Cytoflex 1

Flow Cytometry & Single Cell Analysis
https://fcsc.ku.dk/

Primary Flourochrome (Dim Antigen)	Secondary Flourochrome (Bright Antigen)	BV421	Pac Blue	BV480	BV510	BV605	BV650	FITC	BB515	AF 488	PerCP-Cy5.5	BB700	PE	PE-CF594	PE-Cy5	PE-Cy7	APC	AF 647	AF 700	APC-R700	APC-Cy7	APC-Fire	APC-eflour780
	BV421	450		50%	5%	17%	13%	1%	0%	0%	0%	0%	0%	2%	1%	2%	2%	3%	3%	1%	2%	2%	2%
	Pacific Blue		54	50%	5%	17%	13%	1%	0%	0%	0%	0%	0%	2%	1%	2%	2%	3%	3%	1%	2%	2%	2%
	BV480	5%	1%	127		0%	0%	2%	0%	0%	3%	0%	0%	0%	0%	0%	0%	2%	6%	3%	3%	2%	1%
	BV510	5%	1%		60	0%	0%	2%	0%	0%	3%	0%	0%	0%	0%	0%	0%	2%	6%	3%	3%	2%	1%
	BV605	3%	28%	47%	59%	303	58%	5%	0%	26%	10%	23%	74%	83%	49%	0%	16%	25%	5%	4%	4%	26%	0%
	BV650	0%	0%	26%	35%	69%	118	1%	0%	0%	14%	34%	28%	43%	59%	0%	45%	0%	1%	3%	4%	0%	0%
	FITC	0%	7%	42%	2%	3%	3%	85			12%	18%	23%	17%	28%	17%	16%	24%	22%	25%	20%	25%	22%
	BB515	0%	7%	42%	2%	3%	3%		393		12%	18%	23%	17%	28%	17%	16%	24%	22%	25%	20%	25%	22%
	Alexa Fluor 488	0%	7%	42%	2%	3%	3%			115	12%	18%	23%	17%	28%	17%	16%	24%	22%	25%	20%	25%	22%
	PerCP-Cy5.5	0%	0%	6%	0%	83%	74%	46%	17%	9%	95		91%	97%	99%	32%	92%	77%	58%	92%	47%	3%	17%
	BB700	0%	0%	6%	0%	83%	74%	46%	17%	9%		893	91%	97%	99%	32%	92%	77%	58%	92%	47%	3%	17%
	PE	0%	3%	4%	8%	81%	9%	4%	3%	7%	4%	2%	1243	76%	46%	24%	8%	9%	6%	8%	4%	8%	6%
	PE-CF594	2%	9%	16%	23%	92%	32%	3%	1%	7%	3%	6%	80%	706	23%	6%	4%	6%	0%	0%	0%	6%	1%
	PE-Cy5	3%	0%	2%	0%	70%	77%	33%	0%	0%	89%	86%	72%	83%	1331	16%	92%	82%	59%	93%	47%	1%	17%
	PE-Cy7	0%	1%	0%	0%	53%	36%	1%	0%	0%	59%	67%	55%	74%	88%	1222	62%	51%	40%	81%	89%	89%	89%
	APC	1%	0%	6%	3%	31%	72%	7%	2%	1%	40%	55%	15%	27%	95%	9%	296		23%	65%	52%	22%	45%
	Alexa Fluor 647	1%	0%	6%	3%	31%	72%	7%	2%	1%	40%	55%	15%	27%	95%	9%		155	23%	65%	52%	22%	45%
	Alexa Fluor 700	0%	9%	0%	9%	49%	61%	6%	1%	11%	74%	90%	68%	86%	96%	8%	82%	85%	66		37%	28%	31%
	APC-R700	0%	9%	0%	10%	49%	61%	6%	1%	11%	74%	90%	68%	86%	96%	8%	82%	85%		480	37%	28%	31%
	APC-Cy7	0%	0%	0%	0%	0%	42%	2%	0%	0%	46%	83%	6%	30%	84%	89%	76%	73%	64%	89%	225		
	APC-Fire	0%	0%	0%	0%	0%	42%	2%	0%	0%	46%	83%	6%	30%	84%	89%	76%	73%	64%	89%		212	
	APC-eflour780	0%	0%	0%	0%	0%	42%	2%	0%	0%	46%	83%	6%	30%	84%	89%	76%	73%	64%	89%			232

How the matrix was made
Mouse spleens was stained with individual anti-CD8 labeled antibodies with the indicated flourochrome and analyzed on the indicated instrument.
Each calculated value was arbitrarily assigned a color code according to the legend to show where the biggest spreading was situated.

How to use the resolution impact matrix
You find the color of interest on the top of the matrix, go down till you find the channel that you need to combine the color with and read the impact of spreading.

Consider this for panel design
When you are designing larger panels the task of making correct combinations becomes more difficult, but using the list below can help you:

- The lineage markers such as CD4, CD19 etc. should be found on the top of the matrix.
- Make sure the lineage marker has as many green cells as possible.
- For an important marker you should find the color on the left of the matrix.
- Make sure the marker has as many green cells as possible when you move across the matrix.
- Notice that spreading occurs between different laser lines.

Amount of spreading (%)

0-20
20-40
40-60
60-80
80-100
NA

Relative fluorochrome brightness (AU)

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