abcam

**ab133549**

# Anti-RALBP1 antibody [EPR6472]

Recombinant | RabMAb | KO Validated

## Key facts

| | |
|---|---|
| **Isotype** | IgG |
| **Host species** | Rabbit |
| **Storage buffer** | pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA |
| **Form** | Liquid |
| **Clonality** | Monoclonal |
| **Immunogen** | The exact immunogen used to generate this antibody is proprietary information. |
| **Clone number** | EPR6472 |
| **Purification technique** | Affinity purification Protein A |
| **Dissociation constant** | 5.42 x $10^{-11}$ M |
| **Concentration** | 0.177 mg/mL The concentration of this product may be batch-dependent<br>Batch concentration finder → |

## Reactivity data

## IHC-P

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/250 - 1/500 |
| **Notes** | - |

### Predicted

| | |
|---|---|
| **Species** | Mouse, Rat |
| **Dilution info** | - |
| **Notes** | - |

## IP

### Not recommended

| | |
|---|---|
| **Species** | Human, Mouse, Rat |
| **Dilution info** | - |
| **Notes** | - |

## WB

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/10000 - 1/50000 |
| **Notes** | - |

### Predicted

| | |
|---|---|
| **Species** | Mouse, Rat |
| **Dilution info** | - |
| **Notes** | - |

## ICC/IF

### Not recommended

| | |
|---|---|
| **Species** | Human, Mouse, Rat |
| **Dilution info** | - |
| **Notes** | - |

## Flow Cyt (Intra)

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/10 - 1/1000 |
| **Notes** | ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody. |

### Predicted

| | |
|---|---|
| **Species** | Mouse, Rat |
| **Dilution info** | - |
| **Notes** | - |

## Target data

See full target information RALBP1

| | |
|---|---|
| **Function** | Multifunctional protein that functions as a downstream effector of RALA and RALB (PubMed:7673236). As a GTPase-activating protein/GAP can inactivate CDC42 and RAC1 by stimulating their GTPase activity (PubMed:7673236). As part of the Ral signaling pathway, may also regulate ligand-dependent EGF and insulin receptors-mediated endocytosis (PubMed:10910768, PubMed:12775724). During mitosis, may act as a scaffold protein in the phosphorylation of EPSIN/EPN1 by the mitotic kinase cyclin B-CDK1, preventing endocytosis during that phase of the cell cycle (PubMed:12775724). During mitosis, also controls mitochondrial fission as an effector of RALA (PubMed:21822277). Recruited to mitochondrion by RALA, acts as a scaffold to foster the mitotic kinase cyclin B-CDK1-mediated phosphorylation and activation of DNM1L (PubMed:21822277). Could also function as a primary ATP-dependent active transporter for glutathione conjugates of electrophiles. May also actively catalyze the efflux of a wide range of substrates including xenobiotics like doxorubicin (DOX) contributing to cell multidrug resistance. |

## Storage

| | |
|---|---|
| **Shipped at conditions** | Conditional Ambient |
| **Appropriate short-term storage conditions** | +4°C |
| **Appropriate long-term storage conditions** | -20°C |
| **Storage information** | Stable for 12 months at -20°C |

## Notes

**Patented technology**
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

**What are the advantages of a recombinant monoclonal antibody?**
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 batch production

For more information, read more on recombinant antibodies.

## Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

| | |
|---|---|
| **Activity summary** | RALBP1 also known as ralA binding protein 1 or RLIP76 is a multifunctional protein involved in various cellular processes. It has a molecular mass of approximately 76 kDa. RALBP1 often associates with cell membranes and is expressed ubiquitously in different tissues indicating its widespread role in cellular functioning. The protein interacts with small GTPases such as RalA playing critical roles in signal transduction processes. |
| **Biological function summary** | RALBP1 serves as an effector protein involved in several cellular activities including endocytosis and transport of glutathione conjugates. It functions as part of a complex with partner proteins facilitating the movement of substrates across cellular membranes. This role is essential for maintaining cellular homeostasis as it helps to eliminate toxic compounds and regulate intracellular signals. |
| **Pathways** | RALBP1 participates in the Rho family small GTPase signaling and the Ras signaling pathways. Within these pathways the protein coordinates with RalA and other GTPases to control the trafficking of vesicles and stress response mechanisms. Through these interactions RALBP1 plays an important role in cytoskeletal reorganization and cellular response to extracellular cues. |
| **Associated diseases and disorders** | RALBP1 has associations with cancer and metabolic conditions. Alterations in its function or expression levels may contribute to oncogenic processes as RALBP1 interacts with factors like the epidermal growth factor receptor (EGFR) which is often dysregulated in cancerous cells. Moreover its role in detoxification processes implicates RALBP1 in metabolic disorders potentially affecting the body's capability to manage oxidative stress. |

## Product promise

**Tested**

We have tested this species and application combination and it works. It is covered by our product promise.

**Expected**

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

**Predicted**

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

**Not recommended**

We do not recommend this combination. It is not covered by our product promise.

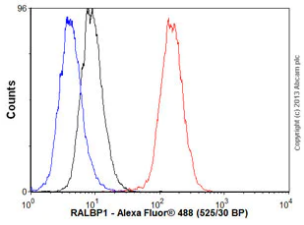
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In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
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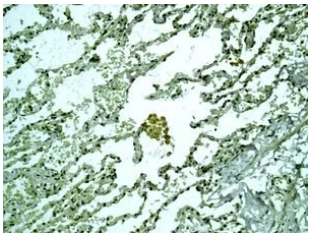
## 5 product images




### Flow Cytometry (Intracellular) - Anti-RALBP1 antibody [EPR6472] (ab133549)

RALBP1 Flow Cytometry (Intracellular) staining using rabbit Anti-RALBP1 antibody

Overlay histogram showing HEK293 cells stained with ab133549 (red line). The cells were fixed with 80% methanol (5 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 (ab133549, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H&L) (ab150077) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (0.1µg/1x$10^6$ cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This antibody gave a positive signal in HEK293 cells fixed with 4% paraformaldehyde (10 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.

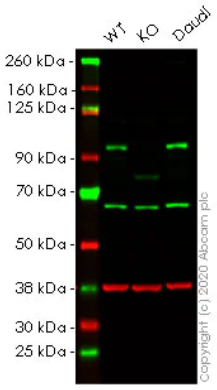


### Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-RALBP1 antibody [EPR6472] (ab133549)

RALBP1 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-RALBP1 antibody

Immunohistochemical analysis of paraffin embedded Human lung tissue labelling RALBP1 with ab133549 antibody at a dilution of 1/250.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.




## Western blot - Anti-RALBP1 antibody [EPR6472] (ab133549)

RALBP1 Western blot staining using rabbit Anti-RALBP1 antibody

**Lanes 1-3:** Merged signal (red and green). Green - ab133549 observed at 95 kDa. Red - loading control ab8245 observed at 37 kDa.
ab133549 Anti-RALBP1 antibody [EPR6472] was shown to specifically react with PDE10A in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab265404 (knockout cell lysate ab258167) was used. Wild-type and PDE10A knockout samples were subjected to SDS-PAGE. ab133549 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-RALBP1 antibody [EPR6472] (ab133549) at 1/1000 dilution

Lane 1:
Wild-type HeLa cell lysate at 20 µg

Lane 2:
RALBP1 knockout HeLa cell lysate at 20 µg

Lane 2:
Western blot - Human RALBP1 knockout HeLa cell line (ab265404)

Lane 3:
Daudi cell lysate at 20 µg

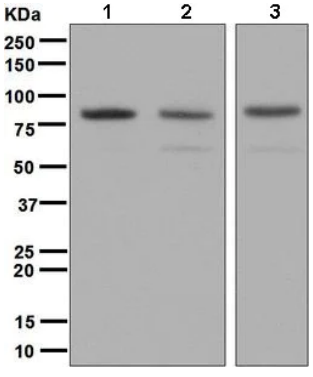
Secondary

All lanes:
Western blot - Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/20000 dilution

Performed under reducing conditions.

Predicted band size: 76 kDa

Observed band size: 95 kDa




## Western blot - Anti-RALBP1 antibody [EPR6472] (ab133549)

All lanes:
Western blot - Anti-RALBP1 antibody [EPR6472] (ab133549) at 1/10000 dilution

Lane 1:
MCF7 cell lysate at 10 µg

Lane 2:
HT-29 cell lysate at 10 µg

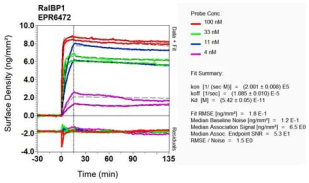
Lane 3:
HeLa cell lysate at 10 µg

Secondary

All lanes:
HRP labelled goat anti-rabbit at 1/2000 dilution

Predicted band size: 76 kDa

Observed band size: 95 kDa




## OI-RD Scanning - Anti-RALBP1 antibody [EPR6472] (ab133549)

We have systematically measured KD (the equilibrium dissociation constant between the antibody and its antigen), of more than 840 recombinant antibodies to assess not only their individual KD values but also to see the average affinity of antibody. Based on the comparison with published literature values for mouse monoclonal antibodies, Recombinant antibodies appear to be on average 1-2 order of magnitude higher affinity.

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