# abcam

### Product datasheet

# Anti-LYVE1 antibody [EPR21771] ab218535

Recombinant RabMAb

★★★★ 1 Abreviews 6 References 10 Images

#### Overview

**Product name** Anti-LYVE1 antibody [EPR21771]

**Description** Rabbit monoclonal [EPR21771] to LYVE1

**Host species** Rabbit

**Tested applications** Suitable for: IP, WB, IHC-P, IHC-Fr, Flow Cyt

Species reactivity Reacts with: Mouse

**Immunogen** Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

Positive control WB: Mouse lymph node and lung lysates; bEnd.3 whole cell lysate. IHC-P: Mouse liver, lung and

colon tissues. IHC-Fr: Mouse liver and stomach tissues. Flow Cyt: bEnd.3 cells. IP: Mouse lung

lysate.

**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: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

**Purity** Protein A purified

Clonality Monoclonal Clone number EPR21771

**Isotype** IgG

#### **Applications**

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab218535 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
IP		1/30.
WB		1/1000. Detects a band of approximately 34-70 kDa (predicted molecular weight: 35 kDa).
IHC-P	<b>★</b> ★ ★ ★ ★ <b>(1)</b>	1/5000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
IHC-Fr		1/500. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
Flow Cyt		1/60.

**Function** 

Ligand-specific transporter trafficking between intracellular organelles (TGN) and the plasma membrane. Plays a role in autocrine regulation of cell growth mediated by growth regulators containing cell surface retention sequence binding (CRS). May act as a hyaluronan (HA) transporter, either mediating its uptake for catabolism within lymphatic endothelial cells themselves, or its transport into the lumen of afferent lymphatic vessels for subsequent re-uptake and degradation in lymph nodes.

Tissue specificity

Mainly expressed in endothelial cells lining lymphatic vessels.

Sequence similarities

Contains 1 Link domain.

Post-translational modifications

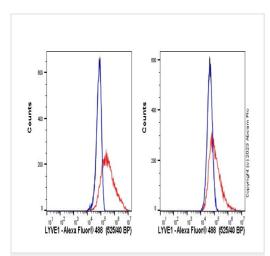
O-glycosylated.

Cellular localization

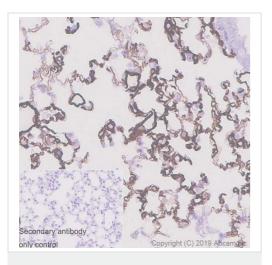
Membrane. Localized to the plasma membrane and in vesicles near extranuclear membranes which may represent trans-Golgi network (TGN) and endosomes/prelysosomeal compartments.

Undergoes ligand-dependent internalization and recycling at the cell surface.

#### **Images**



Flow Cytometry - Anti-LYVE1 antibody [EPR21771] (ab218535)



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-LYVE1 antibody
[EPR21771] (ab218535)

Flow cytometry overlay histogram showing left, bEND.3 treated with 100ng/mL TNF-alpha for 24h and right, negative untreated bEND.3 stained with ab218535 (red line). The cells were incubated in 1x PBS containing 10% normal goat serum to block non-specific protein-protein interactionfollowed by the antibody (ab218535) (1x  $10^6$  in  $100\mu$ l at  $10.0\mu$ g/ml (1/209)) for 30min on ice.

The secondary antibody Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min on ice

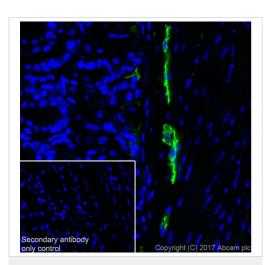
Isotype control antibody (black line) was Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control used at the same concentration and conditions as the primary antibody. Unlabelled sample (blue line) was also used as a control.

Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.

Immunohistochemical analysis of paraffin-embedded mouse lung tissue labeling LYVE1 with ab218535 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Positive staining on the lymphatic endothelial cells of mouse lung is observed.

Counter stained with hematoxylin. Heat mediated antigen retrieval using ab93684 (Tris/EDTA buffer, pH 9.0).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.

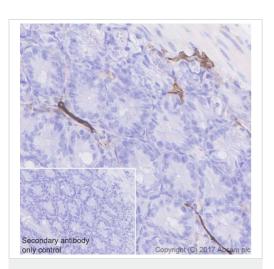


Immunohistochemistry (Frozen sections) - Anti-LYVE1 antibody [EPR21771] (ab218535)

Immunohistochemical analysis of 4% paraformaldehyde-fixed, 0.2% Triton X-100 permeabilized frozen mouse stomach tissue labeling LYVE1 with ab218535 at 1/5000 dilution (green), followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor<sup>®</sup> 488) (ab150077) secondary antibody at 1/1000 dilution (green). Positive staining of the endothelium of lymph vessels in the submucosae on mouse stomach tissue section (PMID: 15705793).

The nuclear counter stain is DAPI (blue).

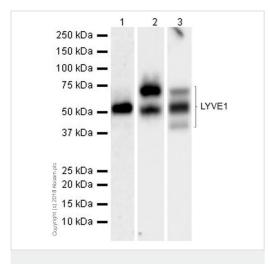
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (Alexa Fluor<sup>®</sup> 488) (ab150077) secondary antibody at 1/1000 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-LYVE1 antibody
[EPR21771] (ab218535)

Immunohistochemical analysis of paraffin-embedded mouse colon tissue labeling LYVE1 with ab218535 at 1/5000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Positive staining on the lymphatic endothelium of mouse colon (PMID: 14722766). Counter stained with Hematoxylin. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) Ready to use.



Western blot - Anti-LYVE1 antibody [EPR21771] (ab218535)

**All lanes :** Anti-LYVE1 antibody [EPR21771] (ab218535) at 1/1000 dilution

Lane 1: Mouse lymph node lysate

Lane 2: bEnd.3 (mouse brain endothelioma cell line) whole cell

lysate

Lane 3: Mouse lung lysate

Lysates/proteins at 20 µg per lane.

#### Secondary

**All lanes :** Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/10000 dilution

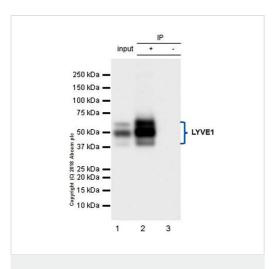
Developed using the ECL technique.

**Predicted band size:** 35 kDa **Observed band size:** 34-70 kDa

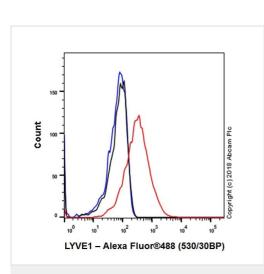
**Exposure time :** Lane 1: 3 minutes; Lane 2: 32 seconds; Lane 3: 5 seconds.

Blocking/Dilution buffer: 5% NFDM/TBST.

Several bands are observed including soluble, glycosylated and non-glycosylated forms which are consistent with the literature (PMID: 26966180).



Immunoprecipitation - Anti-LYVE1 antibody [EPR21771] (ab218535)



Flow Cytometry - Anti-LYVE1 antibody [EPR21771] (ab218535)

LYVE1 was immunoprecipitated from 0.35 mg mouse lung lysate with ab218535 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab218535 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/5000 dilution.

Lane 1: Mouse lung lysate 10 µg (Input).

Lane 2: ab218535 IP in mouse lung lysate.

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab218535 in mouse lung lysate.

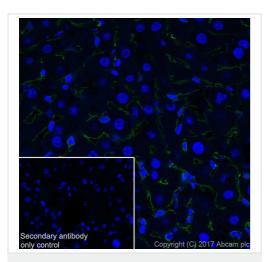
Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 8 seconds.

Several bands are observed including soluble, glycosylated and non-glycosylated forms which are consistent with the literature (PMID: 26966180).

Flow cytometric analysis of bEnd.3 (mouse brain endothelioma cell line) cells labeling LYVE1 with ab218535 at 1/60 dilution (red) compared with a Rabbit lgG, monoclonal [EPR25A] - Isotype Control (ab172730) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) (ab150077) at 1/2000 dilution was used as the secondary antibody.

Gated on total viable cells.

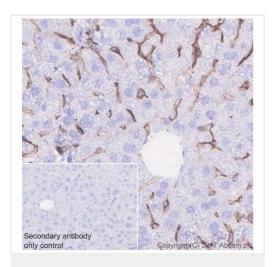


Immunohistochemistry (Frozen sections) - Anti-LYVE1 antibody [EPR21771] (ab218535)

Immunohistochemical analysis of 4% paraformaldehyde-fixed, 0.2% Triton X-100 permeabilized frozen mouse liver tissue labeling LYVE1 with ab218535 at 1/500 dilution (green), followed by Goat Anti-Rabbit lgG H&L (Alexa Fluor<sup>®</sup> 488) (ab150077) secondary antibody at 1/1000 dilution (green). Positive staining of the endothelium of sinusoid blood vessels on mouse liver tissue section (PMID: 11719431).

The nuclear counter stain is DAPI (blue).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (Alexa Fluor<sup>®</sup> 488) (ab150077) secondary antibody at 1/1000 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-LYVE1 antibody
[EPR21771] (ab218535)

Immunohistochemical analysis of paraffin-embedded mouse liver tissue labeling LYVE1 with ab218535 at 1/5000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Positive staining on the endothelial surface of mouse hepatic sinusoids (PMID: 16353487; PMID: 11719431). Counter stained with Hematoxylin. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use.



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