Technical Data Sheet

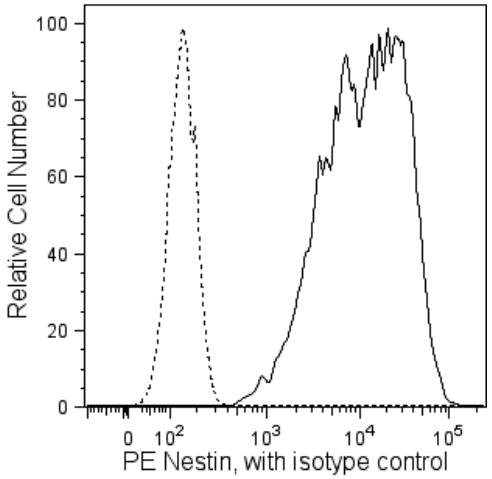
PE Mouse anti-Nestin

Product Information

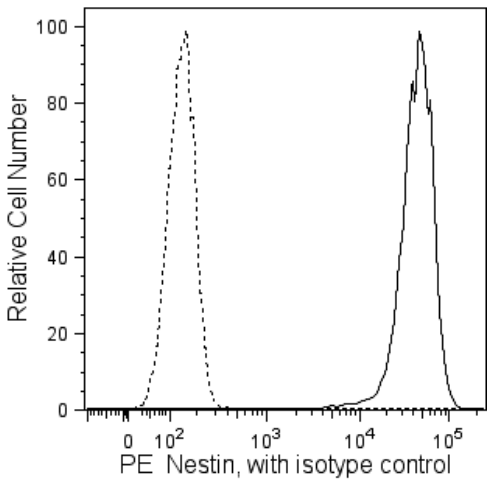
Material Number:	**561230**
Size:	50 tests
Vol. per Test:	5 µl
Clone:	25/NESTIN
Immunogen:	Rat Nestin aa. 402-604 Recombinant Protein
Isotype:	Mouse IgG1, κ
Reactivity:	QC Testing: Rat Tested in Development: Human
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The cytoskeleton consists primarily of core structural proteins that include microfilaments, microtubules, and intermediate filaments (IFs). IFs contain more than 50 distinct proteins that are organized into six different subtypes: Type I/II keratins expressed in epithelia, type III vimentin/desmin, type IV neurofilament proteins, type V nuclear lamins, and type VI nestin expressed primarily in embryonic cells. Nestin has a conserved core region (amino acids 7 to 314), which contains an α helical domain that is involved in coiled-coil assembly of IFs. The C-terminal region of nestin is similar to type IV IFs, since it contains highly charged amino acids, many glutamate residues, and an 11 amino acid repeat motif. Nestin is expressed in the cerebrum during embryonic development, in the cerebellum during early postnatal development, and in dermatomal cells and myoblasts during myogenesis. In vitro, nestin forms homodimers and homotetramers, but not IFs, and can co-assemble with type III vimentin and type IV internexin proteins. Thus, nestin is a core IF protein that is essential for proper cytoskeletal formation during neurogenesis and myogenesis.



Analysis of Nestin expression in rat glioma. *C6 cells (ATCC, Cat. No. CCL-107) were fixed (BD Cytofix™ buffer, Cat. No. 554655) for 20 minutes at room temperature, permeabilized with BD Phosflow™ Perm Buffer I (Cat. No. 557885), and then stained with either PE Mouse anti-Nestin (solid line) or PE Mouse IgG1, κ Isotype Control (Clone MOPC-21, Cat. No. 554680, dashed line). Flow cytometry was performed on a BD LSR™ II flow cytometry system. This antibody conjugate is also compatible with BD Phosflow™ Perm Buffers II and III.*



Analysis of Nestin expression in human Neural Stem Cells (NSC). *NSC were derived from H9 human embryonic stem cells (WiCell, Wisconsin) and grown for 8 passages, fixed (BD Cytofix™ buffer, Cat. No. 554655) for 20 minutes at room temperature, permeabilized with BD Phosflow™ Perm Buffer I (Cat. No. 557885), and then stained with either PE Mouse anti-Nestin (solid line) or PE Mouse IgG1, κ Isotype Control (Clone MOPC-21, Cat. No. 554680, dashed line). Flow cytometry was performed on a BD LSR™ II flow cytometry system. This antibody conjugate is also compatible with BD Phosflow™ Perm Buffers II and III.*

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.
The antibody was conjugated with R-PE under optimum conditions, and unconjugated antibody and free PE were removed.
Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

BD Biosciences

bdbiosciences.com

United States	Canada	Europe	Japan	Asia Pacific	Latin America/Caribbean
877.232.8995	888.259.0187	32.53.720.550	0120.8555.90	65.6861.0633	55.11.5185.9995

For country-specific contact information, visit **bdbiosciences.com/how_to_order/**

Application Notes

Application

Intracellular staining (flow cytometry)	Routinely Tested

Recommended Assay Procedure:

This antibody conjugate is suitable for intracellular staining of human and rat cell lines using BD Cytofix™ Fixation Buffer and the BD Phosflow™ Permeabilization Buffers: Perm/Wash Buffer I, Perm Buffer II, or Perm Buffer III (see Suggested Companion Products).

Suggested Companion Products

Catalog Number	Name	Size	Clone
554680	PE Mouse IgG1, κ Isotype Control	0.1 mg	MOPC-21
554655	Fixation Buffer	100 ml	(none)
557885	Perm/Wash Buffer I	125 ml	(none)
558052	Perm Buffer II	125 ml	(none)
558050	Perm Buffer III	125 ml	(none)

Product Notices

1. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
2. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1 × 10^6 cells in a 100-µl experimental sample (a test).
3. An isotype control should be used at the same concentration as the antibody of interest.
4. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
5. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
6. Source of all serum proteins is from USDA inspected abattoirs located in the United States.

References

Jin K, Zhu Y, Sun Y, Mao XO, Xie L, Greenberg DA. Vascular endothelial growth factor (VEGF) stimulates neurogenesis in vitro and in vivo. *Proc Natl Acad Sci U S A.* 2002; 99(18):11946-11950. (Clone-specific: Immunofluorescence)

Kachinsky AM, Dominov JA, Miller JB. Myogenesis and the intermediate filament protein, nestin. *Dev Biol.* 1994; 165(1):216-228. (Biology)

Kernie SG, Erwin TM, Parada LF. Brain remodeling due to neuronal and astrocytic proliferation after controlled cortical injury in mice. *J Neurosci Res.* 2001; 66(3):317-326. (Clone-specific: Immunofluorescence)

Lendahl U, Zimmerman LB, McKay RD. CNS stem cells express a new class of intermediate filament protein. *Cell.* 1990; 60(4):585-595. (Biology)

Steinert PM, Chou YH, Prahlad V, et al. A high molecular weight intermediate filament-associated protein in BHK-21 cells is nestin, a type VI intermediate filament protein. Limited co-assembly in vitro to form heteropolymers with type III vimentin and type IV alpha-internexin. *J Biol Chem.* 1999; 274(14):9881-9890. (Biology)

Wu D, Tadano M, Edamatsu H, et al. Neuronal lineage-specific induction of phospholipase Cepsilon expression in the developing mouse brain. *Eur J Neurosci.* 2003; 17(8):1571-1580. (Clone-specific: Immunofluorescence)