abcam

Product datasheet

Anti-PABPN1 antibody [EP3000Y] ab75855



★★★★★ 2 Abreviews 39 References 14 Images

Overview

Product name Anti-PABPN1 antibody [EP3000Y]

Rabbit monoclonal [EP3000Y] to PABPN1 **Description**

Host species Rabbit

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

Species reactivity Reacts with: Mouse, Rat, Human

Immunogen Synthetic peptide within Human PABPN1 aa 1-100 (N terminal). The exact sequence is

proprietary.

Positive control WB: Raw264.7, MCF-7, 293T, Mouse spleen, Rat brain and HeLa whole cell lysate (ab150035).

ICC/IF: MCF-7 cells. Flow Cyt (intra): MCF-7 cells. IHC-P: Squamous cell cervical carcinoma

tissue, human bladder carcinoma, mouse kidney, rat kidney.

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 long

term. Avoid freeze / thaw cycle. Stable for 12 months at -20°C.

Storage buffer pH: 7.20

Preservative: 0.01% Sodium azide

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

Purity Protein A purified

Clonality Monoclonal Clone number EP3000Y

Isotype IgG

Applications

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab75855 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) | | 1/40. ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody. For unpurified use at 1:80. |
| ICC/IF | | 1/100 - 1/250. |
| WB | **** <u>(1)</u> | 1/1000 - 1/10000. Predicted molecular weight: 33 kDa. |
| IP | | 1/30. Fur unpurified use at 1:50. |
| IHC-P | | 1/1000. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See IHC antigen retrieval protocols. For unpurified use at 1/100 -1/250. |

Target

Function

Involved in the 3'-end formation of mRNA precursors (pre-mRNA) by the addition of a poly(A) tail of 200-250 nt to the upstream cleavage product. Stimulates poly(A) polymerase (PAPOLA) conferring processivity on the poly(A) tail elongation reaction and controls also the poly(A) tail length. Increases the affinity of poly(A) polymerase for RNA. Is also present at various stages of mRNA metabolism including nucleocytoplasmic trafficking and nonsense-mediated decay (NMD) of mRNA. Cooperates with SKIP to synergistically activate E-box-mediated transcription through MYOD1 and may regulate the expression of muscle-specific genes. Binds to poly(A) and to poly(G) with high affinity. May protect the poly(A) tail from degradation.

Tissue specificity

Ubiquitous.

Involvement in disease

Defects in PABPN1 are the cause of oculopharyngeal muscular dystrophy (OPMD) [MIM:164300]. OPMD is a form of late-onset slowly progressive myopathy characterized by eyelid ptosis, dysphagia and, sometimes by other cranial and limb-muscle involvement.

Sequence similarities

Contains 1 RRM (RNA recognition motif) domain.

Domain

The RRM domain is essential for specific adenine bases recognition in the poly(A) tail but not

sufficient for poly(A) binding.

Post-translational modifications

Arginine dimethylation is asymmetric and involves PRMT1 and PRMT3. It does not influence the

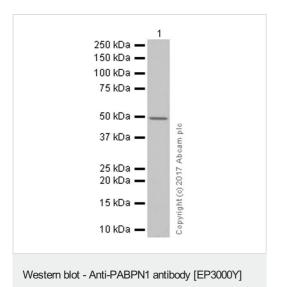
RNA binding properties.

Cellular localization

Nucleus. Cytoplasm. Localized in cytoplasmic mRNP granules containing untranslated mRNAs. Shuttles between the nucleus and the cytoplasm but predominantly found in the nucleus. Its nuclear import may involve the nucleocytoplasmic transport receptor transportin and a RAN-GTP-sensitive import mechanism. Is exported to the cytoplasm by a carrier-mediated pathway that is

Images

(ab75855)



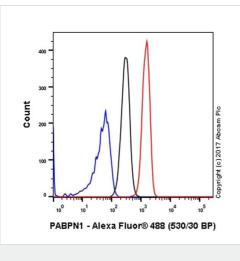
Anti-PABPN1 antibody [EP3000Y] (ab75855) at 1/2000 dilution (purified) + Rat brain lysates at 15 µg

Secondary

Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

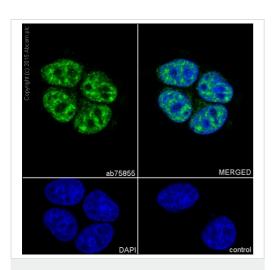
Predicted band size: 33 kDa

Blocking and diluting buffer: 5% NFDM/TBST



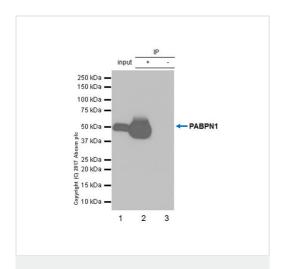
Flow Cytometry (Intracellular) - Anti-PABPN1 antibody [EP3000Y] (ab75855)

Intracellular Flow Cytometry analysis of MCF7 (Human breast adenocarcinoma epithelial cell) cells labeling PABPN1 with purified ab75855 at 1/40 dilution (10 ug/ml) (red). Cells were fixed with 80% Methanol and permeabilized with 0.1% Tween-20. A Goat anti rabbit lgG (Alexa Fluorr®488) secondary antibody was used at 1/2000 dilution. Isotype control - Rabbit monoclonal lgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Immunocytochemistry/ Immunofluorescence - Anti-PABPN1 antibody [EP3000Y] (ab75855)

Immunocytochemistry/ Immunofluorescence analysis of MCF7 (Human breast adenocarcinoma epithelial cell) cells labeling PABPN1 with purified ab75855 at 1:100 dilution (4.1µg/ml). Cells were fixed in 4% paraformaldehyde and permeabilized with 0.1% tritonX-100. ab150077 Goat anti rabbit IgG(Alexa Fluor[®] 488) was used as the secondary antibody at 1:1000 dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Immunoprecipitation - Anti-PABPN1 antibody [EP3000Y] (ab75855)

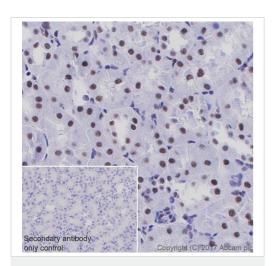
ab75855 (purified) at 1:30 dilution (5ug) immunoprecipitating PABPN1 in MCF7 (Human breast adenocarcinoma epithelial cell) whole cell lysate.

Lane 1 (input): MCF7 (Human breast adenocarcinoma epithelial cell) whole cell lysate 10ug

Lane 2 (+): ab75855 & MCF7 (Human breast adenocarcinoma epithelial cell) whole cell lysate

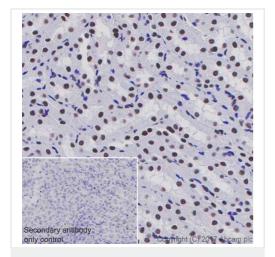
Lane 3 (-): Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab75855 in MCF7 (Human breast adenocarcinoma epithelial cell) whole cell lysate

For western blotting, VeriBlot for IP Detection Reagent (HRP) (ab131366) was used for detection at 1:10,000 dilution.



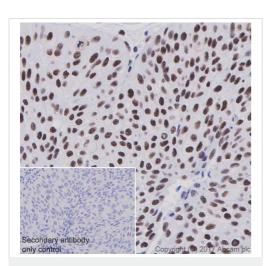
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PABPN1 antibody
[EP3000Y] (ab75855)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat kidney tissue sections labeling PABPN1 with Purified ab75855 at 1:1000 dilution (0.41 µg/ml). Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. ImmunoHistoProbe one step HRP Polymer (ready to use) secondary antibody was used at 1:0 dilution. PBS instead of the primary antibody was used as the negative control.



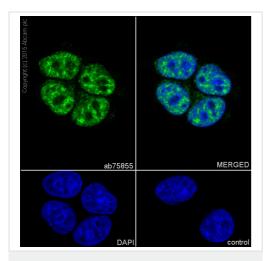
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PABPN1 antibody
[EP3000Y] (ab75855)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse kidney tissue sections labeling PABPN1 with Purified ab75855 at 1:1000 dilution (0.41 µg/ml). Heat mediated antigen retrieval was performed using **ab93684** (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. ImmunoHistoProbe one step HRP Polymer (ready to use) secondary antibody was used at 1:0 dilution. PBS instead of the primary antibody was used as the negative control.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PABPN1 antibody
[EP3000Y] (ab75855)

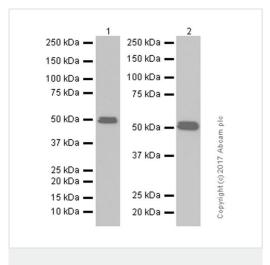
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human bladder carcinoma tissue sections labeling PABPN1 with Purified ab75855 at 1:1000 dilution (0.41 µg/ml). Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. ImmunoHistoProbe one step HRP Polymer (ready to use) secondary antibody was used at 1:0 dilution. PBS instead of the primary antibody was used as the negative control.



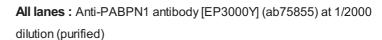
Immunocytochemistry/ Immunofluorescence - Anti-PABPN1 antibody [EP3000Y] (ab75855)

Immunocytochemistry/Immunofluorescence analysis of MCF-7 cells labelling PABPN1 with unpurified ab75855 at 1/500. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. **ab150077**, an Alexa Fluor[®] 488-conjugated goat antirabbit lgG (1/1000) was used as the secondary antibody. Control: PBS only.

Nuclear counter stain: DAPI.



Western blot - Anti-PABPN1 antibody [EP3000Y] (ab75855)



Lane 1 : RAW 264.7 (Mouse Abelson murine leukemia virusinduced tumor macrophage) whole cell lysates

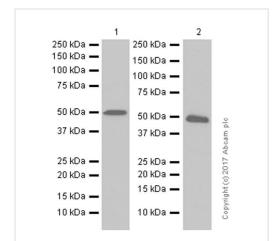
Lane 2: Mouse spleen lysates

Lysates/proteins at 15 µg per lane.

Secondary

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

Predicted band size: 33 kDa



Western blot - Anti-PABPN1 antibody [EP3000Y] (ab75855)

Blocking and diluting buffer: 5% NFDM/TBST

All lanes : Anti-PABPN1 antibody [EP3000Y] (ab75855) at 1/2000 dilution (purified)

Lane 1: 293T (Human embryonic kidney epithelial cell) whole cell lysates

Lane 2 : MCF7 (Human breast adenocarcinoma epithelial cell) whole cell lysates

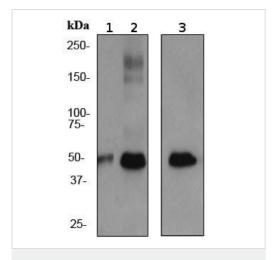
Lysates/proteins at 15 µg per lane.

Secondary

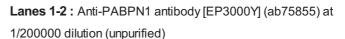
All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 33 kDa **Observed band size:** 49 kDa

Blocking and diluting buffer: 5% NFDM/TBST



Western blot - Anti-PABPN1 antibody [EP3000Y] (ab75855)



Lane 3 : Anti-PABPN1 antibody [EP3000Y] (ab75855) at 1/1000000 dilution (unpurified)

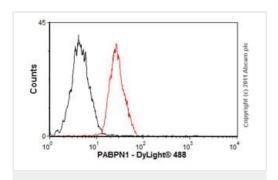
Lane 1 : Raw264.7 cell lysate
Lane 2 : MCF-7 cell lysate
Lane 3 : HeLa cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

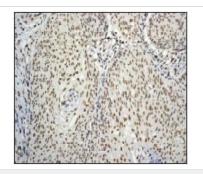
All lanes: HRP-conjugated goat anti-rabbit lgG at 1/1000 dilution

Predicted band size: 33 kDa **Observed band size:** 49 kDa



Flow Cytometry (Intracellular) - Anti-PABPN1 antibody [EP3000Y] (ab75855)

Overlay histogram showing MCF-7 cells stained with unpurified ab75855 (red line). The cells were fixed with 80% 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 (ab75855, 1 μ g/1x10⁶ cells) 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 lgG (monoclonal) (1 μ g/1x10⁶ cells) used under the same conditions. Acquisition of >5,000 events was performed.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-PABPN1 antibody
[EP3000Y] (ab75855)

Unpurified ab75855, at 1/100 dilution, staining PABPN1 in squamous cell cervical carcinoma, by Immunohistochemistry using formalin-fixed, paraffin-embedded tissue.

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.



Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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