

Technical Data Sheet

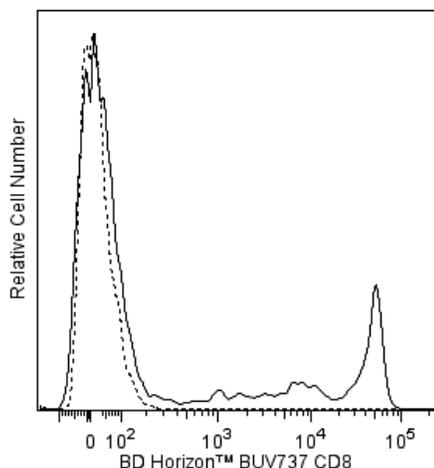
BUV737 Mouse Anti-Human CD8**Product Information**

Material Number:	564628
Alternate Name:	CD8 α ; CD8A; CD8 alpha; Leu2a; MAL; T8; p32
Size:	25 Tests
Vol. per Test:	5 μ l
Clone:	SK1
Immunogen:	Human Peripheral Blood T Cells
Isotype:	Mouse (BALB/c) IgG1, κ
Reactivity:	QC Testing: Human Tested in Development: Rhesus, Cynomolgus, Baboon
Workshop:	I T51,74; III T118,152,571
Storage Buffer:	Aqueous buffered solution containing $\leq 0.09\%$ sodium azide.

Description

The SK1 monoclonal antibody specifically binds to CD8 alpha (CD8 α). CD8 α is a type I transmembrane glycoprotein and a member of the immunoglobulin superfamily. CD8 α is expressed by the majority of thymocytes, by subpopulations of $\alpha\beta$ T cells and $\gamma\delta$ T cells and by some NK cells. Cell surface CD8 α is expressed either as a disulfide-linked homodimer (CD8 $\alpha\alpha$) or as a heterodimer (CD8 $\alpha\beta$) when disulfide-bonded to a CD8 beta chain (CD8 β). CD8-positive $\alpha\beta$ T cells coexpress both CD8 $\alpha\alpha$ homodimers and CD8 $\alpha\beta$ heterodimers whereas some $\gamma\delta$ T cells and NK cells express CD8 $\alpha\alpha$ homodimers. CD8 plays important roles in T cell activation and selection. The extracellular IgSF domain of CD8 α binds to a non-polymorphic determinant on HLA class I molecules ($\alpha 3$ domain) and enables CD8 to function as a co-receptor with MHC class I-restricted TCR during T cell recognition of antigen. The cytoplasmic domain of CD8 α associates with Lck, a Src family protein tyrosine kinase that is involved in intracellular signaling.

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.



Flow cytometric analysis of CD8 α expression on human peripheral blood lymphocytes. Whole blood was stained with either BD Horizon™ BUV737 Mouse IgG1, κ Isotype Control (Cat. No. 564299; dashed line histogram) or BD Horizon BUV737 Mouse Anti-Human CD8 antibody (Cat. No. 564628/564629; solid line histogram). Erythrocytes were lysed with BD FACS Lysing Solution (Cat. No. 349202). The fluorescence histograms were derived from gated events with the forward and side light-scatter characteristics of intact lymphocytes. Flow cytometric analysis was performed using a BD™ LSR II Flow Cytometer System.

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

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564628 Rev. 3



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
564299	BUV737 Mouse IgG1, κ Isotype Control (to be replaced with 612758)	50 μ g	X40
564629	BUV737 Mouse Anti-Human CD8 (to be replaced with 612754 & 612755)	100 Tests	SK1
349202	BD FACSTM Lysing Solution	100 mL	(none)
555899	Lysing Buffer	100 mL	(none)
554656	Stain Buffer (FBS)	500 mL	(none)
554657	Stain Buffer (BSA)	500 mL	(none)
563794	Brilliant Stain Buffer	100 Tests	(none)
566349	Brilliant Stain Buffer	1000 Tests	(none)
566385	Brilliant Stain Buffer Plus	1000 Tests	(none)

Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1×10^6 cells in a 100- μ l experimental sample (a test).
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. Species testing during development may have been performed with a different format of the same clone. Selected applications have been tested for cross-reactivity.
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 <http://regdocs.bd.com> to access safety data sheets (SDS).
7. 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.
8. 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.
9. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

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