# **Technical Data Sheet**

# Purified Mouse Anti-Human CD324 (E-Cadherin)

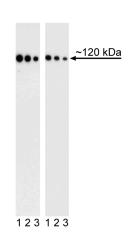
#### **Product Information**

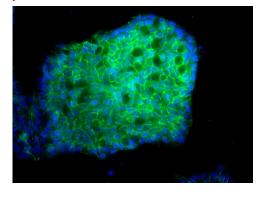
Target MW: ~120 kDa

**Storage Buffer:** Aqueous buffered solution containing ≤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.





Western blot analysis of CD324 (E-cadherin) expression in human breast adenocarcinoma and human embryonic stem (ES) cells. Cell Lysates from a human breast adenocarcinoma cell line MCF-7 (ATCC HTB-22, left blot) and H9 human ES Cells (WiCell, Madison, WI, right blot) were probed with Purified Mouse Anti-Human CD324 (E-Cadherin) monoclonal antibody at titrations of 0.125 (lanes 1), 0.063 (lanes 2), 0.032 (lanes 3) µg/ml. Proteins were detected using HRP Goat Anti-Mouse Ig (Cat. No. 554002). CD324 (E-cadherin) is identified as a band of ~120 kDa in MCF7 and Human ES cells

Immunoflourescent staining of CD324 (E-cadherin) on human embryonic stem (ES) cells. H9 human ES cells (WiCell, Madison, WI) passage 45 grown in mTESR™1 media (StemCell Technologies) on BD Matrigel™ hESC-qualified Matrix (Cat. No. 354277) were fixed with BD Cytofix™ Fixation Buffer (Cat. No. 554655). Cells were stained with Purified Mouse Anti-Human CD324 (E-Cadherin) monoclonal antibody (pseudo-colored green) at 2.5 µg/ml. The second-step reagent was Alexa Fluor® 488 goat anti-mouse Ig (Life Technologies), and cell nuclei were stained 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**

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography. Store undiluted at 4°C.

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## **Application Notes**

#### Application

PT ·····	
Western blot	Routinely Tested
Flow cytometry	Tested During Development
Bioimaging	Tested During Development
Immunofluorescence	Tested During Development

## **Suggested Companion Products**

Catalog Number	Name	Size	Clone	
554002	HRP Goat Anti-Mouse Ig	1.0 ml	(none)	
562870	PE Mouse Anti-Human CD324 (E-Cadherin)	100 tests	67A4	
554655	Fixation Buffer	100 ml	(none)	

#### **Product Notices**

- Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- 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.
- Sodium azide is a reversible inhibitor of oxidative metabolism; therefore, antibody preparations containing this preservative agent must not be used in cell cultures nor injected into animals. Sodium azide may be removed by washing stained cells or plate-bound antibody or dialyzing soluble antibody in sodium azide-free buffer. Since endotoxin may also affect the results of functional studies, we recommend the NA/LE (No Azide/Low Endotoxin) antibody format, if available, for in vitro and in vivo use.
- Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
- All other brands are trademarks of their respective owners. 6

#### 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)

Bühring HJ, Müller T, Herbst R, et al. The adhesion molecule E-cadherin and a surface antigen recognized by the antibody 9C4 are selectively expressed on erythroid cells of defined maturational stages, Leukemia, 1996; 10(1):106-116, (Clone-specific; Flow cytometr)

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)

D'Amour KA, Agulnick AD, Eliazer S, Kelly OG, Kroon E, Baetge EE. Efficient differentiation of human embryonic stem cells to definitive endoderm. Nat Biotechnol. 2005; 23(12):1534-1541. (Biology)

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

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