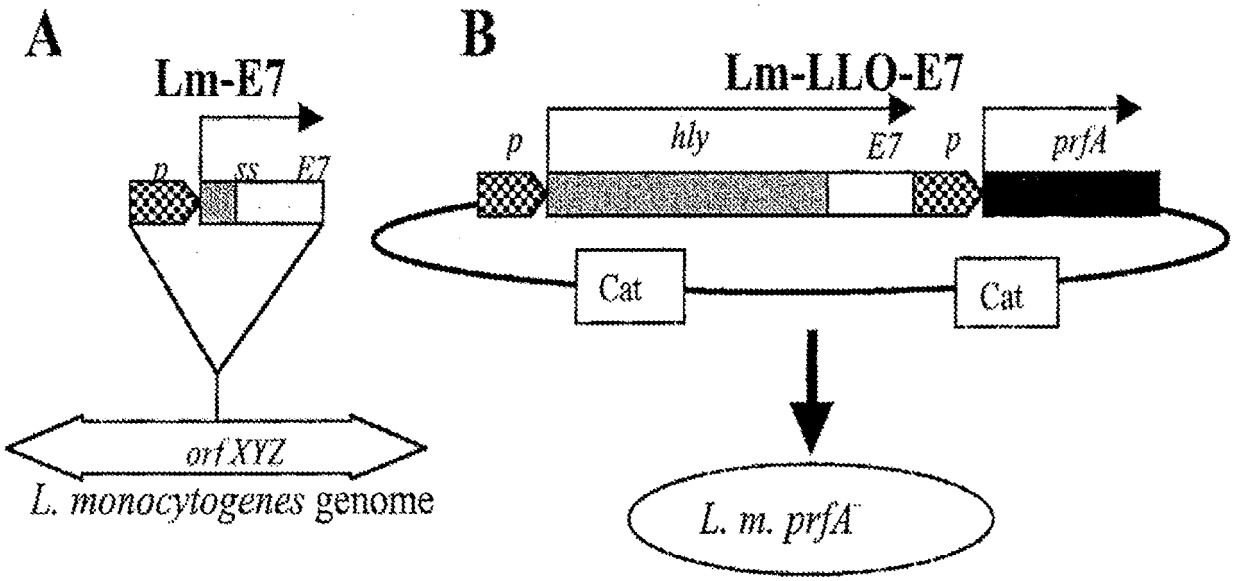


Figure 1

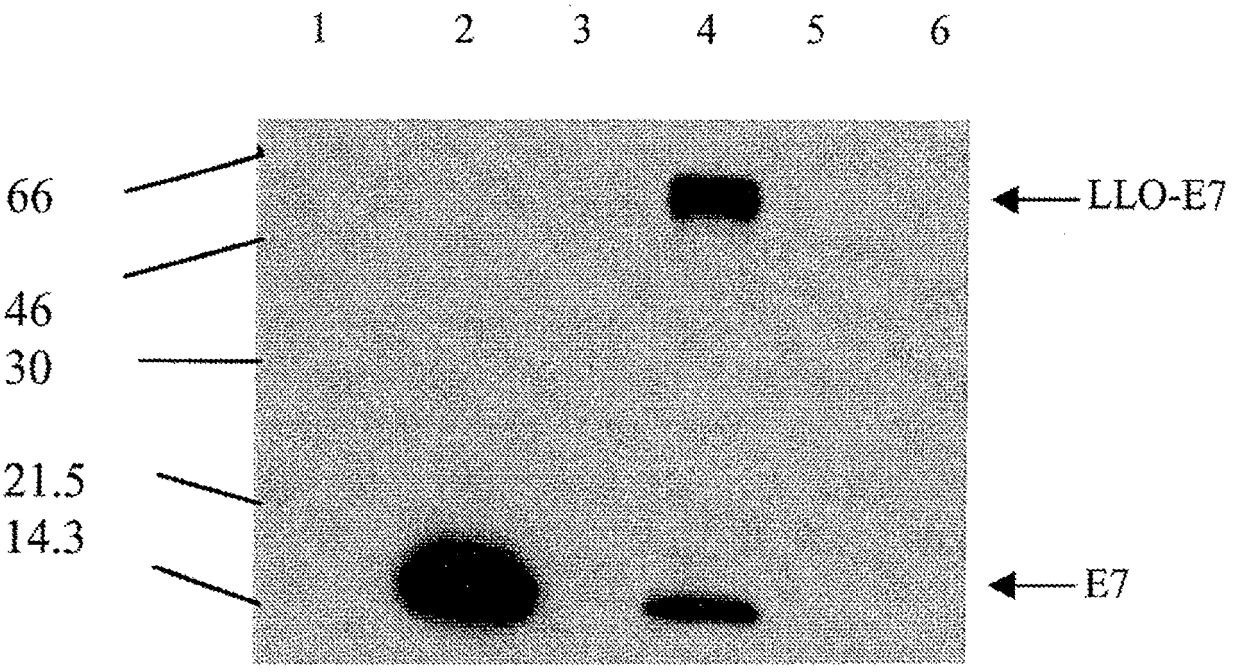


Figure 2

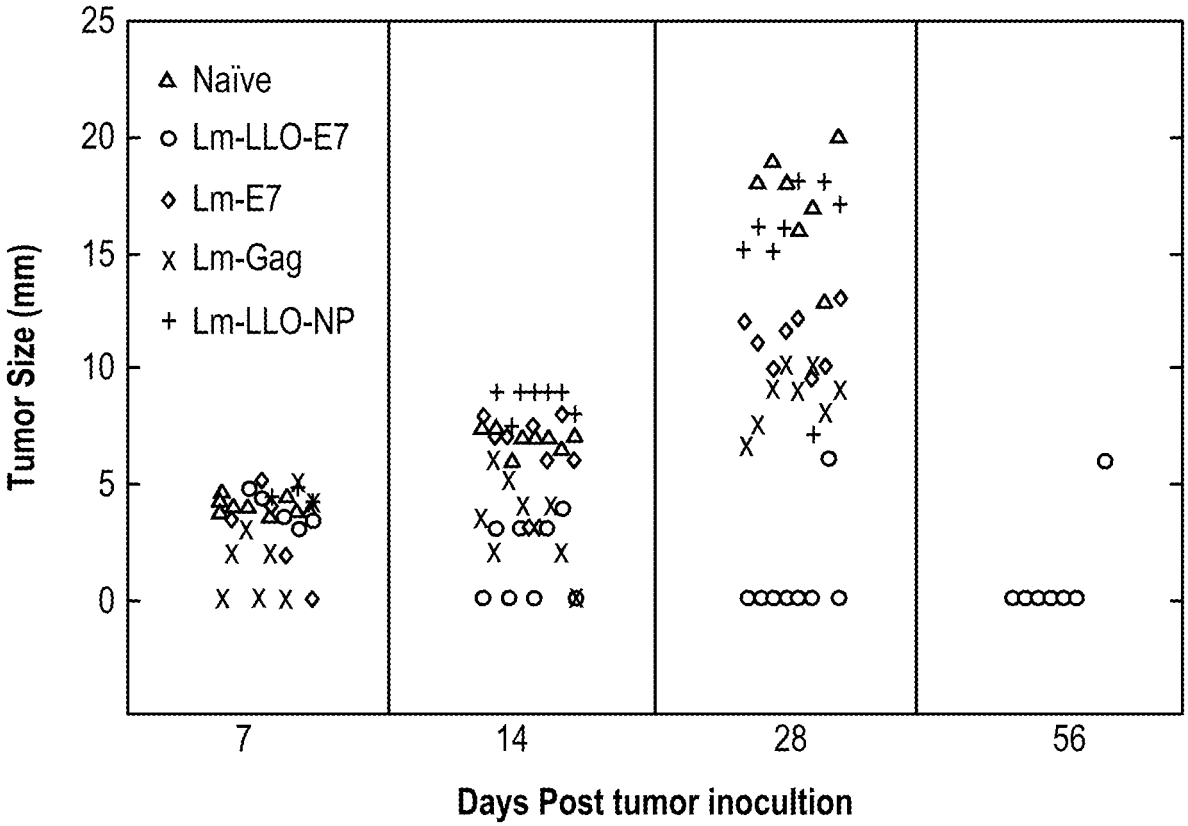

25
20
15
10
5
0
Tumor Size (mm)
Naïve
Lm-LLO-E7
Lm-E7
Lm-Gag
Lm-LLO-NP
7
14
28
56
Days Post tumor inocultion

Figure 3

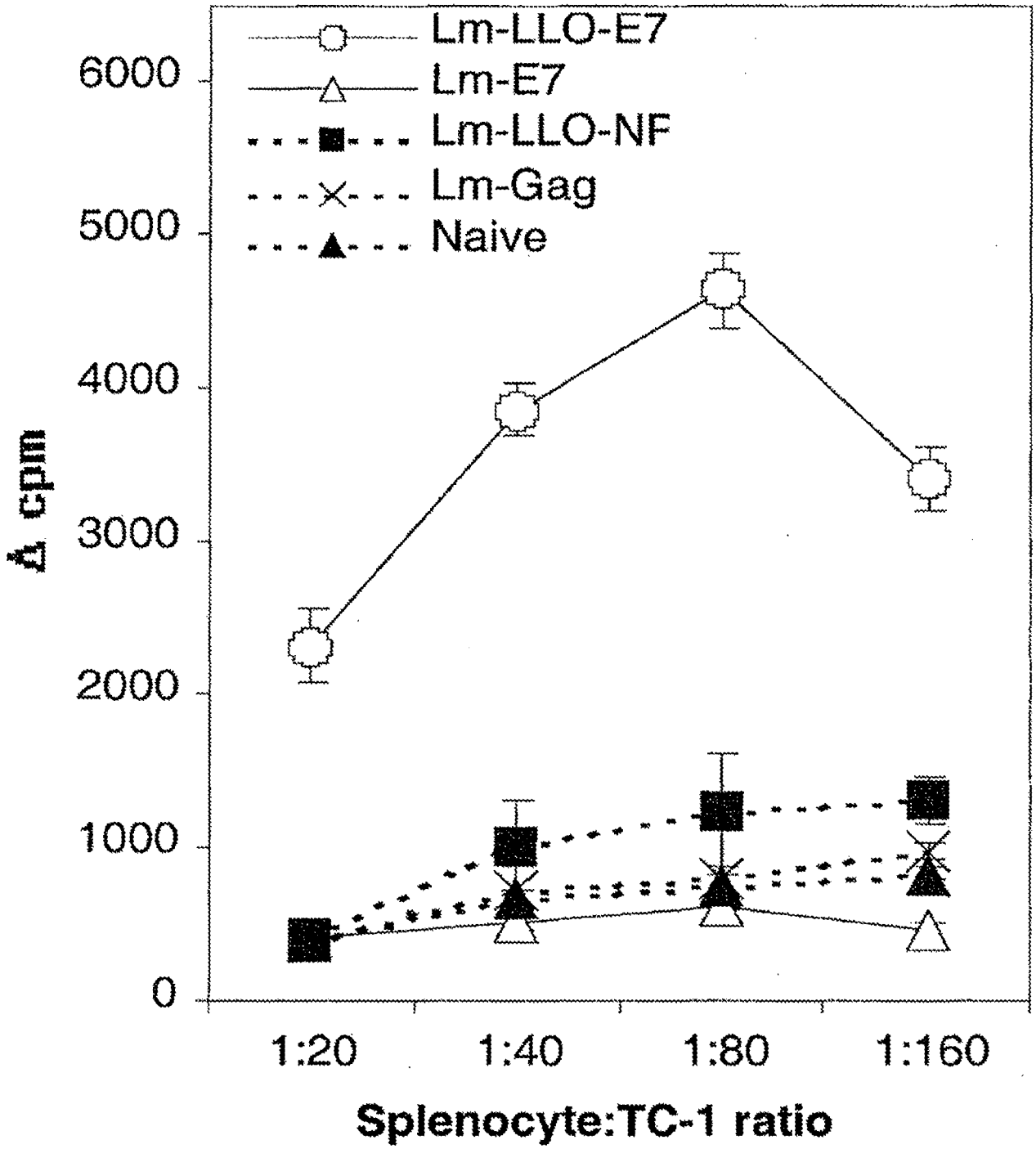

Lm-LLO-E7
Lm-E7
Lm-LLO-NF
Lm-Gag
Naive
6000
5000
4000
3000
2000
1000
0
Δ cpm
1:20
1:40
1:80
1:160
Splenocyte:TC-1 ratio

Figure 4

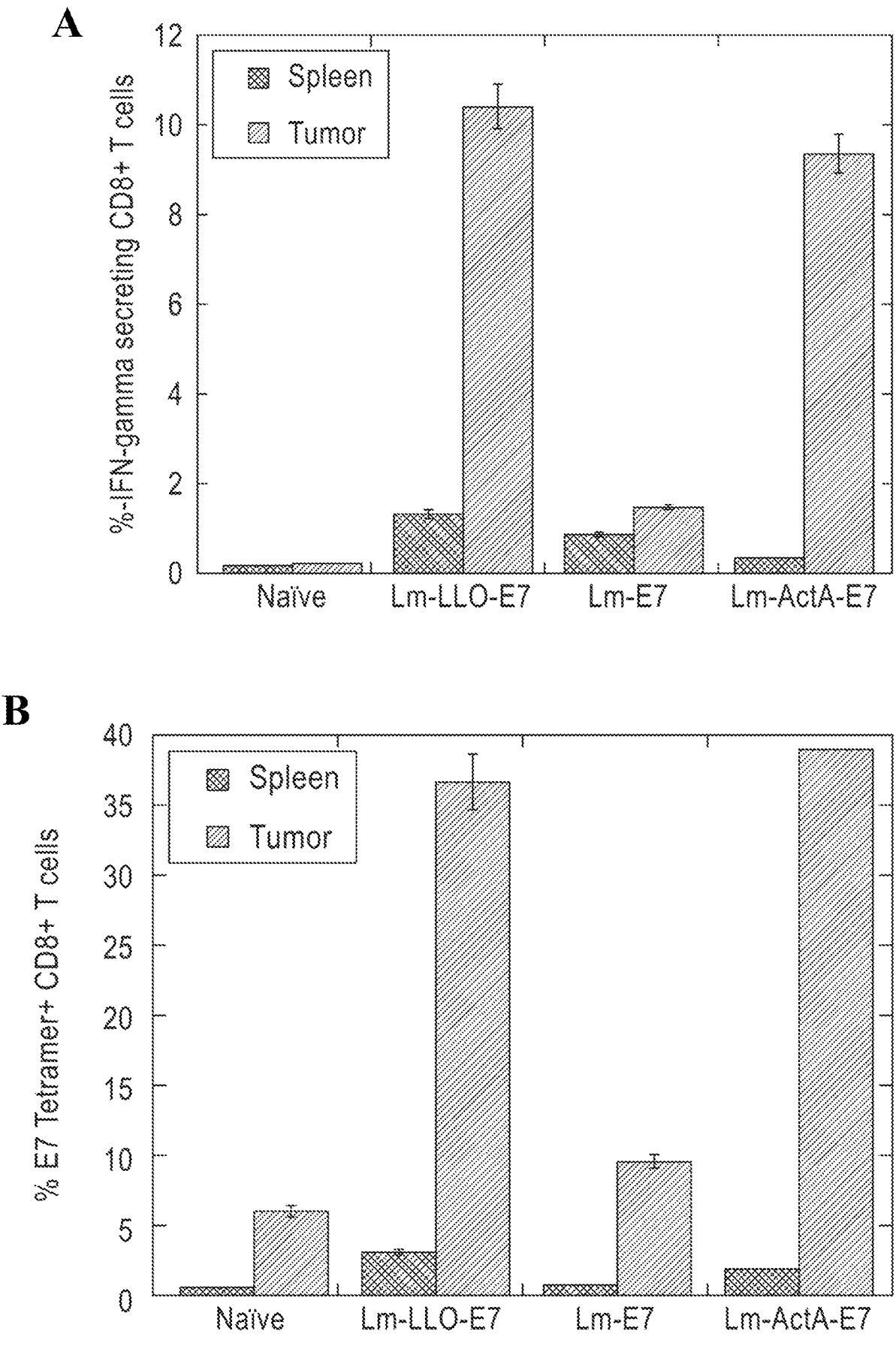

A
%-IFN-gamma secreting CD8+ T cells
Spleen
Tumor
0
2
4
6
8
10
12
Naïve
Lm-LLO-E7
Lm-E7
Lm-ActA-E7
B
% E7 Tetramer+ CD8+ T cells
Spleen
Tumor
0
5
10
15
20
25
30
35
40
Naïve
Lm-LLO-E7
Lm-E7
Lm-ActA-E7

Figure 5

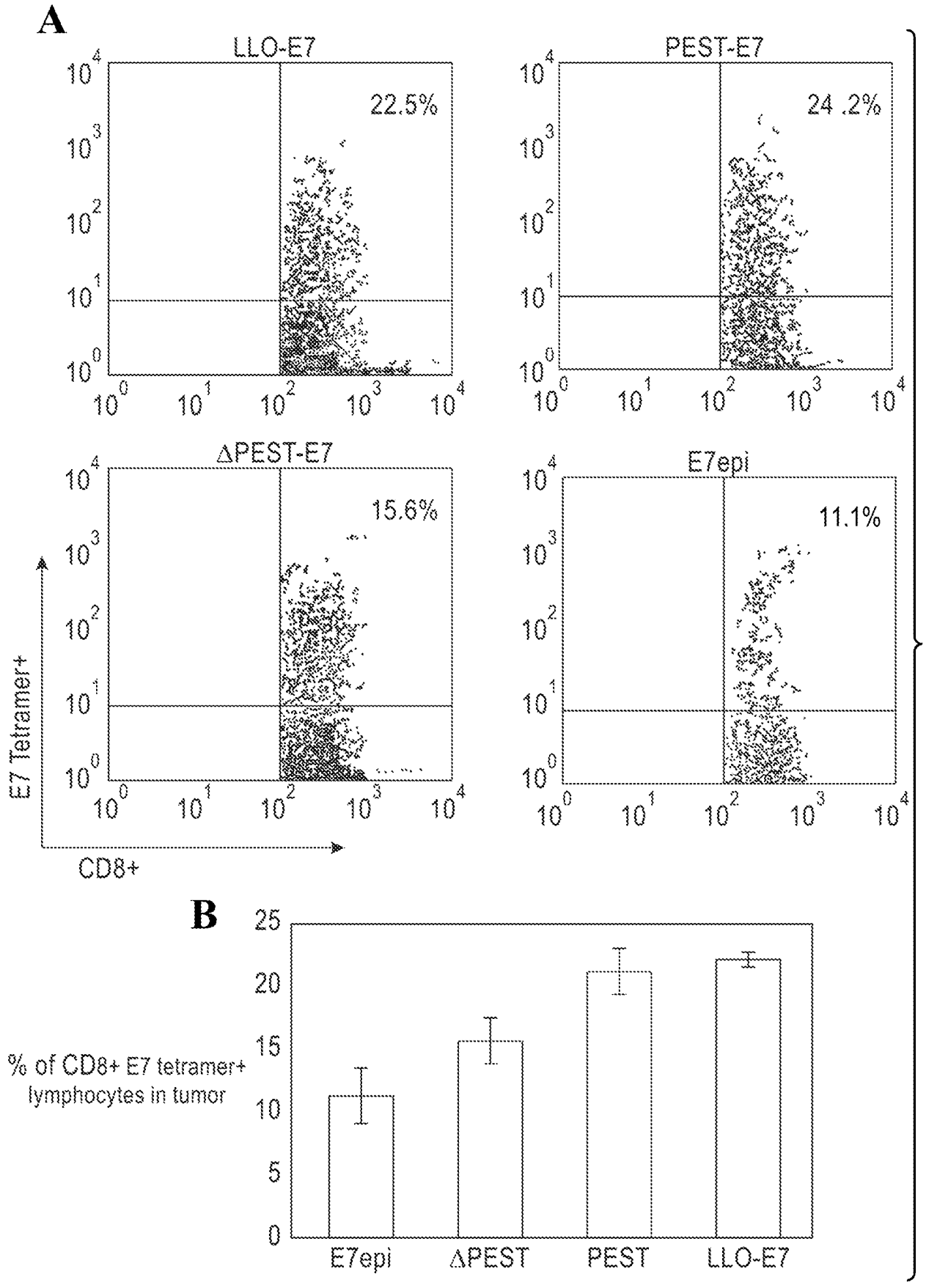

A
LLO-E7
22.5%
PEST-E7
24 .2%
ΔPEST-E7
15.6%
E7epi
11.1%
E7 Tetramer+
CD8+
B
% of CD8+ E7 tetramer+ lymphocytes in tumor
25
20
15
10
5
0
E7epi
ΔPEST
PEST
LLO-E7

Figure 6

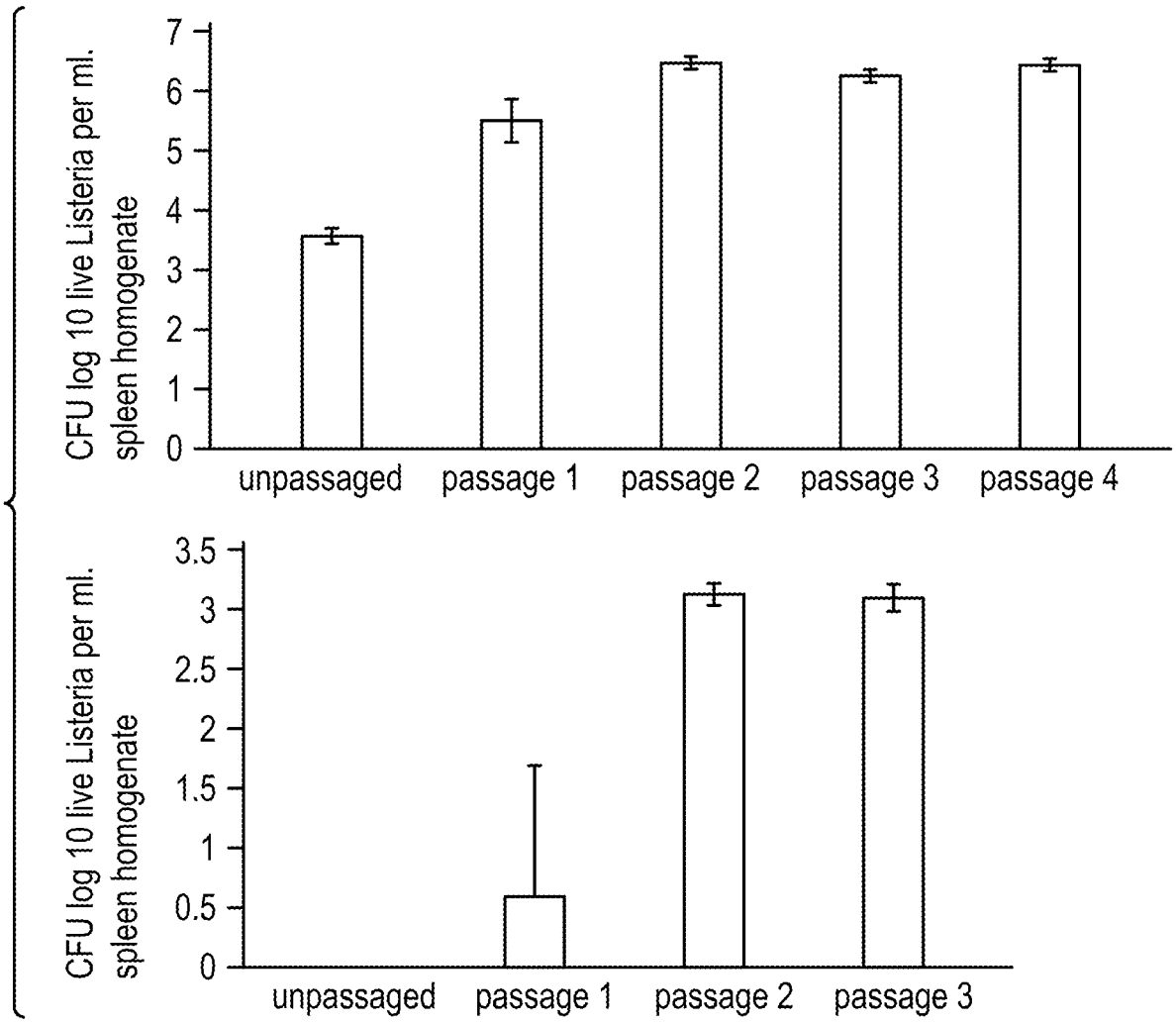


Figure 7A

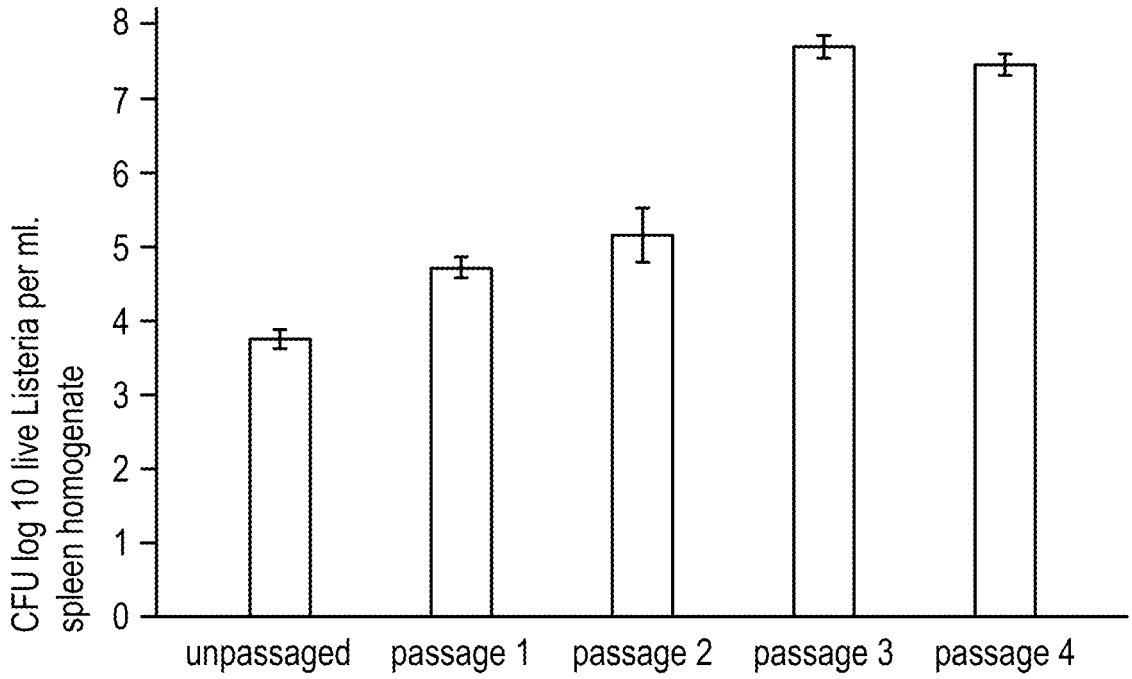


Figure 7B

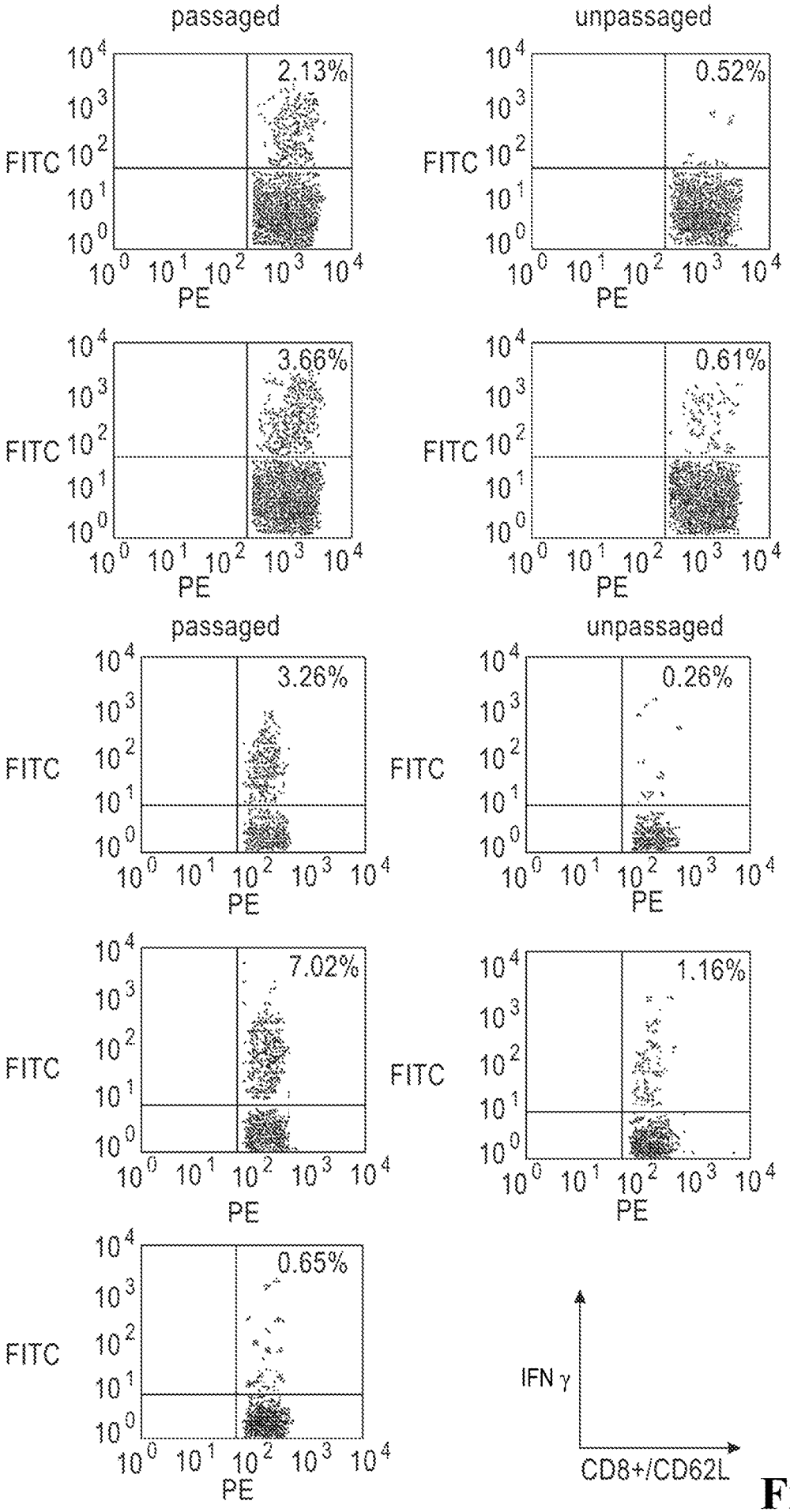
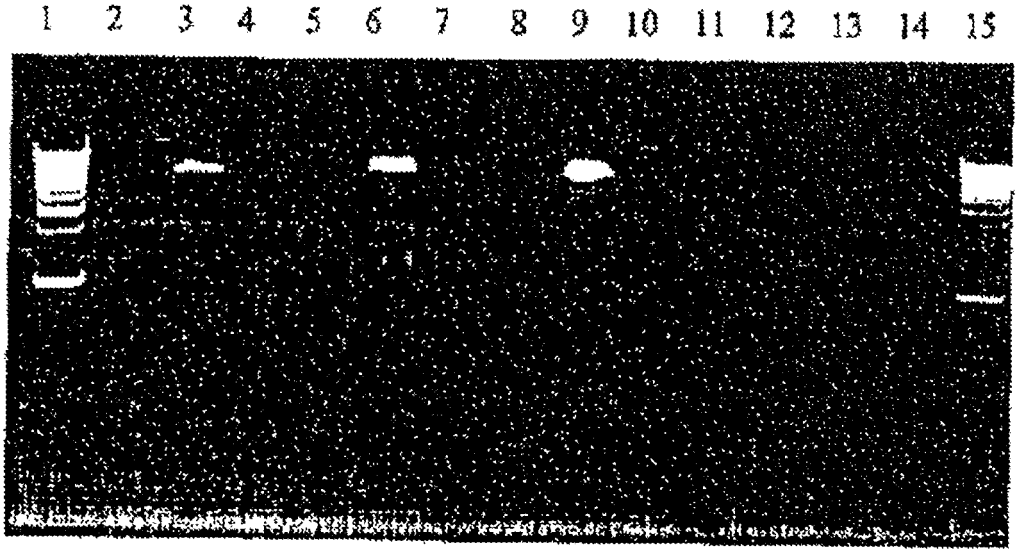

passaged
unpassaged
2.13%
0.52%
3.66%
0.61%
passaged
unpassaged
3.26%
0.26%
7.02%
1.16%
0.65%
FITC
PE
IFN γ
CD8+/CD62L

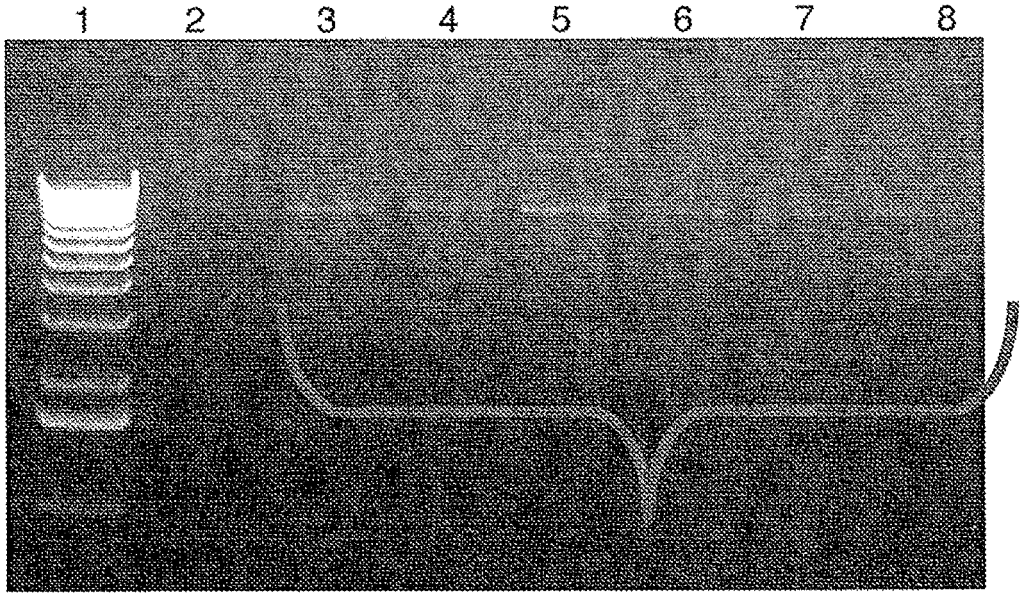
Figure 8



Lane	Sample	Lane	Sample
1	1Kb ladder	9	LB B, generation 5
2	100ng reference pGG55	10	LB A, generation 9
3	LB A, generation 5	11	LB B, generation 14
4	LB A, generation 9	12	LB B, generation 19*
5	LB A, generation 14	13	LB B, generation 24*
6	LB A, generation 19	14	LB B, generation 29*
7	LB A, generation 24	15	1Kb ladder
8	LB A, generation 29		

* Residual ethanol remaining in sample, therefore the majority of the sample did not load into the well, resulting in a less intense plasmid band

Figure 9A



Lane	Sample		
1	1Kb ladder	5	TB, generation 21
2	100ng reference pGG55	6	TB, generation 28
3	TB, generation 7	7	TB, generation 35
4	TB, generation 14	8	TB, generation 42

Figure 9B

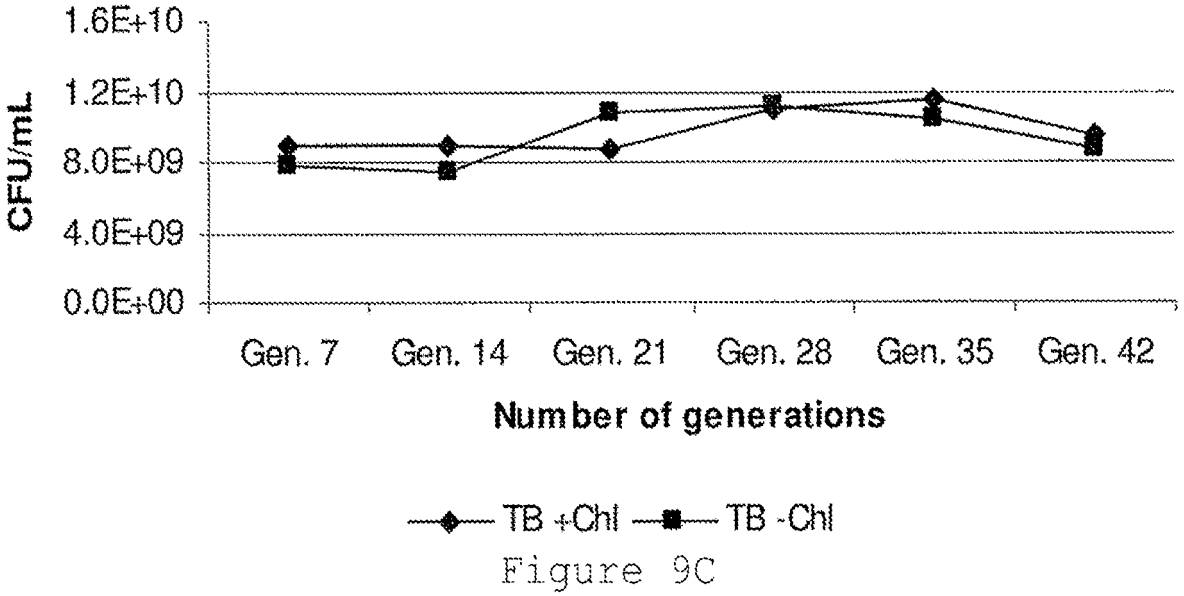

1.6E+10
1.2E+10
8.0E+09
4.0E+09
0.0E+00
CFU/mL
Gen. 7
Gen. 14
Gen. 21
Gen. 28
Gen. 35
Gen. 42
Number of generations
TB +Chl
TB -Chl

Figure 9C

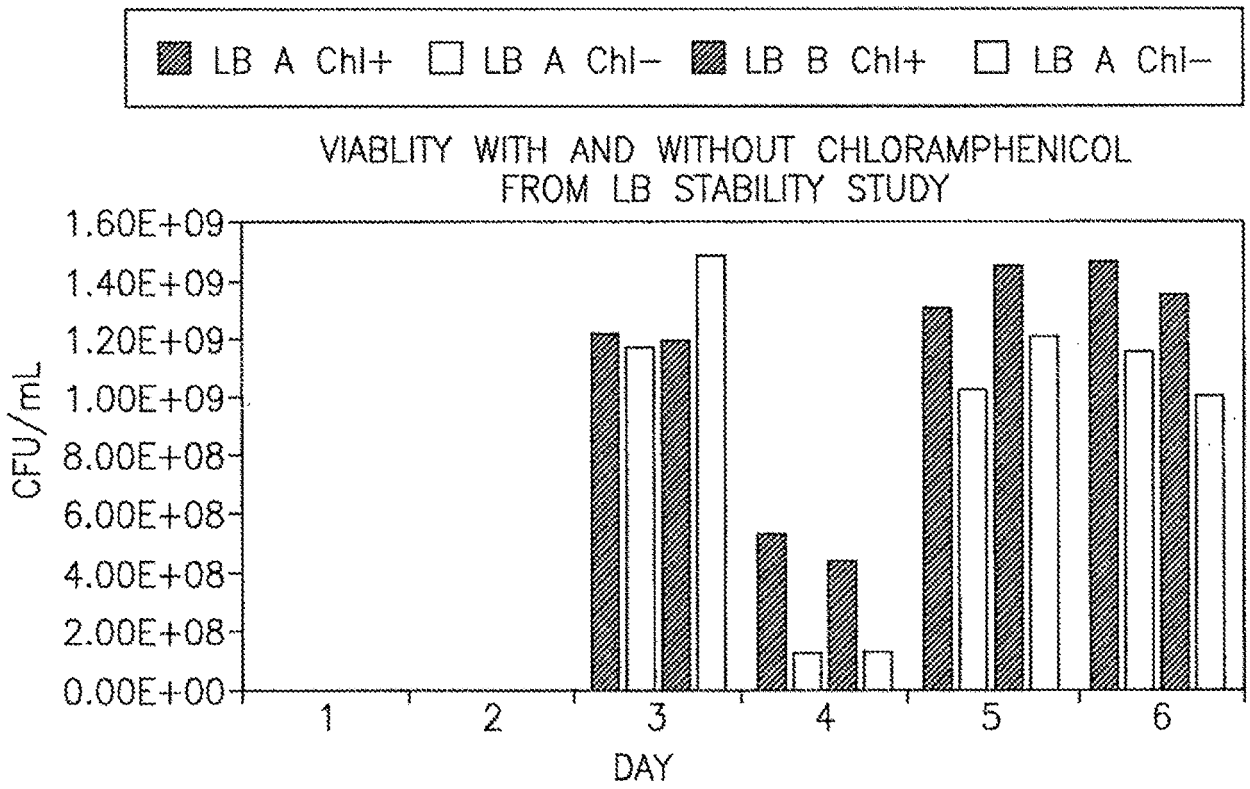


Figure 10

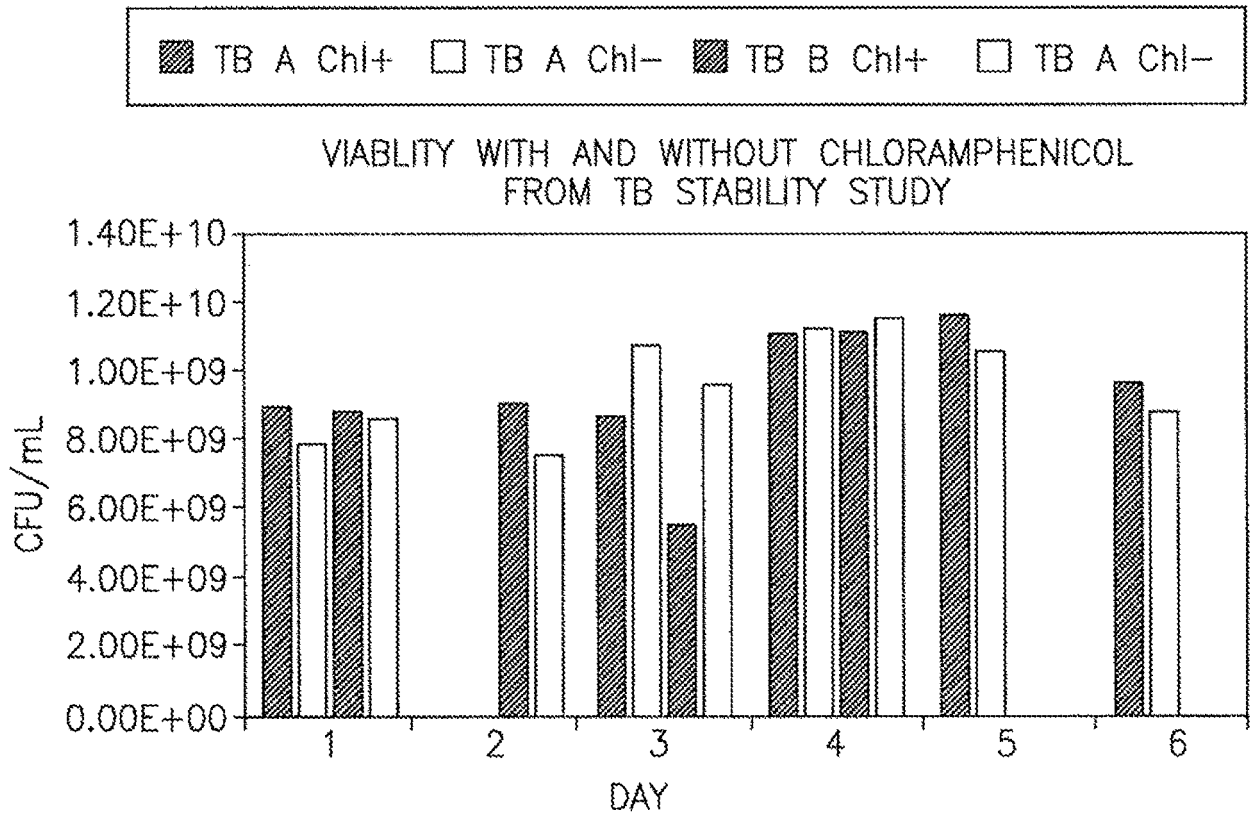
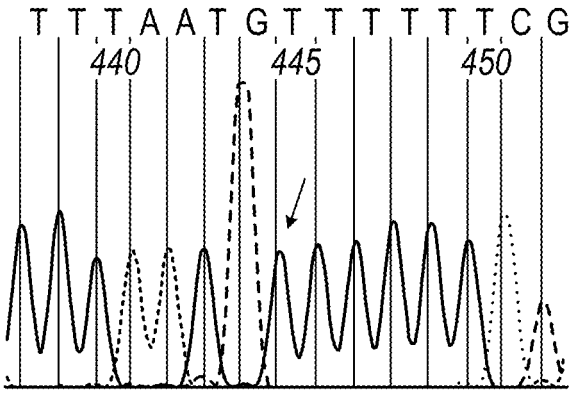
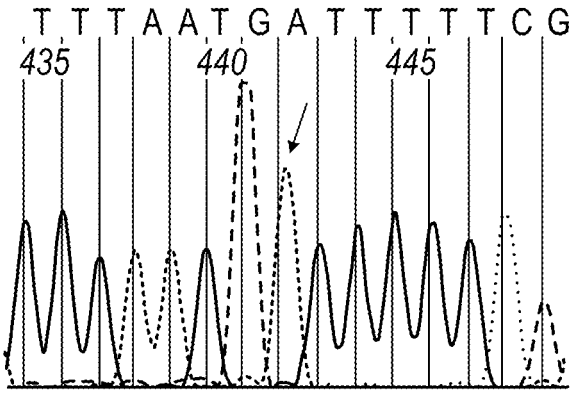


Figure 11

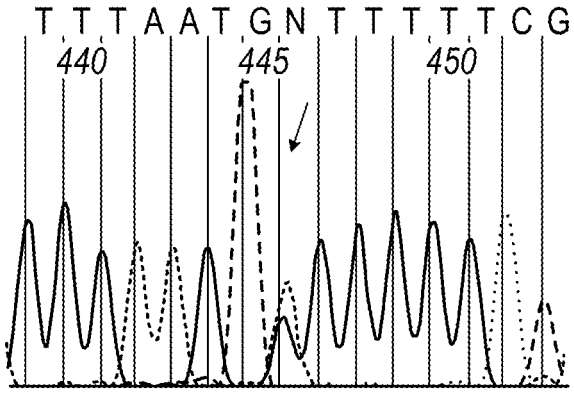
pGG55 D133V only (1:0)



pGG55 WT only (0:1)

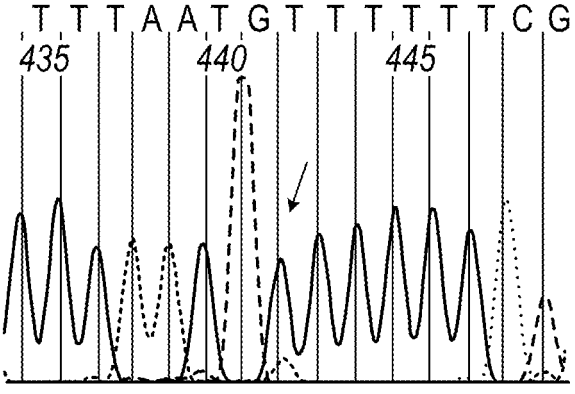


pGG55 D133V + pGG55 WT (1:1)



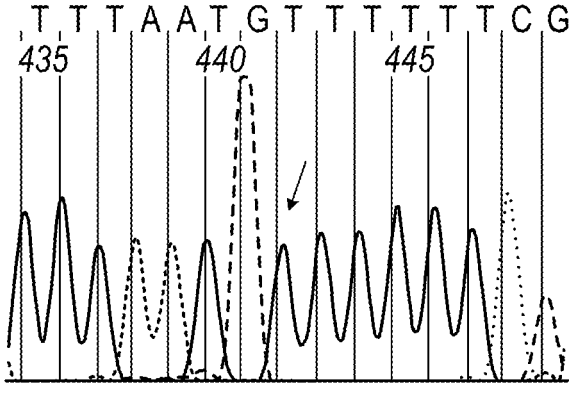
Two peaks are clearly visible and indicated by the arrow when plasmids are mixed in equimolar amounts.

pGG55 D133V + pGG55 WT (10:1)



1 in 10 is the maximum limit of detection and the peak from the wild-type sequence is still clearly detectable.

pGG55 D133V + pGG55 WT (100:1)



The peak from the wild-type sequence has disappeared completely when the wild-type prfA plasmid is diluted 1 to 100 or more.

Figure 12

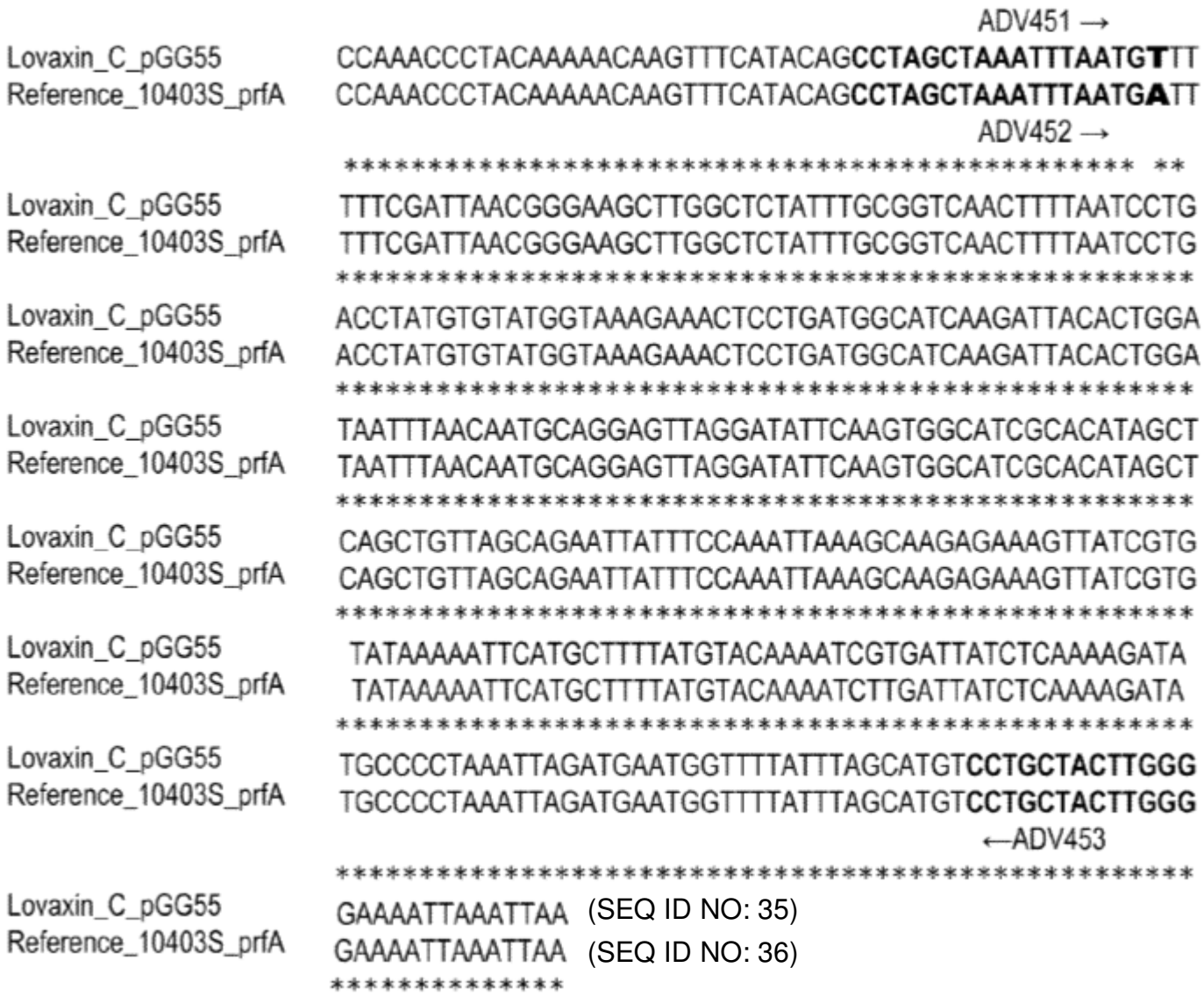
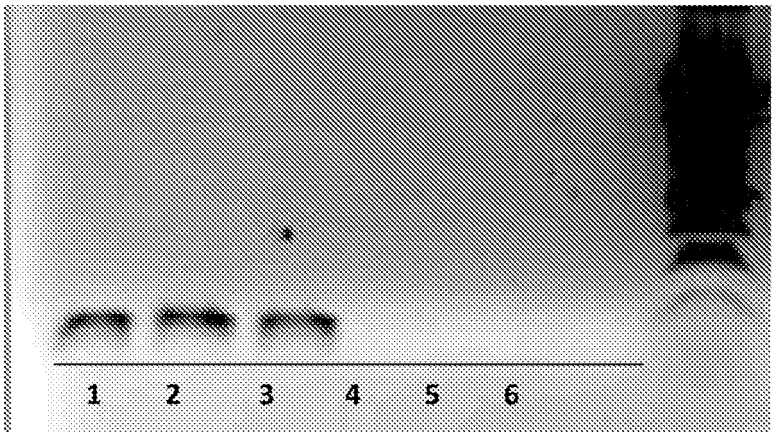
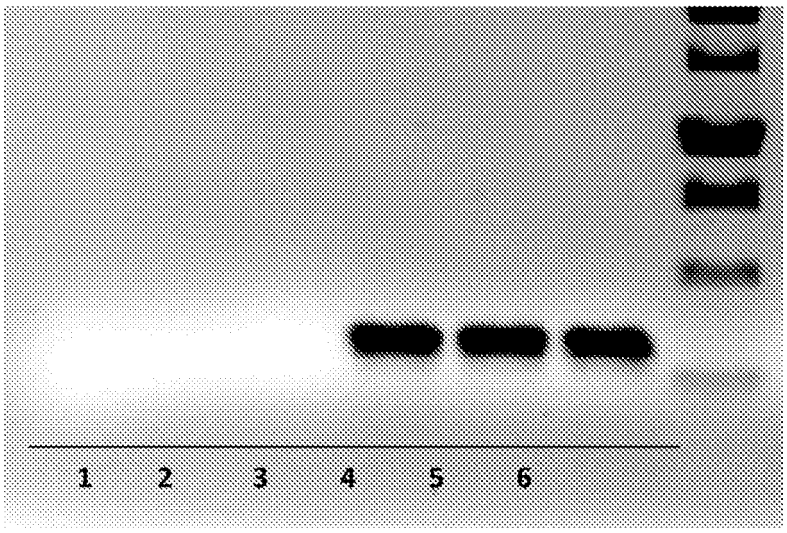

ADV451 →
Lovaxin_C_pGG55 CCAAACCCTACAAAAACAAGTTTCATACAGCCTAGCTAAATTTAATGTTT
Reference_10403S_prfA CCAAACCCTACAAAAACAAGTTTCATACAGCCTAGCTAAATTTAATGATT
ADV452 →
Lovaxin_C_pGG55 TTTCGATTAACGGGAAGCTTGGCTCTATTTGCGGTCAACTTTTAATCCTG
Reference_10403S_prfA TTTCGATTAACGGGAAGCTTGGCTCTATTTGCGGTCAACTTTTAATCCTG
Lovaxin_C_pGG55 ACCTATGTGTATGGTAAAGAAACTCCTGATGGCATCAAGATTACACTGGA
Reference_10403S_prfA ACCTATGTGTATGGTAAAGAAACTCCTGATGGCATCAAGATTACACTGGA
Lovaxin_C_pGG55 TAATTTAACAATGCAGGAGTTAGGATATTCAAGTGGCATCGCACATAGCT
Reference_10403S_prfA TAATTTAACAATGCAGGAGTTAGGATATTCAAGTGGCATCGCACATAGCT
Lovaxin_C_pGG55 CAGCTGTTAGCAGAATTATTTCCAAATTAAAGCAAGAGAAAGTTATCGTG
Reference_10403S_prfA CAGCTGTTAGCAGAATTATTTCCAAATTAAAGCAAGAGAAAGTTATCGTG
Lovaxin_C_pGG55 TATAAAAATTCATGCTTTTATGTACAAAATCGTGATTATCTCAAAAGATA
Reference_10403S_prfA TATAAAAATTCATGCTTTTATGTACAAAATCTTGATTATCTCAAAAGATA
Lovaxin_C_pGG55 TGCCCCTAAATTAGATGAATGGTTTTATTTAGCATGTCCTGCTACTTGGG
Reference_10403S_prfA TGCCCCTAAATTAGATGAATGGTTTTATTTAGCATGTCCTGCTACTTGGG
←ADV453
Lovaxin_C_pGG55 GAAAATTAAATTAA (SEQ ID NO: 35)
Reference_10403S_prfA GAAAATTAAATTAA (SEQ ID NO: 36)

Figure 13



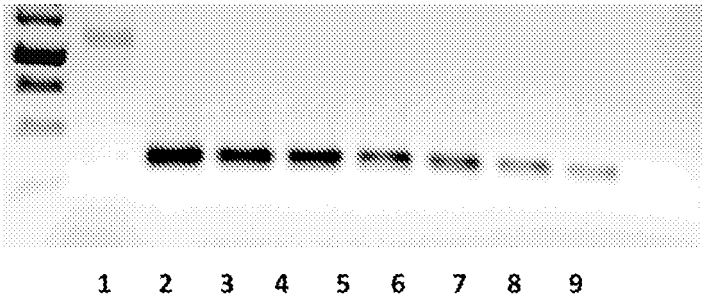
1. 5 ng of pGG55 D133V;
2. 1 ng of pGG55 D133V;
3. 0.2 ng of pGG55 D133V;
4. 5 ng of pGG55 wild-type;
5. 1 ng of pGG55 wild-type;
6. 0.2 ng of pGG55 wild-type;

Figure 14



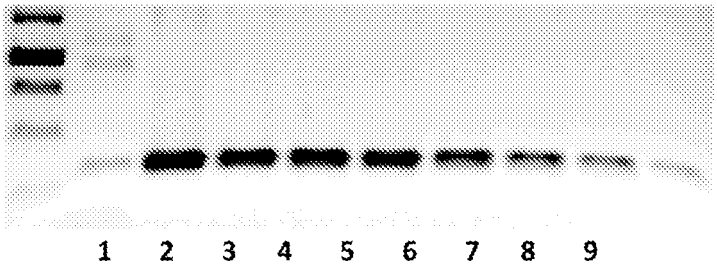
1) 5 ng of pGG55 D133V;
2) 1 ng of pGG55 D133V;
3) 0.2 ng of pGG55 D133V;
4) 5 ng of pGG55 wild-type;
5) 1 ng of pGG55 wild-type;
6) 0.2 ng of pGG55 wild-type;

Figure 15



1. 1 ng of pGG55 D133V
2. 1 ng of pGG55 wild-type
3. 100 pg of pGG55 wild-type (10^{-1})
4. 10 pg of pGG55 wild-type (10^{-2})
5. 1 pg of pGG55 wild-type (10^{-3})
6. 100 fg of pGG55 wild-type (10^{-4})
7. 10 fg of pGG55 wild-type (10^{-5})
8. 1 fg of pGG55 wild-type (10^{-6})

Figure 16



1. 5 ng of pGG55 D133V
2. 5 ng of pGG55 wild-type
3. 500 pg of pGG55 wild-type (10^{-1})
4. 50 pg of pGG55 wild-type (10^{-2})
5. 5 pg of pGG55 wild-type (10^{-3})
6. 500 fg of pGG55 wild-type (10^{-4})
7. 50 fg of pGG55 wild-type (10^{-5})
8. 5 fg of pGG55 wild-type (10^{-6})

Figure 17

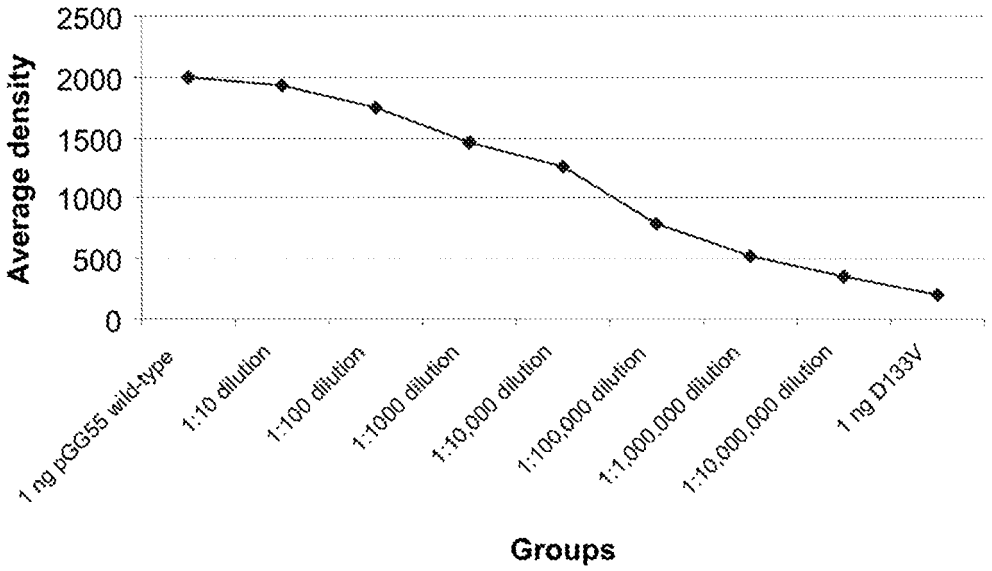


Figure 18

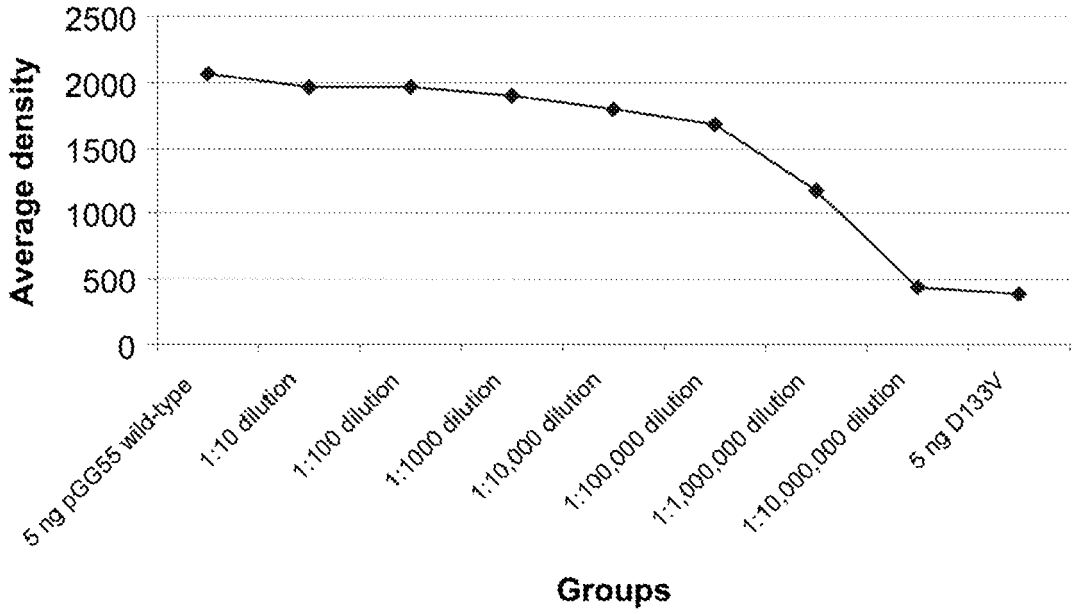


Figure 19

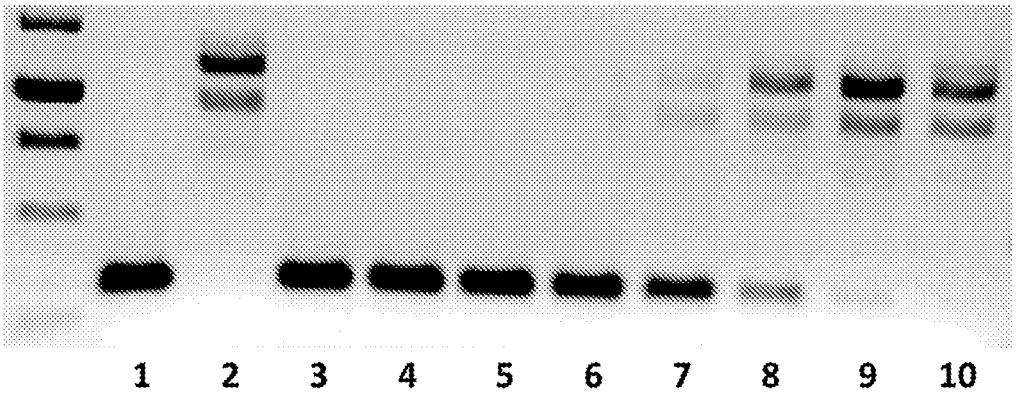


Figure 20

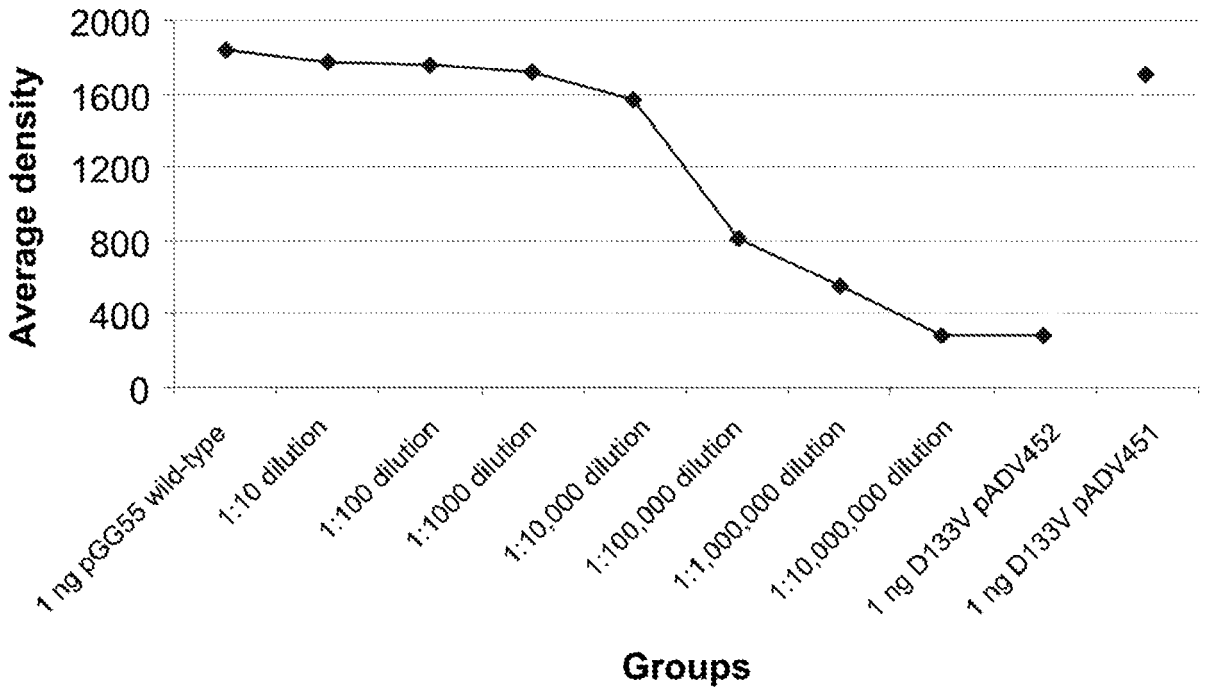
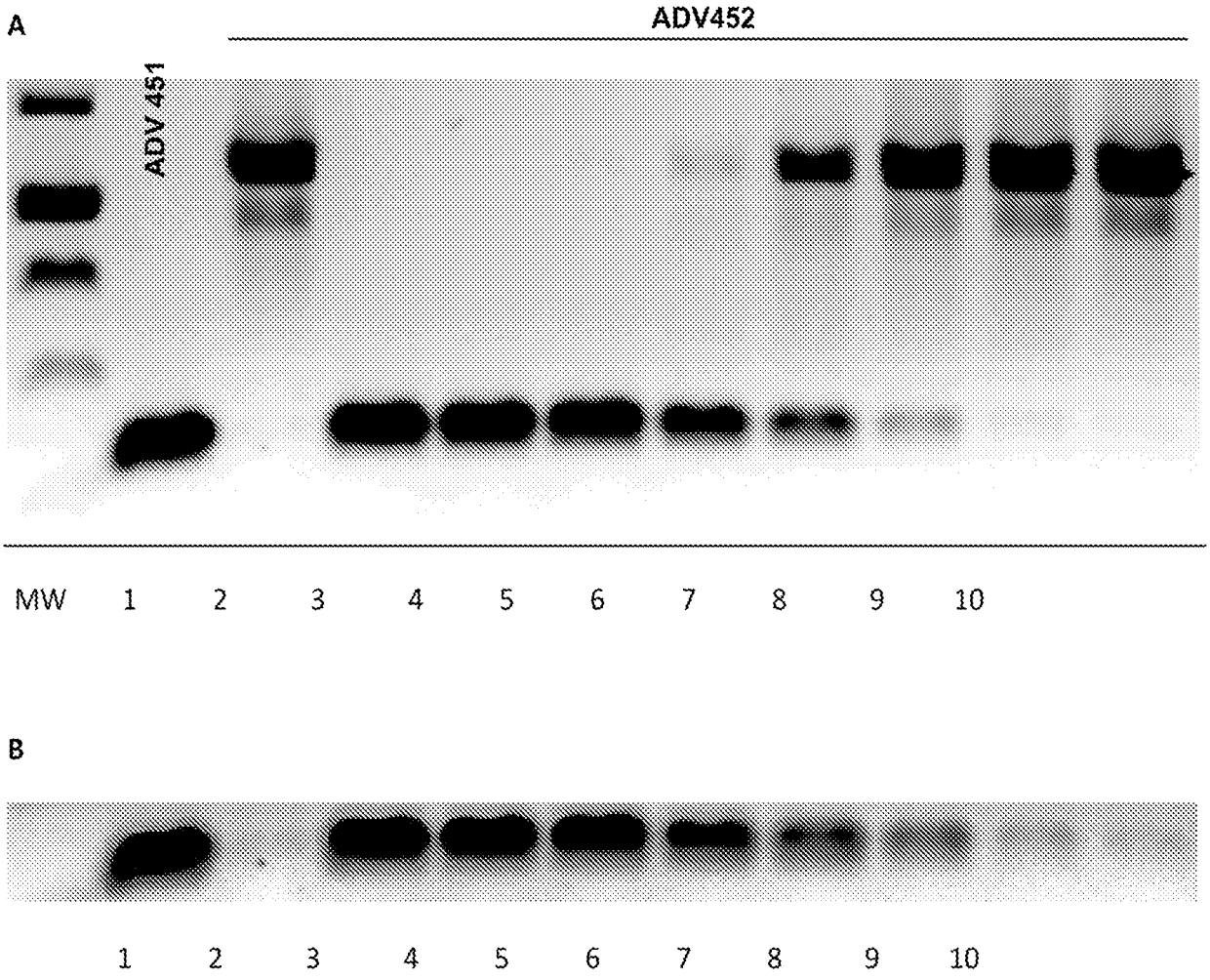

Average density
2000
1600
1200
800
400
0
1 ng pGG55 wild-type
1:10 dilution
1:100 dilution
1:1000 dilution
1:10,000 dilution
1:100,000 dilution
1:1,000,000 dilution
1:10,000,000 dilution
1 ng D133V pADV452
1 ng D133V pADV451
Groups

Figure 21



1) 1 ng of pGG55 D133V only

2) 1 ng of pGG55 D133V only

3) 1 ng of pGG55 D133V + 1 ng of pGG55 wild-type prfA

4) 1 ng of pGG55 D133V + 100 pg of pGG55 wild-type prfA

6) 1 ng of pGG55 D133V + 1 pg of pGG55 wild-type prfA

7) 1 ng of pGG55 D133V + 100 fg of pGG55 wild-type prfA

8) 1 ng of pGG55 D133V + 10 fg of pGG55 wild-type prfA

9) 1 ng of pGG55 D133V + 1 fg of pGG55 wild-type prfA

Figure 22