abcam

Product datasheet

Anti-Syntenin antibody [EPR8102] - BSA and Azide free ab236071

KO VALIDATED Recombinant RabMAb

3 References 11 Images

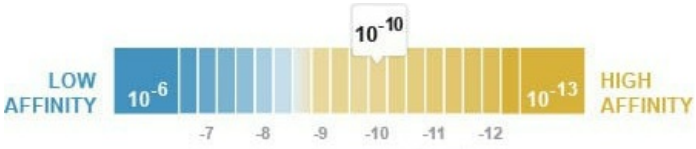
Overview

Product name	Anti-Syntenin antibody [EPR8102] - BSA and Azide free
Description	Rabbit monoclonal [EPR8102] to Syntenin - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** Flow Cyt (Intra), IHC-P, IP, ICC/IF, WB
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: HAP1, A549 and HeLa cell lysates.
General notes	ab236071 is the carrier-free version of **ab133267**. Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications. 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. This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. 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**. Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with

these species. Please contact us for more information.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Dissociation constant (K_D)	$K_D = 1.24 \times 10^{-10}$ M



Learn more about K_D

Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR8102
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab236071 in the following tested applications.

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

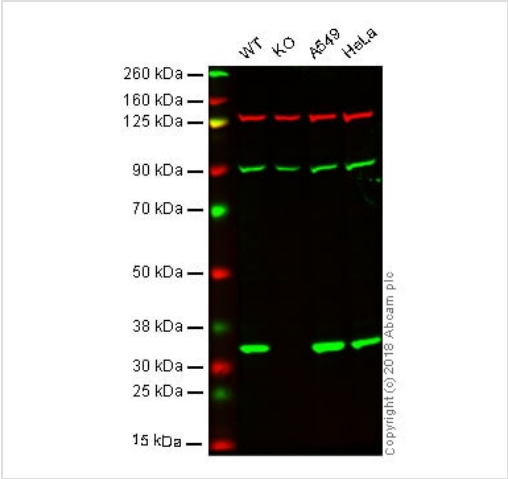
Application	Abreviews	Notes
Flow Cyt (Intra)		Use at an assay dependent concentration. **ab172730** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. See **IHC antigen retrieval protocols**.
IP		Use at an assay dependent concentration.
ICC/IF		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 32 kDa (predicted molecular weight: 32 kDa).

Target

Function Seems to function as an adapter protein. In adherens junctions may function to couple syndecans

	to cytoskeletal proteins or signaling components. Seems to couple transcription factor SOX4 to the IL-5 receptor (IL5RA). May also play a role in vesicular trafficking. Seems to be required for the targeting of TGFA to the cell surface in the early secretory pathway.
Tissue specificity	Widely expressed. Expressed in fetal kidney, liver, lung and brain. In adult highest expression in heart and placenta.
Sequence similarities	Contains 2 PDZ (DHR) domains.
Post-translational modifications	Phosphorylated on tyrosine residues.
Cellular localization	Cell junction > focal adhesion. Cell junction > adherens junction. Cell membrane. Endoplasmic reticulum membrane. Nucleus. Melanosome. Cytoplasm > cytosol. Cytoplasm > cytoskeleton. Mainly membrane-associated. Localized to adherens junctions, focal adhesions and endoplasmic reticulum. Colocalized with actin stress fibers. Also found in the nucleus. Identified by mass spectrometry in melanosome fractions from stage I to stage IV.

Images



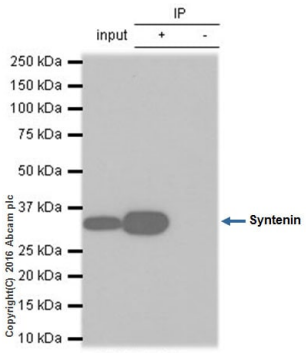
Western blot - Anti-Syntenin antibody [EPR8102] - BSA and Azide free (ab236071)

Lane 1: Wild-type HAP1 whole cell lysate (20 µg)
Lane 2: Syntenin knockout HAP1 whole cell lysate (20 µg)
Lane 3: A549 whole cell lysate (20 µg)
Lane 4: HeLa whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - **ab133267** observed at 32 kDa. Red - loading control, **ab130007**, observed at 130 kDa.

ab133267 was shown to recognize Syntenin in wild-type HAP1 cells as signal was lost at the expected MW in Syntenin knockout cells. Additional cross-reactive bands were observed in the wild-type and knockout cells. Wild-type and Syntenin knockout samples were subjected to SDS-PAGE. **ab133267** and **ab130007** (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 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 1/20000 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 (**ab133267**).



Immunoprecipitation - Anti-Syntenin antibody [EPR8102] - BSA and Azide free (ab236071)

ab133267 (purified) at 1/40 immunoprecipitating Syntenin in HeLa whole cell lysate.

Lane 1 (input): HeLa whole cell lysate (10µg)

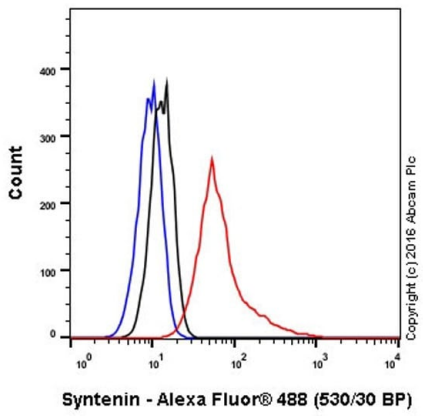
Lane 2 (+): **ab133267** + HeLa whole cell lysate.

Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of **ab133267** in HeLa whole cell lysate.

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.

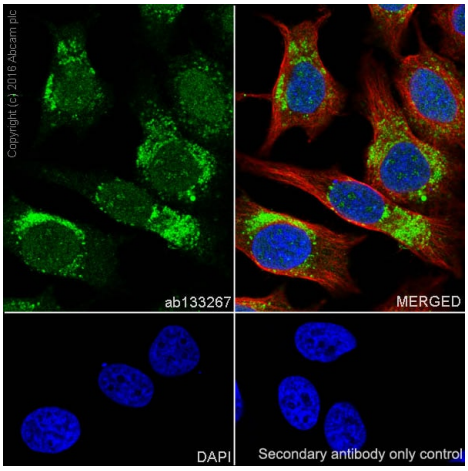
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab133267**).



Flow Cytometry (Intracellular) - Anti-Syntenin antibody [EPR8102] - BSA and Azide free (ab236071)

Intracellular Flow Cytometry analysis of HeLa (Human epithelial cell line from cervix adenocarcinoma) labeling Syntenin with purified **ab133267** at 1/50 (red). Cells were fixed with 4% paraformaldehyde. A goat anti rabbit IgG (Alexa Fluor® 488) 1/2000 was used as the secondary antibody. Black - Isotype control, rabbit monoclonal IgG. Blue - Unlabelled control, cells without incubation with primary and secondary antibodies.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab133267**).

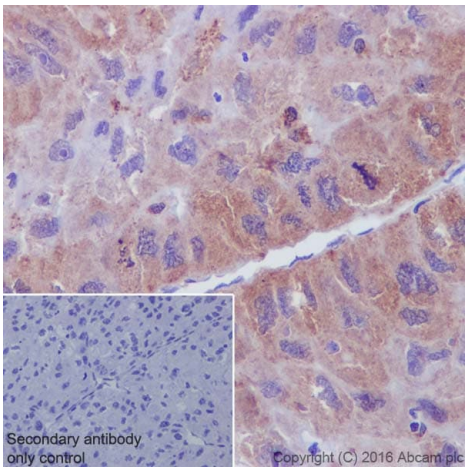


Immunocytochemistry/ Immunofluorescence - Anti-Syntenin antibody [EPR8102] - BSA and Azide free (ab236071)

Immunocytochemistry/Immunofluorescence analysis of HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling Syntenin with purified **ab133267** at 1/200 dilution. Cells were fixed with 4% paraformaldehyde and permeabilized with 0.1% Triton X-100. A goat anti rabbit IgG (Alexa Fluor® 488) (**ab150077**) was used as the secondary antibody at a dilution of 1/1000. DAPI was used as a nuclear counterstain.

Negative control 1: PBS only.

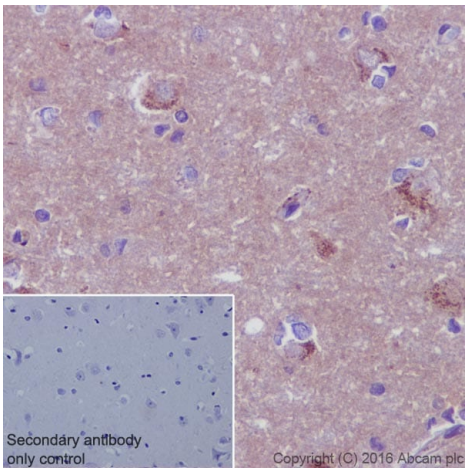
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab133267**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Syntenin antibody [EPR8102] - BSA and Azide free (ab236071)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human hepatocellular carcinoma tissue labeling Syntenin with purified **ab133267** at 1/50 dilution. Heat mediated antigen retrieval was performed using EDTA buffer pH 9. **ab97051**, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin

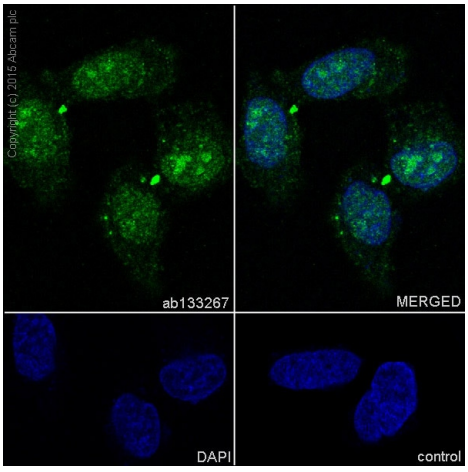
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab133267**).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Syntenin antibody [EPR8102] - BSA and Azide free (ab236071)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human cerebral cortex tissue labeling Syntenin with purified **ab133267** at 1/50 dilution. Heat mediated antigen retrieval was performed using EDTA buffer pH 9. **ab97051**, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab133267**).

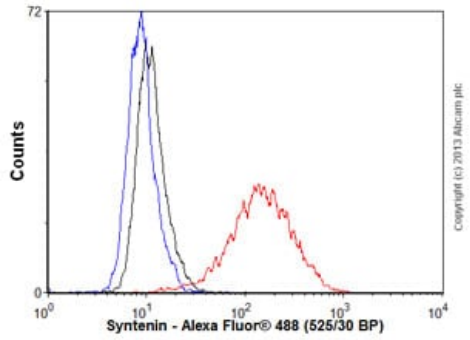


Immunocytochemistry/ Immunofluorescence - Anti-Syntenin antibody [EPR8102] - BSA and Azide free (ab236071)

ab133267 staining Syntenin in HeLa (human cervix adenocarcinoma) cells by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with 4% Paraformaldehyde and permeabilized with 0.1% Triton X-100. Samples were incubated with primary antibody at a dilution of 1/500. A goat anti rabbit IgG (Alexa Fluor® 488) (**ab150077**) was used as the secondary antibody at a dilution of 1/1000. DAPI was used as a nuclear counterstain.

Negative control 1: PBS only.

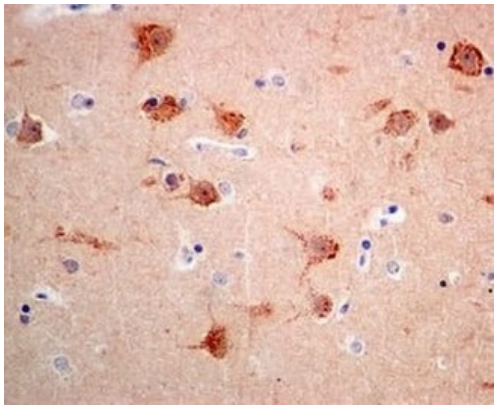
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab133267**).



Flow Cytometry (Intracellular) - Anti-Syntenin antibody [EPR8102] - BSA and Azide free (ab236071)

Overlay histogram showing SHSY-5Y cells stained with unpurified **ab133267** (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (**ab133267**, 1/10000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H&L) (**ab150077**) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (0.1µg/1x10^6 cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter. This antibody gave a positive signal in SHSY-5Y cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab133267**).

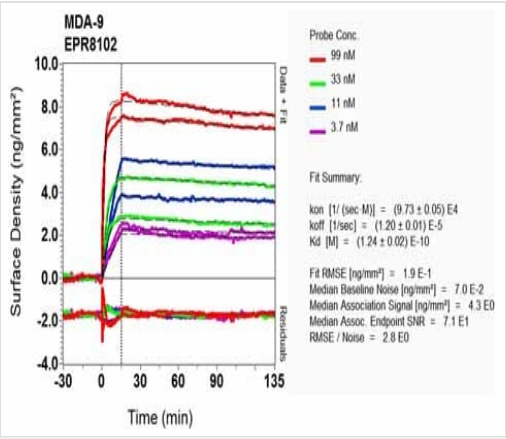


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Syntenin antibody [EPR8102] - BSA and Azide free (ab236071)

Immunohistochemical analysis of Syntenin in paraffin embedded Human brain tissue, using unpurified **ab133267** at a 1/50 dilution.

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab133267**).

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



OI-RD Scanning - Anti-Syntenin antibody [EPR8102] - BSA and Azide free (ab236071)

Anti-Syntenin antibody [EPR8102] - BSA and Azide free (ab236071)

Equilibrium disassociation constant (K_D)

Learn more about K_D

Click here to learn more about K_D

This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (**ab133267**).

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