# abcam

## Product datasheet

## Anti-LYRIC/AEG1 antibody [EPR20797] ab227981

Recombinant RabMAb

#### 13 References 10 Images

#### Overview

**Product name** Anti-LYRIC/AEG1 antibody [EPR20797]

**Description** Rabbit monoclonal [EPR20797] to LYRIC/AEG1

**Host species** Rabbit

**Tested applications** Suitable for: Flow Cyt (Intra), WB, ICC/IF, IP, IHC-P

Species reactivity Reacts with: Mouse, Rat, Human

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

Positive control WB: MCF7, T47D, HeLa, PC-3 and PC-12 whole cell lysates; Human lung cancer lysate. IHC-P:

Human astrocytoma and ovarian cancer tissues; Mouse and rat cerebrum tissues. ICC/IF: HeLa

and PC-3 cells. Flow Cyt (intra): HeLa cells. IP: HeLa whole cell 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: 0.05% BSA, 40% Glycerol (glycerin, glycerine), PBS

**Purity** Protein A purified

Clonality Monoclonal Clone number EPR20797

**Isotype** IgG

### **Applications**

#### The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab227981 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
Flow Cyt (Intra)		1/500.
WB		1/1000. Detects a band of approximately 75, 80 kDa (predicted molecular weight: 64 kDa).
ICC/IF		1/100.
IP		1/30.
IHC-P		1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

I	a	rg	et

#### **Function**

Downregulates SLC1A2/EAAT2 promoter activity when expressed ectopically. Activates the nuclear factor kappa-B (NF-kappa-B) transcription factor. Promotes anchorage-independent growth of immortalized melanocytes and astrocytes which is a key component in tumor cell expansion. Promotes lung metastasis and also has an effect on bone and brain metastasis, possibly by enhancing the seeding of tumor cells to the target organ endothelium. Induces chemoresistance.

## Tissue specificity

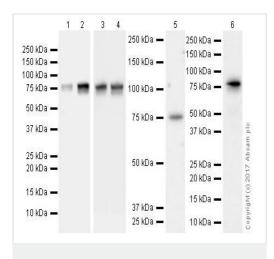
Widely expressed with highest levels in muscle-dominating organs such as skeletal muscle, heart, tongue and small intestine and in endocrine glands such as thyroid and adrenal gland.

Overexpressed in various cancers including breast, brain, prostate, melanoma and glioblastoma multiforme.

## Cellular localization

Endoplasmic reticulum membrane. Nucleus membrane. Cell junction > tight junction. Nucleus > nucleolus. Cytoplasm > perinuclear region. In epithelial cells, recruited to tight junctions (TJ) during the maturation of the TJ complexes. A nucleolar staining may be due to nuclear targeting of an isoform lacking the transmembrane domain (By similarity). TNF-alpha causes translocation from the cytoplasm to the nucleus.

## **Images**



Western blot - Anti-LYRIC/AEG1 antibody [EPR20797] (ab227981)

**All lanes :** Anti-LYRIC/AEG1 antibody [EPR20797] (ab227981) at 1/1000 dilution

**Lane 1 :** MCF7 (human breast adenocarcinoma cell line) whole cell lysate at 20  $\mu g$ 

**Lane 2 :** T-47D (human ductal breast epithelial tumor cell line) whole cell lysate at 20  $\mu$ g

Lane 3: HeLa (human epithelial cell line from cervix adenocarcinoma) whole cell lysate at 20 µg

**Lane 4 :** PC-3 (human prostate adenocarcinoma cell line) whole cell lysate at 20  $\mu g$ 

Lane 5: Human lung cancer lysate at 10 µg

**Lane 6 :** PC-12 (rat adrenal gland pheochromocytoma cell line) whole cell lysate at 10  $\mu g$ 

## Secondary

**Lanes 1-4 & 6 :** Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Lane 5: VeriBlot for IP Detection Reagent (HRP) (ab131366) at 1/4000 dilution

Predicted band size: 64 kDa

Observed band size: 75.80 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure times.

Lanes 1, 2 & 5: 3 minutes.

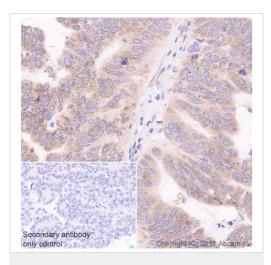
Lanes 3 & 4: 15 seconds.

Lane 6: 58 seconds.

The blot was developed on a BIO-RAD® ChemiDoc™ MP instrument.

This antibody detected LYRIC/AEG1 in rat PC-12 cells only, not in other rat tissues or cell lines tested.

The protein migrates as a 75/80kDa doublet, as has been observed in the literature (PMID: 23835593).

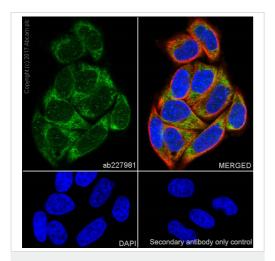


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-LYRIC/AEG1 antibody
[EPR20797] (ab227981)

Immunohistochemical analysis of paraffin-embedded human ovarian cancer tissue labeling LYRIC/AEG1 with ab227981 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Cytoplasmic staining in human ovarian cancer tissue (PMID: 27143933) is observed. Counter stained with hematoxylin.

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

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

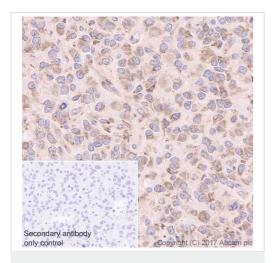


Immunocytochemistry/ Immunofluorescence - Anti-LYRIC/AEG1 antibody [EPR20797] (ab227981)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (human epithelial cell line from cervix adenocarcinoma) cells labeling LYRIC/AEG1 with ab227981 at 1/100 dilution followed by Goat Anti-Rabbit lgG H&L (Alexa Fluor<sup>®</sup> 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing cytoplasmic and weakly nuclear staining in HeLa cell line (PMID: 21750868).

The nuclear counter stain is DAPI (blue). Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor<sup>®</sup> 594) (ab195889) (red) at 1/200 dilution.

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

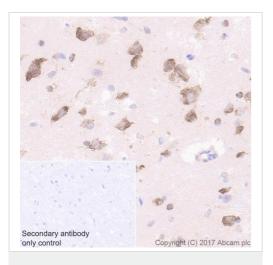


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-LYRIC/AEG1 antibody
[EPR20797] (ab227981)

Immunohistochemical analysis of paraffin-embedded human astrocytoma tissue labeling LYRIC/AEG1 with ab227981 at 1/2000 dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) Ready to use. Cytoplasmic staining in human astrocytoma tissue (PMID: 25197376) is observed. Counter stained with hematoxylin.

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

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

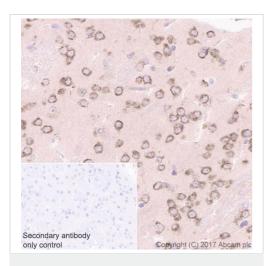


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-LYRIC/AEG1 antibody
[EPR20797] (ab227981)

Immunohistochemical analysis of paraffin-embedded mouse cerebrum tissue labeling LYRIC/AEG1 with ab227981 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Cytoplasmic staining in mouse cerebrum tissue (PMID:25197376) 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) Ready to use.

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



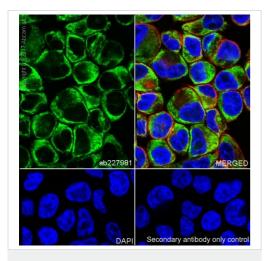
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-LYRIC/AEG1 antibody
[EPR20797] (ab227981)

Immunohistochemical analysis of paraffin-embedded rat cerebrum tissue labeling LYRIC/AEG1 with ab227981 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

Cytoplasmic staining in rat cerebrum tissue (PMID:25197376) is observed. Counter stained with hematoxylin.

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

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

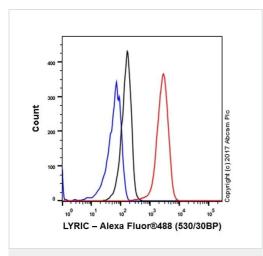


Immunocytochemistry/ Immunofluorescence - Anti-LYRIC/AEG1 antibody [EPR20797] (ab227981)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized PC-3 (human prostate adenocarcinoma cell line) cells labeling LYRIC/AEG1 with ab227981 at 1/100 dilution followed by Goat Anti-Rabbit lgG H&L (Alexa Fluor<sup>®</sup> 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing cytoplasmic staining in PC-3 cell line.

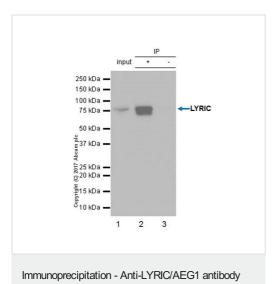
The nuclear counter stain is DAPI (blue). Tubulin is detected with Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor<sup>®</sup> 594) (ab195889) (red) at 1/200 dilution.

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.



Flow Cytometry (Intracellular) - Anti-LYRIC/AEG1 antibody [EPR20797] (ab227981)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed, 90% methanol-permeabilized HeLa (human epithelial cell line from cervix adenocarcinoma) cell line labeling LYRIC/AEG1 with ab227981 at 1/500 dilution (red) compared with a Rabbit IgG, monoclonal [EPR25A] - Isotype Control (ab172730) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) at 1/2000 dilution was used as the secondary antibody.



[EPR20797] (ab227981)

LYRIC/AEG1 was immunoprecipitated from 0.35 mg of HeLa (human epithelial cell line from cervix adenocarcinoma) whole cell lysate with ab227981 at 1/30 dilution. Western blot was performed from the immunoprecipitate using ab227981 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10000 dilution.

Lane 1: HeLa whole cell lysate 10 µg (Input).

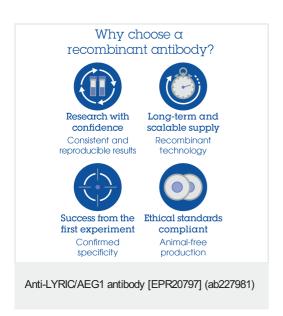
Lane 2: ab227981 IP in HeLa whole cell lysate.

Lane 3: Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab227981 in HeLa whole cell lysate.

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

Exposure time: 1 second.

The protein migrates as a 75/80kDa doublet, as has been observed in the literature (PMID: 23835593).



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