

## Technical Data Sheet

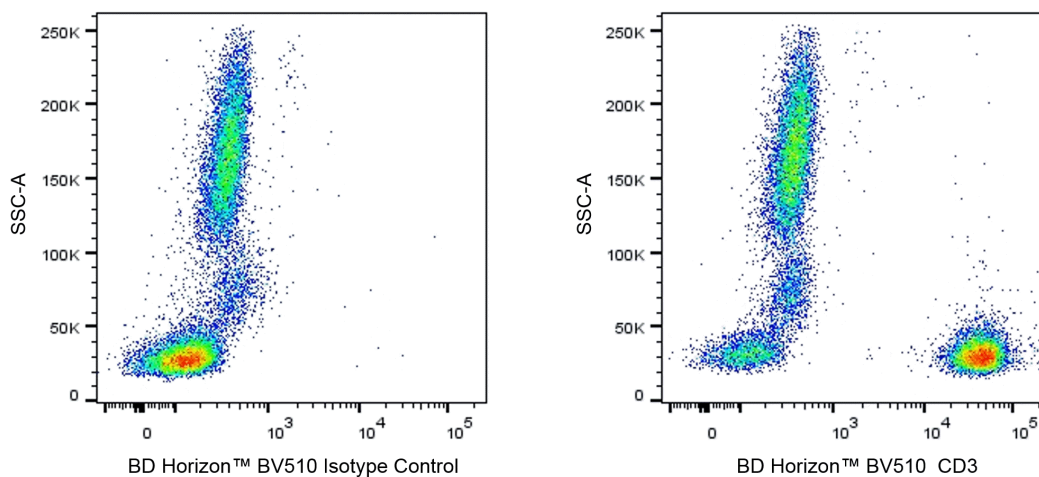
**BV510 Mouse Anti-Human CD3****Product Information**

<b>Material Number:</b>	<b>566780</b>
<b>Alternate Name:</b>	CD3E; CD3ε; T-cell surface antigen T3/Leu-4 epsilon; T3E; TCRE
<b>Size:</b>	25 Tests
<b>Vol. per Test:</b>	5 µl
<b>Clone:</b>	OKT3
<b>Immunogen:</b>	Sheep Erythrocyte Rosette-purified Human T Cells
<b>Isotype:</b>	Mouse (BALB/c x A/J, also known as CAF1) IgG2a, κ
<b>Reactivity:</b>	QC Testing: Human
<b>Storage Buffer:</b>	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

**Description**

The OKT3 monoclonal antibody specifically recognizes the CD3 epsilon subunit (CD3ε/CD3ε) of the CD3 complex which consists of four transmembrane proteins (γ, δ, ε, ζ) that are associated with the T cell antigen receptor (TCR) to form the CD3/TCR complex. The CD3 complex associates with either TCR αβ or TCR γδ heterodimers that are alternatively expressed by some thymocytes, T cells or NKT cells. The CD3 complex is required for the cell surface expression and signal-transducing functions of the TCR. The CD3 complex is expressed by ~60-85% thymocytes and by all peripheral mature T cells. CD3ε is also known as T3E or TCRE. CD3ε is a ~20 kDa unglycosylated type I transmembrane protein that is encoded by *CD3E* which belongs to the immunoglobulin superfamily (IgSF). CD3ε has an Ig-like extracellular domain (ECD) and an immunoreceptor tyrosine-based activation motif (ITAM) in its cytoplasmic domain. The OKT3 antibody can reportedly fix complement, stimulate T cell proliferation and cytokine production, and block the binding of other human CD3ε-specific antibodies including UCHT1 and SK7.

The antibody was conjugated to BD Horizon BV510 which is part of the BD Horizon Brilliant™ Violet family of dyes. With an Ex Max of 405-nm and Em Max at 510-nm, BD Horizon BV510 can be excited by the violet laser and detected in the BD Horizon V500 (525/50-nm) filter set. BD Horizon BV510 conjugates are useful for the detection of dim markers off the violet laser.



**Multiparameter flow cytometric analysis of CD3 expression on human peripheral blood lymphocytes.** Human whole blood was stained with either BD Horizon™ BV510 Mouse IgG2a, κ Isotype Control (Cat. No. 563027; Left Plot) or BD Horizon BV510 Mouse Anti-Human CD3 antibody (Cat. No. 566779/566780; Right Plot). Erythrocytes were lysed with BD Pharm Lyse™ Lysing Buffer (Cat. No. 555899). The two-parameter pseudocolor dot plot showing the correlated expression of CD3 (or Ig Isotype control staining) versus side light-scatter (SSC) signals was derived from gated events with the forward and side light-scatter characteristics of viable leucocyte populations. Flow cytometry and data analysis were performed using a BD FACSCelesta™ 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 with BD Horizon™ BV510 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV510 were removed.

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## Application Notes

### Application

Flow cytometry

Routinely Tested

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

### 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)
566385	Brilliant Stain Buffer Plus	1000 Tests	(none)
555899	Lysing Buffer	100 mL	(none)
349202	BD FACS™ Lysing Solution	100 mL	(none)
563027	BV510 Mouse IgG2a, $\kappa$ Isotype Control	50 $\mu$ g	G155-178
566779	BV510 Mouse Anti-Human CD3	100 Tests	OKT3

### Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use  $1 \times 10^6$  cells in a 100- $\mu$ l experimental sample (a test).
2. An isotype control should be used at the same concentration as the antibody of interest.
3. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
4. 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.
5. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at [www.bdbiosciences.com/colors](http://www.bdbiosciences.com/colors).
6. BD Horizon Brilliant Violet 510 is covered by one or more of the following US patents: 8,575,303; 8,354,239.
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. Please refer to <http://regdocs.bd.com> to access safety data sheets (SDS).
9. Please refer to [www.bdbiosciences.com/us/s/resources](http://www.bdbiosciences.com/us/s/resources) for technical protocols.

### References

Horibe K, Knowles RW, Naito K, Morishima Y, Dupont B. Analysis of T lymphocyte antibody specificities: Comparison of serology with immunoprecipitation patterns. In: Bernard A. A. Bernard .. et al., ed. *Leucocyte typing : human leucocyte differentiation antigens detected by monoclonal antibodies*. Berlin New York: Springer-Verlag; 1984:212-224. (Clone-specific: Flow cytometry)

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, Radioimmunoassay)

Semnani R, Nutman TB, Corrado G, Hochman P, Shaw S, Van Severter GA. Costimulation mediated by purified ICAM-1 and LFA-3 regulates differential stimulation and cytokine secretion of human 'naive' and 'memory' CD4+ T cells. In: Schlossman SF, Stuart F, Schlossman .. et al., ed. *Leucocyte typing V : white cell differentiation antigens*. Oxford: Oxford University Press; 1995:1488-1491. (Clone-specific: Functional assay)

Ernst DN, Shih CC. CD3 complex. *J Biol Regul Homeost Agents*. 2000; 14(3):226-229. (Biology)

Kung P, Goldstein G, Reinherz EL, Schlossman SF. Monoclonal antibodies defining distinctive human T cell surface antigens. *Science*. 1979; 206(4416):347-349. (Immunogen: Cytotoxicity, Flow cytometry, Radioimmunoassay)

Li B, Wang H, Dai J, et al. Construction and characterization of a humanized anti-human CD3 monoclonal antibody 12F6 with effective immunoregulation functions. *Immunology*. 2005; 116(4):487-498. (Clone-specific: Blocking, Flow cytometry)

Touraine JL, Favrot MC, Ansary ME, Cordier G, de bouteiller O. Phenotype of prothymocytes from human bone marrow determined by monoclonal antibodies: Modification induced by thymic factors. In: Bernard A. A. Bernard .. et al., ed. *Leucocyte typing : human leucocyte differentiation antigens detected by monoclonal antibodies*. Berlin New York: Springer-Verlag; 1984:298-311. (Clone-specific: Flow cytometry)

Emmrich F. Selective stimulation of human CD4 and CD8 T-cells by crosslinking the T-cell receptor with subset-specific differentiation antigens. In: McMichael AJ, A.J. McMichael .. et al., ed. *Leucocyte typing III : white cell differentiation antigens*. Oxford New York: Oxford University Press; 1987:203-206. (Clone-specific: Functional assay)

Kurrie R, Seyfert W, Trautwein A, Seiler FR. T cell activation by CD3 antibodies. In: Reinherz EL, Ellis L, Reinherz .. et al., ed. *Leucocyte typing II*. New York: Springer-Verlag; 1986:137-146. (Clone-specific: Functional assay)

Tunnacliffe A, Olsson C, Traunacker A, Krissansen GW, Karjalainen K, de la Hera A. The majority of CD3 epitopes are conferred by the epsilon chain. In: Knapp W. W. Knapp .. et al., ed. *Leucocyte typing IV : white cell differentiation antigens*. Oxford New York: Oxford University Press; 1989:295-296. (Clone-specific: Immunoprecipitation)

Van Wauwe JP, De Mey JR, Goossens JG. OKT3: a monoclonal anti-human T lymphocyte antibody with potent mitogenic properties. *J Immunol*. 1980; 124:2708-2713. (Clone-specific: Functional assay)

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: Functional assay)