

### MOLECULAR PROBES®

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# PRODUCT INSERT MONOCLONAL ANTIBODIES TO THE HUMAN CD3 ANTIGEN

Product	Form	Volume	Antibody*	Tests	Excitation (nm)	Peak Emission (nm)
CD0300	Purified	0.5 ml	100 μg		N/A	N/A
CD0328	Pacific Blue <sup>TM</sup>	0.5 ml		100 min.	405	455
CD0326	Alexa Fluor® 405	0.5 ml		100 min.	405	421
CD0330	Pacific Orange <sup>TM</sup>	0.5 ml		100 min.	405	551
CD0301	FITC	0.5 ml		100 min.	488	525
CD0304	R-PE	0.5 ml		100 min.	488	575
CD0324	PE- Alexa Fluor® 700	0.5 ml		100 min.	488	723
CD0305	APC	0.5 ml		100 min.	600-650	660
CD0329	Alexa Fluor® 700	0.5 ml		100 min.	630-702	723

#### PRODUCT DESCRIPTION

Mouse monoclonal antibody to the human CD3 antigen. For intracellular staining, Product Code GM4013-5 is recommended.

**Clone:** UCHT1 **Isotype:** Mouse IgG1

Lot No.: See label Expiration: See label

**Buffer:** Phosphate buffered saline (PBS)

**Preservative:** 0.1% *sodium azide*. Sodium azide is an extremely toxic and dangerous compound particularly when combined with acids or metals. Solutions containing sodium azide should be disposed of properly.

**Stabilizer:** For conjugated products only, a highly purified grade of BSA has been added as a stabilizing agent to bring the final protein concentration to 4-5 mg/ml after conjugation.

### STORAGE & HANDLING

Store reagents at 2-8°C. Light exposure should be avoided with fluorochrome conjugated reagents. Use dim light during handling, incubation with cells and prior to analysis. It is recommended that cells be analyzed within 18 hours of staining. If the reagent is being diluted, it is recommended that only the quantity to be used within one week be diluted.

#### PRODUCT CHARACTERIZATION

#### **Antigen Specificity:**

The UCHT1 monoclonal antibody reacts with human CD3e, a 20 kDa subunit of the TCR/CD3 complex  $^1$ . Along with the other CD3 subunits  $\gamma$  and  $\delta$ , the  $\epsilon$  chain is required for proper assembly, trafficking and surface expression of the TCR complex  $^2$ . CD3 is expressed by developing thymocytes and by all mature T cells. Crosslinking of TCR via immobilized UCHT1 results in T cell proliferation.

Leukocyte Workshop Status: Leukocyte Typing III, IV and V.

### PRODUCT QUALITY CONTROL

Each lot is tested by flow cytometry using human peripheral blood leukocytes (PBL). This testing was performed using 5  $\mu$ l of antibody per 1 x  $10^6$  cells in a 100  $\mu$ l staining volume. Because conditions may vary, it is recommended that each investigator determine the optimal amount of antibody to be used for each application. See reverse for representative flow cytometry data.

#### **REFERENCES:**

- Beverley, P., and R. E. Callard. 1981. Euro. J. Immunol. 11(4):329-334.
- Schlossman, S. F., L. Boumsell, W. Gilks, J. M. Harlan, T. Kishimoto, C.Morimoto, J. Ritz, S. Shaw, R. Silverstein, T. Springer, T. F. Tedder, and R. F.Todd eds. 1995. Leukocyte Typing V. Oxford University Press Inc., NewYork.
- Garson, J. A., P. C. Beverley, H. B. Coakham, and E. I. Harper. 1982. Nature 298(5872):375-7.
- Dornan, S., Z. Sebestyen, J. Gamble, P. Nagy, A. Bodnar, L. Alldridge, S. Doe, N. Holmes, L. K. Goff, P. Beverely, J. Szollosi, and D. R. Alexander
- \* Antibody value assigned is based on the Optical Density at 280 nm.

TR, Texas Red®

TC, TRI-COLOR®, PE-Cy®5

The efficiency of energy transfer in tandem dyes can be significantly decreased by exposure to visible light. We recommend that longer wavelength fluorochrome conjugates, e.g. PE-Cy®7, PE-Alexa Fluor® 700, be protected from light during staining and while awaiting analysis, e.g. cover with aluminum foil.

The Texas Red<sup>®</sup>, Alexa Fluor<sup>®</sup>, Pacific Blue<sup>®</sup>, and Pacific Orange<sup>TM</sup> dye conjugates in this product are sold under license from Molecular Probes, Inc., for research use only or as analyte specific reagents, except for use in combination with microarrays or high content screening, and are covered by pending and issued patents.

Cy® is a trademark of GE/Amersham Biosciences.

#### ANALYTE SPECIFIC REAGENT, ANALYTICAL AND PERFORMANCE CHARACTERISTICS ARE NOT ESTABLISHED.

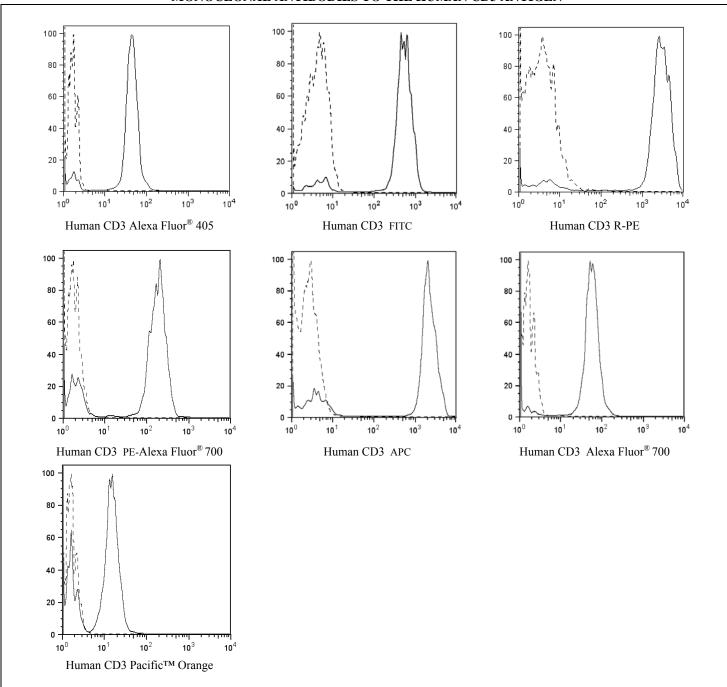
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Log fluorescence intensity profiles of human peripheral blood lymphocytes analyzed on a FACSCalibur<sup>TM</sup>, FACScan<sup>TM</sup>, FACS Vantage<sup>TM</sup> or BD<sup>TM</sup> LSR II flow cytometer, and analyzed using CellQuest<sup>TM</sup> software, BD Biosciences, San Jose, CA or FlowJo<sup>©</sup> software, TreeStar, Inc. (www.flowjo.com)

Negative control profiles represent unstained cells.

**Note:** Flow cytometric data shown may not necessarily have been generated using the enclosed lot of reagent. For this reason, and due to differences in flow cytometers and cytometer settings, results may vary from those illustrated above. It is suggested that investigators titrate reagents to determine optimal conditions for use in their systems.

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