

Anti-RAB11B Antibody Picoband™ (monoclonal, 6C5)

Catalog # ABO14921

Specification

Anti-RAB11B Antibody Picoband™ (monoclonal, 6C5) - Product Information

Application WB, IF, ICC, FC

Primary Accession
Host

O15907

Mouse

Isotype Mouse IgG2b
Reactivity Rat, Human, Mouse
Clansity Monoclanal

Clonality Monoclonal Format Lyophilized

Description

Anti-RAB11B Antibody Picoband™ (monoclonal, 6C5) . Tested in Flow Cytometry, IF, ICC, WB applications. This antibody reacts with Human, Mouse, Rat.

Reconstitution

Add 0.2ml of distilled water will yield a concentration of 500 µg/ml.

Anti-RAB11B Antibody Picoband™ (monoclonal, 6C5) - Additional Information

Gene ID 9230

Other Names

Ras-related protein Rab-11B, 3.6.5.2, GTP-binding protein YPT3, RAB11B, YPT3

Calculated MW

24 kDa KDa

Application Details

Western blot, 0.1-0.5 μg/ml, Human, Mouse, Rat

br> Immunocytochemistry/Immunofluorescence, 2 μg/ml, Human

br> Flow Cytometry, 1-3 μg/1x10^6 cells, Human

cells, Human

br>

Subcellular Localization

Recycling endosome membrane; Lipid-anchor; Cytoplasmic side. Cytoplasmic vesicle, secretory vesicle, synaptic vesicle membrane; Lipid-anchor; Cytoplasmic side. Cytoplasmic vesicle, phagosome membrane; Lipid-anchor; Cytoplasmic side. Recruited to phagosomes containing S. aureus.

Protein Name

Ras-related protein Rab-11B

Contents

Each vial contains 4mg Trehalose, 0.9mg NaCl, 0.2mg Na2HPO4, 0.05mg NaN3.

Immunogen

A synthetic peptide corresponding to a sequence at the C-terminus of human RAB11B, which shares 97.4% and 100% amino acid (aa) sequence identity with mouse and rat RAB11B, respectively.



Purification Immunogen affinity purified.

Cross ReactivityNo cross-reactivity with other proteins.

Storage

Store at -20°C for one year from date of receipt. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20°C for six months. Avoid repeated freeze-thaw cycles.

Anti-RAB11B Antibody Picoband™ (monoclonal, 6C5) - Protein Information

Name RAB11B (HGNC:9761)

Synonyms YPT3

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 (PubMed:14627637, PubMed: 19029296, PubMed:19244346, PubMed:20717956, PubMed: 21248079, PubMed:22129970, PubMed:26032412). The small Rab GTPase RAB11B plays a role in endocytic recycling, regulating apical recycling of several transmembrane proteins including cystic fibrosis transmembrane conductance regulator/CFTR, epithelial sodium channel/ENaC, potassium voltage-gated channel, and voltage-dependent L-type calcium channel. May also regulate constitutive and regulated secretion, like insulin granule exocytosis. Required for melanosome transport and release from melanocytes. Also regulates V-ATPase intracellular transport in response to extracellular acidosis (PubMed: 14627637, PubMed:19029296, PubMed:19244346, PubMed:20717956, PubMed:21248079, PubMed:22129970). Promotes Rabin8/RAB3IP preciliary vesicular trafficking to mother centriole by forming a ciliary targeting complex containing Rab11, ASAP1, Rabin8/RAB3IP, RAB11FIP3 and ARF4, thereby regulating ciliogenesis initiation (PubMed:25673879). On the contrary, upon LPAR1 receptor signaling pathway activation, interaction with phosphorylated WDR44 prevents Rab11-RAB3IP-RAB11FIP3 complex formation and cilia growth (PubMed: 31204173).

Cellular Location

Recycling endosome membrane {ECO:0000250|UniProtKB:P46638}; Lipid-anchor {ECO:0000250|UniProtKB:P46638}; Cytoplasmic side {ECO:0000250|UniProtKB:P46638}. Cytoplasmic vesicle, secretory vesicle, synaptic vesicle membrane {ECO:0000250|UniProtKB:O35509}; Lipid-anchor {ECO:0000250|UniProtKB:O35509}; Cytoplasmic side {ECO:0000250|UniProtKB:O35509}. Cytoplasmic vesicle, phagosome membrane;



Lipid-anchor; Cytoplasmic side. Cytoplasmic vesicle. Note=Recruited to phagosomes containing S.aureus. Colocalizes with RAB11AFIP1 on punctate vesicles (PubMed:26032412).

Anti-RAB11B Antibody Picoband™ (monoclonal, 6C5) - Protocols

Provided below are standard protocols that you may find useful for product applications.

- Western Blot
- Blocking Peptides
- Dot Blot
- <u>Immunohistochemistry</u>
- Immunofluorescence
- Immunoprecipitation
- Flow Cytomety
- Cell Culture

Anti-RAB11B Antibody Picoband™ (monoclonal, 6C5) - Images

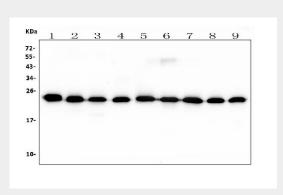


Figure 2. Western blot analysis of RAB11B using anti-RAB11B antibody (M04526). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.

Lane 1: rat brain tissue lysates,

Lane 2: rat lung whole cell lysates,

Lane 3: rat spleen whole cell lysates,

Lane 4: rat C6 whole cell lysates,

Lane 5: mouse brain whole cell lysates.

Lane 6: mouse lung whole cell lysates.

Lane 7: mouse spleen whole cell lysates.

Lane 8: mouse Neuro-2a whole cell lysates.

Lane 9: mouse RAW246.7 whole cell lysates.

After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-RAB11B antigen affinity purified polyclonal antibody (Catalog # M04526) at 0.5 g/mL overnight at 4C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-mouse IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1001) with Tanon 5200 system. A specific band was detected for RAB11B at approximately 24KD. The expected band size for RAB11B is at 24KD.



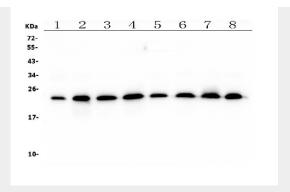


Figure 1. Western blot analysis of RAB11B using anti-RAB11B antibody (M04526). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions.

Lane 1: human HEK293 tissue lysates,

Lane 2: human Hela whole cell lysates,

Lane 3: human A549 whole cell lysates,

Lane 4: human placenta whole cell lysates,

Lane 5: human HepG2 whole cell lysates.

Lane 6: human Caco-2 whole cell lysates.

Lane 7: human THP-1 whole cell lysates.

Lane 8: human Raji whole cell lysates.

After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA for 50-90 minutes. Blocked the membrane with 5% Non-fat Milk/ TBS for 1.5 hour at RT. The membrane was incubated with mouse anti-RAB11B antigen affinity purified polyclonal antibody (Catalog # M04526) at 0.5 μ g/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-mouse IgG-HRP secondary antibody at a dilution of 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1001) with Tanon 5200 system. A specific band was detected for RAB11B at approximately 24KD. The expected band size for RAB11B is at 24KD.

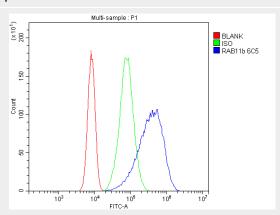


Figure 3.Flow Cytometry analysis of Caco-2 cells using anti-RAB11B antibody (M04526). Overlay histogram showing Caco-2 cells stained with M04526 (Blue line). The cells were blocked with 10% normal goat serum. And then incubated with mouse anti-RAB11B Antibody (M04526, 1 μ g/1x10⁶ cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-mouse IgG (BA1126, 5-10 μ g/1x10⁶ cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was mouse IgG (1 μ g/1x10⁶) used under the same conditions. Unlabelled sample (Red line) was also used as a control.



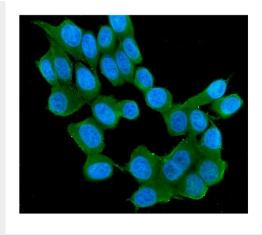


Figure 4. IF analysis of RAB11B using anti-RAB11B antibody (M04526).

RAB11B was detected in immunocytochemical section of MCF7 cells. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 2 μ g/mL mouse anti-RAB11B Antibody (M04526) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Mouse IgG (BA1126) was used as secondary antibody at 1:100 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.

Anti-RAB11B Antibody Picoband™ (monoclonal, 6C5) - Background

Ras-related protein Rab-11B is a protein that in humans is encoded by the RAB11B gene. It is mapped to 19p13.2. The Ras superfamily of small GTP-binding proteins, which includes the Ras, Ral, Rho, Rap, and Rab families, is involved in controlling a diverse set of essential cellular functions. The Rab family, including RAB11B, appears to play a critical role in regulating exocytotic and endocytotic pathways.