Technical Data Sheet

PerCP-Cy™5.5 Rat Anti-Mouse Ly-6G and Ly-6C

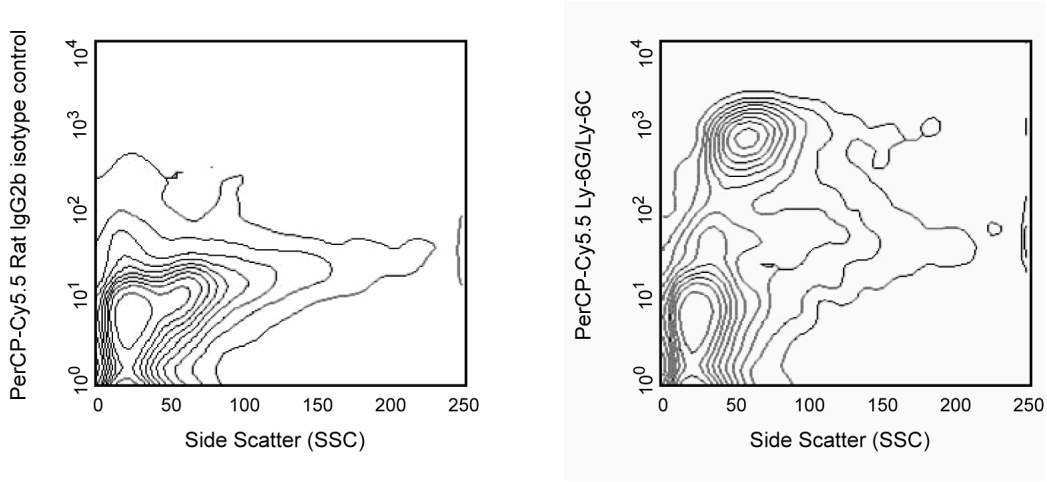
Product Information

Material Number:	**552093**
Alternate Name:	Ly6c, Lymphocyte antigen 6C2; Lymphocyte antigen 6G, Ly6g, Gr-1
Size:	0.1 mg
Concentration:	0.2 mg/ml
Clone:	RB6-8C5
Isotype:	Rat IgG2b, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing protein stabilizer and ≤0.09% sodium azide.

Description

The RB6-8C5 antibody reacts with 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 mAb is a component of the "lineage cocktail" used in studies of hematopoietic lineages. The mAb 1A8 (Cat. No. 551461) specifically recognizes Ly-6G, but not Ly-6C.

Based on the comparison of the staining patterns of mAbs clones 1A8 and RB6-8C5 on total blood leukocytes, it is evident that mAb 1A8 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 TDS Cat. No. 551459 and 553128 for more detail information.



Two-parameter analysis of the expression of Ly-6G and/or Ly-6C on bone marrow myeloid cells. *BALB/c bone-marrow leukocytes were stained with either PerCP-Cy™5.5 Rat IgG2b, κ Isotype Control (Cat. No. 550764; left panel) or PerCP-Cy™5.5 Rat Anti-Mouse Ly-6G and Ly-6C (Cat. No. 552093/561103; right panel). Please note that the population of cells having the lowest SSC (erythroid and lymphoid cells) shows little expression of Ly-6G/Ly-6C, while most of the leukocytes with moderate to high SSC (myeloid cells) are Ly-6G/Ly-6C positive. Flow cytometry was performed on a BD FACSCalibur™ Flow Cytometry 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 PerCP-Cy5.5 under optimum conditions, and unconjugated antibody and free PerCP-Cy5.5 were removed. Storage of PerCP-Cy5.5 conjugates in unoptimized diluent is not recommended and may result in loss of signal intensity.

Application Notes

Application

Flow cytometry	Routinely Tested

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Suggested Companion Products

Catalog Number	Name	Size	Clone
550764	PerCP-Cy™5.5 Rat IgG2b, κ Isotype Control	0.1 mg	A95-1
554656	Stain Buffer (FBS)	500 ml	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
3. Please observe the following precautions: Absorption of visible light can significantly alter the energy transfer occurring in any tandem fluorochrome conjugate; therefore, we recommend that special precautions be taken (such as wrapping vials, tubes, or racks in aluminum foil) to prevent exposure of conjugated reagents, including cells stained with those reagents, to room illumination.
4. PerCP-Cy5.5–labelled antibodies can be used with FITC- and R-PE–labelled reagents in single-laser flow cytometers with no significant spectral overlap of PerCP-Cy5.5, FITC, and R-PE fluorescence.
5. PerCP-Cy5.5 is optimized for use with a single argon ion laser emitting 488-nm light. Because of the broad absorption spectrum of the tandem fluorochrome, extra care must be taken when using dual-laser cytometers, which may directly excite both PerCP and Cy5.5™. We recommend the use of cross-beam compensation during data acquisition or software compensation during data analysis.
6. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
7. Cy is a trademark of Amersham Biosciences Limited. This conjugated product is sold under license to the following patents: US Patent Nos. 5,486,616; 5,569,587; 5,569,766; 5,627,027.
8. This product is subject to proprietary rights of Amersham Biosciences Corp. and Carnegie Mellon University and made and sold under license from Amersham Biosciences Corp. This product is licensed for sale only for research. It is not licensed for any other use. If you require a commercial license to use this product and do not have one return this material, unopened to BD Biosciences, 10975 Torreyana Rd, San Diego, CA 92121 and any money paid for the material will be refunded.
9. 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.
10. An isotype control should be used at the same concentration as the antibody of interest.

References

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Greimers R, Trebak M, Moutschen M, Jacobs N, Boniver J. Improved four-color flow cytometry method using fluo-3 and triple immunofluorescence for analysis of intracellular calcium ion ([Ca2+]i) fluxes among mouse lymph node B- and T-lymphocyte subsets. *Cytometry.* 1996; 23(3):205-217. (Methodology)

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Shapiro HM. *Practical Flow Cytometry, 3rd Edition.* New York: Wiley-Liss, Inc; 1995:280-281. (Methodology)

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

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