



## Mouse monoclonal antibody to Nestin [4D11] (317-630)

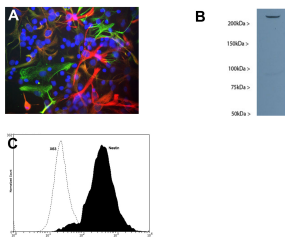
<b>Catalogue No.:</b>	M-1385-100
<b>Description:</b>	Nestin is a member of the class IV intermediate filament protein family which is expressed in neuronal stem cells. The molecular weight of human Nestin as determined by SDS-PAGE mobility is about 240kDa. However the real molecular weight is considerably less than this, at 177kDa, the disparity being likely due to the highly charged region of the C-terminal segment. Nestin is relatively poorly conserved in protein sequence across species boundaries, so that the mouse and human proteins have an overall identity of only 62%. As a result antibodies to the human protein often fail to recognize the rodent homologue and vice versa. However this antibody stains both rodent and human Nestin. Antibodies to Nestin are widely used to identify neural stem cells.
<b>Batch No.:</b>	See product label
<b>Unit size:</b>	100 uL
<b>Antigen:</b>	Partial segment (region 317-630 aa) of human Nestin expressed in E.coli
<b>Antigen Location:</b>	317-630
<b>Antibody Type:</b>	Monoclonal
<b>Isotype:</b>	IgG
<b>Clone:</b>	4D11
<b>Other Names:</b>	Nestin; NES;
<b>Accession:</b>	P48681 NEST_HUMAN;
<b>Produced in:</b>	Mouse, IgG1 kappa
<b>Purity:</b>	Ascites fluid
<b>Applications:</b>	Western Blotting (WB), Immunocytochemistry (IC) and Flow Cytometry. Suggested dilution for WB is 1:1,000-5,000 and 1:250-500 for IC. Use 2ug/10 <sup>6</sup> cells for Flow Cytometry. Biosensis recommends optimal dilutions/concentrations should be determined by the end user.
<b>Comments:</b>	Ready-to-use reagent for in-vitro laboratory research use only.
<b>Specificity:</b>	This antibody is specific for the 240 kDa Nestin protein by WB on developing rat brain (P18) homogenate. A much weaker band at approx. 90 kDa may also be seen. This is suggested to be a breakdown product of the 240 kDa band.
<b>Antibody Against:</b>	Nestin
<b>Cross-reactivity:</b>	Human, Rodent
<b>Form:</b>	Lyophilised from ascites fluid.
<b>Appearance:</b>	White powder
<b>Reconstitution:</b>	Reconstitute in sterile distilled water. Centrifuge to remove any insoluble material.
<b>Storage:</b>	After reconstitution of lyophilised antibody, aliquot and store at -20C for a higher stability. Avoid freeze-thaw cycles.
<b>Expiry Date:</b>	12 months after purchase
<b>Specific References:</b>	Schomann T, Mezzanotte L, De Groot JCMJ, Rivolta MN, Hendriks SH, Frijns JHM, Huisman

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MA (2017) Neuronal differentiation of hair-follicle-bulge-derived stem cells co-cultured with mouse cochlear modiolus explants. PLoS One. 12(10):e0187183. Application: ICC/IF; Species: Mouse, Hair follicle bulge-derived neural crest-derived stem cells (HFBCs).

Gho CG, Schomann T, de Groot SC, Frijns JH, Rivolta MN, Neumann MH, Huisman MA.(2015) Isolation, expansion and neural differentiation of stem cells from human plucked hair- a further step towards autologous nerve recovery. Cytotechnology In press. Application: IF; Species: Human, Hair follicle bulge-derived neural crest-derived stem cells (HFBCs), Keywords: Hair follicle stem cell, Regeneration, Neural crest, Neuron, Glia, Cryopreservation



A: Mixed cultures of neonatal rat neurons and glia stained with Mouse monoclonal antibody to Nestin [4D11] M-1385-100 (red), Chicken polyclonal antibody to vimentin C-1409-50 (green) and DNA (DAPI stain, blue). Astrocytes and neuronal stem cells stain strongly and specifically in a clearly filamentous fashion with the Nestin antibody. The filamentous staining pattern is as expected as both Nestin and Vimentin are components of 10nm filaments. Note that some cells contain Nestin, but do not stain strongly for Vimentin and so appear red. Others contain Vimentin and not Nestin and so appear green- these are likely to be fibroblastic or endothelial cells. Some cells express both proteins and so appear yellowish. The presence of Nestin indicates that the cells are developing astrocytes, neuroblasts or undifferentiated neural stem cells. B: Western blot in of developing rat brain (P18) homogenate probed with mouse monoclonal antibody to Nestin M-1385-100. A single strong band running at ~240kDa is seen. C: Analysis of nesting expression in human neuroblastoma SH-SY5Y by Flow Cytometry. Fixing and Permeabilization of cells: Absolute methanol (10 minutes in ice) and 0.1% Tween-20 in PBS, Blocking: 1% BSA, Primary antibody: Mouse Monoclonal antibody to Nestin (cat # M-1385-100, 2µg per  $\sim 10^6$  cells) for 30 minutes at room temperature, Secondary antibody: Goat anti-mouse PE labeled secondary antibody (1:100 fold dilution) with incubation for 20 minutes in dark at room temperature. Non-specific Control IgG, clone X63 (cat # M-1249-200) was used as negative control under same conditions (black dashed). Flow cytometry data and results were generated using Orflo Moxiflow™ instrument and protocols.

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