

### FC4: Cell & Tissue Analysis BD FACS Aria Configuration

	<u>Position</u>	<u>LP</u>	<u>Filter</u>	<u>PMT Range</u>	<u>Fluorochrome</u>
<b>Blue Laser</b> (488nm Em) 20mW	A	755	780/60	750nm – 810nm	PE-Cy7
	B	690	710/50	685nm – 735nm	PerCP-Cy5.5, PE-Cy5.5
	C	640	660/20	650nm – 670nm	PE-Cy5, (PerCP option*), AO-Red*, 7AAD**
	D	600	610/20	600nm – 620nm	PE-Texas Red (ECD), PI**
	E	550	575/25	562.5nm – 587.5nm	PE, TMRh, DsRed
	F	505	530/30	515nm – 545nm	FITC, GFP, Alexa488, Sytox Gr, DCFDA, YFP, NDBG*, AO-Green*
	G		488/10		SSC
	H				None
<b>Red Laser</b> (638nm Em) 30mW	A	755	780/60	750nm – 810nm	APC-Cy7, Alexa750
	B	690	710/50	685nm – 735nm	Alexa 680, APC-Cy5.5
	C		660/20	650nm – 670nm	APC, TO-PRO-3**
<b>Violet Laser</b> (405nm Em) 50mW	A	545	605/40	585nm – 625nm	QDot 605
	B	505	525/50	500nm – 550nm	Alexa 430, AmCyan, Cyan Fluorescent Protein, Pacific Orange
	C		450/50	425nm – 475nm	DAPI**, Pacific Blue, Cascade Blue, Alexa 405, Hoechst
<b>UV 20mW</b> (355nm Em)	A	635	670LP	> 670nm	Hoechst Red
	B		450/50	425nm – 475nm	Hoechst Blue, DAPI, Indo1-Violet

Optional Filters:

	<u>Position</u>	<u>LP</u>	<u>Filter</u>	<u>PMT Range</u>	<u>Fluorochrome</u>
Blue Laser	C	640	675/20	665nm – 685nm	PerCP
	E	525	550/30	535nm – 565nm	YFP
	F	495	510/21	499.5nm – 520.5nm	GFP
Violet Laser	A	635	660/40	640nm – 680nm	QDot655
	B	545	605/40	585nm – 625nm	QDot 605
UV Laser	A	505	530/30	515nm – 545nm	Indo1-Blue, GFP, Dansyl Cadaverine, ASBMS**
		450			
		535			
		635			
			405/20	395nm – 415nm	Indo-1 Violet
		605/12	599nm – 611nm	QDot 605	

\* AO – Acridine Orange used to discriminate autophagy

NDBG 6-[N-(7-nitrobenz-2-oxa-1,3-diazol-4-yl)amino]-6-deoxyglucose used to discriminate glucose/hexose transport

\*\* Dye can be used for viability exclusion on unfixed (non-permeabilized) samples

Blue Laser PI and 7AAD approx. 2ug/mL for 1X10<sup>6</sup> cells

UV Laser DAPI 0.05ug/mL – 0.2ug/mL for approx. 1.5X10<sup>5</sup> cells

UV Laser ASBMS (aminostilbamidine, methanesulfonate) 10uM (from 10mM stock DMSO -80<sup>C</sup>) for 10' at RT then place on ice

Red Laser TO-PRO-3 [1-5nM]<sub>F</sub>