# abcam

# Product datasheet

# Anti-RAP1GAP antibody [Y134] ab32373



重组 RabMAb

★★★★★ 1 Abreviews 11 References 12 图像

#### 概述

产品名称 Anti-RAP1GAP抗体[Y134]

描述 兔单克隆抗体[Y134] to RAP1GAP

宿主 Rabbit

特异性 ab32373 recognises Rap1 GTPase-activating protein 1

经测试应用 适用于: Flow Cyt (Intra), ICC/IF, WB, IHC-P, IP

种属反应性 与反应: Mouse, Rat, Human

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

阳性对照 WB: Jurkat, SH-SY5Y cell lysate, mouse and rat brain lysate IHC: Human breast cancer ICC/IF:

HeLa cells IP: C6 cell lysate

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**.

# 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.

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 Y134

**同种型** lgG

# 应用

# The Abpromise guarantee Abpromise™承诺保证使用ab32373于以下的经测试应用

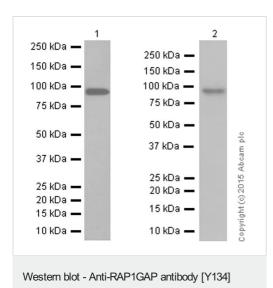
"应用说明"部分下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
Flow Cyt (Intra)		1/30 - 1/50.  ab172730 - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
ICC/IF		1/100 - 1/250.
WB		1/10000 - 1/50000. Detects a band of approximately 95 kDa (predicted molecular weight: 73 kDa).
IHC-P	**** (1)	1/1000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
IP		1/20 - 1/60.

## 靶标

功能	GTPase activator for the nuclear Ras-related regulatory protein RAP-1A (KREV-1), converting it to the putatively inactive GDP-bound state.
组织 <b>特异性</b>	Significant expression seen in the brain, kidney and pancreas. Abundant in the cerebral cortex and expressed at much lower levels in the spinal cord. Not detected in the lymphoid tissues.
序列相似性	Contains 1 GoLoco domain.  Contains 1 Rap-GAP domain.
细 <b>胞定位</b>	Golgi apparatus membrane.

## 图片



(ab32373)

**All lanes :** Anti-RAP1GAP antibody [Y134] (ab32373) at 1/10000 dilution (purified)

Lane 1 : Mouse brain lysate

Lane 2 : Rat brain lysate

Lysates/proteins at 20 µg per lane.

#### **Secondary**

**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) ( $\underline{ab97051}$ ) at 1/20000

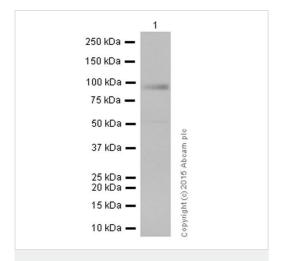
dilution

**Predicted band size:** 73 kDa **Observed band size:** 95 kDa

Blocking buffer: 5% NFDM/TBST Dilution buffer: 5% NFDM/TBST

The molecular weight observed is consistent with what has been described in the literature (PMID: 9346962, PMID: 15632203,

PMID: 30144784).



Anti-RAP1GAP antibody [Y134] (ab32373) at 1/10000 dilution (purified) + Jurkat cell lysate at 20  $\mu g$ 

#### Secondary

Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

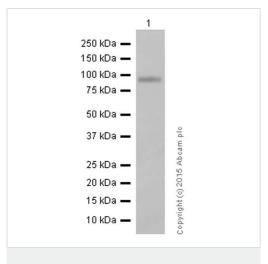
**Predicted band size:** 73 kDa **Observed band size:** 95 kDa

Western blot - Anti-RAP1GAP antibody [Y134] (ab32373)

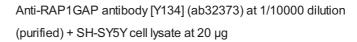
Blocking buffer: 5% NFDM/TBST Dilution buffer: 5% NFDM/TBST

The molecular weight observed is consistent with what has been described in the literature (PMID: 9346962, PMID: 15632203,

PMID: 30144784).



Western blot - Anti-RAP1GAP antibody [Y134] (ab32373)



#### Secondary

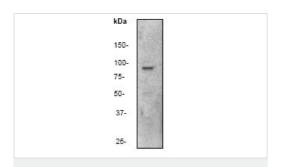
Anti-rabbit lgG (HRP), specific to the non-reduced form of lgG at 1/1000 dilution

**Predicted band size:** 73 kDa **Observed band size:** 95 kDa

Blocking buffer: 5% NFDM/TBST Dilution buffer: 5% NFDM/TBST

The molecular weight observed is consistent with what has been described in the literature (PMID: 9346962, PMID: 15632203,

PMID: 30144784).

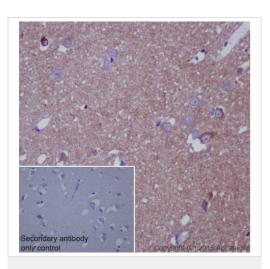


Western blot - Anti-RAP1GAP antibody [Y134] (ab32373)

Anti-RAP1GAP antibody [Y134] (ab32373) at 1/50000 dilution (unpurified) + Jurkat cell lysate

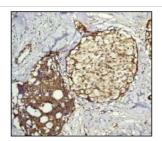
**Predicted band size:** 73 kDa **Observed band size:** 95 kDa

The molecular weight observed is consistent with what has been described in the literature (PMID: 9346962, PMID: 15632203, PMID: 30144784).



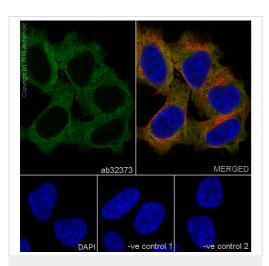
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RAP1GAP antibody
[Y134] (ab32373)

Immunohistochemical staining of paraffin embedded human cerebral cortex with purified ab32373 at a working dilution of 1/1000. The secondary antibody used is **ab97051**, a goat antirabbit lgG (H&L) at a dilution of 1/500. The sample is counterstained with hematoxylin. Antigen retrieval was performed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-RAP1GAP antibody
[Y134] (ab32373)

Immunohistochemical staining of paraffin-embedded human breast cancer tissue using unpurified ab32373 at 1/100 dilution.



Immunocytochemistry/ Immunofluorescence - Anti-

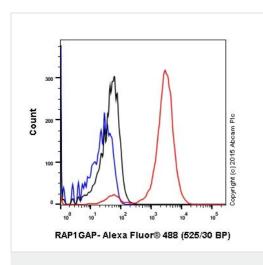
RAP1GAP antibody [Y134] (ab32373)

Immunocytochemistry/ Immunofluorescence - Anti-RAP1GAP antibody [Y134] (ab32373)

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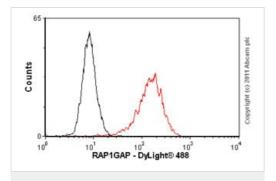
Immunofluorescence staining of HeLa cells with purified ab32373 at a working dilution of 1/250, counter-stained with DAPI. The secondary antibody was Alexa Fluor® 488 goat anti-rabbit (ab150077), used at a dilution of 1/1000. ab7291, a mouse antitubulin antibody (1/1000), was used to stain tubulin along with ab150120 (Alexa Fluor® 594 goat anti-mouse, 1/1000), shown in the top right hand panel. The cells were fixed in 4 % PFA and permeabilized using 0.1% Triton X 100. The negative controls are shown in bottom middle and right hand panels - for negative control 1, purified ab32373 was used at a dilution of 1/500 followed by an Alexa Fluor® 594 goat anti-mouse antibody (ab150120) at a dilution of 1/500. For negative control 2, ab7291 (mouse antitubulin) was used at a dilution of 1/500 followed by an Alexa Fluor® 488 goat anti-rabbit antibody (ab150077) at a dilution of 1/400.

ICC/IF image of unpurified ab32373 stained HeLa cells. The cells were 4% formaldehyde fixed (10 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab32373, 1µg/ml) overnight at +4°C. The secondary antibody (green) was ab96899, DyLight® 488 goat anti-rabbit lgG (H+L) used at a 1/250 dilution for 1h.Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.



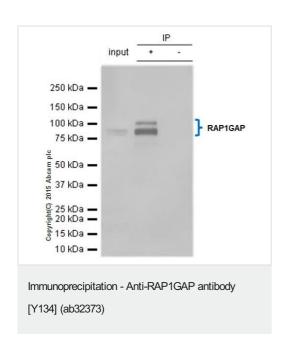
Flow Cytometry (Intracellular) - Anti-RAP1GAP antibody [Y134] (ab32373)

Overlay histogram showing Jurkat cells fixed in 4% PFA and stained with purified ab32373 at a dilution of 1/30 (red line). The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit at a dilution of 1/500. Rabbit monoclonal lgG was used as an isotype control (black line) and cells incubated in the absence of both primary and secondary antibody were used as a negative control (blue line).

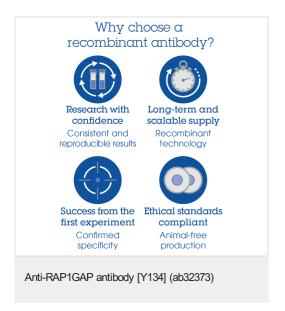


Flow Cytometry (Intracellular) - Anti-RAP1GAP antibody [Y134] (ab32373)

Overlay histogram showing SH-SY5Y cells stained with unpurified ab32373 (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab32373, 1/100) for 30 min at 22°C. The secondary antibody used was DyLight 488 goat anti-rabbit  $\log (H+L) (ab96899)$  at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit  $\log (monoclonal) (1 \mu g/1 x 10^6)$  cells) used under the same conditions. Acquisition of >5,000 events was performed.



ab32373 (purified) at 1/20 immunoprecipitating RAP1GAP in 10  $\mu$ g C6 cell lysate (Lanes 1 and 2, observed at 82-95 kDa). Lane 3 - Rabbit monoclonal IgG (<u>ab172730</u>). For western blotting, HRP Veriblot for IP Detection Reagent (<u>ab131366</u>) was used for detection (1/1000). Blocking buffer and concentration: 5% NFDM/TBST Dilution buffer and concentration: 5% NFDM/TBST



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