abcam

Product datasheet

Anti-LAMP2 antibody [EPR19531] ab199947

KO VALIDATED Recombinant RabMAb

2 References 7 Images

Overview

Product name	Anti-LAMP2 antibody [EPR19531]
Description	Rabbit monoclonal [EPR19531] to LAMP2
Host species	Rabbit
Tested applications	**Suitable for:** IHC-P, WB
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: Human fetal kidney, fetal spleen and placenta lysates; HeLa, THP-1, HepG2, HEK-293, JAR and Jurkat whole cell lysates. IHC-P: Human liver and breast cancer tissues.
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 +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR19531
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab199947 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
IHC-P		1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB		1/1000. Detects a band of approximately 110-130 kDa (predicted molecular weight: 45 kDa).

Target

Function Implicated in tumor cell metastasis. May function in protection of the lysosomal membrane from autodigestion, maintenance of the acidic environment of the lysosome, adhesion when expressed on the cell surface (plasma membrane), and inter-and intracellular signal transduction. Protects cells from the toxic effects of methylating mutagens.

Tissue specificity Isoform LAMP-2A is highly expressed in placenta, lung and liver, less in kidney and pancreas, low in brain and skeletal muscle. Isoform LAMP-2B is highly expressed in skeletal muscle, less in brain, placenta, lung, kidney and pancreas, very low in liver.

Involvement in disease Defects in LAMP2 are the cause of Danon disease (DAND) [MIM:300257]; also known as glycogen storage disease type 2B (GSD2B). DAND is a lysosomal glycogen storage disease characterized by the clinical triad of cardiomyopathy, vacuolar myopathy and mental retardation. It is often associated with an accumulation of glycogen in muscle and lysosomes.

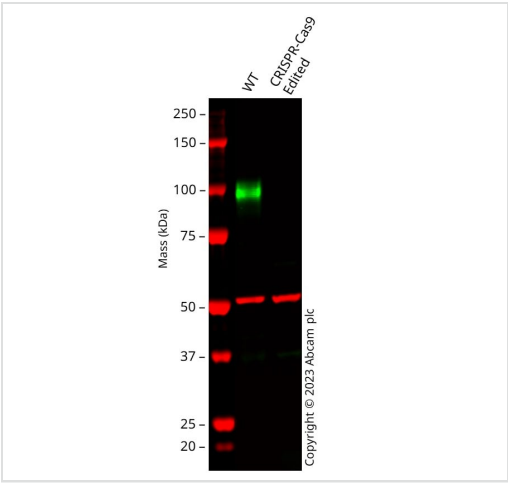
Sequence similarities Belongs to the LAMP family.

Post-translational modifications O- and N-glycosylated; some of the 16 N-linked glycans are polylactosaminoglycans.

Cellular localization Cell membrane. Endosome membrane. Lysosome membrane. This protein shuttles between lysosomes, endosomes, and the plasma membrane.

Form Alternative splicing produces 3 isoforms.

Images



Western blot - Anti-LAMP2 antibody [EPR19531] (ab199947)

All lanes : Anti-LAMP2 antibody [EPR19531] (ab199947) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell
Lane 2 : LAMP2 CRISPR-Cas9 edited HeLa cell lysate

Lysates/proteins at 20 µg per lane.

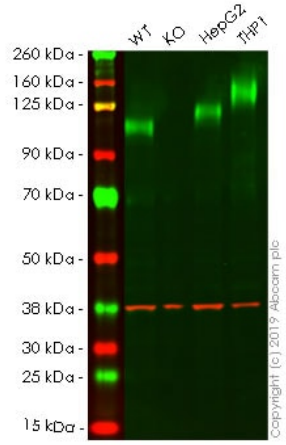
Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 45 kDa
Observed band size: 100 kDa

False colour image of Western blot: Anti-LAMP2 antibody [EPR19531] 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, ab199947 was shown to bind specifically to LAMP2. A band was observed at 100 kDa in wild-type HeLa cell lysates with no signal observed at this size in LAMP2 CRISPR-Cas9 edited cell line **ab255402** (CRISPR-Cas9 edited cell lysate **ab263861**). The band observed in the CRISPR-Cas9 edited lysate lane below 100 kDa is likely to represent a truncated form of LAMP2. This has not been investigated further and the functional properties of the gene product have not been determined. To generate this image, wild-type and LAMP2 CRISPR-Cas9 edited 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® 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.



Western blot - Anti-LAMP2 antibody [EPR19531] (ab199947)

All lanes : Anti-LAMP2 antibody [EPR19531] (ab199947) at 1/1000 dilution

Lane 1 : Wild-type HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate
Lane 2 : LAMP2 knockout HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate
Lane 3 : Hep G2 (Human liver hepatocellular carcinoma cell line) whole cell lysate
Lane 4 : THP-1 (Human monocytic leukemia cell line) whole cell lysate

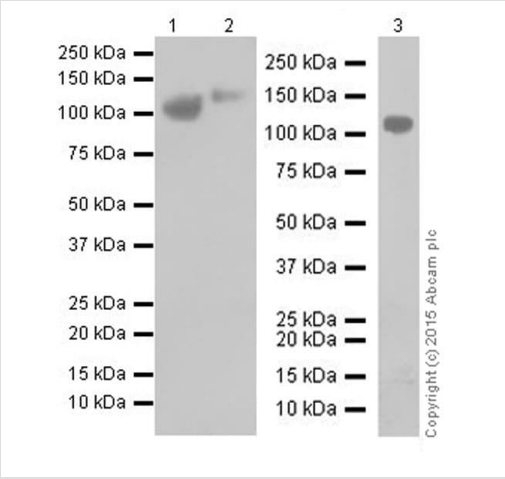
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 45 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab199947 observed at 45 kDa. Red - loading control, **ab130007**, observed at 130 kDa.

ab199947 was shown to specifically react with LAMP2 in wild-type HEK-293 cells as signal was lost in LAMP2 knockout cells. Wild-type and LAMP2 knockout samples were subjected to SDS-PAGE. Ab199947 and **ab130007** (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 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 1/20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-LAMP2 antibody [EPR19531] (ab199947)

All lanes : Anti-LAMP2 antibody [EPR19531] (ab199947) at 1/1000 dilution

Lane 1 : Human fetal kidney lysate
Lane 2 : Human fetal spleen lysate
Lane 3 : Human placenta lysate

Lysates/proteins at 10 µg per lane.

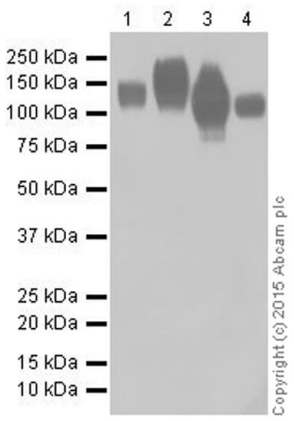
Secondary
All lanes : Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/100000 dilution

Predicted band size: 45 kDa
Observed band size: 110-130 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.

The molecular weight observed is consistent with what has been described in the literature (PMID: 19828315) and lots of other products.



Western blot - Anti-LAMP2 antibody [EPR19531] (ab199947)

All lanes : Anti-LAMP2 antibody [EPR19531] (ab199947) at 1/1000 dilution

Lane 1 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate
Lane 2 : THP-1 (Human monocytic leukemia cell line) whole cell lysate
Lane 3 : JAR (Human placenta choriocarcinoma cell line) whole cell lysate
Lane 4 : Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate

Lysates/proteins at 20 µg per lane.

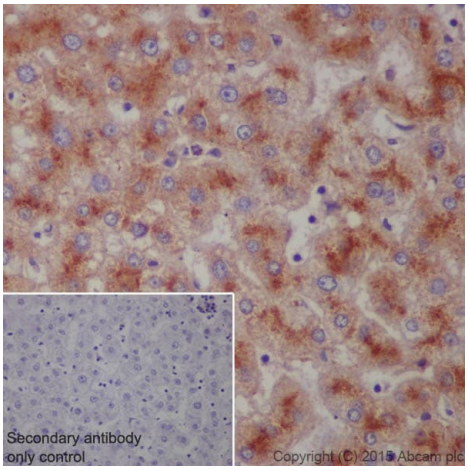
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 45 kDa
Observed band size: 110-130 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.

The molecular weight observed is consistent with what has been described in the literature (PMID: 19828315) and lots of other products.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-LAMP2 antibody [EPR19531] (ab199947)

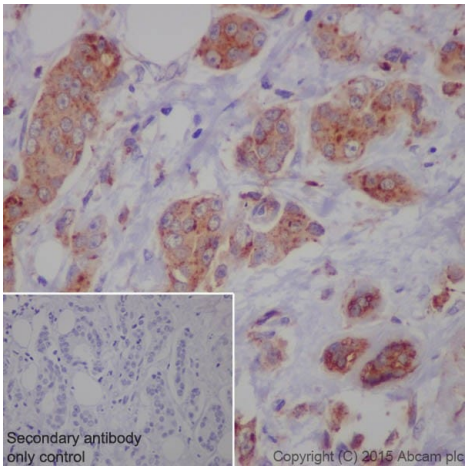
Immunohistochemical analysis of paraffin-embedded Human liver tissue labeling LAMP2 with ab199947 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

Cytoplasm staining on Human liver is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-LAMP2 antibody [EPR19531] (ab199947)

Immunohistochemical analysis of paraffin-embedded Human breast cancer tissue labeling LAMP2 with ab199947 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

Cytoplasm staining on Human breast cancer is observed.

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Anti-LAMP2 antibody [EPR19531] (ab199947)

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