# **Technical Data Sheet**

# **BUV395 Mouse Anti-Human CD3**

#### **Product Information**

Material Number: 563548

Alternate Name: CD3e; CD3E; T3E; TCRE; T-cell surface antigen T3/Leu-4 epsilon

Immunogen: Human infant thymocytes and peripheral blood lymphocytes from a Sézary

Syndrome donor

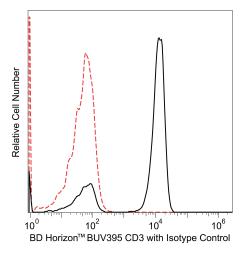
Workshop: III 471

**Storage Buffer:** Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

#### Description

The UCHT1 monoclonal antibody specifically binds to the human CD3ε-chain, a 20-kDa subunit of the CD3/T cell antigen receptor complex. CD3ε is expressed on 70-80% of normal human peripheral blood lymphocytes and 60-85% of thymocytes. Studies from the HLDA Workshop show that this antibody is mitogenic for CD3ε-positive cells when used in conjunction with costimulatory agents such as pokeweed mitogen or anti-CD28 antibody. CD3 plays a central role in signal transduction during antigen recognition. The UCHT1 antibody stains both surface and intracellular CD3ε unlike the other CD3 clone, HIT3a, that stains only extracellular CD3ε.

The antibody was conjugated to BD Horizon™ BUV395 which has been exclusively developed by BD Biosciences as an optimal dye for use on a 355 nm laser equipped instrument. With an Ex Max at 348 nm and an Em Max at 395 nm, this dye has virtually no spillover into any other detector. BD Horizon™ BUV395 can be excited with a 355 nm laser and detected with a 379/28 filter.



Flow cytometric analysis of CD3 expression on human peripheral blood lymphocytes. Human whole blood was stained with the BD Horizon™ BUV395 Mouse Anti-Human CD3 antibody (Cat. No. 563546/563548; solid line histogram) or with BD Horizon™ BUV395 Mouse IgG1, κ Isotype Control (Cat. No. 563547; dashed line histogram). The erythrocytes were lysed with BD FACS™ Lysing Solution (Cat. No. 349202). The fluorescence histograms were derived from events with the forward and side light-scatter characteristics of intact lymphocytes. Flow cytometric analysis was performed using a BD LSR™ II Flow Cytometry 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™ BUV395 under optimum conditions, and unconjugated antibody and free BD Horizon™ BUV395 were removed.

#### **Application Notes**

Application

Flow cytometry Routinely Tested

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### **Suggested Companion Products**

Catalog Number	Name	Size	Clone
554656	Stain Buffer (FBS)	500 ml	(none)
563547	BUV395 Mouse IgG1, k Isotype Control	50 μg	X40
563546	BUV395 Mouse Anti-Human CD3	100 tests	UCHT1
349202	BD FACS <sup>TM</sup> Lysing Solution	100 ml	(none)

### **Product Notices**

- 1. An isotype control should be used at the same concentration as the antibody of interest.
- 2. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
- 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.
- For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
- 5. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1 × 10<sup>6</sup> cells in a 100-µl experimental sample (a test).

#### References

Beverley PC, Callard RE. Distinctive functional characteristics of human "T" lymphocytes defined by E rosetting or a monoclonal anti-T cell antibody. *Eur J Immunol*. 1981; 11(4):329-334. (Clone-specific: Flow cytometry, Fluorescence activated cell sorting)

Burns GF, Boyd AW, Beverley PC. Two monoclonal anti-human T lymphocyte antibodies have similar biologic effects and recognize the same cell surface antigen. *J Immunol.* 1982; 129(4):1451-1457. (Clone-specific: Blocking, Functional assay, Immunoprecipitation, Inhibition, Radioimmunoassay) Ernst DN, Shih CC. CD3 complex. *J Biol Regul Homeost Agents*. 2000; 14(3):226-229. (Biology)

Knapp W, Dörken B, Gilks WR, et al, ed. Leucocyte Typing IV. New York, NY: Oxford University Press; 1989:1-1182. (Clone-specific: Flow cytometry)

McMichael AJ, Beverly PCL, Gilks W, et al, ed. Leukocyte Typing III: White Cell Differentiation Antigens. New York: Oxford University Press; 1987. (Clone-specific: Flow cytometry)

Van Wauwe JP, Goossens JG, Beverley PC. Human T lymphocyte activation by monoclonal antibodies; OKT3, but not UCHT1, triggers mitogenesis via an interleukin 2-dependent mechanism. *J Immunol.* 1984; 133(1):129-132. (Clone-specific: Flow cytometry, Functional assay, Stimulation)

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