

December 03-4th, 2021

HEU: D17 $\gamma\delta$ CBMC panels #11

510c/pl
180c/pl
650c/pl
645c/pl
730c/pl
700c/pl
620c/pl
620c/pl
680c/pl
510c/pl

Specimen	Status	Date	Conc	Tasks	Volume	Lymphocytes (1:50) %2	Total	.66E+6	1E+6	.5E+6
Inf 179	IL-2					4.77	2.39	276 μ l	418 μ l	
	Zol					4.08	2.04	326 μ l		245 μ l
	BCG					5.80	2.90	227 μ l		173 μ l
Inf 287	IL-2					3.10	1.55	426 μ l	645 μ l	
	Zol					2.75	1.38	478 μ l		362 μ l
	BCG					2.52	1.26	524 μ l		397 μ l
Inf 191	IL-2					4.74	2.37	278 μ l	422 μ l	
	Zol					3.97	1.96	337 μ l		255 μ l
	BCG					4.27	2.14	308 μ l		234 μ l
N4062	Zol					4.38	2.19	301 μ l		228 μ l

<47=183 μ l

	Expanded Gd-p4b	Surface μ L x 9.5	Intra μ L x 9.5
BV421	PD1*	2	19
BV510	Aqua L/D	1:500	
BV650	CD56*	1.8	17.1
Alexa 488 (FITC)	Perforin (dG9)	-	3 28.5
PerCPeF710	CD3	1.5	14.3
PE	-	-	-
PE Dazzle			
PE Vio770	NKG2A	0.6	5.7
APC	V82	1	9.5
APC Fire750	CD16	1	9.5
20.5 μ l/rxn;	PBS	12.6	119.7 17.5 166.3

	Expanded Gd-CK PMA	Surface μ L x 9.5	Intra μ L x 9.5
BV421	PD1	2	19
BV510	Aqua L/D	1:500	
BV650	CD56	1.5	14.3
FITC	CD107a	<3 μ L added separately>	
PerCPeF710	TNF α		2 19
PE	V82	1	9.5
PE Dazzle			
PE Vio770	IFN γ	-	0.6 5.7
Alexa 647 (APC)	CD27	1.5	14.3
APC Fire750	CD45RO	1.8	17.1
20.5 μ l/rxn;	PBS	12.7	120.7 17.9 170

	Expanded Gd-p1	Surface μ L x 9.5
V450 BV421	PD1	2 19
V525 BV510	Aqua L/D	1:500
V670 BV650	CD16	1.5 14.3
B530 FITC	V82	1.2 11.04
B710		
PerCPeF710	CD25	2 19
Y590 PE	CD28	2 19
Y615 PE Dazzle	CD27	1.5 14.3
Y780 PE		
Vio770	CD3	0.5 4.8
R670 APC	V81	1 9.5
R780		
APC Fire750	CD45RA	1.8 17.1
20.5 μ l/rxn;	PBS	7 66.5

	Expanded Gd-p2	Surface μ L x 9.5	Intra μ L x 9.5
BV421	V82	1 9.5	
BV510	CD3	1.5 14.3	
BV650	NKG2D	2 19	
	Streptavidin	{1.5} 14.2	{19} 180.5
Alexa 488 (FITC)	dg9	-	3 28.5
PerCPeF710	CD56	1 9.5	
PE	PD1	1.5 14.3	
PE Dazzle			
PE Vio770	NKG2A	0.6 5.7	
APC	V81	1 9.5	
APC Fire750	Horizon L/D	1:1000	
20.5 μ l/rxn;	PBS	11.9 113	17.5 166.3

cont'd 3:04
Plate 5:14
Prep done @ 8:21
8:45 spin
W/O @ 9:03
S&S @ 20
surface @ 20
-> 10:06
strept @ 10:24
@ 10:51
Fixperm @ 21
23:41-21
Intra m
Done @ 00:42

27 x 36 3600
x 2 18 2200
210 -> 23 x 2 = 46 L/D Aqua
10 μ l - 10 ml 40 Horizon
126 mls FBS-PBS
~108 ml Perm Wash

Finished Nonopaid Perm Wash

W/O @ 23:47 -> 10:2
Surface @ 00:16
Fixperm @ 00:50 -> 00:710
Intra @ 1:29 am
Done @ 02:17 am

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scs are identical as the D17 #9 setup in plating (run fresh)

13.02	12.97
00	.01
.38	→ .38
.00	.04
.00	.02
8.78	8.74

Starting w/ P1 \uparrow Fsc to 560 @ 12:46 pm
~ 3:40 sec hand out to FF @ 4 min

Inf 174 Zol had weird CD25 x PDI distribution

\uparrow 520

9
x4
36 min
16
42 min

~~Inf 287 BCG P1 failed BCG stim?~~ ~~few VD₂~~

\nearrow early culture
same perhaps

same PDI vs naive IL-2
CD25

Beads when alternatives
≠ exist, or biologic
alternative is just asked

P2 \uparrow Fsc to 600

$\star \rightarrow$ i Inf 287 Zol P2 mismatch w/ other outcome shear VD₂? \star

BCG culture pattern repeats ✓

oops just low amount, underlying
cell viability?

cell gate Fsc fine @ 200 ✓

KNR1 OP-12

Needs 6-EP validate CD161 - cell.

287 \neq great VD₂/Zol \rightarrow BCG/IL-2
but kpid.

Does 191 have more VD₂?
yes HEU-hi, but?

Done @ 2:57pm

