

Technical Data Sheet

BV605 Rat Anti-Mouse CD21/CD35

Product Information

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| Material Number: | 563176 |
| Alternate Name: | CR2/CR1 |
| Size: | 50 µg |
| Concentration: | 0.2 mg/ml |
| Clone: | 7G6 |
| Immunogen: | Purified Mouse CR1 |
| Isotype: | Rat (SD) IgG2b, κ |
| Reactivity: | QC Testing: Mouse |
| Storage Buffer: | Aqueous buffered solution containing BSA and ≤0.09% sodium azide. |

Description

The 7G6 antibody recognizes an epitope shared by 145-150-kDa and 190-kDa complement receptor proteins, originally designated CR2 (CD21) and CR1 (CD35), respectively. In the mouse, CD21 and CD35 are expressed on the majority of peripheral B lymphocytes, on the majority of resident peritoneal macrophages and mast cells, on peripheral blood granulocytes after treatment with N-formyl-Met-Leu-Phe, and on follicular dendritic cells, but not on thymocytes, T cells, erythrocytes, or platelets. CD21 is a ligand-binding component of the CD19/CD21/CD81 signal-transduction complex associated with the antigen receptor on B lymphocytes. CD21/CD35 also co-localizes with CD19 on the surface of peritoneal mast cells. Cr2null mice display impaired inflammatory and humoral immune responses in vivo. The 7G6 mAb has been reported to inhibit rosette formation by C3d-bearing sheep erythrocytes, to block the complement dependent trapping of immune complexes by follicular dendritic cells, and to down-regulate mouse CD21/CD35 expression upon in vivo application, thus inhibiting primary antibody responses to immunization. Co-stimulation of B-cell differentiation via Sepharose-coupled 7G6 antibody has also been observed. The 7G6 mAb recognizes an epitope on CD35 distinct from the epitope recognized by anti-mouse CD35, clone 8C12 (Cat. No. 558768, for the purified antibody), and it does not block binding of 8C12 mAb to mouse CD35.

This antibody is conjugated to BD Horizon BV605 which is part of the BD Horizon Brilliant™ Violet family of dyes. With an Ex Max of 407-nm and Em Max of 602-nm, BD Horizon BV605 can be excited by a violet laser and detected with a standard 610/20-nm filter set. BD Horizon BV605 is a tandem fluorochrome of BD Horizon BV421 and an acceptor dye with an Em max at 605-nm. Due to the excitation of the acceptor dye by the green (532 nm) and yellow-green (561 nm) lasers, there will be significant spillover into the PE and BD Horizon PE-CF594 detectors off the green or yellow-green lasers. BD Horizon BV605 conjugates are very bright, often exhibiting brightness equivalent to PE conjugates and can be used as a third color off of the violet laser.

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).

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™ BV605 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV605 were removed.

Application Notes

Application

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| Flow cytometry | Routinely Tested |
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Suggested Companion Products

| Catalog Number | Name | Size | Clone |
|----------------|--|--------|---------|
| 554656 | Stain Buffer (FBS) | 500 mL | (none) |
| 563145 | BV605 Rat IgG2b, κ Isotype Control | 50 µg | R35-38 |
| 555899 | Lysing Buffer | 100 mL | (none) |
| 553092 | APC Rat Anti-Mouse CD45R/B220 | 0.1 mg | RA3-6B2 |
| 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 |
| 563794 | Brilliant Stain Buffer | 5 mL | (none) |

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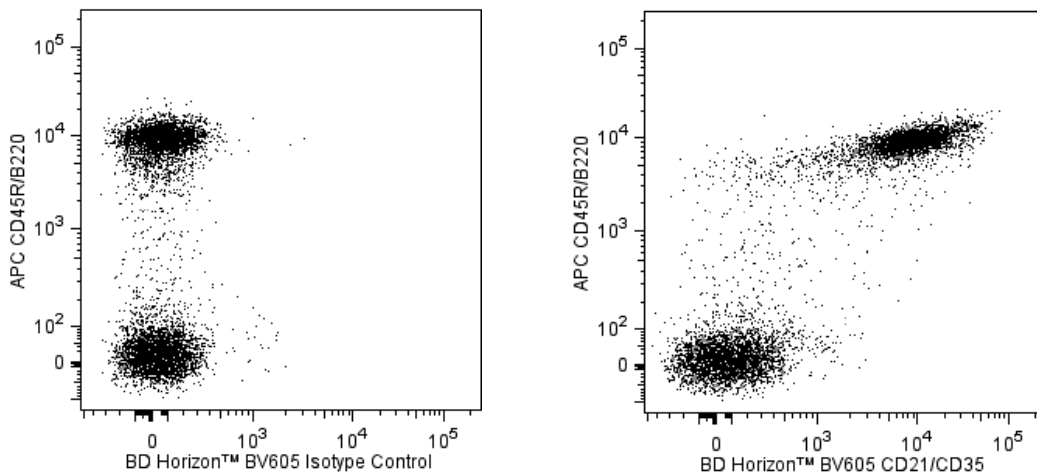
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Two-color flow cytometric analysis of CD21/CD35 expression on BALB/c mouse splenocytes. BALB/c splenic leukocytes were simultaneously stained with APC Rat Anti-Mouse CD45R/B220 antibody (Cat. No. 553092/561880) and either BD Horizon™ BV605 Rat IgG2b, κ Isotype Control (Cat. No. 563145, Left Panel) or BD Horizon™ BV605 Rat Anti-Mouse CD21/CD35 antibody (Cat. No. 563176, Right Panel) in the presence of Purified Rat Anti-Mouse CD16/CD32 antibody (Mouse BD Fc Block™) (Cat. No. 553141/553142). Two-color flow cytometric dot plots showing the correlated expression of CD21/CD35 (or Ig Isotype control staining) versus CD45R/B220 were derived from gated events with the forward and side light-scatter characteristics of viable splenocytes. Flow cytometric analysis was performed using a BD LSRFortessa™ Cell Analyzer System.

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
3. An isotype control should be used at the same concentration as the antibody of interest.
4. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
5. 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.
6. 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.
7. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
8. Although every effort is made to minimize the lot-to-lot variation in the efficiency of the fluorochrome energy transfer, differences in the residual emission from BD Horizon™ BV421 may be observed. Therefore, we recommend that individual compensation controls be performed for every BD Horizon™ BV605 conjugate.
9. CF™ is a trademark of Biotium, Inc.

References

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- Thyphronitis G, Kinoshita T, Inoue K, et al. Modulation of mouse complement receptors 1 and 2 suppresses antibody responses in vivo. *J Immunol.* 1991; 147(1):224-230. (Clone-specific: Inhibition, In vivo exacerbation)
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