abcam

Product datasheet

Anti-Doublecortin antibody ab18723

39 Abreviews 190 References 14 Images

Overview

Product name	Anti-Doublecortin antibody
Description	Rabbit polyclonal to Doublecortin
Host species	Rabbit
Specificity	Please note: Low dilutions of this antibody can cause high background in IHC. Please use as high a dilution as possible. Optimal working dilutions are batch dependent.
Tested applications	**Suitable for:** WB, ICC, ICC/IF, IHC-FrFl, IHC-FoFr, IHC-Fr, IHC-P
Species reactivity	**Reacts with:** Mouse, Rat, Chicken, Cat, Human, Cynomolgus monkey, Quail, Rhesus monkey
Immunogen	Synthetic peptide conjugated to KLH derived from within residues 300 to the C-terminus of Human Doublecortin. Read Abcam's proprietary immunogen policy (Peptide available as ab19804.)
Positive control	This antibody gave a positive signal in Brain (Mouse) Tissue Lysate - normal tissue, 0 days old ICC-IF: SHSY5Y cells. IHC-P: FFPE Rat brain 6 weeks/FFPE Mouse Brain 8 weeks.
General notes	

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 or -80°C. Avoid freeze / thaw cycle.
Storage buffer	Preservative: 0.02% Sodium Azide Constituents: 1% BSA, PBS, pH 7.4
Purity	Immunogen affinity purified
Clonality	Polyclonal
Isotype	IgG

Applications

Our Abpromise guarantee covers the use of **ab18723** 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
WB	★★★★★	Use a concentration of 1 µg/ml. Detects a band of approximately 45 kDa (predicted molecular weight: 40-45 kDa).
ICC	★★★★★	Use at an assay dependent concentration. We recommend Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed (ab150081) secondary antibody.
ICC/IF	★★★★★	Use a concentration of 1 - 5 µg/ml.
IHC-FrFl	★★★★★	1/1000. Please note: Low dilutions of this antibody can cause high background. Please use as high a dilution as possible. Optimal working dilutions are batch dependent.
IHC-FoFr	★★★★★	1/100 - 1/2000.
IHC-Fr	★★★★☆	1/2000 - 1/7000.
IHC-P	★★★★☆	Use a concentration of 0.05 - 1 µg/ml. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

Target

Function

Seems to be required for initial steps of neuronal dispersion and cortex lamination during cerebral cortex development. May act by competing with the putative neuronal protein kinase DCAMKL1 in binding to a target protein. May in that way participate in a signaling pathway that is crucial for neuronal interaction before and during migration, possibly as part of a calcium ion-dependent signal transduction pathway. May be part with LIS-1 of an overlapping, but distinct, signaling pathways that promote neuronal migration.

Tissue specificity

Highly expressed in neuronal cells of fetal brain (in the majority of cells of the cortical plate, intermediate zone and ventricular zone), but not expressed in other fetal tissues. In the adult, highly expressed in the brain frontal lobe, but very low expression in other regions of brain, and not detected in heart, placenta, lung, liver, skeletal muscles, kidney and pancreas.

Involvement in disease

Defects in DCX are the cause of lissencephaly X-linked type 1 (LISX1) [MIM:300067]; also called X-LIS or LIS. LISX1 is a classic lissencephaly characterized by mental retardation and seizures that are more severe in male patients. Affected boys show an abnormally thick cortex with absent or severely reduced gyri. Clinical manifestations include feeding problems, abnormal muscular tone, seizures and severe to profound psychomotor retardation. Female patients display a less severe phenotype referred to as 'doublecortex'.
Defects in DCX are the cause of subcortical band heterotopia X-linked (SBHX) [MIM:300067]; also known as double cortex or subcortical laminar heterotopia (SCLH). SBHX is a mild brain malformation of the lissencephaly spectrum. It is characterized by bilateral and symmetric plates or bands of gray matter found in the central white matter between the cortex and cerebral ventricles, cerebral convolutions usually appearing normal.
Note=A chromosomal aberration involving DCX is found in lissencephaly. Translocation t(X;2)(q22.3;p25.1).

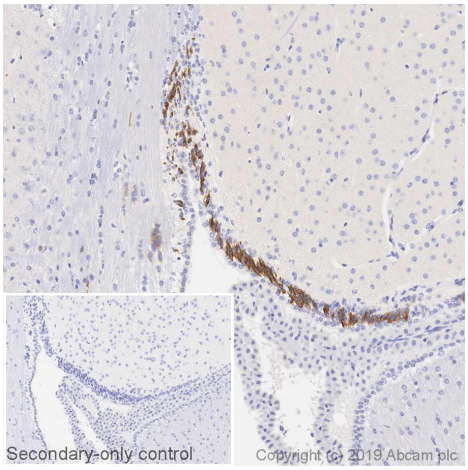
Sequence similarities

Contains 2 doublecortin domains.

Cellular localization

Cytoplasm.

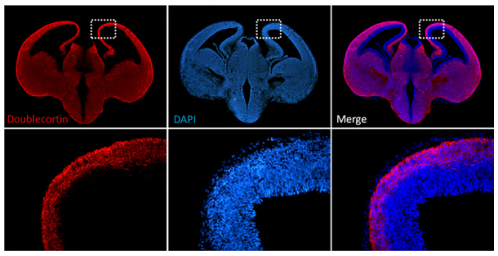
Images



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Doublecortin antibody (ab18723)

IHC image of ab18723 staining in Mouse 8 weeks brain formalin fixed paraffin embedded tissue section, performed on a Leica BOND™ system using the standard protocol F (with no post primary). The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab18723, 0.1µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. Secondary-only control image is shown as insert.

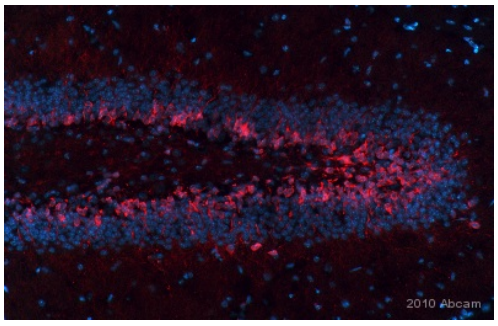
For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.



Immunohistochemistry (PFA perfusion fixed frozen sections) - Anti-Doublecortin antibody (ab18723)

This image is courtesy of an anonymous Abreview

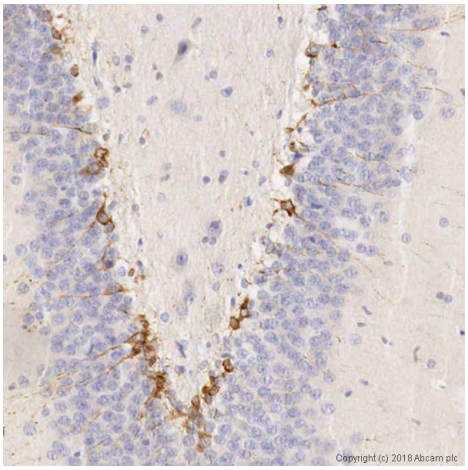
ab18723 staining Doublecortin in mouse embryonic 15 day brain tissue sections by Immunohistochemistry (PFA perfusion fixed frozen sections). Tissue samples were fixed by perfusion with paraformaldehyde, permeablized with 0.5% Triton X-100 in PBS, blocked with 10% serum for 1 hour at 25°C and antigen retrieval was by heat mediation in citrate buffer, pH 6. The sample was incubated with primary antibody (1/500 in PBS + 0.1% Triton X-100 + 1% serum) for 16 hours at 25°C. An Alexa Fluor® 594-conjugated donkey anti-rabbit IgG polyclonal (1/700) was used as the secondary antibody.



Immunohistochemistry (Frozen sections) - Anti-Doublecortin antibody (ab18723)

This image is courtesy of an anonymous abreview.

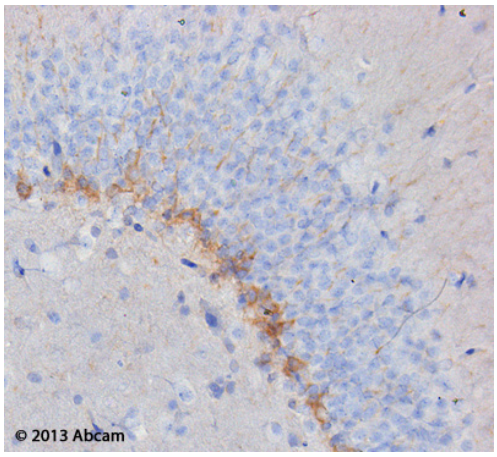
IHC-Fr image of Doublecortin staining in Mouse adult dentate gyrus sections using ab18723(1:100). The sections were fixed in paraformaldehyde and permeabilized using 1x TBST. The sections were then blocked using 10% donkey serum for 1 hour at 25°C. ab18723 was diluted 1:100 and incubated with the sections for 12 hours at 25°C. The secondary antibody used was Donkey anti-rabbit conjugated to Cy3 Dye (1:500). DAPI was used to stain the nuclei.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Doublecortin antibody (ab18723)

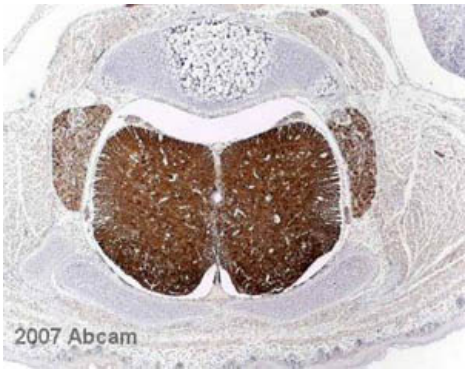
IHC image of ab18723 staining in rat 6 week brain formalin fixed paraffin embedded tissue section, performed on a Leica Bond™ system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6) for 20 mins. The section was then incubated with ab18723, 0.1µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Doublecortin antibody (ab18723)

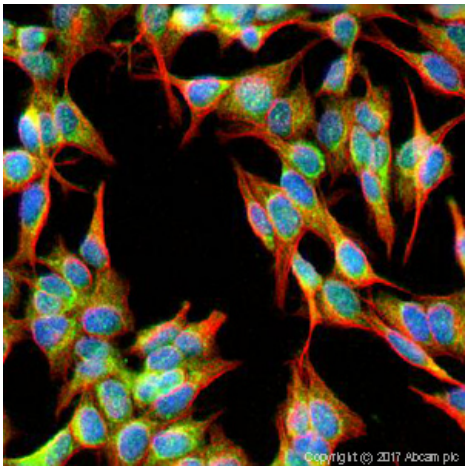
ab18723 staining 6 week rat brain tissue dentate gyrus (DG) by IHC-P using rabbit-specific EXPOSE IHC detection kit (ab80437). Formalin fixed paraffin embedded tissue sections were pre-treated using heat mediated antigen retrieval (using a pressure cooker) with sodium citrate buffer (pH6) for 30 mins. The section was incubated with ab18723, 0.1µg/ml, for 1 hour at room temperature. DAB was used as the chromogen and the section was counterstained with haematoxylin and mounted with DPX.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Doublecortin antibody (ab18723)

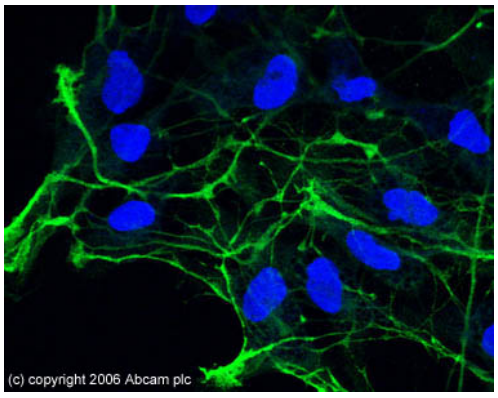
This image is courtesy of Carl Hobbs, King's College London, United Kingdom

ab18723 at 1/200 staining mouse E18 body T/S spinal cord tissue sections by IHC-P. The tissue was formaldehyde fixed and a heat mediated antigen retireval step was perfomed before incubation with the antibody for 24 hours. A biotinylated goat antibody was used as the secondary.



Immunocytochemistry/ Immunofluorescence - Anti-Doublecortin antibody (ab18723)

ab18723 stained in SHSY5Y cells. Cells were fixed with 100% methanol (5 min) at room temperature and incubated with PBS containing 10% goat serum, 0.3 M glycine, 1% BSA and 0.1% triton for 1h at room temperature to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody ab18723 at 5 µg/ml and ab7291 (Mouse monoclonal [DM1A] to alpha Tubulin - Loading Control) at 1/1000 dilution overnight at +4°C. The secondary antibodies were Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) (ab150120) secondary antibody (pseudo-colored red) and Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) preadsorbed (ab150081) secondary antibody (colored green) used at 1 ug/ml for 1hour at room temperature. DAPI was used to stain the cell nuclei (colored blue) at a concentration of 1.43µM for 1hour at room temperature.



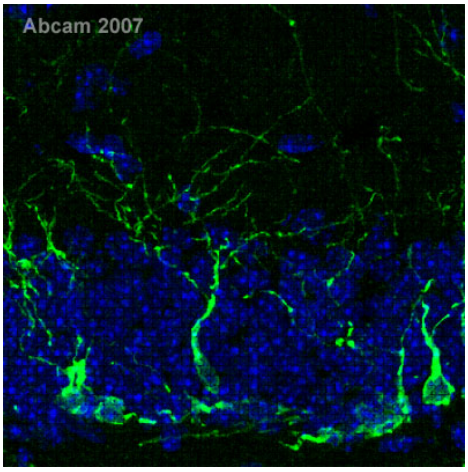
Immunocytochemistry/ Immunofluorescence - Anti-Doublecortin antibody (ab18723)

This image is courtesy of Randal Moldrich, CNRS UMR7637, ESPCI, France

Doublecortin antibody (ab18723; green) labeling cell extensions consistent with dendrite morphology in 4 day old cultures. Preincubation of ab18723 with its immunising peptide (ab19804) quenched immunostaining (see review).

Dorsal root ganglion explants were dissected from 16 day-old rat embryos and cultured for 4 days in vitro with Neurobasal Medium containing 10% fetal calf serum and B27 supplement. Immunocytochemistry: All steps were performed in PBS. Cells or explants were fixed in 4% PFA for 15min, permeabilised with 0.1% TX100 for 10min and blocked with 5% BSA, 0.1% TX100 for 45min. ab18723 was incubated at 5µg/ml for 12h in 5% BSA, 0.1% TX100 at 4°C. For peptide blocking experiments preincubation of the peptide (ab19804; 250µg/ml) and antibody (5µg/ml) was performed for 2h at 37°C. Cultures were washed (3x) of primary

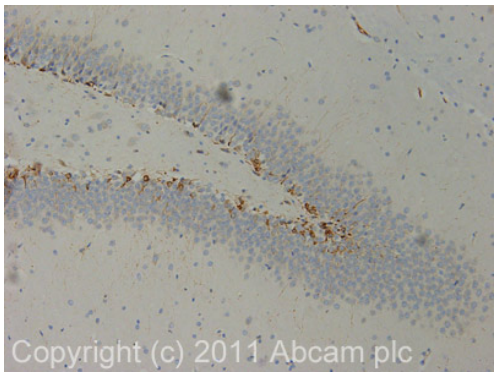
antibody solution. Goat anti-rabbit AlexaFluor 488 was used as secondary antibody (1/400) in 5% BSA, 0.1% TX100 for 2h at R



Immunohistochemistry (Frozen sections) - Anti-Doublecortin antibody (ab18723)

This image is courtesy of Francois Guillemot, NIMR, UK

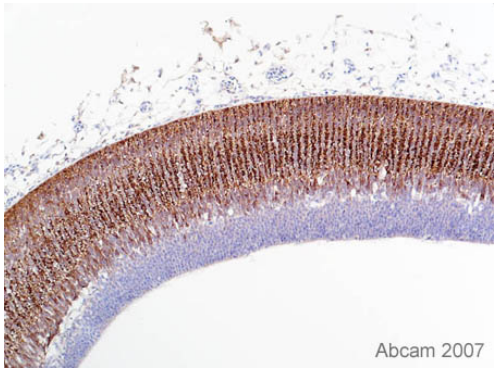
Doublecortin expression in the dentate gyrus of a 1 month-old mouse brain. Doublecortin staining using ab18723 (1/500) in the dentate gyrus of a 1 month-old mouse brain. The mouse has been perfused with paraformaldehyde 4% (50ml). After dissection, the brain has been incubated overnight in sucrose 20%, embedded in OCT and cryosectioned (10 µm). No antigen retrieval was used. The secondary antibody used was a non-Abcam Goat anti-rabbit Alexa488.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Doublecortin antibody (ab18723)

IHC image of ab18723 staining in rat 6 week brain formalin fixed paraffin embedded tissue section, performed on a Leica Bond™ system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6) for 20 mins. The section was then incubated with ab18723, 0.1µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

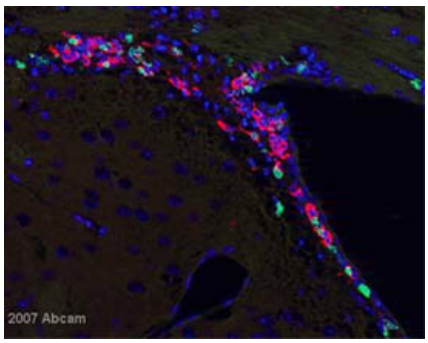
For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Doublecortin antibody (ab18723)

This image is courtesy of Carl Hobbs, King's College London, United Kingdom

Immunohistochemistical staining (on formaldehyde/PFA-fixed paraffin-embedded sections) of Doublecortin antibody - Neuronal Marker (ab18723) on Quail Tissue sections (E6/7 brain (Saggital section). Antigen retrieval step: Heat mediated. Blocking step: 1% BSA for 10 mins at RT. Primary Antibody ab18723 incubated at 1/400 for 2 hours RT. Secondary Antibody: Biotin conjugated goat anti rabbit Ig (1/300).

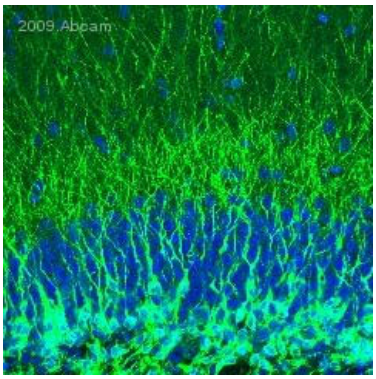


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Doublecortin antibody (ab18723)

This image is courtesy of Carl Hobbs, King's College London, United Kingdom

ab18723 at 1/2000 staining mouse brain svr: progenitor olfactory neurones by IHC-P. The tissue was formaldehyde fixed and a heat mediated antigen retrieval step was performed before incubation with the antibody for 16 hours. An Alexa-Fluor ® 488 conjugated goat antibody was used as the secondary (green). The tissue was also stained for Ki67 (shown in red).

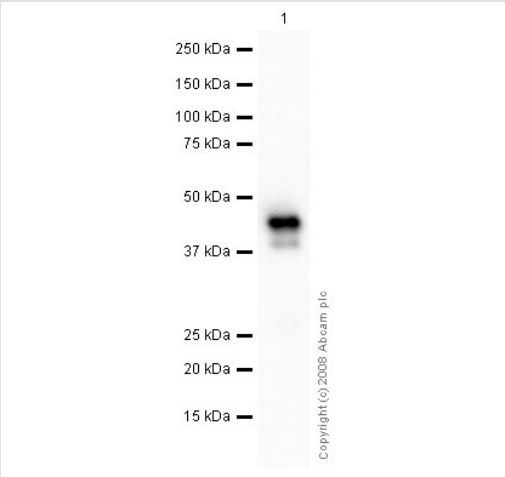
The MIP image was derived from Apotome-generated Z-stacks from the greyscale image of each of the channels in the MIP.



Immunohistochemistry (PFA perfusion fixed frozen sections) - Anti-Doublecortin antibody (ab18723)

This image is courtesy of an anonymous Abreview

ab18723 staining Doublecortin in mouse dentate gyrus hippocampal tissue section by Immunohistochemistry (PFA perfusion fixed frozen sections). Tissue samples were incubated with primary antibody 1/1000 for 16 hours at 4°C. An Alexa Fluor®488 conjugated goat polyclonal to rabbit IgG at 1/400 dilution was used as secondary.



Western blot - Anti-Doublecortin antibody (ab18723)

Anti-Doublecortin antibody (ab18723) at 1 µg/ml + Mouse brain tissue lysate - total protein (0 days) (ab7188) at 10 µg

Secondary

Goat polyclonal to Rabbit IgG - H&L - Pre-Adsorbed (HRP) at 1/3000 dilution

Performed under reducing conditions.

Predicted band size: 40-45 kDa

Observed band size: 45 kDa

why is the actual band size different from the predicted?

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