abcam

Product datasheet

Anti-NISCH antibody ab56849

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Overview

Product name	Anti-NISCH antibody
Description	Mouse monoclonal to NISCH
Host species	Mouse
Tested applications	**Suitable for:** ICC/IF, WB, Flow Cyt
Species reactivity	**Reacts with:** Human
Immunogen	Recombinant fragment: LTGSTPMQVV TCLTRDSYLT HCFLQHLMVV LSSLERTPSP EPVDKDFYSE FGNKTTGKME NYELIHSSRV KFTYPSEEEI GDLTFTVAQK MAEPEKAPAL , corresponding to amino acids 1246-1346 of Human NISCH Run BLAST with ExPASy Run BLAST with NCBI
Positive control	ICC/IF: SKNSH cells.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Upon delivery aliquot and store at -20°C or -80°C. Avoid repeated freeze / thaw cycles.
Storage buffer	Preservative: None PBS, pH 7.2
Purity	Protein G purified
Clonality	Monoclonal
Isotype	IgG1
Light chain type	kappa

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab56849 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 10 µg/ml.

Application	Abreviews	Notes
WB		Use a concentration of 1 - 5 µg/ml. This antibody has only been tested in WB against the recombinant fragment used as immunogen. We have no data on the detection of endogenous protein.
Flow Cyt		Use 0.01-0.1µg for 10^6 cells. ab170190 - Mouse monoclonal IgG1, is suitable for use as an isotype control with this antibody.

Target

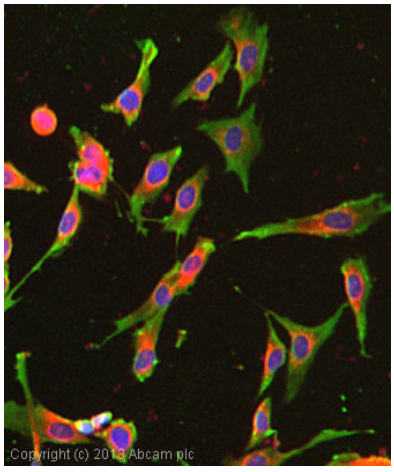
Relevance

Mouse Nischarin (NISCH) has been shown to interact with the alpha-5 subunit of integrin and inhibit cell migration. Its human homologue (imidazoline receptor antisera-selected (IRAS)), contains an NH2-terminal extension and is a larger protein of 1504 amino acids consisting of an NH2-terminal PX domain, 5 putative leucine-rich repeats, a predicted coiled-coil domain, and a long COOH-terminal region. It has the ability to homo-oligomerize via its coiled-coil region. The PX domain of IRAS is essential for association with phosphatidylinositol 3-phosphate-enriched endosomal membranes. NISCH may serve as a functional imidazoline I1-receptor.

Cellular localization

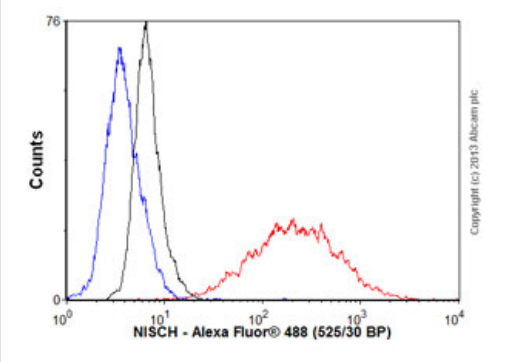
Cell membrane. Cytoplasm. Early endosome. Recycling endosome.

Images



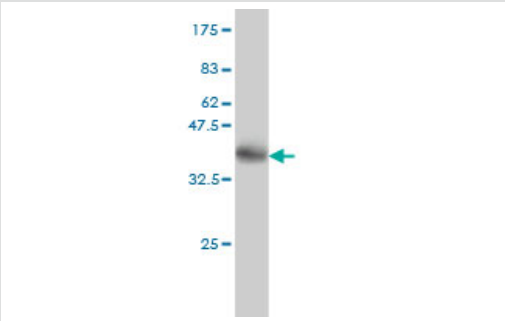
Immunocytochemistry/ Immunofluorescence - Anti-NISCH antibody (ab56849)

ICC/IF image of ab56849 stained SKNSH cells. The cells were 4% formaldehyde fixed (10 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab56849, 10µg/ml) overnight at +4°C. The secondary antibody (green) was ab96879, DyLight® 488 goat anti-mouse IgG (H+L) used at a 1/250 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.



Flow Cytometry - Anti-NISCH antibody (ab56849)

Overlay histogram showing SHSY-5Y cells stained with ab56849 (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 (ab56849, 0.1µg/1x10^6 cells) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-mouse IgG (H&L) (ab150113) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was mouse IgG1 [ICIGG1] (ab91353, 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.



Western blot - Anti-NISCH antibody (ab56849)

Western blot against tagged recombinant protein immunogen using ab56849 NISCH antibody at 1ug/ml. Predicted band size of immunogen is 37 kDa

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