

February 17th, 2023

HEU: D17 $\gamma\delta$ CBMC panel #3 - 2023

Specimen	Status	Location	Conc	Date	Volume	Ly	Ly+M	Total	1E+6	.66E+6	.5E+6
190 ⁺ NDO23	IL-2		44 μ l			2.51 1.295			772 μ l	509 μ l	
450 μ l	Zol		8.3 μ l			1.18 0.59				1.118	
370 μ l	BCG					4.16 2.08				317 μ l	
500 μ l	Inf136	IL-2	18.6 μ l			5.51 2.75			363 μ l	240 μ l	
500 μ l	Zol		17.9 μ l			5.58 2.79				236 μ l	
660 μ l	BCG					5.81 2.90				227 μ l	
430 μ l	Inf587	IL-2	33 μ l			3.35 1.67			598 μ l	395 μ l	
500 μ l	Zol		42 μ l			2.65 1.32				509 μ l	
400 μ l	BCG		50 μ l			2.24 1.12				589 μ l	

10:45pm return incubator

10:53pm count

11:25pm reretained

06:08am plate out

6:28 am cells out plate

6:34 am spin

✓ D @ 6:56am → 7:11am/16

Spin @ 7:21 am

Cells Frozen @ 7:37 am

✓ D wash DBV fridge.

Ab prep 8:00 am

$$\begin{array}{r} 24 \\ \times 8 \\ \hline \end{array}$$

$$\begin{array}{r} 19.2 \\ \times 20 \text{ mls} \rightarrow 400 \mu\text{l} \\ 7 \text{ mls} \end{array}$$

10:47am Intracellular → 10:27 am

11:32 am Final Wash

Done & stored 50 μ l @ 11:49 am

8:39am Surface stain → 9:00 AM

Mixes ✓ 8:47 am

Spin @ 9:07 am

Strept @ 9:24 am → 36 am

Pi done @ 9:26 am / others stored

Strept wash @ 9:39 am

9:51 am → 10 am 11 an

10:17 am (first Permash)

10:30 am 2nd

11-2-117

11-4

11-6

11-8

11-10

11-12

11-14

11-16

11-18

11-20

11-22

11-24

11-26

11-28

11-30

12-2

12-4

12-6

12-8

12-10

12-12

12-14

12-16

12-18

12-20

February 17th, 2023

D17 Gd-P4b

#	Detector	Fluorochrome	Marker	Clone	L/D 15 min (RT)	Surface 20 min @4C	7.2	Intracellular 40min @RT	7.2
1	V450	BV421	PD1*			2.5	18.0		
2	V525	BV510	L/D Aqua		<1:500>				
3	V670	BV650	CD56*			1.8	13.0		
4	B530	Alexa 488	Perforin					3.5	25.2
5	B710	PerCPeF710	CD3			1.5	10.8		
6									
7	Y780	PE-Vio770	NKG2A			0.6	4.3		
8	R670	APC	Vδ2			1.0	7.2		
9	R780	APC-Fire750	CD16			1.0	7.2		
Antibody Total						8.4	60.5	3.5	25.2
PBS						12.1	87.1	17.0	122.4
Pipette draw volume / sample						19.5		19.5	

D17 Gd-P2

#	Detector	Fluorochrome	Marker	Clone	L/D 15 min (RT)	Surface 20 min @4C	7.2	Streptavidin	Intracellular 40min @RT	7.2
1	V450	BV421	Vδ2			1.0	7.2			
2	V525	BV510	CD3			1.5	10.8			
3	V670	BV650	NKG2D			2.0	14.4	1.5		
4	B530	Alexa 488	Perforin						3.5	25.2
5	B710	PerCPVio700	CD56			1.0	7.2			
6	Y590	PE	PD1			1.5	10.8			
7	Y780	PE-Vio770	NKG2A			0.6	4.3			
8	R670	APC	Vδ1			1.0	7.2			
9	R780	APC-Fire750	L/D Horizon		<1:1000>					
Antibody Total						8.6	61.9	10.8	3.5	25.2
PBS						11.9	85.7	136.8	17.0	122.4
Pipette draw volume / sample						19.5		19.5	19.5	

D17 Gd-P1

#	Detector	Fluorochrome	Marker	Clone	L/D 15 min (RT)	Surface 20 min @4C	7.2
1	V450	BV421	PD1			2.5	18.0
2	V525	BV510	L/D Aqua		<1:500>		
3	V670	BV650	CD16			1.5	10.8
4	B530	FITC	Vδ2			1.2	8.6
5	B710	PerCPeF710	CD25			2.0	14.4
6	Y590	PE	CD28			2.5	18.0
7	Y615	PE-Dazzle	CD27			1.5	10.8
8	Y780	PE-Vio770	CD3			0.5	3.6
9	R670	APC	Vδ1			1.0	7.2
10	R780	APC-Fire750	CD45RA			1.8	13.0
Antibody Total						14.5	104.4
PBS						6.0	43.2
Pipette draw volume / sample						19.5	

D17 gd-CK: α-γδTCR

#	Detector	Fluorochrome	Marker	Clone	L/D 15 min (RT)	Surface 20 min @4C	9.5	Intracellular 40min @RT	9.5
1	V450	BV421	PD1			2.5	23.8		
2	V525	BV510	L/D Aqua		<1:500>				
3	V670	BV650	CD56			1.5	14.3		
4	B530	Alexa 488	CD107a						
5	B710	PerCPeF710	TNFα				0.0	2.5	23.8
6	Y590	PE	Vδ2			1.0	9.5		
7	Y780	PE-Vio770	IFNγ					1.1	10.5
8	R670	Alexa 647	CD27			2.0	19.0		
9	R780	APC-Fire750	CD45RO			1.8	17.1		
Antibody Total						8.8	83.6	3.6	34.2
PBS						11.7	111.2	16.9	160.6
Pipette draw volume / sample						19.5		19.5	

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