abcam

Product datasheet

Anti-ARF1 antibody ab183576

KO VALIDATED

1 References 9 Images

Overview

Product name	Anti-ARF1 antibody
Description	Rabbit polyclonal to ARF1
Host species	Rabbit
Tested applications	**Suitable for:** ICC/IF, IP, WB, IHC-P
Species reactivity	**Reacts with:** Mouse, Rat, Dog, Human **Predicted to work with:** Cow
Immunogen	Synthetic peptide corresponding to Human ARF1 aa 150 to the C-terminus (C terminal). Database link: **P84077** Run BLAST with Run BLAST with
Positive control	WB: HeLa, U2 OS, 3T3, NRK, MDA-MB-231, PANC-1 and MDCK cell lysates; HeLa cells. IHC: mouse colon, human breast carcinoma and human colon carcinoma. ICC/IF: MDA-MB-231 cells IP: HeLa
General notes	The Life Science industry has been in the grips of a reproducibility crisis for a number of years. Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets your needs before purchasing. If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be found below, along with publications, customer reviews and Q&As

Properties

Form	Liquid
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	Preservative: 0.05% Sodium azide Constituents: 0.1% BSA, 30% Glycerol, 69% PBS
Purity	Immunogen affinity purified
Clonality	Polyclonal

Isotype IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab183576 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
ICC/IF		Use a concentration of 5 µg/ml.
IP		Use at 2 µg/mg of lysate.
WB		1/3000. Predicted molecular weight: 21 kDa.
IHC-P		1/10 - 1/100. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

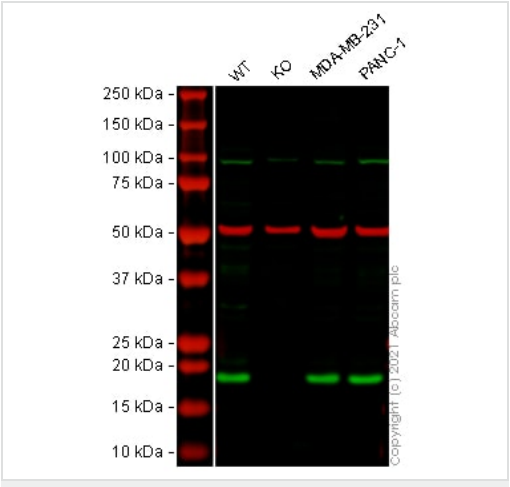
Target

Function GTP-binding protein that functions as an allosteric activator of the cholera toxin catalytic subunit, an ADP-ribosyltransferase. Involved in protein trafficking among different compartments. Modulates vesicle budding and uncoating within the Golgi complex. Deactivation induces the redistribution of the entire Golgi complex to the endoplasmic reticulum, suggesting a crucial role in protein trafficking. In its GTP-bound form, its triggers the association with coat proteins with the Golgi membrane. The hydrolysis of ARF1-bound GTP, which is mediated by ARFGAPs proteins, is required for dissociation of coat proteins from Golgi membranes and vesicles.

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

Cellular localization Golgi apparatus. Cytoplasm > perinuclear region.

Images



Western blot - Anti-ARF1 antibody (ab183576)

All lanes : Anti-ARF1 antibody (ab183576) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : ARF1 knockout HeLa cell lysate
Lane 3 : MDA-MB-231 cell lysate
Lane 4 : PANC-1 cell lysate

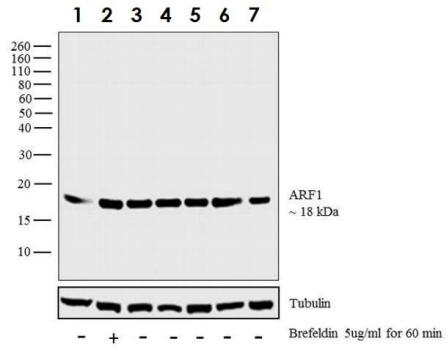
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 21 kDa
Observed band size: 18 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab183576 observed at 18 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55 kDa.

ab183576 was shown to react with ARF1 in wild-type HeLa cells in Western blot with loss of signal observed in ARF1 knockout cell line **ab264939** (ARF1 knockout cell lysate **ab257353**). Wild-type HeLa and ARF1 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with ab183576 and **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.



Western blot - Anti-ARF1 antibody (ab183576)

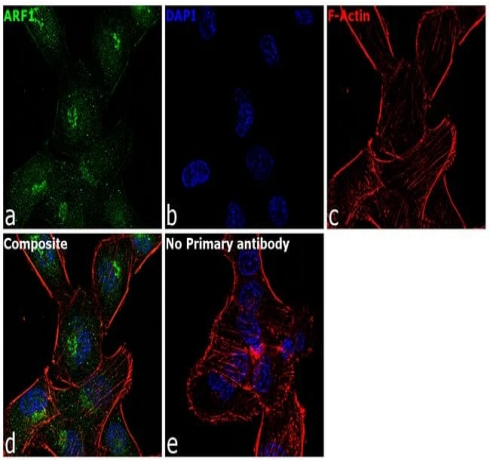
All lanes : Anti-ARF1 antibody (ab183576) at 1/3000 dilution

Lane 1 : MDA-MB-231 whole cell extract (human breast adenocarcinoma cell line)
Lane 2 : MDA-MB-231 whole cell extract (human breast adenocarcinoma cell line) treated with 5ug/ml Brefeldin for 60 minutes
Lane 3 : HeLa whole cell extract
Lane 4 : U2 OS whole cell extract (human bone osteosarcoma epithelial cell line)
Lane 5 : DU 145 whole cell extract (Human prostate carcinoma cell line)
Lane 6 : PC-3 whole cell extract (human prostate cancer cell line)
Lane 7 : SK-OV-3 whole cell extract (Human ovarian cancer cell line)

Lysates/proteins at 30 µg per lane.

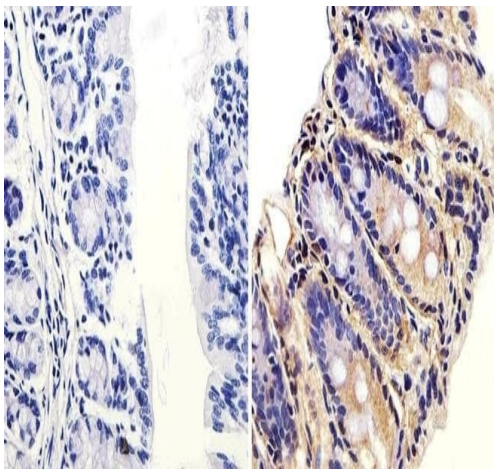
Secondary
All lanes : Goat anti-Rabbit IgG (H+L) Superclonal™ Secondary Antibody, HRP conjugate at 1/4000 dilution

Predicted band size: 21 kDa



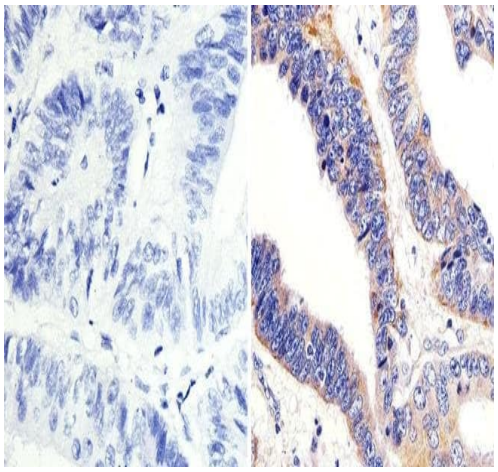
Immunocytochemistry/ Immunofluorescence - Anti-ARF1 antibody (ab183576)

Immunocytochemistry analysis of 4% paraformaldehyde-fixed 0.1% Triton™ X-100 permeabilized MDA-MB-231 cells staining ARF1 with ab183576 at 5µg/ml, and Goat anti-Rabbit IgG (H+L) Superclonal™ Secondary Antibody, Alexa Fluor® 488 conjugate at 1/2000 dilution (green). Nuclear counterstain was ProLong™ Diamond Antifade Mountant with DAPI (blue), and F-actin was stained with Rhodamine Phalloidin (red). Negative control used no primary antibody.



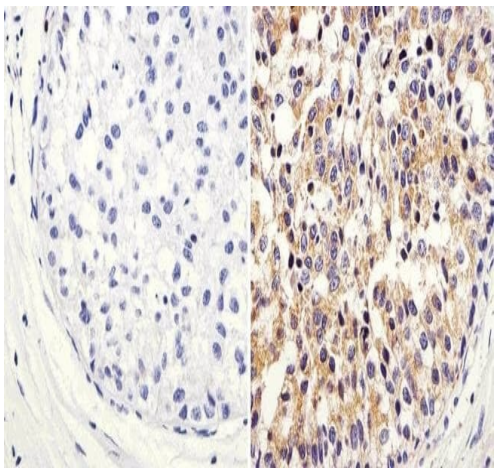
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ARF1 antibody (ab183576)

Immunohistochemistry analysis of paraffin-embedded human mouse colon tissue staining ARF1 with ab183576 at 1/100 dilution (right), and negative control (left) with no primary antibody. Atigen retrieval method using sodium citrate pH6, and detection was with an HRP-conjugated secondary antibody.



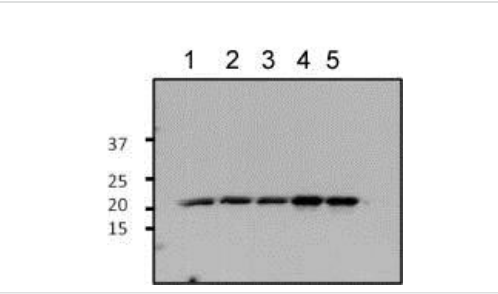
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ARF1 antibody (ab183576)

Immunohistochemistry analysis of paraffin-embedded human colon carcinoma tissue staining ARF1 with ab183576 at 1/100 dilution (right), and negative control (left) with no primary antibody. Atigen retrieval method using sodium citrate pH6, and detection was with an HRP-conjugated secondary antibody.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ARF1 antibody (ab183576)

Immunohistochemistry analysis of paraffin-embedded human breast carcinoma tissue staining ARF1 with ab183576 at 1/100 dilution (right), and negative control (left) with no primary antibody. Atigen retrieval method using sodium citrate pH6, and detection was with an HRP-conjugated secondary antibody.



Western blot - Anti-ARF1 antibody (ab183576)

All lanes : Anti-ARF1 antibody (ab183576) at 1/1000 dilution

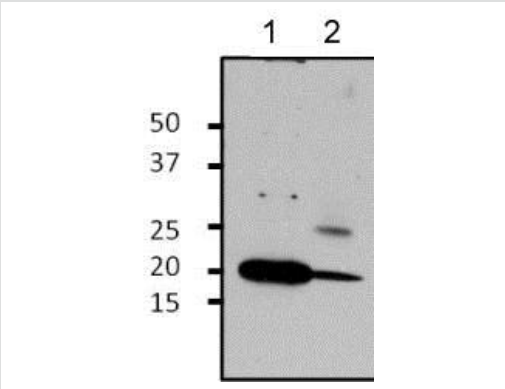
Lane 1 : HeLa cell lysate
Lane 2 : U2 OS cell lysate
Lane 3 : 3T3 cell lysate
Lane 4 : NRK cell lysate
Lane 5 : MDCK cell lysate

Lysates/proteins at 25 µg per lane.

Secondary
All lanes : goat anti-rabbit IgG-HRP at 1/1500 dilution

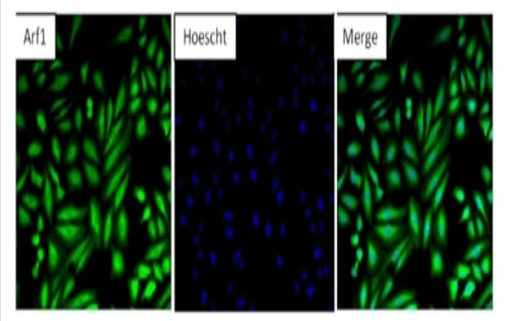
Developed using the ECL technique.

Predicted band size: 21 kDa



Immunoprecipitation - Anti-ARF1 antibody (ab183576)

Detection of ARF1 in Immunoprecipitates of HeLa cell lysate. Antigen-antibody complexes were formed by incubating 500µg of HeLa whole cell lysate with 2µg ab183576 (lane 2) compared with HeLa cell lysate as a positive control (lane 1). For detection, ab183576 was used at 1/1000 dilution.



Immunocytochemistry/ Immunofluorescence - Anti-ARF1 antibody (ab183576)

Immunofluorescent analysis of HeLa cells (formalin-fixed, 0.1% Triton X-100 permeabilized) labeling ARF1 with ab183576 at 1/100 dilution followed with DyLight 488 goat anti-rabbit IgG secondary antibody at 1/400 dilution. Nuclei (blue) were stained with Hoechst 33342 dye.

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