December 19th, 2024

Specimen	Status	Location	Conc	Date	Notes	1			
NVO20	1114					Volume	CD45+	Total	Resuspension
NYOZZ	Adult		20t +6	8/3/2011	Same chapped text issue	1	11E6_	11 EG	1 2.7 Volume
									12.7 Volume
	Г								

Fresh FBS-10% and worsh tibes after lost contamination issue!

· Added I mak of the to water bath.

Reagent prepa 1:15 pm a Mix & FOS-PBS done @ 1:59pm

Than @ 1:20pm

DIVASE

Stain Count @ 2:43pm

Spin @ 3:24 pm

L/D Full strined @ 3:42 pm spin@3: Stepm Hotse's @3.54pm > 4:25 spin@ 4:27 Cad scs @ H:05pm = 74:35 7

#SPIN @4:39 pm - Spin @4:39pm

7.5 cold @ 4:51 pm/ > 5:21 pm spine 5:22pm

Shrept @ 5:30 pm > 5:45 spin @ 5:47 pm/ Tix @ 5:50 -> 08-> 18 ->

Mix Up wy PemWash 1:1 Instead of 1:10 10...

Second wash 1:1 75 ~ 1:10 Ugh...

Soo stipid,

Soo inforced 1:1 mstead of 1:10 for first wash

TBD the effect... ndivada @ 6:46 pm

this one before, LTBD effect? HC016

1.5 El each

¥300 ×600 ×1200 9.300 18600 24 1.200

10.500 Fix

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			And Ur				24				-	23		23 !		20	13	+	18	+	17			1	16	1	T	15	14	: 1	;	Τ	12	T	11	10	9	000	T	П	7	Т		6	Т	T	Τ.	I. T		_	П	_		
		NO. TOTAL	ISTAIN					R5	R4	-		R1	V610	V	500	VC7	CPI	104	YG3	YG2	YG1	B14	B13	B12	810	BO BO	87	B6	84	83	82	V16	V15	V14	V13	-	V10	\ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \	<b>Y7</b>	٧6	V5	+	4536	+	5 5	_	4 UV14		2 UV10	15	9/0	1 00/1	#	₽
		CONTROLS !!!	ED CONTROLO	APC-Fire 810	ADC E: 210	APC-Eiro 750	Zombie NIB	TOTAL PROPERTY.	APC-R700	140 IOU 14/	lexa Elijor 647	VBC OTSAL-31		PE-FIFE /44	PE-Fire /00		PE-Cy5		PE-Dazzle 594		PF	RB780		ND/US	DB70E			RB613	Spark Blue 574	Spark Blue 550	Alexa Fluor 488		BV786	BV750	BV711	BV605	BUCOL	BV570	BISNE		BV480		Pacific Blue		t				9 BUV563		6 6 6 V 395			Filter Fluorochrome
	Pip			CCR7	CD27	1/0	7	CDTO/9		172	CD39	CD56?	HLA-DR	CD25	CD127		CXCR3	2	TNEO	ICOS		CXCBCS		CD26			, CA	PD1	CD45	CD3	FoxP3		CCR6	IENIV	CD8	CD45RA		CD16			CD161	CDTO	CD19			CD4	CD38	CCR4	CD69		CD62L			Marker
	Pippette draw volume	R10 Media	Antibody Total		(0323)			(H4A3) Supernatan		(MQ1- 2			(L243)					7 (TTOWIN	MARTIN	EH12.2H7 1				(M-A261)	9				(01/7)	17/3/		(CHTT)	(11/0) 2	71		(J252D4)				(TCOCOL)	(REAG21)	(HIB19)	(M5E2)		(000)	(6/3)	(B6)	(1G1)	(FN50) 2		(DREG-56)		cione	2
				<u> </u>	1	2		natan 1.2			1	1	2	2		-	١				2		-	7			1	1	-	beads		1		2	1	1	F	2		F	,		2		1	1	2	2			1		Dose	
		ם מ	<u></u>	-				2.4						1	1								1	1	1			-								1	+				-			-	+	-			-		-		stim!!!	ring.
	Pippette draw	Brilliant Ct.	ntibod T			<1:2500>																													1													1					2 40 Min	- 70
31.2	22.8	11.4	1		1			1		1.2			1.2			1			<0.03>	2000							•					1.7								1.5			1		1.5		7.8						10 min 30	
	45.6	22.8	2			1				2.4			2.4			2			\ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \ \							7	3					2/								w		1	1		3	-	3.6			+		40	30min 2	Hot Stain
44.1		15.7		1.5								0.6		1.5									1.0				0.0	7.7	10				1.2	1.0	1.0	0:0	0.9				1.3	1.5			0.5	0.5				1.2		@4C	30min	Calabo
-	62.8	31.4		ω						-		1.2		ω	1	1							3				7.2	2.4		1			2.4	2	2	1.0	10				u	ω			F	, 1				2.4			2	
		1	1							+	-	-	1								-															1			<0.4>	Strep												15 min	4*C for	
	-	1																																																		then Flx/Perm	RBC Lyse,	αβιce
	21.6	-						7:4									1					2						0.5	ω			1.5												0.7		0.1	0.0	00			(		Intranuclear	αβ I cell SFC panel
		L	L		1	_		0									0					0						0	0			0												0		0	c					0		

Wash with 2 ml PBS, spin down 1300rpm 8min 800 ul of LiveDead mix (1:2500) @RT for 15min Wash 1.5 ml 5% PBS-FBS, spin 1300 rpm, 8min

Add CD161 biotin antibody for 10 minutes at RT

Add HotStain mix, incubate @ 37C for 30min Wash 1.5 ml 5% PBS-FBS 1400 rpm, 6min

Add ColdStain mix, incubate @ 4C for 30min Add 300-500 ul 1x RBC Lysis for 3 minutes Wash 1.5 ml 5% PBS-FBS 1400 rpm, 6min

Add Streptavidin Mix, incubate @ 4C for 15min Wash 1.5 ml 5% PBS-FBS 1400 rpm, 6min U.8 ml Nuclear HxPerm, incubate @ 4C for 30min (vortex every 10 minutes)

First PermWash: 1.5 ml NuclearWash 1500 rpm 6 min

Second Perm Wash: 1.5 ml NuclearWash 1500 rpm 6 min

Add intracellular Stain, incubate @ RT for 40min First PermWash: 1.5 ml NuclearWash 1500 rpm 6 min Second Perm Wash: 1.5 ml NuclearWash 1500 rpm 6 min Resuspend in 81 III 14% EFS ADEX

Resuspend in 80 ul 0.4% PFA-PBS
Cap tubes, wrap rack in foil, store at 4\*C

