

Technical Data Sheet

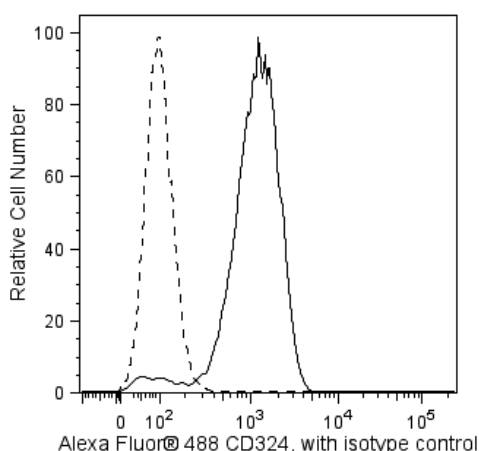
Alexa Fluor® 488 Mouse Anti-Human CD324 (E-Cadherin)

Product Information

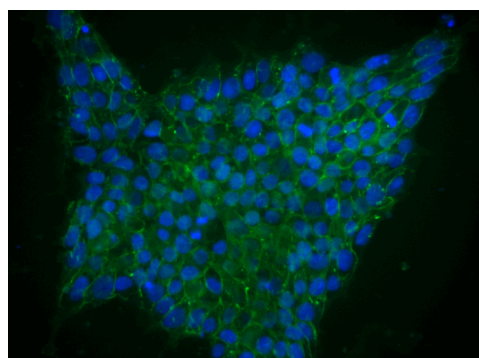
Material Number:	563570
Alternate Name:	E-cadherin
Size:	50 tests
Vol. per Test:	5 µl
Clone:	67A4
Immunogen:	Human Breast Tumor Cell Line
Isotype:	Mouse (BALB/c) IgG1, κ
Reactivity:	QC Testing: Human
Storage Buffer:	Aqueous buffered solution containing BSA, protein stabilizer, and ≤0.09% sodium azide.

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 Alexa Fluor® 488 Mouse anti-Human CD324 antibody (Cat. No. 563570, solid line) or Alexa Fluor® 488 Mouse IgG1, κ Isotype Control (Cat. No. 557782, 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.



Immunofluorescent Staining of CD324 (E-Cadherin) on human embryonic stem (ES) cells. H9 human ES cells (WiCell, Madison, WI) grown in mTESR™ 1 media (Stem Cell Technologies) were fixed with BD Cytofix™ fixation buffer (Cat. No. 554655). Cells were stained with Alexa Fluor® 488 Mouse Anti-Human CD324 (E-Cadherin) monoclonal antibody (Cat. No. 563570, pseudo-colored green) at 5 µg/mL. Counter-staining was with DAPI (pseudo-colored blue). The images were captured on a BD Pathway™ 435 Cell Analyzer and merged using BD AttoVision™ Software.

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 to Alexa Fluor® 488 under optimum conditions, and unreacted Alexa Fluor® 488 was removed.

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Application Notes

Application

Flow cytometry	Routinely Tested
Bioimaging	Tested During Development
Immunofluorescence	Tested During Development

Suggested Companion Products

Catalog Number	Name	Size	Clone
557782	Alexa Fluor® 488 Mouse IgG1 κ Isotype Control	50 tests	MOPC-21
554655	Fixation Buffer	100 ml	(none)

Product Notices

1. 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).
2. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
3. An isotype control should be used at the same concentration as the antibody of interest.
4. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
5. The Alexa Fluor®, Pacific Blue™, and Cascade Blue® dye antibody conjugates in this product are sold under license from Molecular Probes, Inc. for research use only, excluding use in combination with microarrays, or as analyte specific reagents. The Alexa Fluor® dyes (except for Alexa Fluor® 430), Pacific Blue™ dye, and Cascade Blue® dye are covered by pending and issued patents.
6. Alexa Fluor® 488 fluorochrome emission is collected at the same instrument settings as for fluorescein isothiocyanate (FITC).
7. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
8. 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.
9. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
10. mTESR™1 is a trademark of StemCell Technologies.
11. Accutase is a registered trademark of Innovative Cell Technologies, Inc.

References

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

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