abcam

ab275024

Anti-NG2 antibody [EPR23976-145]

Anti-NG2 antibody [EPR23976-145] (ab275024) is a rabbit monoclonal antibody detecting NG2 in **Western Blot, Flow Cytometry, IP, ICC/IF**. Suitable for **Human, Mouse, Rat**.

- Biophysical QC for unrivalled batch-batch consistency

Recombinant | RabMAb | 20ul selling size

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	EPR23976-145
Purification technique	Affinity purification Protein A
Concentration	0.468 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

ICC/IF

Tested

Species	Mouse
Dilution info	1/50

Notes	-

Species	Rat
Dilution info	1/50
Notes	-

Species	Human
Dilution info	1/50
Notes	-

IP

Tested

Species	Mouse
Dilution info	1/30
Notes	-

Expected

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

Flow Cyt

Tested

Species	Mouse
Dilution info	1/500
Notes	-

Species	Human
Dilution info	1/500
Notes	-

Expected

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

WB

Tested

Species	Mouse
Dilution info	1/1000
Notes	-

Species	Rat
Dilution info	1/1000
Notes	-

Species	Human
Dilution info	1/1000
Notes	-

IHC-P

Not recommended

Species	Human, Mouse, Rat
Dilution info	-
Notes	-

Target data

See full target information CSPG4

Function Proteoglycan playing a role in cell proliferation and migration which stimulates endothelial cells motility during microvascular morphogenesis. May also inhibit neurite outgrowth and growth cone collapse during axon regeneration. Cell surface receptor for collagen alpha 2(VI) which may confer cells ability to migrate on that substrate. Binds through its extracellular N-terminus growth factors, extracellular matrix proteases modulating their activity. May regulate MPP16-dependent degradation and invasion of type I collagen participating in melanoma

cells invasion properties. May modulate the plasminogen system by enhancing plasminogen activation and inhibiting angiostatin. Also functions as a signal transducing protein by binding through its cytoplasmic C-terminus scaffolding and signaling proteins. May promote retraction fiber formation and cell polarization through Rho GTPase activation. May stimulate alpha-4, beta-1 integrin-mediated adhesion and spreading by recruiting and activating a signaling cascade through CDC42, ACK1 and BCAR1. May activate FAK and ERK1/ERK2 signaling cascades.

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

Notes

What is this antibody validated in?
Anti-NG2 antibody [EPR23976-145] (ab275024) is a rabbit recombinant monoclonal antibody and is validated for use in Western Blot (WB), Flow Cytometry (Flow Cyt), Immunoprecipitation (IP), Immunocytochemistry/immunofluorescence (ICC/IF) in Human, Mouse, Rat samples.

What is the molecular weight of NG2?
Anti-NG2 [EPR23976-145] (ab275024) specifically detects a band for NG2 (UniProt: Q8VHY0) at a molecular weight of 251kDa.

Other related products
We have a range of other formats of antibody clone [EPR23976-145] also available for your convenience: ab275024, Carrier free - ab275038, Alexa Fluor® 647 - ab283639, PE - ab314252

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.

Trial sizes available!
Test your antibody or perform pre-screening before committing to a larger quantity. Sold in 20µl. Discover our selection of trial-size antibodies.

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

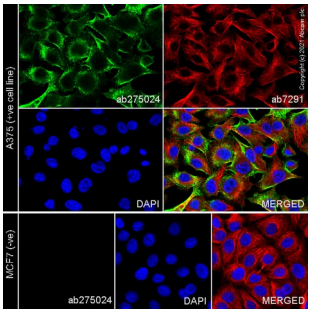
We do not recommend this combination. It is not covered by our product promise.

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.

10 product images



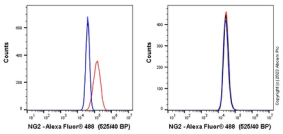
Immunocytochemistry/ Immunofluorescence - Anti-NG2 antibody [EPR23976-145] (ab275024)

NG2 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-NG2 antibody

ab275024 staining NG2 in A375 cells, with negative expression in MCF7 cells. The cells were fixed with 100% methanol (5 min), permeabilised with 0.1% Triton x-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at +4°C with ab275024 at 5 µg/ml and ab7291, Mouse monoclonal [DM1A] to alpha Tubulin at 0.5 µg/ml. Cells were then incubated with ab150081, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488), pre-adsorbed at 1/1000 dilution (shown in green)

and ab150119, Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor® 647), pre-adsorbed at 1/1000 dilution (shown in red). Nuclear DNA was labelled with DAPI (shown in blue). Image was acquired with a confocal microscope (Leica-Microsystems TCS SP8) and a single confocal section is shown.

This product also work with 4% formaldehyde (10 min) fixation under the same testing conditions.



Flow Cytometry - Anti-NG2 antibody [EPR23976-145] (ab275024)

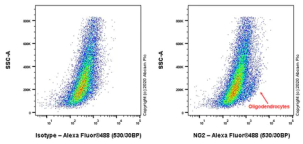
NG2 Flow Cytometry staining using rabbit Anti-NG2 antibody

Flow cytometry overlay histogram showing left A-375 positive cells and right negative MCF7 stained with ab275024 (red line). The cells were incubated in 1x PBS containing 10% normal goat serum to block non-specific protein-protein interactionfollowed by the antibody (ab275024) (1x 106 in 100µl at 5.0 µg/ml (1/402)) for 30min on ice.

The secondary antibody Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed was incubated at 1/4000 for 30min on ice

Isotype control antibody (black line) was Recombinant Rabbit IgG, monoclonal [EPR25A] - Isotype Control used at the same concentration and conditions as the primary antibody. Unlabelled sample (blue line) was also used as a control.

Acquisition of >5000 events were collected using a 50 mW Blue laser (488nm) and 525/40 bandpass filter.

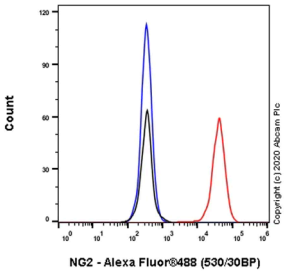


Flow Cytometry - Anti-NG2 antibody [EPR23976-145] (ab275024)

NG2 Flow Cytometry staining using rabbit Anti-NG2 antibody

Flow cytometric analysis of Mouse primary neural glia cell cells labelling NG2 with ab275024 at 1/500 dilution (0.1ug) (Right) compared with a Rabbit monoclonal IgG (ab172730) isotype control (Left).
Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody.

Gated on viable cells.

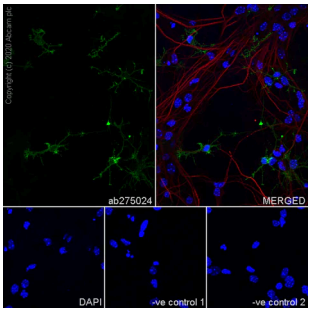


Flow Cytometry - Anti-NG2 antibody [EPR23976-145] (ab275024)

NG2 Flow Cytometry staining using rabbit Anti-NG2 antibody

Flow cytometric analysis of LADMAC (Mouse bone marrow monocyte macrophage) cells labelling NG2 with ab275024 at 1/500 dilution (0.1ug) (Red) compared with a Rabbit monoclonal IgG (ab172730) isotype control (Black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue).
Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody.

Gated on viable cells.

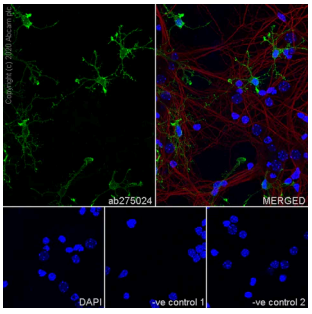


Immunocytochemistry/ Immunofluorescence - Anti-NG2 antibody [EPR23976-145] (ab275024)

NG2 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-NG2 antibody

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized rat primary neural/glia cell cells labelling NG2 with ab275024 at 1/100 (4.67 ug/ml) dilution, followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) antibody at 1/1000 dilution (Green). Confocal image showing positive staining in rat primary glia cells.Confocal scanning Z step was set as 0.3 µm followed by image processing with maximum Z projection. ab11267 Anti-MAP2 mouse monoclonal antibody was used to counterstain MAP2 at 1/500 dilution, followed by ab150120 Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) at 1/1000 dilution (Red). The Nuclear counterstain was DAPI (Blue).
-ve control 1: ab275024 at 1/100 dilution, followed by ab150120 Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) at 1/1000 dilution.

-ve control 2: ab11267 at 1/500 dilution, followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution.

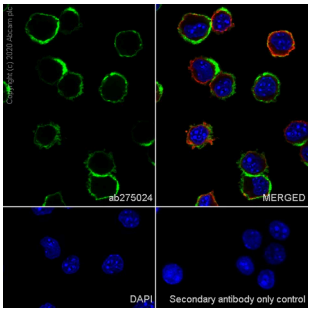


Immunocytochemistry/ Immunofluorescence - Anti-NG2 antibody [EPR23976-145] (ab275024)

NG2 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-NG2 antibody

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized mouse primary neural/glia cell cells labelling NG2 with ab275024 at 1/100 (4.67 ug/ml) dilution, followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) antibody at 1/1000 dilution (Green). Confocal image showing positive staining in mouse primary glia cells. Confocal scanning Z step was set as 0.3 µm followed by image processing with maximum Z projection. is observed. ab11267 Anti-MAP2 mouse monoclonal antibody was used to counterstain MAP2 at 1/500 dilution, followed by ab150120 Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) at 1/1000 dilution (Red). The Nuclear counterstain was DAPI (Blue).
ve control 1: ab275024 at 1/100 dilution, followed by ab150120 Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) at 1/1000 dilution.

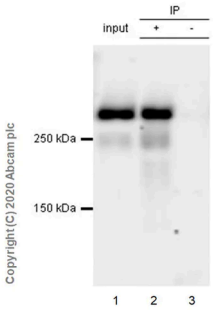
-ve control 2: ab11267 at 1/500 dilution, followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution.



Immunocytochemistry/ Immunofluorescence - Anti-NG2 antibody [EPR23976-145] (ab275024)

NG2 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-NG2 antibody

Immunofluorescent analysis of 4% Paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized LADMAC cells labelling NG2 with ab275024 at 1/50 (9.34 ug/ml) dilution, followed by ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) antibody at 1/1000 dilution (Green). Confocal image showing membranous staining in LADMAC cell line. ab195889 Anti-alpha Tubulin mouse monoclonal antibody - Microtubule Marker (Alexa Fluor® 594) was used to counterstain tubulin at 1/200 dilution (Red). The Nuclear counterstain was DAPI (Blue).
Secondary antibody only control: Secondary antibody is ab150077 Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) at 1/1000 dilution.



Immunoprecipitation - Anti-NG2 antibody [EPR23976-145] (ab275024)

NG2 was immunoprecipitated from 0.35 mg Mouse brain tissue lysate with ab275024 at 1/30 dilution (2ug in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab275024 at 1/500 dilution. VeriBlot for IP Detection Reagent (HRP)(ab131366) was used at 1/1000 dilution.
Lane 1: Mouse brain tissue lysate 10 ug

Lane 2: ab275024 IP in Mouse brain tissue lysate

Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab275024 in mouse brain tissue lysate

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

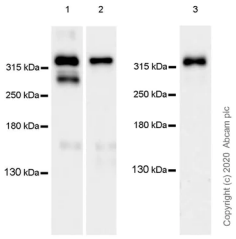
Exposure time: 15 seconds.

Sample loaded onto lane 1 was non-boiled as boiling may cause protein aggregates.

All lanes:
Immunoprecipitation - Anti-NG2 antibody [EPR23976-145] (ab275024)

Predicted band size: 251 kDa

Observed band size: 280 kDa, 330 kDa



Western blot - Anti-NG2 antibody [EPR23976-145] (ab275024)

Blocking and dilution buffer: 5% NFDM/TBST.
Exposure times: Lane 1: 26 seconds; Lane 2: 59 seconds; Lane 3: 125 seconds.

The band of 330KDa represents the intact NG2 proteoglycan modified by chondroitin sulfate, the band of 280KDa represents NG2 core protein.

The molecular weight observed is consistent with what has been described in the literature (PMID: 20455858, 16625365, 23481707).

Samples are non-boiled as boiling may cause protein aggregates.

All lanes:
Western blot - Anti-NG2 antibody [EPR23976-145] (ab275024) at 1/1000 dilution

Lane 1:
A375 (human malignant melanoma epithelial cell) whole cell lysate at 20 µg

Lane 2:
SK-MEL-28 (human malignant melanoma) whole cell lysate at 20 µg

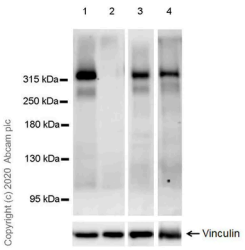
Lane 3:
Human pancreas tissue lysate at 40 µg

Secondary

All lanes:
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (ab97051) at 1/50000 dilution

Predicted band size: 251 kDa

Observed band size: 280 kDa, 330 kDa



Western blot - Anti-NG2 antibody [EPR23976-145] (ab275024)

NG2 Western blot staining using rabbit Anti-NG2 antibody

Blocking and dilution buffer: 5% NFDM/TBST.
Exposure times: Lanes 1-3: 59 seconds; Lane 4: 81 seconds.

The band of 330KDa represents the intact NG2 proteoglycan modified by chondroitin sulfate, the band of 280KDa represents NG2 core protein.
The molecular weight observed is consistent with what has been described in the literature (PMID: 20455858, 16625365, 23481707).

Negative control: Mouse liver (PMID: 23481707).

Samples are non-boiled as boiling may cause protein aggregates.

All lanes:
Western blot - Anti-NG2 antibody [EPR23976-145] (ab275024) at 1/1000 dilution

Lane 1:
Mouse brain tissue lysate at 20 µg

Lane 2:
Mouse liver tissue lysate at 20 µg

Lane 3:
Mouse pancreas tissue lysate at 40 µg

Lane 4:
Rat brain tissue lysate at 40 µg

Secondary

All lanes:
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (ab97051) at 1/50000 dilution

Predicted band size: 251 kDa

Observed band size: 280 kDa, 330 kDa

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