

A

Protocol	FITC	PE	ECD	PC5.5	PC7	APC	APC-700	APC-750	Pac Blu	KrO
T Cell-2	CD154*	CD152*	CD45RO	CD56	CD279	CD3	CD8	CD28	CD4	CD45
Monocytes	CD40*	CD142*	CD14	CD33	CD274	CD86*		CD16	HLA-DR	CD45

B

	FS	SS	FL1	FL2	FL3	FL4	FL5	FL6	FL7	FL8	FL9	FL10
Voltage	205	527	452	421	459	565	624	570	515	498	386	358
Gain	5.0	7.5	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Discriminator	FS	SS	FL1	FL2	FL3	FL4	FL5	FL6	FL7	FL8	FL9	FL10
	90	OFF	OFF	OFF	OFF	OFF	OFF	OFF	OFF	OFF	OFF	OFF
	FL1	FL2	FL3	FL4	FL5	FL6	FL7	FL8	FL9	FL10		
FL1		1.4	0.2	0.1	0.1	0	0	0	0	1.4		
FL2	14.9		7.2	0.7	0.7	0	0	0	0	0		
FL3	7.9	44.5		0.1	0.6	0	0	0	0	0		
FL4	1	9.5	55.9		0	0.6	0.5	0	0	0		
FL5	0	1	10.5	33.6		0.5	0.8	3.6	0	0		
FL6	0	0	0.4	1.1	0		12.1	15.2	0	0		
FL7	0	0	0.1	15.7	0.4	19.7		5.9	0	0		
FL8	0	0	0	6	6.3	5.7	30.3		0	0		
FL9	0	0	0	0	0	0	0	0		2.3		
FL10	1	1.5	0	0	0	0	0	0	5.9			
Lasers	Status	Shutter	Power	Current								
Blue	On	Closed	22 mW	0.43 A								
Red	On	Closed	25 mW	0.07 A								
Violet	On	Closed	40.1 mW	0.07 A								

C

	FS	SS	FL1	FL2	FL3	FL4	FL5	FL6	FL7	FL8	FL9	FL10
Voltage	208	654	432	422	447	581	629	593	516	492	354	326
Gain	5.0	7.5	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0	1.0
Discriminator	FS	SS	FL1	FL2	FL3	FL4	FL5	FL6	FL7	FL8	FL9	FL10
	100	OFF	OFF	OFF	OFF	OFF	OFF	OFF	OFF	OFF	OFF	OFF
	FL1	FL2	FL3	FL4	FL5	FL6	FL7	FL8	FL9	FL10		
FL1		1.4	0.1	0.2	0.1	2	0	0	0	1.4		
FL2	14		8.2	0.5	1.3	0.7	0	0	0	0		
FL3	3.2	44.5		0	0.6	0	0	0	0	0		
FL4	1	9.5	3.8		0.2	0.6	0.5	0	0	0		
FL5	0	1	1	26.2		1.4	0	1.9	0	0		
FL6	22.2	0	0.4	2.3	1.6		6.9	23.3	0	0		
FL7	0	0	0	2.3	0.4	14.7		9	0	0		
FL8	0	0	0	3.2	10.8	2	23.1		0	0		
FL9	0	0	0	0	0	0	0	0		2.3		
FL10	1	1.5	0	0	0	0	0	0	5.9			
Lasers	Status	Shutter	Power	Current								
Blue	On	Closed	22 mW	0.43 A								
Red	On	Closed	24.9 mW	0.07 A								
Violet	On	Closed	40 mW	0.07 A								

Additional File 1. Protocols for T cell and Monocyte phenotypes. A. List of antibodies. All antibodies were purchased from Beckman Coulter unless otherwise designated. *= antibodies purchased from BD Biosciences. **B.** Beckman Coulter Gallios instrument settings for the T-cell-2 protocol. **C.** Beckman Coulter Gallios instrument settings for the Monocytes protocol.