

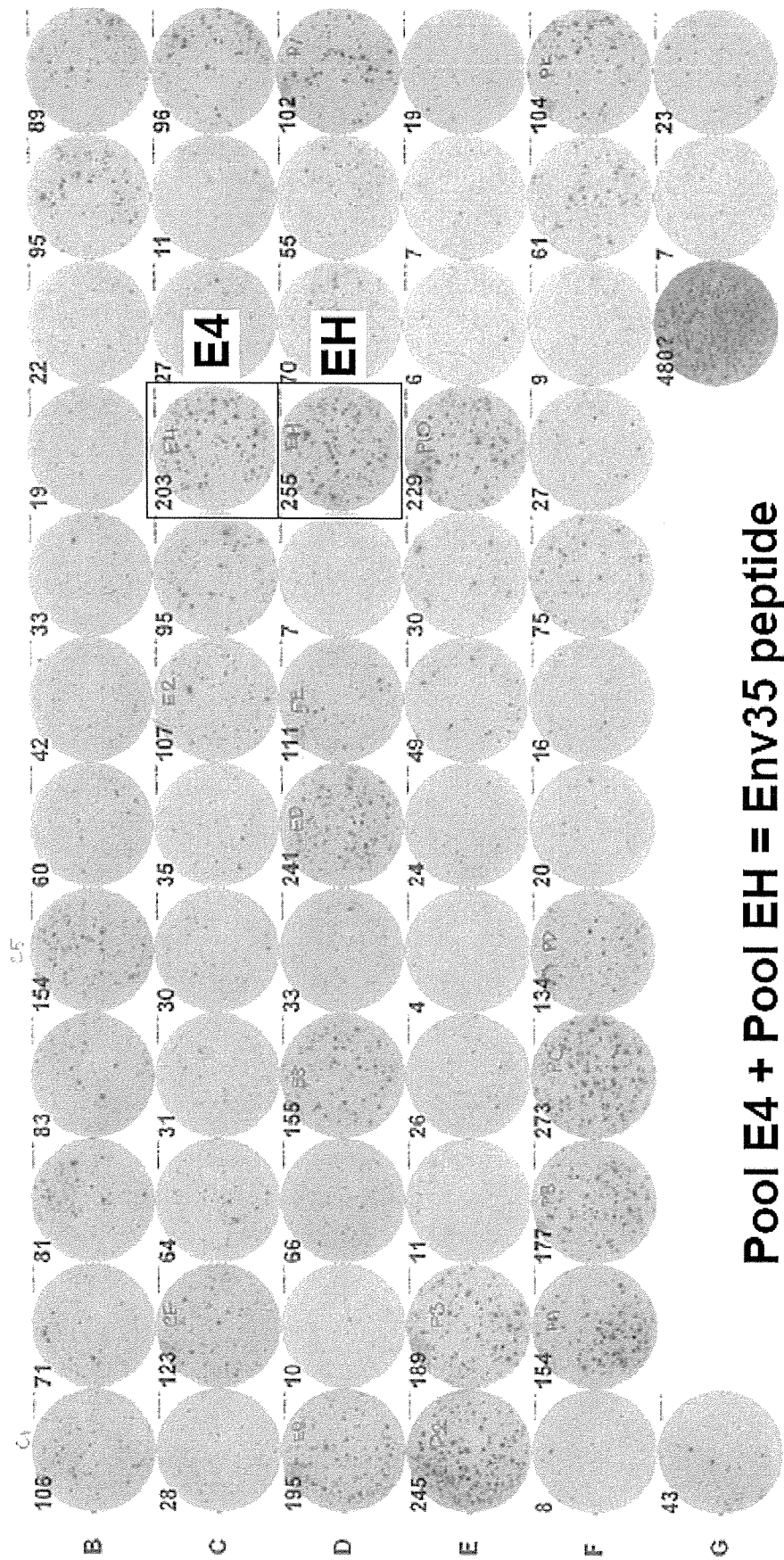


Figure 1

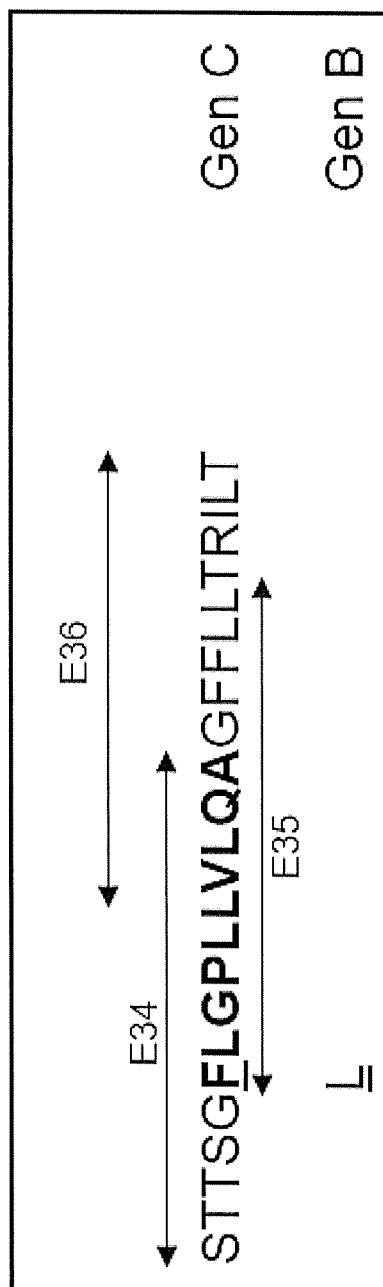
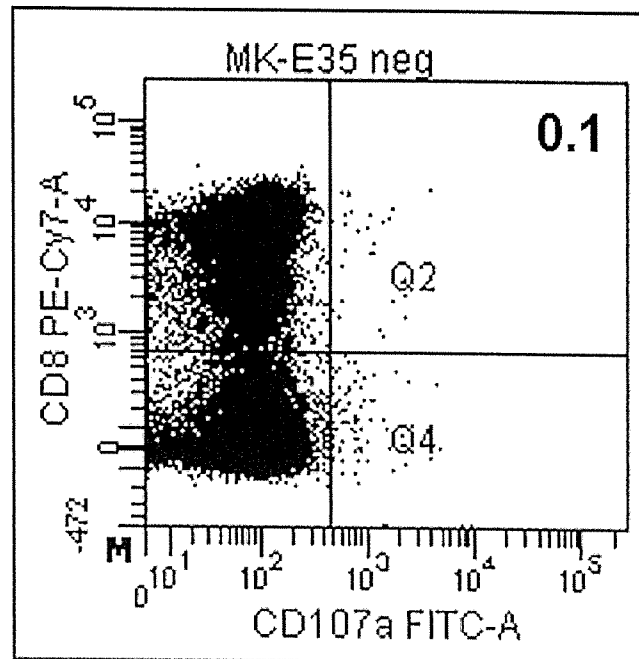


Figure 2

unstimulated



+ E35 peptide

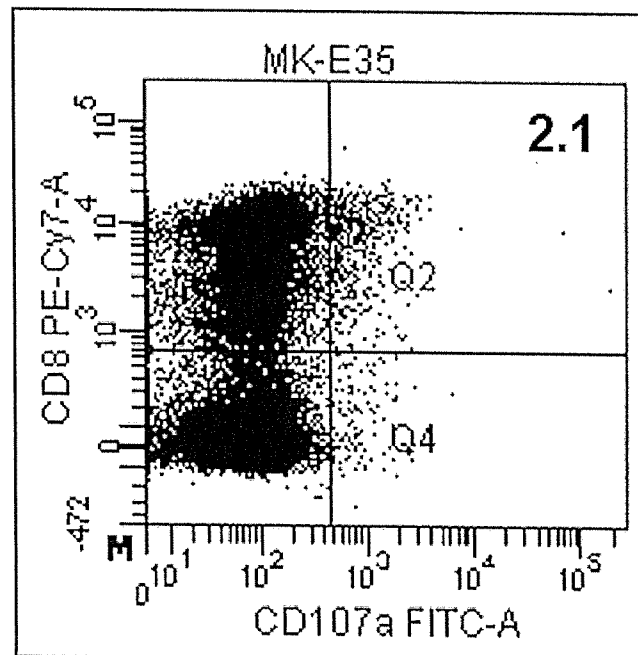


Figure 3

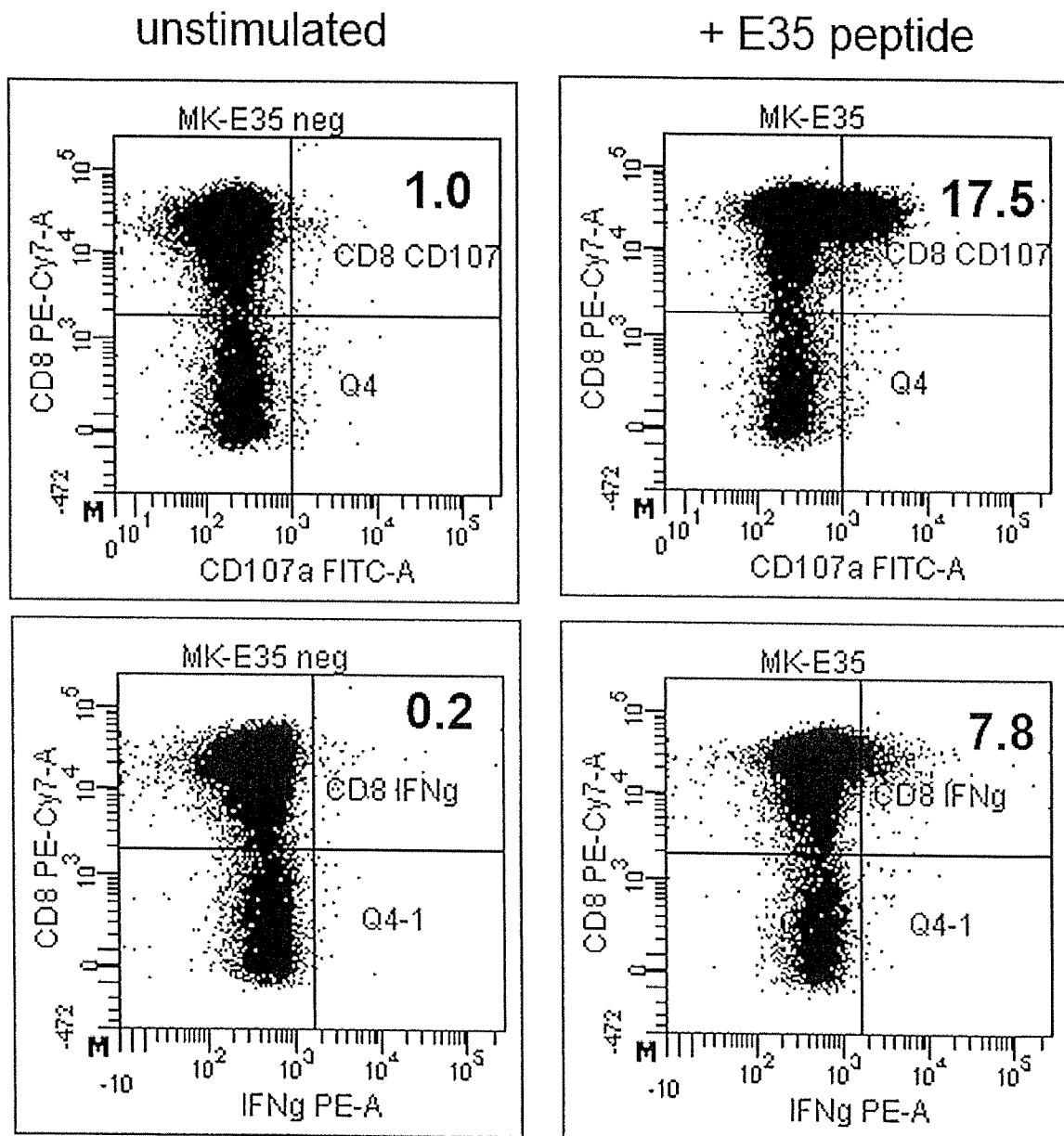


Figure 4

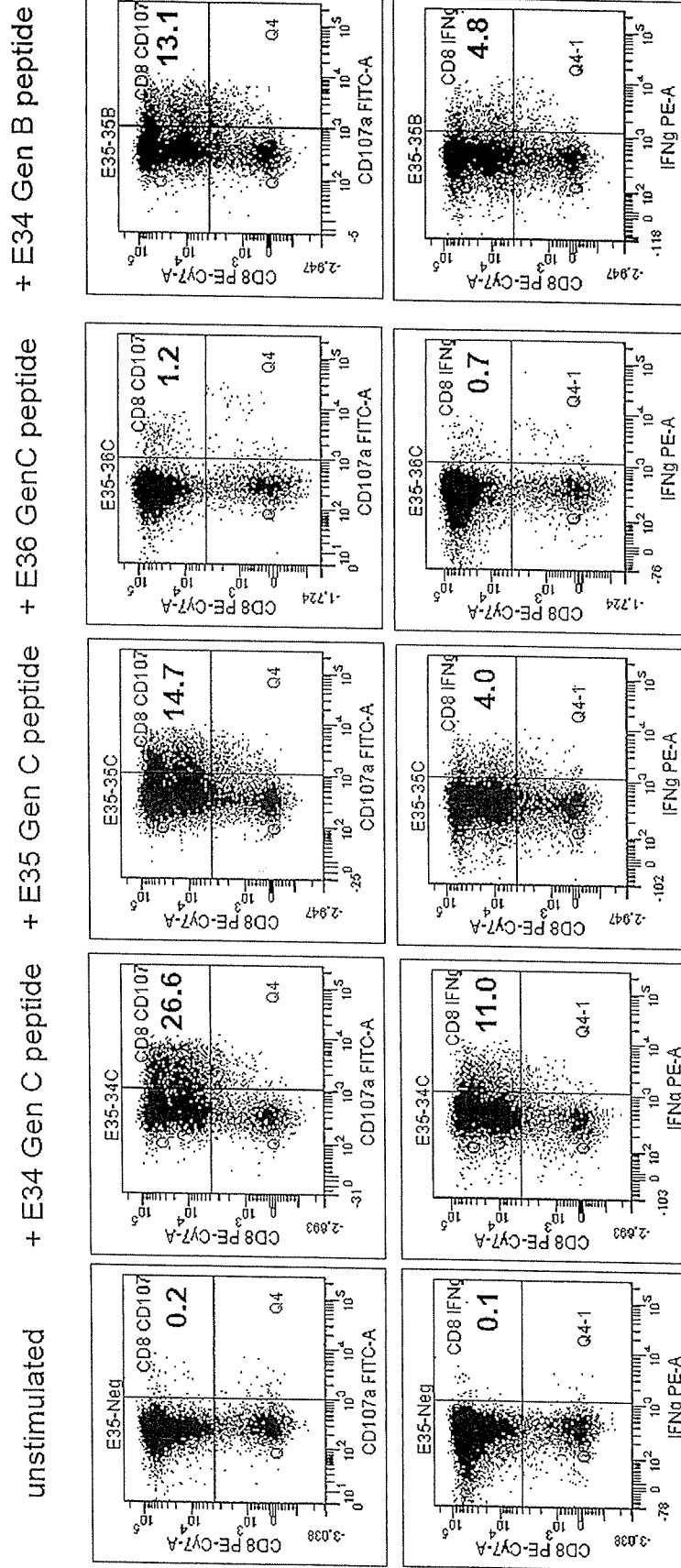


Figure 5

AA no.	Name	Amino Acid Sequence
166-180	15 mer HBVC Env 34	STTSGFLGPLLVLQA
171-185	15 mer HBVC Env 35	FLGPLLVLQAGFLL
170-179	Env 34-1	GLLGPLLVLQ
170-180	Env 34-2	GLLGPLLVLQA
171-180	Env 34-3	LLGPLLVLQA
172-180	Env 34-4	LGPLLVLQA
171-181	Env 34-5	LLGPLLVLQAG
172-181	Env 34-6	LGPLLVLQAG
173-181	Env 34-7	GPLLVLQAG
170-180	Env 34-8	GELGPLLVLQA
171-180	Env 34-9	FLGPLLVLQA
171-181	Env 34-10	FLGPLLVLQAG

Gen B

Gen C/D

Figure 6A

Name	Amino Acid Sequence	% CD8+ CD107a + Response (of total lymphocytes)	% CD8+ IFN- γ + Response (of total lymphocytes)
15 mer HBVC Env 34	STTSGFLGPIILVLQA	58.1	33.1
15 mer HBVC Env 35	FLGPIILVLQAGFFLL	58.8	37.4
HBVB Env 34-1	GLGPIILVLQ	19.4	1.8
HBVB Env 34-2	GLLGPIILVLQA	59.9	45.9
HBVB Env 34-3	LLGPIILVLQA	59.7	56.7
HBVB Env 34-4	LGPIILVLQA	36.0	7.3
HBVB Env 34-5	LLGPIILVLQAG	61.0	58.5
HBVB Env 34-6	LGPIILVLQAG	0.1	0.1
HBVB Env 34-7	GPIILVLQAG	25.9	3.6
HBVC/D Env 34-8	GLGPIILVLQA	61.1	58.0
HBVC/D Env 34-9	FLGPIILVLQA	59.4	57.6
HBVC/D Env 34-10	FLGPIILVLQAG	60.1	58.8

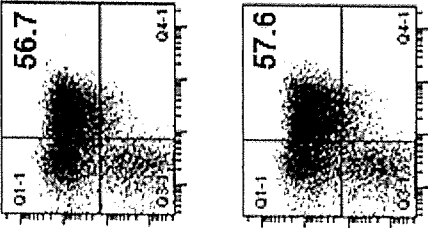


Figure 6B

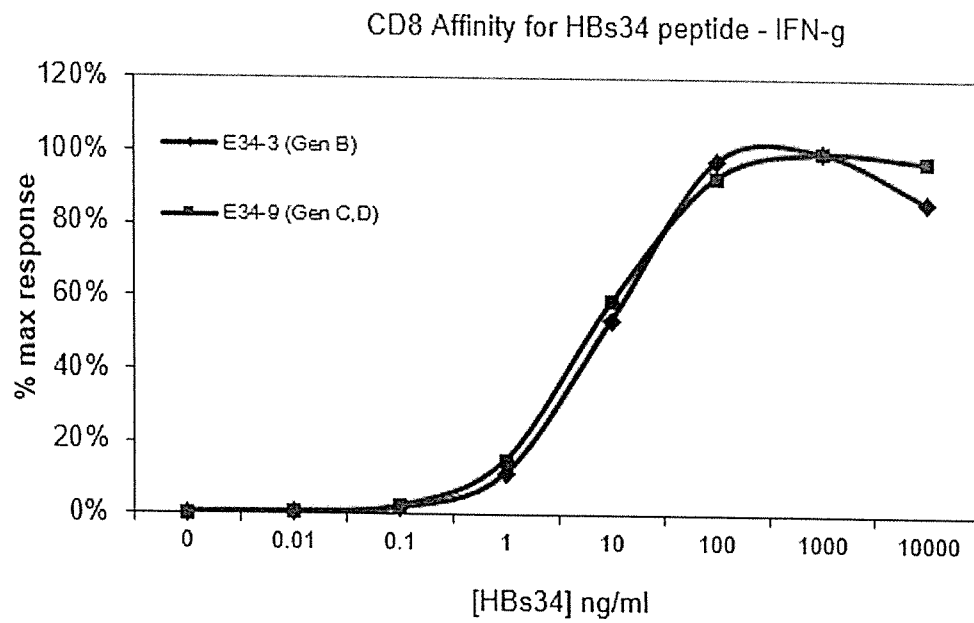
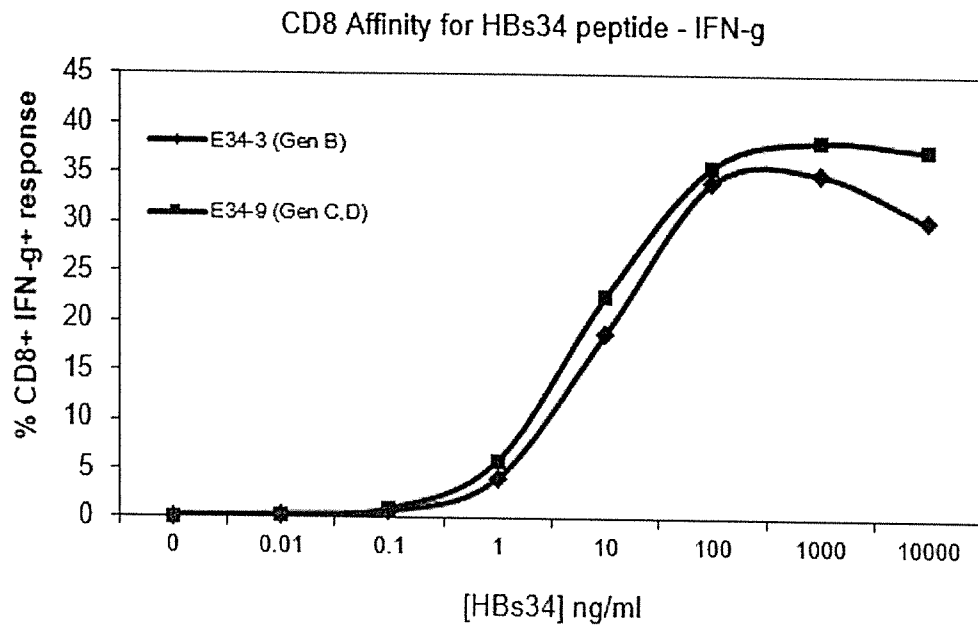


Figure 7

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	HLA - A	HLA - A	HLA - B	HLA - B	HLA - C	HLA - C
MK	A0201	A1101	B1502	B5304	Cw0801	Cw0401
WGP22	-	1101	1502	-	0801	-
CF0515	2601	2402	4006	5502	0801	1203

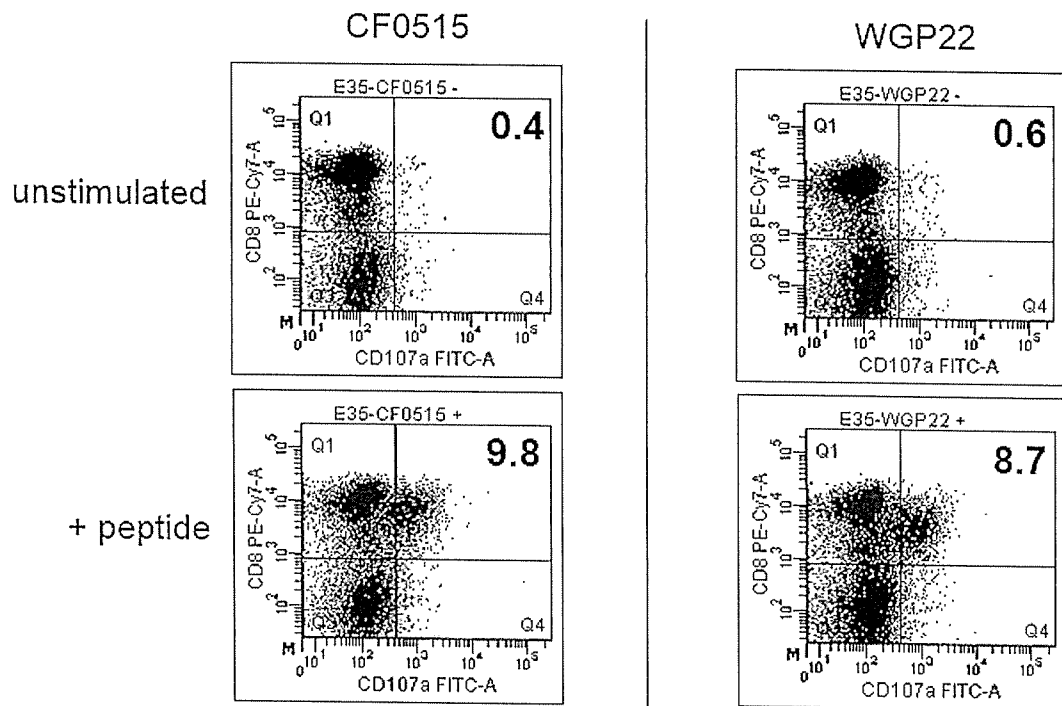


Figure 8

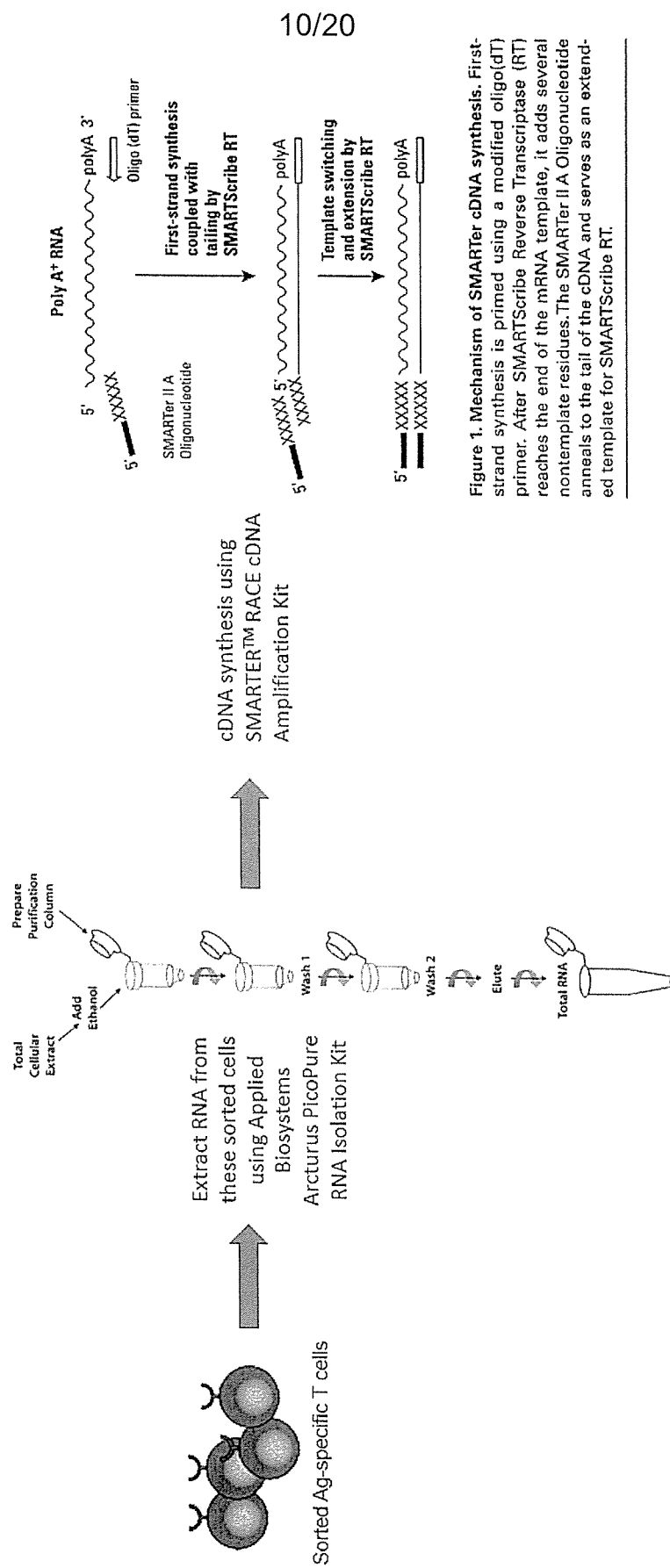


Figure 1. Mechanism of SMARTer cDNA synthesis. First-strand synthesis is primed using a modified oligo(dT) primer. After SMARTScribe Reverse Transcriptase (RT) reaches the end of the mRNA template, it adds several nontemplate residues. The SMARTer II A Oligonucleotide anneals to the tail of the cDNA and serves as an extended template for SMARTScribe RT.

Figure 9

Alpha chain

CDR1a: DXSSTY (SEQ ID NO: 1), or
 DSSSTY (SEQ ID NO:24), or
 DISSTY (SEQ ID NO:25)

CDR2a: IFSNMDM (SEQ ID NO: 2)

CDR3a: AETLDNYGQNFV (SEQ ID NO: 3)

Where X = S or I

Beta chain

CDR1b: DFQATT (SEQ ID NO: 4)

CDR2b: SNEGSKA (SEQ ID NO: 5)

CDR3b: SAVDRDEPFHSNQPQH (SEQ ID NO: 6)

	Before Codon Optimization	After Codon Optimization
CDR1a (where CDR1a = DSSSTY (SEQ ID NO: 24)	gacagctcctccacctac (SEQ ID NO: 7)	gactcctctagctacctac (SEQ ID NO: 13)
CDR2a	atcttttcaaataatggacatg (SEQ ID NO: 8)	atcttttccaacatggacatg (SEQ ID NO: 14)
CDR3a	gcagagaccttggaataactatggatcagaattttgtc (SEQ ID NO: 9)	gccgagaccctggacaactacggccagaatttcgtg (SEQ ID NO: 15)
CDR1b	gacttccaggccacaact (SEQ ID NO: 10)	gacttccaggccaccaca (SEQ ID NO: 16)
CDR2b	tccaatgagggctccaaggcc (SEQ ID NO: 11)	agcaacgaaggatccaaagcc (SEQ ID NO: 17)
CDR3b	agtgtgttagacagggatgaacctttccatagcaatc agccccagcat (SEQ ID NO: 12)	tcagcagtggaccgagatgaacctttccacagcaacca gccacagcal (SEQ ID NO: 18)

Figure 11

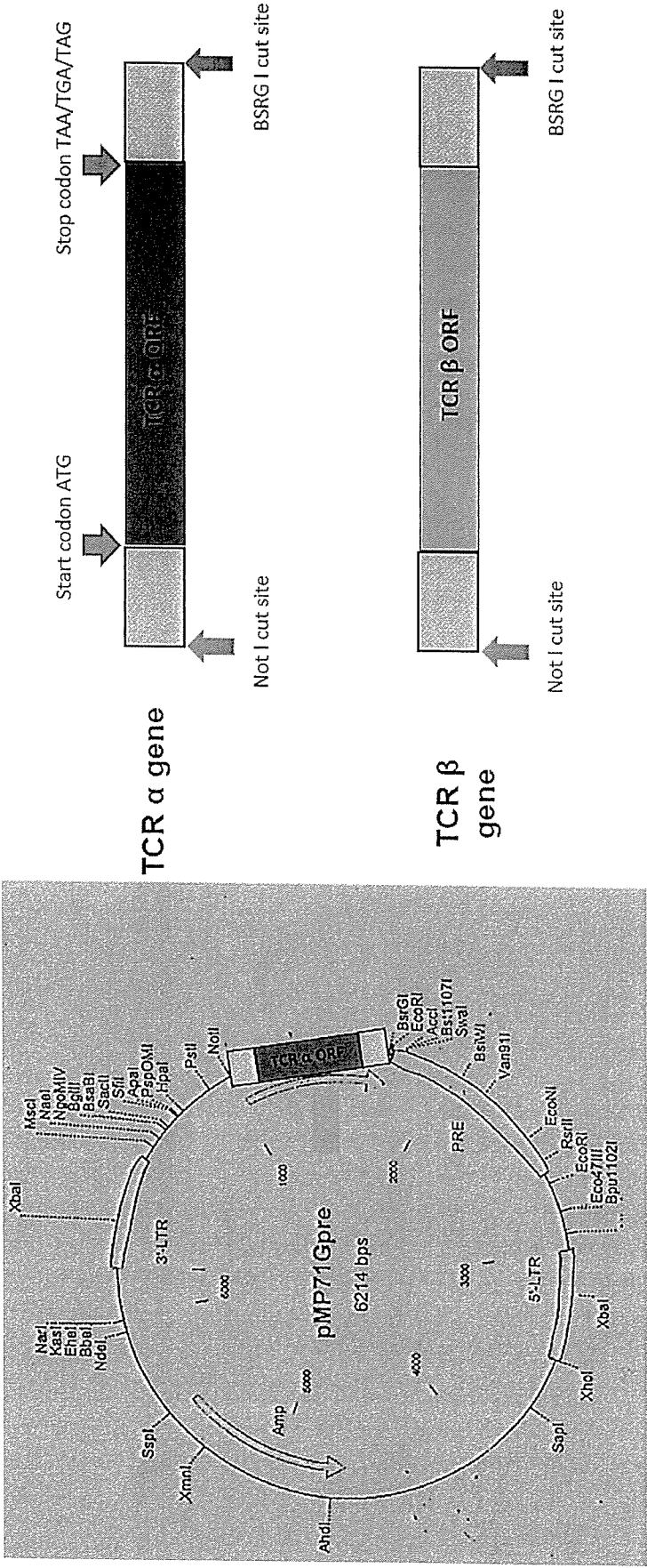


Figure 12

Retroviral Transduction

- Day 0-6: Transduction
- Day 7: Determine transduction efficiency by Vb & Pentamer staining
- Day 10-11: Test functionality of introduced TCR
 - Stimulate transduced T cells with peptide-loaded EBVB line
 - Stain for IFN- γ , TNF- α and IL-2

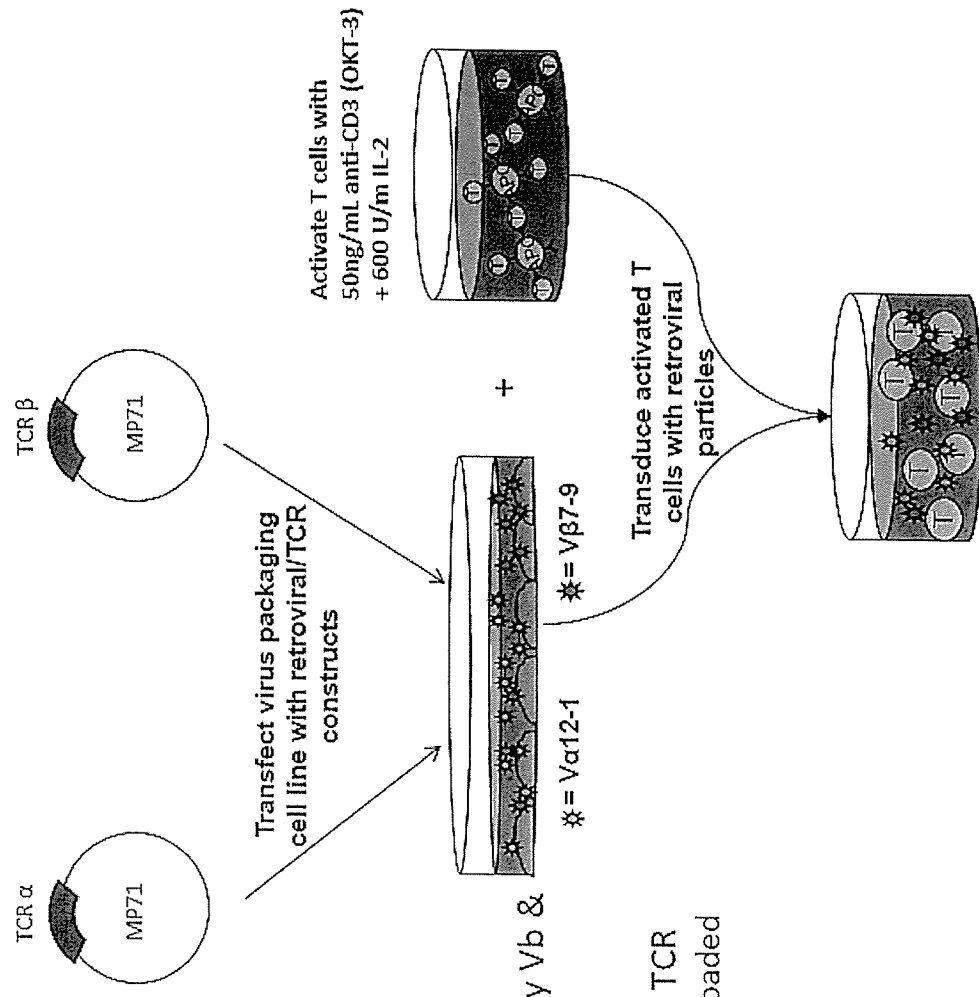
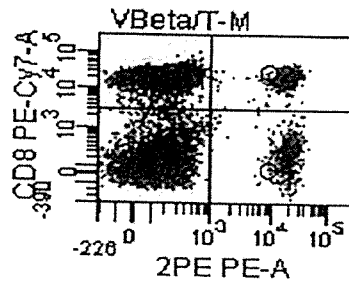


Figure 13

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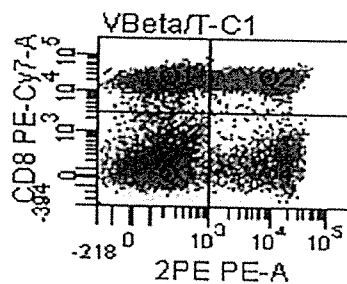
Vb Staining

MOCK



Tube Name: M		
Population	#Events	%Parent
☒ Q1	13,217	63.5
☒ Q2	800	3.8
☒ Q3	6,232	29.9
☒ Q4	589	2.7

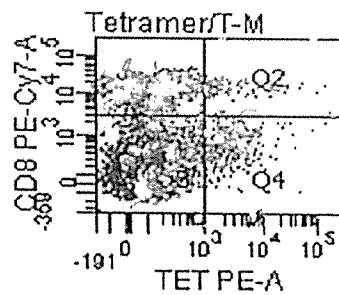
TCR-
Transduced



Tube Name: C1		
Population	#Events	%Parent
☒ Q1	10,200	48.4
☒ Q2	4,052	19.2
☒ Q3	5,178	24.6
☒ Q4	1,660	7.9

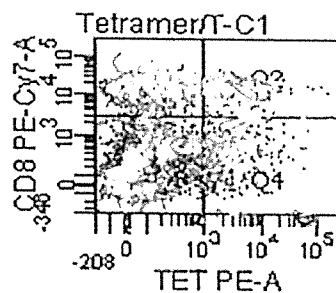
Pentamer Staining

MOCK



Experiment Name: ENV34-9 TCR TDX...		
Specimen Name: Tetramer/T		
Tube Name: M		
Record Date: Aug 1, 2011 12:07:...		
Population	#Events	%Parent
☒ Q1	14,067	66.5
☒ Q2	100	0.5
☒ Q3	6,614	31.3
☒ Q4	364	1.7

TCR-
Transduced



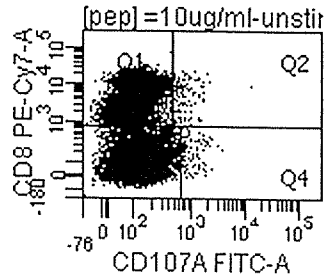
Experiment Name: ENV34-9 TCR TDX...		
Specimen Name: Tetramer/T		
Tube Name: C1		
Record Date: Aug 1, 2011 12:09:...		
Population	#Events	%Parent
☒ Q1	14,586	66.5
☒ Q2	442	2.0
☒ Q3	6,416	29.2
☒ Q4	501	2.3

Figure 14

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CD107a response

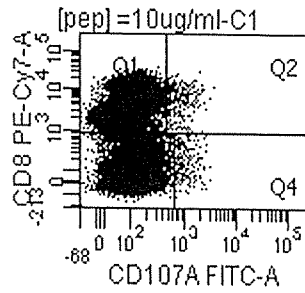
MOCK



Tube Name: unstim

Population	#Events	%Parent
■ Lymphocytes	23,949	47.7
☒ Q1	11,323	47.3
☒ Q2	137	0.6
☒ Q3	12,350	51.6
☒ Q4	139	0.6

TCR-
Transduced

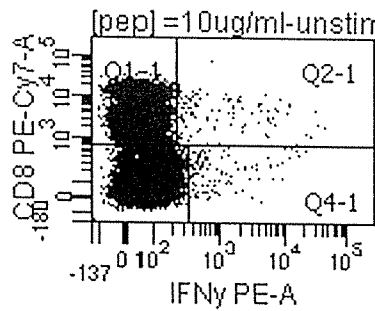


Tube Name: C1

Population	#Events	%Parent
■ Lymphocytes	34,347	45.1
☒ Q1	17,070	49.7
☒ Q2	335	1.0
☒ Q3	16,721	48.7
☒ Q4	221	0.6

IFN-g response

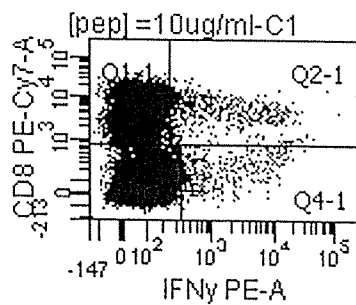
MOCK



Tube Name: unstim

Population	#Events	%Parent
■ Lymphocytes	23,949	47.7
☒ Q1-1	11,382	47.5
☒ Q2-1	123	0.5
☒ Q3-1	12,301	51.4
☒ Q4-1	143	0.6

TCR-
Transduced



Tube Name: C1

Population	#Events	%Parent
■ Lymphocytes	34,347	45.1
☒ Q1-1	16,810	48.9
☒ Q2-1	683	2.0
☒ Q3-1	16,397	47.7
☒ Q4-1	457	1.3

Figure 15

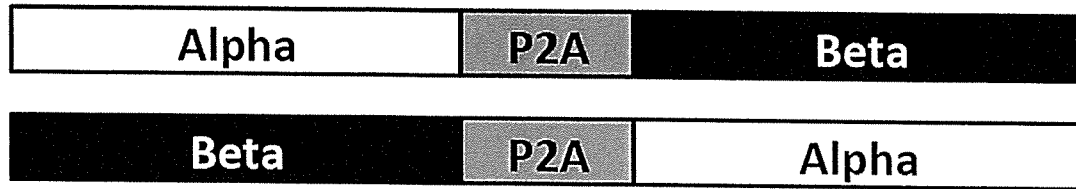


Figure 16

Vb Staining

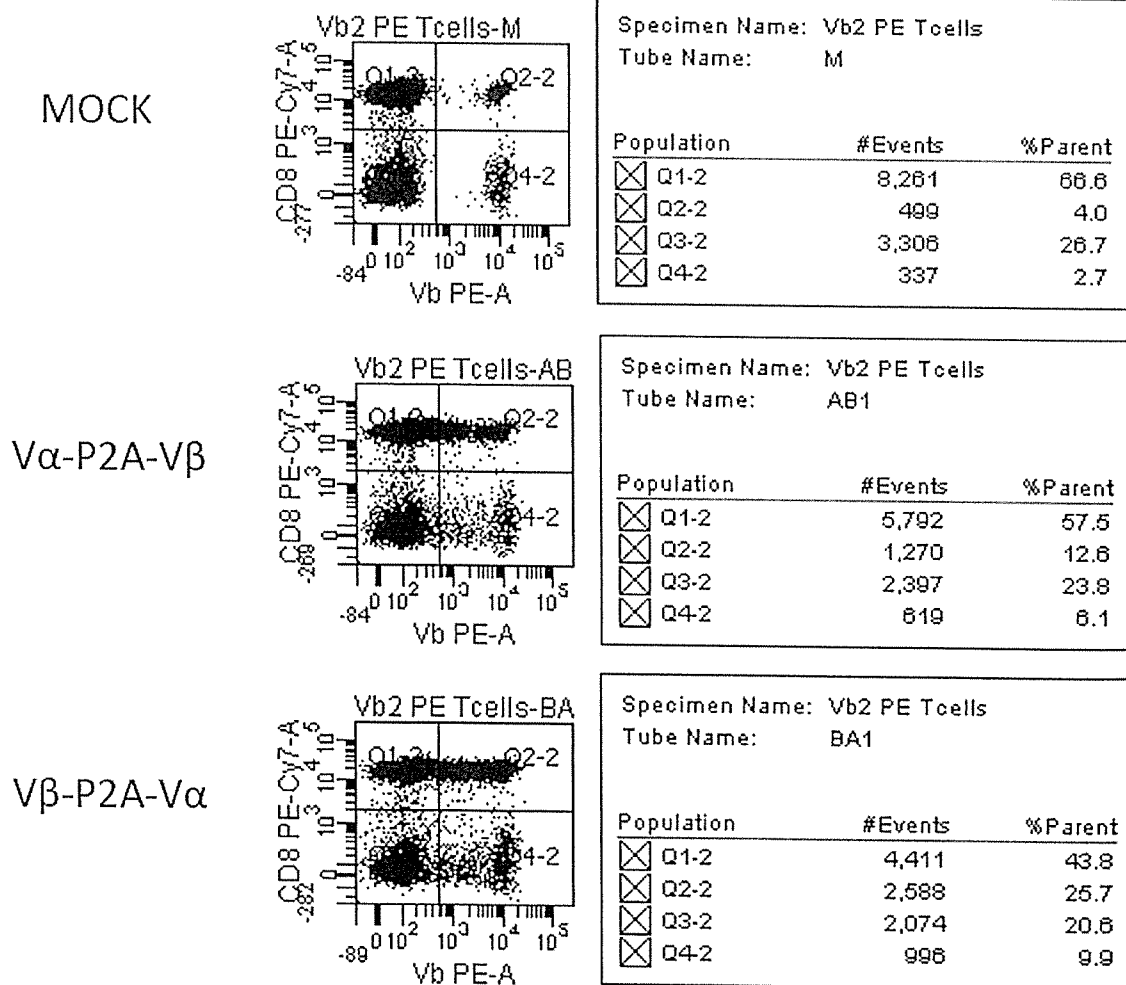
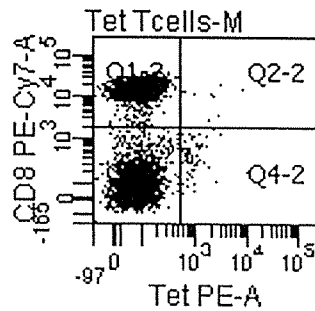


Figure 17

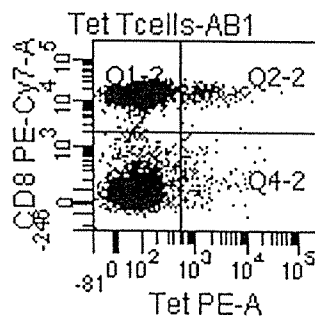
Pentamer Staining

MOCK



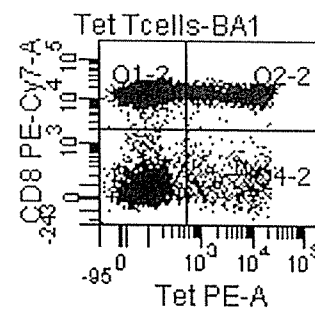
Specimen Name: Tet Tcells
Tube Name: M

Population	#Events	%Parent
☒ Q1-2	6,420	70.0
☒ Q2-2	12	0.1
☒ Q3-2	2,655	29.0
☒ Q4-2	81	0.9

V α -P2A-V β 

Specimen Name: Tet Tcells
Tube Name: AB1

Population	#Events	%Parent
☒ Q1-2	6,864	68.4
☒ Q2-2	215	2.1
☒ Q3-2	2,810	28.0
☒ Q4-2	151	1.5

V β -P2A-V α 

Specimen Name: Tet Tcells
Tube Name: BA1

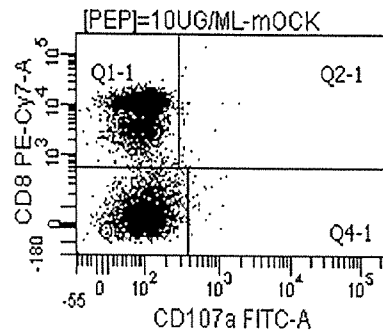
Population	#Events	%Parent
☒ Q1-2	5,521	55.0
☒ Q2-2	1,405	14.0
☒ Q3-2	2,524	25.1
☒ Q4-2	592	5.9

Figure 17 (cont.)

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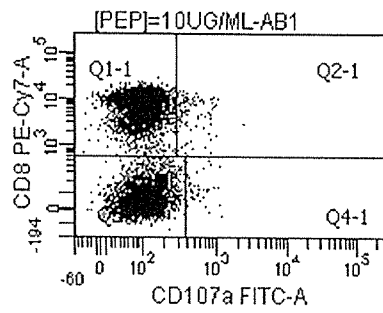
CD107a response

MOCK



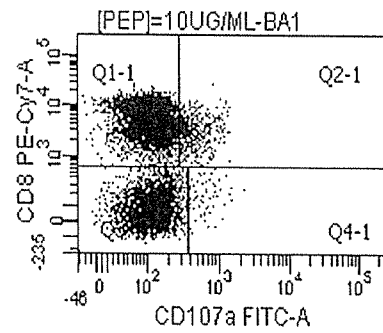
Tube Name: mOCK		
Population	#Events	%Parent
☒ Q1-1	4,741	61.6
☒ Q2-1	26	0.3
☒ Q3-1	2,899	37.7
☒ Q4-1	28	0.4

V α -P2A-V β



Tube Name: AB1		
Population	#Events	%Parent
☒ Q1-1	6,571	64.6
☒ Q2-1	121	1.2
☒ Q3-1	3,423	33.7
☒ Q4-1	53	0.5

V β -P2A-V α



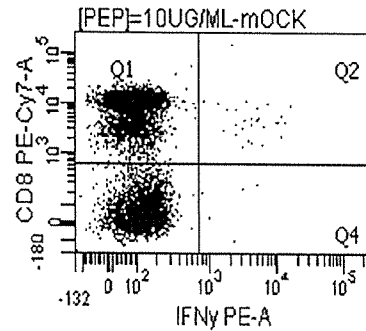
Tube Name: BA1		
Population	#Events	%Parent
☒ Q1-1	6,655	63.8
☒ Q2-1	561	5.4
☒ Q3-1	3,127	30.0
☒ Q4-1	82	0.8

Figure 18

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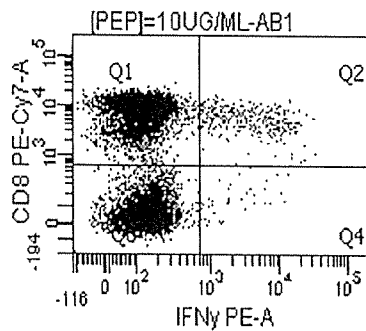
IFN-g response

MOCK



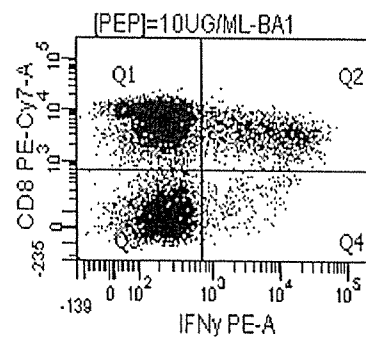
Tube Name: mOCK		
Population	#Events	%Parent
☒ Q1	4,729	61.5
☒ Q2	28	0.4
☒ Q3	2,932	38.1
☒ Q4	5	0.1

Vα-P2A-Vβ



Tube Name: AB1		
Population	#Events	%Parent
☒ Q1	6,384	62.6
☒ Q2	313	3.1
☒ Q3	3,448	33.9
☒ Q4	43	0.4

Vβ-P2A-Vα



Tube Name: BA1		
Population	#Events	%Parent
☒ Q1	5,830	55.9
☒ Q2	1,359	13.0
☒ Q3	3,018	28.9
☒ Q4	218	2.1

Figure 18 (cont.)