### **Technical Data Sheet**

# **BV421 Mouse Anti-Human CD3**

#### **Product Information**

Material Number: 563798

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

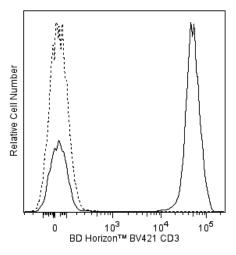
 Workshop:
 II T118; III T492

**Storage Buffer:** Aqueous buffered solution containing BSA and ≤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  $\alpha/\beta$  or  $\gamma/\delta$  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 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<sup>TM</sup> BV421 which is part of the BD Horizon<sup>TM</sup> Brilliant Violet<sup>TM</sup> family of dyes. With an Ex Max of 407-nm and Em Max at 421-nm, BD Horizon<sup>TM</sup> BV421 can be excited by the violet laser and detected in the standard Pacific Blue<sup>TM</sup> filter set (eg, 450/50-nm filter). BD Horizon<sup>TM</sup> BV421 conjugates are very bright, often exhibiting a 10 fold improvement in brightness compared to Pacific Blue<sup>TM</sup> conjugates.



Flow cytometric analysis of CD3 expression on human peripheral blood lymphocytes. Whole blood was stained with BD Horizon™ BV421 Mouse Anti-Human CD3 antibody (Cat. No. 563797/563798; solid line histogram) or BD Horizon™ BV421 Mouse IgG1, κ Isotype Control (Cat. No. 562438; dashed 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™ BV421 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV421 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)
562438	BV421 Mouse IgG1, k Isotype Control	50 μg	X40
563797	BV421 Mouse Anti-Human CD3	25 tests	SK7
349202	BD FACS™ Lysing Solution	100 ml	(none)
555899	Lysing Buffer	100 ml	(none)

### **Product Notices**

- 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).
- 2. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
- 3. An isotype control should be used at the same concentration as the antibody of interest.
- 4. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- 5. 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.
- 7. Pacific Blue<sup>TM</sup> is a trademark of Molecular Probes, Inc., Eugene, OR.
- 8. Brilliant Violet<sup>TM</sup> 421 is a trademark of Sirigen.

#### References

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Kan EA, Wang CY, Wang LC, Evans RL. Noncovalently bonded subunits of 22 and 28 kd are rapidly internalized by T cells reacted with anti-Leu-4 antibody. *J. Immunol.* 1983: 131(2):536-539. (Clone-specific: Flow cytometry. Functional assay. Immunofluorescence. Immunoprecipitation)

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Knowles RW. Immunochemical analysis of the T-cell–specific antigens. In: Reinherz EL, Haynes BF, Nadler LM, Bernstein ID, ed. Leukocyte Typing II: Human T Lymphocytes. New York, NY: Springer-Verlag; 1986:259-288. (Clone-specific: Immunoprecipitation)

Kurrie R, Seyfert W, Trautwein A, Seiler FR. T cell activation by CD3 antibodies. In: Reinherz EL, Haynes BF, Nadler LM, Bernstein ID, ed. *Leukocyte Typing II: Human T Lymphocytes*. New York, NY: Springer-Verlag; 1986:137-146. (Clone-specific: Activation, Flow cytometry, Functional assay, Stimulation)

Lanier LL, Allison JP, Phillips JH. Correlation of cell surface antigen expression on human thymocytes by multi-color flow cytometric analysis: implications for differentiation. *J Immunol.* 1986; 137(8):2501-2507. (Clone-specific: Immunoprecipitation)

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Schlossman SF, Boumsell L, Gilks W, et al, ed. *Leukocyte Typing V: White Cell Differentiation Antigens*. New York: Oxford University Press; 1995. (Biology) van Dongen JJM, Krissansen GW, Wolvers-Tettero ILM, et al. Cytoplasmic expression of the CD3 antigen as a diagnostic marker for immature T-cell malignanacies. *Blood.* 1988; 71(3):603-612. (Clone-specific: Immunofluorescence, Western blot)

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