

Technical Data Sheet

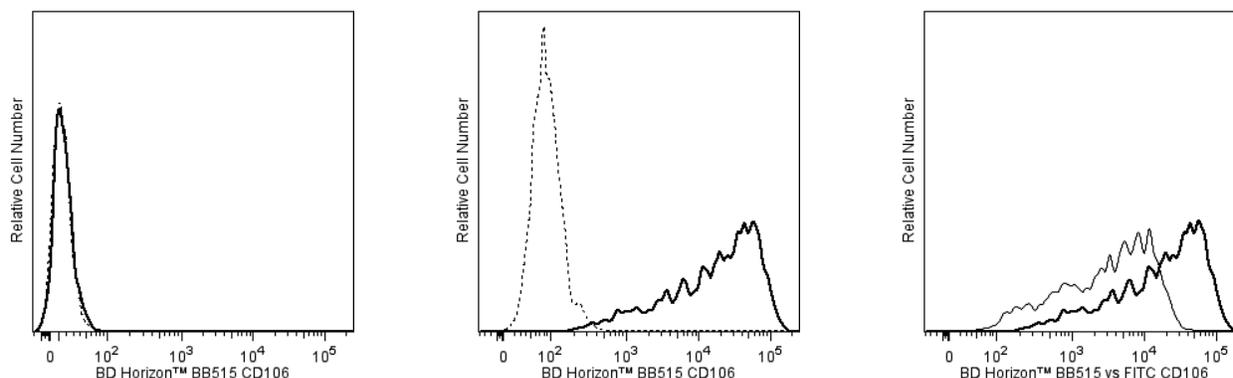
BB515 Mouse Anti-Human CD106**Product Information**

Material Number:	565417
Alternate Name:	VCAM-1; Vascular cell adhesion protein 1; INCAM-100; L1CAM
Size:	50 Tests
Vol. per Test:	5 µl
Clone:	51-10C9
Immunogen:	Human VCAM-1 Recombinant Protein
Isotype:	Mouse IgG1, κ
Reactivity:	QC Testing: Human
Workshop:	V E112
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The 51-10C9 monoclonal antibody specifically binds to CD106. CD106 is a 100-110 kDa type I transmembrane sialoglycoprotein that is also known as Vascular cell adhesion molecule-1 (VCAM-1) and INCAM-110. CD106 is expressed at high levels on the surface of cytokine-stimulated endothelium, and at minimal levels on unstimulated endothelium. VCAM-1 serves as a ligand for the leukocyte integrins $\alpha 4\beta 1$ (CD49d/CD29 complex; VLA-4) and $\alpha 4\beta 7$ (LPAM-1). The 51-10C9 monoclonal antibody inhibits the in vitro binding of lymphocytes and monocytes to VCAM-1 on stimulated endothelium.

The antibody was conjugated to BD Horizon BB515 which is part of the BD Horizon Brilliant™ Blue family of dyes. With an Ex Max near 490 nm and an Em Max near 515 nm, BD Horizon BB515 can be excited by the blue laser (488 nm) laser and detected with a 530/30 nm filter. This dye has been exclusively developed by BD Biosciences and is up to seven times brighter than FITC with less spillover into the PE channel. Due to similar excitation and emission properties, BB515, FITC, and Alexa Fluor® 488 cannot be used simultaneously. It is not recommended to use BB515 in cocktails that include Streptavidin conjugates as it may cause high background.



Flow cytometric analysis of CD106 expression on HUVEC cells - Staining comparisons between BD Horizon™ BB515- and FITC-conjugated antibodies. Human Umbilical Vein Endothelial Cells (HUVEC) were either left untreated (Left Panel) or cultured (24 hours at 37°C) with Recombinant Human TNF protein (Cat. No. 554618; 20 ng/ml; Middle and Right Panels). The cells were then stained with either BD Horizon™ BB515 Mouse IgG1, κ Isotype Control (Cat. No. 564416; dashed line histograms), or BD Horizon BB515 Mouse Anti-Human CD106 antibody (Cat. No. 565417; bold solid line histograms). Alternatively, cells were stained with FITC Mouse Anti-Human CD106 antibody (Cat. No. 551146; thin solid line histogram).

Overlaid histograms are shown to facilitate staining comparisons between: BB515 Anti-CD106 antibody versus its Ig Isotype Control (Left and Middle Panels), and BB515 Anti-CD106 antibody versus FITC Anti-CD106 antibody (Right Panel). The fluorescence histograms showing CD106 expression (or Ig Isotype control staining) were derived from gated events with the forward and side light-scatter characteristics of viable cells. Flow cytometric analysis was performed using a BD LSRFortessa™ Cell Analyzer 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 BD Horizon™ BB515 under optimum conditions and unconjugated antibody was removed.

Application Notes**Application**

Flow cytometry	Routinely Tested
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Recommended Assay Procedure:

BD™ CompBeads can be used as surrogates to assess fluorescence spillover (Compensation). When fluorochrome conjugated antibodies are bound to CompBeads, they have spectral properties very similar to cells. However, for some fluorochromes there can be small differences in spectral emissions compared to cells, resulting in spillover values that differ when compared to biological controls. It is strongly recommended that when using a reagent for the first time, users compare the spillover on cells and CompBead to ensure that BD Comp beads are appropriate for your specific cellular application.

For optimal and reproducible results, BD Horizon Brilliant Stain Buffer should be used anytime two or more BD Horizon Brilliant dyes are used in the same experiment. Fluorescent dye interactions may cause staining artifacts which may affect data interpretation. The BD Horizon Brilliant Stain Buffer was designed to minimize these interactions. More information can be found in the Technical Data Sheet of the BD Horizon Brilliant Stain Buffer (Cat. No. 563794/566349) or the BD Horizon Brilliant Stain Buffer Plus (Cat. No. 566385).

For optimal results, it is recommended to perform 2 washes after staining with antibodies. Cells may be prepared, stained with antibodies and washed twice with wash buffer per established protocols for immunofluorescence staining, prior to acquisition on a flow cytometer. Performing fewer than the recommended wash steps may lead to increased spread of the negative population.

Suggested Companion Products

Catalog Number	Name	Size	Clone
554618	Recombinant Human TNF	10 µg	(none)
564416	BB515 Mouse IgG1, κ Isotype Control	100 µg	X40
554656	Stain Buffer (FBS)	500 mL	(none)
554657	Stain Buffer (BSA)	500 mL	(none)
563794	Brilliant Stain Buffer	100 Tests	(none)
566349	Brilliant Stain Buffer	1000 Tests	(none)
566385	Brilliant Stain Buffer Plus	1000 Tests	(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. An isotype control should be used at the same concentration as the antibody of interest.
3. 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.
4. The manufacture, use, sale, offer for sale, or import of this product is subject to one or more patents or pending applications. This product, and only in the amount purchased by buyer, may be used solely for buyer's own internal research, in a manner consistent with the accompanying product literature. No other right to use, sell or otherwise transfer (a) this product, or (b) its components is hereby granted expressly, by implication or by estoppel. Diagnostic uses require a separate license.
5. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
6. BD Horizon Brilliant Stain Buffer is covered by one or more of the following US patents: 8,110,673; 8,158,444; 8,575,303; 8,354,239.
7. Please refer to <http://regdocs.bd.com> to access safety data sheets (SDS).
8. Please refer to www.bdbiosciences.com/us/s/resources for technical protocols.

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