# abcam

## Product datasheet

## Anti-L1CAM antibody [EPR23241-224] ab270455



★★★★★ 1 Abreviews 1 References 画像数 12

#### 製品の概要

製品名 Anti-L1CAM antibody [EPR23241-224]

製品の詳細 Rabbit monoclonal [EPR23241-224] to L1CAM

由来種 Rabbit

アプリケーション 適用あり: WB, IHC-P, Flow Cyt, ICC/IF, IP

種交差性 交差種: Human

免疫原 Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

ポジティブ・コントロール WB: Human brain tissue lysate; HeLa, A375 and MCF7 whole cell lysates. IHC-P: Human kidney,

spleen and cerebellum tissue. Flow Cyt: A375 and HeLa cells. IP: A375 whole cell lysate. ICC/IF:

A375 and HeLa cells.

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 Liquid

保存方法 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.

バッファー pH: 7.2

Preservative: 0.01% Sodium azide

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

精製度 Protein A purified

ポリ/モノ モノクローナル クローン名 EPR23241-224

## 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ab270455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)	1/1000. Detects a band of approximately 250, 80 kDa (predicted molecular weight: 140 kDa).
IHC-P		1/100. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
Flow Cyt		1/50 - 1/500.
ICC/IF		1/100 - 1/500.
IP		1/30.

#### ターゲット情報

#### 機能

## 関連疾患

Cell adhesion molecule with an important role in the development of the nervous system. Involved in neuron-neuron adhesion, neurite fasciculation, outgrowth of neurites, etc. Binds to axonin on neurons.

Defects in L1CAM are the cause of hydrocephalus due to stenosis of the aqueduct of Sylvius (HSAS) [MIM:307000]. Hydrocephalus is a condition in which abnormal accumulation of cerebrospinal fluid in the brain causes increased intracranial pressure inside the skull. This is usually due to blockage of cerebrospinal fluid outflow in the brain ventricles or in the subarachnoid space at the base of the brain. In children is typically characterized by enlargement of the head, prominence of the forehead, brain atrophy, mental deterioration, and convulsions. In adults the syndrome includes incontinence, imbalance, and dementia. HSAS is characterized by mental retardation and enlarged brain ventricles.

Defects in L1CAM are the cause of mental retardation-aphasia-shuffling gait-adducted thumbs syndrome (MASA) [MIM:303350]; also known as corpus callosum hypoplasia, psychomotor retardation, adducted thumbs, spastic paraparesis, and hydrocephalus or CRASH syndrome. MASA is an X-linked recessive syndrome with a highly variable clinical spectrum. Main clinical features include spasticity and hyperreflexia of lower limbs, shuffling gait, mental retardation, aphasia and adducted thumbs. The features of spasticity have been referred to as complicated spastic paraplegia type 1 (SPG1). Some patients manifest corpus callosum hypoplasia and hydrocephalus. Inter- and intrafamilial variability is very wide, such that patients with hydrocephalus, MASA, SPG1, and agenesis of corpus callosum can be present within the same family.

Defects in L1CAM are the cause of spastic paraplegia X-linked type 1 (SPG1) [MIM:303350]. Spastic paraplegia is a degenerative spinal cord disorder characterized by a slow, gradual, progressive weakness and spasticity of the lower limbs.

Note=Defects in L1CAM may contribute to Hirschsprung disease by modifying the effects of Hirschsprung disease-associated genes to cause intestinal aganglionosis.

Defects in L1CAM are a cause of partial agenesis of the corpus callosum (ACCPX) [MIM:304100]. A syndrome characterized by partial corpus callosum agenesis, hypoplasia of inferior vermis and cerebellum, mental retardation, seizures and spasticity. Other features include microcephaly, unusual facies, and Hirschsprung disease in some patients.

配列類似性

Belongs to the immunoglobulin superfamily. L1/neurofascin/NgCAM family.

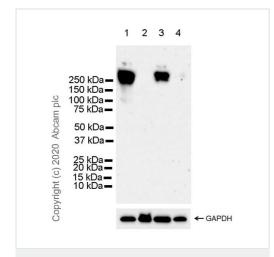
Contains 5 fibronectin type-III domains.

Contains 6 lg-like C2-type (immunoglobulin-like) domains.

細胞内局在

Cell membrane.

### 画像



Western blot - Anti-L1CAM antibody [EPR23241-224] (ab270455) **All lanes :** Anti-L1CAM antibody [EPR23241-224] (ab270455) at 1/1000 dilution

**Lane 1 :** Wild-type HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate

Lane 2: Human L1CAM knockout HeLa cell line (ab255401)

**Lane 3**: A375 (human malignant melanoma epithelial cell), whole cell lysate

**Lane 4**: A549 (human lung carcinoma epithelial cell), whole cell lysate

Lysates/proteins at 20 µg per lane.

#### Secondary

**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Predicted band size: 140 kDa Observed band size: 250 kDa

Exposure time: 3 minutes

## Blocking/Diluting buffer and concentration: 5% NFDM/TBST.

ab270455 was shown to specifically react with L1CAM in wild-type HeLa cells. Loss of signal was observed when knockout sample from Human L1CAM knockout HeLa cell line (ab255401) was used. Wild-type and L1CAM knockout samples were subjected to SDS-PAGE. ab270455 and ab181602 (Rabbit anti-GAPDH loading control) were incubated 1 hour at room temperature at 1/1000 dilution and 1/200,000 dilution respectively. Blots were developed with Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated

(<u>ab97051</u>) secondary antibody at 1/100,000 dilution for 1 hour at room temperature before imaging. The blot was developed on a BIO-RAD® ChemiDoc™ MP instrument using the ECL technique.

A549 is negative or very low expression cell line for L1CAM (PMID: 23511563).

MERGED ab270455

MERGED ab270455

Immunocytochemistry/ Immunofluorescence - Anti-L1CAM antibody [EPR23241-224] (ab270455)

Immunocytochemistry analysis of wild-type and L1CAM knockout HeLa (human epithelial cell line from cervix adenocarcinoma) cells labelling L1CAM with ab270455 at 1  $\mu$ g/ml. Cells were fixed with 100% methanol (5mins). An Alexa Fluor® 488-conjugated goat antirabbit lgG (2  $\mu$ g/ml) (ab150081) was used as the secondary antibody (shown in green). The cells were counterstained with a mouse monoclonal [DM1A] to alpha tubulin antibody (ab195889) at 1  $\mu$ g/ml and a Alexa Fluor® 647-conjugated goat secondary antibody to mouse lgG (ab150119) was used at 2  $\mu$ g/ml to detect the signal (shown in red). Nuclei counterstained with DAPI (blue).

Confocal image showing strong membranous and weak cytoplasmic staining in wild-type HeLa cells, while no staining in L1CAM knockout HeLa cells (ab255401).

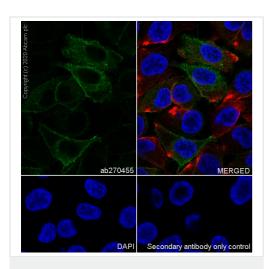


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-L1CAM antibody
[EPR23241-224] (ab270455)

Immunohistochemical analysis of paraffin-embedded Human cerebellum tissue labeling L1CAM with ab270455 at 1/100 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Positive staining on the molecular layer of the human cerebellum (PMID: 12514225). The section was incubated with ab270455 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

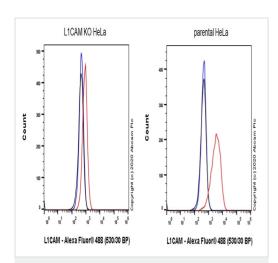
Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.



Immunocytochemistry/ Immunofluorescence - Anti-L1CAM antibody [EPR23241-224] (ab270455)

Immunocytochemistry/Immunofluorescence analysis of A375 (human malignant melanoma cell line) cells labelling L1CAM with ab270455 at 1/100. Cells were fixed with 4% paraformaldehyde. An Alexa Fluor<sup>®</sup> 488-conjugated goat anti-rabbit lgG (1/1000) (ab150077) was used as the secondary antibody. The cells were counterstained with an Alexa Fluor<sup>®</sup> 594-conjugated mouse antialpha tubulin antibody (1/200) (ab195889). Nuclei counterstained with DAPI (blue).

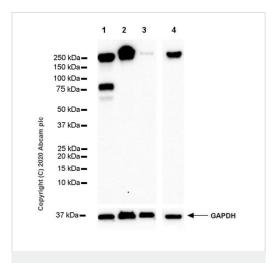
Confocal image showing strong membranous and weak cytoplasmic staining in A375 cells.



Flow Cytometry - Anti-L1CAM antibody [EPR23241-224] (ab270455)

Flow cytometric analysis of L1CAM KO HeLa (Human cervix adenocarcinoma epithelial cell, Left) / Parental HeLa (Right) cells labeling L1CAM with ab270455 at 1/50 dilution (Red) compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (Blue). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody.

Human L1CAM knockout HeLa cell line (<u>ab255401</u>). Gated on viable cells.



Western blot - Anti-L1CAM antibody [EPR23241-224] (ab270455)

**All lanes :** Anti-L1CAM antibody [EPR23241-224] (ab270455) at 1/1000 dilution

Lane 1: Human brain tissue lysate

**Lane 2 :** A375 (human malignant melanoma epithelial cell), whole cell lysate

**Lane 3 :** A549 (human lung carcinoma epithelial cell), whole cell lysate

**Lane 4 :** MCF7 (human breast adenocarcinoma epithelial cell), whole cell lysate

Lysates/proteins at 20 µg per lane.

## Secondary

**All lanes :** Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated (**ab97051**) at 1/100000 dilution

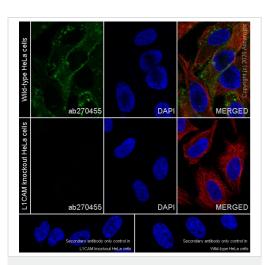
**Predicted band size:** 140 kDa **Observed band size:** 250,80 kDa

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

L1CAM is a glycoprotein. Full length 250-kDa L1CAM and cleaved 80-kDa are observed. The molecular weight observed is consistent with what have been described in literature (PMID 29127326).

A549 is negative or very low expression cell line for L1CAM (PMID: 23511563).

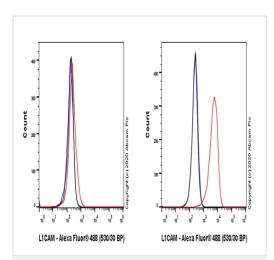
Exposure time: 3 minutes.



Immunocytochemistry/ Immunofluorescence - Anti-L1CAM antibody [EPR23241-224] (ab270455)

Immunocytochemistry/Immunofluorescence analysis of wild-type and L1CAM knockout HeLa (human epithelial cell line from cervix adenocarcinoma) cells labelling L1CAM with ab270455 at 1/500. Cells were fixed with 4% paraformaldehyde. An Alexa Fluor<sup>®</sup> 488-conjugated goat anti-rabbit lgG (1/1000) (ab150077) was used as the secondary antibody. The cells were counterstained with an Alexa Fluor<sup>®</sup> 594-conjugated mouse anti-alpha tubulin antibody (1/200) (ab195889). Nuclei counterstained with DAPI (blue).

Confocal image showing strong membranous and weak cytoplasmic staining in wild-type HeLa cells, while no staining in L1CAM knockout HeLa cells (ab255401).

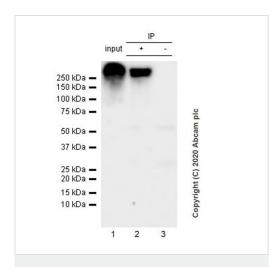


Flow Cytometry - Anti-L1CAM antibody [EPR23241-224] (ab270455)

Flow cytometric analysis of A549 (Human lung carcinoma epithelial cell, Left) / A375 (Human malignant melanoma epithelial cell, Right) cells labelling L1CAM with ab270455 at 1/500 dilution (Red) compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody.

A549 is negative or very low expression cell line for L1CAM (PMID: 23511563).

Gated on viable cells.



Immunoprecipitation - Anti-L1CAM antibody [EPR23241-224] (ab270455)

L1CAM was immunoprecipitated from 0.35 mg A375 (human malignant melanoma epithelial cell), whole cell lysate using ab270455 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab270455 at 1/500 dilution. VeriBlot for IP Detection Reagent (HRP)(ab131366) was used at 1/5000 dilution.

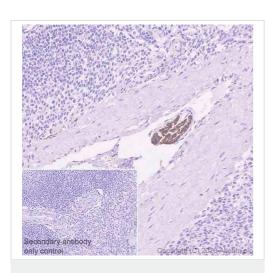
Lane 1: A375 (human malignant melanoma epithelial cell), whole cell lysate 10ug

Lane 2: ab270455 IP in A375 whole cell lysate

Lane 3: Rabbit monoclonal lgG ( $\underline{ab172730}$ ) instead of ab270455 in A375 whole cell lysate

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

Exposure time: 5.5 seconds.

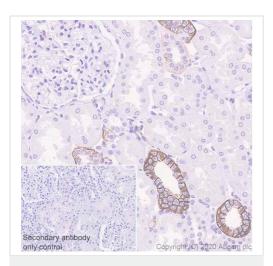


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-L1CAM antibody
[EPR23241-224] (ab270455)

Immunohistochemical analysis of paraffin-embedded Human spleen tissue labeling L1CAM with ab270455 at 1/100 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Positive staining on the nerve in human spleen (PMID: 26743472). The section was incubated with ab270455 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.



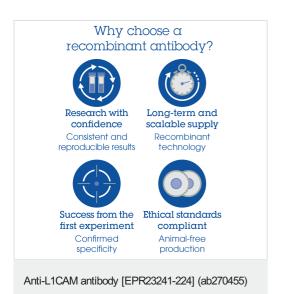
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-L1CAM antibody

[EPR23241-224] (ab270455)

Immunohistochemical analysis of paraffin-embedded Human kidney tissue labeling L1CAM with ab270455 at 1/100 dilution followed by a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101). Membranous staining on distal renal tubules in human kidney. The section was incubated with ab270455 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin.

Secondary antibody only control: Secondary antibody is a ready to use Rabbit specific IHC polymer detection kit HRP/DAB (ab209101).

Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution2) for 20 mins.



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