

## Product datasheet

### Anti-ATG12 antibody [EPR4800] $\alpha$ b109491


KO VALIDATED

Recombinant

RabMAb

[8 References](#) [6 Images](#)

#### Overview

Product name	Anti-ATG12 antibody [EPR4800]
Description	Rabbit monoclonal [EPR4800] to ATG12
Host species	Rabbit
Tested applications	<b>Suitable for:</b> WB, IHC-P <b>Unsuitable for:</b> Flow Cyt, ICC/IF or IP
Species reactivity	<b>Reacts with:</b> Human <b>Predicted to work with:</b> Mouse, Rat 
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	Human fetal kidney, HepG2, and Raji lysates; ATG12 transfected 293T lysates; Human breast carcinoma tissue.
General notes	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"> <li>- High batch-to-batch consistency and reproducibility</li> <li>- Improved sensitivity and specificity</li> <li>- Long-term security of supply</li> <li>- Animal-free production</li> </ul> <p>For more information <a href="#">see here</a>.</p> <p>Our RabMAb<sup>®</sup> technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to <a href="#">RabMAb<sup>®</sup> patents</a>.</p>

#### Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.
Storage buffer	pH: 7.20 Preservative: 0.05% Sodium azide Constituents: 0.1% BSA, 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue culture supernatant
Purity	Protein A purified
Clonality	Monoclonal

**Clone number** EPR4800

**Isotype** IgG

## Applications

**The Abpromise guarantee** Our **Abpromise guarantee** covers the use of ab109491 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
WB		1/1000 - 1/10000. Predicted molecular weight: 15 kDa.
IHC-P		1/250 - 1/500. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

**Application notes** Is unsuitable for Flow Cyt, ICC/IF or IP.

## Target

**Function** Ubiquitin-like protein required for autophagy. Conjugated to ATG3 and ATG5.

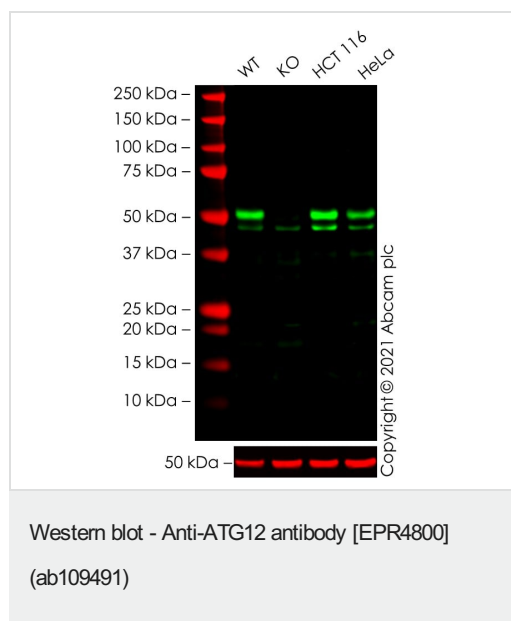
**Tissue specificity** Ubiquitous.

**Sequence similarities** Belongs to the ATG12 family.

**Domain** Shares weak sequence similarity with ubiquitin family, but contains an 'ubiquitin superfold' and the C-terminal Gly is required for isopeptide linkage.

**Cellular localization** Cytoplasm.

## Images



**All lanes** : Anti-ATG12 antibody [EPR4800] (ab109491) at 1/1000 dilution

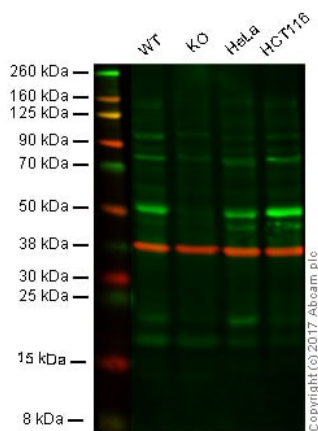
**Lane 1** : Wild-type THP-1 cell lysate  
**Lane 2** : ATG12 knockout THP-1 cell lysate  
**Lane 3** : HCT 116 cell lysate  
**Lane 4** : HeLa cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

**Predicted band size:** 15 kDa  
**Observed band size:** 52 kDa

False colour image of Western blot: Anti-ATG12 antibody [EPR4800] staining at 1/1000 dilution, shown in green; Mouse anti-Alpha Tubulin [DM1A] ([ab7291](#)) loading control staining at 1/20000 dilution, shown in red. In Western blot, ab109491 was shown to bind specifically to ATG12. A band likely to be the unfunctional complex with ATG5 was observed at 52 kDa in wild-type THP-1 cell lysates with no signal observed at this size in Atg12 knockout cell line [ab277831](#) (knockout cell lysate [ab278183](#)) - unconjugated functional form not observed at 15 kDa. To generate this image, wild-type and Atg12 knockout THP-1 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 (IRDye<sup>®</sup> 800CW) preabsorbed ([ab216773](#)) and Goat anti-Mouse IgG H&L (IRDye<sup>®</sup> 680RD) preabsorbed ([ab216776](#)) at 1/20000 dilution.



Western blot - Anti-ATG12 antibody [EPR4800] (ab109491)

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

**Lane 2:** ATG12 knockout HAP1 whole cell lysate (20 µg)

**Lane 3:** HeLa whole cell lysate (20 µg)

**Lane 4:** HCT116 whole cell lysate (20 µg)

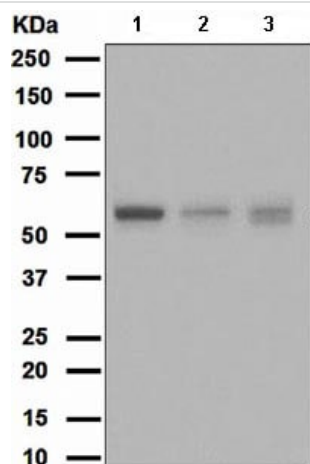
**Lanes 1 - 4:** Merged signal (red and green). Green - ab109491 observed at 55 kDa. Observed band shows ATG12-ATG5 conjugation. Red - loading control, [ab8245](#), observed at 37 kDa.

Ab109491 was shown to specifically react with

ATG12 in wild-type cells along with additional cross-reactive bands.

The band was not seen in ATG12 knockout HAP1 cells. Wild-type and ATG12 knockout samples were subjected to SDS-PAGE.

Ab109491 and [ab8245](#) (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/10000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye<sup>®</sup> 800CW) preabsorbed [ab216773](#) and Goat anti-Mouse IgG H&L (IRDye<sup>®</sup> 680RD) preabsorbed [ab216776](#) secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-ATG12 antibody [EPR4800]  
(ab109491)

**All lanes :** Anti-ATG12 antibody [EPR4800] (ab109491) at 1/1000 dilution

**Lane 1 :** Human fetal kidney tissue lysate

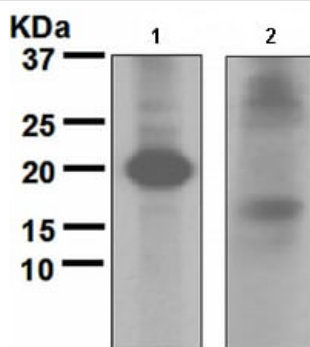
**Lane 2 :** HepG2 cell lysate

**Lane 3 :** Raji lysate

Lysates/proteins at 10 µg per lane.

**Predicted band size:** 15 kDa

Observed band shows ATG12-ATG5 conjugation



Western blot - Anti-ATG12 antibody [EPR4800]  
(ab109491)

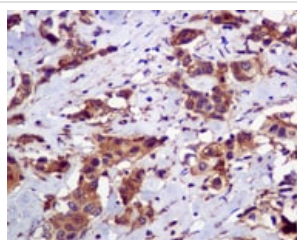
**All lanes :** Anti-ATG12 antibody [EPR4800] (ab109491) at 1/1000 dilution

**Lane 1 :** ATG12 transfected 293T lysate

**Lane 2 :** Non-transfected 293T lysate

Lysates/proteins at 10 µg per lane.

**Predicted band size:** 15 kDa



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ATG12 antibody [EPR4800] (ab109491)

ab109491, at 1/250 dilution, staining ATG12 in paraffin-embedded Human breast carcinoma by Immunohistochemistry.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

### Why choose a recombinant antibody?



**Research with confidence**  
Consistent and reproducible results



**Long-term and scalable supply**  
Recombinant technology



**Success from the first experiment**  
Confirmed specificity



**Ethical standards compliant**  
Animal-free production

Anti-ATG12 antibody [EPR4800] (ab109491)

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

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