abcam

ab138488

Anti-NEK9 antibody [EP7361]

Recombinant | RabMAb | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Clone number	EP7361
Purification technique	Affinity purification Protein A
Concentration	0.384 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Not recommended

Species	Mouse, Rat, Human
Dilution info	-
Notes	-

WB

Tested

Species	Mouse
Dilution info	1/1000 - 1/10000
Notes	-

Species	Rat
Dilution info	1/1000 - 1/10000
Notes	-

Species	Human
Dilution info	1/1000 - 1/10000
Notes	-

ICC/IF

Tested

Species	Mouse
Dilution info	1/100
Notes	**For unpurified use at 1/250 - 1/500.**

Species	Human
Dilution info	1/100
Notes	**For unpurified use at 1/250 - 1/500.**

Expected

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

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	1/10 - 1/100
Notes	ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Expected

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

IHC-P

Tested

Species	Human
Dilution info	1/50 - 1/100
Notes	-

Not recommended

Species	Mouse, Rat
Dilution info	-
Notes	-

Target data

See full target information NEK9

Function Pleiotropic regulator of mitotic progression, participating in the control of spindle dynamics and chromosome separation (PubMed:12101123, PubMed:12840024, PubMed:14660563, PubMed:19941817). Phosphorylates different histones, myelin basic protein, beta-casein, and BICD2 (PubMed:11864968). Phosphorylates histone H3 on serine and threonine residues and beta-casein on serine residues (PubMed:11864968). Important for G1/S transition and S phase progression (PubMed:12840024, PubMed:14660563, PubMed:19941817). Phosphorylates NEK6 and NEK7 and stimulates their activity by releasing the autoinhibitory functions of Tyr-108 and Tyr-97 respectively (PubMed:12840024, PubMed:14660563, PubMed:19941817, PubMed:26522158).

Storage

Shipped at conditions	Conditional Ambient
Appropriate short-term storage duration	1-2 weeks
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, Stable for 12 months at -20°C

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	NEK9 also known as NIMA-related kinase 9 is a serine/threonine kinase with a molecular mass of approximately 118 kDa. It plays a role in the regulation of various cell cycle processes. NEK9 is active in both the cytoplasm and nucleus and expresses broadly in various tissues including the liver kidneys and pancreas. It also has homologs in other eukaryotic organisms indicating its conserved function across species.
Biological function summary	This protein modulates different aspects of mitosis particularly in spindle formation and chromosome segregation. NEK9 forms a part of a complex in conjunction with other kinases like NEK6 and NEK7 coordinating the early events of mitosis. Its activity supports centrosome separation and microtubule nucleation which are important for proper cell division.

Pathways	NEK9 is involved in the cell cycle and microtubule organization pathways. It works in conjunction with proteins such as PLK1 and Aurora A which regulate the entry and progression through mitosis. NEK9 activates NEK6 and NEK7 which further propagate signals necessary for mitotic spindle assembly ensuring efficient cell division.
Associated diseases and disorders	NEK9 has links to cancer and neurological disorders. Abnormal NEK9 activity has associations with tumorigenesis particularly linked with the dysregulation of cell cycle checkpoints and malignant transformation. In neurological disorder contexts NEK9 interacts with proteins like tau perturbing normal cellular functions and potentially contributing to disease pathology.

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

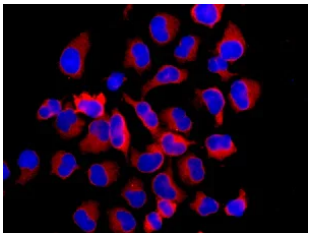
We do not recommend this combination. It is not covered by our product promise.

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.

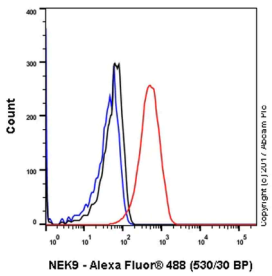
9 product images



Immunocytochemistry/ Immunofluorescence - Anti-NEK9 antibody [EP7361] (ab138488)

NEK9 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-NEK9 antibody

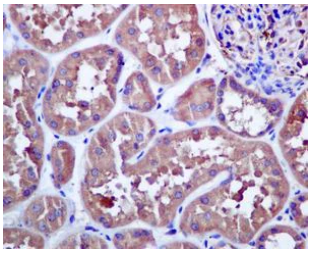
Immunofluorescent analysis of HeLa cells labelling NEK9 with unpurified ab138488 at 1/250 dilution.



Flow Cytometry (Intracellular) - Anti-NEK9 antibody [EP7361] (ab138488)

NEK9 Flow Cytometry (Intracellular) staining using rabbit Anti-NEK9 antibody

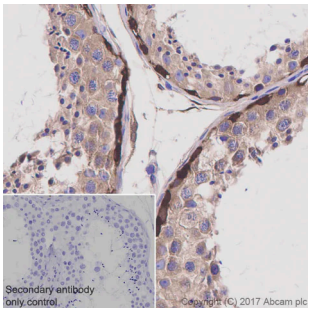
Intracellular Flow Cytometry analysis of HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling Sonic Hedgehog with purified ab53281 at 1/20 dilution (10 ug/ml) (red). Cells were fixed with 4% Paraformaldehyde. A Goat anti rabbit IgG (Alexa Fluor® 488) secondary antibody was used at 1/2000 dilution. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - cells without incubation with primary antibody and secondary antibody (Blue).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NEK9 antibody [EP7361] (ab138488)

NEK9 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-NEK9 antibody

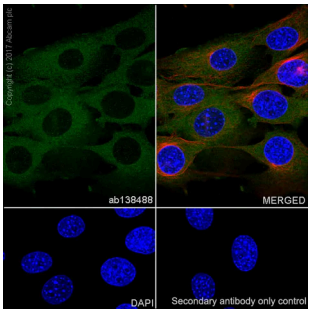
Immunohistochemical analysis of paraffin-embedded Human kidney tissue labelling NEK9 with unpurified ab138488 at 1/50 dilution.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NEK9 antibody [EP7361] (ab138488)

NEK9 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-NEK9 antibody

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human testis tissue sections labeling NEK9 with Purified ab138488 at 1:100 dilution (3.84 µg/ml). Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer pH 9.0). Tissue was counterstained with Hematoxylin. ImmunoHistoProbe one step HRP Polymer (ready to use) secondary antibody was used at 1:0 dilution. PBS instead of the primary antibody was used as the negative control.

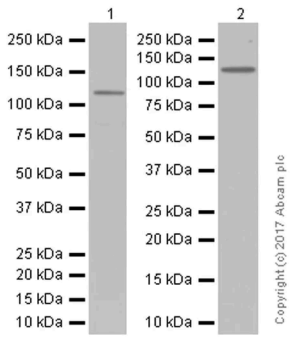


Immunocytochemistry/ Immunofluorescence - Anti-NEK9 antibody [EP7361] (ab138488)

NEK9 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-NEK9 antibody

Immunocytochemistry/ Immunofluorescence analysis of NIH/3T3 (Mouse embryonic fibroblast) cells labeling NEK9 with Purified ab138488 at 1:100 dilution (3.8µg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with

ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 µg/ml). ab150077 Goat anti rabbit IgG(Alexa Fluor® 488) was used as the secondary antibody at 1:1000 dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Western blot - Anti-NEK9 antibody [EP7361] (ab138488)

NEK9 Western blot staining using rabbit Anti-NEK9 antibody

Blocking and diluting buffer: 5% NFDM/TBST
All lanes:
Western blot - Anti-NEK9 antibody [EP7361] (ab138488) at 1/5000 dilution

Lane 1:
NIH/3T3 (Mouse embryonic fibroblast) whole cell lysate at 15 µg

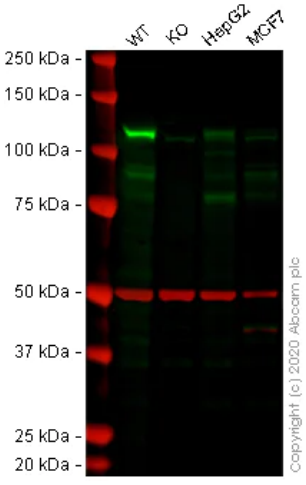
Lane 2:
THP-1 (Human monocytic leukemia monocyte) whole cell lysate at 15 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 107 kDa

Observed band size: 120 kDa



Western blot - Anti-NEK9 antibody [EP7361] (ab138488)

NEK9 Western blot staining using rabbit Anti-NEK9 antibody

Lanes 1 - 4: Merged signal (red and green). Green - ab138488 observed at 120 kDa. Red - loading control, ab7291 (Mouse anti-Alpha Tubulin [DM1A]) observed at 55kDa.
ab138488 was shown to react with NEK9 in wild-type A431 cells in western blot. Loss of signal was observed when NEK9 knockout sample was used. Wild-type and NEK9 knockout A431 cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween®) before incubation with ab138488 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 hour at room temperature before imaging.

All lanes:
Western blot - Anti-NEK9 antibody [EP7361] (ab138488) at 1/1000 dilution

Lane 1:
Wild-type A-431 (Human epidermoid carcinoma cell line) whole cell lysate at 20 µg

Lane 2:
NEK9 knockout A-431 (Human epidermoid carcinoma cell line) whole cell lysate at 20 µg

Lane 2:
Western blot - Human NEK9 knockout A-431 cell line (ab270474)

Lane 3:
Hep G2 (Human liver hepatocellular carcinoma cell line) whole cell lysate at 20 µg

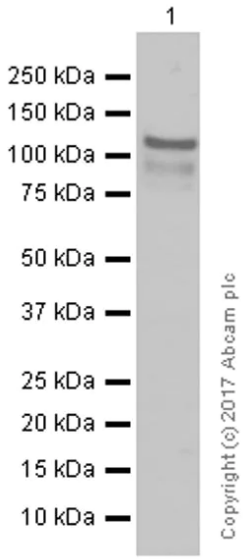
Lane 4:

MCF7 (Human breast adenocarcinoma cell line) whole cell lysate at 20 µg

Performed under reducing conditions.

Predicted band size: 107 kDa

Observed band size: 120 kDa



Western blot - Anti-NEK9 antibody [EP7361] (ab138488)

NEK9 Western blot staining of Rat spleen lysates using rabbit Anti-NEK9 antibody

Blocking and diluting buffer: 5% NFDM/TBST
All lanes:
Western blot - Anti-NEK9 antibody [EP7361] (ab138488) at 1000 Cells

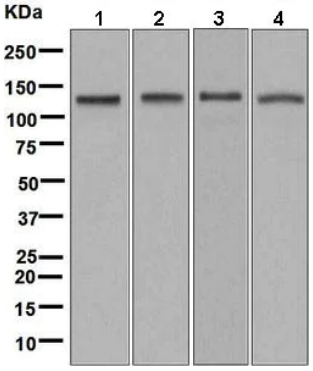
All lanes:
Rat spleen lysates at 15 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 107 kDa

Observed band size: 120 kDa



Western blot - Anti-NEK9 antibody [EP7361] (ab138488)

All lanes:
Western blot - Anti-NEK9 antibody [EP7361] (ab138488) at 1/1000 dilution

Lane 1:
NIH3T3 cell lysate at 10 µg

Lane 2:
HepG2 cell lysate at 10 µg

Lane 3:
HeLa cell lysate at 10 µg

Lane 4:
THP1 cell lysate at 10 µg

Secondary

All lanes:
HRP labelled goat anti-rabbit at 1/2000 dilution

Predicted band size: 107 kDa

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