Technical Data Sheet

BUV615 Mouse Anti-Human CD8

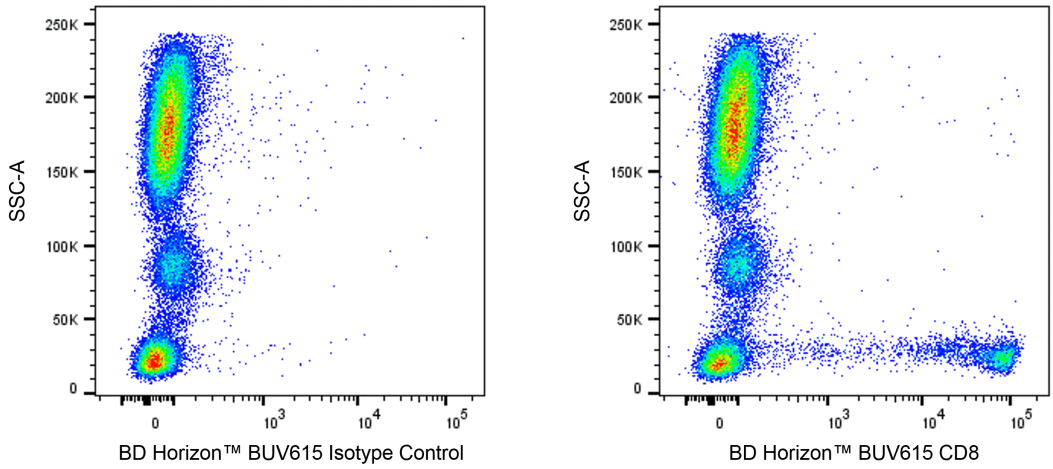
Product Information

Material Number:	**612995**
Alternate Name:	CD8α; CD8A; CD8 alpha; Leu2a; MAL; T8; p32
Size:	25 Tests
Vol. per Test:	5 µl
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 ≤0.09% sodium azide.

Description

The SK1 monoclonal antibody specifically binds to CD8 alpha (CD8α). CD8α is a type I transmembrane glycoprotein and a member of the immunoglobulin superfamily. CD8α is expressed by the majority of thymocytes, by subpopulations of αβ T cells and γδ T cells and by some NK cells. Cell surface CD8α is expressed either as a disulfide-linked homodimer (CD8αα) or as a heterodimer (CD8αβ) when disulfide-bonded to a CD8 beta chain (CD8β). CD8-positive αβ T cells coexpress both CD8αα homodimers and CD8αβ heterodimers whereas some γδ T cells and NK cells express CD8αα homodimers. CD8 plays important roles in T cell activation and selection. The extracellular IgSF domain of CD8α binds to a non-polymorphic determinant on HLA class I molecules (α3 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 CD8α associates with Lck, a Src family protein tyrosine kinase that is involved in intracellular signaling.

The antibody was conjugated to BD Horizon BUV615 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 615 nm. BD Horizon Brilliant BUV615 can be excited by the ultraviolet laser (355 nm) and detected with a 610/20 filter and a 595 nm LP. Due to the excitation of the acceptor dye by the blue/yellow-green laser line, there may be significant spillover into channels detecting PE-CF594 like emissions (eg, 610/20-nm filter).



Multiparameter flow cytometric analysis of CD8 expression on human peripheral blood leucocyte populations. *Whole blood was stained with either BD Horizon BUV615 Mouse IgG1, κ Isotype Control (Cat. No. 612986; Left Plot) or BD Horizon BUV615 Mouse Anti-Human CD8 antibody (Cat. No. 612994/612995; Right Plot). The erythrocytes were lysed with BD FACS™ Lysing Solution (Cat. No. 349202). A two-parameter pseudocolor density plot showing the correlated expression of CD8 (or Ig Isotype control staining) versus side light-scatter (SSC-A) signals was derived from gated events with the forward and side light-scatter characteristics of intact leucocyte populations. Flow cytometry and data analysis were performed using a BD LSRFortessa™ X-20 Cell Analyzer System and FlowJo™ software.*

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 to the dye under optimum conditions and unconjugated antibody and free dye were removed.

Application Notes

Application

Flow cytometry	Routinely Tested

BD Biosciences

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

For country contact information, visit **bdbiosciences.com/contact**

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)
612986	BUV615 Mouse IgG1, κ Isotype Control	50 µg	X40
612994	BUV615 Mouse Anti-Human CD8	100 Tests	SK1
555899	Lysing Buffer	100 mL	(none)
349202	BD FACS™ Lysing Solution	100 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-µl 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. 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 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.
6. BD Horizon Brilliant Ultraviolet 615 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.
7. Please refer to http://regdocs.bd.com to access safety data sheets (SDS).
8. Species testing during development may have been performed with a different format of the same clone. Selected applications have been tested for cross-reactivity.
9. Please refer to www.bdbiosciences.com/us/s/resources for technical protocols.

References

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Evans RL, Wall DW, Platsoucas CD, et al. Thymus-dependent membrane antigens in man: inhibition of cell-mediated lympholysis by monoclonal antibodies to TH2 antigen. *Proc Natl Acad Sci U S A.* 1981; 78(1):544-548. (Immunogen: Flow cytometry, Functional assay, Inhibition)

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