

# Hu-CD45RA/CD45RO/CD3/CD8

## 4 Color FCM Reagent: sc-2947



### BACKGROUND

Human CD45RA/CD45RO/CD3/CD8: sc-2947 is a direct immunofluorescence reagent formatted to identify and determine the percentage of mature T cells and suppressor/cytotoxic (naive and memory) T cells in erythrocyte-lysed whole blood, based on cell-surface antigen expression. CD45 is a major leukocyte cell surface molecule that is essential for the activation of T and B lymphocytes (1,2). In T cells, the alternative splicing of CD45 is regulated so that naive or unprimed T cells predominantly express CD45RA-positive isoforms and switch to expression of CD45RO upon activation. CD45RO expression is correlated with the memory T-cell phenotype (3). CD3 identifies T lymphocytes and non-covalently associates with either  $\alpha/\beta$  or  $\gamma/\delta$  TCR, which recognizes antigens associated with the MHC (4). CD8 identifies suppressor/cytotoxic T lymphocytes and binds class I MHC molecules, which enhances the activation of resting T lymphocytes (5).

Antigen Expression	Cell Type Identified
CD3+	Mature T Cells
CD3+ CD8+	Suppressor/Cytotoxic T Cells
CD3+ CD8+ CD45RA+	Suppressor/Cytotoxic Naive T Cells
CD3+ CD8+ CD45RO+	Suppressor/Cytotoxic Memory T Cells

### 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  $\mu$ L per test ( $1 \times 10^6$  cells). **For research use only. Not for use in diagnostic procedures.**

### INSTRUMENT

Human CD45RA/CD45RO/CD3/CD8: sc-2947 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)
CD45RA	4KB5	IgG <sub>1</sub>	FITC	515-545
CD45RO	UCH-L1	IgG <sub>2a</sub>	PE	562-607
CD3	UCH-T1	IgG <sub>1</sub>	PE-Cy5	>650
CD8	HIT8a	IgG <sub>1</sub>	APC	652-668

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

### ISOTYPE CONTROL

sc-2947 CON (IgG<sub>1</sub> FITC/IgG<sub>2a</sub> PE/IgG<sub>1</sub> PE-Cy5/IgG<sub>1</sub> APC) is the isotype matched negative control for this system and is suitable for 50 tests.

### REFERENCES

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