

Mo-CD4/CD69/CD3

3 Color FCM Reagent: sc-3616



BACKGROUND

Mouse CD4/CD69/CD3: sc-3616 is a direct immunofluorescence reagent formatted to identify activated helper/inducer T lymphocytes in erythrocyte-lysed whole blood. CD4 identifies the helper/inducer T lymphocyte subset that is present on normal peripheral blood lymphocytes (1,2). CD4 binds class II MHC molecules (2) and is the primary receptor for HIV (3). CD3 identifies T lymphocytes and non-covalently associates with either α/β or γ/δ TCR (4). CD3+CD8+ and CD3+CD4+ percentages or counts are used to characterize and monitor some forms of immunodeficiency and autoimmune disease (5,6). In normal peripheral blood, CD69 is variably expressed on lymphocytes (7). Upon activation, CD69 expression increases on T, B, and NK lymphocytes (8). In thymus, CD69 is constitutively expressed on the bright CD3+ subset of T cells, mostly on subpopulations of CD4+ CD8- or CD4- CD8+ T cells (8).

Antigen Expression	Cell Type Identified
CD3+	Mature T Cells
CD3+ CD4+	Helper/Inducer T Cells
CD3+ CD4+ CD8- CD69+	Activated Thymocytes

STORAGE

Store at 4° C. Do not freeze. Stable for one year from the date of shipment. Protect reagents from prolonged exposure to light.

PRODUCT

Supplied in 1.0 ml of PBS containing 0.1% azide and 0.1% gelatin. Sufficient for 50 tests. This product has been titrated for optimal performance. Recommended use is 20 μ l per test (1×10^6 cells). **For research use only. Not for use in diagnostic procedures.**

INSTRUMENT

Mouse CD4/CD69/CD3: sc-3616 is recommended for use with a dual laser Flow Cytometer fitted with appropriate acquisition and analysis software, such as the FACSCalibur™ Flow Cytometer fitted with CellQuest™ Software by Becton Dickinson.

The flow cytometer must be equipped with 635 nm and 488 nm lasers and must be capable of detecting light scatter (forward and side) and four-color fluorescence with emission detectable in four ranges: 515-545 nm, 562-607 nm, >650 nm and 652-668 nm, and it must be able to threshold and discriminate using the >650 channel.

Antigen	Clone	Isotype	Label*	Detection Range (nm)
CD4	H129.19	rat IgG _{2a}	FITC	515-545
CD69	H1.2F3	Armenian Hamster IgG	PE	562-607
CD3	145-2C11	Armenian Hamster IgG	PE-Cy5	>650

*Fluorescent labels include FITC: Fluorescein isothiocyanate; PE: phycoerythrin; PE-Cy5: phycoerythrin-cyanin 5; APC: allophycocyanin

ISOTYPE CONTROL

sc-3616 CON (rat IgG_{2a} FITC/Armenian Hamster IgG PE/Armenian Hamster IgG PE-Cy5) is the isotype matched negative control for this system and is suitable for 50 tests.

REFERENCES

- Reichert, T., DeBruyere, M., Deneys, V., Totterman, T., Lydyard, P., Yuksel, F., Chapel, H., Jewell, D., Van Hove, L., Linden, J., *et al.* 1991. Lymphocyte subset reference ranges in adult Caucasians. *Clin. Immunol. Immunopathol.* **60**: 190-208.
- Gallagher, P.F., Fazekas de St. Groth, B., and Miller, J.F. 1989. CD4 and CD8 molecules can physically associate with the same T-cell receptor. *Proc. Natl. Acad. Sci. USA* **86**: 10044-10048.
- Dalglish, A.G., Beverley, P.C.L., Clapham, P.R., Crawford, D.H., Greaves, M.F., and Weiss, R.A. 1984. The CD4 (T4) antigen is an essential component of the receptor for the AIDS retrovirus. *Nature* **312**: 763-767.
- Exley, M., Terhorst, C., and Wileman, T. 1991. Structure, assembly and intracellular transport of the T cell receptor for antigen. *Semin. Immunol.* **3**: 283-297.
- Foucar, K. and Goeken, J.A. 1982. Clinical Applications of immunologic techniques to the diagnosis of lymphoproliferative and immunodeficiency disorders. *Lab. Med.* **13**: 403-413.
- Smolen, J.S., Chused, T.M., Leiserson, W.M., Reeves, J.P., Alling, D., and Steinberg, A.D. 1982. Heterogeneity of immunoregulatory T-cell subsets in systemic lupus erythematosus. Correlation with clinical features. *Am. J. Med.* **72**: 783-790.
- Schwartz, R., Biedobitek, G., and Stein, H. Cluster report: CD69. Knapp, W., Dörken, B., Gilks, W.R., *et al.*, eds. *Leucocyte Typing IV: White Cell Differentiation Antigens*. New York, NY: Oxford University Press; 1989: 428-432.
- Testi, R., Phillips, J.H., and Lanier, L.L. 1988. Constitutive expression of a phosphorylated activation antigen (Leu 23) by CD3bright human thymocytes. *J. Immunol.* **141**: 2557-2563.