

Technical Data Sheet

BUV661 Rat Anti-Mouse CD8a

Product Information

Material Number:	750023
Size:	50 µg
Clone:	53-6.7
Alternative Name:	Cd8a; CD8 alpha chain; Ly-2; Lyt2; Lyt-2; Ly-35; Ly-B
Reactivity:	Mouse (Tested in Development)
Isotype:	Rat LOU, also known as Louvain, LOU/C, LOU/M IgG2a, κ
Immunogen:	Mouse Spleen Cells or Thymocyte Membranes
Application:	Flow cytometry (Qualified)
Concentration:	0.2 mg/ml
Entrez Gene ID:	12525
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.
Regulatory Status:	RUO

Description

The 53-6.7 monoclonal antibody specifically binds to the 38 kDa α and 34 kDa α' chains of the CD8 differentiation antigen (Ly-2 or Lyt-2) of all mouse strains tested. The CD8 α and α' chains (CD8a) form heterodimers with the CD8 β chain (CD8b, Ly-3, or Lyt-3) on the surface of most thymocytes. A subpopulation of mature T lymphocytes (i.e., MHC class I-restricted T cells, including most T suppressor/cytotoxic cells) expresses almost exclusively the CD8 $\alpha\beta$ heterodimer. Subsets of $\gamma\delta$ TCR-bearing T cells, intestinal intraepithelial lymphocytes, and dendritic cells express CD8a without CD8b. It has been suggested that the expression of the CD8a/CD8b heterodimer is restricted to T lymphocytes which matured in the thymus or in an extrathymic environment that had been influenced by thymus-initiated neuroendocrine signals. CD8 is an antigen coreceptor on the T-cell surface which interacts with MHC class I molecules on antigen-presenting cells or epithelial cells. It participates in T-cell activation through its association with the T-cell receptor complex and protein tyrosine kinase lck (p56 [lck]). The CD8 α and α' chains arise from alternatively spliced messengers of a single CD8a gene. The longer α form associates with p56 [lck] via a CXCP motif in its cytoplasmic domain, which it shares with CD4, but not with CD8b. The truncated α' chain is unable to associate with p56 [lck], and it may function to attenuate the CD8-mediated costimulatory signal during intrathymic T-cell maturation. In vivo and in vitro treatment with 53-6.7 mAb has reportedly been effective at depleting CD8+ peripheral T lymphocytes. The 53-6.7 antibody has also been reported to cross-react with CD8 α - and α' -like polypeptides on subsets of thymic and peripheral lymphocytes in the Egyptian toad, *Bufo regularis*.

The antibody was conjugated to BD Horizon™ BUV661 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 661-nm. BD Horizon Brilliant BUV661 can be excited by the ultraviolet laser (355 nm) and detected with a 670/25 filter and a 630 nm LP. Due to cross laser excitation of this dye, there may be significant spillover into channels detecting APC-like emissions (eg, 670/25-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 BUV661 reagents. Therefore, the use of BD CompBeads or BD CompBeads Plus to determine spillover values for these reagents is not recommended. Different BUV661 reagents (eg, 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.

Preparation and Storage Section

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 BUV661 under optimal conditions that minimize unconjugated dye and antibody.

Recommended Assay Procedure

For optimal and reproducible results, BD Horizon Brilliant Stain Buffer should be used anytime two or more BD Horizon Brilliant dyes (including BD OptiBuild Brilliant reagents) 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).

Suggested Companion Products

Catalog Number	Name	Size	Clone
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™) 2.4G2 RUO	0.1 mg	
565804	Red Nucleic Acid Stain RUO	0.5 mL	
612973	BUV661 Rat IgG2a, κ Isotype Control R35-95 RUO	50 µg	
554656	Stain Buffer (FBS) RUO	500 mL	
554657	Stain Buffer (BSA) RUO	500 mL	
563794	Brilliant Stain Buffer RUO	100 Tests	
555899	Lysing Buffer RUO	100 mL	

Product Notices

1. This antibody was developed for use in flow cytometry.
2. The production process underwent stringent testing and validation to assure that it generates a high-quality conjugate with consistent performance and specific binding activity. However, verification testing has not been performed on all conjugate lots.
3. Researchers should determine the optimal concentration of this reagent for their individual applications.
4. An isotype control should be used at the same concentration as the antibody of interest.
5. 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.
6. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
7. Please refer to www.bdbiosciences.com/us/s/resources for technical protocols.
8. 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.
9. BD Horizon Brilliant Ultraviolet 661 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.

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