abcam

Product datasheet

Anti-ATG12 antibody [EPR4800] ab109491



ייבעדיו RabMAb

8 References 画像数6

製品の概要

製品名 Anti-ATG12 antibody [EPR4800]

製品の詳細 Rabbit monoclonal [EPR4800] to ATG12

由来種 Rabbit

アプリケーション 適用あり: WB, IHC-P

適用なし: Flow Cyt,ICC/IF or IP

種交差性 交差種: Human

交差が予測される動物種: Mouse, Rat 4

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

ポジティブ・コントロール Human fetal kidney, HepG2, and Raji lysates; ATG12 transfected 293T lysates; Human breast

carcinoma tissue.

特記事項 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**.

製品の特性

製品の状態

保存方法 Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.

バッファー pH: 7.20

Preservative: 0.05% Sodium azide

Constituents: 0.1% BSA, 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue

culture supernatant

精製度 Protein A purified

ポリモノ モノクローナル

lgG

アプリケーション

The Abpromise guarantee <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab109491の使用に適用されます アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
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.

追加情報

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

ターゲット情報

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

組織特異性 Ubiquitous.

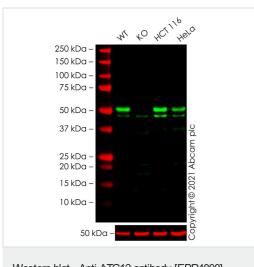
配列類似性 Belongs to the ATG12 family.

ドメイン Shares weak sequence similarity with ubiquitin family, but contains an 'ubiquitin superfold' and the

C-terminal Gly is required for isopeptide linkage.

細胞内局在 Cytoplasm.

画像



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

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

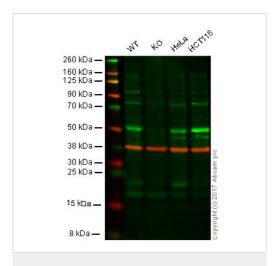
Lane 4: HeLa cell lysate

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[®] 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® 800CW) preabsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye® 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)

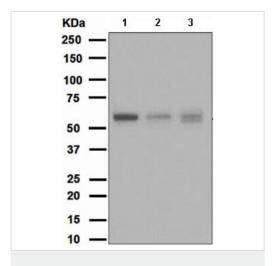
Lane 3: HeLa whole cell lysate (20 µg)

Lane 4: HCT116 whole cell lysate (20 µg)

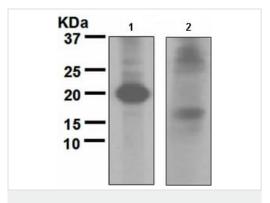
Lanes 1 - 4: Merged signal (red and green). Green - ab109491 observed at 55 kDa. Observed band shows ATG12-ATG5 conjugation. Red - loading control, **ab8245**, observed at 37 kDa.

Ab109491 was shown to specifically react with

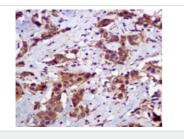
ATG12 in wild-type cells along with additional cross-reactive bands. The band was not seen in ATG12 knockout HAP1 cells. Wild-type and ATG12 knockout samples were subjected to SDS-PAGE. Ab109491 and ab8245 (Mouse anti GAPDH loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/10000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed ab216776 secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.



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



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



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - 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

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

Lane 1 : ATG12 transfected 293T lysate

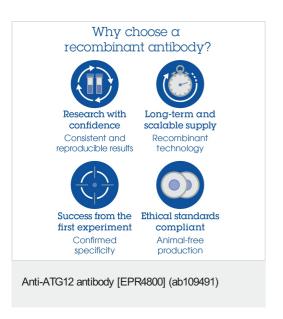
Lane 2 : Non-transfected 293T lysate

Lysates/proteins at 10 µg per lane.

Predicted band size: 15 kDa

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.



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