



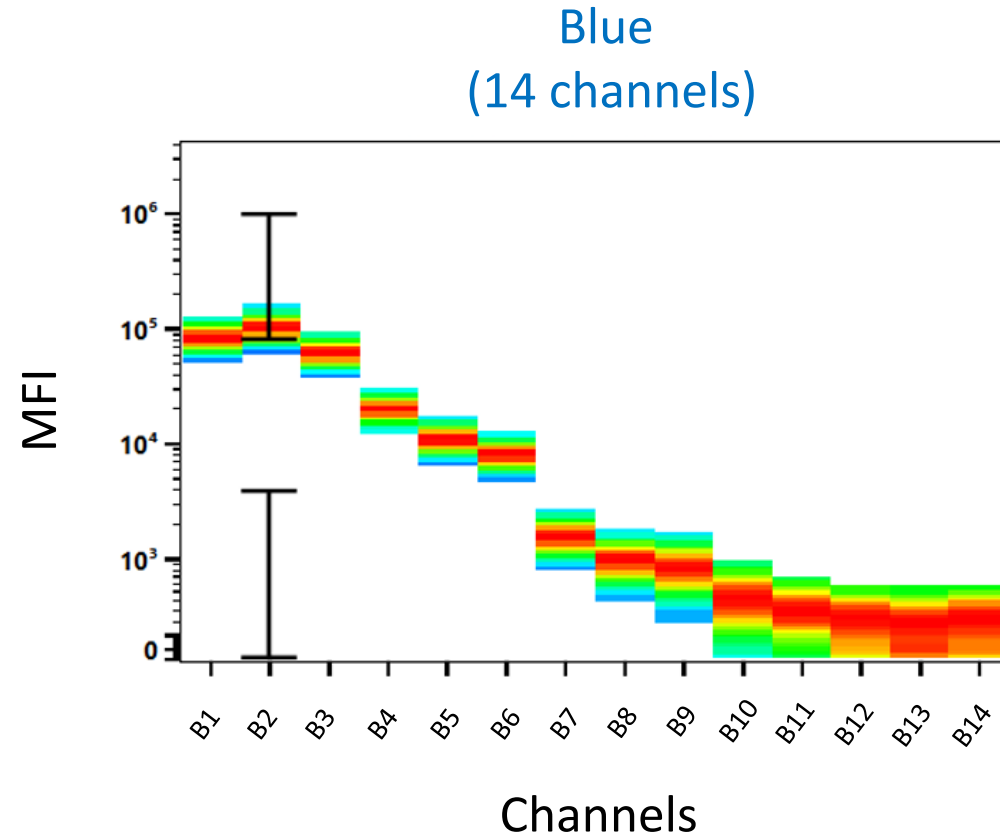
Cytek® Aurora Fluorochrome Selection Guidelines 1 Laser 14B

Fluorochrome Signatures

Dyes can be used in combination if they have unique spectrum signatures.

Look for dyes with unique spectra and consider spread introduced by the dyes when designing multicolor panels (see slide 15).

How to Read Full Spectrum Fluorochrome Signatures

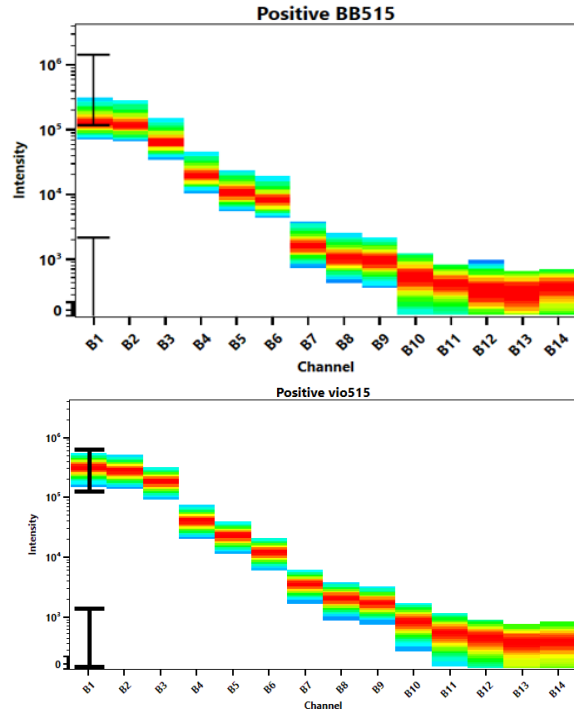


This dye is excited by the blue laser. The peak channel (indicated by the black bar) is in channel B2.

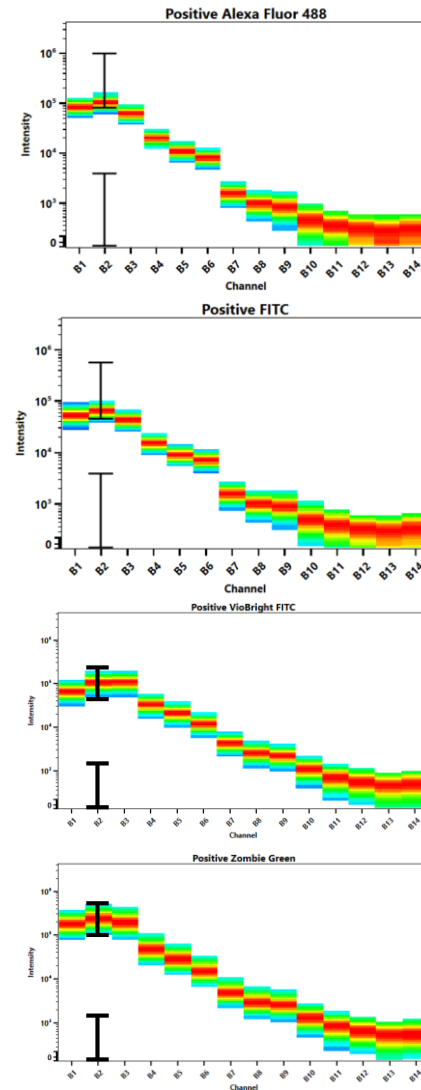
Dyes Primarily Excited by the Blue Laser

Blue Laser Excitable Dyes with Similar Signatures (1 of 2)

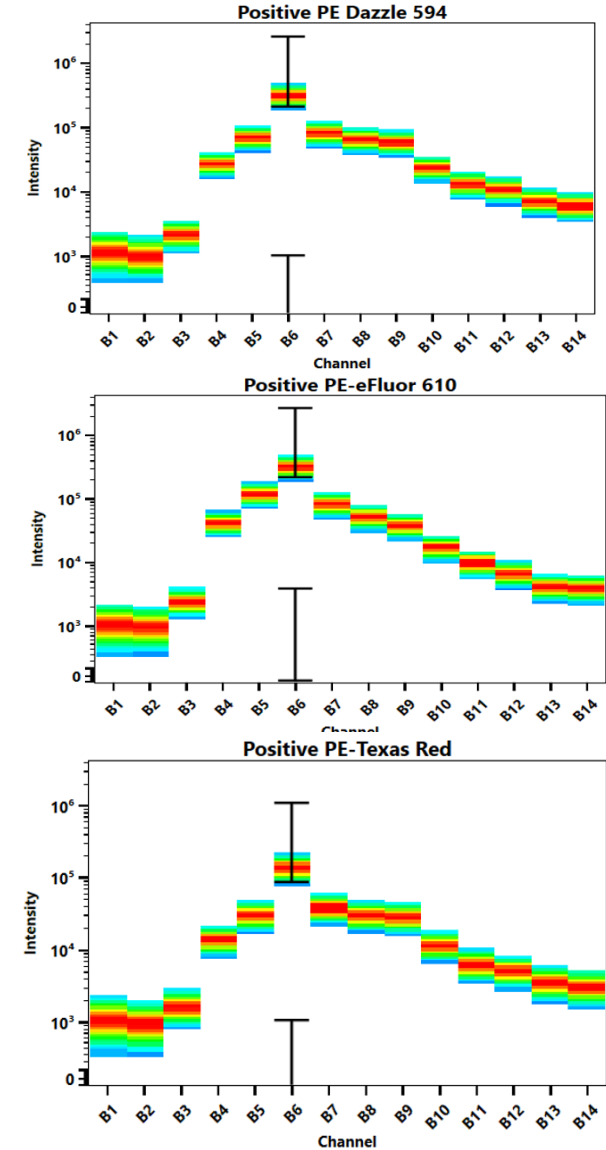
BB515 and vio515



Alexa Fluor 488, FITC, VioBright FITC, and Zombie Green

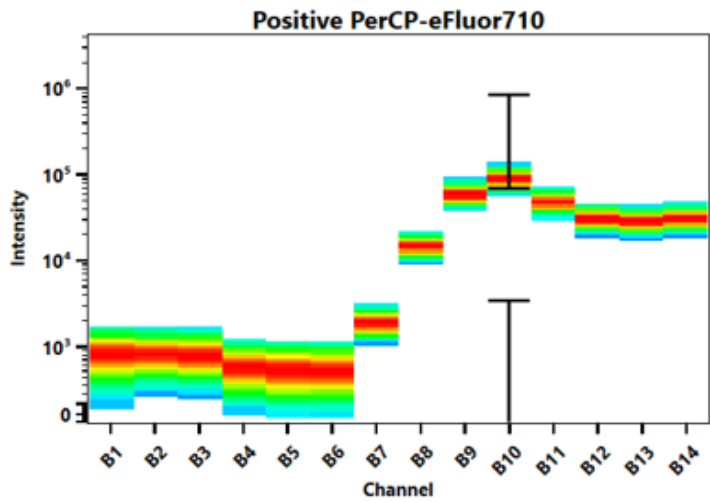
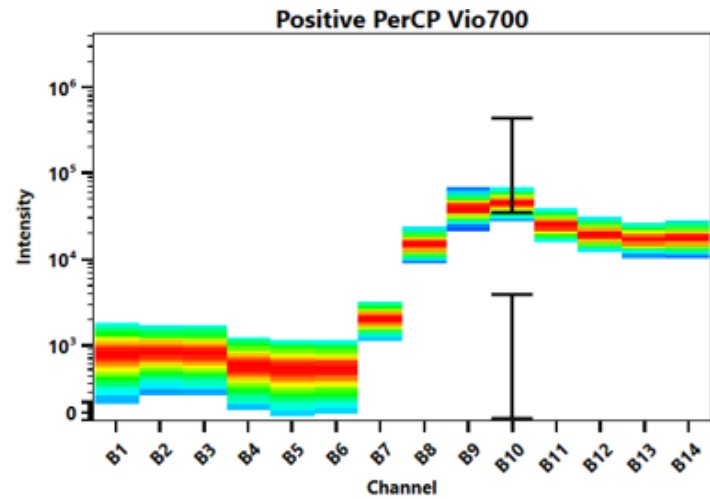


PE/Dazzle 594, PE-eFluor 610 and PE-Texas Red

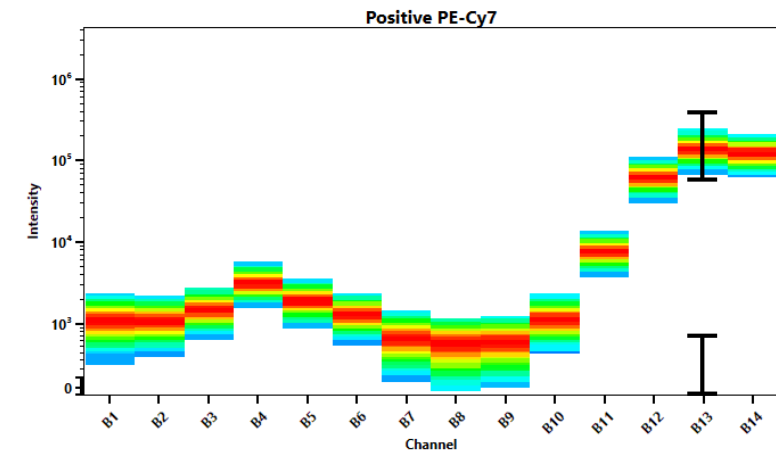
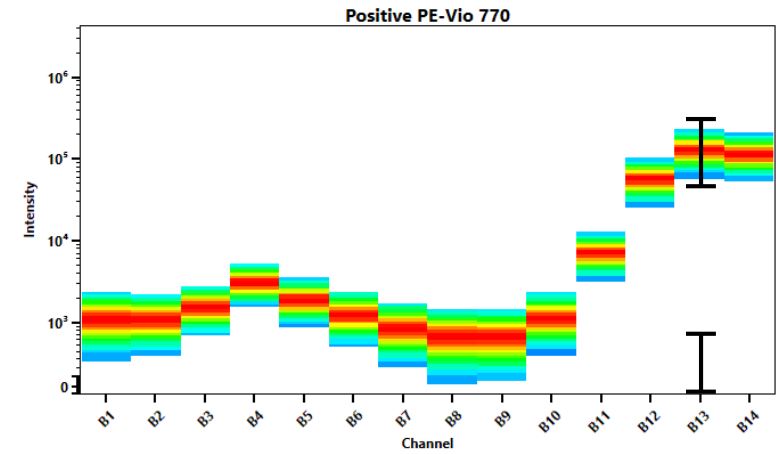


Blue Laser Excitable Dyes with Similar Signatures (2 of 2)

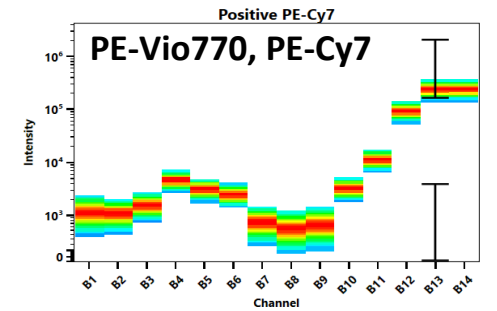
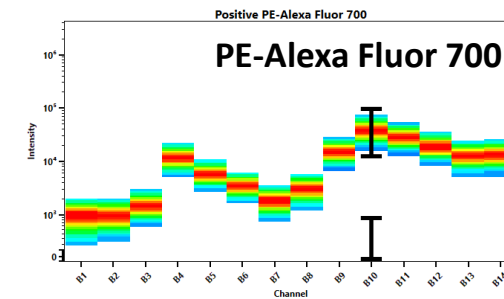
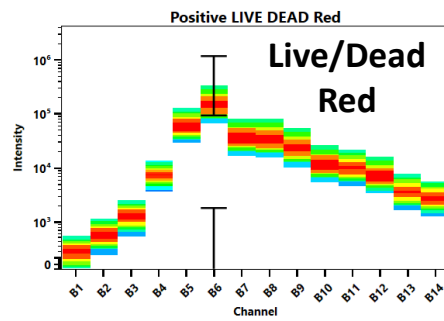
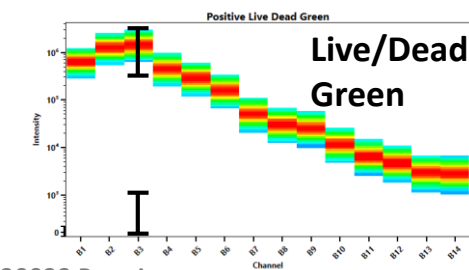
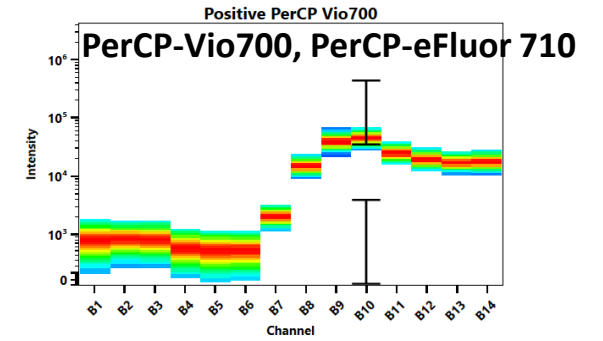
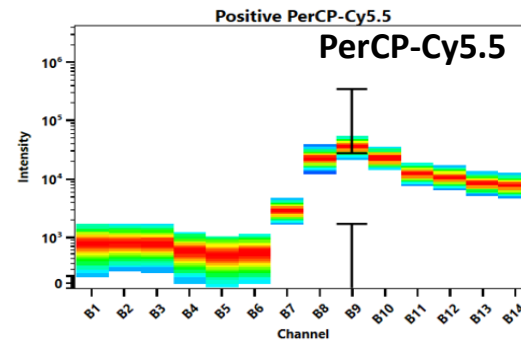
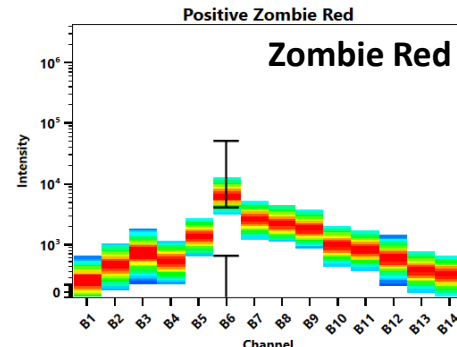
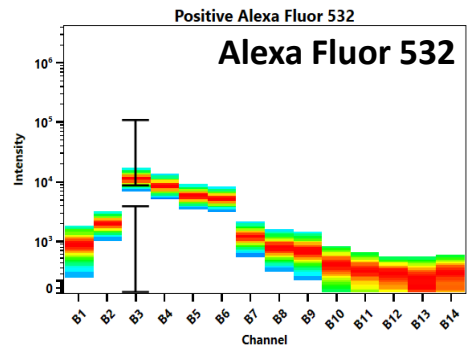
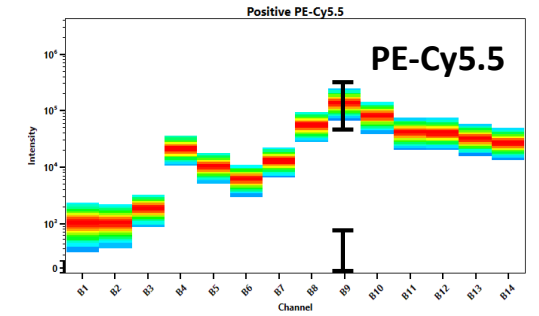
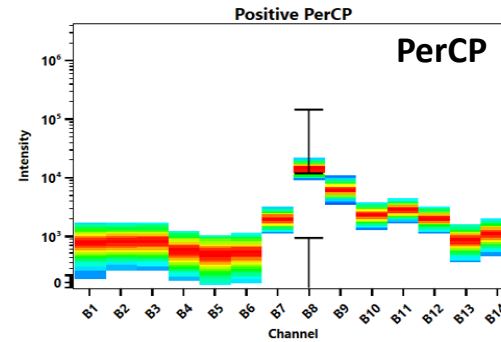
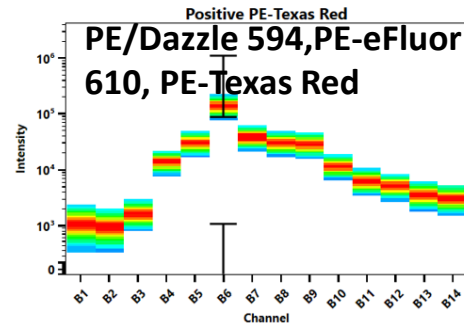
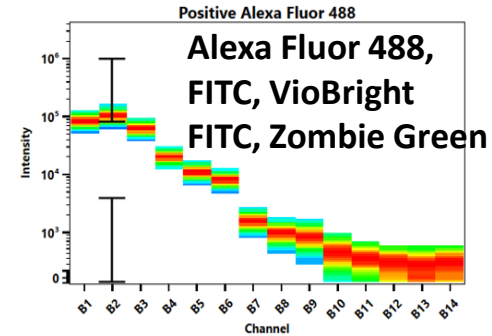
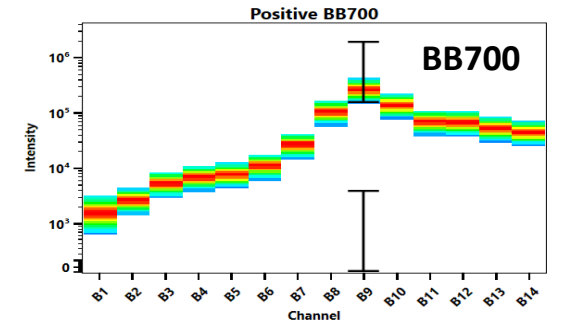
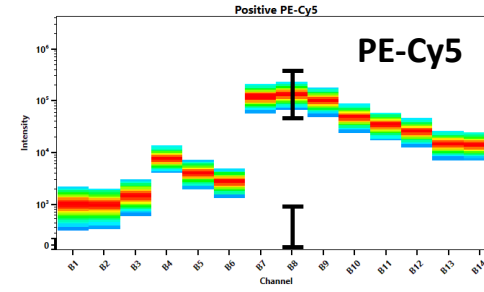
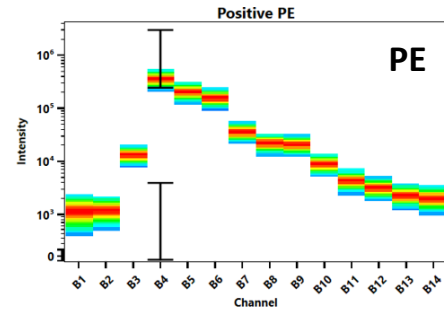
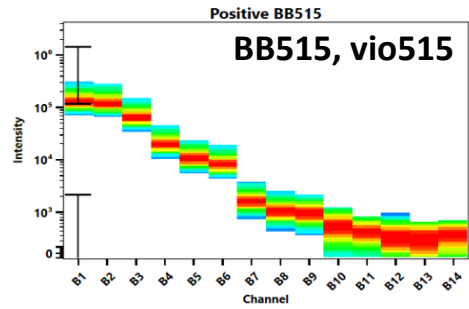
PerCP-Vio700, PerCP-eFluor 710



PE-Vio770, PE-Cy7

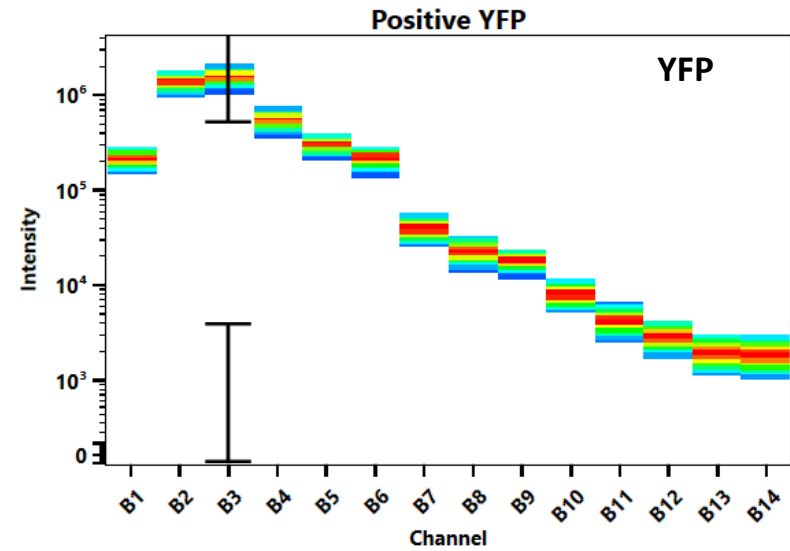
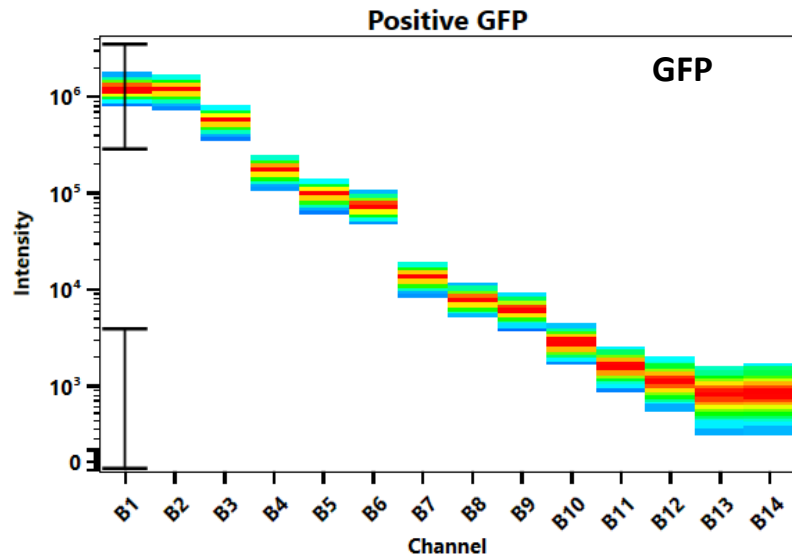
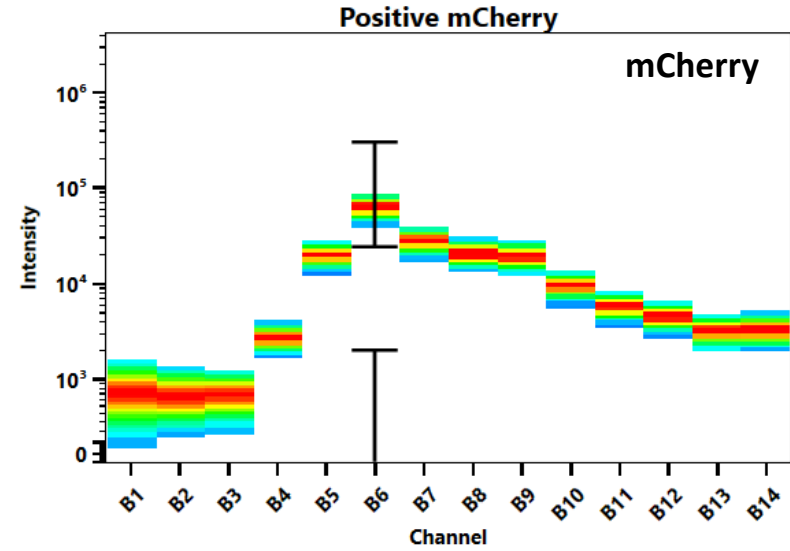
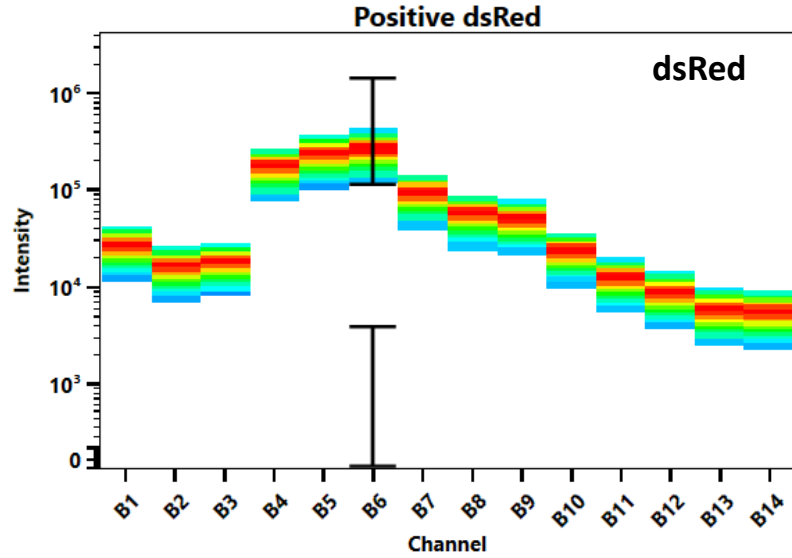


Blue Laser Excitable Dyes with Unique Signatures



Fluorescent Protein Signatures

Fluorescent Proteins



Peak Channels & Possible Combination of Dyes

Fluorochrome Peak Channels

Blue Excited Fluors	Peak Channel
BB515, sVio515, Vio515	B1
Alexa Fluor 488, FITC, VioBright FITC, Zombie Green	B2
Alexa Fluor 532, Live/Dead Green	B3
PE	B4
PE/Dazzle 594, PE-CF594, PE-eFluor 610, PE-Texas Red	B6
PE-Cy5, PerCP	B8
PE-Cy5.5, PerCP-Cy5.5, BB700	B9
PerCP Vio700, PerCP-eFluor 710	B10
PE Vio770, PE-Cy7	B13

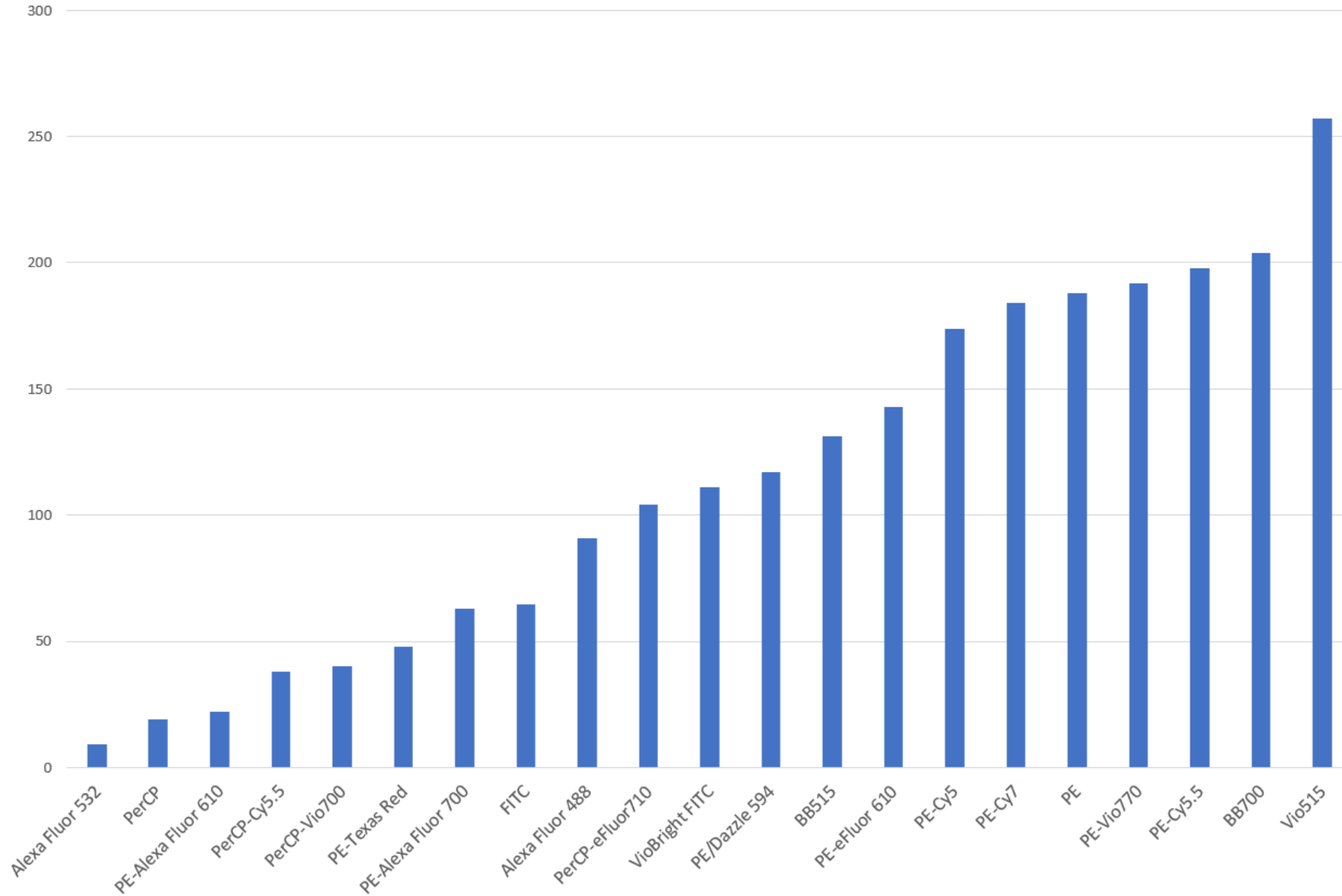
Example of 7 dyes that can be Used in Combination

Fluorophore
FITC
Alexa Fluor 532
PE
PE/Dazzle 594 or equivalent
PerCP-Cy5.5
PE-Cy7
PerCP-eFluor 710

Stain Indexes

Data generated using CD4 staining in human PBMCs

Stain Index Ranking - 21 Dyes



Cross-Stain Index Matrix

Dyes used in combination need to have unique spectra AND need to be assessed in terms of spread that they introduce to other dyes.

For example PE and PE/Dazzle 594 have distinct signatures, but since both dyes emit in the same wavelength range and significant spread is introduced by PE, careful panel design is needed when used in combination.

Spread Matrix for 5 Fluors that can be Used in Combination

	Alexa Fluor 488	PerCP-Cy5.5	PE	PE-Dazzle594	PE-Cy7
Alexa Fluor 488	Black	White	Light Pink	White	White
PerCP-Cy5.5	White	Black	White	White	Light Pink
PE	White	Light Pink	Black	Light Pink	White
PE-Dazzle594	White	Light Pink	Dark Pink	Black	White
PE-Cy7	White	White	White	White	Black

To read this table: spread of fluor in the row impacts the resolution of the fluor in the column. Pink means the fluor in that row has spread into the dye in the column (for example PE into PE/Dazzle 594). Areas in dark pink are where more attention to panel design is needed.

Spread Matrix for 9 Fluors that can be Used in Combination

	BB515	Alexa Fluor 488	Alexa Fluor 532	PerCP-Cy5.5	PerCP-eFluor 710	PE	PE-Dazzle594	PE-Cy5	PE-Cy7
BB515	Black	Dark Pink	Dark Pink	Light Pink	Light Pink	Light Pink	Light Pink	Light Pink	Light Pink
Alexa Fluor 488	Light Pink	Black	Light Pink	Light Pink	Light Pink	Light Pink	Light Pink	Light Pink	Light Pink
Alexa Fluor 532	Light Pink	Light Pink	Black	Light Pink	Light Pink	Light Pink	Light Pink	Light Pink	Light Pink
PerCP-Cy5.5	Light Pink	Light Pink	Light Pink	Black	Light Pink	Light Pink	Light Pink	Light Pink	Light Pink
PerCP-eFluor 710	Light Pink	Light Pink	Light Pink	Light Pink	Black	Light Pink	Light Pink	Light Pink	Light Pink
PE	Light Pink	Light Pink	Light Pink	Light Pink	Light Pink	Black	Light Pink	Light Pink	Light Pink
PE-Dazzle594	Light Pink	Light Pink	Light Pink	Light Pink	Light Pink	Light Pink	Black	Light Pink	Light Pink
PE-Cy5	Light Pink	Light Pink	Light Pink	Dark Pink	Dark Pink	Light Pink	Light Pink	Black	Light Pink
PE-Cy7	Light Pink	Light Pink	Light Pink	Light Pink	Light Pink	Light Pink	Light Pink	Light Pink	Black

To read this table: spread of fluor in the row impacts resolution of the fluor in the column. Pink means the fluor in that row has spread into the dye in the column (for example PE into PE/Dazzle 594). Areas in dark pink are where more attention to panel design is needed.

Document Revision History

Effective Date	Description of Change	Revision	EC No.
10/21/2019	Initial Release	A	EC-00265