abcam

ab220816

Anti-NEDD8 antibody [Y297] - BSA and Azide free

Recombinant | RabMAb

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	Y297
Purification technique	Affinity purification Protein A
Specificity	This antibody detects NEDD8 bound to cullins proteins (neddylated cullins) as well as free NEDD8.
Concentration	0.957 - 1.01 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Human

Dilution info	-
Notes	-

Predicted

Species	Mouse, Rat
Dilution info	-
Notes	-

WB

Expected

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

ICC/IF

Tested

Species	Human
Dilution info	-
Notes	-

Predicted

Species	Mouse, Rat
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-

Notes	-

Predicted

Species	Mouse, Rat
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human
Dilution info	-
Notes	-

Predicted

Species	Mouse, Rat
Dilution info	-
Notes	-

Target data

See full target information NEDD8

Function	Ubiquitin-like protein which plays an important role in cell cycle control and embryogenesis via its conjugation to a limited number of cellular proteins, such as cullins or p53/TP53 (PubMed:10318914, PubMed:10597293, PubMed:11953428, PubMed:14690597, PubMed:15242646, PubMed:9694792, PubMed:38605244, PubMed:38316879). Attachment of NEDD8 to cullins is critical for the recruitment of E2 to the cullin-RING-based E3 ubiquitin-protein ligase complex, thus facilitating polyubiquitination and proteasomal degradation of cyclins and other regulatory proteins (PubMed:10318914, PubMed:10597293, PubMed:11953428, PubMed:20688984, PubMed:9694792, PubMed:38605244, PubMed:38316879). Attachment of NEDD8 to p53/TP53 inhibits p53/TP53 transcriptional activity (PubMed:15242646). Covalent attachment to its substrates requires prior activation by the E1 complex UBE1C-APPBP1 and linkage to the E2 enzyme UBE2M (PubMed:14690597).

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

ab220816 is the carrier-free version of ab81264.

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.

Conjugation ready

Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. 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.

Compatibility

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.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	NEDD8 also known as neural precursor cell expressed developmentally downregulated protein 8 is a small ubiquitin-like protein with a molecular weight of approximately 9 kDa. It is expressed in various tissues prominently in the brain and muscles. NEDD8 conjugates to target proteins modifying their functions by

	altering their stability activity or location. It plays a role in the regulation of protein degradation.
Biological function summary	The NEDD8 protein is part of a modification system that activates cullin-RING ligase complexes. This activation promotes the ubiquitination and subsequent proteasomal degradation of targeted proteins. As a post-translational modification system similar to ubiquitin NEDD8 is important for maintaining protein homeostasis by ensuring timely degradation and turnover of cellular proteins. These processes impact cell cycle progression signal transduction and stress responses.
Pathways	NEDD8 is a central player in the ubiquitin-proteasome system an essential pathway for protein turnover. It is particularly important in the NEDDylation pathway where it regulates protein functions by transiently attaching to substrate proteins. Through these processes NEDD8 interacts closely with proteins like cullins and ubiquitin affecting cellular responses and maintaining cellular integrity under different conditions.
Associated diseases and disorders	NEDD8 influences several pathological states. Aberrant NEDD8 conjugation can lead to cancer by disrupting the controlled degradation of proteins involved in cell cycle regulation such as p27 and p21. Additionally improper NEDDylation has links to neurodegenerative diseases like Alzheimer's as it affects proteins related to synaptic function and neuronal survival. Understanding the role of NEDD8 in these diseases aids in the development of potential therapeutic strategies.

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

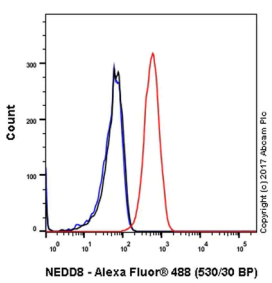
We do not recommend this combination. It is not covered by our product promise.

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:

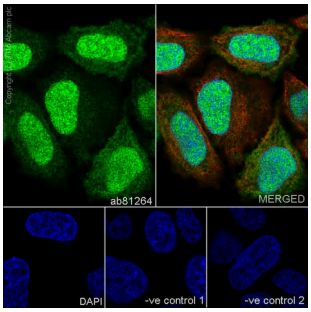
6 product images



Flow Cytometry (Intracellular) - Anti-NEDD8 antibody [Y297] - BSA and Azide free (ab220816)

NEDD8 Flow Cytometry (Intracellular) staining using rabbit Anti-NEDD8 antibody

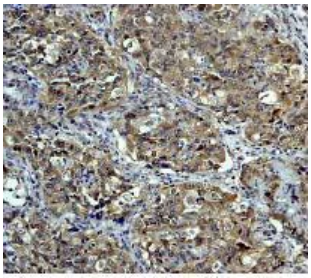
Intracellular Flow Cytometry analysis of A-673 (Human muscle Ewing's Sarcoma) cells labeling NEDD8 (red) with purified ab81264 at a 1/300 dilution (1ug/mL). Cells were fixed with 4% paraformaldehyde and permeabilized with 90% methanol. A goat anti rabbit IgG (Alexa Fluor® 488) (ab150077) was used as the secondary antibody at a 1/2000 dilution. Black - Rabbit monoclonal IgG (Black) (ab172730). Blue (unlabeled control) - Cell without incubation with primary antibody and secondary antibody (Blue).
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab81264).



Immunocytochemistry/ Immunofluorescence - Anti-NEDD8 antibody [Y297] - BSA and Azide free (ab220816)

NEDD8 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-NEDD8 antibody

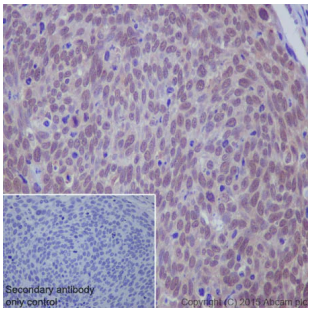
Immunofluorescence staining of HeLa cells with purified ab81264 at a working dilution of 1/100, counter-stained with DAPI. The secondary antibody was Alexa Fluor® 488 goat anti-rabbit (ab150077), used at a dilution of 1/1000. ab7291, a mouse anti-tubulin antibody (1/1000), was used to stain tubulin along 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 ab81264 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.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab81264).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NEDD8 antibody [Y297] - BSA and Azide free (ab220816)

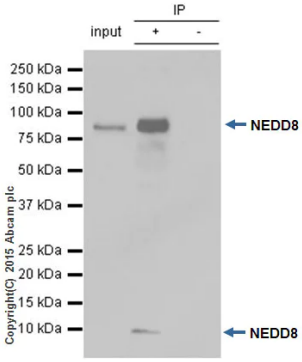
NEDD8 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-NEDD8 antibody

Unpurified ab81264, at 1/100 dilution, staining NEDD8 in human lung carcinoma tissue by Immunohistochemistry using formalin-fixed, paraffin-embedded section.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab81264).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-NEDD8 antibody [Y297] - BSA and Azide free (ab220816)

Immunohistochemical staining of paraffin embedded human cervical carcinoma with purified ab81264 at a working dilution of 1/400. 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.This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab81264).

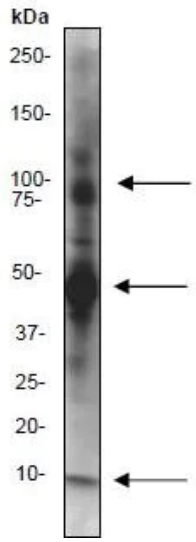


Immunoprecipitation - Anti-NEDD8 antibody [Y297] - BSA and Azide free (ab220816)

ab81264 (purified) at 1/20 immunoprecipitating NEDD8 in 10 µg K562 cell lysate (Lanes 1 and 2, observed at 10 and 90 kDa). Lane 3 - Rabbit monoclonal IgG (ab172730). For western blotting, HRP Veriblot for IP (ab131366) was used for detection at 1/1000 dilution. Blocking buffer and concentration: 5% NFDM/TBST Dilution 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 (ab81264).

All lanes:
Immunoprecipitation - Anti-NEDD8 antibody [Y297] (ab81264)

Predicted band size: 9 kDa



Immunoprecipitation - Anti-NEDD8 antibody [Y297] - BSA and Azide free (ab220816)

Western blot analysis of immunoprecipitation pellet from HeLa cell lysate using unpurified ab81264 at 1/500 dilution. Band at 90 kDa shows Neddylated cullins, 50 kDa the IgG fraaction and 9 kDa Free NEDD8.
This data was developed using the same antibody clone in a different buffer formulation containing PBS, BSA, glycerol, and sodium azide (ab81264).

All lanes:
Immunoprecipitation - Anti-NEDD8 antibody [Y297] (ab81264)

Predicted band size: 9 kDa

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