Technical Data Sheet

BB700 Mouse Anti-Human CD3

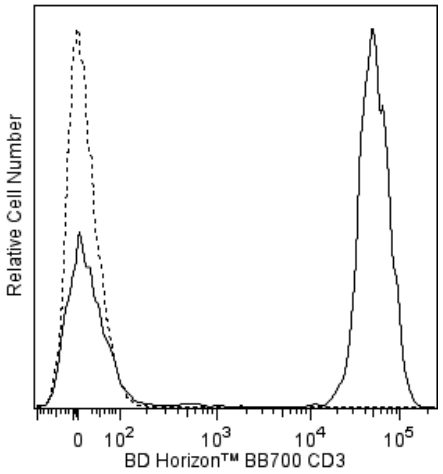
Product Information

Material Number:	**566575**
Alternate Name:	CD3-epsilon; CD3E; Leu4; T-cell surface antigen T3/Leu-4 epsilon chain; T3E
Size:	100 Tests
Vol. per Test:	5 µl
Clone:	SK7 (also known as Leu-4)
Immunogen:	Human Thymocytes
Isotype:	Mouse (BALB/c) IgG1, κ
Reactivity:	QC Testing: Human
Workshop:	II T118; III T492
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The SK7 (Leu-4) monoclonal antibody specifically binds to the epsilon chain of the CD3 antigen/T-cell antigen receptor (TCR) complex. This complex is composed of at least six proteins that range in molecular weight from 20 to 30 kDa. The antigen recognized by CD3 antibodies is noncovalently associated with either α/β or γ/δ TCR (70 to 90 kDa). The CD3 antigen is present on 61% to 85% of normal peripheral blood lymphocytes 60% to 85% of thymocytes and on Purkinje cells in the cerebellum. The soluble form of this antibody has a mitogenic effect on most peripheral blood T lymphocytes, provided appropriate functional monocytes are present.

The antibody was conjugated to BD Horizon BB700, which is part of the BD Horizon Brilliant™ Blue family of dyes. It is a polymer-based tandem dye developed exclusively by BD Biosciences. With an excitation max of 485 nm and an emission max of 693 nm, BD Horizon BB700 can be excited by the 488 nm laser and detected in a standard PerCP-Cy™5.5 set (eg, 695/40-nm filter). This dye provides a much brighter alternative to PerCP-Cy5.5 with less cross laser excitation off the 405 nm and 355 nm lasers.



Flow cytometric analysis of CD3 expression on human peripheral blood lymphocytes. *Whole blood was stained with either BD Horizon™ BB700 Mouse IgG1, κ Isotype Control (Cat. No. 566404; dashed line histogram) or BD Horizon BB700 Mouse Anti-Human CD3 antibody (Cat. No. 566575/566576; solid line histogram). Erythrocytes were lysed with BD FACS™ Lysing Solution (Cat. No. 349202). The fluorescence histogram showing CD3 expression (or Ig Isotype control staining) was derived from gated events with the forward and side light-scatter characteristics of intact lymphocytes. Flow cytometric analysis was performed using a BD FACSCelesta™ 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™ BB700 under optimum conditions, and unconjugated antibody and free BD Horizon BB700 were removed.

Application Notes

Application

Flow cytometry	Routinely Tested

BD Biosciences

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Recommended Assay Procedure:

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

When setting up compensation, it is recommended to compare spillover values obtained from cells and BD™ CompBeads to ensure that beads will provide sufficiently accurate spillover values.

For optimal results, it is recommended to perform two washes after staining with antibodies. Cells may be prepared, stained with antibodies and washed twice with wash buffer per established protocols for immunofluorescent staining, prior to acquisition on a flow cytometer. Performing fewer than the recommended wash steps may lead to increased spread of the negative population.

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	100 Tests	(none)
566349	Brilliant Stain Buffer	1000 Tests	(none)
349202	BD FACS™ Lysing Solution	100 mL	(none)
555899	Lysing Buffer	100 mL	(none)
566576	BB700 Mouse Anti-Human CD3	25 Tests	SK7
566404	BB700 Mouse IgG1, κ Isotype Control	50 µg	X40
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. The manufacture, use, sale, offer for sale, or import of this product is subject to one or more patents or pending applications. This product, and only in the amount purchased by buyer, may be used solely for buyer's own internal research, in a manner consistent with the accompanying product literature. No other right to use, sell or otherwise transfer (a) this product, or (b) its components is hereby granted expressly, by implication or by estoppel. Diagnostic uses require a separate license.
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. BD Horizon Brilliant Blue 700 is covered by one or more of the following US patents: 8,455,613 and 8,575,303.
8. Cy is a trademark of GE Healthcare.
9. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

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