abcam

ab32120

Anti-Nck antibody [Y531]

Recombinant | RabMAb | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 50% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	Y531
Purification technique	Affinity purification Protein A
Concentration	0.118 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

Species	Human
Dilution info	-
Notes	-

Expected

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

Predicted

Species	Rat
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	1/2000 - 1/10000
Notes	-

Expected

Species	Mouse
Dilution info	1/2000 - 1/10000
Notes	-

Predicted

Species	Rat
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human

Dilution info	1/250
Notes	-

Expected

Species	Mouse
Dilution info	1/250
Notes	-

Predicted

Species	Rat
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/1000
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Expected

Species	Mouse
Dilution info	1/1000
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Predicted

Species	Rat
Dilution info	-
Notes	-

Target data

See full target information NCK1

Function	Adapter protein which associates with tyrosine-phosphorylated growth factor receptors, such as KDR and PDGFRB, or their cellular substrates. Maintains low levels of EIF2S1 phosphorylation by promoting its dephosphorylation by PP1. Plays a role in the DNA damage response, not in the detection of the damage by ATM/ATR, but for efficient activation of downstream effectors, such as that of CHEK2. Plays a role in ELK1-dependent transcriptional activation in response to activated Ras signaling. Modulates the activation of EIF2AK2/PKR by dsRNA. May play a role in cell adhesion and migration through interaction with ephrin receptors.

Storage

Shipped at conditions	Conditional Ambient
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	The Nck protein also referred to as Nck1 n c k or Nck is an adaptor protein integral in cell signaling pathways. It consists of a molecular mass around 45 kDa and exists mainly within the cytoplasm. The Nck proteins including Nck1 and its closely related form Nck2 are particularly prominent in cells of the immune and nervous systems. Functionally they facilitate interactions between receptor tyrosine kinases and downstream signaling molecules translating extracellular stimuli into cellular responses.
Biological function summary	The Nck proteins play significant roles in actin cytoskeleton reorganization influencing cell shape movement and adhesion. These proteins often participate as part of larger signaling complexes where they serve as scaffolds bringing together key molecules. Nck proteins directly interact with partners containing SH2 and SH3 domains which are important for various cellular dynamic processes. Their role in cytoskeletal dynamics aligns them with many cellular mechanisms including endocytosis and cell motility.
Pathways	Nck proteins significantly influence the regulation of the actin cytoskeleton pathway and the Rho GTPases pathway. Through these pathways they interact with proteins such as WASP and Rac regulating actin polymerization and cell migration. Nck's ability to form complex signaling networks positions it as a central actor in transducing signals that affect cell shape and movement. It acts collaboratively with multiple signaling pathways to maintain cellular plasticity and response to extracellular signals.
Associated diseases and disorders	Nck proteins have associations with cancer and immunological conditions. Alterations in their signaling functions may contribute to tumor progression or metastasis given their role in cell movement and adhesion. In immune disorders abnormal Nck signaling could disrupt normal immune cell functions potentially leading to autoimmune conditions. For example Nck proteins may interact with oncogenic proteins like BCR-ABL in chronic myeloid leukemia highlighting their relevance in disease contexts.

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

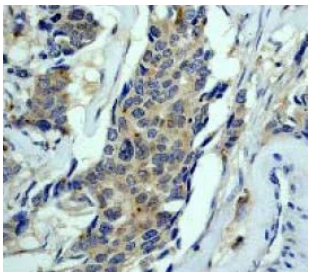
We do not recommend this combination. It is not covered by our product promise.

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.

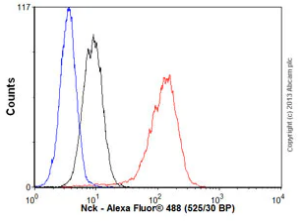
5 product images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Nck antibody [Y531] (ab32120)

Nck Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Nck antibody

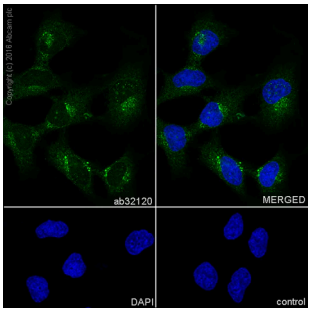
ab32120 (1/100), staining human breast carcinoma by immunohistochemistry, Paraffin embedded tissue.
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-Nck antibody [Y531] (ab32120)

Nck Flow Cytometry (Intracellular) staining using rabbit Anti-Nck antibody

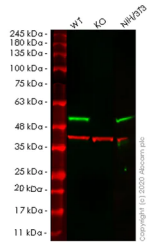
Overlay histogram showing Jurkat cells stained with ab32120 (red line). The cells were fixed with 4% paraformaldehyde (10 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab32120, 1/1000 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 Jurkat cells fixed with 80% methanol (5 min)/permeabilized with 0.1% PBS-Tween for 20 min used under the same conditions.



Immunocytochemistry/ Immunofluorescence - Anti-Nck antibody [Y531] (ab32120)

Nck Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Nck antibody

Immunocytochemistry/ Immunofluorescence analysis of HeLa (human cervix adenocarcinoma epithelial cell) cells labeling Nck with purified ab32120 at 1/250 dilution (8 µg/mL). Cells were fixed in 100% Methanol. Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) was used as the secondary antibody at 1/1000 (2 µg/mL) dilution. DAPI (blue) was used as nuclear counterstain.



Western blot - Anti-Nck antibody [Y531] (ab32120)

Nck Western blot staining using rabbit Anti-Nck antibody

Lanes 1-3: Merged signal (red and green). Green - ab32120 observed at 50 kDa. Red - loading control ab8245 observed at 36 kDa.
ab32120 Anti-Nck antibody [Y531] was shown to specifically react with Nck in wild-type HeLa cells. Loss of signal was observed when knockout cell line ab265323 (knockout cell lysate ab258059) was used. Wild-type and Nck knockout samples were subjected to SDS-PAGE. ab32120 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated at room temperature for 2. 5 hours at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-Nck antibody [Y531] (ab32120) at 1/1000 dilution

Lane 1:
Wild-type HeLa cell lysate at 20 µg

Lane 2:
NCK1 knockout HeLa cell lysate at 20 µg

Lane 2:
Western blot - Human NCK1 (Nck) knockout HeLa cell line (ab265323)

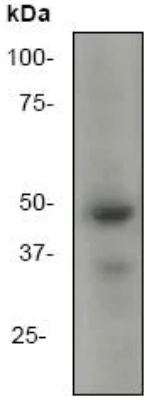
Lane 3:
NIH/3T3 cell lysate at 20 µg

Secondary

All lanes:
Western blot - Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (ab216773) at 1/10000 dilution

Predicted band size: 43 kDa

Observed band size: 50 kDa



Western blot - Anti-Nck antibody [Y531] (ab32120)

All lanes:
Western blot - Anti-Nck antibody [Y531] (ab32120) at 1/10000 dilution

All lanes:
Hela cell lysate

Predicted band size: 43 kDa

Observed band size: 35 kDa, 47 kDa

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