Technical Data Sheet

BUV563 Rat Anti-Mouse Ly-6G and Ly-6C

Product Information

Material Number:	741226
Size:	50 µg
Clone:	RB6-8C5
Alternative Name:	Ly6c, Lymphocyte antigen 6C2; Lymphocyte antigen 6G, Ly6g, Gr-1
Reactivity:	Mouse (Tested in Development)
Isotype:	Rat IgG2b, κ
Immunogen:	Not Reported
Application:	Flow cytometry (Qualified)
Concentration:	0.2 mg/ml
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.
Regulatory Status:	RUO

Description

The RB6-8C5 monoclonal antibody recognizes a common epitope on Ly-6G and Ly-6C, previously known as the myeloid differentiation antigen Gr-1. In the bone marrow, the level of antigen expression is directly correlated with granulocyte differentiation and maturation. The antigen is also expressed on the monocyte lineage in the bone marrow, but not on erythroid cells. In the periphery, RB6-8C5 antibody recognizes granulocytes (neutrophils and eosinophils) and monocytes. The RB6-8C5 antibody is a component of the "lineage cocktail" used in studies of hematopoietic cell lineages. The 1A8 antibody (Cat. No. 551461) specifically recognizes Ly-6G, but not Ly-6C.
Based on comparison of the staining patterns given by 1A8 versus RB6-8C5 antibodies on total blood leucocytes, it is evident that the 1A8 antibody stains the RB6-8C5-bright population, corresponding to Ly-6G-expressing granulocytes; whereas, the RB6-8C5-dim population is 1A8-negative and corresponds to Ly-6C-expressing lymphocytes and monocytes. Please refer to the Technical Data Sheets for Cat. No. 551459 and 553128 for more detailed information.

The antibody was conjugated to BD Horizon™ BUV563 which is part of the BD Horizon Brilliant™ Ultraviolet family of dyes. This dye is a tandem fluorochrome of BD Horizon BUV395 which has an Ex Max of 348 nm and an acceptor dye. The tandem has an Em Max at 563 nm. BD Horizon BUV563 can be excited by the 355 nm ultraviolet laser. On instruments with a 561 nm Yellow-Green laser, the recommended bandpass filter is 585/15 nm with a 535 nm long pass to minimize laser light leakage. When BD Horizon BUV563 is used with an instrument that does not have a 561 nm laser, a 560/40 nm filter with a 535 nm long pass may be more optimal. Due to the excitation and emission characteristics of the acceptor dye, there may be spillover into the PE and PE-CF594 detectors. However, the spillover can be corrected through compensation as with any other dye combination.

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 BUV563 under optimal conditions that minimize unconjugated dye and antibody.

Recommended Assay Procedure

For optimal and reproducible results, BD Horizon Brilliant Stain Buffer should be used anytime two or more BD Horizon Brilliant dyes (including BD OptiBuild Brilliant reagents) 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).

Suggested Companion Products

Catalog Number	Name	Size	Clone
612925	BUV563 Rat IgG2b, κ Isotype Control	50 µg	R35-38
554656	Stain Buffer (FBS)	500 mL	
554657	Stain Buffer (BSA)	500 mL	

563794	Brilliant Stain Buffer	100 Tests	
555899	Lysing Buffer	100 mL	
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.1 mg	2.4G2

Product Notices

1. This antibody was developed for use in flow cytometry.
2. The production process underwent stringent testing and validation to assure that it generates a high-quality conjugate with consistent performance and specific binding activity. However, verification testing has not been performed on all conjugate lots.
3. Researchers should determine the optimal concentration of this reagent for their individual applications.
4. An isotype control should be used at the same concentration as the antibody of interest.
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.
7. Please refer to www.bdbiosciences.com/us/s/resources for technical protocols.
8. 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.

References

Brummer E, Sugar AM, Stevens DA. Immunological activation of polymorphonuclear neutrophils for fungal killing: studies with murine cells and blastomyces dermatitidis in vitro. J Leukoc Biol. 1984; 36(4):505-520. (Clone-specific: Flow cytometry).

Conlan JW, North RJ. Neutrophils are essential for early anti-Listeria defense in the liver, but not in the spleen or peritoneal cavity, as revealed by a granulocyte-depleting monoclonal antibody. J Exp Med. 1994; 179(1):259-268. (Clone-specific: Flow cytometry).

Czuprynski CJ, Brown JF, Maroushek N, Wagner RD, Steinberg H. Administration of anti-granulocyte mAb RB6-8C5 impairs the resistance of mice to Listeria monocytogenes infection. J Immunol. 1994; 152(4):1836-1846. (Clone-specific: Flow cytometry).

Fleming TJ, Fleming ML, Malek TR. Selective expression of Ly-6G on myeloid lineage cells in mouse bone marrow. RB6-8C5 mAb to granulocyte-differentiation antigen (Gr-1) detects members of the Ly-6 family. J Immunol. 1993; 151(5):2399-2408. (Clone-specific: Flow cytometry).

Gumley TP, McKenzie IF, Sandrin MS. Tissue expression, structure and function of the murine Ly-6 family of molecules. Immunol Cell Biol. 1995; 73(4):277-296. (Biology: Flow cytometry).

Hestdal K, Ruscetti FW, Ihle JN, et al. Characterization and regulation of RB6-8C5 antigen expression on murine bone marrow cells. J Immunol. 1991; 147(1):22-28. (Biology: Flow cytometry).

Lagasse E, Weissman IL. Flow cytometric identification of murine neutrophils and monocytes. J Immunol Methods. 1996; 197(1-2):139-150. (Biology: Flow cytometry).

Lewinsohn DM, Bargatze RF, Butcher EC. Leukocyte-endothelial cell recognition: evidence of a common molecular mechanism shared by neutrophils, lymphocytes, and other leukocytes. J Immunol. 1987; 138(12):4313-4321. (Biology: Flow cytometry).

Stoppacciaro A, Melani C, Parenza M, et al. Regression of an established tumor genetically modified to release granulocyte colony-stimulating factor requires granulocyte-T cell cooperation and T cell-produced interferon gamma. J Exp Med. 1993; 178(1):151-161. (Clone-specific: Flow cytometry).

Tepper RI, Coffman RL, Leder P. An eosinophil-dependent mechanism for the antitumor effect of interleukin-4. Science. 1992; 257(5069):548-551. (Biology: Flow cytometry).

Tumpey TM, Chen SH, Oakes JE, Lausch RN. Neutrophil-mediated suppression of virus replication after herpes simplex virus type 1 infection of the murine cornea. J Virol. 1996; 70(2):898-904. (Clone-specific: Flow cytometry).

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