

Fig. 1

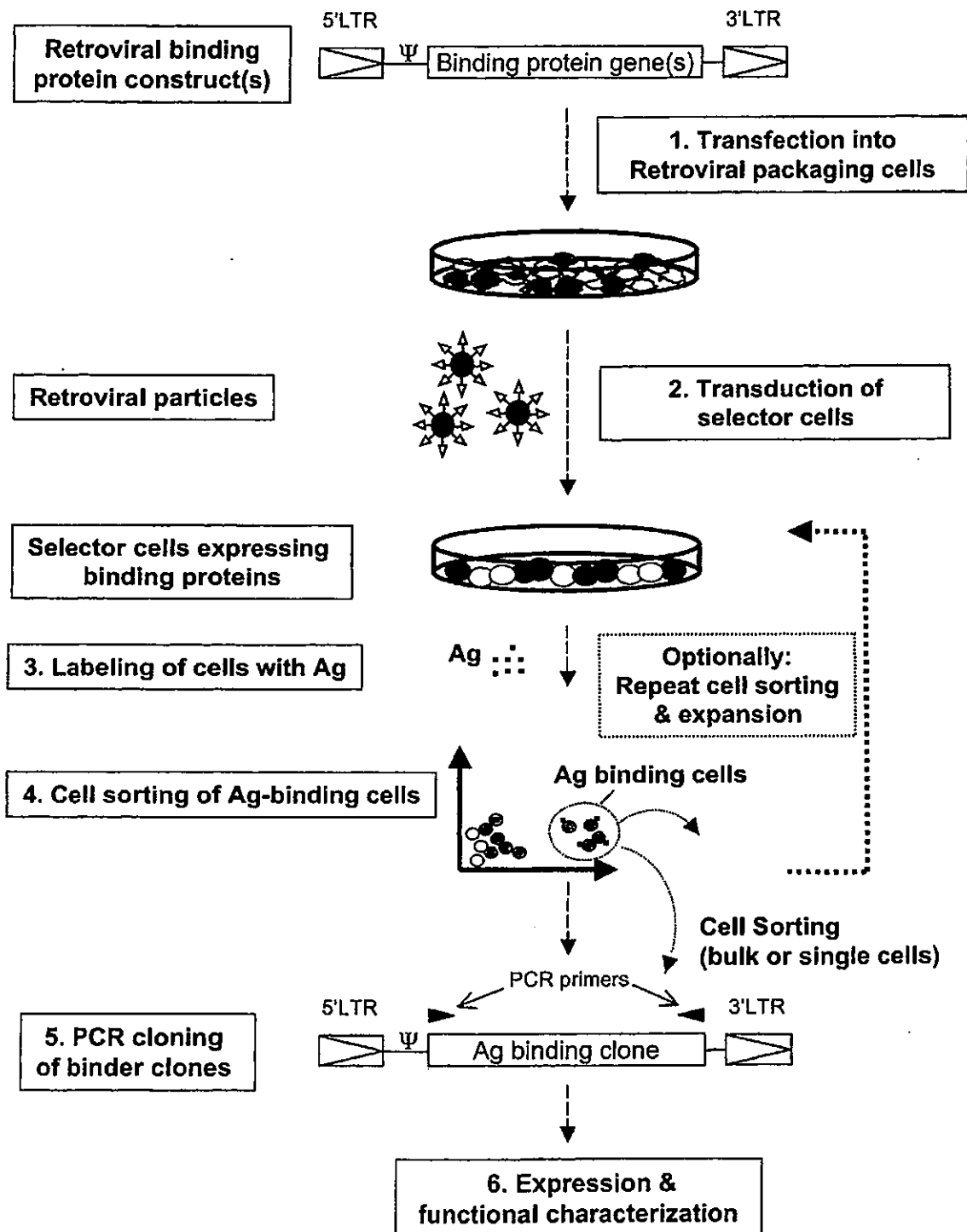


Fig. 2a)

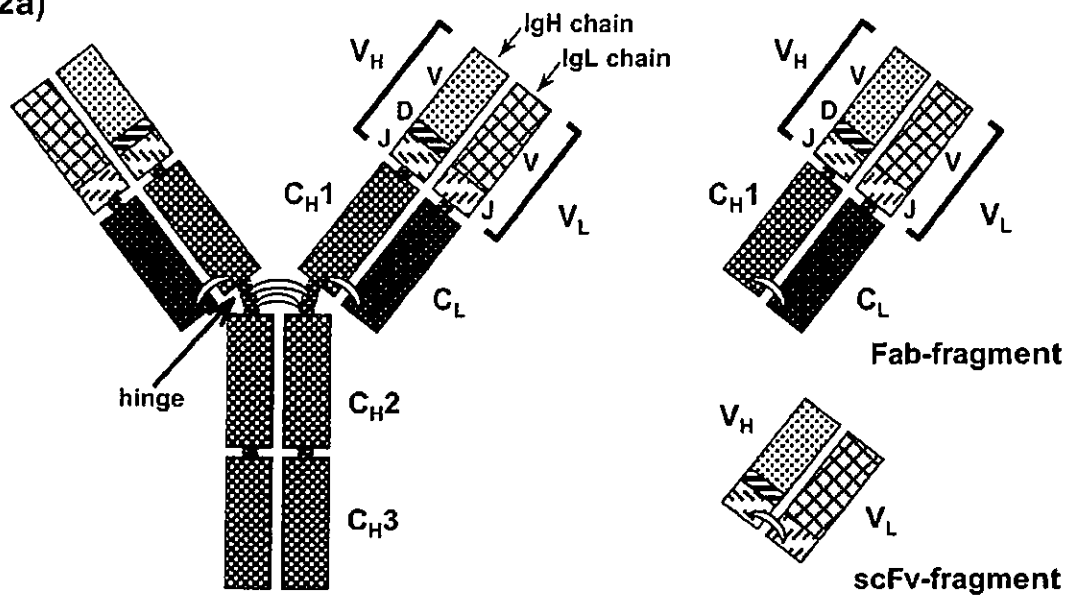


Fig. 2b)

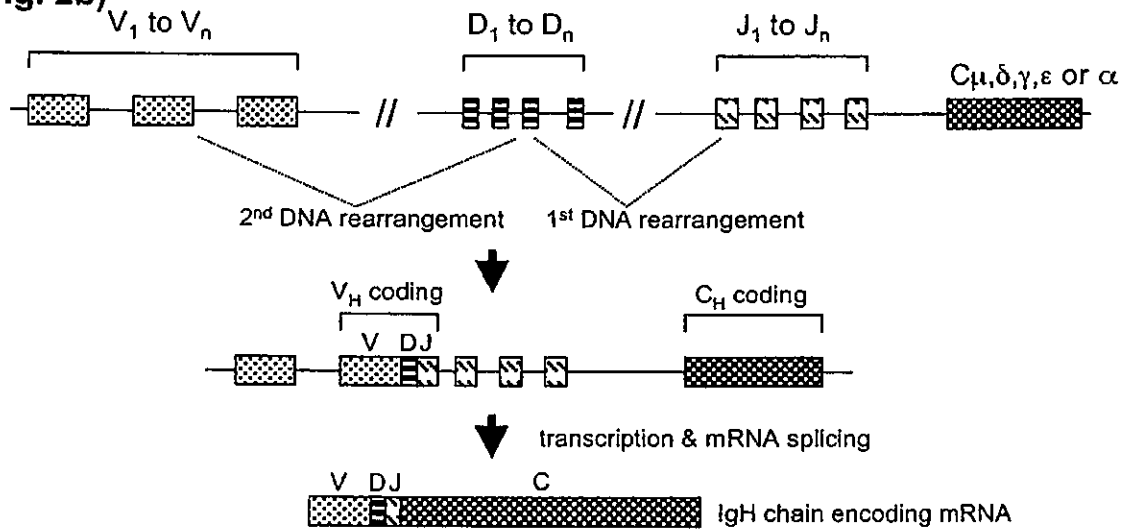


Fig. 2c)

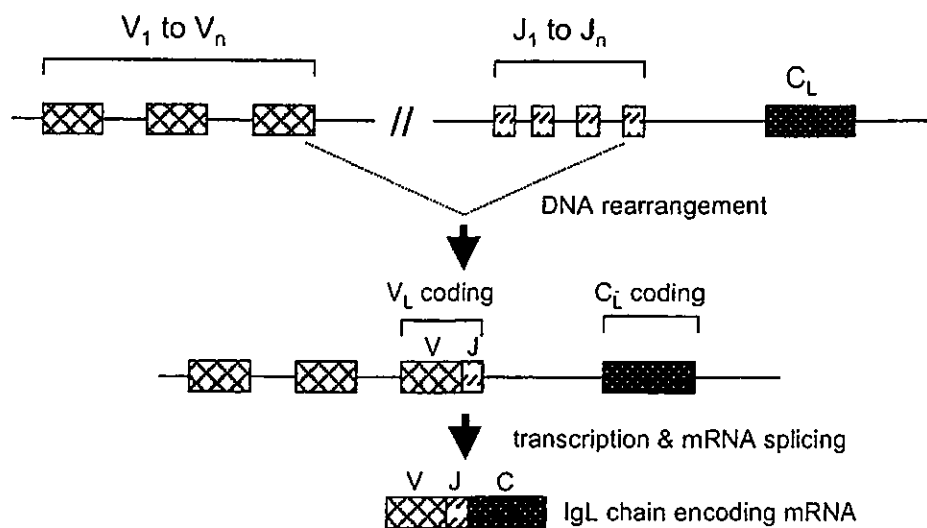


Fig. 3a)

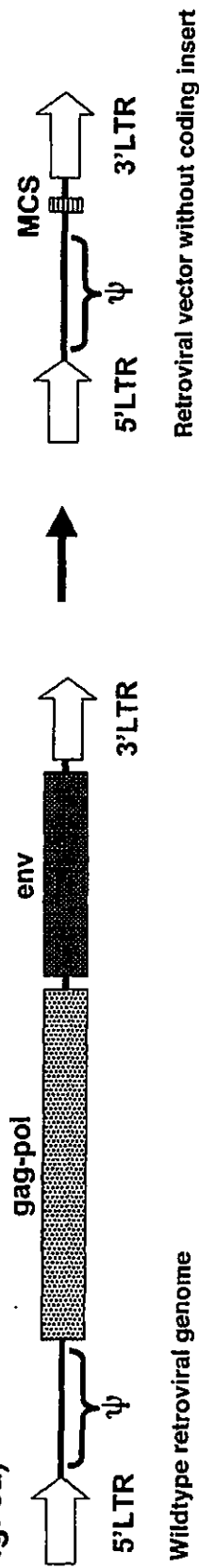


Fig. 3b)

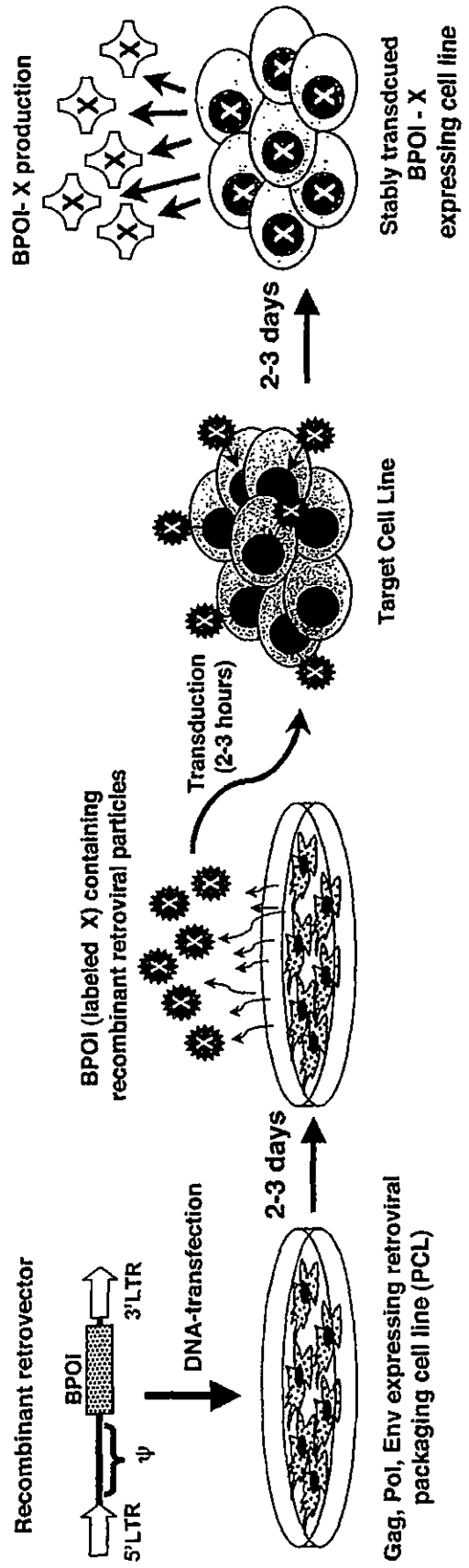


Fig. 4a+b

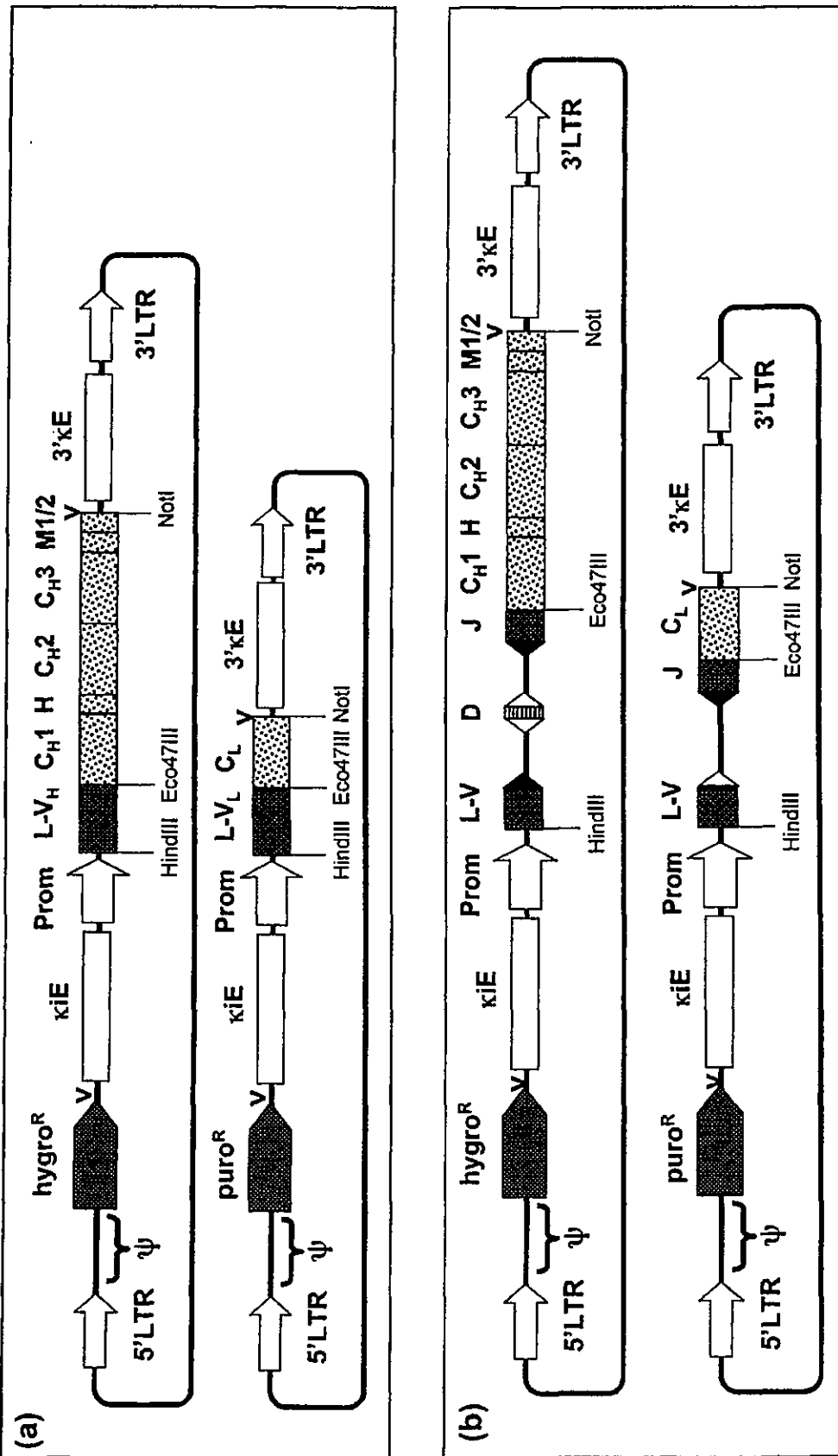
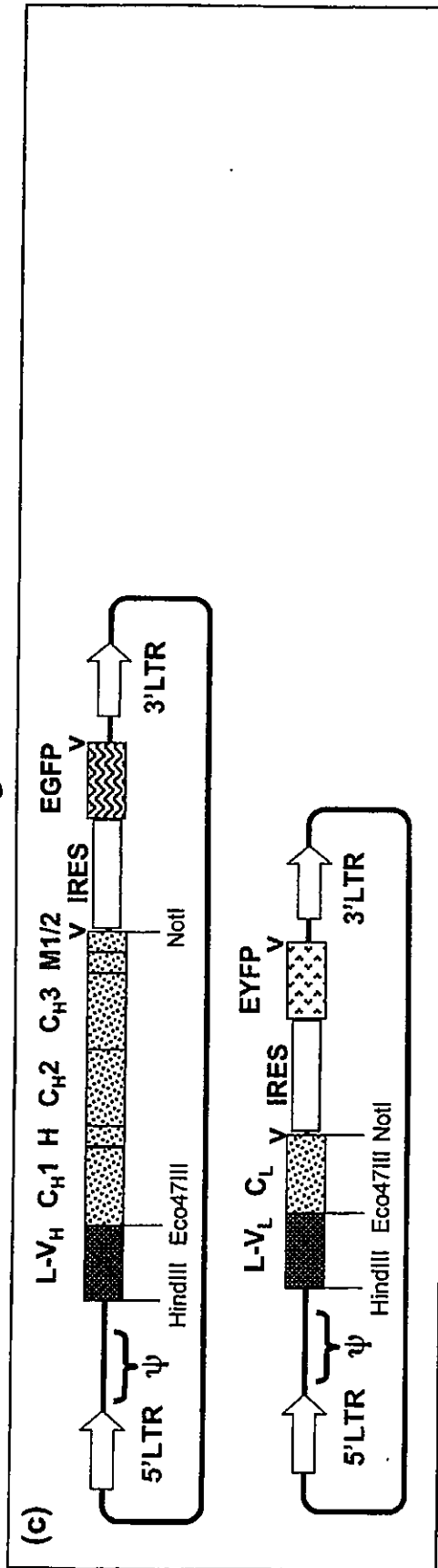


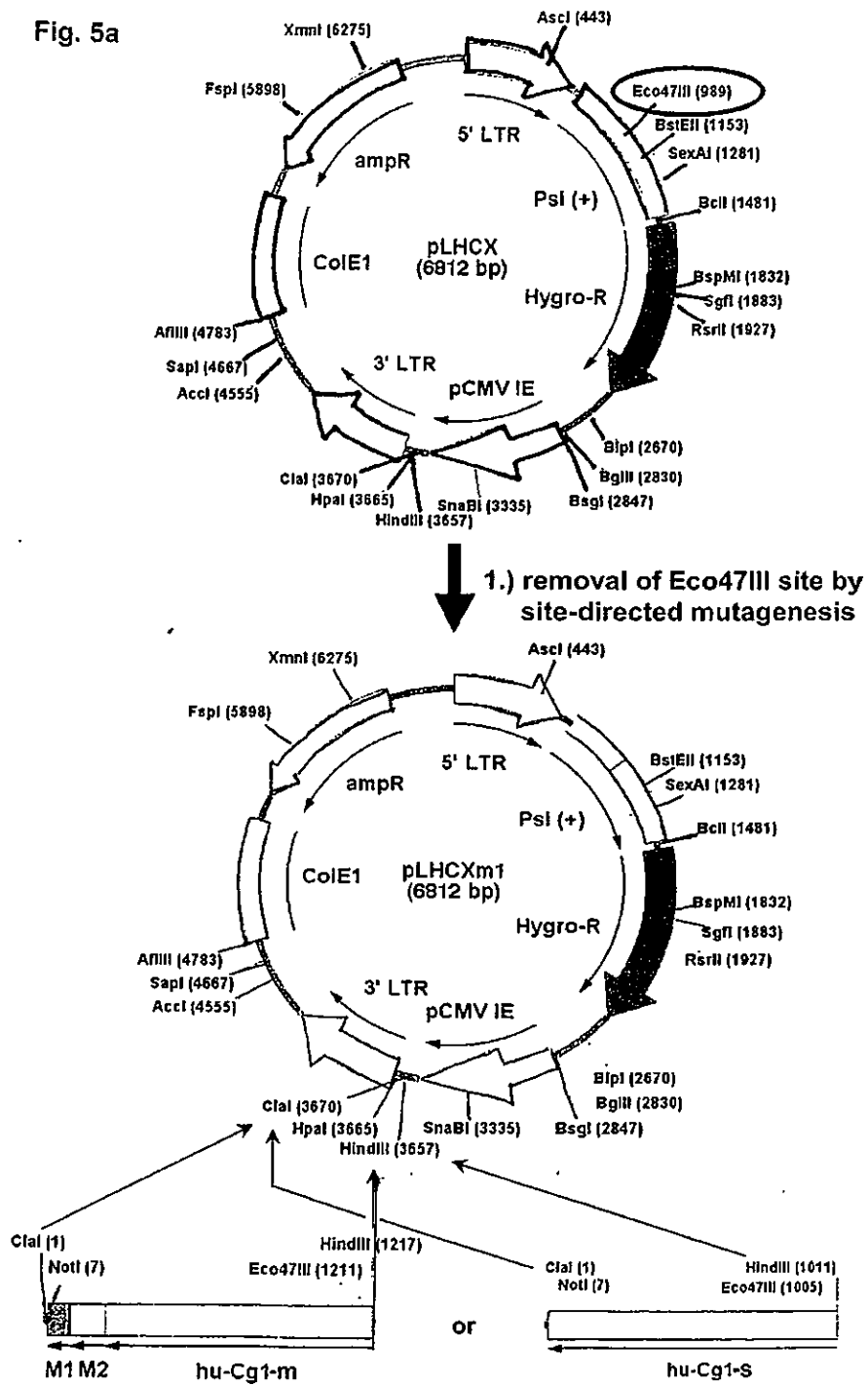
Fig. 4c

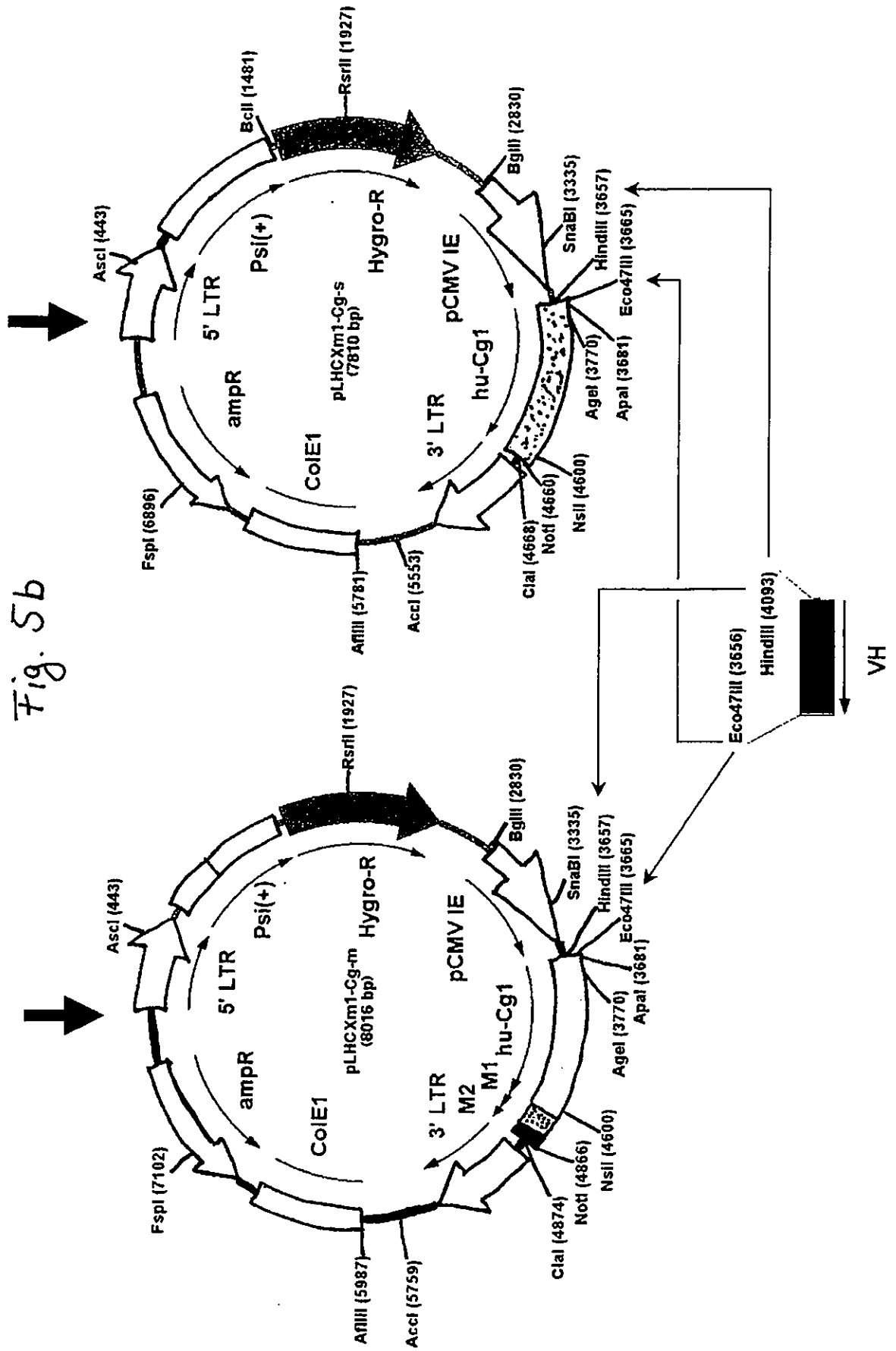


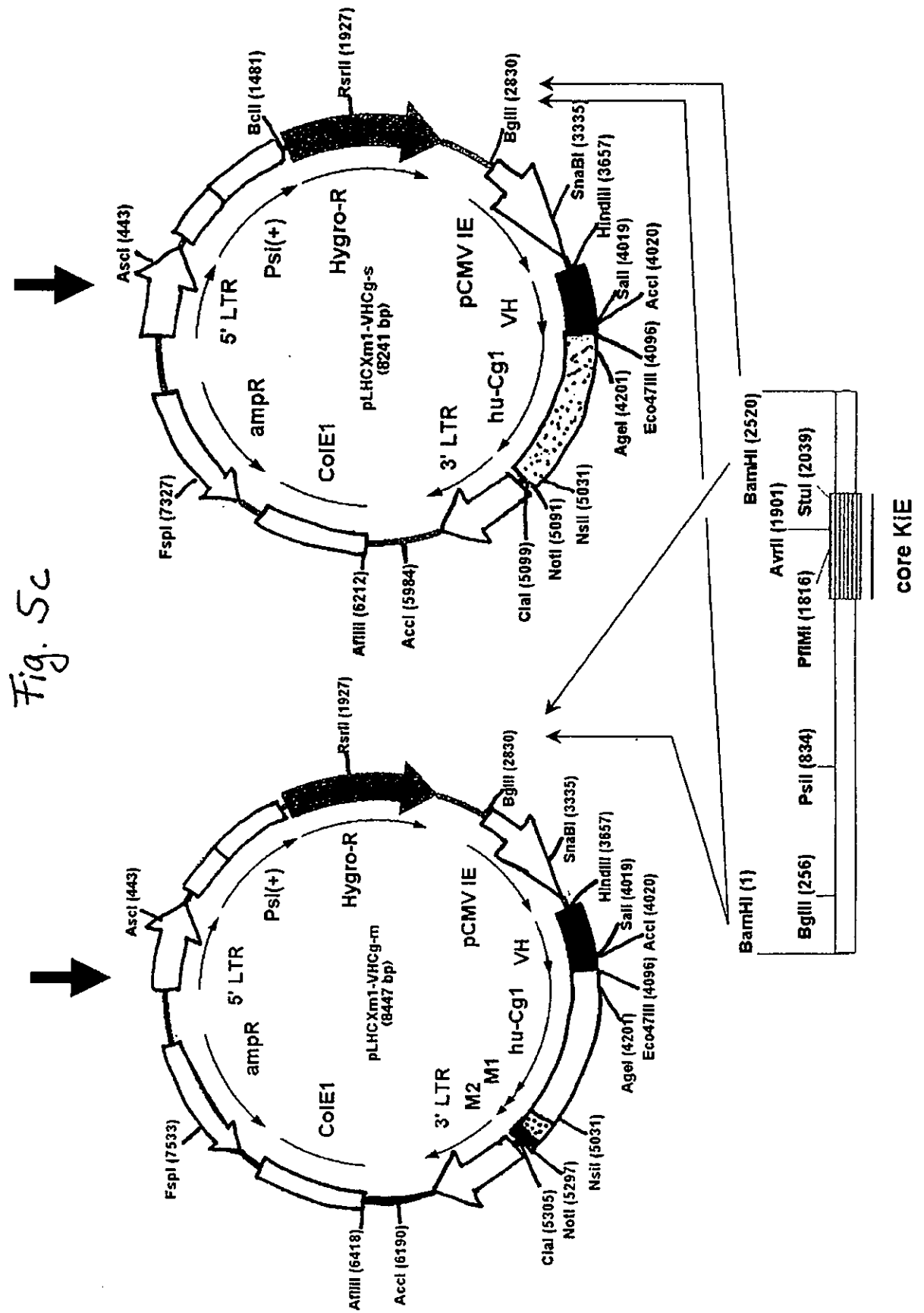
► and ◄ : 23 basepair recombination signal sequences (RSSs) ▷ and ◄ : 12 basepair RSSs

LTR : long terminal repeat ψ : packaging signal v : stop codon

Fig. 5a







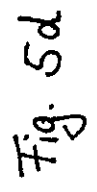
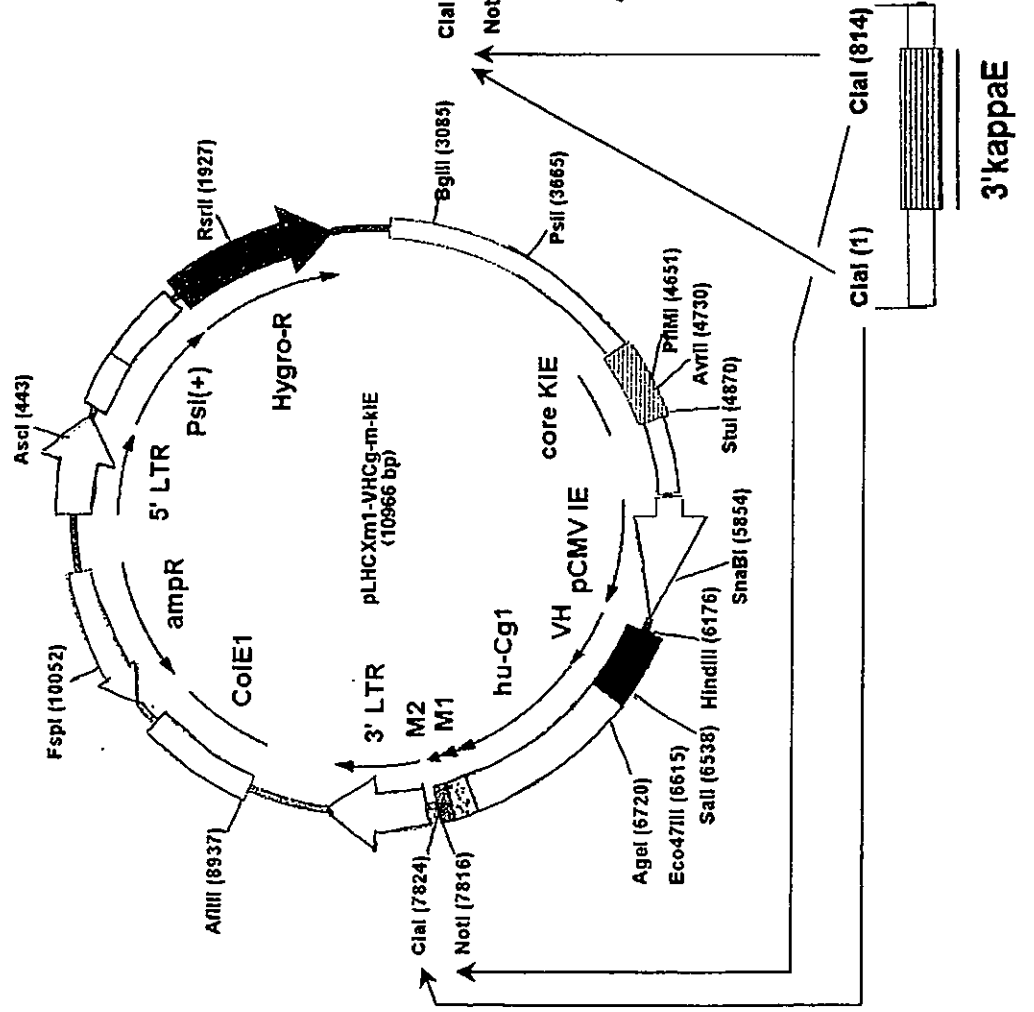
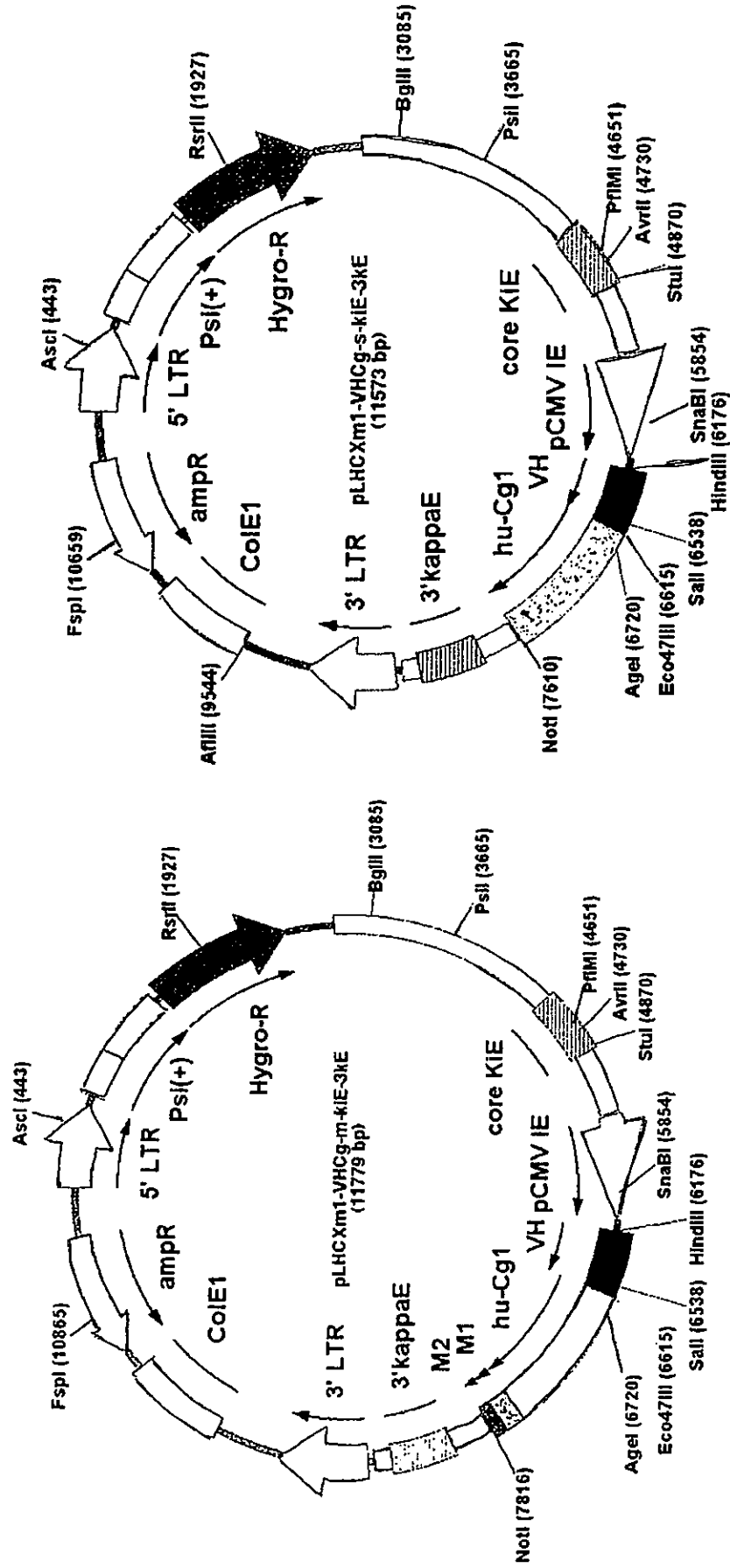


Fig. 5e



Retroviral expression construct for membrane bound $\gamma 1H$

Retroviral expression construct for secreted $\gamma 1H$ chains

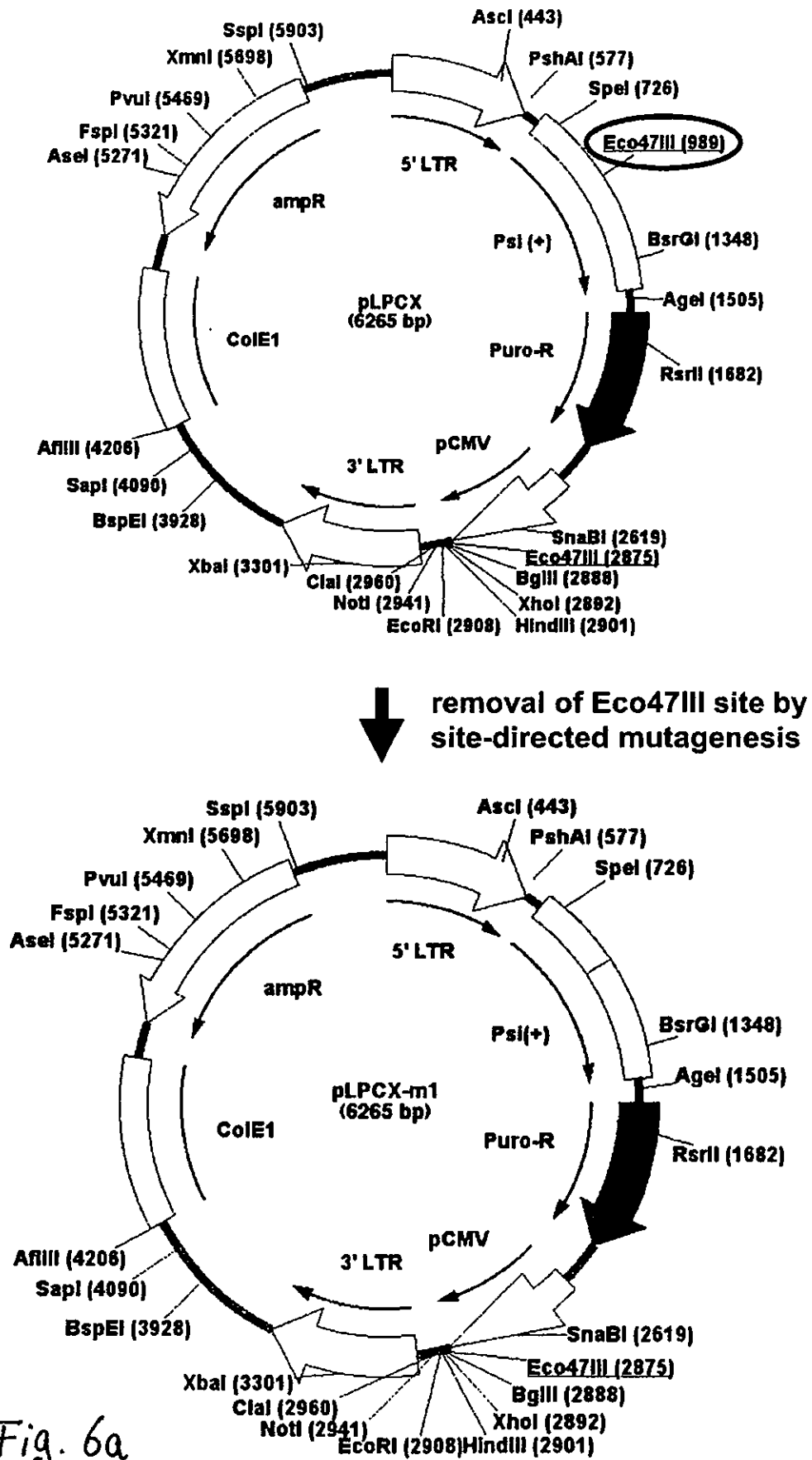
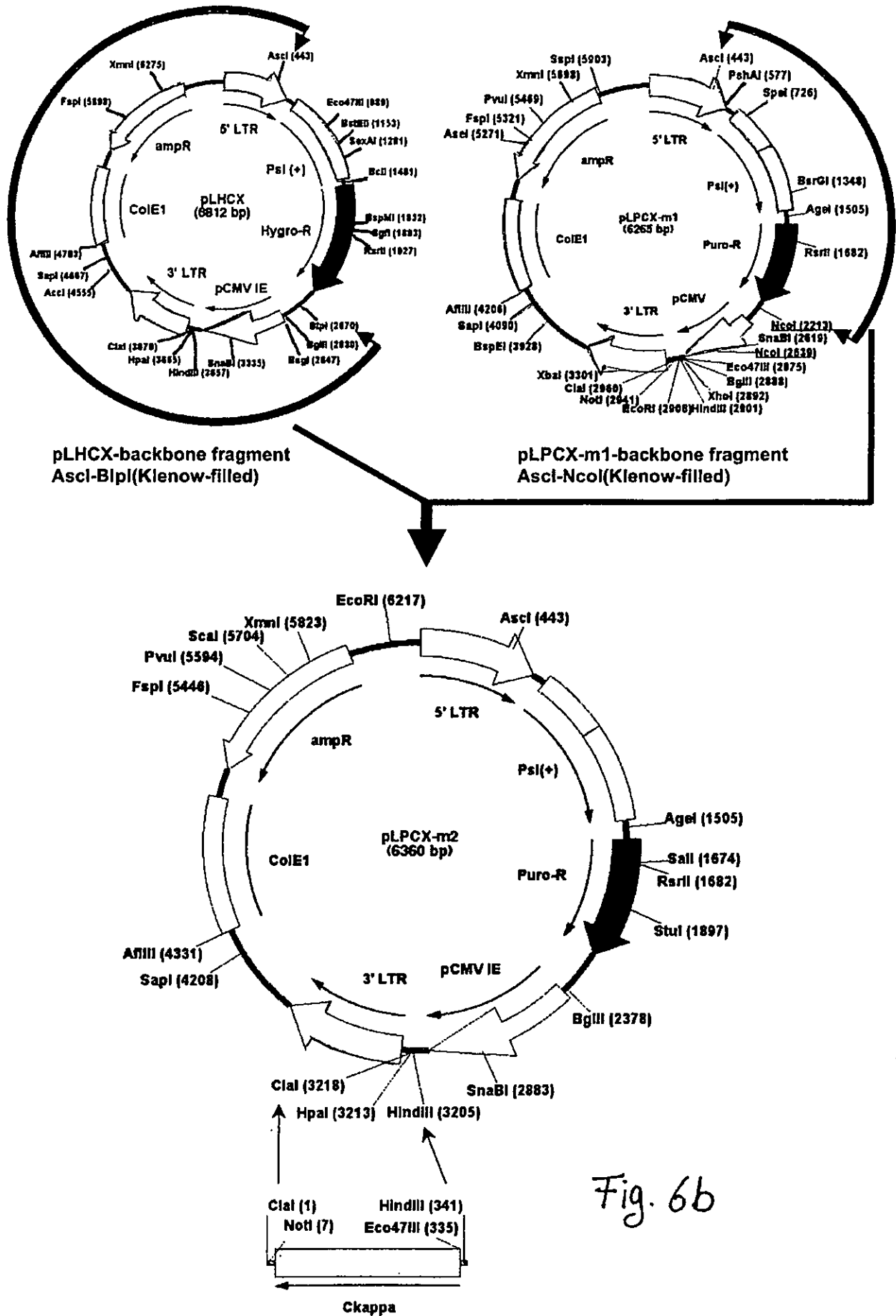


Fig. 6a



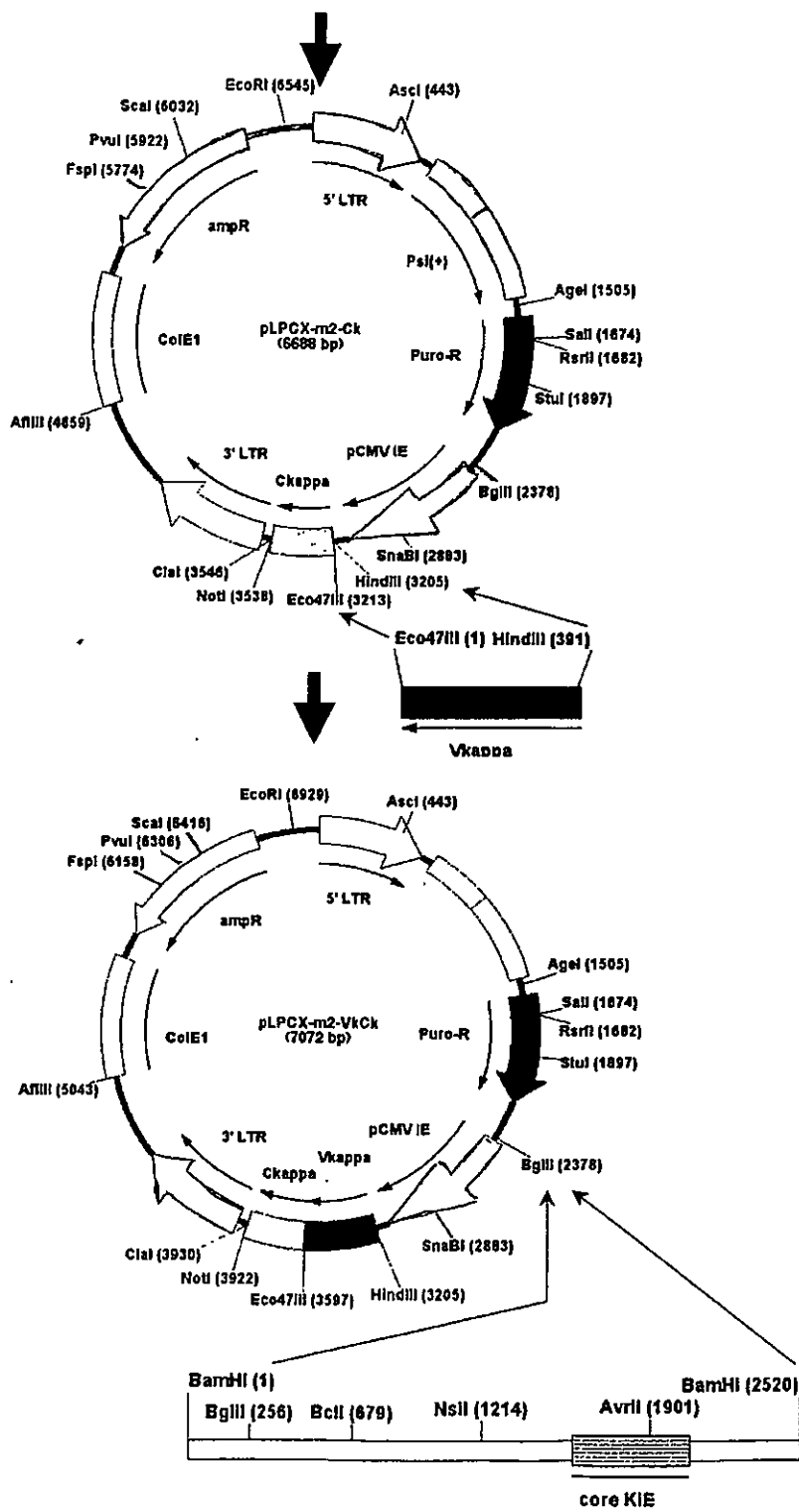


Fig. 6c

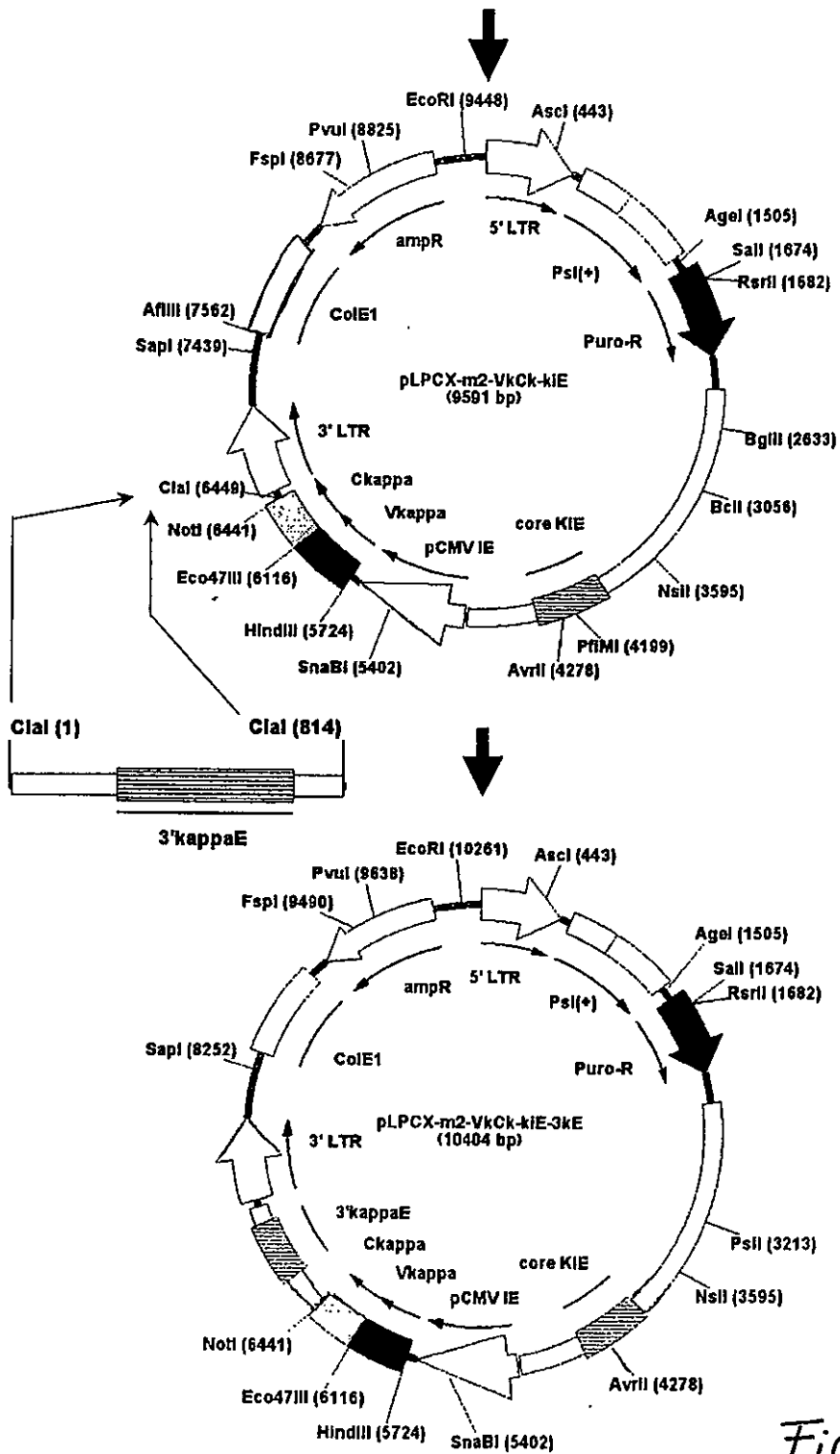


Fig. 6d

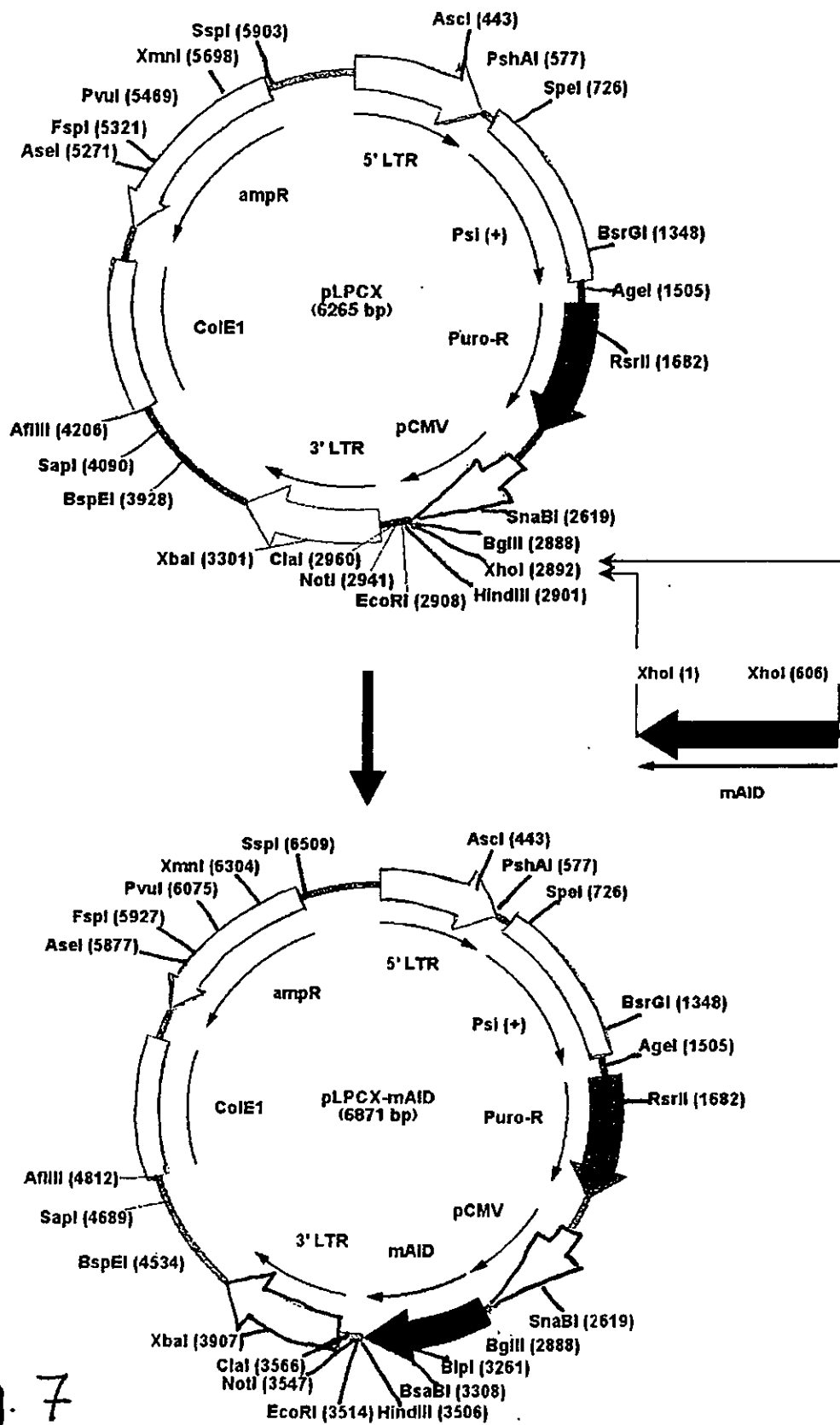
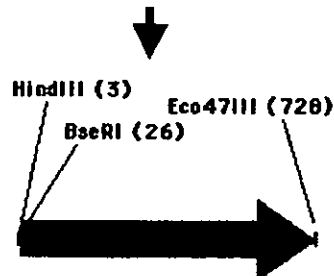
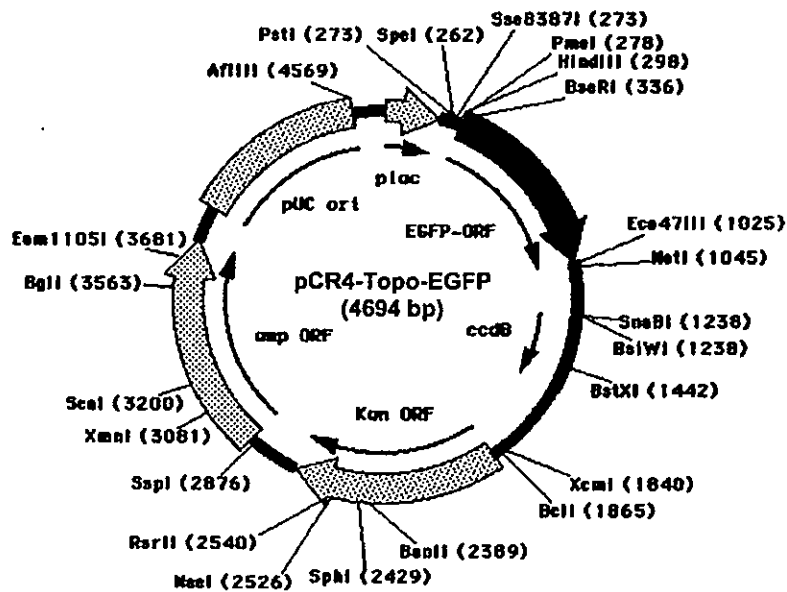


Fig. 7

- 1.) PCR amplification of EGFP open reading frame using
template: pIRES-EGFP + PCR primers: Seq-ID 17 & Seq-ID 18



- 2.) ligation of PCR product into pCR4-Topo (Invitrogen), sequence verification



- 3.) site-directed mutagenesis to introduce stop mutation and SpeI site at aa 107/108 of EGFP-ORF (see Fig. 10 a. & b.)

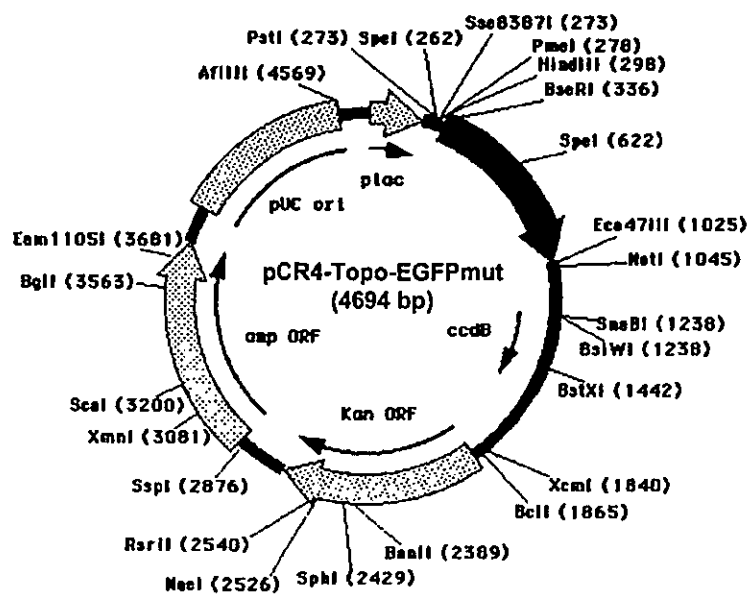


Fig. 8a

Fig. 8b

4.) Ligation of mutated EGFP ORF
as HindIII/Eco47III fragments from
pCR4-Topo EGFPmut into
pLHCXm1-VHC γ -s and into
pLHCXm1-VHC γ -s- κ E-3'kE

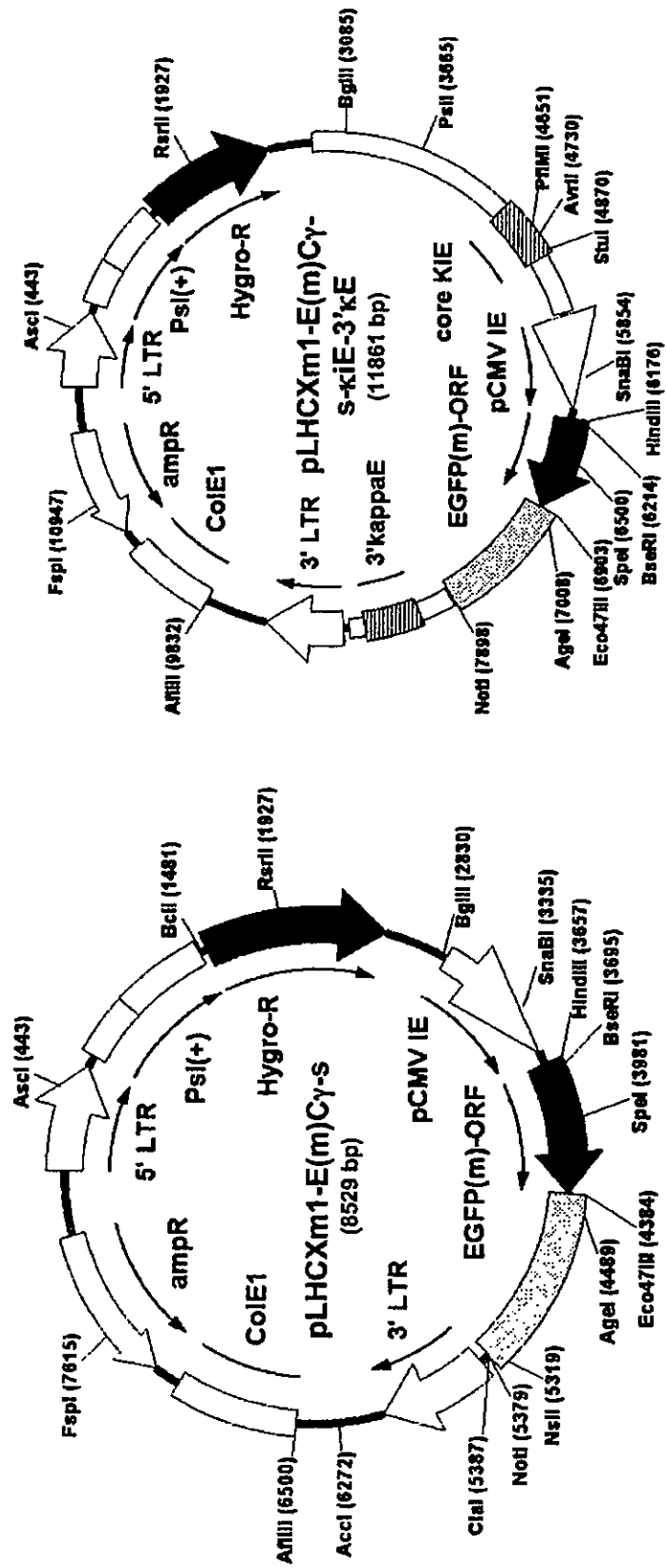


Fig. 9 a)

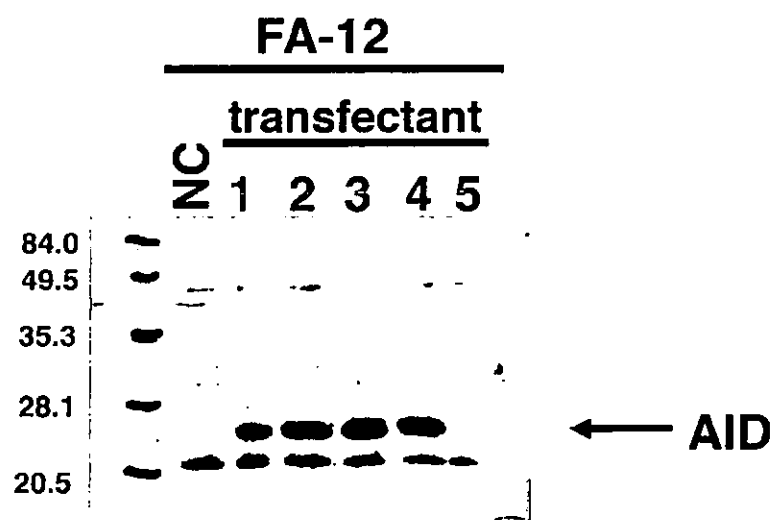


Fig. 9 b)

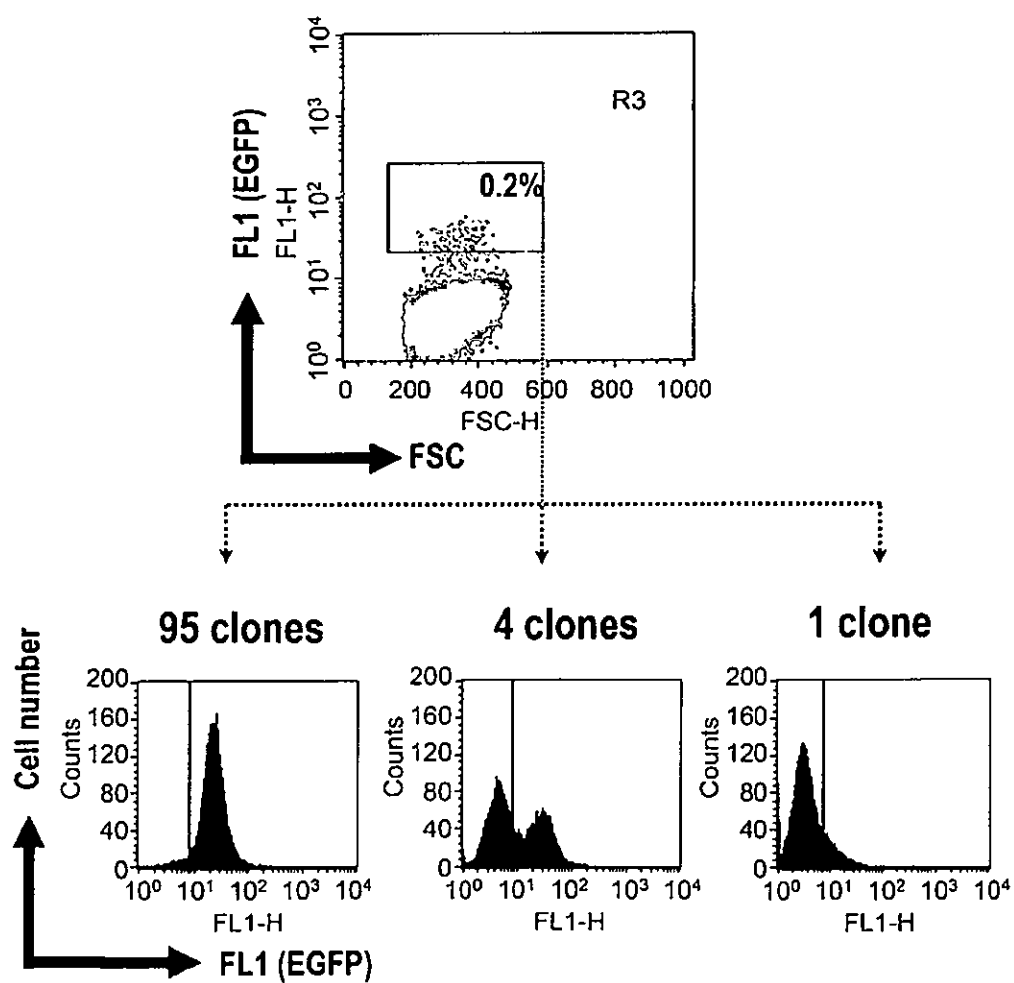


Fig. 10

a)

nt 313-> .. GGC AAC TAC AAG ACC CGC .. nt 313-> .. GGC AAC TAG tat ACC CGC ..
 aa 105-> G N Y K T R aa 105-> G N • Y T R

wt EGFP sequence  mutated EGFP sequence

Spel

b)

1 ATGGTGAACAGGGCGAGGAGCTGTTACCCGGGGTGGTCCCATCTCGTGAAGCTGACGGGACGTAAACGGCCACAGGTTCAAGCTGTCCGGCGAGG
 TACCACTGGTTCCCGCTCCTCGACACAGTGGCCCGACCGGTAGGACACAGCTGACCCCTGCATTTGCCGGTGTTCARAGTCCACAGGCCCGCTCC
 1▶ M V S K G E E L F T G V Y P I L V E L D G D V N G H K F S V S G E

101 CGAGGGCGATGCCACCTACGGCAGCTGACCTGAGGTTTCATCTGACCCACCGGACAGCTGCCCTGGCCACCCCTCGTACACACCCCTGACCTA
 CGCTCCCGCTACGGTGGATGCCGTTCCGACTGGGACTTCAGGTAGACGTGGTGGCCGTTTCGACGGGACCGGGTGGGAGGACCTGGTGGGACTGGAT
 34▶ G E G D A T Y G K L T L K F I C T T G K L P V P W P T L V T T L T Y

201 CGGCGTGCGGTGCTTCAGCCGCTACCCCGACACATGAGGACACGACTTCTTCAGTCCGCGATGCCCGGAGGCTACGTCAGGAGGCGGACCATCTTC
 GCCGACGTCACGAGTCCGGGATGGGCTGGTGTACTTCGTCGCTGACGAGGTTCAAGGCGGTACGGGCTTCGATGAGGTTCTCGCGTGGTAGAG
 67▶ G V Q C F S R Y P D H M K Q H D F F K S A M P E G Y V Q E R T I F

Spel (316)

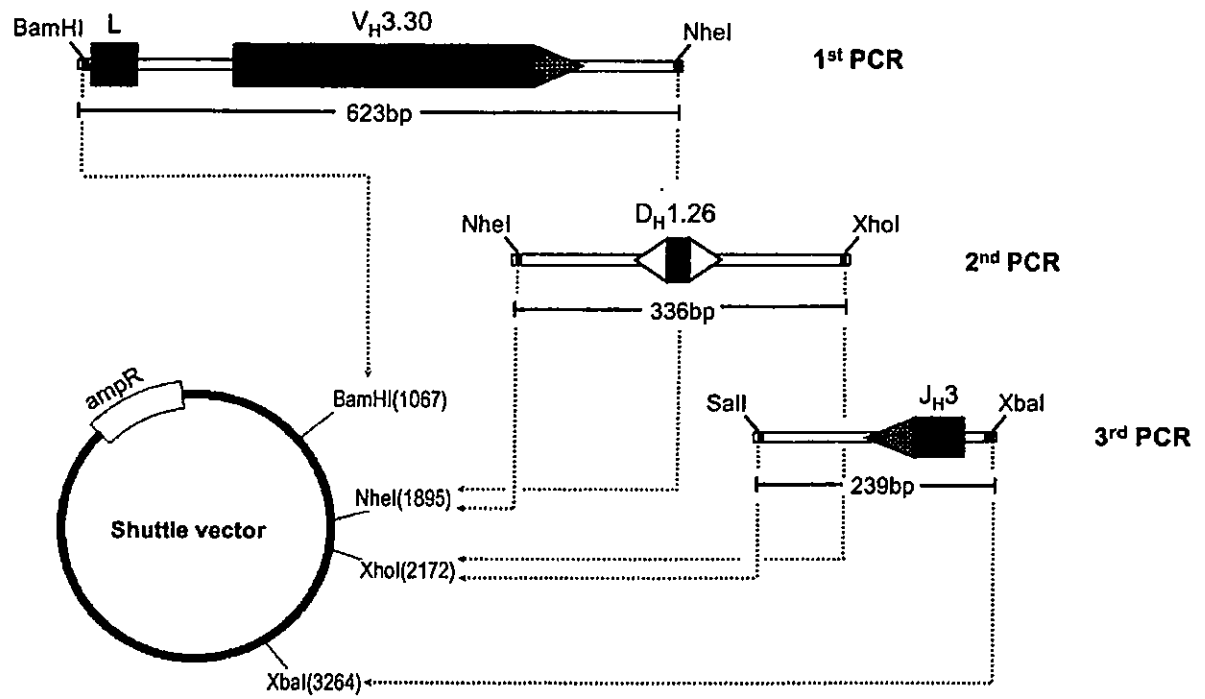
301 TTCAGGGACGACGGGACACTAGtatatACCCCGCGCGCTCGAGGTCGAGGTCGACCGGACCTGGTGAACCGCATCGAGCTGAAGGGCATCGACTTCAGGGAG
 AAGTTCCCTGCTGCCGTTGATcatatAGGCGCGCTCCACTTCAGGCTCCGCTGTGGGACCACTTGGCGTAGCTCGACTTCCCGTAGCTGAAGTTCCCTC
 101▶ F K D D G N • Y T R A E Y K F E G D T L V N R I E L K G I D F K E

400 GACGGCAGCATCTGGGGCAGCAGCTGGAGTACACTACACAGCCACAGGCTCTATATCATGSCCGACAGACGAGAGACGGCATCAGGTGACATTCA
 CTGCCGTTGTAGGACCCCGTGTTCGACCTCATGTTGATGTTGCGGTTGTGAGATATAGTACCGGCTGTCTCTCTTCCCGTAGTTCACATTGAGGT
 134▶ D G N I L G H K L E Y N Y N S H N V Y I M A D K Q K N G I K V N F

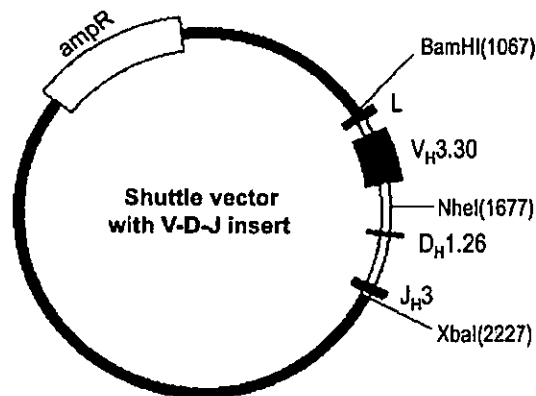
500 AGATCCGACACATCGAGGACGGGAGCGTGACGCTGCCGACCACTACACAGACACACCCCATCGGCGAGGCCCCGCTGCTGCTGCCGACACCA
 TCTAGGCGGTGTTGATGCTCTGCCGTCGACGTCGAGCGGGTGGTGTGCTCTCTGTGGGGTAGCCGCTGCCGGGACACGACGACCGGCTGTTGGT
 167▶ K I R H N I E D G S V Q L A D H Y Q Q N T P I G D G P V L L P D N H

600 CTACCTGAGCACCCGCTCGGCCCTGAGCAGAGACCCGACGAGGAGCGGATGATGCTGCTGAGGTTCTGACCCGCGGATCACTCTCGGC
 GATGAGCTGCTGGGTGAGGCGGGGACTCGTTTCTGGGGTTGCTCTTCGCGCTAGTGTACAGGACGACCTCAGGACCTGGCGGCGGCGCTAGTGAGGCGCG
 200▶ Y L S T Q S A L S K D P N E K R D H M V L L E F V T A A G I T L G

700 ATGGACGAGCTGTACAG
 TACCTGCTCGACATGTT
 234▶ M D E L Y K



Sequential ligation of 1st, 2nd and 3rd PCR product



PCR re-amplification of V-D-J insert with primers Seq-ID29 and Seq-ID30



Fig. Ma

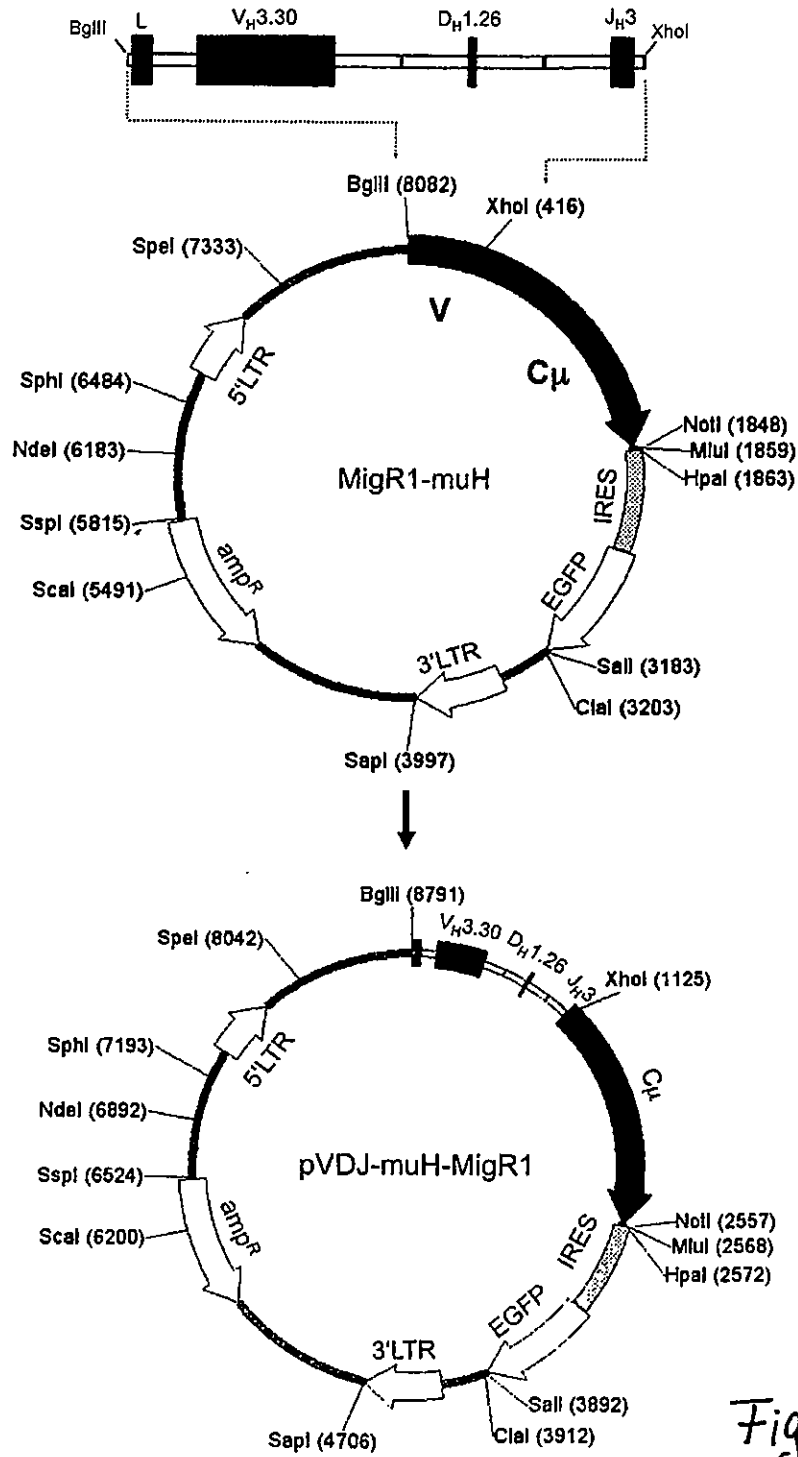
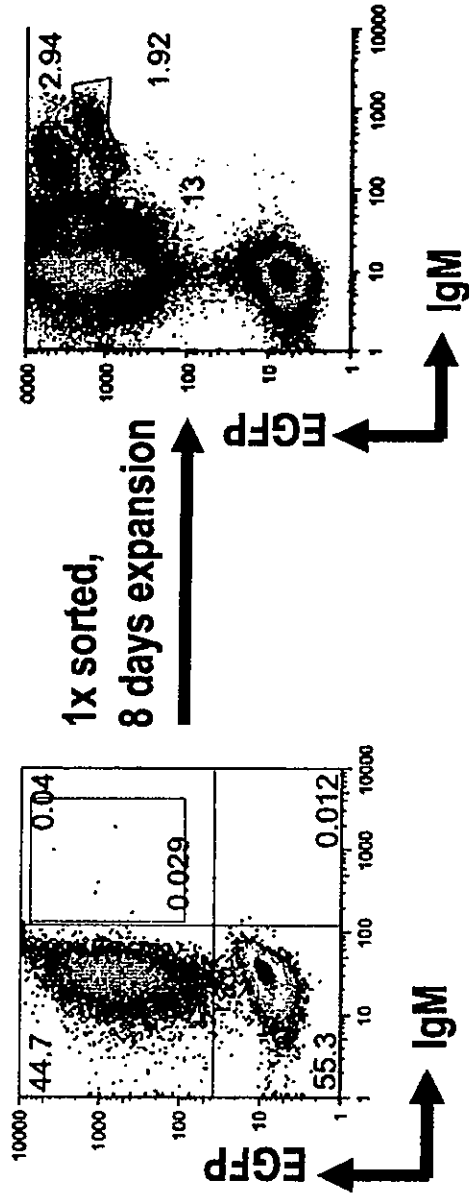


Fig. 12

(a)



(b)

germline	Y Y C A K TAT TAC TGT GCG AAA GA G GTA TAG TGG GAG CTA CTA C..... T GAT GCT TTT GAT ATC TGG	D A F D I W J _H 3
	V _H 3.30	D _H 1.26
clone 225	Y Y C A K D Q TAT TAC TGT GCG AAA GA TCAA N-region	W E L D A F D I W J _H 3
	V _H 3.30	D _H 1.26

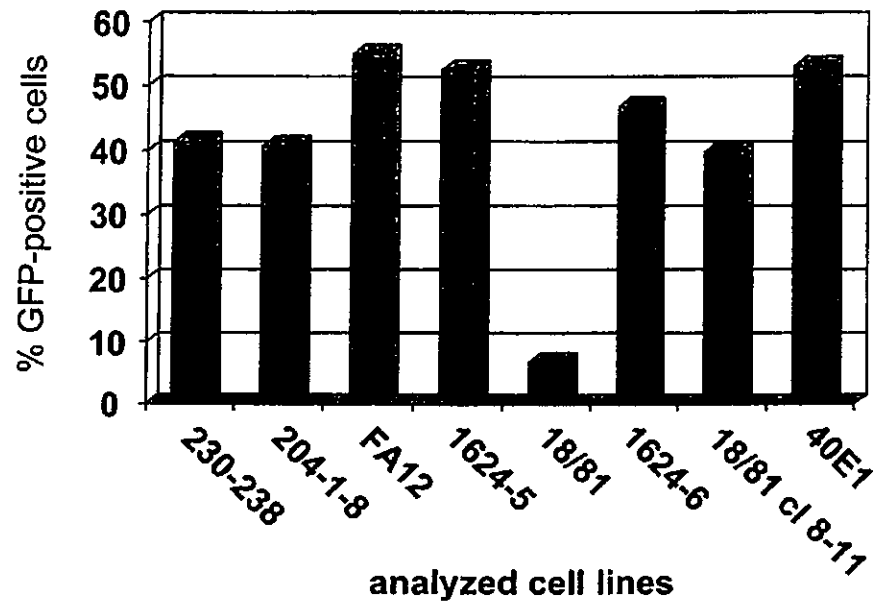


Fig. 13

Fig. 14

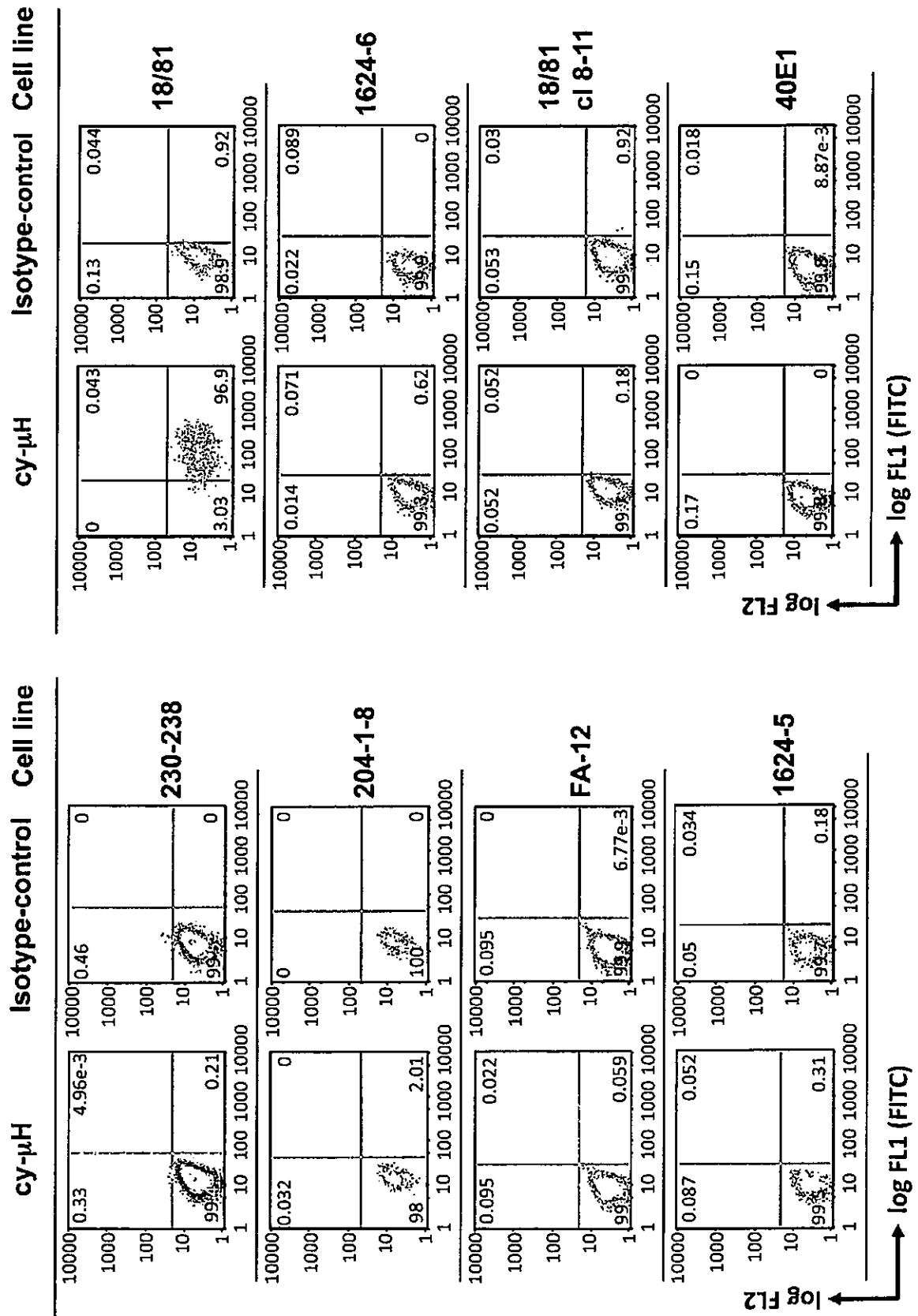


Fig. 15

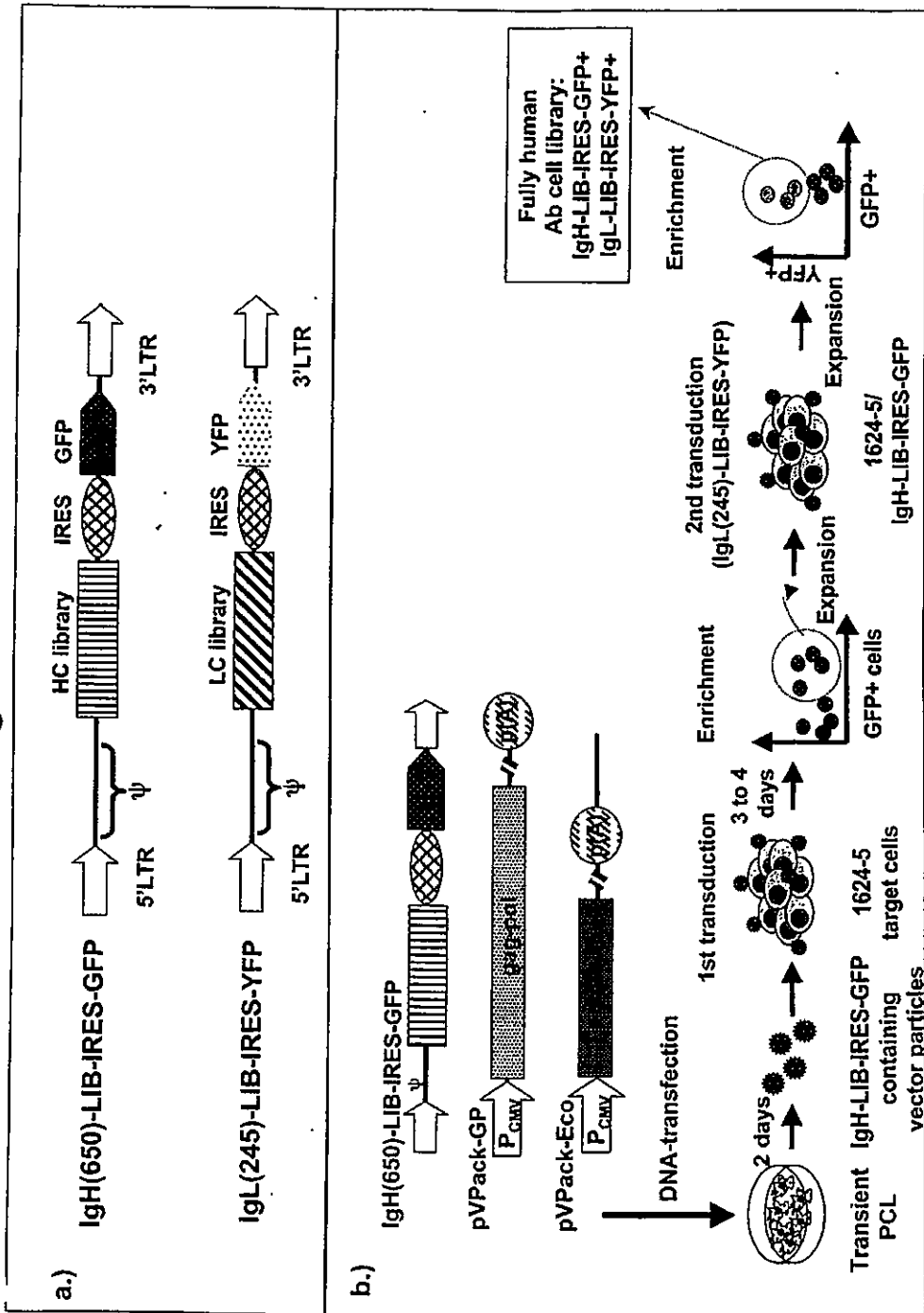


Fig. 16

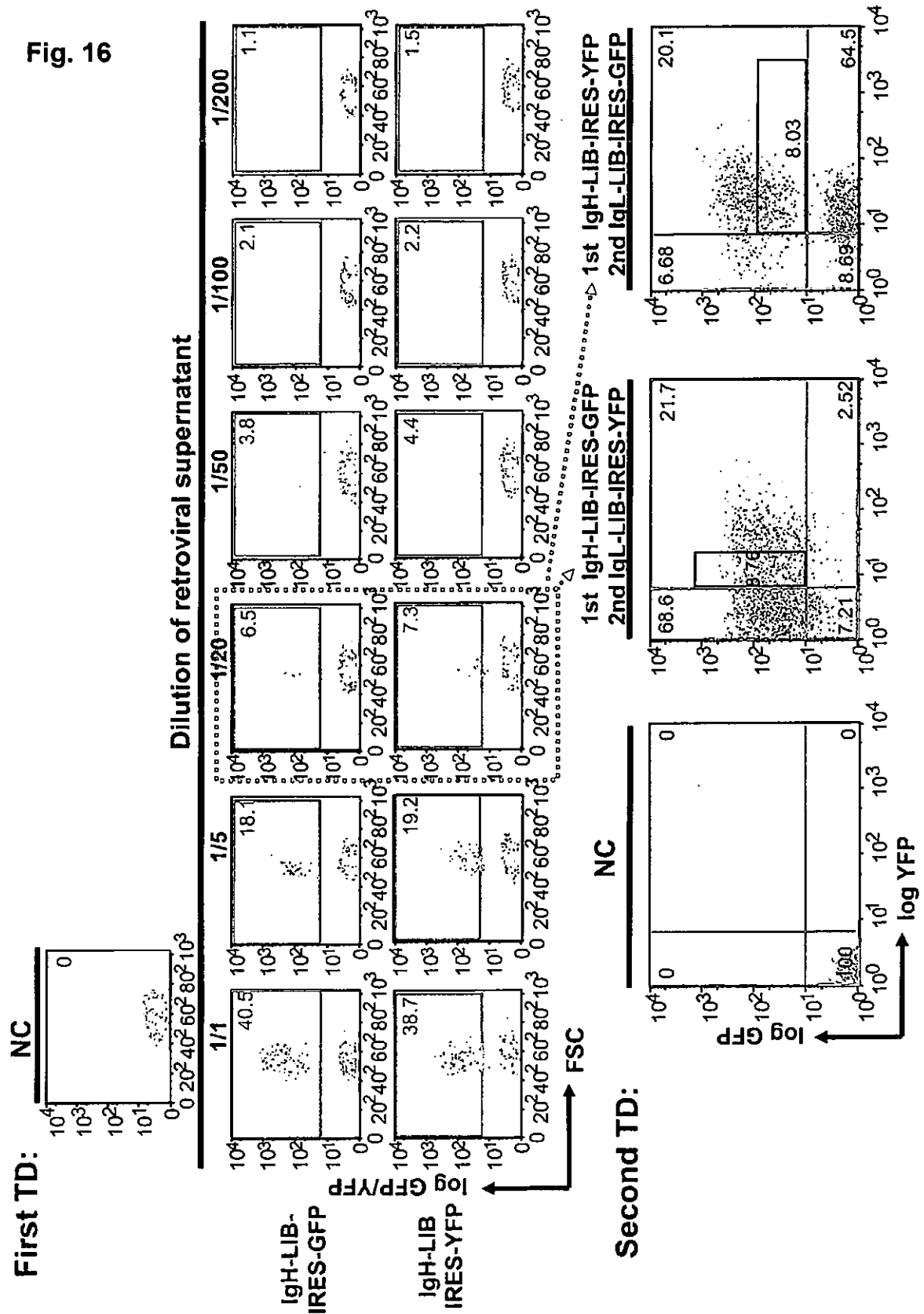


Fig. 17

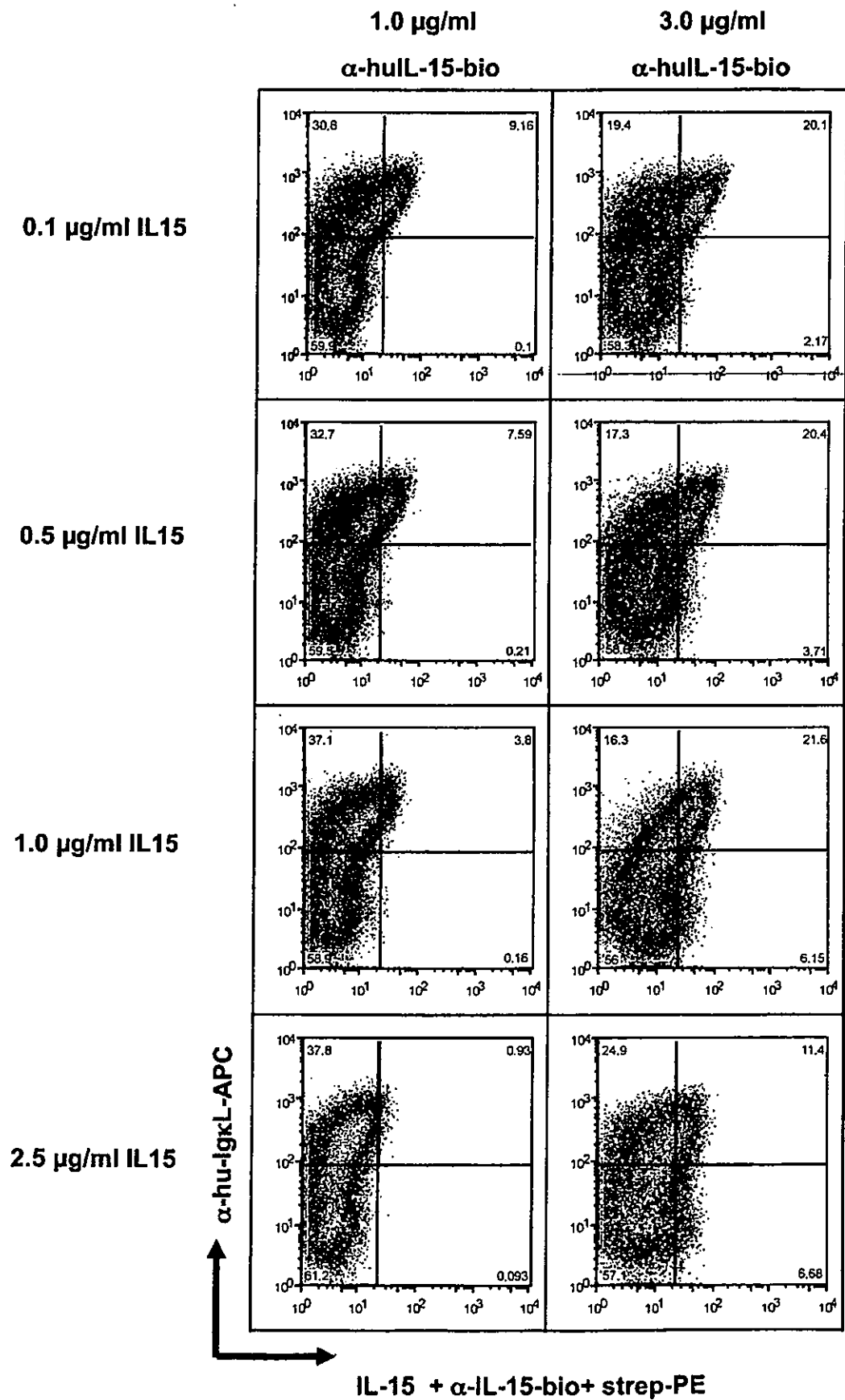


Fig. 18

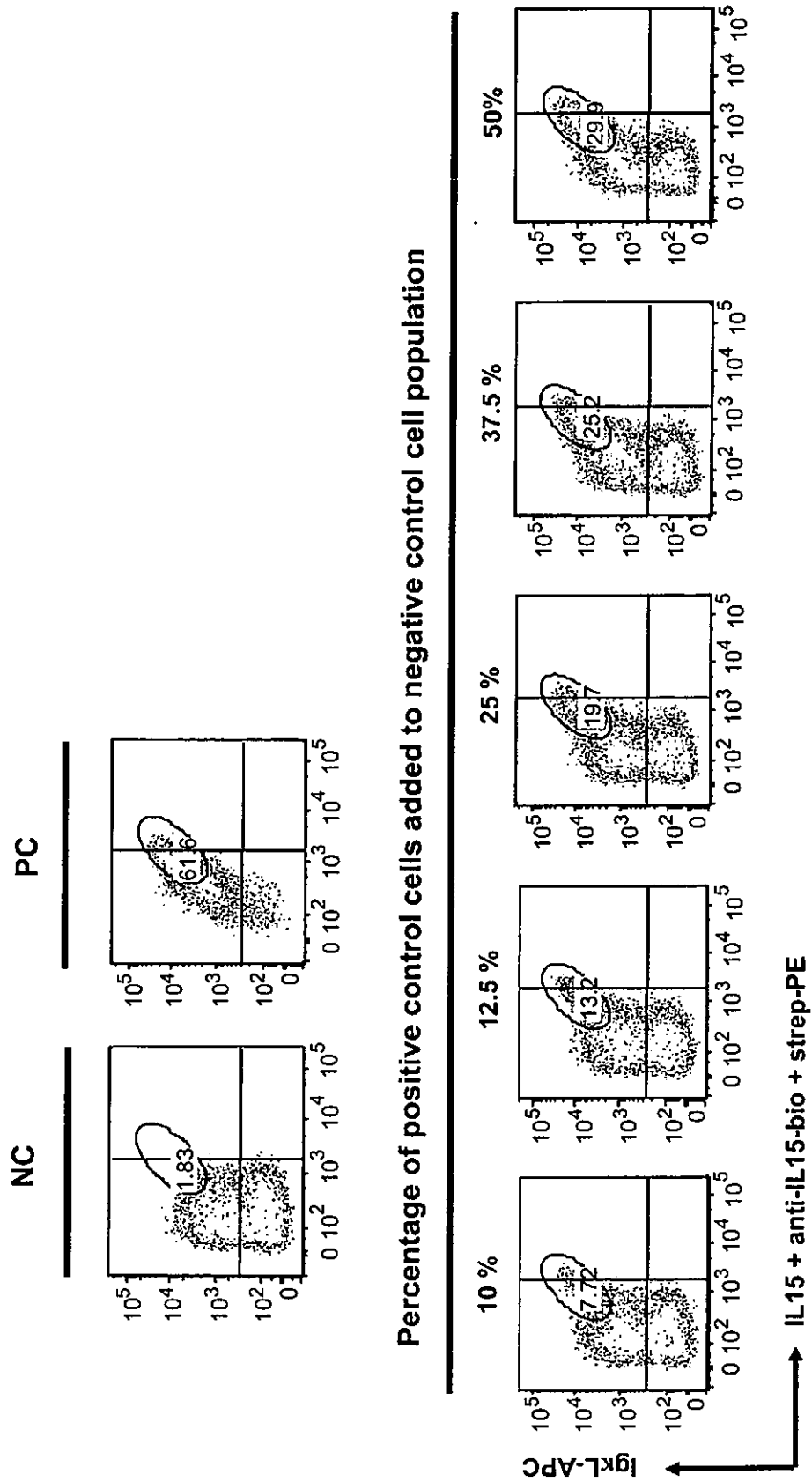
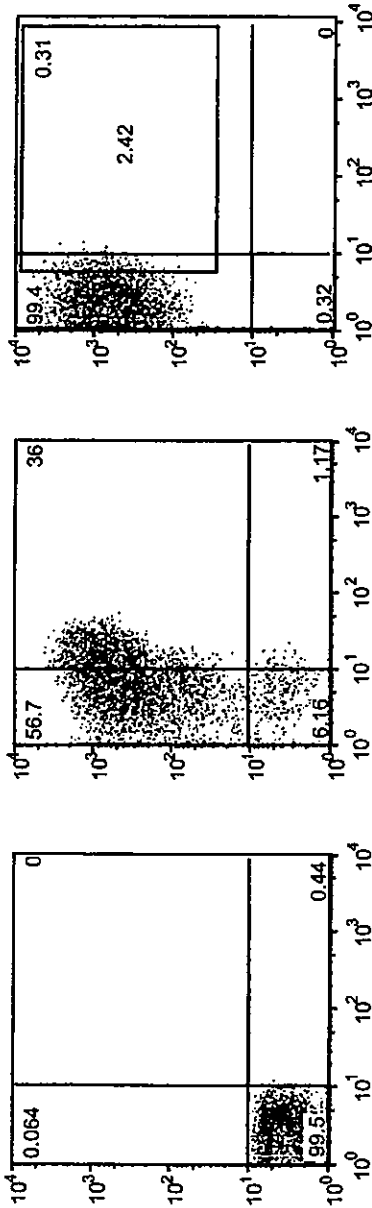


Fig. 19

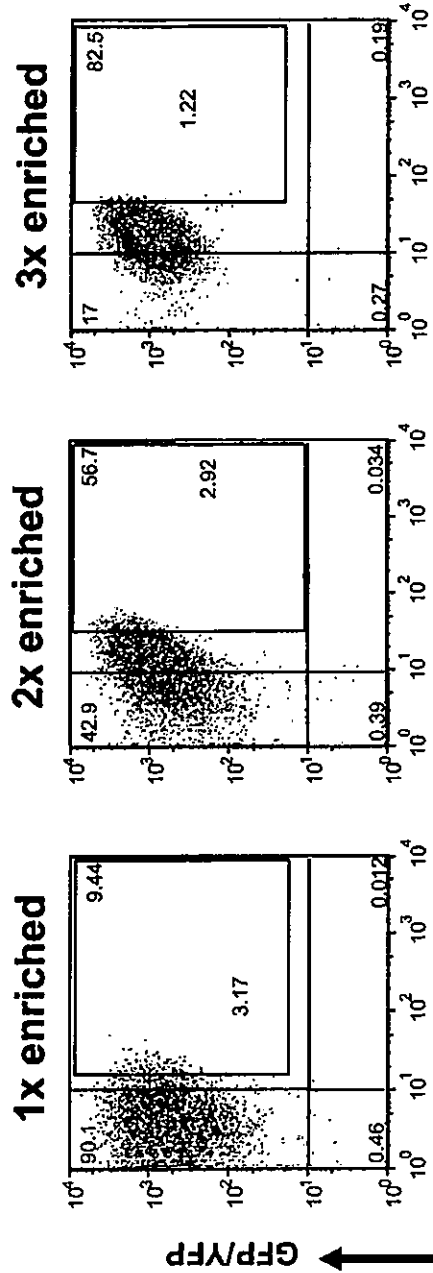
IL15 IgH + IgL-LIB
transduced

IL15 IgH+IgL
transduced
PC

un-transduced
NC



IL15 IgH + IgL-LIB transduced



IL-15 + α-IL-15-biot strep-APC

Fig. 20

IL15 IgH+IgL
transduced

un-transduced

NC

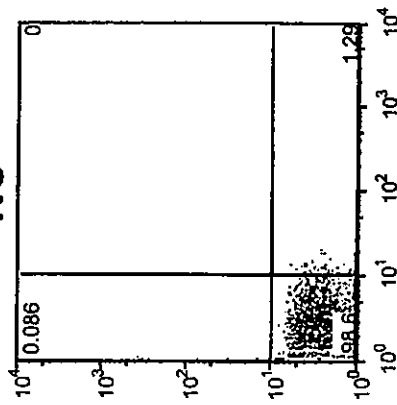
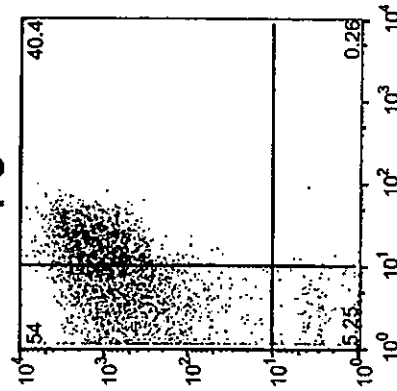
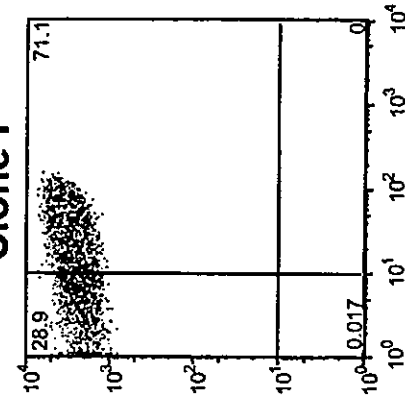
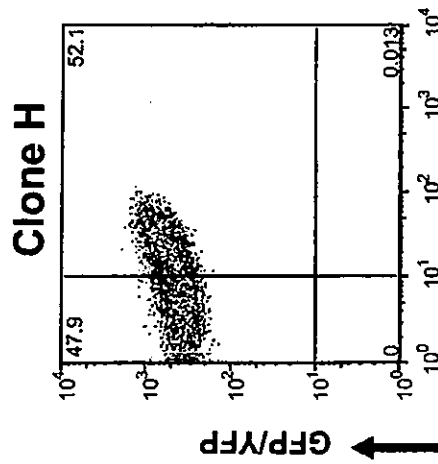
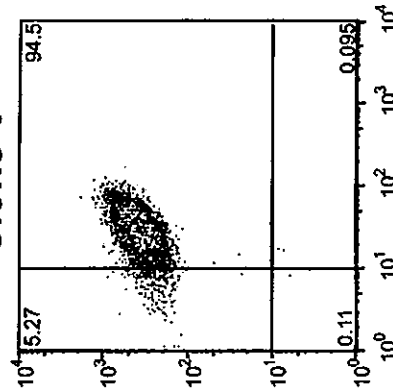
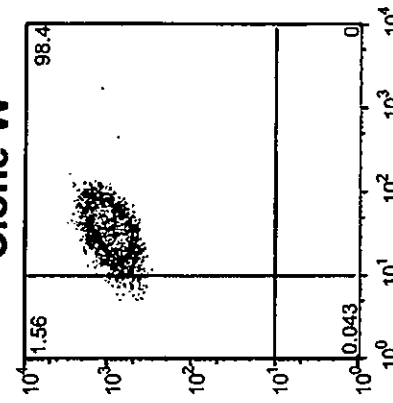
PC

Clone F

Clone H

Clone V

Clone W



GFP/FP

IL-15 + α-IL-15-biot strep-APC

Fig. 21

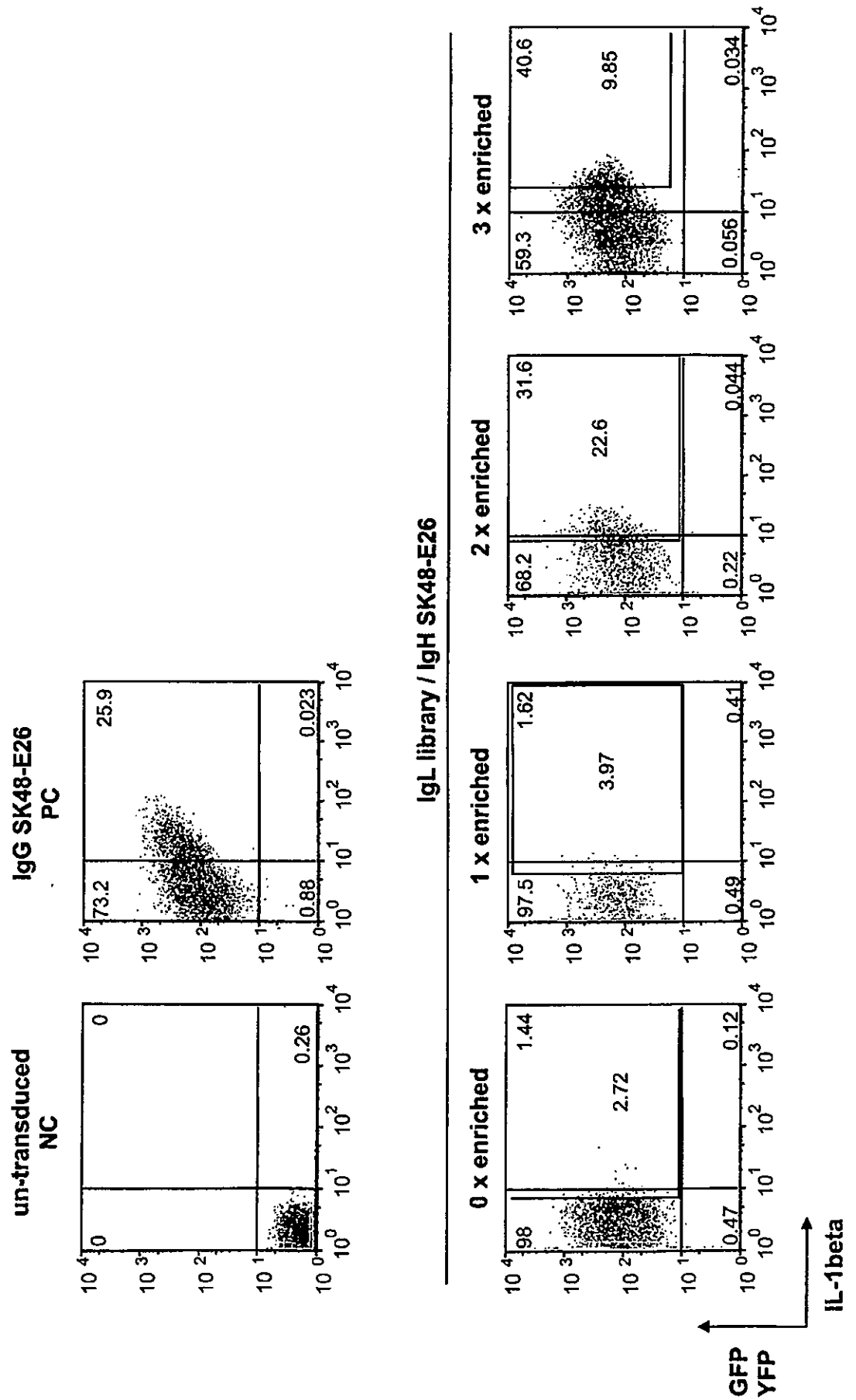


Fig. 22

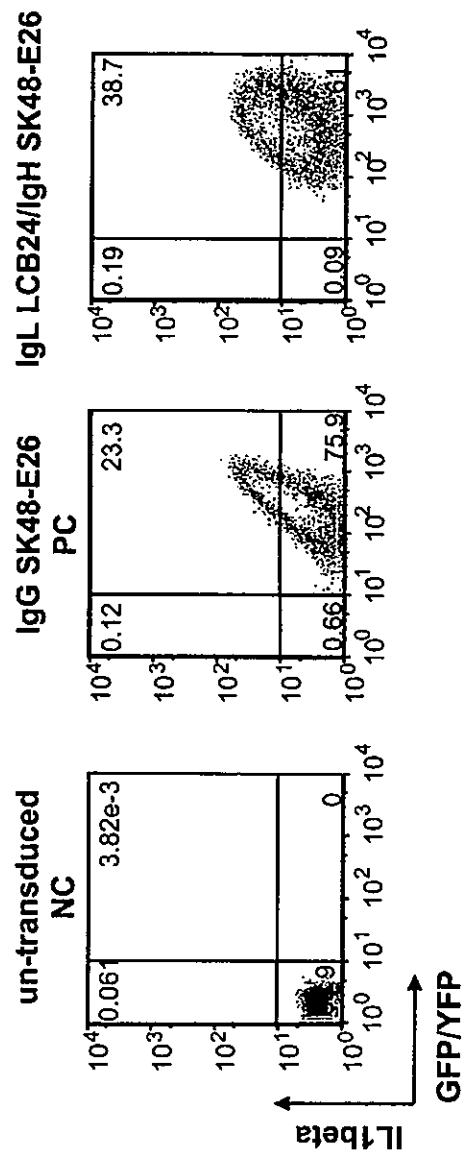


Fig. 23

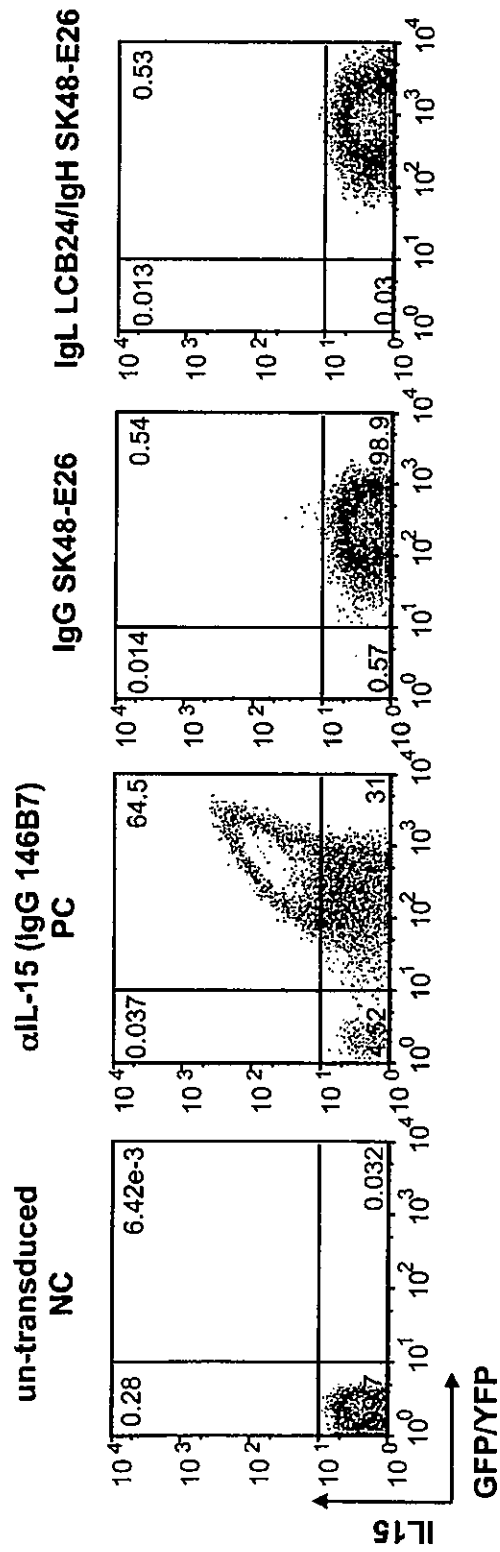


Fig. 24

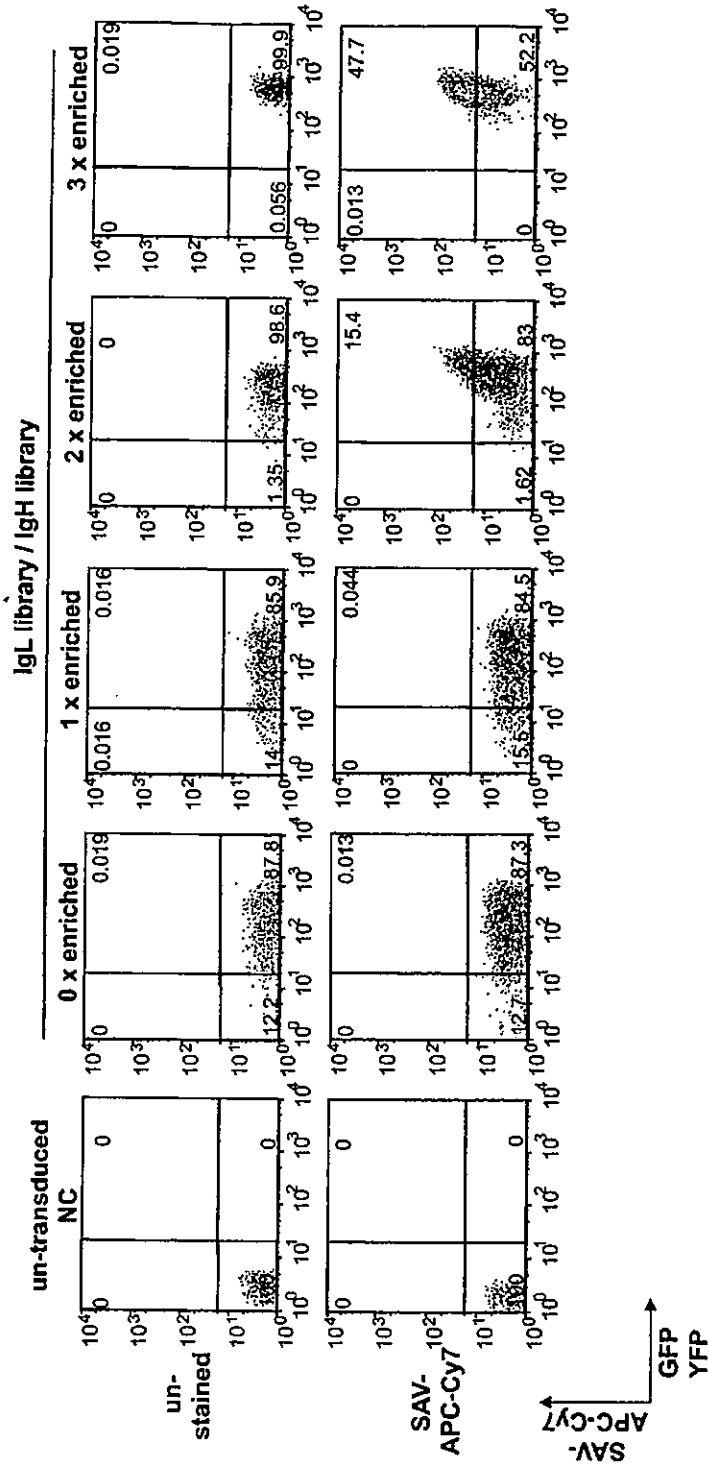


Fig. 25

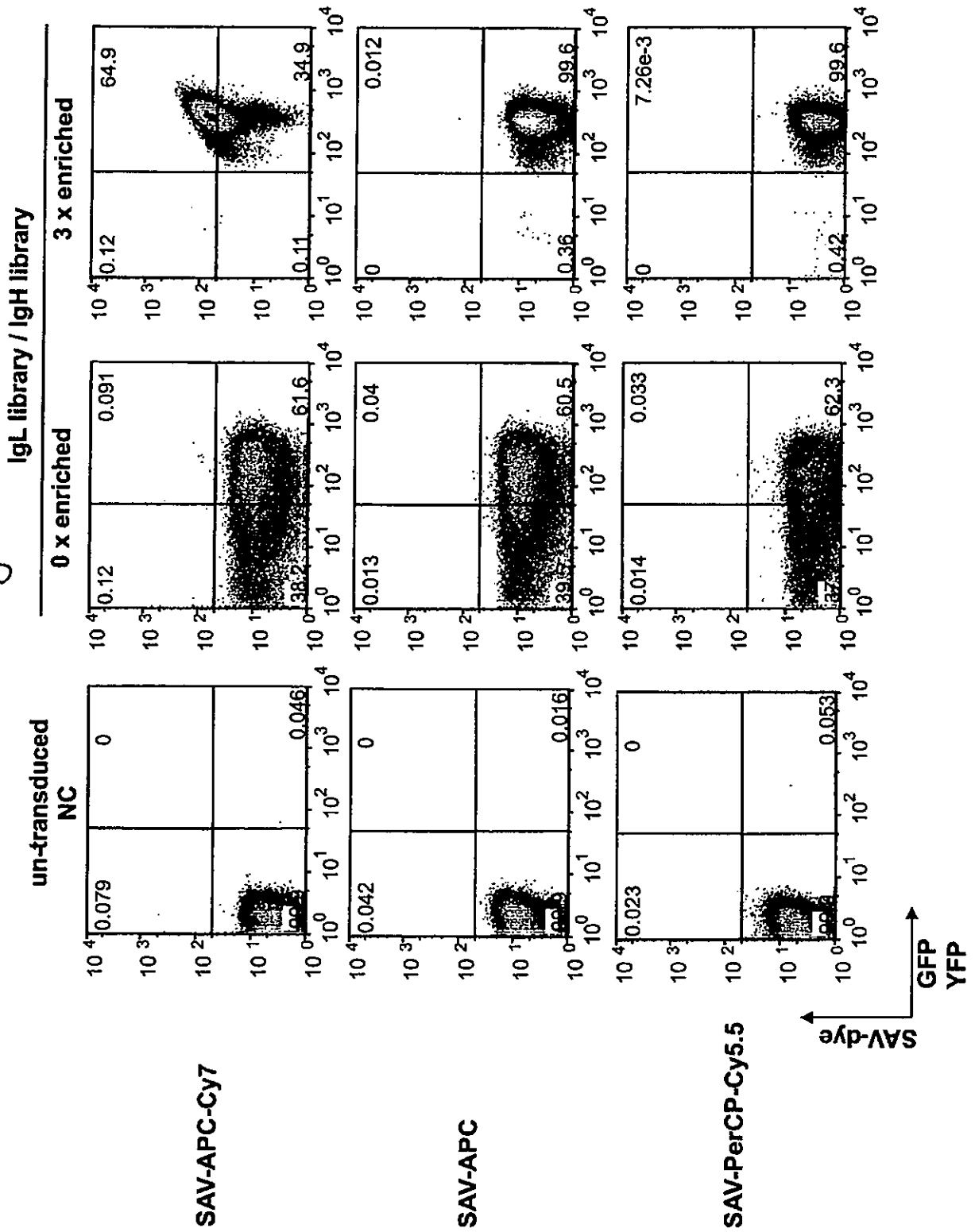


Fig. 26

