

February 19th, 2025

Mehnaz Mbrayles

Specimen	Status	Location	Conc	Date	Notes	Volume	CD45+	Total	Resuspension
301 ^u Inf10775 a-1	Cord HU ♂	VCQU			Lysed? lysed ✓	0.5 mL			
201 ^u ND006	Adult ♀		20EG	11/22/20		1 ML			
Inf 978	Cord HU ♀				Blood ✓	2 mL			

9:34 AM prep 5
ND006 then

9:42 AM Inf1077 then

9:46 AM spin ✓

10:22 AM count

11:15 spin

2nd stain count @ 11:42 am

12:33 AM

3:41 pm se's cold → 4:11 pm
1/6 spin @

FcBlock @ 4:56 pm

Samples cold @ 4:11 pm
spin @ 4:44 pm

59 µL Done @ 4:54 pm
80 µL for
samples

W741570

~~Inf ND050~~
~~2~~ 11
~~270 µL~~
~~270 µL~~
cool Buu PE-Fire
Adult Buu PE-Fire 700

ND050
~~2~~ 13
230 µL
Adult Buu PE-Fire 700

~~430 µL~~
640

Inf 3 / 5.4 E6
555 µL
OO
92 µL ~ 500K

ND050 3 / 13.0 E6
15 230 µL
OO
38 µL ~ 500K

8.43

PE-Fire 700 @ 12 µL
is too much,
but separable/discernable/
workable

February 19th, 2025

Inf1077-5 4 HU♂ 2022-06-13 497 28 A, G
Inf1140-6 5 HU♂ 2022-07-11

February 19th, 2025

February 19th, 2025

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Good cells

#	Filter	Fluorochrome	Clone	Dose	L/D RT 15 min	Surface 30min @4C	4
1	UV1	BUV395	CD18			0.4	1.6
2	UV2	BUV496	HLA-DR			<0.2>	#####
3	UV3	BUV563	CCR2			0.8	3.2
4	UV4	BUV615	CD89			0.7	2.8
5	UV5	BUV661	CX3CR1			1.0	4
6	UV6	BUV737	CD38			0.5	2
7	UV7	BUV805	CD38			0.8	3.2
8	V1	Zombie Violet	Viability		<1:1000>		
9	V2						
10	V3						
11	V4						
12	V5						
13	V6						
14	V7						
15	V8	BV570	CD16			0.9	3.6
16	V9	BV605	CD56			0.6	2.4
17	V10	BV650	CD11c			0.4	1.6
18	V11	BV711	FCER1a			1.2	4.8
19	V12	BV750	CD303			0.3	1.2
20	V13	BV785	CD141			0.4	1.6
21	V14	CD11b				0.1	0.4
22	V15	CD14				0.2	0.6
23	B4	Spark Blue 550					
24	B6						
25	B7						
26	B8						
27	B9						
28	B10	PerCP-Cy5.5	CD123			2.0	8
29	B12						
30	B13						
31	B14						
32	YG1	PE	CD45			0.3	1.2
33	YG2	PE-Dazzle 594	CD1c			0.3	1.2
34	YG3	PE-Cy5	CD40			1.3	5.2
35	YG4						
36	YG5	PE-Fire 700	HLA-DR			<1.2>	#####
37	YG6	cFluor BV6750	CD19			0.8	3.2
38	YG7	PE-Cy7	CD88			0.3	1.2
39	YG8	PE-Fire810	CD5			0.0	0.06
40	YG9						
41	R1	Alexa Fluor 647	CD163			1.2	4.8
42	R2						
43	R3	R718	CD32			0.4	1.6
44	R4						
45	R5						
46	R6						
47	R7	APC-Fire 750	CD4			0.5	2
48	R8	APC-Fire 810	CD64			0.8	3.2
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Reads

Wash with 2 ml PBS, spin down 1300rpm 8min
 800 ul of Live/Dead mix (1:1000) @RT for 15min
 Wash 1.5 ml 5% PBS-FBS, spin 1300 rpm, 8min
 Add ColdStain mix, incubate @ 4C for 30min
 Add 300-500 ul 1x RBC Lysis for 3 minutes
 Resuspend in 80 ul 0.4% PFA-PBS
 Cap tubes, wrap rack in foil, store at 4°C

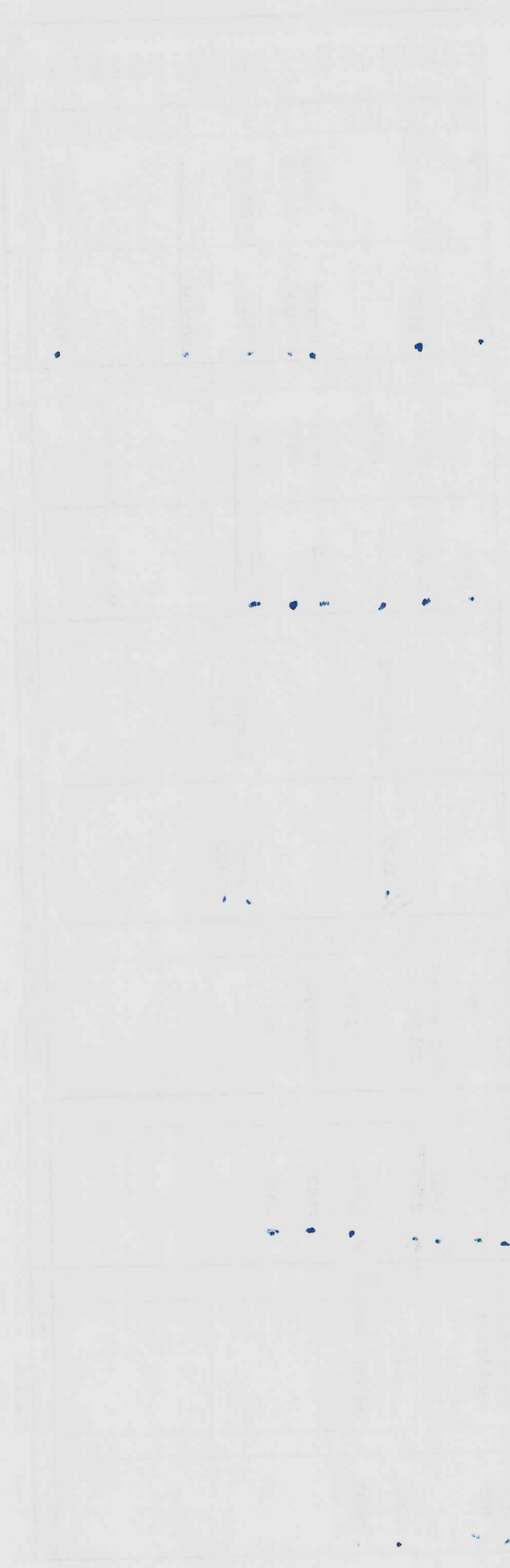
460
 7540

20
 150
 1000

2/18/2025

Spectrum		Myeloid Panel		Blue		Yellow-Green		Red	
	UV		Violet						
373	UV1								
388	UV2	BUV395							
428	UV3								
443	UV4								
458	UV5								
473	UV6								
508	UV7								
514	UV7								
525	UV8								
542	UV9								
582	UV9	BUV563							
598	UV10								
613	UV10	BUV615							
664	UV11	BUV661							
679	UV12								
697	UV13								
717	UV14	BUV737							
738	UV15								
750	UV16								
760	UV16								
783	UV16								
812	UV16								

$$\begin{array}{r} 1:10 \\ \hline 1:10 \text{ CDs} \end{array}$$



100 200 300 400 500 600 700 800 900 1000

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PE-Fic 100

Buugas

2b pop FcERla BV711 ✓

PerCP-cy5.3 ~

Pe-cy5 ✓

C Fluor BV6750 ✓

PerCP-cy5.5

Pe-cy5

