

## Technical Data Sheet

## BV421 Rat Anti-Mouse CD326

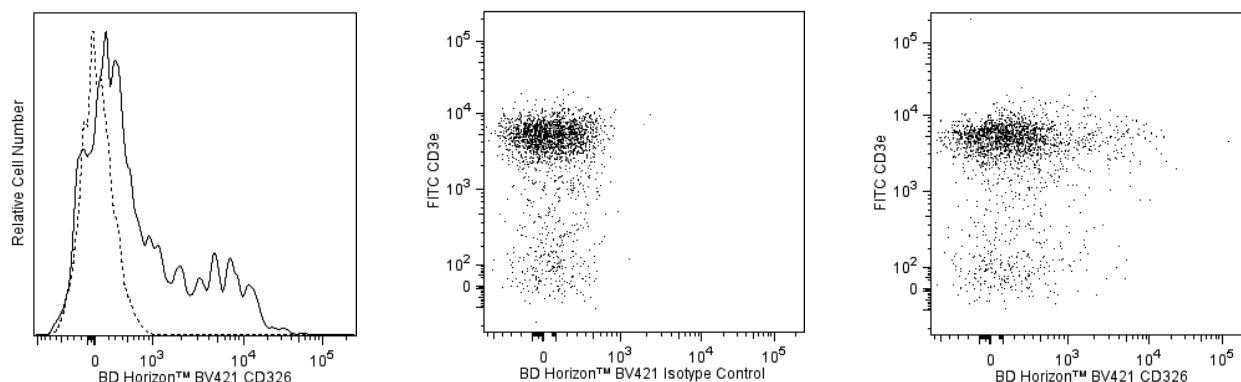
## Product Information

<b>Material Number:</b>	<b>563214</b>
<b>Alternate Name:</b>	Ep-CAM; EGP; EGP-2; Egp314; GA733-2; TROP1; Tacsd1; Tacstd1; Ly74; gp40
<b>Size:</b>	50 µg
<b>Concentration:</b>	0.2 mg/ml
<b>Clone:</b>	G8.8
<b>Immunogen:</b>	Glycoconjugates from BALB/c mouse-derived TE-71 medullary thymic epithelial cell line
<b>Isotype:</b>	Rat (SD) IgG2a, κ
<b>Reactivity:</b>	QC Testing: Mouse
<b>Storage Buffer:</b>	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

## Description

The G8.8 antibody reacts with CD326/Ep-CAM (*Epithelial Cell Adhesion Molecule*), also known as gp40 in the mouse and by a variety of names (including GA733-2, CO17-1A, and EGP) in the human. In the mouse, Ep-CAM is a 40-42 kDa cell-surface type 1 transmembrane glycoprotein expressed on thymic epithelial cells, thymic dendritic cells, immature thymocytes, a small subset of peripheral T lymphocytes, intestinal epithelium, kidney-collecting tubule epithelium, keratinocytes, Langerhans cells and lymph node and splenic dendritic cells. Profiles of Ep-CAM expression on fetal thymocytes and on the CD4[-] CD8[-], CD4[+] CD8[+], CD4[-] CD8[+], and CD4[+] CD8[-] subsets of adult thymocytes have been published. In unrelated studies, mouse Ep-CAM mRNA was detected in tissues containing epithelial cells (kidney, stomach, intestine, lung, and thymus) and in plasma cells and plasmacytomas, but not in heart, muscle, liver, brain, spleen, B lymphomas, or pre-B lymphomas. Ep-CAM is a Ca<sup>2+</sup> independent homophilic adhesion molecule that is proposed to play roles in the development and normal function of epithelial tissues and in the progression of carcinomas.

The antibody was conjugated to BD Horizon™ BV421 which is part of the BD Horizon™ Brilliant Violet™ family of dyes. With an Ex Max of 407-nm and Em Max at 421-nm, BD Horizon™ BV421 can be excited by the violet laser and detected in the standard Pacific Blue™ filter set (eg, 450/50-nm filter). BD Horizon™ BV421 conjugates are very bright, often exhibiting a 10 fold improvement in brightness compared to Pacific Blue™ conjugates.



**Multicolor analysis of CD326 expression on mouse thymocytes and splenic T lymphocytes.** BALB/c mouse thymocytes and splenic leucocytes were preincubated with Purified Rat Anti-Mouse CD16/CD32 antibody (Mouse BD Fc Block™) (Cat. No. 553141/553142) and stained with BD Horizon™ BV421 Rat IgG2a, κ Isotype Control (Cat. No. 562602) or BD Horizon™ BV421 Rat Anti-Mouse CD326 antibody (Cat. No. 563214).

**Left Panel:** Thymocytes were further stained with FITC Rat Anti-Mouse CD4 (553047/553046/561835) and PE Rat Anti-Mouse CD8a (553033/553032/561095) antibodies. The CD326 (solid line) and Ig Isotype Control (dashed line) fluorescence histograms were derived from CD4- and CD8-negative gated events with the forward and side light-scatter characteristics of viable thymocytes.

**Middle and Right Panels:** The splenic leucocytes were further stained with FITC Hamster Anti-Mouse CD3e (Cat. No. 553062/553061/561827) and PE Rat Anti-Mouse CD25 (Cat. No. 553866/561065) antibodies. Two-color flow cytometric dot plots show the correlated expression patterns of Ig Isotype control staining (Middle Plot) or CD326 (Right Plot) versus CD3e for CD25+ gated events with the forward and side light-scatter characteristics of viable splenic leucocytes. A small population of CD3+CD25+CD326+ cells were detected (Right Panel), whereas the CD25- T cells do not express detectable levels of CD326 (data not shown).

Flow cytometric analysis was performed using a BD™ LSR II Flow Cytometer System.

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## 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 BD Horizon™ BV421 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV421 were removed.

## Application Notes

### Application

Flow cytometry

Routinely Tested

## Suggested Companion Products

Catalog Number	Name	Size	Clone
554656	Stain Buffer (FBS)	500 ml	(none)
562602	BV421 Rat IgG2a, κ Isotype Control	50 µg	R35-95
555899	Lysing Buffer	100 ml	(none)
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.1 mg	2.4G2
553142	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.5 mg	2.4G2
553062	FITC Hamster Anti-Mouse CD3e	0.5 mg	145-2C11
553061	FITC Hamster Anti-Mouse CD3e	0.1 mg	145-2C11
561827	FITC Hamster Anti-Mouse CD3e	25 µg	145-2C11
553866	PE Rat Anti-Mouse CD25	0.2 mg	PC61
561065	PE Rat Anti-Mouse CD25	25 µg	PC61
553047	FITC Rat Anti-Mouse CD4	0.5 mg	RM4-5
553046	FITC Rat Anti-Mouse CD4	0.1 mg	RM4-5
561835	FITC Rat Anti-Mouse CD4	25 µg	RM4-5
553033	PE Rat Anti-Mouse CD8a	0.2 mg	53-6.7
553032	PE Rat Anti-Mouse CD8a	0.1 mg	53-6.7
561095	PE Rat Anti-Mouse CD8a	25 µg	53-6.7

## Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
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/pharming/en/protocols](http://www.bdbiosciences.com/pharming/en/protocols) for technical protocols.
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. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at [www.bdbiosciences.com/colors](http://www.bdbiosciences.com/colors).
7. Pacific Blue™ is a trademark of Molecular Probes, Inc., Eugene, OR.
8. Brilliant Violet™ 421 is a trademark of Sirigen.

## References

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Farr A, Nelson A, Truex J, Hosier S. Epithelial heterogeneity in the murine thymus: a cell surface glycoprotein expressed by subcapsular and medullary epithelium. *J Histochem Cytochem.* 1991; 39(5):345-353. (Immunogen: Electron microscopy, Immunohistochemistry, Immunoprecipitation)

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