abcam

ab251389

Anti-PUS1 antibody [EPR20181] - BSA and Azide free

Recombinant | RabMAb | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR20181
Concentration	0.982 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Human
Dilution info	-
Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

WB

Tested

Species	Mouse, Rat, Human
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	-
Notes	This antibody was not successful when we used it on RAW 264.7 (Mouse macrophage cell line transformed with Abelson murine leukemia virus) cells in ICC application. This antibody was not tested on rat cells in ICC.

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Target data

See full target information PUS1

Function	Pseudouridylate synthase that catalyzes pseudouridylation of tRNAs and mRNAs (PubMed:15772074, PubMed:24722331). Acts on positions 27/28 in the anticodon stem and also positions 34 and 36 in the anticodon of an intron containing tRNA (PubMed:24722331). Also catalyzes pseudouridylation of mRNAs: mediates pseudouridylation of mRNAs with the consensus sequence 5'-UGUAG-3' (PubMed:31477916, PubMed:35051350). Acts as a regulator of pre-mRNA splicing by mediating pseudouridylation of pre-mRNAs at locations associated with alternatively spliced regions (PubMed:35051350). Pseudouridylation of pre-mRNAs near splice sites directly regulates mRNA splicing and mRNA 3'-end processing (PubMed:35051350). Involved in regulation of nuclear receptor activity through pseudouridylation of SRA1 mRNA (PubMed:24722331).

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

ab251389 is the carrier-free version of ab203010.

Patented technology

Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?

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 batch production

For more information, read more on recombinant antibodies.

Conjugation ready
Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with 1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

Compatibility
This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	PUS1 also known as pseudouridine synthase 1 is an enzyme responsible for catalyzing the isomerization of uridine to pseudouridine in RNA molecules. This protein has a mass of approximately 50 kDa and expresses widely in both the cytoplasm and mitochondria of human cells. Pseudouridine is the most abundant RNA modification and plays an important role in RNA stability and structure. The presence of PUS1 can vary among cell types supporting its necessity in diverse cellular contexts.
Biological function summary	The modification of RNA by PUS1 improves the stability and folding of the RNA structure aiding its functionality. The enzyme is not part of a larger complex but acts independently to achieve its role in RNA modification. Pseudouridine created by PUS1 permits enhanced base stacking and hydrogen bonding which influences the translation process and the overall efficiency of protein synthesis. This modification is important for the proper functioning of tRNA and rRNA ensuring fidelity during protein translation.
Pathways	PUS1 plays an essential role in the RNA processing and modification pathways. Its activity integrates into pathways such as ribosome biogenesis and RNA quality control. Particularly it interacts with other RNA-binding proteins and enzymes like TRUB1 another pseudouridine synthase which also contributes to the maturation of structured RNAs. These pathways are vital for maintaining cellular homeostasis and responding to various physiological demands.
Associated diseases and disorders	Mutations in PUS1 are linked to mitochondrial myopathy and sideroblastic anemia a rare condition characterized by muscle weakness and anemia. These disorders illustrate the critical impact of RNA modification defects on cellular energy production and red blood cell formation. In these conditions PUS1 may interact with defective proteins leading to impaired pseudouridylation disrupting normal cellular functions and contributing to disease pathology. Understanding the role of

PUS1 might provide insights into potential therapeutic strategies for such disorders.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

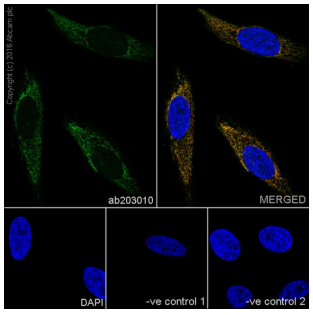
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

9 product images



Immunocytochemistry/ Immunofluorescence - Anti-PUS1 antibody [EPR20181] - BSA and Azide free (ab251389)

PUS1 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-PUS1 antibody

This data was developed using ab203010, the same antibody clone in a different buffer formulation.

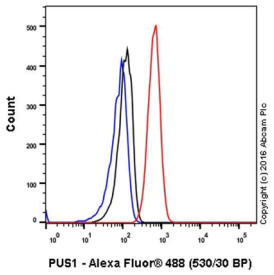
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling PUS1 with ab203010 at 1/500 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing mitochondrial staining on HeLa cell line.

The nuclear counterstain is DAPI (blue). COXIV is detected with ab33985 (Anti-COX IV(mouse mAb)) at 1/1000 dilution followed by Goat anti-mouse IgG (Alexa Fluor® 594) (ab150120) secondary antibody at 1/1000 dilution (red).

The negative controls are as follows:-

-ve control 1: ab203010 at 1/500 dilution followed by ab150120 (Alexa Fluor® 594 Goat anti-Mouse secondary) at 1/1000 dilution.

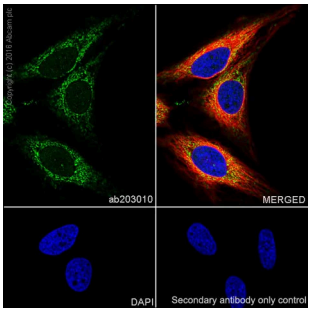
-ve control 2: ab33985 (anti-COX IV(mouse mAb)) at 1/1000 dilution followed by ab150077 (Alexa Fluor® 488 Goat Anti-Rabbit IgG H&L) at 1/1000 dilution.



Flow Cytometry (Intracellular) - Anti-PUS1 antibody [EPR20181] - BSA and Azide free (ab251389)

PUS1 Flow Cytometry (Intracellular) staining using rabbit Anti-PUS1 antibody

This data was developed using ab203010, the same antibody clone in a different buffer formulation.
Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling PUS1 with ab203010 at 1/400 dilution (red) compared with a rabbit monoclonal IgG isotype control (ab172730; black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit IgG (Alexa Fluor® 488) at 1/2000 dilution was used as the secondary antibody.



Immunocytochemistry/ Immunofluorescence - Anti-PUS1 antibody [EPR20181] - BSA and Azide free (ab251389)

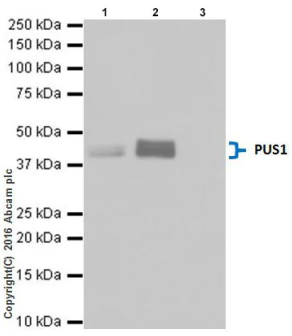
PUS1 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-PUS1 antibody

This data was developed using ab203010, the same antibody clone in a different buffer formulation.
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling PUS1 with ab203010 at 1/500 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing cytoplasmic staining on HeLa cell line.

The nuclear counterstain is DAPI (blue). Tubulin is detected with ab195889 (Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594)) at 1/200 dilution (red).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) at 1/1000 dilution.

This antibody was not successful when we used it on RAW 264.7 (Mouse macrophage cell line transformed with Abelson murine leukemia virus) cells in ICC application. This antibody was not tested on rat cells in ICC.



Immunoprecipitation - Anti-PUS1 antibody [EPR20181] - BSA and Azide free (ab251389)

This data was developed using ab203010, the same antibody clone in a different buffer formulation.
PUS1 was immunoprecipitated from 0.35 mg of HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate with ab203010 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab203010 at 1/1000 dilution. VeriBlot for IP Detection Reaction (HRP) (ab131366), was used for detection at 1/10000 dilution.

Lane 1: HEK-293 whole cell lysate, 10µg (Input).

Lane 2: ab203010 IP in HEK-293 whole cell lysate.

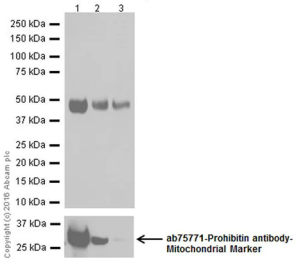
Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab203010 in HEK-293 whole cell lysate.

Blocking and dilution buffer: 5% NFDM/TBST.

Exposure time: 5 seconds.
All lanes:
Immunoprecipitation - Anti-PUS1 antibody [EPR20181] (ab203010)

Predicted band size: 47 kDa

Observed band size: 44 kDa, 47 kDa



Western blot - Anti-PUS1 antibody [EPR20181] - BSA and Azide free (ab251389)

This data was developed using ab203010, the same antibody clone in a different buffer formulation.
Blocking and dilution buffer: 5% NFDM/TBST.

The expression profile observed is consistent with what has been described in UniProt.

All lanes:
Western blot - Anti-PUS1 antibody [EPR20181] (ab203010) at 1/5000 dilution

Lane 1:
HeLa (Human epithelial cell line from cervix adenocarcinoma) mitochondria lysate at 20 µg

Lane 2:
HeLa cytoplasm fraction lysate at 20 µg

Lane 3:
A431 (Human epidermoid carcinoma cell line) whole cell lysate at 20 µg

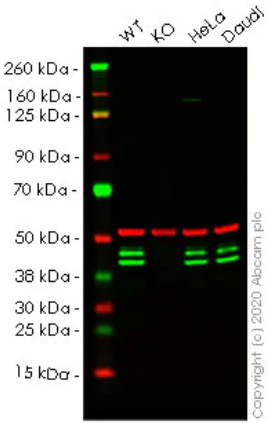
Secondary

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

Predicted band size: 47 kDa

Observed band size: 47 kDa

Exposure time: 8s



Western blot - Anti-PUS1 antibody [EPR20181] - BSA and Azide free (ab251389)

PUS1 Western blot staining using rabbit Anti-PUS1 antibody

This data was developed using the same antibody clone in a different buffer formulation (ab203010).
Lanes 1- 4: Merged signal (red and green). Green - ab203010 observed at 45 kDa. Red - Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) observed at 50 kDa.

ab203010 was shown to react with PUS1 in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line ab266091 (knockout cell lysate ab258158) was used. Wild-type HEK-293T and PUS1 knockout HEK-293T cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab203010 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye®800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye®680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-PUS1 antibody [EPR20181] (ab203010) at 1/1000 dilution

Lane 1:
Wild-type HEK-293T cell lysate at 20 µg

Lane 2:
PUS1 knockout HEK-293T cell lysate at 20 µg

Lane 2:
Western blot - Human PUS1 knockout HEK-293T cell line (ab266091)

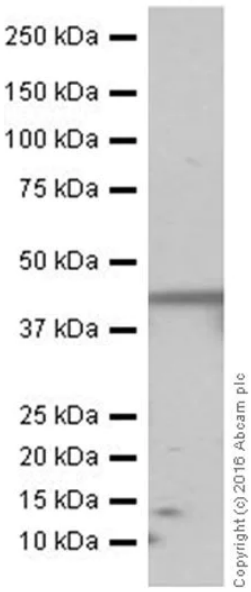
Lane 3:
HeLa cell lysate at 20 µg

Lane 4:
Daudi cell lysate at 20 µg

Performed under reducing conditions.

Predicted band size: 47 kDa

Observed band size: 45 kDa



Western blot - Anti-PUS1 antibody [EPR20181] - BSA and Azide free (ab251389)

This data was developed using ab203010, the same antibody clone in a different buffer formulation.
Blocking and dilution buffer: 5% NFDM/TBST.

All lanes:
Western blot - Anti-PUS1 antibody [EPR20181] (ab203010) at 1/1000 dilution

All lanes:
Human skeletal muscle lysate at 10 µg

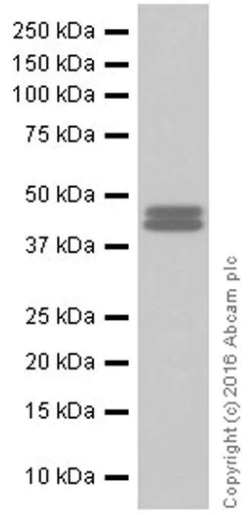
Secondary

All lanes:
Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/1000 dilution

Predicted band size: 47 kDa

Observed band size: 47 kDa

Exposure time: 3min



Western blot - Anti-PUS1 antibody [EPR20181] - BSA and Azide free (ab251389)

PUS1 Western blot staining of HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate using rabbit Anti-PUS1 antibody

This data was developed using ab203010, the same antibody clone in a different buffer formulation.
Blocking and dilution buffer: 5% NFDM/TBST.

Based on sequence alignment, the antibody can recognize 2 isoforms, the predicted MW are 47kDa and 44kDa, respectively (PMID: 17056637).

All lanes:
Western blot - Anti-PUS1 antibody [EPR20181] (ab203010) at 1/1000 dilution

All lanes:
HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate at 10 µg

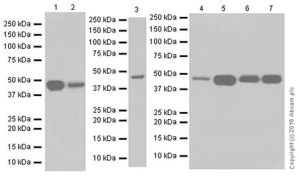
Secondary

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

Predicted band size: 47 kDa

Observed band size: 44 kDa, 47 kDa

Exposure time: 3s



Western blot - Anti-PUS1 antibody [EPR20181] - BSA and Azide free (ab251389)

This data was developed using ab203010, the same antibody clone in a different buffer formulation.
Blocking and dilution buffer: 5% NFDM/TBST.

Exposure time: Lanes 1-2: 10 seconds; Lane 3: 3minutes; Lanes 4-7: 10 seconds.

All lanes:
Western blot - Anti-PUS1 antibody [EPR20181] (ab203010) at 1/2000 dilution

Lane 1:
Mouse heart tissue lysate at 10 µg

Lane 2:
Mouse spleen tissue lysate at 10 µg

Lane 3:
Rat spleen tissue lysate at 10 µg

Lane 4:
C6 (Rat glial tumor cell line) whole cell lysate at 10 µg

Lane 5:
RAW 264.7 (Mouse macrophage cell line transformed with Abelson murine leukemia virus) whole cell lysate at 10 µg

Lane 6:
PC-12 (Rat adrenal gland pheochromocytoma cell line) whole cell lysate at 10 µg

Lane 7:
NIH/3T3 (Mouse embryonic fibroblast cell line) whole cell lysate at 10 µg

Secondary

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

Predicted band size: 47 kDa

Observed band size: 47 kDa

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.