution (Green). Confocal image showing cytoplasmic staining in NIH/3T3 cell line is observed. ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor[®] 594) was used to counterstain tubulin at 1/200 (2.5 ug/ml) dilution (Red). The Nuclear counterstain was DAPI (Blue).

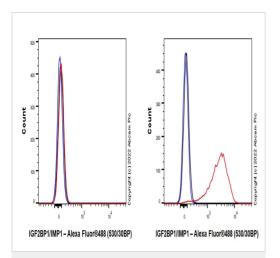
Secondary antibody only control: Secondary antibody is **ab150081**Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) preadsorbed at 1/1000 (2 ug/ml) dilution.



Immunocytochemistry/ Immunofluorescence - Anti-IGF2BP1/IMP1 antibody [EPR26408-18] (ab290736)

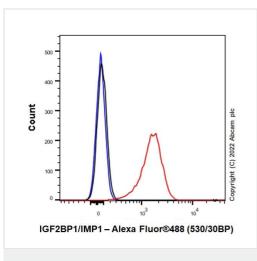
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeilized Caco-2 (human colorectal adenocarcinoma epithelial cell) cells lebelling IGF2BP1/IMP1 with ab290736 at 1/50 (11.34 ug/ml) dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 (2 ug/ml) dilution (Green). Confocal image showing cytoplasmic staining in Caco-2 cell line. Negative control: U-2 OS (PMID: 23069990). ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 (2.5 ug/ml) dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) preadsorbed at 1/1000 (2 ug/ml) dilution.



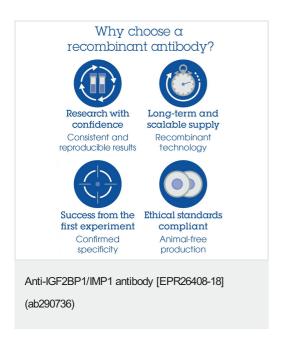
Flow Cytometry (Intracellular) - Anti-IGF2BP1/IMP1 antibody [EPR26408-18] (ab290736)

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized U-2 OS (human bone osteosarcoma epithelial cell, Left) / Caco-2 (human colorectal adenocarcinoma epithelial cell, Right) cells labelling IGF2BP1/IMP1 with ab290736 at 1/500 dilution (0.1ug) (Red) compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/2000 dilution was used as the secondary antibody. Negative control: U-2 OS (PMID: 26917013)



Flow Cytometry (Intracellular) - Anti-IGF2BP1/IMP1 antibody [EPR26408-18] (ab290736)

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized NIH/3T3 (Mouse embryonic fibroblast) cells labelling IGF2BP1/IMP1 with ab290736 at 1/500 dilution (0.1ug) (Red) compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/2000 dilution was used as the secondary antibody.



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