abcam

Product datasheet

Anti-RAB10 (phospho T73) antibody [MJF-R21] ab230261



Recombinant

RabMAb

50 References 6 Images

Overview

Product name Anti-RAB10 (phospho T73) antibody [MJF-R21]

Description Rabbit monoclonal [MJF-R21] to RAB10 (phospho T73)

Host species Rabbit

Tested applications

Suitable for: WB, Dot blot

Species reactivity

Reacts with: Mouse, Human

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

Positive control WB: Wild-type MEF whole cell lysate; LRRK2 [R1441C] knock-in MEF whole cell lysate, 293T

transfected with RAB10 expression vector containing a myc-His-tag®, whole cell lysate. Dot Blot:

Rab10 (phospho T73) peptide.

General notes Please see PMID: 29127256. Lis P et al. Development of phospho-specific Rab protein

antibodies to monitor in vivo activity of the LRRK2 Parkinson's disease kinase. Biochem J 475:1-

22 (2018).

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.



Properties

Form Liquid

1

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: PBS, 40% Glycerol, 0.05% BSA

Purity Protein A purified

Clonality Monoclonal
Clone number MJF-R21

Isotype IgG

Applications

The Abpromise guarantee

Our Abpromise guarantee covers the use of ab230261 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
WB		1/1000. Detects a band of approximately 23 kDa (predicted molecular weight: 23 kDa).
Dot blot		1/1000.

Target

Function

The small GTPases Rab are key regulators of intracellular membrane trafficking, from the formation of transport vesicles to their fusion with membranes. Rabs cycle between an inactive GDP-bound form and an active GTP-bound form that is able to recruit to membranes different set of downstream effectors directly responsible for vesicle formation, movement, tethering and fusion (By similarity). That Rab is mainly involved in the biosynthetic transport of proteins from the Golgi to the plasma membrane. Regulates, for instance, SLC2A4/GLUT4 glucose transporter-enriched vesicles delivery to the plasma membrane. In parallel, it regulates the transport of TLR4, a toll-like receptor to the plasma membrane and therefore may be important for innate immune response. Plays also a specific role in asymmetric protein transport to the plasma membrane within the polarized neuron and epithelial cells. In neurons, it is involved in axonogenesis through regulation of vesicular membrane trafficking toward the axonal plasma membrane while in epithelial cells, it regulates transport from the Golgi to the basolateral membrane. Moreover, may play a role in the basolateral recycling pathway and in phagosome maturation. According to PubMed:23263280, may play a role in endoplasmic reticulum dynamics and morphology controlling tubulation along microtubules and tubules fusion.

Sequence similarities

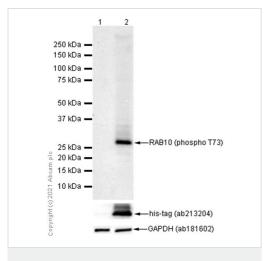
es Belongs to the small GTPase superfamily. Rab family.

Cellular localization

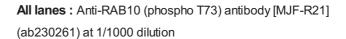
Cytoplasmic vesicle membrane. Golgi apparatus membrane. Golgi apparatus, trans-Golgi network membrane. Endosome membrane. Recycling endosome membrane. Cytoplasmic vesicle, phagosome membrane. Cell projection, cilium. Endoplasmic reticulum membrane. Associates with SLC2A4/GLUT4 storage vesicles (PubMed:22908308). Localizes to the base of the cilium (PubMed:20576682). Transiently associates with phagosomes (By similarity).

Localizes to the endoplasmic reticulum at domains of new tubule growth (PubMed:23263280).

Images



Western blot - Anti-RAB10 (phospho T73) antibody [MJF-R21] (ab230261)



Lane 1 : 293T (Human embryonic kidney epithelial cell) transfected with an empty vector, containing a myc-His-tag®, whole cell lysate **Lane 2 :** 293T transfected with RAB10 expression vector containing a myc-His-tag®, whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

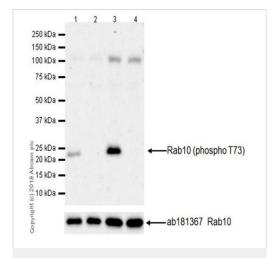
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 23 kDa

Exposure time: 180 seconds

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

Observed MW 28 kDa



Western blot - Anti-RAB10 (phospho T73) antibody [MJF-R21] (ab230261)

All lanes : Anti-RAB10 (phospho T73) antibody [MJF-R21] (ab230261) at 1/1000 dilution

Lane 1 : Wild-type MEF (mouse embryonic fibroblast cell line) whole cell lysate

Lane 2: Wild-type MEF treated with 100 nM MLi-2 for 90 minutes, whole cell lysate

Lane 3: LRRK2 [R1441C] knock-in MEF whole cell lysate

Lane 4: LRRK2 [R1441C] knock-in MEF treated with 100 nM MLi-2 for 90 minutes, whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Developed using the ECL technique.

Predicted band size: 23 kDa **Observed band size:** 23 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.

The LRRK2 pathogenic mutation R1441C increases LRRK2 activity and markedly elevates Rab10 phosphorylation in MEF (mouse embryonic fibroblasts).

The expression pattern is consistent with the literature (PMID: 29127256).

The cell lysates were kindly provided by our collaborator, Dr. Dario Alessi.

All lanes : Anti-RAB10 (phospho T73) antibody [MJF-R21] (ab230261) at 1/1000 dilution

Lane 1 : Wild-type A549 (human lung carcinoma cell line) whole cell lysate

Lane 2: Wild-type A549 treated with 100 nM MLi-2 for 90 minutes, whole cell lysate

Lane 3: Rab8A knock-out A549 whole cell lysate

Lane 4: Rab8A knock-out A549 treated with 100 nM MLi-2 for 90 minutes, whole cell lysate

Lane 5: Rab10 knock-out A549 whole cell lysate

Lane 6: Rab10 knock-out A549 treated with 100 nM MLi-2 for 90

minutes, whole cell lysate

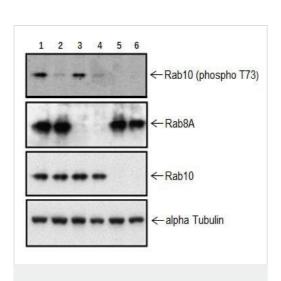
Lysates/proteins at 20 µg per lane.

Secondary

All lanes: HRP-labeled secondary antibody at 1/2500 dilution

Predicted band size: 23 kDa **Observed band size:** 23 kDa

Blocking buffer: 5% NFDM/TBST.

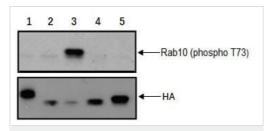


Western blot - Anti-RAB10 (phospho T73) antibody [MJF-R21] (ab230261)

Dilution buffer: 5% BSA/TBST.

The images were kindly provided by our collaborator, Dr. Dario Alessi, and have been published (PMID: 29127256).

Scanned with Licor Odyssey CLx.



Western blot - Anti-RAB10 (phospho T73) antibody [MJF-R21] (ab230261)

All lanes : Anti-RAB10 (phospho T73) antibody [MJF-R21] (ab230261) at 1/1000 dilution

Lane 1: HEK-293 (human epithelial cell line from embryonic kidney) cells transfected with LRRK2 [Y1699C] and HA-tagged Rab3A expression vectors, were treated with 150 nM MLi-2 for 90 minutes, whole cell lysate

Lane 2: HEK-293 cells transfected with LRRK2 [Y1699C] and HAtagged Rab8A expression vectors, were treated with 150 nM MLi-2 for 90 minutes, whole cell lysate

Lane 3: HEK-293 cells transfected with LRRK2 [Y1699C] and HAtagged Rab10 expression vectors, were treated with 150 nM MLi-2 for 90 minutes, whole cell lysate

Lane 4: HEK-293 cells transfected with LRRK2 [Y1699C] and HAtagged Rab35 expression vectors, were treated with 150 nM MLi-2 for 90 minutes, whole cell lysate

Lane 5: HEK-293 cells transfected with LRRK2[Y1699C] and HAtagged Rab43 expression vectors, were treated with 150 nM MLi-2 for 90 minutes, whole cell lysate

Lysates/proteins at 0.1 µg per lane.

Secondary

All lanes: IRDye 800CW secondary antibody at 1/25000 dilution

Predicted band size: 23 kDa
Observed band size: 23 kDa

Blocking buffer: 5% NFDM/TBST.

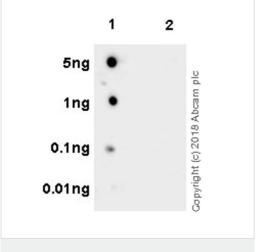
Dilution buffer: 5% BSA/TBST.

The LRRK2 pathogenic mutation Y1699C increases LRRK2 activity and markedly elevates the phosphorylation of Rab proteins.

The images were kindly provided by our collaborator Dr. Dario

Alessi, and have been published (PMID: 29127256).

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Dot Blot - Anti-RAB10 (phospho T73) antibody [MJF-R21] (ab230261)

Dot blot analysis of Rab10 (phospho T73) labeled with ab230261 at 1/1000 dilution.

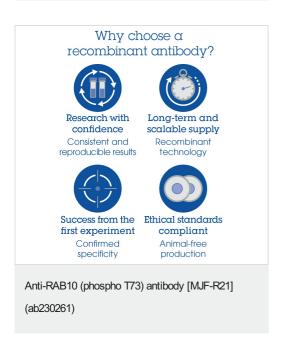
Lane 1: Rab10 (phospho T73) peptide;

Lane 2: Rab10 non-phospho peptide.

Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/100,000 dilution was used as secondary antibody.

Blocking/Dilution buffer: 5% NFDM/TBST.

Exposure time: 32 seconds.



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