

Spectral

BD Horizon RealBlue™ 545 Reagents

Enhance your data and move to higher parameter with confidence

Excited by the 488-nm blue laser and offer minimal cross-laser excitation off the 561-nm yellow-green laser

BD Horizon RealBlue™ 545 (RB545) Reagents are part of a comprehensive family of laser-specific reagents. The RB545 fluorochrome was specially designed to produce less spillover, which improves resolution and enables high-parameter experiments for spectral flow instruments.

RB545 reagents are well suited for high-expression surface and intracellular markers, can be used together with FITC and easily integrated into existing spectral flow cytometer panels.

RB545 Specifications		
Ex_{max}	Em_{max}	Relative Brightness
496 nm	545 nm	

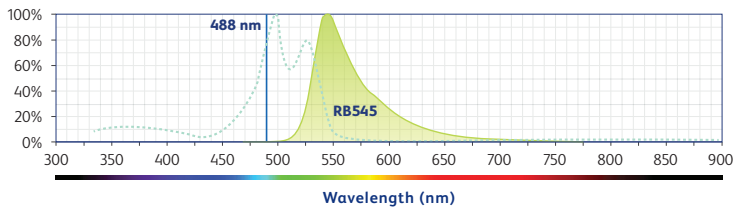

Figure 1. Excitation and emission spectra of the RB545 fluorochrome.

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Comparable brightness and greater resolution relative to FITC

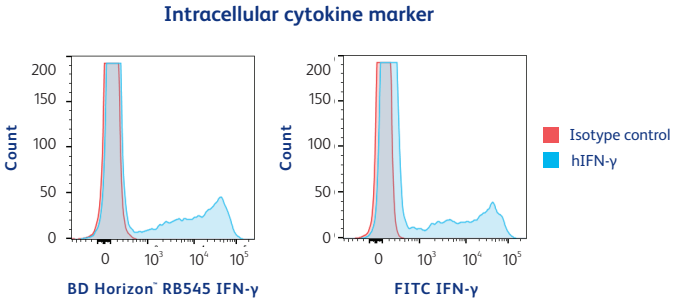

Figure 2. High-expression intracellular markers can be resolved comparably with FITC and BD Horizon™ RB545 Reagents.

PBMCs were stimulated with PMA and ionomycin in the presence of BD GolgiStop™ Protein Transport Inhibitor for 5 hours and then stained with either BD Horizon™ RB545 Reagents or FITC IFN-γ (B27) and matched isotype controls. Samples were acquired on a BD FACSymphony™ A5 SE Cell Analyzer.

Stable performance with lot-to-lot consistency

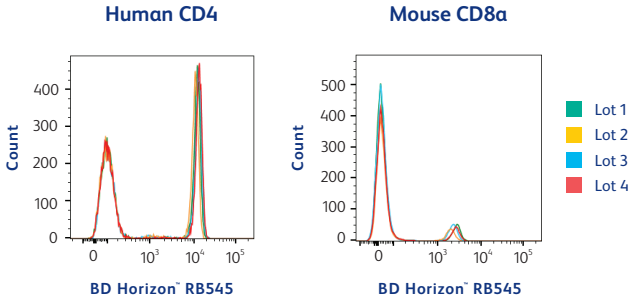

Figure 3. RB545 lot-to-lot consistency is demonstrated across specificities.

Human whole blood was stained with human CD4 (SK3) RB545, followed by lysis with BD FACS™ Lysing Solution using four different batches of reagent. Mouse splenocytes were stained with mouse CD8a (53-6.7) RB545 using four different batches of reagent. All specificities were run on a BD FACSymphony™ A5 SE Cell Analyzer.

RB545 reagents can be used together with FITC to expand parameters in spectral flow cytometry

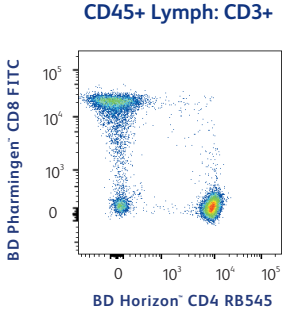

Figure 4. RB545 and FITC used together in a multicolor TBNK panel.

Human whole blood was stained with eight dyes and lysed with BD FACS™ Lysing Solution. Samples were then acquired on a BD FACSymphony™ A5 SE Cell Analyzer and spectrally unmixed using FlowJo™ Software.

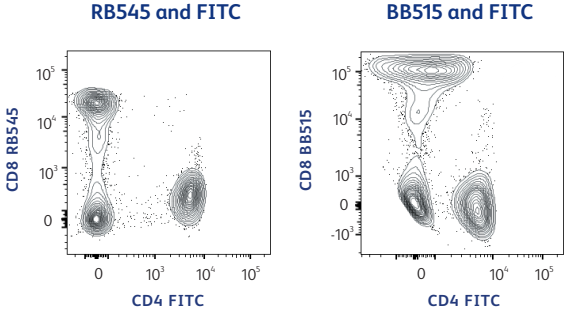

Figure 5. Using RB545 with FITC or the BD Horizon Brilliant™ Blue 515 (BB515) Reagent is a superior alternative to using FITC with BB515.

Human PBMCs were stained with two sets of fluorochromes, moderately similar RB545 and FITC, and highly similar BB515 and FITC. RB545 with FITC provides better population resolution compared to FITC with BB515. Samples were acquired on the BD FACSymphony™ A5 SE Cell Analyzer and spectrally unmixed with FlowJo™ Software.

BD flow cytometers are Class 1 Laser Products.
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