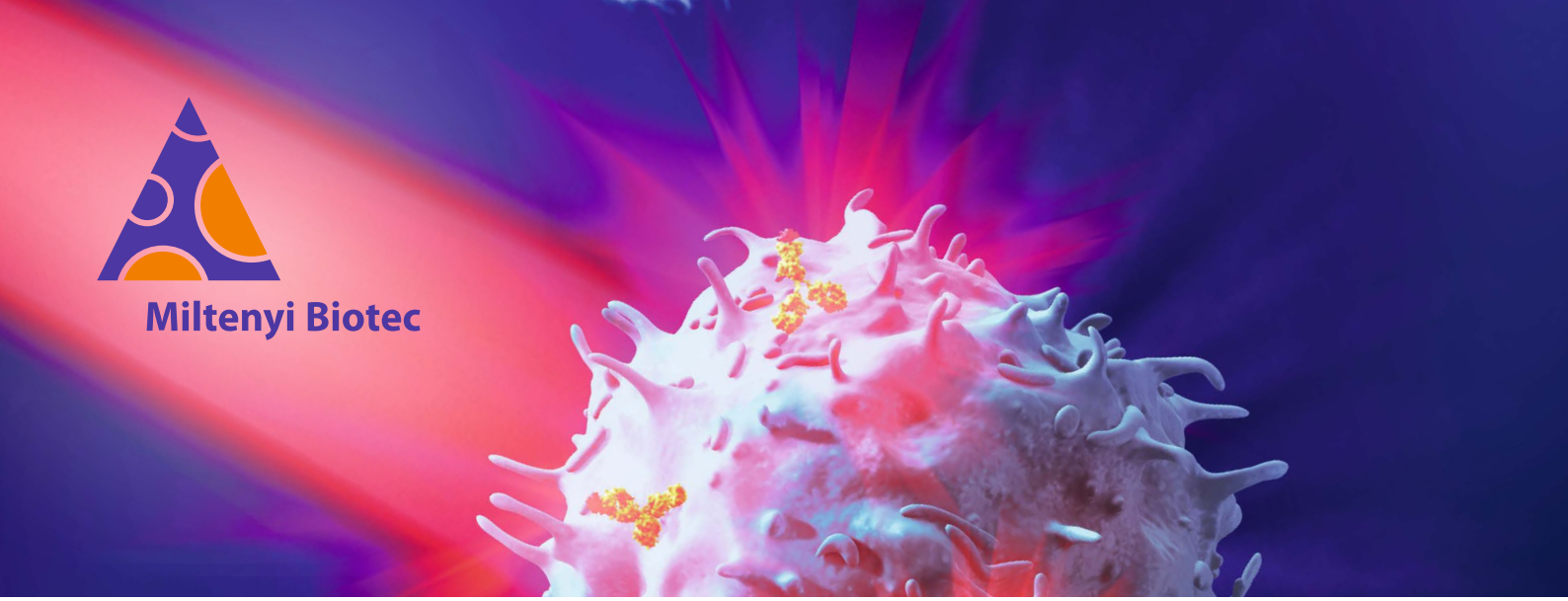

Fluorochrome brightness index

Find the brightest option for your flow cytometry experiments

Flow cytometry has an inherent need to continuously increase the number of fluorochromes to meet the growing demand of sophisticated multicolor analysis. However, fluorochromes vary greatly with regard to their fluorescence intensity. In particular for antigens with low expression levels, it is crucial to select an appropriate fluorochrome-conjugated antibody to obtain sufficiently bright signals for reliable analysis.

In flow cytometric analysis, fluorescence intensity for individual conjugates can be evaluated (table 1) according to the stain index (SI) and the mean fluorescence intensity (MFI). SI is a measurement that takes into account both the distance (D) between the positive and background peaks and the spread (W) of the background peak (fig. 1). SI is defined by the quotient of D/W. MFI defines the mean intensity of the fluorescence measured in a specific channel. MFI does not take into account the spread of the background peak, only the brightness of the positive peak.

SI and MFI values can differ due to a number of variables, including antigen target and antibody clone, the flow cytometer's filters, settings, and laser power, as well as buffer conditions. Therefore, the evaluation in table 1 should be regarded only as an indicator of brightness.

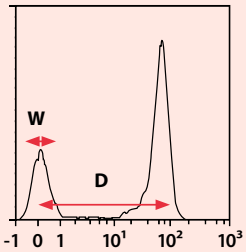

Figure 1: SI is defined by the quotient of D/W, whereas MFI is defined only by the brightness of the positive peak.

In all cases, the individual product pages on our website, including example histograms or dot plots, should be consulted for details on particular fluorochrome-conjugated antibodies. Comprehensive information is available at **www.miltenyibiotec.com/antibodies**

Dye	MFI	SI
Vio® Bright R667	5	5
PE	5	5
Vio R667	5	5
Vio Bright B515	5	5
PE-Vio 615	5	5
Vio Bright R720	4	4
Vio Bright V423	5	4
Vio B515	4	4
Vio Bright FITC	4	4
APC	4	3
PE-Vio 770	4	3
FITC	3	3
Vio R720	3	3
VioBlue®	3	3
PerCP-Vio 700	3	3
APC-Vio 770	3	3
VioGreen™	1	2
PerCP	1	1

Table 1: Data was generated using the MACSQuant® Analyzer 16. Human peripheral blood mononuclear cells were stained with CD4 (REA623) antibodies conjugated to the indicated fluorochromes. Stain indices and MFI values may vary depending on instrument, instrument configuration, reagents, and cell type used (5 = brightest, 1=dim).

Miltenyi Biotec B.V. & Co. KG | Phone +49 2204 8306-0 | Fax +49 2204 85197 | macsde@miltenyi.com | **www.miltenyibiotec.com**
Miltenyi Biotec provides products and services worldwide. Visit **www.miltenyibiotec.com/local** to find your nearest Miltenyi Biotec contact.

V.07