

Sept 18-20, 2020

"Contamination" experiment

HEU: New TT Optimization #5

12 hr Rest, 2hr Stim, 18 hr Incubation.

Adult	Location		Date	Tasks	Volume	Ly	Ly+Mon	Total
ND050		15E6	08/12/20	Costim diff vs TT def	3 ml	4.17E4	5.29E4	12.51E6
					4 ml	2.15E4	3.39E4	11.00E6

Adult 1.0E+6 cells	Ex vivo TT	Extracellular $\times 10$		Intracellular $\times 10$		
V450 BV421	CD69	.5	5	CD69	1	10
V525 BV510	Aqua	1:500		Aqua	-	
V670 BV650	CD4	1.8	18	CD4	0.5	5
B530 FITC	CD8	1.5	15	CD8	0.5	5
B695 PerCPeF710	TNFA	-		TNFA	3.0	30
Y590 PE	CD62L	2	20	CD62L	-	
Y780 PE Cy7	IFNg	-		IFNg	0.8	8
R670 APC	CD3	1.5	15	CD3	0.5	5
R780 APC-Fire750	CD45RA	1.5	15	CD45RA	-	
PBS		9.7	97		14.7	147

Costim CD28+ CD49d+ Costim + T

TT only TT+ CD28 TT+ CD49d TT+ Costim + T

Costim: .5 μ l into 9.5 μ l \rightarrow 5 μ l each condition

+ 25 μ l over 500 μ l
~ .5 μ g conc

1.785 μ l ~ 5 μ g TT over 500 μ l
~ 10 μ g TT

Plated @ 2M/ml ~ 500 μ l ~ 1M/ml

10/19

11:47am Incubation start

1:47pm Gdgi. Mix added

7:52am Processing starts, spindown

8:05am L/D started ~ 8:20am

8:25am L/D spindown

10:03am Intracellular start

10:43 Intra out centrif

10/20

