

Technical Data Sheet

BUV737 Rat Anti-Mouse CD44 (to be replaced with 612799)**Product Information**

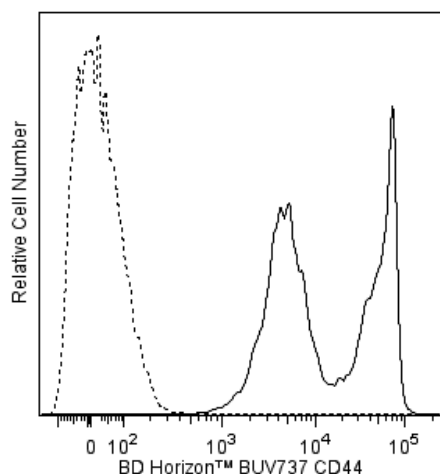
Material Number:	564392
Alternate Name:	Pgp-1; Ly-24; H-CAM; HERMES; ECMR-III; Hyaluronate Receptor
Size:	50 µg
Concentration:	0.2 mg/ml
Clone:	IM7
Immunogen:	Dexamethasone-induced, SJL mouse spontaneous myeloid leukemia M1 cells myeloid leukemia M1
Isotype:	Rat IgG2b, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The IM7 antibody specifically recognizes an epitope on both alloantigens and all isoforms of the CD44 glycoprotein (Pgp-1, Ly-24). The standard form of CD44, lacking variable exons and referred to as CD44H or CD44s, is widely expressed on hematopoietic and non-hematopoietic cells. CD44 isoforms encoded by variable exons are expressed on epithelial cells, but only at low levels on most leukocytes. Mice with the Ly-24.1 alloantigen (e.g., BALB/c, CBA/J, DBA/1, DBA/2) have relatively large subsets of CD44H+ T lymphocytes, while Ly-24.2 strains (e.g., A, AKR, CBA/N, C3H/He, C57BL, C57BR, C57L, C58, NZB, SJL, SWR, 129) have fewer CD44H+ T cells. CD44 is a cell adhesion receptor, and its principal ligand, hyaluronate, is a common component of extracellular matrices. Differential glycosylation of CD44 influences its binding to hyaluronate. Additional ligands include the cell surface form of CD74 and the cytokine osteopontin (Eta-1). Bone marrow- and thymus-derived progenitor cells capable of repopulating the thymus express CD44. In the periphery, the level of CD44 expression increases upon activation of B lymphocytes, CD4+ T cells, and CD8+ T cells; memory cells can be recognized by their CD44[hi] phenotype. The IM7 mAb inhibits established collagen-induced arthritis in DBA/1 mice. Moreover, it prevents CNS inflammation and clinical symptoms of experimental autoimmune encephalomyelitis. In contrast, the same antibody exacerbates experimental autoimmune thyroiditis in CBA/J mice. The IM7 mAb recognizes a different epitope from that recognized by mAb KM114, and the antibody pair can be used in ELISA to detect soluble CD44. It has been observed that IM7 antibody crossreacts with human, dog, cat, horse, cow, and pig leukocytes. Anti-human CD44, clone G44-26, and IM7 antibody compete for binding to human peripheral blood lymphocytes.

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 with an Ex Max near 350 nm and an Em Max near 737 nm. BD Horizon Brilliant BUV737 can be excited by the ultraviolet laser (355 nm) and detected with a 740/35 nm filter. Due to the excitation of the acceptor dye by the red laser line, there may be significant spillover into red laser detectors with filters in the 700-720 nm range.

This product will be discontinued in July 2019. 612799 is the reformulated replacement for this catalog number and it is available to be purchased. Visit bdbiosciences.com/newbu for more information.



Flow cytometric analysis of CD44 expression on mouse bone-marrow cells. Mouse bone-marrow cells were preincubated with Purified Rat Anti-Mouse CD16/CD32 antibody (Mouse BD Fc Block™) (Cat. No. 553141/553142). The cells were then stained with either BD Horizon™ BUV737 Rat IgG2b, κ Isotype Control (Cat. No. 564295; dashed line histogram) or BD Horizon BUV737 Rat Anti-Mouse CD44 antibody (Cat. No. 564392; solid line histogram). The fluorescence histogram showing CD44 expression (or Ig Isotype control staining) was derived from gated events with the forward and side light-scatter characteristics of viable bone marrow cells. Flow cytometric analysis was performed using a BD™ LSR II Flow Cytometer System.

BD Biosciences

bdbiosciences.com

United States 877.232.8995 Canada 866.979.9408 Europe 32.2.400.98.95 Japan 0120.8555.90 Asia Pacific 65.6861.0633 Latin America/Caribbean 55.11.5185.9995

For country contact information, visit bdbiosciences.com/contact

Conditions: The information disclosed herein is not to be construed as a recommendation to use the above product in violation of any patents. BD Biosciences will not be held responsible for patent infringement or other violations that may occur with the use of our products. Purchase does not include or carry any right to resell or transfer this product either as a stand-alone product or as a component of another product. Any use of this product other than the permitted use without the express written authorization of Becton, Dickinson and Company is strictly prohibited.

For Research Use Only. Not for use in diagnostic or therapeutic procedures. Not for resale.
© 2017 BD. BD, the BD Logo and all other trademarks are property of Becton, Dickinson and Company.



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

Recommended Assay Procedure:

BD™ CompBeads can be used as surrogates to assess fluorescence spillover (Compensation). When fluorochrome conjugated antibodies are bound to BD CompBeads, they have spectral properties very similar to cells. However, for some fluorochromes there can be small differences in spectral emissions compared to cells, resulting in spillover values that differ when compared to biological controls. It is strongly recommended that when using a reagent for the first time, users compare the spillover on cells and BD CompBead to ensure that BD CompBeads are appropriate for your specific cellular application.

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. 563794/566349) or the BD Horizon Brilliant Stain Buffer Plus (Cat. No. 566385).

Note: When using high concentrations of antibody, background binding of this dye to erythroid cell subsets (mature erythrocytes and precursors) has been observed. For researchers studying these cell populations, or in cases where light scatter gating does not adequately exclude these cells from the analysis, this background may be an important factor to consider when selecting reagents for panel(s).

Suggested Companion Products

Catalog Number	Name	Size	Clone
554656	Stain Buffer (FBS)	500 mL	(none)
554657	Stain Buffer (BSA)	500 mL	(none)
564295	BUV737 Rat IgG2b, κ Isotype Control (to be replaced with 612762)	50 µg	R35-38
563794	Brilliant Stain Buffer	100 Tests	(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
566385	Brilliant Stain Buffer Plus	1000 Tests	(none)
566349	Brilliant Stain Buffer	1000 Tests	(none)

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. 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.
4. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
5. BD Horizon Brilliant Ultraviolet 737 is covered by one or more of the following US patents: 8,110,673; 8,158,444; 8,227,187; 8,575,303; 8,354,239.
6. 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.
7. Please refer to <http://regdocs.bd.com> to access safety data sheets (SDS).
8. Please refer to www.bdbiosciences.com/pharming/en/protocols for technical protocols.

References

Brocke S, Piercy C, Steinman L, Weissman IL, Veromaa T. Antibodies to CD44 and integrin alpha4, but not L-selectin, prevent central nervous system inflammation and experimental encephalomyelitis by blocking secondary leukocyte recruitment. *Proc Natl Acad Sci U S A*. 1999; 96(12):6896-6901. (Clone-specific: Blocking)

Budd RC, Cerottini JC, Horvath C, et al. Distinction of virgin and memory T lymphocytes. Stable acquisition of the Pgp-1 glycoprotein concomitant with antigenic stimulation. *J Immunol*. 1987; 138(10):3120-3129. (Clone-specific: Flow cytometry, Fluorescence activated cell sorting, Immunoprecipitation)

Camp RL, Scheynius A, Johansson C, Pure E. CD44 is necessary for optimal contact allergic responses but is not required for normal leukocyte extravasation. *J Exp Med*. 1993; 178(2):497-507. (Clone-specific: Induction, Inhibition, Radioimmunoassay)

Ernst DN, Weigle WO, Noonan DJ, McQuitty DN, Hobbs MV. The age-associated increase in IFN-γ synthesis by mouse CD8+ T cells correlates with shifts in the frequencies of cell subsets defined by membrane CD44, CD45RB, 3G11, and MEL-14 expression. *J Immunol*. 1993; 151(2):575-587. (Clone-specific: Flow cytometry)

Godfrey DI, Kennedy J, Suda T, Zlotnik A. A developmental pathway involving four phenotypically and functionally distinct subsets of CD3-CD4-CD8-triple-negative adult mouse thymocytes defined by CD44 and CD25 expression. *J Immunol*. 1993; 150(10):4244-4252. (Clone-specific: Flow cytometry, Fluorescence activated cell sorting)

Hathcock KS, Hirano H, Murakami S, Hodes RJ. CD44 expression on activated B cells. Differential capacity for CD44-dependent binding to hyaluronic acid. *J Immunol*. 1993; 151(12):6712-6722. (Clone-specific: Flow cytometry, Immunoprecipitation)

Hyman R, Lesley J, Schulte R, Trotter J. Progenitor cells in the thymus: most thymus-homing progenitor cells in the adult mouse thymus bear Pgp-1 glycoprotein but not interleukin-2 receptor on their cell surface. *Cell Immunol.* 1986; 101(2):320-327. (Clone-specific: Flow cytometry)

Katoh S, McCarthy JB, Kincade PW. Characterization of soluble CD44 in the circulation of mice. Levels are affected by immune activity and tumor growth. *J Immunol.* 1994; 153(8):3440-3449. (Clone-specific: ELISA)

Katoh S, Zheng Z, Oritani K, Shimozato T, Kincade PW. Glycosylation of CD44 negatively regulates its recognition of hyaluronan. *J Exp Med.* 1995; 182(2):419-429. (Clone-specific: Blocking)

Lesley J, Hyman R, Kincade PW. CD44 and its interaction with extracellular matrix. *Adv Immunol.* 1993; 54:271-335. (Biology)

Lesley J, Trowbridge IS. Genetic characterization of a polymorphic murine cell-surface glycoprotein. *Immunogenetics.* 1982; 15(3):313-320. (Immunogen: Flow cytometry, Immunoprecipitation)

Lynch F, Ceredig R. Mouse strain variation in Ly-24 (Pgp-1) expression by peripheral T cells and thymocytes: implications for T cell differentiation. *Eur J Immunol.* 1989; 19(2):223-229. (Clone-specific: Flow cytometry)

MacDonald HR, Budd RC, Cerottini JC. Pgp-1 (Ly 24) as a marker of murine memory T lymphocytes. *Curr Top Microbiol Immunol.* 1990; 159:97-109. (Biology)

Matsumoto G, Nghiem MP, Nozaki N, Schmits R, Penninger JM. Cooperation between CD44 and LFA-1/CD11a adhesion receptors in lymphokine-activated killer cell cytotoxicity. *J Immunol.* 1998; 160(12):5781-5789. (Clone-specific: Flow cytometry)

Naor D, Sionov RV, Ish-Shalom D. CD44: structure, function, and association with the malignant process. *Adv Cancer Res.* 1997; 71:241-319. (Biology)

Nedvetzki S, Walmsley M, Alpert E, Williams RO, Feldmann M, Naor D. CD44 involvement in experimental collagen-induced arthritis (CIA). *J Autoimmun.* 1999; 13(1):39-47. (Clone-specific: Blocking)

Trowbridge IS, Lesley J, Schulte R, Hyman R, Trotter J. Biochemical characterization and cellular distribution of a polymorphic, murine cell-surface glycoprotein expressed on lymphoid tissues. *Immunogenetics.* 1982; 15:299-312. (Immunogen: Cytotoxicity, Immunoprecipitation)

Vremec D, Zorbas M, Scollay R, et al. The surface phenotype of dendritic cells purified from mouse thymus and spleen: investigation of the CD8 expression by a subpopulation of dendritic cells. *J Exp Med.* 1992; 176(1):47-58. (Clone-specific: Flow cytometry)