Technical Data Sheet

PerCP-Cy[™]5.5 Mouse Anti-Human CD324 (E-Cadherin)

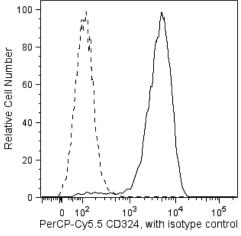
Product Information

Immunogen:Human Breast Tumor Cell LineIsotype:Mouse (BALB/c) IgG1, κ Reactivity:QC Testing: Human

Storage Buffer: Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The 67A4 monoclonal antibody recognizes the extracellular domain of human E-Cadherin (CD324). E-Cadherin is a 120-kDa transmembrane glycoprotein that is localized in the adherens junctions of epithelial cells. There it interacts with the cytoskeleton through the associated cytoplasmic catenin proteins. In addition to being a calcium-dependent adhesion molecule, E-Cadherin is also a critical regulator of epithelial junction formation. Its association with catenins is necessary for cell-to-cell adhesion. These E-Cadherin/catenin complexes associate with cortical actin bundles at both the zonula adherens and the lateral adhesion plaques. Tyrosine phosphorylation can disrupt these complexes, leading to changes in cell adhesion properties. E-Cadherin expression is often down-regulated in highly invasive, poorly differentiated carcinomas. Increased expression of E-Cadherin in these cells reduces their invasiveness. Thus, loss of expression or function of E-Cadherin appears to be an important step in tumorigenic progression. Pluripotent stem cells express E-Cadherin. Upon differentiation, an epithelial to mesenchymal transition results in the loss of E-cadherin expression and a gain in the expression of N-cadherin.



Flow cytometric analysis of CD324 (E-cadherin) expression in human embryonic stem (ES) cells. H9 human ES cells (WiCell, Madison, WI) were harvested with Cell Dissociation Buffer (Life Technologies). Please note that the epitope is sensitive to Accutase™ detachment. The cells were stained with either PerCP-Cy™5.5 Mouse anti-Human CD324 antibody (Cat. No. 563573, solid line) or PerCP-Cy™5.5 Mouse IgG1, κ Isotype Control (Cat. No. 550795, dashed line) at matched concentrations. Histograms were derived from gated events based on light scattering characteristics of H9 human ES cells. Flow cytometry was performed on a BD LSRFortessa™ Flow Cytometry System.

Preparation and Storage

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated with PerCP-Cy5.5 under optimum conditions, and unconjugated antibody and free PerCP-Cy5.5 were removed. Storage of PerCP-Cy5.5 conjugates in unoptimized diluent is not recommended and may result in loss of signal intensity.

Application Notes

Application

Flow cytometry Routinely Tested	

Suggested Companion Products

Catalog Number	Name	Size	Clone	
550795	PerCP-Cy TM 5.5 Mouse IgG1 κ Isotype Control	0.1 mg	MOPC-21	
554656	Stain Buffer (FBS)	500 ml	(none)	

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Product Notices

- 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).
- Source of all serum proteins is from USDA inspected abattoirs located in the United States. 2
- An isotype control should be used at the same concentration as the antibody of interest. 3.
- 4. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- Cy is a trademark of Amersham Biosciences Limited. This conjugated product is sold under license to the following patents: US Patent Nos. 5,486,616; 5,569,587; 5,569,766; 5,627,027.
- Please observe the following precautions: Absorption of visible light can significantly alter the energy transfer occurring in any tandem fluorochrome conjugate; therefore, we recommend that special precautions be taken (such as wrapping vials, tubes, or racks in aluminum foil) to prevent exposure of conjugated reagents, including cells stained with those reagents, to room illumination.
- 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.
- PerCP-Cy5.5-labelled antibodies can be used with FITC- and R-PE-labelled reagents in single-laser flow cytometers with no significant spectral overlap of PerCP-Cy5.5, FITC, and R-PE fluorescence.
- PerCP-Cy5.5 is optimized for use with a single argon ion laser emitting 488-nm light. Because of the broad absorption spectrum of the tandem fluorochrome, extra care must be taken when using dual-laser cytometers, which may directly excite both PerCP and Cy5.5TM. We recommend the use of cross-beam compensation during data acquisition or software compensation during data analysis.
- For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
- 11. This product is subject to proprietary rights of Amersham Biosciences Corp. and Carnegie Mellon University and made and sold under license from Amersham Biosciences Corp. This product is licensed for sale only for research. It is not licensed for any other use. If you require a commercial license to use this product and do not have one return this material, unopened to BD Biosciences, 10975 Torreyana Rd, San Diego, CA 92121 and any money paid for the material will be refunded.
- Accutase is a registered trademark of Innovative Cell Technologies, Inc.

Behrens J, Vakaet L, Friis R, et al. Loss of epithelial differentiation and gain of invasiveness correlates with tyrosine phosphorylation of the E-cadherin/beta-catenin complex in cells transformed with a temperature-sensitive v-SRC gene. J Cell Biol. 1993; 120(3):757-766. (Biology)

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Cepek KL, Shaw SK, Parker CM, et al. Adhesion between epithelial cells and T lymphocytes mediated by E-cadherin and the aEb7 integrin. Nature. 1994; 372:190-193. (Biology)

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Takeichi M. The cadherins: cell-cell adhesion molecules controlling animal morphogenesis. Development. 1988; 102(4):639-655. (Biology)

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