

Technical Data Sheet

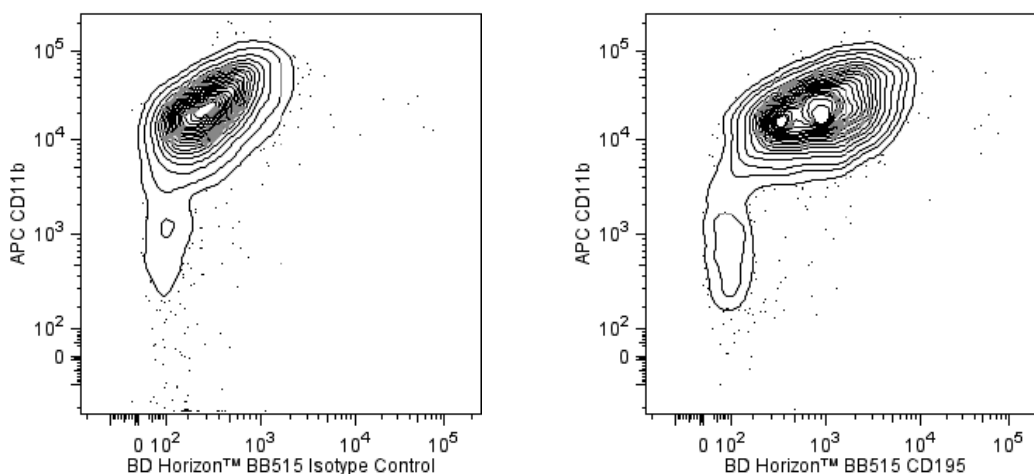
BB515 Rat Anti-Mouse CD195**Product Information**

Material Number:	565093
Alternate Name:	Ccr5; chemokine (C-C) receptor 5; C-C CKR-5; CC-CKR-5; CCR-5; AM4-7
Size:	50 µg
Concentration:	0.2 mg/ml
Clone:	C34-3448
Immunogen:	Mouse CCR5 aa. 9-30
Isotype:	Rat IgG2c, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The C34-3448 monoclonal antibody specifically binds to CD195 which is also known as, C-C chemokine receptor type 5 (CCR5). CD195 is a seven transmembrane-spanning G-protein-coupled receptor that belongs to the β-chemokine receptor family. CD195 regulates lymphocyte chemotaxis activation and transendothelial migration during inflammation. It signals in response to at least three chemokines: CCL3/MIP-1α, CCL4/MIP-1β, and CCL5/RANTES. CD195 is expressed on macrophages and some T-lymphocytes.

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.



Two-color flow cytometric analysis of CD195 expression on mouse peritoneal exudate cells. Mouse peritoneal exudate cells (PEC) were isolated 3 days post-stimulation by intraperitoneal injection of a 3% thioglycollate solution. The PEC were preincubated with Purified Rat Anti-Mouse CD16/CD32 antibody (Mouse BD Fc Block™) (Cat. No. 553141/553142). The cells were then stained with APC Rat Anti-Mouse CD11b (Cat. No. 553312/561690) and either BD Horizon™ BB515 Rat IgG2c, κ Isotype Control (Cat. No. 565091; Left Panel) or BD Horizon BB515 Rat Anti-Mouse CD195 (Cat. No. 565093; Right Panel). Two-color flow cytometric contour plots showing the correlated expression of CD195 (or Ig Isotype control staining) versus CD11b were derived from gated events with the forward and side light-scatter characteristics of viable PEC. Flow cytometric analysis was performed using a BD™ LSR II Flow Cytometer 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

BD Biosciences

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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
553142	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.5 mg	2.4G2
553312	APC Rat Anti-CD11b	0.1 mg	M1/70
561690	APC Rat Anti-CD11b	25 µg	M1/70
554656	Stain Buffer (FBS)	500 mL	(none)
554657	Stain Buffer (BSA)	500 mL	(none)
563794	Brilliant Stain Buffer	100 Tests	(none)
565091	BB515 Rat IgG2c, κ Isotype Control	50 µg	A23-1
566385	Brilliant Stain Buffer Plus	1000 Tests	(none)
566349	Brilliant Stain Buffer	1000 Tests	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
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. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
5. Please refer to <http://regdocs.bd.com> to access safety data sheets (SDS).
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 www.bdbiosciences.com/us/s/resources for technical protocols.

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