abcam

Product datasheet

Anti-Flotillin 1 antibody [EPR6041] - BSA and Azide free ab232405

KO VALIDATED | Recombinant | RabMAb

6 Images

Overview

Product name	Anti-Flotillin 1 antibody [EPR6041] - BSA and Azide free
Description	Rabbit monoclonal [EPR6041] to Flotillin 1 - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** WB, ICC/IF, IP, Flow Cyt, IHC-P
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide within Human Flotillin 1 aa 400-500 (C terminal). The exact sequence is proprietary. Database link: O75955
Positive control	HeLa membrane extract lysate (ab29547) can be used as a positive control in WB. Wild-type HAP1 whole cell lysate
General notes	Ab232405 is the carrier-free version of ab133497. This format is designed for use in antibody labeling, including fluorochromes, metal isotopes, oligonucleotides, enzymes. Our carrier-free formats are supplied in a buffer free of BSA, sodium azide and glycerol for higher conjugation efficiency. Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with <1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold. ab232405 is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm. *Maxpar® is a trademark of Fluidigm Canada Inc.* 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.

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	Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR6041
Isotype	IgG

Applications

Our Abpromise guarantee covers the use of **ab232405** in the following tested applications.

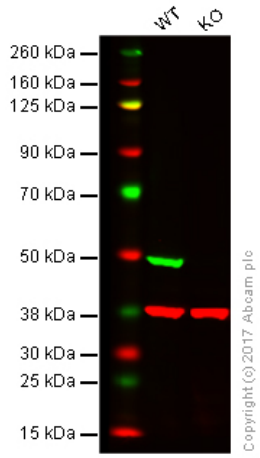
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB		Use at an assay dependent concentration. Predicted molecular weight: 47 kDa.
ICC/IF		Use at an assay dependent concentration.
IP		Use at an assay dependent concentration.
Flow Cyt		Use at an assay dependent concentration.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

Target

Function	May act as a scaffolding protein within caveolar membranes, functionally participating in formation of caveolae or caveolae-like vesicles.
Sequence similarities	Belongs to the band 7/mec-2 family. Flotillin subfamily.
Cellular localization	Cell membrane. Membrane > caveola. Melanosome. Endosome. Membrane-associated protein of caveolae. Identified by mass spectrometry in melanosome fractions from stage I to stage IV.

Images



Western blot - Anti-Flotillin 1 antibody [EPR6041] - BSA and Azide free (ab232405)

Lane 1: Wild-type HAP1 whole cell lysate (20 µg)
Lane 2: Flotillin 1 knockout HAP1 whole cell lysate (20 µg)

Lanes 1 - 2: Merged signal (red and green). Green - ab133497 observed at 47 kDa. Red - loading control, ab9484, observed at 37 kDa.

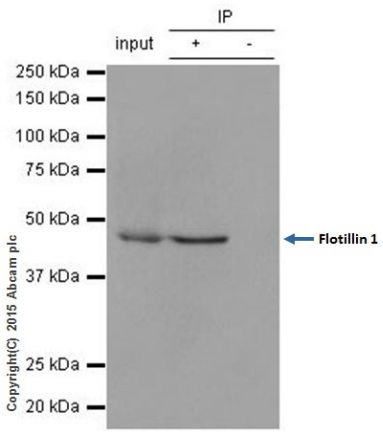
ab133497 was shown to specifically react with Flotillin 1 in wild-type HAP1 cells as signal was lost in Flotillin 1 knockout cells. Wild-type and Flotillin 1 knockout samples were subjected to SDS-PAGE. ab133497 and ab9484 (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/10,000 dilution and 1/20,000 dilution respectively. Blots were developed 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 1/20,000 dilution for 1 hour at room temperature before imaging.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab133497).

Flow Cytometry - Anti-Flotillin 1 antibody [EPR6041] - BSA and Azide free (ab232405)

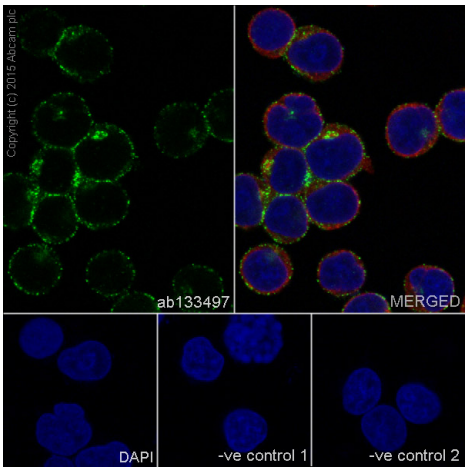
Flow Cytometry analysis of Jurkat (human acute T cell leukemia) cells labeling Flotillin 1 with purified ab133497 at 1/50 dilution (10ug/mL) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit IgG (Alexa Fluor®488)(1/2000 dilution) was used as the secondary antibody. Rabbit monoclonal IgG (Black) was used as the isotype control, cells without incubation with primary antibody and secondary antibody (Blue) was used as the unlabeled control.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab133497).



Immunoprecipitation - Anti-Flotillin 1 antibody [EPR6041] - BSA and Azide free (ab232405)

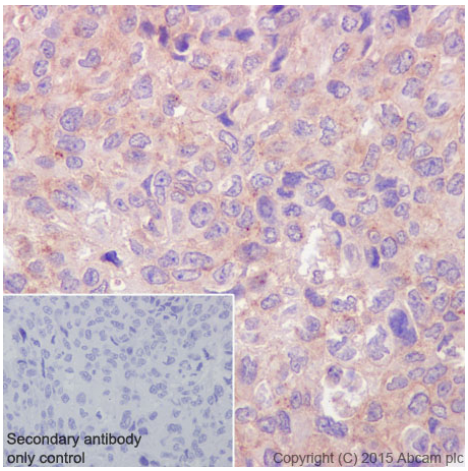
ab133497 (purified) at 1/60 immunoprecipitating Flotillin 1 in 10 µg K562 (Lanes 1 and 2, observed at 48 kDa). Lane 3 - PBS. For western blotting, a HRP-conjugated anti-rabbit IgG, specific to the non-reduced form of IgG was used as the secondary antibody (1/1500). Blocking buffer and concentration: 5% NFDM/TBST Dilution buffer and concentration: 5% NFDM/TBST
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab133497).



Immunocytochemistry/ Immunofluorescence - Anti-Flotillin 1 antibody [EPR6041] - BSA and Azide free (ab232405)

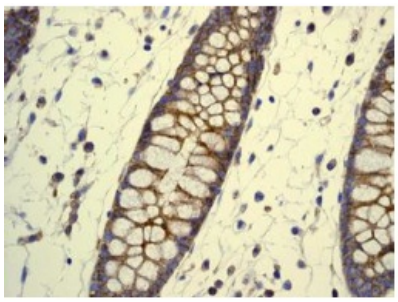
Immunofluorescence staining of Jurkat cells with purified ab133497 at a working dilution of 1/500, counter-stained with DAPI. The secondary antibody was Alexa Fluor® 488 goat anti-rabbit (ab150077), used at a dilution of 1/1000. ab7291, a mouse anti-tubulin antibody (1/1000), was used to stain tubulin along with ab150120 (Alexa Fluor® 594 goat anti-mouse, 1/1000), shown in the top right hand panel. The cells were fixed in 4% PFA and permeabilized using 0.1% Triton X 100. The negative controls are shown in bottom middle and right hand panels - for negative control 1, purified ab133497 was used at a dilution of 1/500 followed by an Alexa Fluor® 594 goat anti-mouse antibody (ab150120) at a dilution of 1/500. For negative control 2, ab7291 (mouse anti-tubulin) was used at a dilution of 1/500 followed by an Alexa Fluor® 488 goat anti-rabbit antibody (ab150077) at a dilution of 1/400.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab133497).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Flotillin 1 antibody [EPR6041] - BSA and Azide free (ab232405)

Immunohistochemical staining of paraffin embedded human cervical carcinoma with purified ab133497 at a working dilution of 1/100. The secondary antibody used is HRP goat anti-rabbit IgG H&L (ab97051) at 1/500. The sample is counter-stained with hematoxylin. Antigen retrieval was perfomed using Tris-EDTA buffer, pH 9.0. PBS was used instead of the primary antibody as the negative control, and is shown in the inset.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab133497).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Flotillin 1 antibody [EPR6041] - BSA and Azide free (ab232405)

Immunohistochemical analysis of paraffin embedded Human colon tissue labelling Flotillin 1 using unpurified ab133497 at a dilution of 1/100.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab133497).

Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

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