# abcam

### Product datasheet

# Anti-ATG4B antibody [EPR6436(2)] ab154843





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#### Overview

**Product name** Anti-ATG4B antibody [EPR6436(2)]

**Description** Rabbit monoclonal [EPR6436(2)] to ATG4B

**Host species** Rabbit

**Tested applications** Suitable for: Flow Cyt (Intra), WB

Unsuitable for: ICC/IF,IHC-P or IP

Species reactivity Reacts with: Mouse. Human

Predicted to work with: Rat

**Immunogen** Synthetic peptide within Human ATG4B. The exact sequence is proprietary.

Positive control Recombinant Human ATG4B protein (ab123195) can be used as a positive control in WB.

Ramos, 293T, Jurkat and HeLa cell lysates; Jurkat cells.

**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 -20°C.

Storage buffer pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: 9% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA, 50% Tissue culture

supernatant

**Purity** Tissue culture supernatant

Clonality Monoclonal

Clone number EPR6436(2)

**Isotype** IgG

#### **Applications**

The Abpromise guarantee Our Abpromise guarantee covers the use of ab154843 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)		1/100 - 1/500. <b>ab172730</b> - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB	*** <u>*</u> (1)	1/1000 - 1/10000. Predicted molecular weight: 44 kDa.

**Application notes** Is unsuitable for ICC/IF,IHC-P or IP.

#### **Target**

Function

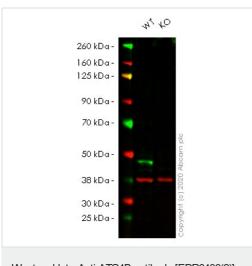
Cysteine protease required for autophagy, which cleaves the C-terminal part of either MAP1LC3, GABARAPL2 or GABARAP, allowing the liberation of form I. A subpopulation of form I is subsequently converted to a smaller form (form II). Form II, with a revealed C-terminal glycine, is considered to be the phosphatidylethanolamine (PE)-conjugated form, and has the capacity for the binding to autophagosomes.

**Tissue specificity**Mainly expressed in the skeletal muscle, followed by brain, heart, liver and pancreas.

**Sequence similarities**Belongs to the peptidase C54 family.

Cellular localization Cytoplasm.

#### **Images**



Western blot - Anti-ATG4B antibody [EPR6436(2)] (ab154843)

**All lanes :** Anti-ATG4B antibody [EPR6436(2)] (ab154843) at 1/1000 dilution

Lane 1: Wild-type HeLa cell lysate

Lane 2: ATG4B knockout HeLa cell lysate

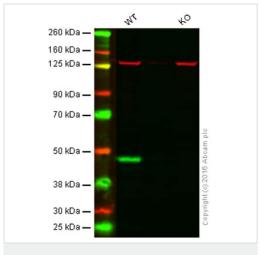
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Predicted band size:** 44 kDa **Observed band size:** 47 kDa

**Lanes 1 - 2:** Merged signal (red and green). Green - ab154843 observed at 47 kDa. Red - loading control <u>ab8245</u> (Mouse anti-GAPDH antibody [6C5]) observed at 37 kDa.

ab154843 was shown to react with ATG4B in wild-type HEK293T cells in Western blot with loss of signal observed in ATG4B knockout cell line <a href="mailto:ab265814">ab265814</a> (ATG4B knockout cell lysate <a href="mailto:ab257364">ab257364</a>). Wild-type and ATG4B knockout HEK293T cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with ab154843 and <a href="mailto:ab8245">ab8245</a> (Mouse anti-GAPDH antibody [6C5]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preabsorbed (<a href="mailto:ab216773">ab216773</a>) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preabsorbed (<a href="mailto:ab216776">ab216776</a>) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.



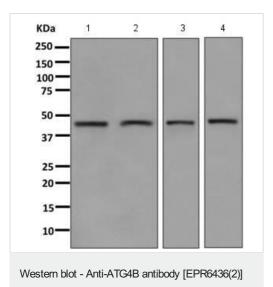
Western blot - Anti-ATG4B antibody [EPR6436(2)] (ab154843)

Lane 1: Wild-type HAP1 cell lysate (20 µg)

Lane 2: ATG4B knockout HAP1 cell lysate (20 µg)

**Lanes 1 - 2:** Merged signal (red and green). Green - ab154843 observed at 47 kDa. Red - loading control, **ab18058**, observed at 130 kDa.

ab154843 was shown to specifically react with ATG4B when ATG4B knockout samples were used. Wild-type and ATG4B knockout samples were subjected to SDS-PAGE. ab154843 and ab18058 (loading control to Vinculin) were diluted at 1/1000 and 1/10000 respectively and incubated overnight at 4°C. 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 1/10000 dilution for 1 hour at room temperature before imaging.



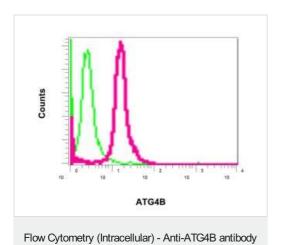
(ab154843)

All lanes: Anti-ATG4B antibody [EPR6436(2)] (ab154843) at 1/1000 dilution

Lane 1: Ramos cell lysate Lane 2: 293T cell lysate Lane 3: Jurkat cell lysate Lane 4: HeLa cell lysate

Lysates/proteins at 10 µg per lane.

Predicted band size: 44 kDa



labeling ATG4B (red), using ab154843 at a 1/100 dilution, and negative control cells probed with a Rabbit lgG (green)

Intracellular Flow Cytometry analysis of permeabilized Jurkat cells



Success from the Ethical standards first experiment Confirmed specificity

[EPR6436(2)] (ab154843)

compliant Animal-free production

Anti-ATG4B antibody [EPR6436(2)] (ab154843)

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

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