abcam

Product datasheet

Anti-Nestin antibody [EPR1301(2)] ab176571

Recombinant RabMAb

6 References 8 Images

Overview

Product name Anti-Nestin antibody [EPR1301(2)]

Description Rabbit monoclonal [EPR1301(2)] to Nestin

Host species Rabbit

Tested applications Suitable for: Flow Cyt (Intra), ICC/IF, IHC-P

Unsuitable for: IP or WB

Species reactivity Reacts with: Human

Immunogen Synthetic peptide within Human Nestin aa 550-650 (Cysteine residue). The exact sequence is

proprietary.

Database link: P48681

Positive control IHC-P: Human kidney, spleen and melanoma tissues. ICC/IF: This antibody gave a positive result

when used in the following methanol fixed cell lines: SKNSH, U-87 MG Flow Cyt (intra): SH-SY5Y

General notesThis 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 production

For more information see here.

Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb**[®] **patents**.

Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with

these species. Please contact us for more information.

Properties

Form Liquid

Storage instructions Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long

term. Avoid freeze / thaw cycle.

Storage buffer Preservative: 0.01% Sodium azide

Constituents: PBS, 0.05% BSA, 40% Glycerol (glycerin, glycerine)

Purity Protein A purified

1

ClonalityMonoclonalClone numberEPR1301(2)

Isotype IgG

Applications

The Abpromise guarantee

Our <u>Abpromise guarantee</u> covers the use of ab176571 in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		1/160.
ICC/IF		1/50 - 1/100. For unpurified use at 1/50 - 1/100.
IHC-P		1/1000. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol. The use of an HRP/AP polymerized secondary antibody is recommended. See IHC antigen retrieval protocols. For unpurified use at 1/250 - 1/500.

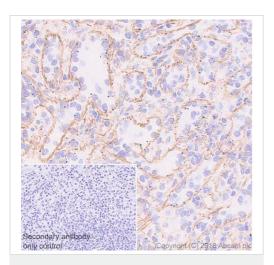
Application notes

Is unsuitable for IP or WB.

Target

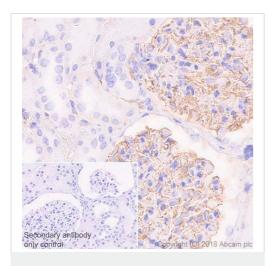
Function	Required for brain and eye development. Promotes the disassembly of phosphorylated vimentin intermediate filaments (IF) during mitosis and may play a role in the trafficking and distribution of IF proteins and other cellular factors to daughter cells during progenitor cell division. Required for survival, renewal and mitogen-stimulated proliferation of neural progenitor cells.	
Tissue specificity	CNS stem cells.	
Sequence similarities	Belongs to the intermediate filament family.	
Developmental stage	Upon terminal neural differentiation, nestin is down-regulated and replaced by neurofilaments.	
Post-translational modifications	Constitutively phosphorylated. This increases during mitosis when the cytoplasmic intermediate filament network is reorganized.	

Images



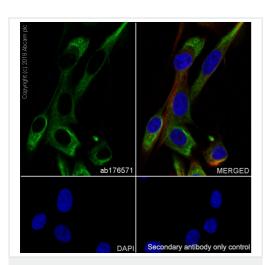
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Nestin antibody
[EPR1301(2)] (ab176571)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human spleen tissue sections labeling Nestin with purified ab176571 at 1:1000 dilution (1.65 μ g/ml). Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use)was used as the secondary antibody. Negative control:PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



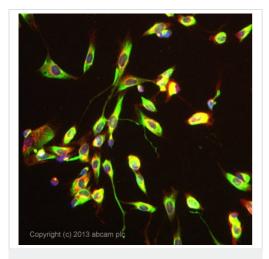
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Nestin antibody
[EPR1301(2)] (ab176571)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human kidney tissue sections labeling Nestin with purified ab176571 at 1:1000 dilution (1.65 µg/ml). Heat mediated antigen retrieval was performed using ab93684 (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use)was used as the secondary antibody. Negative control:PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Immunocytochemistry/ Immunofluorescence - Anti-Nestin antibody [EPR1301(2)] (ab176571)

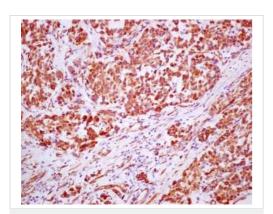
Immunocytochemistry/ Immunofluorescence analysis of U-87 MG (Human glioblastoma-astrocytoma epithelial cell) cells labeling Nestin with purified ab176571 at 1:150 dilution (10 μ g/ml). Cells were fixed in 100% Methanol and permeabilized with None. Cells were counterstained with Ab195889 Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) 1:200 (2.5 μ g/ml). Goat anti rabbit lgG (Alexa Fluor® 488, <u>ab150077</u>) was used as the secondary antibody at 1:1000 (2 μ g/ml) dilution. DAPI nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



Immunocytochemistry/ Immunofluorescence - Anti-Nestin antibody [EPR1301(2)] (ab176571)

ICC/IF image of ab176571 stained SK-N-SH (Human neuroblastoma cell line) cells.

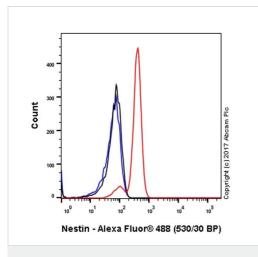
The cells were fixed in 100% methanol (5 min) and then incubated in 1% BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilize the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody ab176571 at 5 μ g/ml overnight at +4°C. The secondary antibody (pseudo-colored green) was Alexa Fluor[®] 488 goat antirabbit (ab150081) lgG (H+L) preadsorbed, used at a 1/1000 dilution for 1h. Alexa Fluor[®] 594 WGA was used to label plasma membranes (pseudo-colored red) at a 1/200 dilution for 1h at room temperature. DAPI was used to stain the cell nuclei (pseudo-colored blue) at a concentration of 1.43 μ M for 1hour at room temperature.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Nestin antibody
[EPR1301(2)] (ab176571)

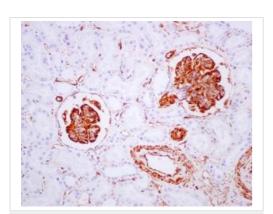
Immunohistochemical analysis of paraffin-embedded human melanoma tissue labeling Nestin using ab176571 at a 1/250 dilution.

Heat mediated antigen retrieval was performed with citrate buffer pH 6 before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-Nestin antibody [EPR1301(2)] (ab176571)

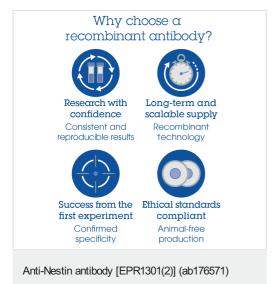
Intracellular Flow Cytometry analysis of SH-SY5Y (human neuroblastoma) cells labeling Nestin with purified ab176571 at 1/160 dilution (10µg/ml) (red). Cells were fixed with 4% paraformaldehyde and permeabilized with 90% methanol. A Goat anti rabbit lgG (Alexa Fluor $^{\&}$ 488) (1/2000 dilution) was used as the secondary antibody. Rabbit monoclonal lgG (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 paraffinembedded sections) - Anti-Nestin antibody
[EPR1301(2)] (ab176571)

Immunohistochemical analysis of paraffin-embedded human kidney tissue labeling Nestin using ab176571 at a 1/250 dilution.

Heat mediated antigen retrieval was performed with citrate buffer pH 6 before commencing with IHC staining protocol.



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