abcam

Product datasheet

Anti-PABPN1 antibody [EP3000Y] ab75855

Recombinant RabMAb

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Overview

Product name	Anti-PABPN1 antibody [EP3000Y]
Description	Rabbit monoclonal [EP3000Y] to PABPN1
Host species	Rabbit
Tested applications	**Suitable for:** ICC/IF, WB, IP, IHC-P, Flow Cyt
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 cell lysates. ICC/IF: MCF-7 cells. Flow Cyt: MCF-7 cells. IHC-P: Squamous cell cervical carcinoma tissue, human bladder carcinoma, mouse kidney, rat kidney.
General notes	Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMab® patents **We are constantly working hard to ensure we provide our customers with best in class antibodies. As a result of this work we are pleased to now offer this antibody in purified format. We are in the process of updating our datasheets. The purified format is designated 'PUR' on our product labels. If you have any questions regarding this update, please contact our Scientific Support team.** This product is a recombinant rabbit monoclonal antibody.

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, 0.05% BSA
Purity	Protein A purified

Clonality	Monoclonal
Clone number	EP3000Y
Isotype	IgG

Applications

Our Abpromise guarantee 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.

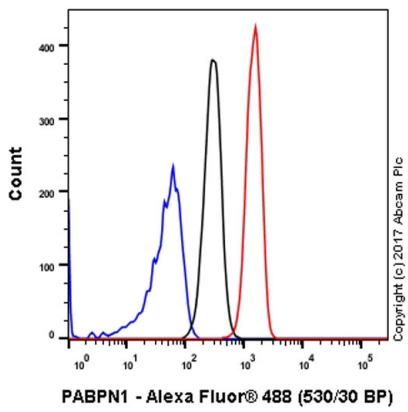
Application	Abreviews	Notes
ICC/IF		1/100 - 1/250.
WB	★★★★★	1/1000 - 1/10000. Predicted molecular weight: 33 kDa.
IP		1/30. **Fur unpurified use at 1:50.**
IHC-P		1/1000. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See IHC antigen retrieval protocols. **For unpurified use at 1/100 -1/250.**
Flow Cyt		1/40. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody. **For unpurified use at 1:80.**

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.

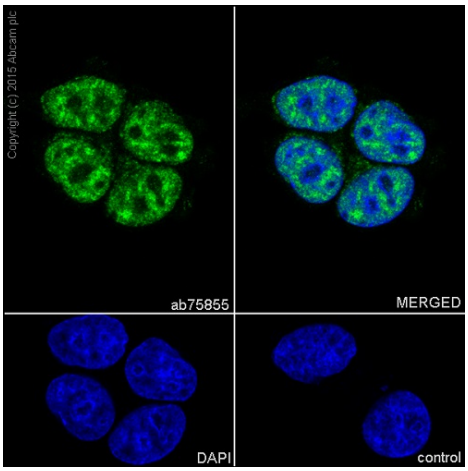
Shuttles between the nucleus and the cytoplasm but predominantly found in the nucleus. Its nuclear import may involve the nucleocytoplasmic transport receptor transportin and a RAN-GTP-sensitive import mechanism. Is exported to the cytoplasm by a carrier-mediated pathway that is independent of mRNA traffic. Nucleus; nuclear speckle. Colocalizes with SKIP and poly(A) RNA in nuclear speckles.

Images



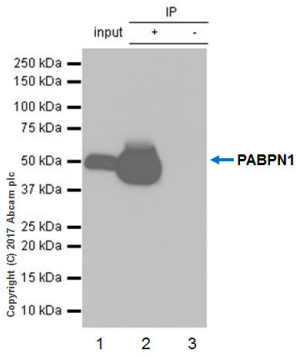
Flow Cytometry - Anti-PABPN1 antibody [EP3000Y] (ab75855)

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 IgG (Alexa Fluor®488) secondary antibody was used at 1:2000 dilution. Isotype control - Rabbit monoclonal IgG (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)

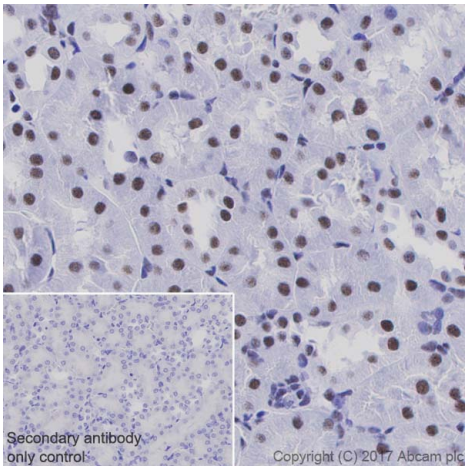
ab75855 (purified) at 1:30 dilution (5ug) immunoprecipitating PABPN1 in MCF7 (Human breast adenocarcinoma epithelial cell) whole cell lysate.

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

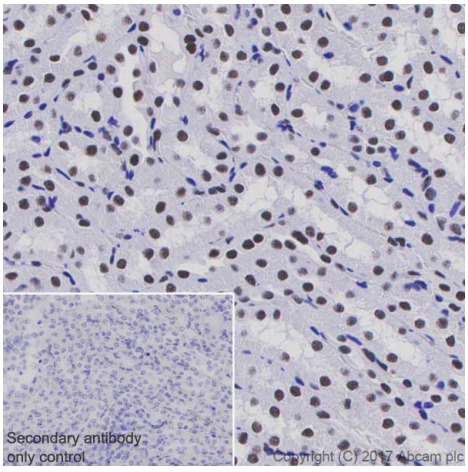
Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab75855 in MCF7 (Human breast adenocarcinoma epithelial cell) whole cell lysate

For western blotting, VeriBlot for IP secondary antibody (HRP) (ab131366) was used as the secondary antibody at 1:10,000 dilution.



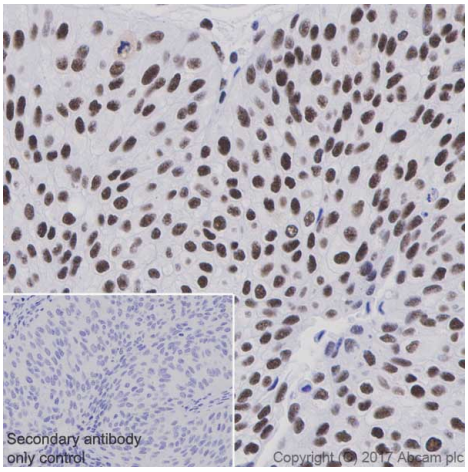
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PABPN1 antibody [EP3000Y] (ab75855)

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.



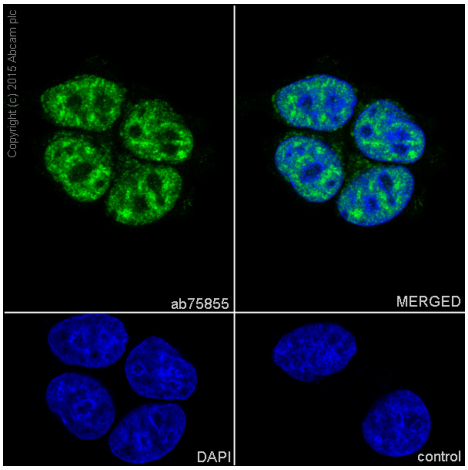
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PABPN1 antibody [EP3000Y] (ab75855)

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.



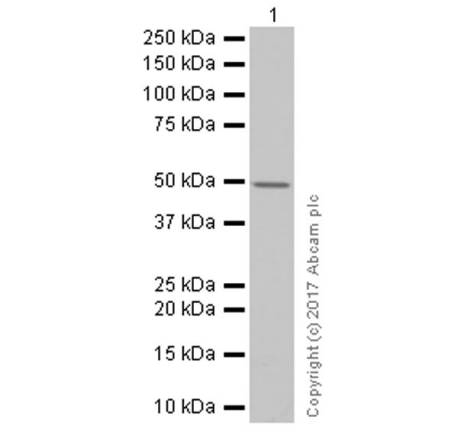
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-PABPN1 antibody [EP3000Y] (ab75855)

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 anti-rabbit IgG (1/1000) was used as the secondary antibody.
Control: PBS only.
Nuclear counter stain: DAPI.



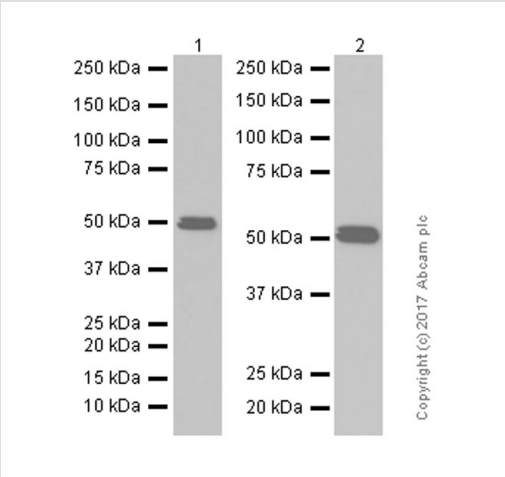
Western blot - Anti-PABPN1 antibody [EP3000Y] (ab75855)

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



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

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

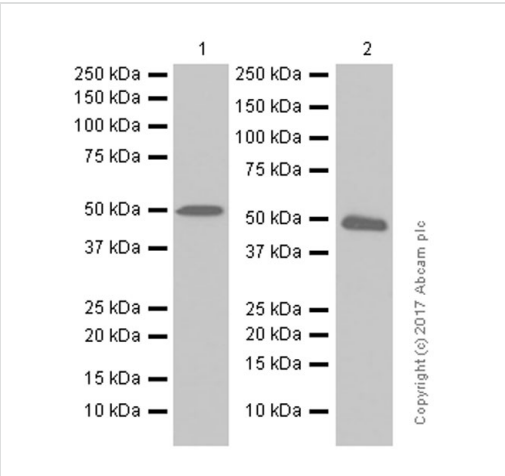
Lane 1 : RAW 264.7 (Mouse Abelson murine leukemia virus-induced tumor macrophage) whole cell lysates
Lane 2 : Mouse spleen lysates

Lysates/proteins at 15 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 33 kDa

Blocking and diluting buffer: 5% NFDM/TBST



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

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

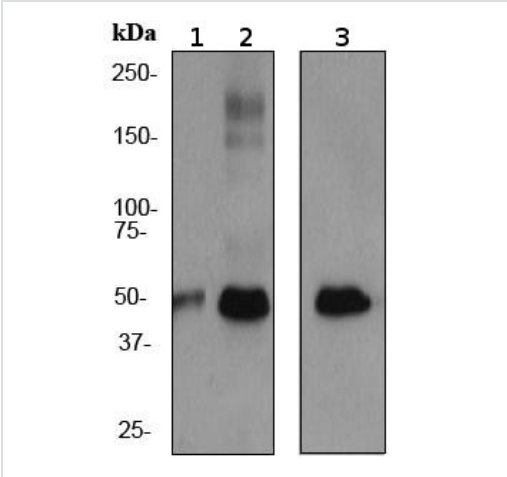
Lane 1 : 293T (Human embryonic kidney epithelial cell) whole cell lysates
Lane 2 : MCF7 (Human breast adenocarcinoma epithelial cell) whole cell lysates

Lysates/proteins at 15 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) 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)

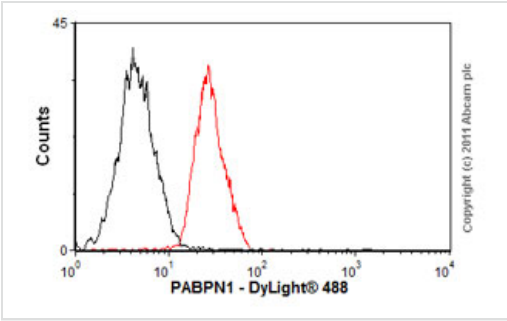
Lanes 1-2 : Anti-PABPN1 antibody [EP3000Y] (ab75855) at 1/200000 dilution (unpurified)
Lane 3 : Anti-PABPN1 antibody [EP3000Y] (ab75855) at 1/1000000 dilution (unpurified)

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

Lysates/proteins at 10 µg per lane.

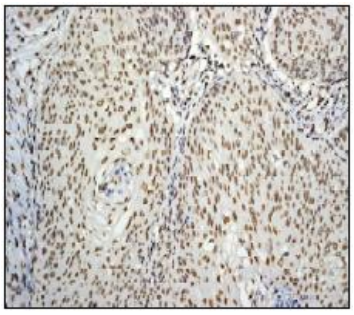
Secondary
All lanes : HRP-conjugated goat anti-rabbit IgG at 1/1000 dilution

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



Flow Cytometry - 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, 1µg/1x10^6 cells) for 30 min at 22ºC. The secondary antibody used was DyLight® 488 goat anti-rabbit IgG (H+L) (ab96899) at 1/500 dilution for 30 min at 22ºC. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10^6 cells) used under the same conditions. Acquisition of >5,000 events was performed.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded 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.

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