Technical Data Sheet

BB515 Rat Anti-Mouse CD43

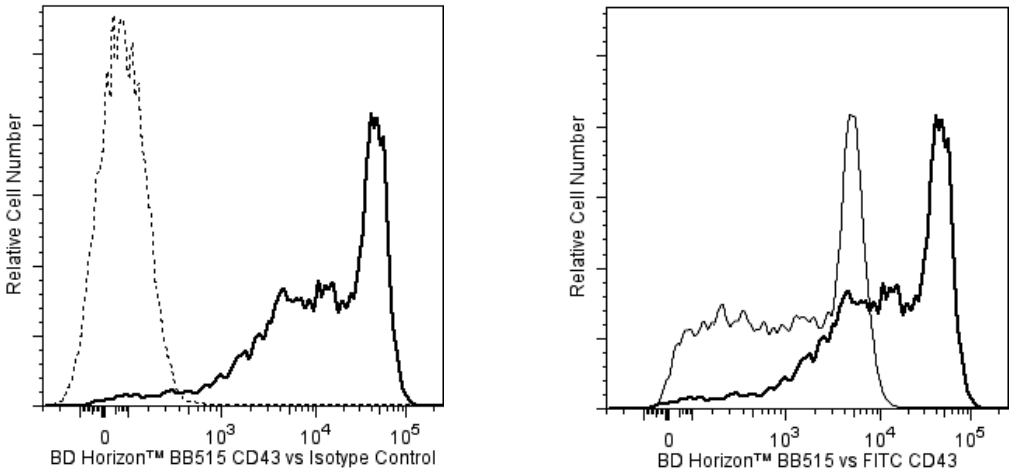
Product Information

Material Number:	**564646**
Alternate Name:	Spn; Sialophorin; Leukosialin; Ly-48; Ly48; Galgp; LEUK
Size:	50 µg
Concentration:	0.2 mg/ml
Clone:	S7
Immunogen:	Mouse Plasmacytoma MOPC-315
Isotype:	Rat (DA x LOU) IgG2a, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The S7 monoclonal antibody specifically binds to the 115 kDa glycosylated form of CD43 (Ly-48, Leukosialin). CD43 is expressed on IL-7-responsive pro-B cells, plasma cells, peritoneal and splenic CD5+ B cells (B-1 cells), granulocytes, monocytes, macrophages, platelets, natural killer cells, thymocytes, peripheral T cytotoxic/suppressor cells, and most T helper cells, but not resting conventional peripheral B cells. CD43 expression has also been detected on pluripotent hematopoietic stem cells and myeloid, lymphoid, and NK-cell progenitors in the bone marrow. Studies of CD43-deficient mice indicate that CD43 participates in the negative regulation of T-cell activation and adhesion.

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 CD43 expression on mouse bone marrow cells - Staining comparisons between BD Horizon™ BB515- and FITC-conjugated antibodies. *Mouse bone marrow cells were preincubated with Purified Rat Anti-Mouse CD16/CD32 antibody (Mouse BD Fc Block™) (Cat. No. 553141/553142). The cells were then stained with either BD Horizon™ BB515 Rat IgG2a, κ Isotype Control (Cat. No. 564418; dashed line histogram) or BD Horizon BB515 Rat Anti-Mouse CD43 antibody (Cat. No. 564646; bold solid line histogram). Alternatively, cells were stained with FITC Anti-Mouse CD43 antibody (Cat. No. 553270/561856; thin solid line histogram).*

Overlaid histograms are shown to facilitate staining comparisons between: BB515 Anti-CD43 antibody versus its Ig Isotype Control (Left Panel), and BB515 Anti-CD43 antibody versus FITC Anti-CD43 antibody (Right Panel). The fluorescence histograms showing CD43 expression (or Ig Isotype control staining) were derived from gated events with the forward and side light-scatter characteristics of viable bone marrow cells. 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.

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Application Notes

Application

Flow cytometry	Routinely Tested

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
564418	BB515 Rat IgG2a, κ Isotype Control	0.1 mg	R35-95
554656	Stain Buffer (FBS)	500 mL	(none)
554657	Stain Buffer (BSA)	500 mL	(none)
563794	Brilliant Stain Buffer	100 Tests	(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
566349	Brilliant Stain Buffer	1000 Tests	(none)
566385	Brilliant Stain Buffer Plus	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. 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.
6. Please refer to www.regdocs.bd.com to access safety data sheets (SDS).
7. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

Gulley ML, Ogata LC, Thorson JA, Dailey MO, Kemp JD. Identification of a murine pan-T cell antigen which is also expressed during the terminal phases of B cell differentiation. *J Immunol.* 1988; 140(11):3751-3757. (Immunogen: Flow cytometry, Fluorescence activated cell sorting, Immunofluorescence, Western blot)

Hardy R, Hayakawa K. Generation of Ly-1 B cells from developmentally distinct precursors. Enrichment by stromal-cell culture or cell sorting. *Ann N Y Acad Sci.* 1992; 651:99-111. (Biology: Western blot)

Hardy RR, Carmack CE, Shinton SA, Kemp JD, Hayakawa K. Resolution and characterization of pro-B and pre-pro-B cell stages in normal mouse bone marrow. *J Exp Med.* 1991; 173(5):1213-1225. (Clone-specific: Flow cytometry, Fluorescence activated cell sorting)

Jones AT, Federsppiel B, Ellies LG, et al. Characterization of the activation-associated isoform of CD43 on murine T lymphocytes. *J Immunol.* 1994; 153(8):3426-3439. (Clone-specific: Western blot)

Moore T, Bennett M, Kumar V. Transplantable NK cell progenitors in murine bone marrow. *J Immunol.* 1995; 154(4):1653-1663. (Clone-specific: Flow cytometry)

Rolink A, ten Boekel E, Melchers F, Fearon DT, Krop I, Andersson J. A subpopulation of B220+ cells in murine bone marrow does not express CD19 and contains natural killer cell progenitors. *J Exp Med.* 1996; 183(1):187-194. (Clone-specific: Flow cytometry)

Stockton BM, Cheng G, Manjunath N, Ardman B, von Andrian UH. Negative regulation of T cell homing by CD43. *Immunity.* 1998; 8(3):373-381. (Clone-specific: Flow cytometry)

Thurman EC, Walker J, Jayaraman S, Manjunath N, Ardman B, Green JM. Regulation of in vitro and in vivo T cell activation by CD43. *Int Immunol.* 1998; 10(5):691-701. (Biology)

Wells SM, Kantor AB, Stall AM. CD43 (S7) expression identifies peripheral B cell subsets. *J Immunol.* 1994; 153(12):5503-5515. (Clone-specific: Flow cytometry, Fluorescence activated cell sorting)