

## Technical Data Sheet

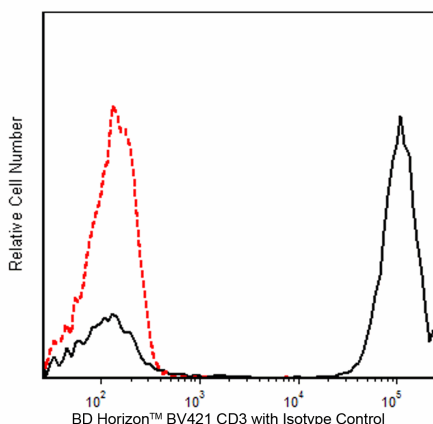
**BV421 Mouse Anti-Human CD3****Product Information**

<b>Material Number:</b>	<b>562426</b>
<b>Alternate Name:</b>	CD3ε; CD3E; T3E; TCRE ; T-cell surface antigen T3/Leu-4 epsilon
<b>Size:</b>	100 tests
<b>Vol. per Test:</b>	5 µl
<b>Clone:</b>	UCHT1
<b>Immunogen:</b>	Human infant thymocytes and peripheral blood lymphocytes from a Sézary Syndrome donor
<b>Isotype:</b>	Mouse (BALB/c) IgG1, κ
<b>Reactivity:</b>	QC Testing: Human
<b>Workshop:</b>	III 471
<b>Storage Buffer:</b>	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™ BV421 which is part of the BD Horizon™ Brilliant Violet™ family of dyes. With an Ex Max of 407-nm and Em Max at 421-nm, BD Horizon™ BV421 can be excited by the violet laser and detected in the standard Pacific Blue™ filter set (eg, 450/50-nm filter). BD Horizon™ BV421 conjugates are very bright, often exhibiting a 10 fold improvement in brightness compared to Pacific Blue™ conjugates.



**Flow cytometric analysis of CD3 expression on human peripheral blood lymphocytes.** Human whole blood was stained with the BD Horizon™ BV421 Mouse anti-Human CD3 antibody (Cat. No. 562426/562427; solid line histogram) or with BD Horizon™ BV421 Mouse IgG1, κ Isotype Control (Cat. No. 562438; dashed line histogram). The erythrocytes were lysed with BD Pharm Lyse™ Lysing Buffer (Cat. No. 555899). The fluorescence histograms were derived from events with the forward and side light-scatter characteristics of viable lymphocytes. Flow cytometry was performed using a BD FACSCanto™ 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)
555899	Lysing Buffer	100 ml	(none)
562438	BV421 Mouse IgG1, k Isotype Control	50 µg	X40
562427	BV421 Mouse Anti-Human CD3	25 tests	UCHT1

## 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-µl experimental sample (a test).
2. An isotype control should be used at the same concentration as the antibody of interest.
3. Brilliant Violet™ 421 is a trademark of Sirigen.
4. Pacific Blue™ is a trademark of Molecular Probes, Inc., Eugene, OR.
5. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
6. 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.
7. 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).
8. Please refer to [www.bdbiosciences.com/pharmingen/protocols](http://www.bdbiosciences.com/pharmingen/protocols) for technical protocols.

## 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)

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

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. (Biology)

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

Schlossman SF, Boumsell L, Gilks W, et al, ed. *Leukocyte Typing V: White Cell Differentiation Antigens*. New York: Oxford University Press; 1995. (Clone-specific)

Zola H, Swart B, Nicholson I, Voss E. *Leukocyte and Stromal Cell Molecules. The CD Markers*. Hoboken, New Jersey: John Wiley & Sons, Inc.; 2007:1-581. (Biology)

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