Technical Data Sheet

BUV737 Rat Anti-Mouse CD21/CD35

Product Information

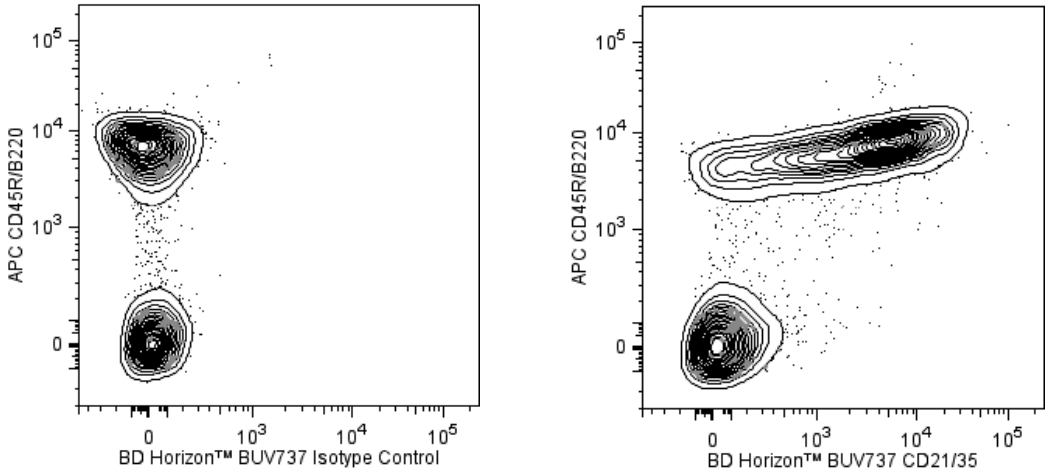
Material Number:	**565090**
Alternate Name:	CR2/CR1
Size:	50 µg
Concentration:	0.2 mg/ml
Clone:	7G6
Immunogen:	Purified Mouse CR1
Isotype:	Rat (SD) IgG2b, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

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.

The antibody was conjugated to BD Horizon BUV737 which is part of the BD Horizon Brilliant™ Ultraviolet family of dyes. This dye is a tandem fluorochrome of BD Horizon BUV395 with an Ex Max of 348-nm and an acceptor dye with an Em Max at 737-nm. BD Horizon Brilliant BUV737 can be excited by the ultraviolet laser (355 nm) and detected with a 740/35 filter. Due to the excitation of the acceptor dye by other laser lines, there may be significant spillover into channels detecting Alexa Fluor® 700-like dyes (e.g., 712/20-nm filter).

Due to spectral differences between labeled cells and beads, using BD™ CompBeads can result in incorrect spillover values when used with BD Horizon BUV737 reagents. Therefore, the use of BD CompBeads or BD CompBeads Plus to determine spillover values for these reagents is not recommended. Different BUV737 reagents (e.g., CD4 vs. CD45) can have slightly different fluorescence spillover therefore, it may also be necessary to use clone specific compensation controls when using these reagents.



Two-color flow cytometric analysis of CD21/CD35 expression on mouse splenocytes. *BALB/c splenic leucocytes were preincubated with Purified Rat Anti-Mouse CD16/CD32 antibody (Mouse BD Fc Block™) (Cat. No. 553141/553142). The cells were then stained with APC Rat Anti-Mouse CD45R/B220 antibody (Cat. No. 553092/561880) and either BD Horizon™ BUV737 Rat IgG2b, κ Isotype Control (Cat. No. 564295; Left Panel) or BD Horizon BUV737 Rat Anti-Mouse CD21/CD35 antibody (Cat. No. 565090, Right Panel). Two-color flow cytometric contour 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.*

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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™ BUV737 under optimum conditions, and unconjugated antibody and free BD Horizon BUV737 were removed.

Application Notes

Application

Flow cytometry	Routinely Tested

Suggested Companion Products

Catalog Number	Name	Size	Clone
554656	Stain Buffer (FBS)	500 mL	(none)
554657	Stain Buffer (BSA)	500 mL	(none)
563794	Brilliant Stain Buffer	5 mL	(none)
564295	BUV737 Rat IgG2b, κ Isotype Control	50 µg	R35-38
555899	Lysing Buffer	100 mL	(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
553092	APC Rat Anti-Mouse CD45R/B220	0.1 mg	RA3-6B2
561880	APC Rat Anti-Mouse CD45R/B220	25 µg	RA3-6B2

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. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
4. 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.
5. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
6. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

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Cariappa A, Tang M, Parng C, et al. The follicular versus marginal zone B lymphocyte cell fate decision is regulated by Aiolos, Btk, and CD21. *Immunity.* 2001; 14(5):603-615. (Clone-specific: Flow cytometry)

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Gommerman JL, Oh DY, Zhou X, et al. A role for CD21/CD35 and CD19 in responses to acute septic peritonitis: a potential mechanism for mast cell activation. *J Immunol.* 2000; 165(12):6915-6921. (Clone-specific: Flow cytometry, Fluorescence microscopy, Immunofluorescence)

Heyman B, Wiersma EJ, Kinoshita T. In vivo inhibition of the antibody response by a complement receptor-specific monoclonal antibody. *J Exp Med.* 1990; 172(2):665-668. (Clone-specific: Inhibition, In vivo exacerbation)

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Kinoshita T, Takeda J, Hong K, Kozono H, Sakai H, Inoue K. Monoclonal antibodies to mouse complement receptor type 1 (CR1). Their use in a distribution study showing that mouse erythrocytes and platelets are CR1-negative. *J Immunol.* 1988; 140(9):3066-3072. (Immunogen: Immunoprecipitation, Radioimmunoassay)

Kinoshita T, Thyphronitis G, Tsokos GC, et al. Characterization of murine complement receptor type 2 and its immunological cross-reactivity with type 1 receptor. *Int Immunol.* 1990; 2(7):651-659. (Clone-specific)

Molina H, Holers VM, Li B, et al. Markedly impaired humoral immune response in mice deficient in complement receptors 1 and 2. *Proc Natl Acad Sci U S A.* 1996; 93(8):3357-3361. (Clone-specific: Flow cytometry)

Oliver AM, Martin F, Gartland GL, Carter RH, Kearney JF. Marginal zone B cells exhibit unique activation, proliferative and immunoglobulin secretory responses. *Eur J Immunol.* 1997; 27(9):2366-2374. (Clone-specific: Flow cytometry)

Oliver AM, Martin F, Kearney JF. IgMhighCD21high lymphocytes enriched in the splenic marginal zone generate effector cells more rapidly than the bulk of follicular B cells. *J Immunol.* 1999; 162(12):7198-7207. (Clone-specific: Flow cytometry)

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Wiersma EJ, Kinoshita T, Heyman B. Inhibition of immunological memory and T-independent humoral responses by monoclonal antibodies specific for murine complement receptors. *Eur J Immunol.* 1991; 21(10):2501-2506. (Clone-specific: Inhibition, In vivo exacerbation)

Yoshida K, van den Berg TK, Dijkstra CD. Two functionally different follicular dendritic cells in secondary lymphoid follicles of mouse spleen, as revealed by CR1/2 and FcR gamma II-mediated immune-complex trapping. *Immunology.* 1993; 80(1):34-39. (Clone-specific: Inhibition)

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