abcam

ab126626

Anti-N WASP antibody [EPR6959]

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	EPR6959
Purification technique	Affinity purification Protein A
Dissociation constant	7.4×10^{-11} M
Concentration	1.235 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IHC-P

Tested

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

Perform heat-mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Expected

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

Predicted

Species	Pig
Dilution info	-
Notes	-

WB

Tested

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

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

Predicted

Species	Pig
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	1/300
Notes	**For unpurified, use 1/50 - 1/100.**

Expected

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

Predicted

Species	Pig
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	Purified format

Expected

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

Predicted

Species	Pig
Dilution info	-
Notes	-

Target data

See full target information Neural Wiskott-Aldrich syndrome protein

Function	Regulates actin polymerization by stimulating the actin-nucleating activity of the Arp2/3 complex (PubMed:16767080, PubMed:19366662, PubMed:19487689, PubMed:22847007, PubMed:22921828, PubMed:9422512). Involved in various processes, such as mitosis and cytokinesis, via its role in the regulation of actin polymerization (PubMed:19366662, PubMed:19487689, PubMed:22847007, PubMed:22921828, PubMed:9422512). Together with CDC42, involved in the extension and maintenance of the formation of thin, actin-rich surface projections called filopodia (PubMed:9422512). In addition to its role in the cytoplasm, also plays a role in the nucleus by regulating gene transcription, probably by promoting nuclear actin polymerization (PubMed:16767080). Binds to HSF1/HSTF1 and forms a complex on heat shock promoter elements (HSE) that negatively regulates HSP90 expression (By similarity). Plays a role in dendrite spine morphogenesis (By similarity). Decreasing levels of DNMBP (using antisense RNA) alters apical junction morphology in cultured enterocytes, junctions curve instead of being nearly linear (PubMed:19767742).

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	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	The protein N-WASP also known as neural Wiskott-Aldrich syndrome protein plays an important role in actin cytoskeleton dynamics. It weighs about 65 kDa and is widely expressed in diverse tissues but shows higher expression levels in the brain. In its mechanical function N-WASP regulates the polymerization of actin by activating the Arp2/3 complex. Through these interactions it helps in the formation of branched actin networks which are essential for cellular movement and shape.
Biological function summary	N-WASP functions are critical in cell signaling endocytosis and neurite extension. N-WASP often acts within a complex and interacts with various proteins like Cdc42 to transmit signals from cell surface to the actin cytoskeleton. These activities position N-WASP as a fundamental component for cellular architecture and mobility impacting processes like immune response and neuronal development. Its interaction with Arp2/3 contributes to dynamic changes in cell movement and shape.
Pathways	N-WASP plays a role in Rac and Cdc42 signaling pathways important for actin reorganization. Within these pathways it interacts with proteins such as Rho family GTPases influencing cytoskeletal arrangements. The proper functioning of these pathways is vital for numerous cellular processes including the migration of cells which affects tissue development and repair.
Associated diseases and disorders	Alterations in N-WASP are linked to immunodeficiencies and neurological disorders. Mutations or dysfunction in N-WASP can lead to Wiskott-Aldrich syndrome characterized by immune defects and eczema due to its role in immune cell signaling. Additionally N-WASP's interaction with proteins such as WASP contributes to its involvement in neurodevelopmental disorders like autistic spectrum disorders reflecting the importance of this protein in maintaining normal cellular function.

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.

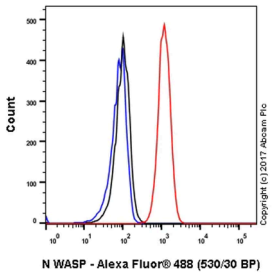
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

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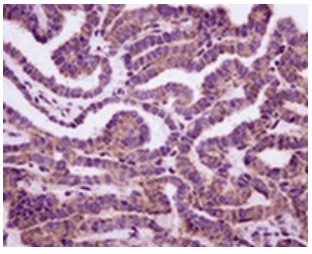
10 product images



Flow Cytometry (Intracellular) - Anti-N WASP antibody [EPR6959] (ab126626)

N WASP Flow Cytometry (Intracellular) staining using rabbit Anti-N WASP antibody

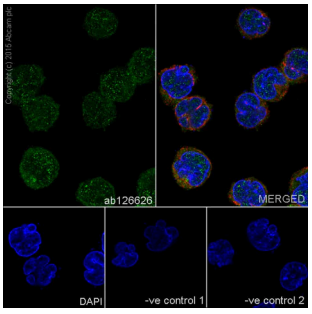
Intracellular Flow Cytometry analysis of K562 (human chronic myelogenous leukemia) cells labeling N WASP with purified ab126626 at 1/150 (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat Anti-Rabbit IgG H&L (Alexa Fluorr® 488) (ab150077)was used as the secondary antibody. Rabbit IgG, monoclonal [EPR25A] - Isotype Control (ab172730) (Black) was used as the isotype control, cells without incubation with primary antibody and secondary antibody (Blue) were used as the unlabeled control.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-N WASP antibody [EPR6959] (ab126626)

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

Unpurified ab126626, at 1/50, staining N WASP in formalin fixed, paraffin embedded human papillary carcinoma tissue by Immunohistochemistry
Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

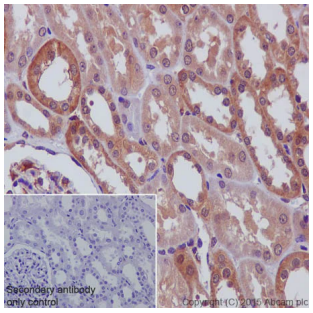


Immunocytochemistry/ Immunofluorescence - Anti-N WASP antibody [EPR6959] (ab126626)

N WASP Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-N WASP antibody

Immunofluorescence staining of K562 cells with purified ab126626 at a working dilution of 1/300, 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

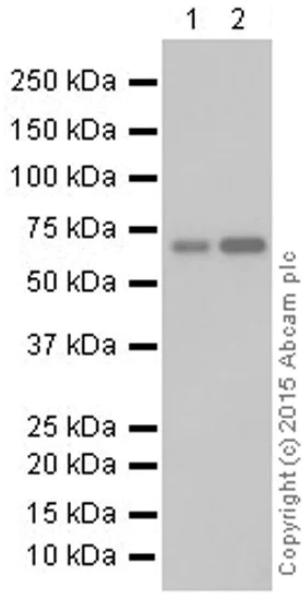
the top right hand panel. The cells were fixed in 4% PFA and permeabilized using 0.1% Triton X 100. The negative controls are shown in bottom middle and right hand panels - for negative control 1, purified ab126626 was used at a dilution of 1/500 followed by an Alexa Fluor® 594 goat anti-mouse antibody (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.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-N WASP antibody [EPR6959] (ab126626)

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

Immunohistochemical staining of paraffin embedded human kidney with purified ab126626 at a working dilution of 1/100. The secondary antibody used is ab97051, a goat anti-rabbit IgG (H&L) at a dilution of 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.



Western blot - Anti-N WASP antibody [EPR6959] (ab126626)

Blocking buffer: 5% NFDM/TBST
Dilution buffer: 5% NFDM/TBST
All lanes:
Western blot - Anti-N WASP antibody [EPR6959] (ab126626) at 1/10000 dilution

Lane 1:
HeLa whole cell lysate at 10 µg

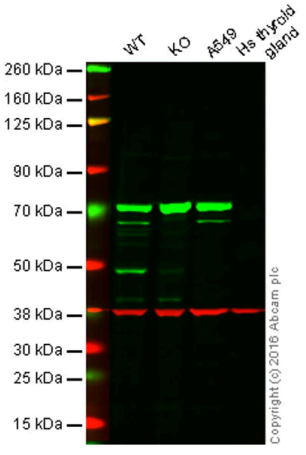
Lane 2:
K562 whole cell lysate at 10 µg

Secondary

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

Predicted band size: 35 kDa, 38 kDa, 42 kDa, 50 kDa, 55 kDa

Observed band size: 42 kDa, 65 kDa



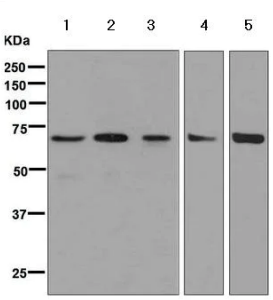
Western blot - Anti-N WASP antibody [EPR6959] (ab126626)

N WASP Western blot staining using rabbit Anti-N WASP antibody

Lane 1: Wild-type HAP1 cell lysate (20 µg)
Lane 2: N WASP knockout HAP1 cell lysate (20 µg)
Lane 3: A549 cell lysate (20 µg)
Lane 4: Human thyroid tissue lysate (20 µg)
Lanes 1 - 4: Merged signal (red and green). Green - ab126626 observed at 67 kDa. Red - loading control, ab8245, observed at 37 kDa.
ab126626 was shown to recognize N WASP when N WASP knockout samples were used, along with additional cross-reactive bands. Wild-type and N WASP knockout samples were subjected to SDS-PAGE. ab126626 and ab8245 (loading control to GAPDH) were diluted 1/1000 and 1/2000 respectively and incubated overnight at 4°C. 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 1/10000 dilution for 1 h at room temperature before imaging.
All lanes:
Western blot - Anti-N WASP antibody [EPR6959] (ab126626)

Predicted band size: 55 kDa



Western blot - Anti-N WASP antibody [EPR6959] (ab126626)

All lanes:
Western blot - Anti-N WASP antibody [EPR6959] (ab126626) at 1/1000 dilution

Lane 1:
NIH 3T3 cell lysate at 10 µg

Lane 2:
F9 cell lysate at 10 µg

Lane 3:
L929 cell lysate at 10 µg

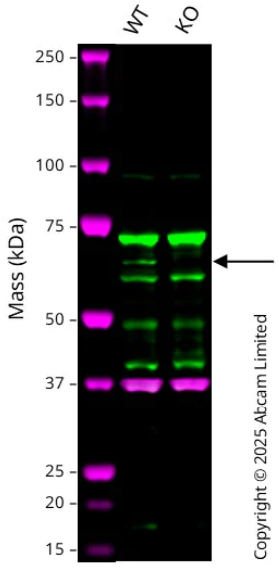
Lane 4:
HeLa cell lysate at 10 µg

Lane 5:
K562 cell lysate at 10 µg

Secondary

All lanes:
Goat anti-rabbit HRP conjugated antibody at 1/2000 dilution

Predicted band size: 55 kDa



Western blot - Anti-N WASP antibody [EPR6959] (ab126626)

N WASP Western blot staining using rabbit Anti-N WASP antibody

Western blot: Rabbit Monoclonal[EPR6959] to N WASP ab126626 staining at 1/1000 dilution, shown in green; Mouse anti GAPDH ab8245 loading control staining at 1/20000 dilution, shown in magenta. A band was observed at 62 kDa in Wild-type U-87 MG cell lysates with no signal observed at this size in WASL knockout U-87 MG cell line. To generate this image, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3pc Milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit 800CW and Goat anti-Mouse 680RD at 1/20,000 dilution.
All lanes:
Western blot - Anti-N WASP antibody [EPR6959] (ab126626) at 1/1000 dilution

Lane 1:
Wild-type U-87 MG at 20 µg

Lane 2:
WASL knockout U-87 MG at 20 µg

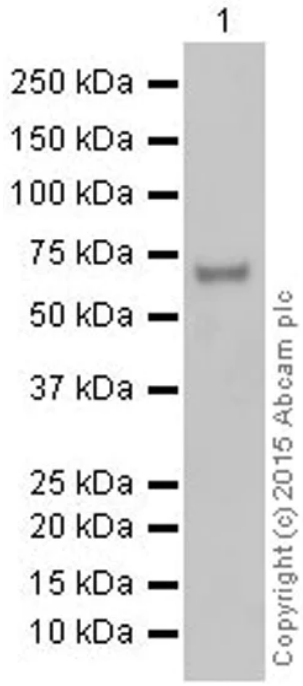
Secondary

All lanes:
Goat anti-Rabbit 800CW & Goat anti-Mouse 680RD at 1/20000 dilution

Performed under reducing conditions.

Predicted band size: 55 kDa

Observed band size: 62 kDa, 36 kDa



Western blot - Anti-N WASP antibody [EPR6959] (ab126626)

All lanes:
Western blot - Anti-N WASP antibody [EPR6959] (ab126626) at 1/10000 dilution

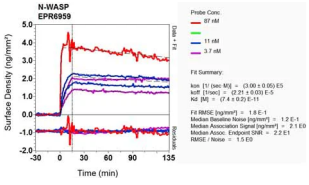
All lanes:
L929 whole cell lysate at 10 µg

Secondary

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

Predicted band size: 55 kDa

Observed band size: 65 kDa



OI-RD Scanning - Anti-N WASP antibody [EPR6959] (ab126626)

We have systematically measured KD (the equilibrium dissociation constant between the antibody and its antigen), of more than 840 recombinant antibodies to assess not only their individual KD values but also to see the average affinity of antibody. Based on the comparison with published literature values for mouse monoclonal antibodies, Recombinant antibodies appear to be on average 1-2 order of magnitude higher affinity.

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