# Product datasheet

## Anti-RhoA antibody [1B12] ab54835

- **Product name**: Anti-RhoA antibody [1B12]
- **Description**: Mouse monoclonal [1B12] to RhoA
- **Host species**: Mouse
- **Tested applications**: Suitable for: WB, IHC-P, Flow Cyt, ICC/IF, ELISA
- **Species reactivity**: Reacts with: Rat, Human
- **Immunogen**: Recombinant full length protein, corresponding to amino acids 1-194 of Human RhoA
- **Positive control**: HeLa cells HL-60 cell lysate
- **General notes**: This product was changed from ascites to tissue culture supernatant on 15 May 2019. Please note that the dilutions may need to be adjusted accordingly. If you have any questions, please do not hesitate to contact our scientific support team.

## Properties

<table>
<thead>
<tr>
<th>Property</th>
<th>Details</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Form</strong></td>
<td>Liquid</td>
</tr>
<tr>
<td><strong>Storage instructions</strong></td>
<td>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.</td>
</tr>
<tr>
<td><strong>Storage buffer</strong></td>
<td>pH: 7.20</td>
</tr>
<tr>
<td></td>
<td>Constituent: 100% PBS</td>
</tr>
<tr>
<td><strong>Purity</strong></td>
<td>Tissue culture supernatant</td>
</tr>
<tr>
<td><strong>Purification notes</strong></td>
<td>Purified from TCS.</td>
</tr>
<tr>
<td><strong>Clonality</strong></td>
<td>Monoclonal</td>
</tr>
<tr>
<td><strong>Clone number</strong></td>
<td>1B12</td>
</tr>
<tr>
<td><strong>Isotype</strong></td>
<td>IgG1</td>
</tr>
<tr>
<td><strong>Light chain type</strong></td>
<td>lambda</td>
</tr>
</tbody>
</table>

## Applications

Our Abpromise guarantee covers the use of ab54835 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.
Regulates a signal transduction pathway linking plasma membrane receptors to the assembly of focal adhesions and actin stress fibers. Serves as a target for the yopT cysteine peptidase from Yersinia pestis, vector of the plague, and Yersinia pseudotuberculosis, which causes gastrointestinal disorders. May be an activator of PLCE1. Activated by ARHGEF2, which promotes the exchange of GDP for GTP.

Sequence similarities
Belongs to the small GTPase superfamily. Rho family.

Domain
The basic-rich region is essential for yopT recognition and cleavage.

Post-translational modifications
Substrate for botulinum ADP-ribosyltransferase.
Cleaved by yopT protease when the cell is infected by some Yersinia pathogens. This removes the lipid attachment, and leads to its displacement from plasma membrane and to subsequent cytoskeleton cleavage.
AMPylation at Tyr-34 and Thr-37 are mediated by bacterial enzymes in case of infection by H. somnus and V. parahaemolyticus, respectively. AMPylation occurs in the effector region and leads to inactivation of the GTPase activity by preventing the interaction with downstream effectors, thereby inhibiting actin assembly in infected cells. It is unclear whether some human enzyme mediates AMPylation; FICD has such ability in vitro but additional experiments remain to be done to confirm results in vivo.
Ubiquitinated by the BCR(BACURD1) and BCR(BACURD2) E3 ubiquitin ligase complexes, leading to its degradation by the proteasome, thereby regulating the actin cytoskeleton and cell migration.

Cellular localization
Cell membrane. Cytoplasm > cytoskeleton.

Images

<table>
<thead>
<tr>
<th>Application</th>
<th>Abreviews</th>
<th>Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>WB</td>
<td>⭐⭐⭐⭐⭐</td>
<td>Use at an assay dependent concentration. Predicted molecular weight: 22 kDa.</td>
</tr>
<tr>
<td>IHC-P</td>
<td>⭐⭐⭐⭐⭐</td>
<td>Use at an assay dependent concentration.</td>
</tr>
<tr>
<td>Flow Cyt</td>
<td>⭐⭐⭐⭐⭐</td>
<td>Use at an assay dependent concentration. ab170190 - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.</td>
</tr>
<tr>
<td>ICC/IF</td>
<td>⭐⭐⭐⭐⭐</td>
<td>Use at an assay dependent concentration.</td>
</tr>
<tr>
<td>ELISA</td>
<td></td>
<td>Use at an assay dependent concentration.</td>
</tr>
</tbody>
</table>

Target

Function
Regulates a signal transduction pathway linking plasma membrane receptors to the assembly of focal adhesions and actin stress fibers. Serves as a target for the yopT cysteine peptidase from Yersinia pestis, vector of the plague, and Yersinia pseudotuberculosis, which causes gastrointestinal disorders. May be an activator of PLCE1. Activated by ARHGEF2, which promotes the exchange of GDP for GTP.

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Ubiquitinated by the BCR(BACURD1) and BCR(BACURD2) E3 ubiquitin ligase complexes, leading to its degradation by the proteasome, thereby regulating the actin cytoskeleton and cell migration.

Cellular localization
Cell membrane. Cytoplasm > cytoskeleton.
RhoA antibody (ab54835) at 1ug/lane + HL-60 cell lysate (ab7914) at 25ug/lane.

This image was generated using the ascites version of the product.

**All lanes**: Anti-RhoA antibody [1B12] (ab54835) at 1 µg

**Lane 1**: RHOA transfected 293T lysate  
**Lane 2**: Non-transfected lysate

Lysates/proteins at 25 µg per lane.

**Secondary**

**All lanes**: Goat anti-mouse IgG

**Predicted band size**: 22 kDa

This image was generated using the ascites version of the product.
Immunocytochemistry/Immunofluorescence - Anti-RhoA antibody [1B12] (ab54835)

ICC/IF image of ab54835 stained HeLa cells (ab150035). The cells were 4% PFA 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 (ab54835, 1µg/ml) overnight at +4°C. The secondary antibody (green) was Alexa Fluor® 488 goat anti-mouse IgG (H+L) used at a 1/1000 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.

This image was generated using the ascites version of the product.

Flow Cytometry - Anti-RhoA antibody [1B12] (ab54835)

Overlay histogram showing HeLa cells (ab150035) stained with ab54835 (red line). The cells were fixed with 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 (ab54835, 1µg/1x10⁶ cells) for 30 min at 22°C. The secondary antibody used was DyLight® 488 goat anti-mouse IgG (H+L) (ab96879) at 1/500 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse IgG1 [ICIGG1] (ab91353, 2µg/1x10⁶ cells) used under the same conditions. Acquisition of >5,000 events was performed. This antibody gave a decreased signal in HeLa cells fixed with 4% paraformaldehyde (10 min)/permeabilized in 0.1% PBS-Tween used under the same conditions.

This image was generated using the ascites version of the product.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-RhoA antibody [1B12] (ab54835)

RhoA antibody (ab54835) used in immunohistochemistry at 5µg/ml on formalin fixed and paraffin embedded human lymphoma tissue.

This image was generated using the ascites version of the product.
ab54835 at 10 µg/ml staining RhoA in human Hela cells (ab150035) by Immunocytochemistry / Immunofluorescence.

This image was generated using the ascites version of the product.

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