# abcam

# Product datasheet

# Anti-TOM70 antibody [EPR26576-162] - BSA and Azide free ab290002





RabMAb

# 17 Images

#### Overview

Product name Anti-TOM70 antibody [EPR26576-162] - BSA and Azide free

**Description** Rabbit monoclonal [EPR26576-162] to TOM70 - BSA and Azide free

Host species Rabbit

Specificity This antibody does not react with Mouse and Rat species for IHC

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

**Species reactivity** Reacts with: Mouse, Rat, Human

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

Positive control WB: NIH/3T3, C6, HepG2, HeLa and U-2 OS whole cell lysates; Mouse brain and liver tissue

lysates; Rat brain, liver and breast tissue lysates. IHC-P: Human colon, esophagus, breast cancer and cerebrum tissues. ICC/IF: U-2 OS, NIH/3T3 and C6 cells. Flow Cyt: U-2 OS, NIH/3T3 and C6

cells. IP: C6, NIH/3T3 and HeLa whole cell lysates.

**General notes** ab290002 is the carrier-free version of <u>ab289977</u>.

Our <u>carrier-free</u> antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency.

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.

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

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

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For more information see here.

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

### **Properties**

Form Liquid

**Storage instructions** Shipped at 4°C. Store at +4°C.

Storage buffer pH: 7.20

Constituent: 100% PBS

Carrier free Yes

Purity Protein A purified

**Clonality** Monoclonal

Clone number EPR26576-162

**Isotype** IgG

# **Applications**

The Abpromise guarantee Our <u>Abpromise guarantee</u> covers the use of ab290002 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           |           | Use at an assay dependent concentration.   |
| Flow Cyt (Intra) |           | Use at an assay dependent concentration.   |
| WB               |           | Use at an assay dependent concentration. Detects a band of approximately 70 kDa (predicted molecular weight: 67 kDa).  |
| IHC-P            |           | Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Only suitable for Human. |
| IP               |           | Use at an assay dependent concentration.   |

# **Target**

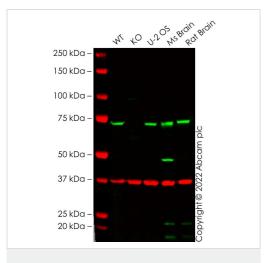
**Function** Receptor that accelerates the import of all mitochondrial precursor proteins.

**Sequence similarities** Belongs to the Tom70 family.

Contains 10 TPR repeats.

**Cellular localization** Mitochondrion outer membrane.

#### **Images**



Western blot - Anti-TOM70 antibody [EPR26576-162] - BSA and Azide free (ab290002)

**All lanes :** Anti-TOM70 antibody [EPR26576-162] (<u>ab289977</u>) at 1/1000 dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: TOM70 knockout HeLa cell lysate

Lane 3: U-2 OS cell lysate

Lane 4: Mouse Brain cell lysate

Lane 5: Rat Brain cell lysate

Lysates/proteins at 20 µg per lane.

### Secondary

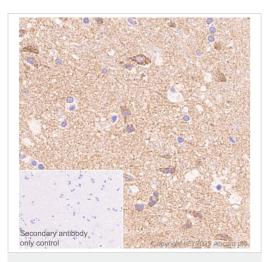
**All lanes :** Goat anti-Rabbit lgG H&L 800CW and Goat anti-Mouse lgG H&L 680RD at 1/20000 dilution

Performed under reducing conditions.

Predicted band size: 67 kDa
Observed band size: 72 kDa

False colour image of Western blot: Anti-TOM70 antibody [EPR26576-162] staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (ab8245) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab289977 was shown to bind specifically to TOM70. A band was observed at 72 kDa in wild-type HeLa cell lysates with no signal observed at this size in TOMM70 knockout cell line ab265396 (knockout cell lysate ab258732). To generate this image, wild-type and TOMM70 knockout HeLa cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween<sup>®</sup> 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature. washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.

This data was developed using <u>ab289977</u>, the same antibody clone in a different buffer formulation.



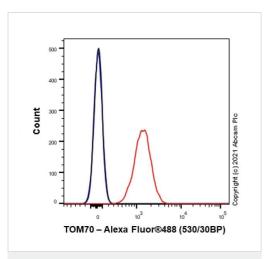
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TOM70 antibody

[EPR26576-162] - BSA and Azide free (ab290002)

Immunohistochemical analysis of paraffin-embedded Human cerebrum tissue labelling TOM70 with <u>ab289977</u> at 1/2000 dilution followed by Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>). Positive staining on human cerebrum. The section was incubated with <u>ab289977</u> for 10 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND<sup>®</sup> RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

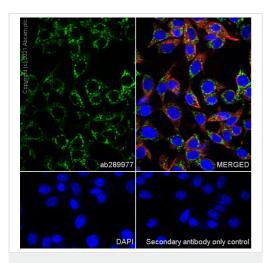
Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 10 mins.



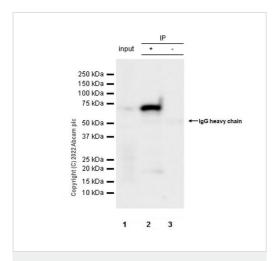
Flow Cytometry (Intracellular) - Anti-TOM70 antibody [EPR26576-162] - BSA and Azide free (ab290002)

This data was developed using <u>ab289977</u>, the same antibody clone in a different buffer formulation.

Flow cytometric analysis of 4% paraformaldehyde fixed, 90% methanol permeabilized C6 (rat glial tumor glial cell) cells labelling TOM70 with <u>ab289977</u> at 1/500 dilution (0.1µg) (Red) (Red) compared with a Rabbit monoclonal IgG (<u>ab172730</u>) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat Anti-Rabbit IgG (Alexa Fluor<sup>®</sup> 488, <u>ab150081</u>) at 1/2000 dilution was used as the secondary antibody.



Immunocytochemistry/ Immunofluorescence - Anti-TOM70 antibody [EPR26576-162] - BSA and Azide free (ab290002)



Immunoprecipitation - Anti-TOM70 antibody
[EPR26576-162] - BSA and Azide free (ab290002)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized C6 cells labeling TOM70 with <a href="mailto:ab289977">ab289977</a> at 1/100 dilution, followed by <a href="mailto:ab150081">ab150081</a> Goat Anti-Rabbit IgG H&L (Alexa Fluor<sup>®</sup> 488) preadsorbed antibody at 1/1000 dilution (Green). Confocal image showing cytoplasmic staining in C6 cell line. <a href="mailto:ab195889">ab195889</a> Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor<sup>®</sup> 594) was used to counterstain tubulin at 1/200 dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is <u>ab150081</u> Goat Anti-Rabbit lgG H&L (Alexa Fluor<sup>®</sup> 488) preadsorbed at 1/1000 dilution.

This data was developed using <u>ab289977</u>, the same antibody clone in a different buffer formulation.

TOM70 was immunoprecipitated from C6 (rat glial tumor glial cell) whole cell lysate with <u>ab289977</u> at 1/30 dilution (2µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using <u>ab289977</u> at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP)(<u>ab131366</u>) was used at 1/5000 dilution.

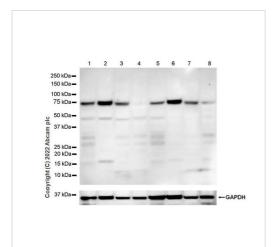
Lane 1: C6 (rat glial tumor glial cell) whole cell lysate 10 µg

Lane 2: ab289977 IP in C6 whole cell lysate

Lane 3: Rabbit monoclonal IgG ( $\underline{ab172730}$ ) instead of  $\underline{ab289977}$  in C6 whole cell lysate

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

Exposure time: 3.25 seconds



Western blot - Anti-TOM70 antibody [EPR26576-162] - BSA and Azide free (ab290002)

**All lanes :** Anti-TOM70 antibody [EPR26576-162] (<u>ab289977</u>) at 1/1000 dilution

Lane 1: NIH/3T3 (mouse embryonic fibroblast) whole cell lysate

Lane 2 : Mouse brain tissue lysate
Lane 3 : Mouse liver tissue lysate

Lane 4: Mouse breast tissue lysate

Lane 5: C6 (rat glial tumor glial cell) whole cell lysate

Lane 6 : Rat brain tissue lysate
Lane 7 : Rat liver tissue lysate
Lane 8 : Rat breast tissue lysate

Lysates/proteins at 20 µg per lane.

### Secondary

**All lanes :** Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated (ab97051) at 1/100000 dilution

**Predicted band size:** 67 kDa **Observed band size:** 70 kDa

This data was developed using 289977, the same antibody clone in a different buffer formulation.

Blocking and diluting buffer and concentration: 5%

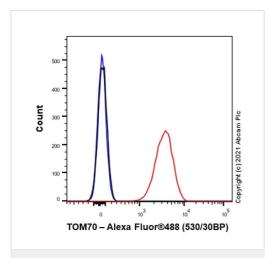
NFDM/TBST

The expression profile/molecular weight observed is consistent with that described in the literature (PMID: 32356556)

Low expression: Breast (human protein

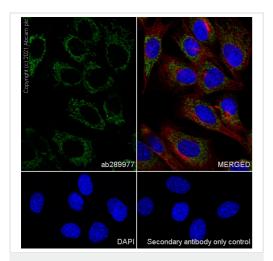
atlas: https://www.proteinatlas.org/ENSG00000154174-TOMM70)

Exposure time: 15 seconds



Flow Cytometry (Intracellular) - Anti-TOM70 antibody [EPR26576-162] - BSA and Azide free (ab290002)

Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol, permeabilized NIH/3T3 (Mouse embryonic fibroblast) cells labelling TOM70 with <u>ab289977</u> at 1/500 dilution (0.1µg) (Red) (Red) compared with a Rabbit monoclonal IgG (<u>ab172730</u>) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat Anti-Rabbit IgG (Alexa Fluor<sup>®</sup> 488, <u>ab150081</u>) at 1/2000 dilution was used as the secondary antibody.

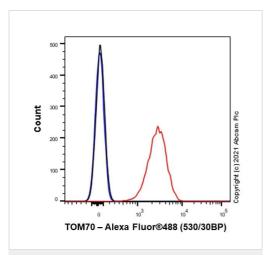


Immunocytochemistry/ Immunofluorescence - Anti-TOM70 antibody [EPR26576-162] - BSA and Azide free (ab290002)

This data was developed using <u>ab289977</u>, the same antibody clone in a different buffer formulation.

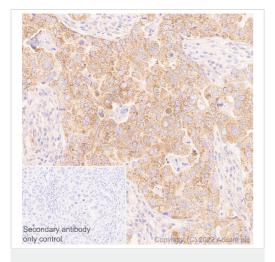
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized NIH/3T3 cells labeling TOM70 with **ab289977** at 1/100 dilution, followed by **ab150081** Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 (2 µg/ml) dilution (Green). Confocal image showing 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 dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is <a href="mailto:ab150081">ab150081</a> Goat Anti-Rabbit IgG H&L (Alexa Fluor<sup>®</sup> 488) preadsorbed at 1/1000 dilution.



Flow Cytometry (Intracellular) - Anti-TOM70 antibody [EPR26576-162] - BSA and Azide free (ab290002)

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol, permeabilized U-2 OS (human bone osteosarcoma epithelial cell) cells labelling TOM70 with <u>ab289977</u> at 1/500 dilution (0.1µg) (Red) (Red) compared with a Rabbit monoclonal lgG (<u>ab172730</u>) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat Anti-Rabbit lgG (Alexa Fluor<sup>®</sup> 488, <u>ab150081</u>) at 1/2000 dilution was used as the secondary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TOM70 antibody

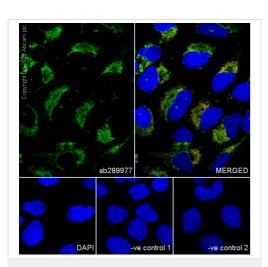
[EPR26576-162] - BSA and Azide free (ab290002)

This data was developed using <u>ab289977</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Human breast cancer tissue labelling TOM70 with <u>ab289977</u> at 1/2000 dilution followed by Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>). Positive staining on human breast cancer. The section was incubated with <u>ab289977</u> for 10 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND<sup>®</sup> RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

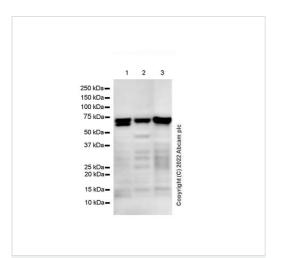
Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 10 mins



Immunocytochemistry/ Immunofluorescence - Anti-TOM70 antibody [EPR26576-162] - BSA and Azide free (ab290002)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized U-2 OS cells labeling TOM70 with **ab289977** at 1/100 dilution, followed by **ab150081** Goat Anti-Rbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 dilution (Green). Confocal image showing mitochondrial staining in U-2 OS cell line. **ab33985** Anti-COX IV mouse monoclonal antibody - Mitochondrial Marker was used at 1/1000 dilution (Red). The Nuclear counterstain was DAPI (Blue).

Secondary antibody only control: Secondary antibody is <u>ab150081</u> Goat Anti-Rabbit lgG H&L (Alexa Fluor<sup>®</sup> 488) preadsorbed at 1/1000 dilution.



Western blot - Anti-TOM70 antibody [EPR26576-162] - BSA and Azide free (ab290002)

**All lanes :** Anti-TOM70 antibody [EPR26576-162] (<u>ab289977</u>) at 1/1000 dilution

**Lane 1 :** HepG2 (human hepatocellular carcinoma epithelial cell) whole cell lysate

Lane 2 : HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate

Lane 3: U-2 OS (human bone osteosarcoma epithelial cell) whole cell lysate

Lysates/proteins at 20 µg per lane.

# **Secondary**

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

Predicted band size: 67 kDa
Observed band size: 70 kDa

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

The expression profile/molecular weight observed is consistent with that described in the literature (PMID: 31907385, PMID:11956321).

Exposure time: 3.25 seconds

This data was developed using <u>ab289977</u>, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Human esophagus tissue labelling TOM70 with <u>ab289977</u> at 1/2000 dilution followed by Rabbit specific IHC polymer detection kit HRP/DAB (<u>ab209101</u>). Positive staining on human esophagus. The section was incubated with <u>ab289977</u> for 10 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND<sup>®</sup> RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 10 mins

This data was developed using <u>ab289977</u>, the same antibody clone in a different buffer formulation.

TOM70 was immunoprecipitated from 0.35 mg HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate 10 µg with ab290002 at 1/30 dilution (2µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab290002 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP) (ab131366) was used at 1/5000 dilution.

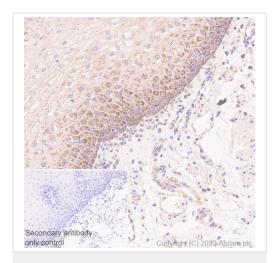
Lane 1: HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate 10  $\mu g$ 

Lane 2: ab289977 IP in HeLa whole cell lysate

Lane 3:Rabbit monoclonal lgG ( $\underline{ab172730}$ ) instead of  $\underline{ab289977}$  in HeLa whole cell lysate

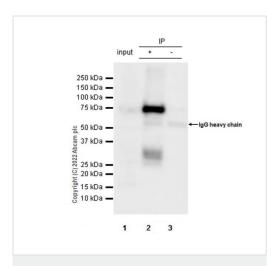
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 3.25 seconds



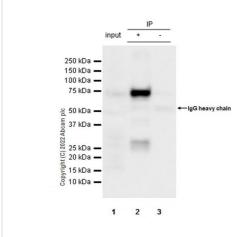
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TOM70 antibody

[EPR26576-162] - BSA and Azide free (ab290002)

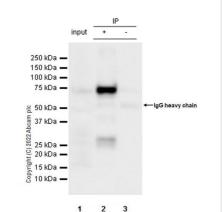


Immunoprecipitation - Anti-TOM70 antibody

[EPR26576-162] - BSA and Azide free (ab290002)



Immunoprecipitation - Anti-TOM70 antibody [EPR26576-162] - BSA and Azide free (ab290002)



NIH/3T3 whole cell lysate Blocking and dilution buffer and concentration: 5% NFDM/TBST.

This data was developed using ab289977, the same antibody

TOM70 was immunoprecipitated from NIH/3T3 (mouse embryonic

fibroblast) whole cell lysate with ab289977 at 1/30 dilution (2µg in

immunoprecipitate using ab289977 at 1/1000 dilution. VeriBlot for

Lane 1: NIH/3T3 (mouse embryonic fibroblast) whole cell lysate 10

Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab289977 in

IP secondary antibody(HRP)(ab131366) was used at 1/5000

0.35mg lysates). Western blot was performed on the

Lane 2: ab289977 IP in NIH/3T3 whole cell lysate

clone in a different buffer formulation.

Exposure time: 3.25 seconds

dilution.



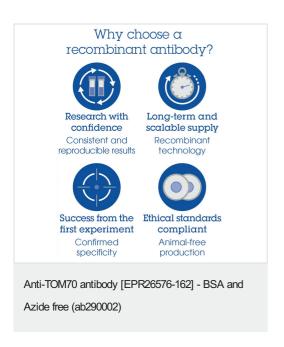
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-TOM70 antibody [EPR26576-162] - BSA and Azide free (ab290002)

This data was developed using ab289977, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Human colon tissue labelling TOM70 with ab289977 at 1/2000 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Positive staining on human colon. The section was incubated with ab289977 for 10 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 Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 10 mins



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