

## Technical Data Sheet

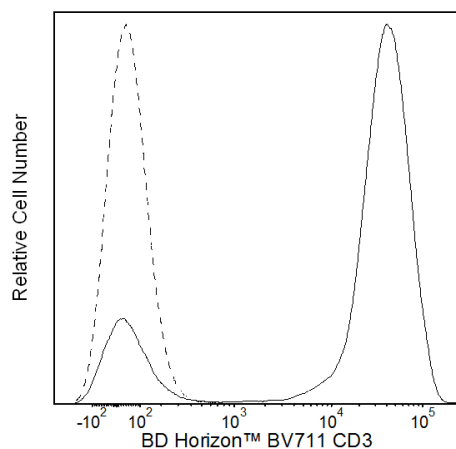
**BV711 Mouse Anti-Human CD3****Product Information**

|                         |                                                                                       |
|-------------------------|---------------------------------------------------------------------------------------|
| <b>Material Number:</b> | <b>563724</b>                                                                         |
| <b>Alternate Name:</b>  | CD3ε; CD3E; T3E; TCRE ; T-cell surface antigen T3/Leu-4 epsilon                       |
| <b>Size:</b>            | 25 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™ BV711 which is part of the BD Horizon™ Brilliant Violet™ family of dyes. This dye is a tandem fluorochrome of BD Horizon™ BV421 with an Ex Max of 405-nm and an acceptor dye with an Em Max at 711-nm. BD Horizon™ BV711 can be excited by the violet laser and detected in a filter used to detect Cy™5.5 / Alexa Fluor® 700-like dyes (eg, 712/20-nm filter). Due to the excitation and emission characteristics of the acceptor dye, there may be moderate spillover into the Alexa Fluor® 700 and PerCP-Cy™5.5 detectors. However, the spillover can be corrected through compensation as with any other dye combination.



**Flow cytometric analysis of CD3 expression on human peripheral blood lymphocytes.** Human whole blood was stained with either BD Horizon™ BV711 Mouse Anti-Human CD3 antibody (Cat. No. 563724/563725; solid line histogram) or BD Horizon™ BV711 Mouse IgG1, κ Isotype Control (Cat. No. 563044; 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 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™ BV711 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV711 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) |
| 563044         | BV711 Mouse IgG1, k Isotype Control | 50 µg  | X40    |
| 349202         | BD FACS™ Lysing Solution            | 100 ml | (none) |
| 555899         | Lysing Buffer                       | 100 ml | (none) |

## 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. 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. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
5. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
6. Cy is a trademark of Amersham Biosciences Limited.
7. Brilliant Violet™ 711 is a trademark of Sirigen.
8. 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).
9. Please refer to [www.bdbiosciences.com/pharming/protocols](http://www.bdbiosciences.com/pharming/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. (Immunogen: 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, Beverley PCL, Gilks W, et al, ed. *Leucocyte 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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