SONY

Product Data Sheet

Brilliant Violet 570™ anti-mouse Ly-6G/Ly-6C (Gr-1)

Catalog # / Size: 1142155 / 125 µl

Clone: RB6-8C5

Isotype: Rat IgG2b, κ

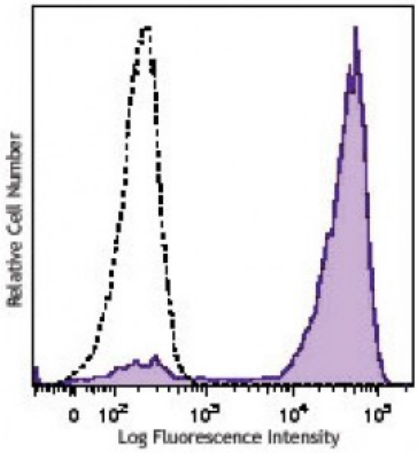
Immunogen: Raised against granulocytes of mouse origin

Reactivity: Mouse

Preparation: The antibody was purified by affinity chromatography and conjugated with Brilliant Violet 570™ under optimal conditions. The solution is free of unconjugated Brilliant Violet 570™ and unconjugated antibody.

Formulation: Phosphate-buffered solution, pH 7.2, containing 0.09% sodium azide and BSA (origin USA).

Concentration: Lot-specific



C57BL/6 mouse bone marrow cells were stained with Ly-6G/Ly6C (clone RB6-8C5) Brilliant Violet 570™ (filled histogram) or rat IgG2b, κ Brilliant Violet 570™ isotype control (open histogram). Data shown was gated on myeloid cell population

Applications:

Applications: Flow Cytometry

Recommended Usage: Each lot of this antibody is quality control tested by immunofluorescent staining with flow cytometric analysis. For flow cytometric staining, the suggested use of this reagent is ≤5 microL per million cells or 5 microL per 100 microL of whole blood. It is recommended that the reagent be titrated for optimal performance for each application.

Brilliant Violet 570™ excites at 405 nm and emits at 570 nm. The bandpass filter 585/42 nm is recommended for detection, although filter optimization may be required depending on other fluorophores used. **Be sure to verify that your cytometer configuration and software setup are appropriate for detecting this channel.** Refer to your instrument manual or manufacturer for support. Brilliant Violet 570™ is a trademark of Sirigen Group Ltd.

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Application Notes: Clone RB6-8C5 binds with high affinity to mouse Ly-6G molecules and to a lower extent to Ly-6C[19]. Clone RB6-8C5 impairs the binding of anti-mouse Ly-6G clone 1A8[19]. However, clone RB6-8C5 is able to stain in the presence of anti-mouse Ly-6C clone HK1.4[20].

The RB6-8C5 antibody has been used to identify peripheral blood neutrophils and deplete granulocytes *in vivo*. Additional reported applications (for relevant formats of this clone) include: *in vitro* complement-mediated cytotoxicity2, *in vivo* depletion[3-5,9], immunoprecipitation1, immunohistochemical staining[6] (including

paraffin-embedded sections[9,16], acetone-fixed frozen sections[11] and zinc-fixed sections[15]), and Western blotting[7]. RB6-8C5 is not suitable for depletion of hepatic myeloid derived suppressor cells (MDSCs)[20].

Special Note: The LEAF™ purified antibody (Endotoxin <0.1 EU/µg, Azide-Free, 0.2 µm filtered) is recommended for functional assays (Cat. No. 108414). For *in vivo* studies or highly sensitive assays, we recommend Ultra-LEAF™ purified antibody (Cat. No. 108436) with a lower endotoxin limit than standard LEAF™ purified antibodies (Endotoxin <0.01 EU/microg).

Application References:

1. Fleming TJ, *et al.* 1993. *J. Immunol.* 151:2399. (IP)
2. Brummer E, *et al.* 1984. *J. Leukocyte Biol.* 36:505. (CMCD)
3. Stoppacciaro A, *et al.* 1993. *J. Exp. Med.* 178:151. (Deplete)
4. Tumpey TM, *et al.* 1996. *J. Virol.* 70:898. (Deplete)
5. Czuprynski CJ, *et al.* 1994. *J. Immunol.* 152:1836. (Deplete)
6. Nitta H, *et al.* 1997. *Cell Vision* 4:73. (IHC)
7. Jutila MA, *et al.* 1988. *Eur. J. Immunol.* 18:1819. (WB)
8. Engwerda CR, *et al.* 2004. *Am. J. Pathol.* 165:2123.
9. Brown CR, *et al.* 2004. *Infect. Immun.* 72:4956. (Deplete, IHC)
10. Andoniou CE, *et al.* 2005. *Nature Immunology* 6:1011. (FC) PubMed
11. Li M, *et al.* 2006. *P. Natl. Acad. Sci USA* 103:11736. (IHC)
12. Dzhagalov I, *et al.* 2007. *Blood* 109:1620. (FC) PubMed
13. Fazilleau N, *et al.* 2007. *Nature Immunol.* 8:753. (FC) PubMed
14. Heuser M, *et al.* 2007. *Blood* 110:1639. (FC) PubMed
15. Wang T, *et al.* 2007. *Infect. Immun.* 75:1144. (IHC)
16. Bosio CM, *et al.* 2007. *J. Immunol.* 178:4538. (IHC)
17. Boehme SA, *et al.* 2009. *Int. Immunol.* 21:81. (IHC)
18. Piao Y, *et al.* 2012. *Neuro Oncol.* 14:1379. PubMed
19. Ribechini E, *et al.* 2009. *Eur. J. Immunol.* 39:3538.
20. Ma C, *et al.* 2012. *J. Leukoc. Biol.* 92:1199.

Description: Gr-1 is a 21-25 kD protein also known as Ly-6G/Ly-6C. This myeloid differentiation antigen is a glycosylphosphatidylinositol (GPI)-linked protein expressed on granulocytes and macrophages. In bone marrow, the expression levels of Gr-1 directly correlate with granulocyte differentiation and maturation; Gr-1 is also transiently expressed on bone marrow cells in the monocyte lineage. Immature Myeloid Gr-1+ cells play a role in the development of antitumor immunity.

Antigen References:

1. Fleming TJ, *et al.* 1993. *J. Immunol.* 151:2399.
2. Jutila MA, *et al.* 1988. *Eur. J. Immunol.* 18:1819.
3. Goni O, *et al.* 2002. *Int. Immunol.* 14:1125.