abcam

ab313575

Anti-RAB27A + RAB27B antibody [EPR27120-68]

Recombinant | RabMAb | 20ul selling size

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	EPR27120-68
Purification technique	Affinity purification Protein A
Concentration	0.504 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Human
Dilution info	1/5000
Notes	Perform heat-mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Not recommended

Species	Mouse, Rat, Transfected cell lysate - Human, Transfected cell line - Human
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	1/1000
Notes	-

Species	Mouse
Dilution info	1/1000
Notes	-

Species	Rat
Dilution info	1/1000
Notes	-

Species	Transfected cell lysate - Human
Dilution info	1/1000
Notes	-

Not recommended

Species	Transfected cell line - Human
Dilution info	-
Notes	-

IP

Tested

Species	Human

Dilution info	1/30
Notes	-

Expected

Species	Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Not recommended

Species	Mouse, Transfected cell lysate - Human, Transfected cell line - Human
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/100 - 1/500
Notes	-

Species	Transfected cell line - Human
Dilution info	1/100
Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Not recommended

Species	Transfected cell lysate - Human

Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/500
Notes	-

Expected

Species	Mouse, Rat
Dilution info	Use at an assay dependent concentration.
Notes	-

Not recommended

Species	Transfected cell lysate - Human, Transfected cell line - Human
Dilution info	-
Notes	-

Target data

See full target information RAB27A

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 sets of downstream effectors directly responsible for vesicle formation, movement, tethering and fusion (PubMed:30771381). RAB27A regulates homeostasis of late endocytic pathway, including endosomal positioning, maturation and secretion (PubMed:30771381). Plays a role in cytotoxic granule exocytosis in lymphocytes. Required for both granule maturation and granule docking and priming at the immunologic synapse (PubMed:18812475).

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	-20°C
Aliquoting information	Upon delivery aliquot
Storage information	Avoid freeze / thaw cycle

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	RAB27A and RAB27B are members of the Rab protein family which are small GTPases involved in membrane trafficking. These proteins also known as GTP-binding proteins are critical for vesicle transport and membrane fusion processes. RAB27A has a molecular mass of approximately 25 kDa and RAB27B has a molecular mass around 24.5 kDa. Both proteins express widely in various cell types yet show higher expression in secretory cells particularly in the brain pancreas and melanocytes. RAB27A and RAB27B localize to the membrane of secretory granules aiding in their mobilization.
Biological function summary	The proteins RAB27A and RAB27B facilitate the movement of secretory vesicles toward the cell membrane for exocytosis. RAB27A is a component of protein complexes known as exocyst complexes which are involved in tethering vesicles to the plasma membrane. The proteins recruit effector molecules such as Myosin Va and Slp2-a to the vesicle surface enhancing their docking and fusion with the cell

	membrane. This process is essential for regulated secretion of hormones neurotransmitters and other important molecules.
Pathways	RAB27A and RAB27B play important roles in the vesicular transport systems that govern secretion pathways including the exocytosis and endocytosis pathways. During exocytosis RAB27A works closely with SNARE proteins to stabilize vesicle fusion. It also interacts with other Rab proteins like RAB3D contributing to efficient vesicle trafficking. In addition RAB27B has a role in synaptic vesicle pathways which are key to neurotransmitter release in neural communication.
Associated diseases and disorders	Mutations in RAB27A lead to Griscelli syndrome type 2 characterized by immune system dysfunction and pigment dilution. This syndrome occurs due to defective vesicle transport in melanocytes and immune cells. RAB27A is linked with LYST a protein involved in lysosomal trafficking which causes Chediak-Higashi syndrome when mutated. Moreover altered RAB27B expression associates with colorectal cancer progression emphasizing its importance in cellular secretion dysfunctions.

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

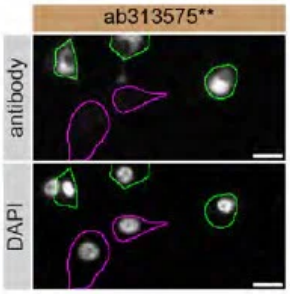
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

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:
Terms & Conditions.

15 product images

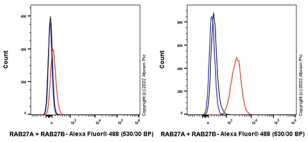


Immunocytochemistry/ Immunofluorescence - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575)

RAB27A + RAB27B Immunocytochemistry/ Immunofluorescence staining of U-87 MG cells using rabbit Anti-RAB27A + RAB27B antibody

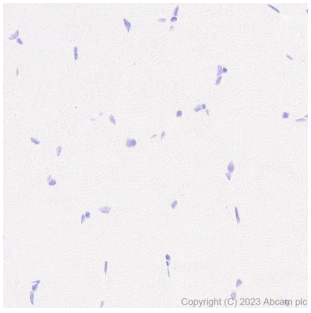
ab313575 was shown to react with Rab27A in wild-type U-87 MG cells in immunocytochemistry with loss of signal observed in a Rab27A siRNA knockdown cell line. Wild-type and siRNA knockdown cells were mixed and pelleted at a 1:1 ratio on coverslips. The cells were fixed with 4% paraformaldehyde (15 min) then permeabilized with 0.1% Triton X-100 (10min) and then blocked with 1x PBS, 0.01% Triton X-100, 5% BSA, 5% NGS. The cells were then incubated with ab313575 at 1/500 dilution overnight at 4°C followed by a further incubation at room temperature for 1h with a goat anti-rabbit secondary antibody to (Alexa Fluor® 555) at 0.5 µg/ml. Acquisition of the green (wild-type), red (antibody staining) and far-red (siRNA knockdown) channels was performed. Representative grayscale images of the red channel are shown. Wild-type and siRNA knockdown cells are outlined with yellow and magenta dashed line, respectively. Schematic representation of the mosaic strategy used is shown on the bottom-right panel. Image was acquired with a Zeiss(LSM-880).

This data was provided by YCharOS Inc., an open science company with the mission of characterizing commercially available antibody reagents for all human proteins. Abcam and YCharOS are working together to help address the reproducibility crisis by enabling the life science community to better evaluate commercially available antibodies.



Flow Cytometry (Intracellular) - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575)

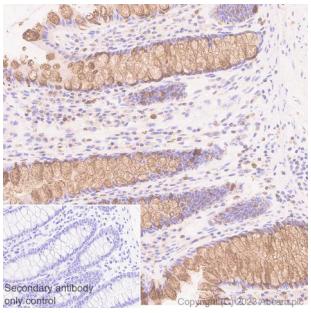
Flow cytometric analysis of 4% paraformaldehyde fixed 90% methanol permeabilized Raji (human Burkitt's lymphoma B lymphocyte, Left) / SK-MEL-28 (human malignant melanoma cell, Right) cells labelling RAB27A + RAB27B with ab313575 at 1/500 dilution (0.1 ug) (Red) compared with a Rabbit monoclonal IgG (ab172730) (Black) isotype control and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue). A Goat Anti-Rabbit IgG (Alexa Fluor® 488, ab150081) at 1/5000 dilution was used as the secondary antibody. Negative control: Raji (PMID: 15548590).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575)

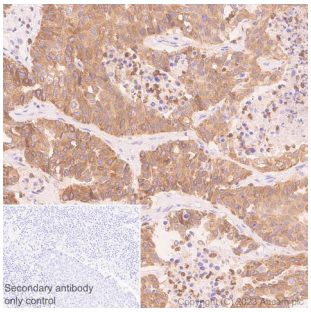
RAB27A + RAB27B Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining of Human skeletal muscle using rabbit Anti-RAB27A + RAB27B antibody

Immunohistochemical analysis of paraffin-embedded human skeletal muscle tissue labeling RAB27A + RAB27B with ab313575 at 1/5000 (0.101 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Negative control: no staining on human skeletal muscle. The section was incubated with ab313575 for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.



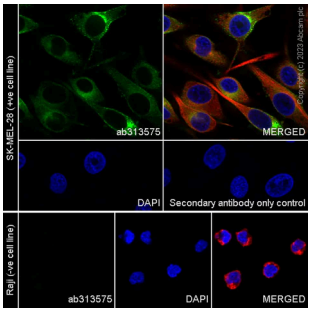
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575)

Immunohistochemical analysis of paraffin-embedded human colon tissue labeling RAB27A + RAB27B with ab313575 at 1/5000 (0.101 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection) was used. Positive staining on human colon (PMID: 26070933). The section was incubated with ab313575 for 30 mins at room temperature. The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection) was used. Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.



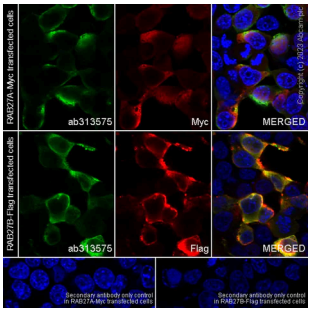
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575)

Immunohistochemical analysis of paraffin-embedded human lung adenocarcinoma tissue labeling RAB27A + RAB27B with ab313575 at 1/5000 (0.101 ug/ml) followed by a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Positive staining on human lung adenocarcinoma (PMID: 30480360). The section was incubated with ab313575 for 30 mins at room temperature.The immunostaining was performed on a Leica Biosystems BOND® RX instrument. Counterstained with Hematoxylin. Secondary antibody only control: Secondary antibody is a ready to use LeicaDS9800 (Bond™ Polymer Refine Detection). Heat mediated antigen retrieval was performed with Tris-EDTA buffer (pH 9.0, Epitope Retrieval Solution2) for 20 mins.



Immunocytochemistry/ Immunofluorescence - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575)

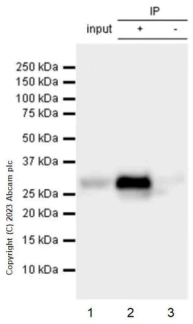
Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized SK-MEL-28 (human malignant melanoma cell) cells labelling RAB27A + RAB27B with ab313575 at 1/100 (5.04 ug/ml) dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 2 ug/mL dilution (Green). Confocal image showing cytoplasmic staining in SK-MEL-28. Negative control: Raji (PMID: 15548590). Image was taken with a confocal microscope(Leica-Microsystems, TCS SP8). ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 2.5ug/ml dilution (Red). The Nuclear counterstain was DAPI (Blue). Secondary antibody only control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 2 ug/mL dilution.



Immunocytochemistry/ Immunofluorescence - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575)

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% TritonX-100 permeabilized 293T (human embryonic kidney epithelial cell) cells labelling RAB27A + RAB27B with ab313575 at 1/100 (5.04 ug/ml) dilution, followed by ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed antibody at 1/1000 2 ug/mL dilution (Green). Confocal image showing both positive staining in 293T cells transfected with human RAB27A and RAB27B expression vectors containing a Myc tag and a Flag tag respectively. Counter stain:ab223894 Anti-Myc tag mouse monoclonal antibody (Alexa Fluor® 594), 1:100 5ug/ml. Anti-FLAG mouse monoclonal antibody, 1:500 2ug/ml. Image was taken with a confocal microscope(Leica-Microsystems, TCS SP8). The Nuclear

counterstain was DAPI (Blue). Secondary antibody only control: Secondary antibody is ab150081 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed at 1/1000 2 ug/mL dilution.



Immunoprecipitation - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575)

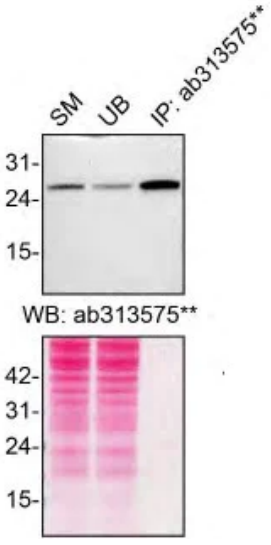
RAB27A + RAB27B was immunoprecipitated from 0.35 mg K562 (human chronic myelogenous leukemia lymphoblast) whole cell lysate with ab313575 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab313575 at 1/1000 dilution. VeriBlot for IP secondary antibody(HRP)(ab131366) was used at 1/5000 dilution. Lane 1: K562 (human chronic myelogenous leukemia lymphoblast) whole cell lysate Lane 2: K562 (human chronic myelogenous leukemia lymphoblast) whole cell lysate Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab313575 in K562 whole cell lysate Blocking and dilution buffer and concentration: 5% NFDM/TBST. Exposure time: 10 seconds
All lanes:
Immunoprecipitation - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575) at 1/30 dilution

All lanes:
K562 (human chronic myelogenous leukemia lymphoblast) whole cell lysate

Secondary

All lanes:
Immunoprecipitation - VeriBlot for IP Detection Reagent (HRP) (ab131366) at 1/5000 dilution

Exposure time: 10s



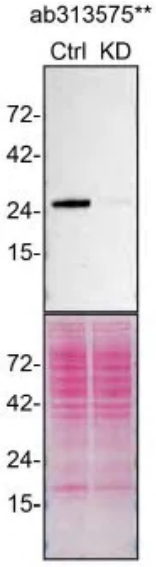
Immunoprecipitation - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575)

Immunoprecipitation of Rab27A in U-87 MG cells. Lysates were prepared and immunoprecipitation was performed using 2 µg of ab313575 pre-coupled to Protein A beads. Samples were then washed and processed for western blot.

This data was provided by YCharOS Inc., an open science company with the mission of characterizing commercially available antibody reagents for all human proteins. Abcam and YCharOS are working together to help address the reproducibility crisis by enabling the life science community to better evaluate commercially available antibodies.
All lanes:
Immunoprecipitation - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575) at 2 µg

Lanes 1 and 3:
U-87 MG cells

Lane 2:
Unbound fraction: Rabbit monoclonal IgG instead of ab313575 in U-87 MG cells



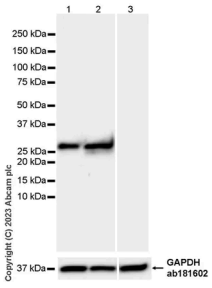
Western blot - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575)

ab313575 was shown to react with Rab27A in wild-type U-87 MG cells in Western blot with loss of signal observed in a Rab27A siRNA knockdown cell line. Cell lysates from wild-type U-87 MG transfected with either scrambled siRNA or Rab27A siRNA were subjected to SDS-PAGE. Membranes were blocked in 5% milk in TBST for 1 hr before incubation with ab313575 overnight at 4 °C at a 1/1000 dilution. Blots were incubated with secondary antibodies at 0.2 µg/mL before imaging.

This data was provided by YCharOS Inc., an open science company with the mission of characterizing commercially available antibody reagents for all human proteins. Abcam and YCharOS are working together to help address the reproducibility crisis by enabling the life science community to better evaluate commercially available antibodies.
All lanes:
Western blot - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575) at 1/1000 dilution

Lane 1:
Wild-type U-87 MG transfected with scrambled siRNA control lysate at 20 µg

Lane 2:
U-87 MG transfected with siRNA specifically targeting Rab27A cell lysate at 20 µg



Western blot - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575)

Blocking and diluting buffer and concentration: 5% NFDM/TBST Negative control: Raji (PMID:15548590) In Western blot, anti-GAPDH antibody (ab181602) loading control staining at 1/200000 dilution. Exposure time: 15 seconds
All lanes:
Western blot - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575) at 1/1000 dilution

Lane 1:
K562 (human chronic myelogenous leukemia lymphoblast) whole cell lysate at 20 µg

Lane 2:
SK-MEL-2 (human skin malignant melanoma cell) whole cell lysate at 20 µg

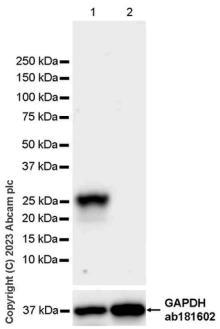
Lane 3:
Raji (human Burkitts lymphoma B lymphocyte) whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Observed band size: 27 kDa

Exposure time: 15s



Western blot - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575)

Blocking and diluting buffer and concentration: 5% NFDM/TBST Negative control: skeletal muscle
In Western blot, anti-GAPDH antibody (ab181602) loading control staining at 1/200000 dilution.
Exposure time: 26 seconds
All lanes:
Western blot - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575) at 1/1000 dilution

Lane 1:
Human stomach tissue lysate at 20 µg

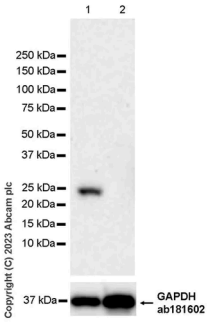
Lane 2:
Human skeletal muscle tissue lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Observed band size: 27 kDa

Exposure time: 26s



Western blot - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575)

Blocking and diluting buffer and concentration: 5% NFDM/TBST Negative control: skeletal muscle.
In Western blot, anti-GAPDH antibody (ab181602) loading control staining at 1/200000 dilution.
Exposure time: 180 seconds
All lanes:
Western blot - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575) at 1/1000 dilution

Lane 1:
Mouse stomach tissue lysate at 20 µg

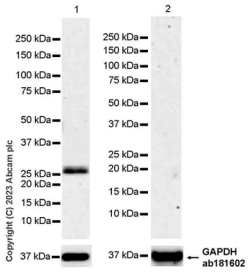
Lane 2:
Mouse skeletal muscle tissue lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Observed band size: 27 kDa

Exposure time: 180s



Western blot - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575)

Blocking and diluting buffer and concentration: 5% NFDM/TBST Negative control: skeletal muscle.
In Western blot, anti-GAPDH antibody (ab181602) loading control staining at 1/200000 dilution.
Exposure time: 180 seconds
All lanes:
Western blot - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575) at 1/1000 dilution

Lane 1:
Rat stomach tissue lysate at 20 µg

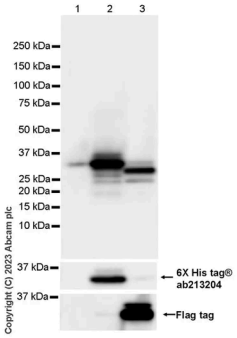
Lane 2:
Rat skeletal muscle tissue lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Observed band size: 27 kDa

Exposure time: 180s



Western blot - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575)

Blocking and diluting buffer and concentration: 5% NFDM/TBST This antibody recognizes both RAB27A and RAB27B. In Western blot, anti-His antibody (ab213204) staining at 1/5000 dilution.
Exposure time: 3 seconds
All lanes:
Western blot - Anti-RAB27A + RAB27B antibody [EPR27120-68] (ab313575) at 1/1000 dilution

Lane 1:
293T (human embryonic kidney epithelial cell) cells transfected with an empty vector containi a His-tag whole cell lysate at 20 µg

Lane 2:
293T cells transfected with a human RAB27A expression vector containi a His-tag whole cell lysate at 20 µg

Lane 3:
293T cells transfected with a human RAB27B expression vector containi a Flag-tag whole cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Observed band size: 27 kDa

Exposure time: 3s

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.