# abcam

## Product datasheet

## Anti-RILPL1 antibody [MJF-R41-21] ab302492





重组 RabMAb

11 图像

产品名称 Anti-RILPL1抗体[MJF-R41-21]

描述 兔单克隆抗体[MJF-R41-21] to RILPL1

宿主 Rabbit

经测试应用 适用于: ICC/IF, IP, WB

不适用于: Flow Cyt,IHC-Fr or IHC-P

种属反应性 与反应: Mouse, Rat, Human

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

阳性对照 WB: Wild-type A549 whole cell lysate. HeLa, NIH/3T3, C6, SH-SY5Y, HEK-293, Neuro-2a, and

> PC-12 whole cell lysates; rat and mouse brain tissue lysate. Human cerebellum, hypothalamus and heart tissue lysate. ICC: SH-SY5Y and Neuro-2a cells. IP: A549 whole cell lysate, mouse and

rat brain tissue lysates.

常规说明 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**.

This antibody was developed with support from The Michael J. Fox Foundation.

性能

形式 Liquid

存放说明 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.

**存储溶液** pH: 7.20

Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

纯**度** Protein A purified

**克隆** 单克隆

**克隆编号** MJF-R41-21

**同种型** lgG

#### 应用

## The Abpromise guarantee Abpromise™承诺保证使用ab302492于以下的经测试应用

"应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
ICC/IF		1/250.
IP		1/30.
WB		1/1000. Detects a band of approximately 47, 42 kDa (predicted molecular weight: 47 kDa).

应**用说明** Is unsuitable for Flow Cyt,IHC-Fr or IHC-P.

靶标

功能 Neuroprotective protein, which acts by sequestring GAPDH in the cytosol and prevent the

apoptotic function of GAPDH in the nucleus. Competes with SIAH1 for binding GAPDH (By

similarity). Does not regulate lysosomal morphology and distribution.

组织**特异性** Widely expressed. Expressed at lower level in liver and kidney.

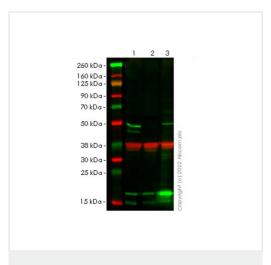
序列相似性 Belongs to the RILPL family.

Contains 1 RILP-like domain.

翻译后修饰 S-nitrosylation is required for the interaction with GAPDH.

细胞定位 Cytoplasm > cytosol.

#### 图片



Western blot - Anti-RILPL1 antibody [MJF-R41-21] (AB302492)

**All lanes**: Anti-RILPL1 antibody [MJF-R41-21] (ab302492) at 1/1000 dilution

**Lane 1 :** Wild-type A549 (human lung carcinoma epithelial cell), whole cell lysate

Lane 2: RILPL1 knockout A549 whole cell lysate

Lane 3: HeLa (human cervical adenocarcinoma epithelial cell), whole cell lysate

Lysates/proteins at 20 µg per lane.

#### Secondary

All lanes: Goat Anti-Rabbit lgG H&L (IRDye® 800CW)
(ab216773) and Goat Anti-Mouse lgG H&L (IRDye® 680RD)
(ab216776) at 1/10000 dilution

Performed under reducing conditions.

**Predicted band size:** 47 kDa **Observed band size:** 42,47 kDa

Blocking / Diluting buffer and:  $Intercept^{®}$  (TBS) Blocking Buffer diluted with an equal volume of 0.1% TBS.

False colour image of Western blot: Anti-RILPL1 antibody [MJF-R41-21] (ab302492) staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (ab8245) loading control staining at 1/20000 dilution, shown in red.

In Western blot, ab302492 was shown to bind specifically to RILPL1. Two bands were observed at 47/42 kDa in wild-type A549 cell lysates whereas no signal observed at this size in RILPL1 knockout cell line. To generate this image, wild-type and RILPL1 knockout A549 cell lysates were analyzed. First, samples were run on an SDS-PAGE gel and then transferred onto an immobilon-FL PVDF membrane. Membranes were blocked in Intercept<sup>®</sup> (TBS) Blocking Buffer diluted with an equal volume of 0.1% TBS 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.

The doublet bands represent two isoforms of RILPL1.

Wild-type and RILPL1 knockout A549 cell lysates were kindly provided by Dr. Dario Alessi, University of Dundee.

**All lanes :** Anti-RILPL1 antibody [MJF-R41-21] (ab302492) at 1/1000 dilution

**Lane 1 :** Wild-type A549 (human lung carcinoma epithelial cell), whole cell lysate

Lane 2: RILPL1 knockout A549 whole cell lysate

Lane 3: NIH/3T3 (mouse embryonic fibroblast), whole cell lysate

Lane 4: C6 (rat glial tumor glial cell), whole cell lysate

Lane 5 : Rat brain tissue lysate
Lane 6 : Rat kidney tissue lysate

Lysates/proteins at 20 µg per lane.

1 2 3 4 5 6

250 kDa—
150 kDa—
100 kDa—
75 kDa—
20 kDa—
20 kDa—
20 kDa—
21 th kDa—
21 th kDa—
22 th kDa—
23 th kDa—
24 th kDa—
25 th kDa—
26 th kDa—
27 th kDa—
27 th kDa—
28 th kDa—
29 th kDa—
20 kDa—
37 kDa—
4 th kD

Western blot - Anti-RILPL1 antibody [MJF-R41-21] (AB302492)

## **Secondary**

**All lanes :** Goat Anti-Rabbit  $\lg G \ H\&L \ (HRP) \ (\underline{ab97051})$  at 1/100000 dilution

**Predicted band size:** 47 kDa **Observed band size:** 42,47 kDa

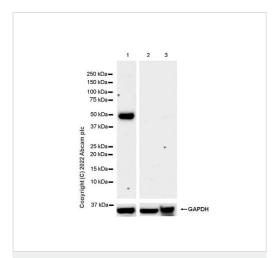
Exposure time: 3 minutes

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

The doublet bands represent two isoforms of RILPL1.

Negative control: kidney (PMID: 14668488).

Wild-type and RILPL1 knockout A549 cell lysates were kindly provided by Dr. Dario Alessi, University of Dundee.



Western blot - Anti-RILPL1 antibody [MJF-R41-21] (AB302492)

**All lanes :** Anti-RILPL1 antibody [MJF-R41-21] (ab302492) at 1/1000 dilution

Lane 1 : Mouse brain tissue lysate

Lane 2 : Mouse kidney tissue lysate

Lane 3 : Mouse liver tissue lysate

Lysates/proteins at 20 µg per lane.

#### **Secondary**

**All lanes :** Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity with human IgG at 1/20000 dilution

**Predicted band size:** 47 kDa **Observed band size:** 47 kDa

Exposure time: 103 seconds

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

Negative control: kidney, liver (PMID: 14668488).

**All lanes :** Anti-RILPL1 antibody [MJF-R41-21] (ab302492) at 1/1000 dilution

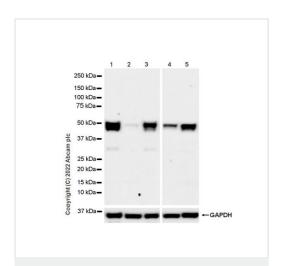
**Lane 1 :** SH-SY5Y (human neuroblastoma epithelial cell), whole cell lysate

**Lane 2 :** U937 (human histiocytic lymphoma monocyte), whole cell lysate

**Lane 3 :** HEK-293 (human embryonic kidney epithelial cell), whole cell lysate

**Lane 4 :** Neuro-2a (mouse neuroblastoma neuroblast), whole cell lysate

Lane 5 : PC-12 (rat adrenal gland pheochromocytoma), whole cell lysate



Western blot - Anti-RILPL1 antibody [MJF-R41-21] (AB302492)

Lysates/proteins at 20 µg per lane.

#### **Secondary**

All lanes : Goat Anti-Rabbit  $\lg G \ H\&L \ (HRP) \ (\underline{ab97051})$  at

1/100000 dilution

**Predicted band size:** 47 kDa **Observed band size:** 42,47 kDa

Exposure time: 37 seconds

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

The doublet bands represent two isoforms of RILPL1.

Low expression: U937 (Human Protein Atlas).

1 2 3

250 kDa—
150 kDa—
150 kDa—
100 kDa—
75 kDa—
50 kDa—
37 kDa—
25 kDa—
20 kDa—
15 kDa—
10 kDa—
10 kDa—

Western blot - Anti-RILPL1 antibody [MJF-R41-21] (AB302492)

**All lanes :** Anti-RILPL1 antibody [MJF-R41-21] (ab302492) at 1/1000 dilution

Lane 1: Human cerebellum tissue lysate

Lane 2: Human hypothalamus tissue lysate

Lane 3: Human heart tissue lysate

Lysates/proteins at 20 µg per lane.

#### **Secondary**

All lanes: Goat Anti-Rabbit IgG (HRP) with minimal cross-reactivity

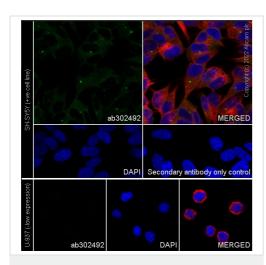
with human IgG at 1/2000 dilution

**Predicted band size:** 47 kDa **Observed band size:** 42,47 kDa

Exposure time: 26 seconds

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

The doublet bands represent two isoforms of RILPL1.

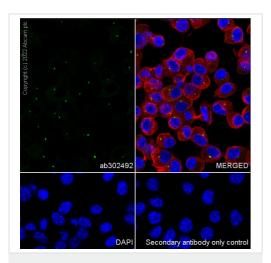


Immunocytochemistry/ Immunofluorescence - Anti-RILPL1 antibody [MJF-R41-21] (ab302492)

Immunofluorescent analysis of 4% Paraformaldehyde fixed, 0.1% Triton X-100 permeabilized SH-SY5Y (human neuroblastoma epithelial cell) labelling RILPL1 with ab302492 at 1/250 dilution followed by <u>ab150081</u> Goat Anti-Rabbit lgG H&L (Alexa Fluor  $^{(8)}$  488) preadsorbed secondary antibody at 1/1000 (2 µg/ml) dilution (green). Confocal image showing centrosome and cytoplasmic staining in SH-SY5Y cell line. The nuclear counterstain is DAPI (blue).

Tubulin is detected with  $\underline{ab195889}$  Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor  $^{\circledR}$  594) (red) at 1/200 dilution.

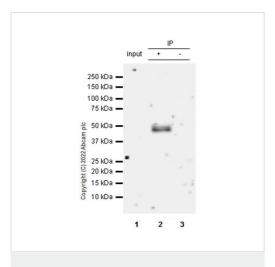
Low expression: U-937



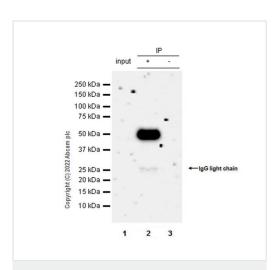
Immunocytochemistry/ Immunofluorescence - Anti-RILPL1 antibody [MJF-R41-21] (ab302492)

Immunofluorescent analysis of 4% Paraformaldehyde fixed, 0.1% Triton X-100 permeabilized Neuro-2a (mouse neuroblastoma neuroblast) labelling RILPL1 with ab302492 at 1/250 dilution followed by <a href="mailto:abb150081">ab150081</a> Goat Anti-Rabbit lgG H&L (Alexa Fluor® 488) preadsorbed secondary antibody at 1/1000 (2 µg/ml) dilution (green). Confocal image showing centrosome staining in Neuro-2a cell line. The nuclear counterstain is DAPI (blue).

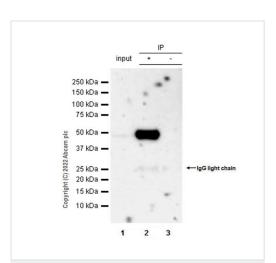
Tubulin is detected with <u>ab195889</u> Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor<sup>®</sup> 594) (red) at 1/200 dilution.



Immunoprecipitation - Anti-RILPL1 antibody [MJF-R41-21] (AB302492)



Immunoprecipitation - Anti-RILPL1 antibody [MJF-R41-21] (AB302492)



Immunoprecipitation - Anti-RILPL1 antibody [MJF-R41-21] (AB302492)

RILPL1 was immunoprecipitated from 0.35 mg A549 (human lung carcinoma epithelial cell), whole cell lysate with ab302492 at 1/30 dilution (2  $\mu$ g in 0.35 mg lysates). Western blot was performed on the immunoprecipitate using ab302492 at 1/1000 dilution. VeriBlot for IP secondary antibody (HRP)(ab131366) was used at 1/5000 dilution.

Lane 1 (Input): A549 whole cell lysate 10 µg

Lane 2 (+): A549 whole cell lysate

Lane 3 (-): Rabbit monoclonal IgG (ab172730) instead of ab302492 in A549 whole cell lysate

Blocking and dilution buffer: 5% NFDM/TBST.

Exposure time: 3 minutes.

RILPL1 was immunoprecipitated from 0.35 mg mouse brain tissue lysate with ab302492 at 1/30 dilution (2 µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab302492 at 1/1000 dilution. VeriBlot for IP secondary antibody (HRP)(ab131366) was used at 1/5000 dilution.

Lane 1 (Input): Mouse brain tissue lysate 10 μg

Lane 2 (+): Mouse brain tissue lysate

**Lane 3** (-): Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab302492 in mouse brain tissue lysate

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

Exposure time: 3 minutes.

RILPL1 was immunoprecipitated from 0.35 mg rat brain tissue lysate with ab302492 at 1/30 dilution (2 µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab302492 at 1/1000 dilution. VeriBlot for IP secondary antibody (HRP)(ab131366) was used at 1/5000 dilution.

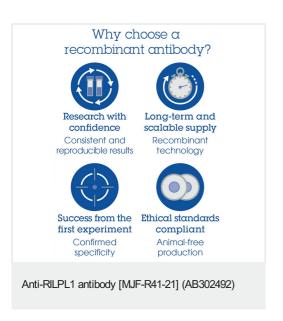
Lane 1 (Input): Rat brain tissue lysate 10 µg

Lane 2 (+): Rat brain tissue lysate

**Lane 3** (-): Rabbit monoclonal IgG (<u>ab172730</u>) instead of ab302492 in rat brain tissue lysate

Blocking and dilution buffer: 5% NFDM/TBST.

Exposure time: 3 minutes.



Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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