1/22

Figure 1

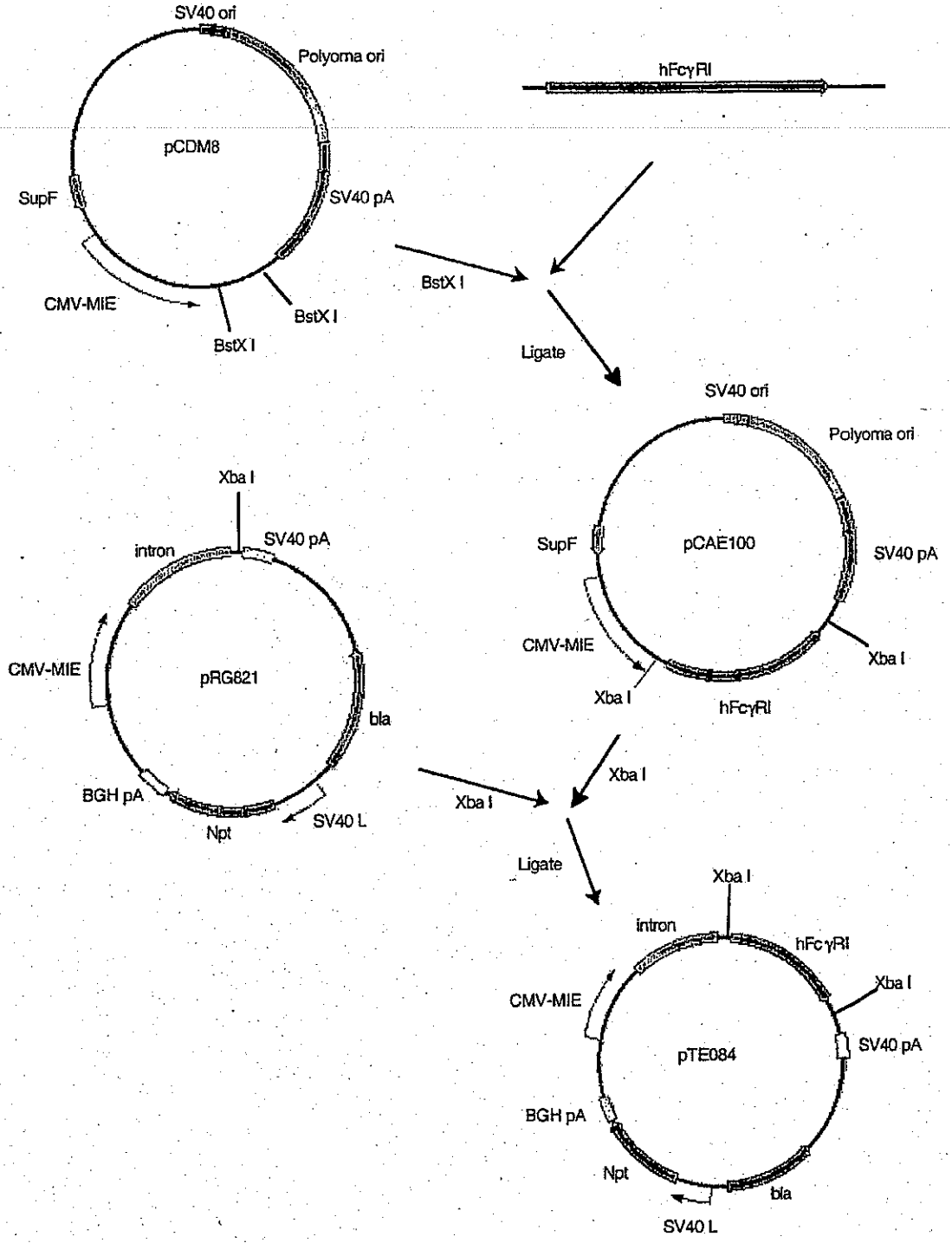

SV40 ori
Polyoma ori
pCDM8
SupF
SV40 pA
CMV-MIE
BstX I
BstX I
hFcγRI
BstX I
Ligate
SV40 ori
Polyoma ori
SupF
pCAE100
SV40 pA
CMV-MIE
Xba I
Xba I
hFcγRI
Xba I
intron
SV40 pA
CMV-MIE
pRG821
bla
BGH pA
Npt
SV40 L
Xba I
Xba I
Ligate
Xba I
intron
hFcγRI
CMV-MIE
Xba I
pTE084
SV40 pA
BGH pA
Npt
bla
SV40 L

Figure 2A.

CHO K1

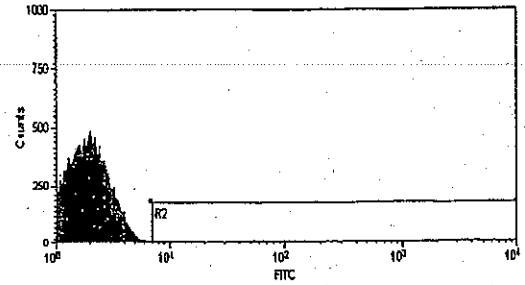


Figure 2B.

CHO K1 + FITC-hFc

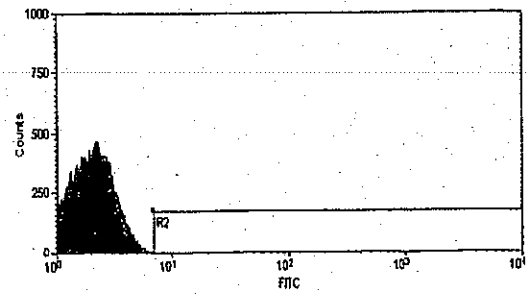


Figure 2C.

CHO K1/hFcγRI pool + FITC-hFc

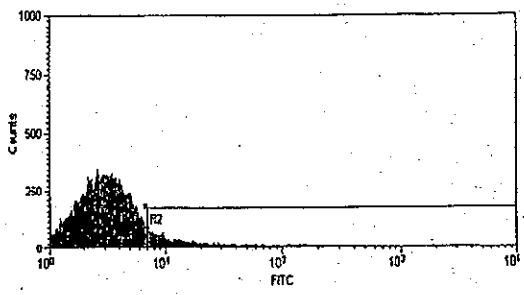


Figure 2D.

RGC3 + FITC-hFc

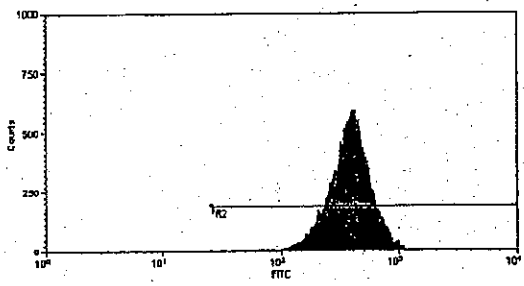


Figure 3

4SC622	IgG Species	Cells Labeled (%)
–	–	0.7
+	–	93.2
+	human	0.8
+	rat	2.0
+	rabbit	(1.0)
+	canine	4.8
+	bovine	92.0
+	ovine	91.7

Figure 4A.

RGC1

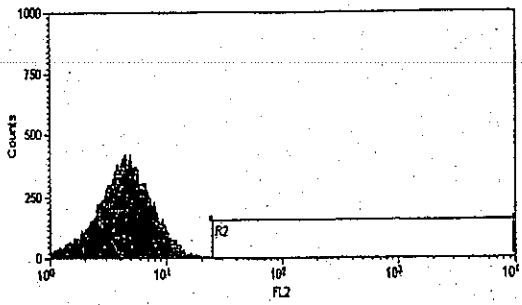


Figure 4B.

RGC1 + 4SC622

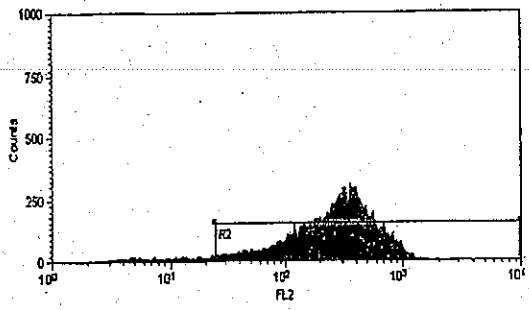


Figure 4C.

RGC1+ 4SC622 + rat IgG

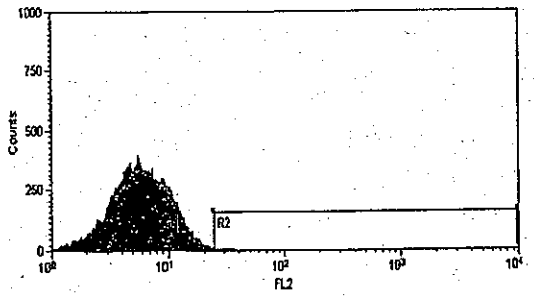


Figure 5A. RGC2

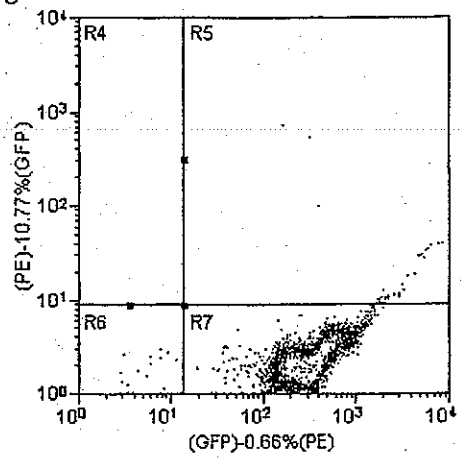


Figure 5B. RGC2 + 4SC622

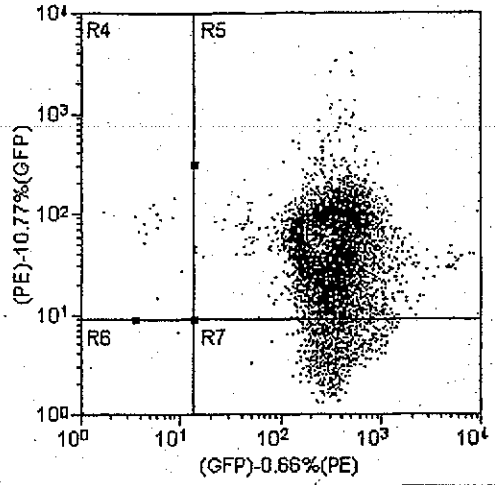


Figure 5C. RGC4

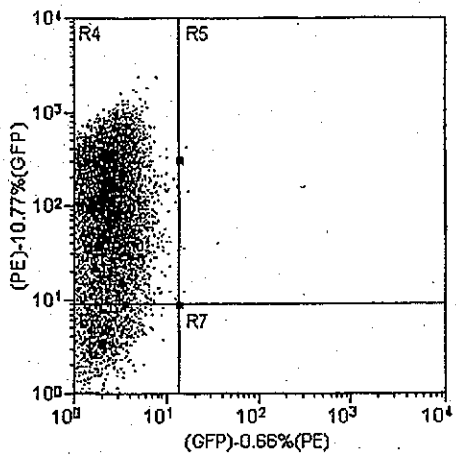


Figure 5D. RGC4 + rat IgG

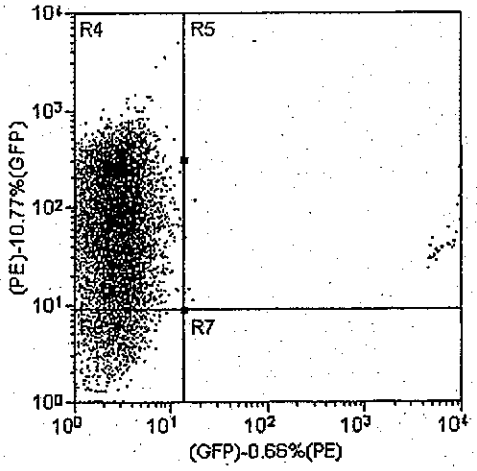


Figure 5E. RGC4 + RGC2

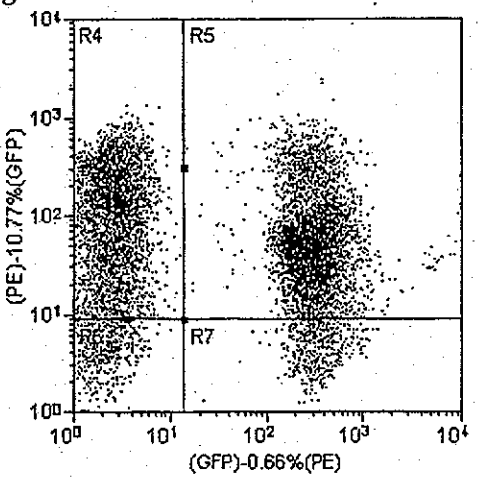


Figure 5F. RGC4 + RGC2 + rat IgG

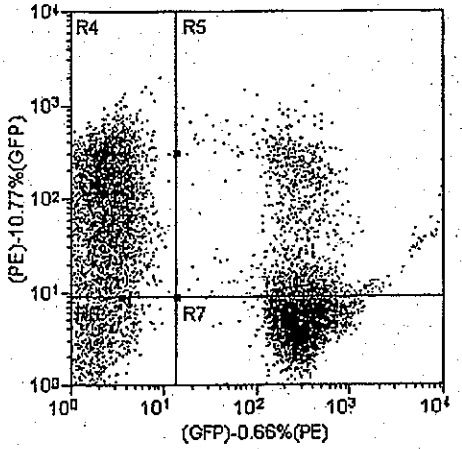


Figure 6

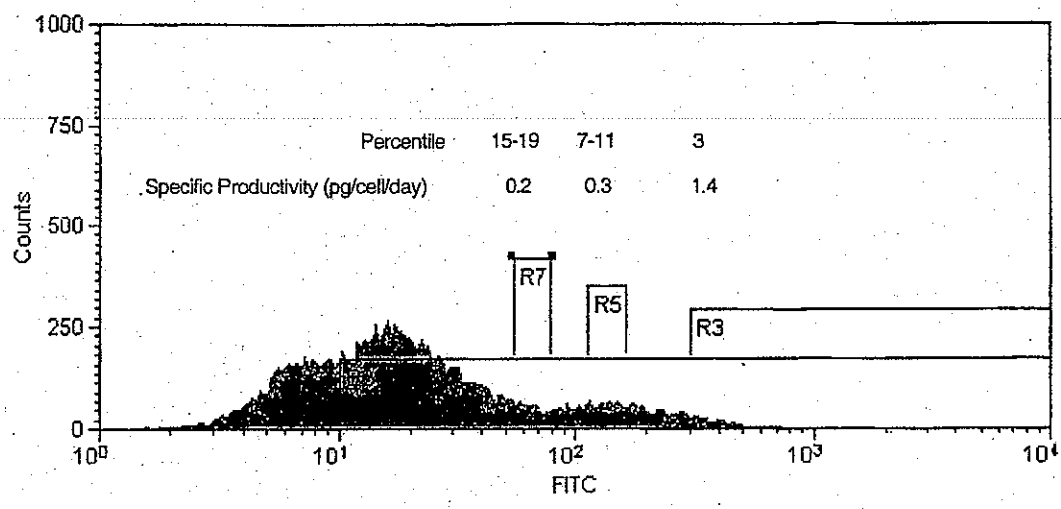

1000
750
500
250
0
Counts
Percentile 15-19 7-11 3
Specific Productivity (pg/cell/day) 0.2 0.3 1.4
R7
R5
R3
10^0
10^1
10^2
10^3
10^4
FITC

Figure 7

Protein	Transient	Hand-picked CHO K1 Stable Cell Lines		RGC1-derived Stable Cell Lines	
	ug/ml	Sp.Prod. (pg/cell/day)	# clones screened	Sp. Prod. (pg/cell/day)	# clones screened
4SC622	1.1	12	2700	12	6
hVEGF-R1R2	33	68	190	77	62
hVEGF-R1R3	27	5	100	22.6	42

Figure 8A. RGC1

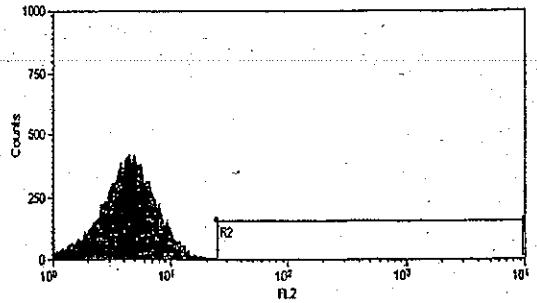


Figure 8B. RGC1 + 4SC622

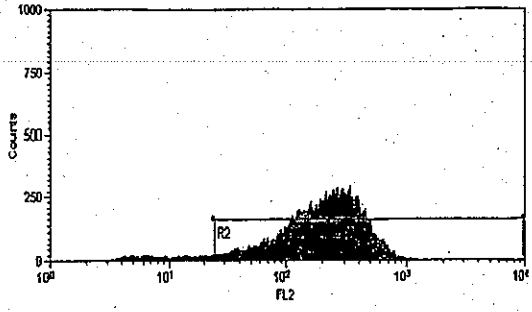


Figure 8C. RGC1 + 4SC622 + 18 hrs.

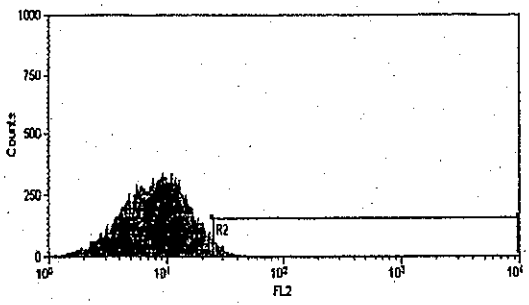


Figure 9

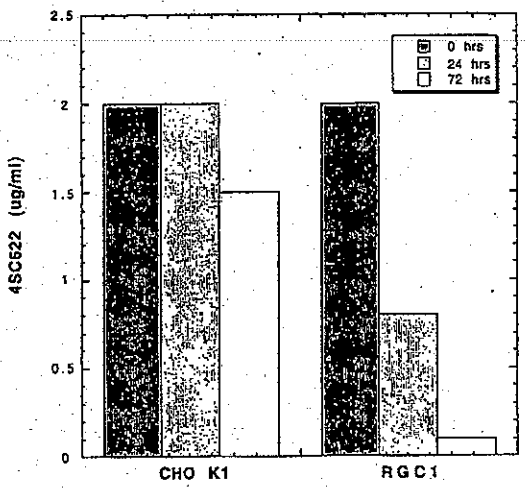


Figure 10

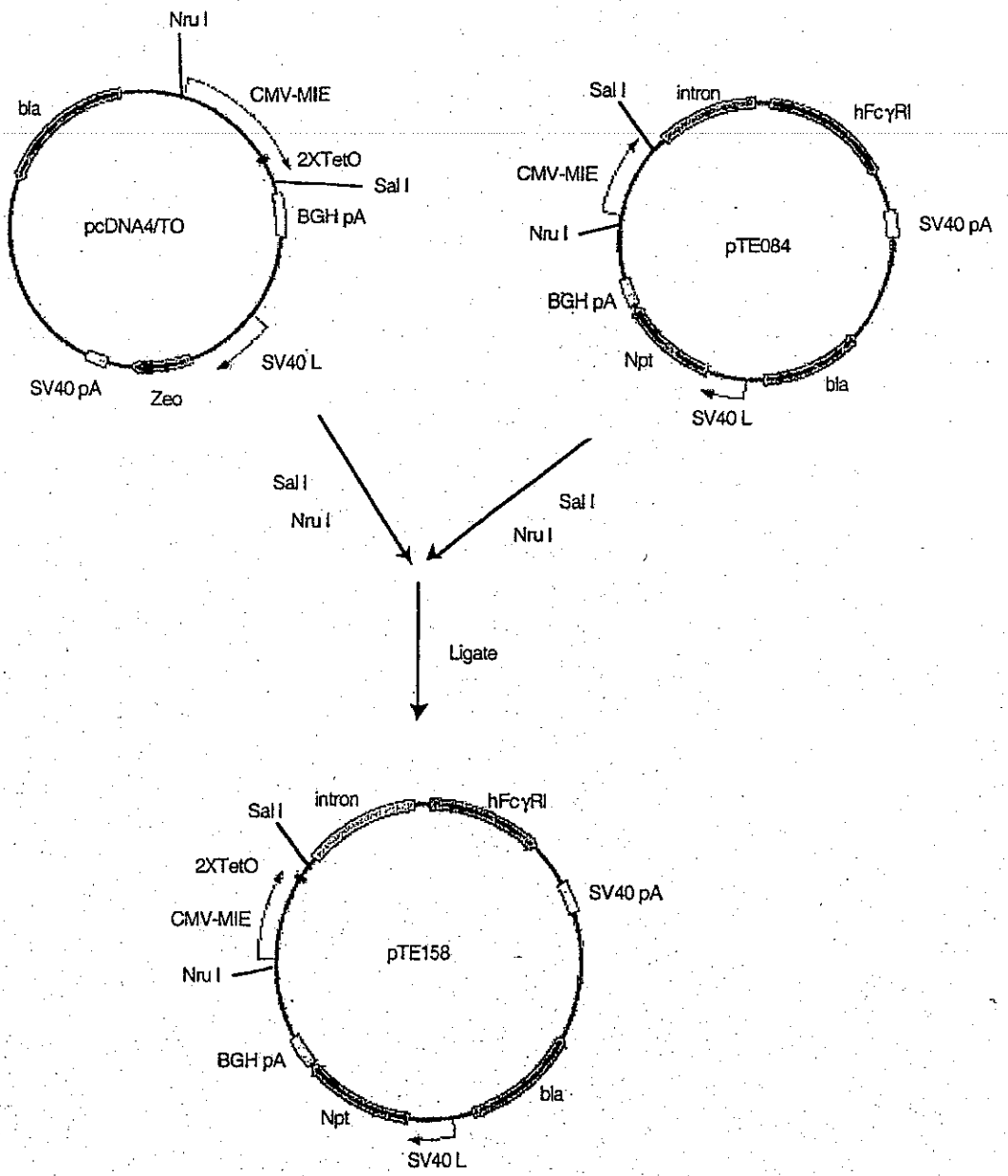

Nru I
CMV-MIE
bla
2XTetO
Sal I
pcDNA4/TO
BGH pA
SV40 L
SV40 pA
Zeo
Sal I
intron
hFcγRI
CMV-MIE
SV40 pA
Nru I
pTE084
BGH pA
Npt
bla
SV40 L
Sal I
Nru I
Sal I
Nru I
Ligate
Sal I
intron
hFcγRI
2XTetO
SV40 pA
CMV-MIE
pTE158
Nru I
BGH pA
bla
Npt
SV40 L

Figure 11A. CHO K1

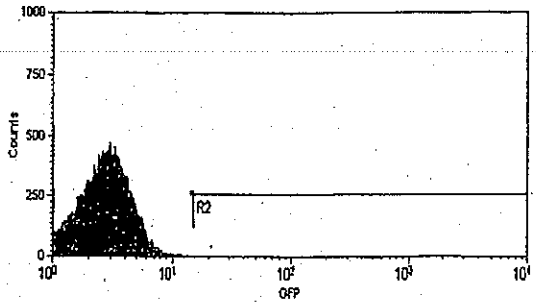


Figure 11B. RGC10 - DOX

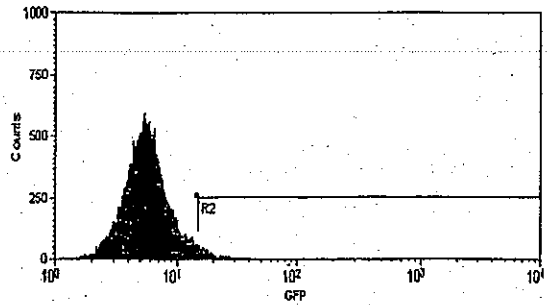


Figure 11C. RGC10 + DOX

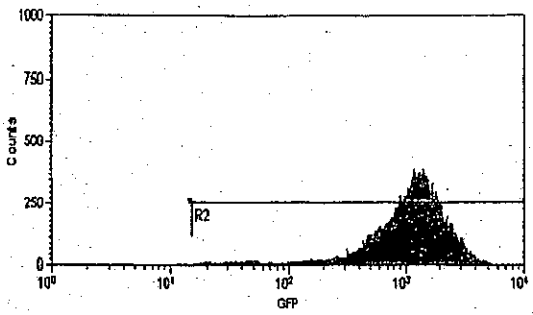


Figure 12A. CHO K1

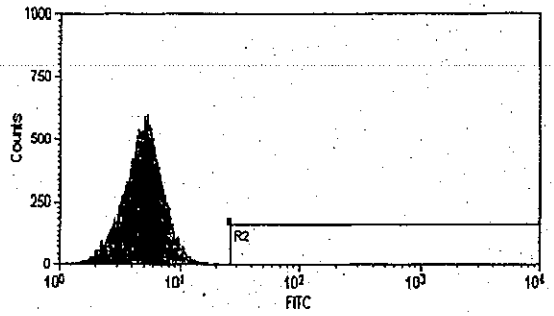


Figure 12B. RGC10/4SC622 pool-Dox

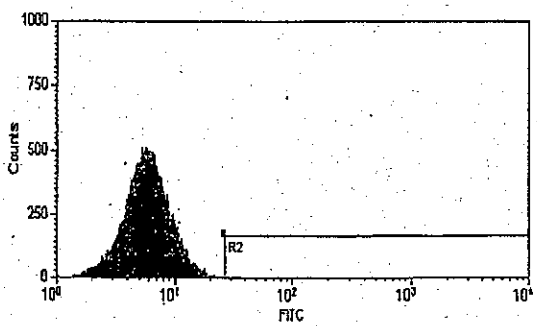


Figure 12C. RGC10/4SC622 pool + Dox

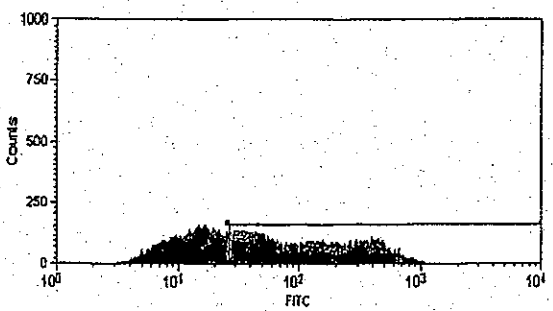


Figure 13

Clone	Sp. Prod.	Clone	Sp. Prod.
1A2	12.15	3A1	9.7
1A3	10.3	3B3	6.75
1B4	11.0	3B6	10.8
1C2	17.8	3C1	11.9
1D3	11.8	3C2	7.7
1D5	10.4	3C6	8.3
		3D1	10.5

Figure 14

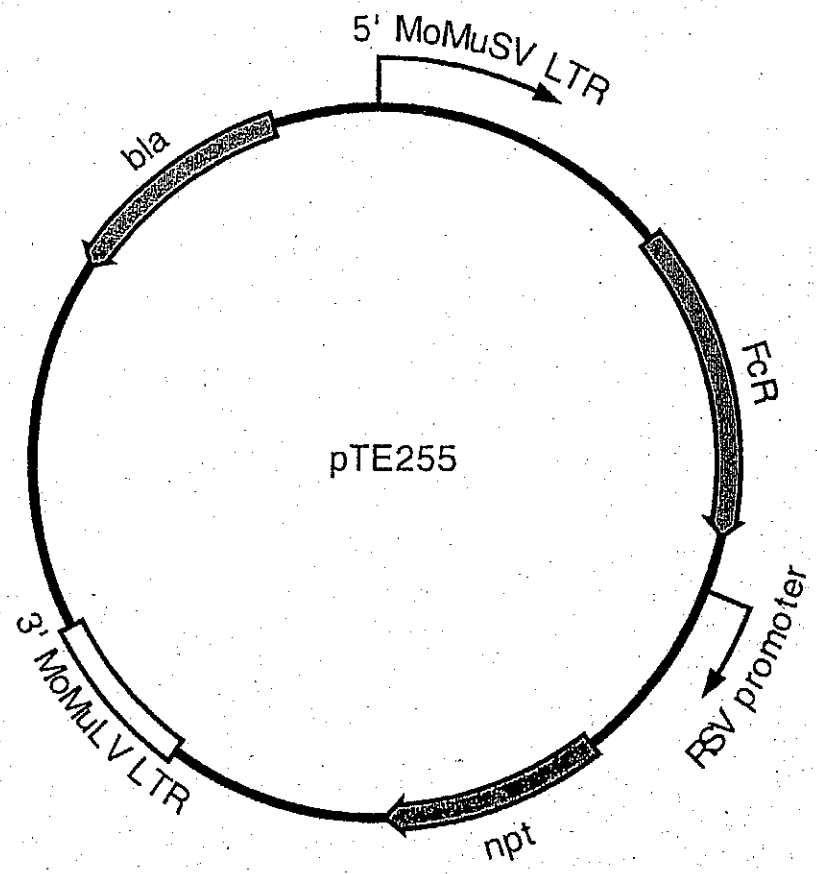


Figure 15A

SP2/0

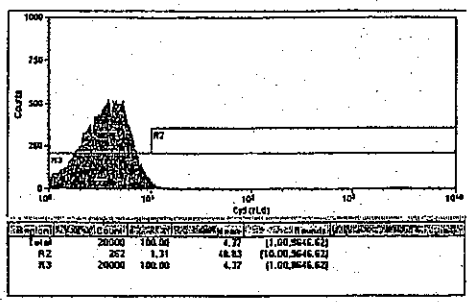


Figure 15B

SP2/0 + Cy5-hFc

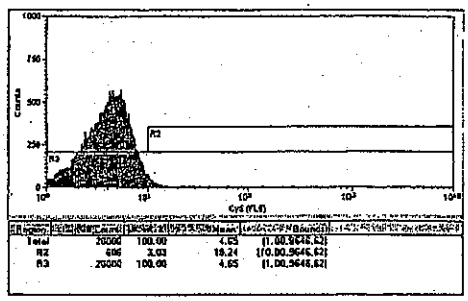


Figure 15C

SP2/0-hFcγRI-4

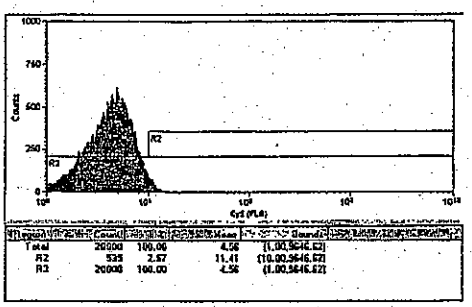


Figure 15D

SP2/0- hFcγRI-4 + Cy5-hFc

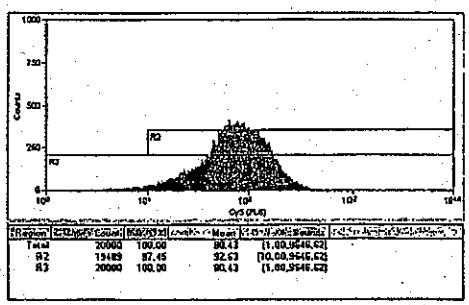


Figure 16

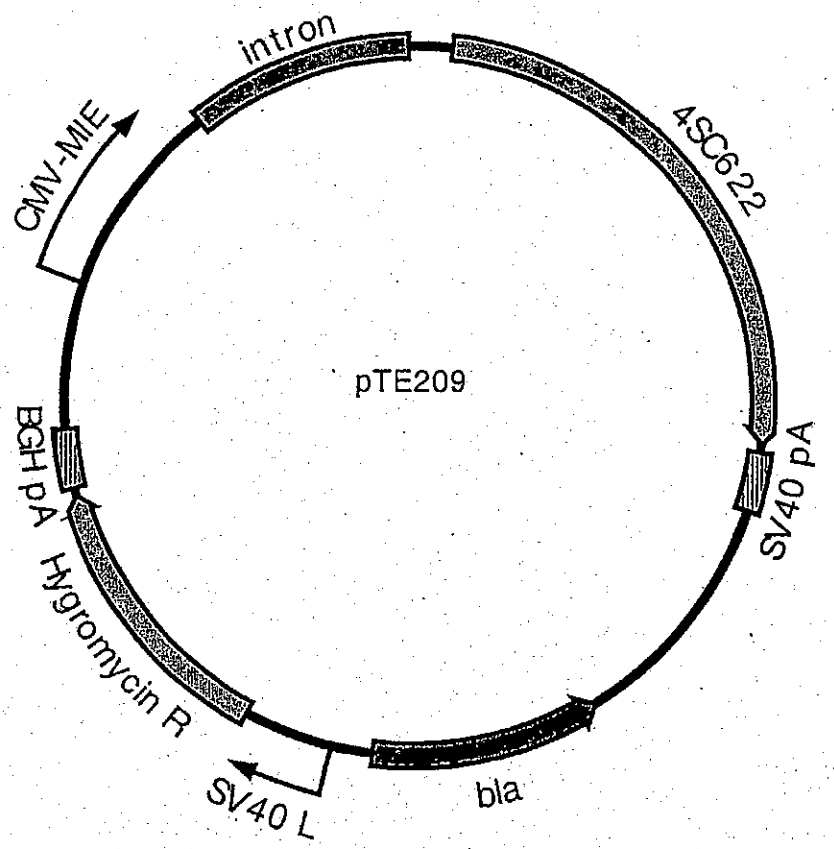
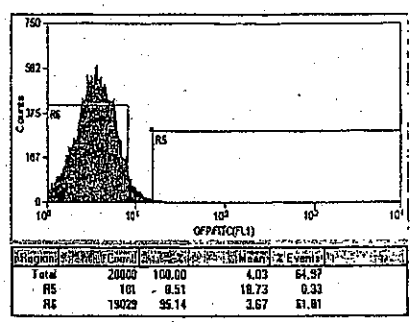

intron
CMV-MIE
4SC622
pTE209
BGH pA
Hygromycin R
SV40 pA
SV40 L
bla

Figure 17A

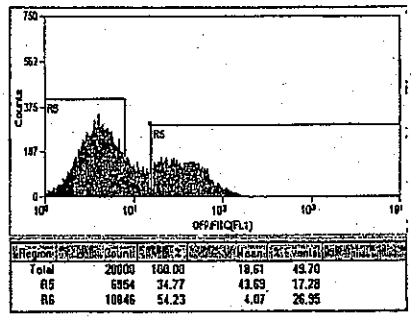
SP2/0-hFcγRI-4/4SC622 pool



Region	Count		Mean	
Total	20000	100.00	4.03	54.57
R5	101	0.51	18.73	0.33
R6	19029	95.14	3.67	51.81

Figure 17B

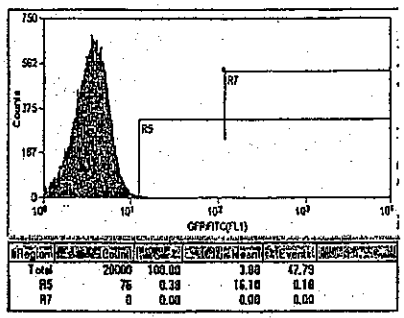
SP2/0-hFcγRI-4/4SC622 pool + FITC α–hIgG



Region	Count		Mean	
Total	20000	100.00	19.61	49.70
R5	6954	34.77	43.69	17.28
R6	10846	54.23	4.07	26.95

Figure 17C

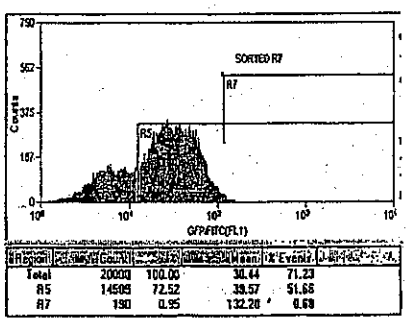
SP2/0-hFcγRI-4/4SC622 top 1% pool



Region	Count		Mean	
Total	20000	100.00	3.88	42.79
R5	76	0.38	16.16	0.16
R7	0	0.00	0.00	0.00

Figure 17D

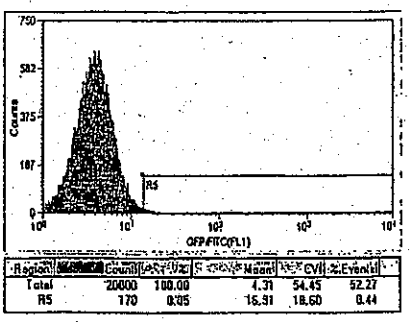
SP2/0-hFcγRI-4/4SC622 top 1% pool + FITC α–hIgG



Region	Count		Mean	
Total	20000	100.00	30.44	71.23
R5	14505	72.52	39.57	51.66
R7	190	0.95	132.28	0.69

Figure 17E

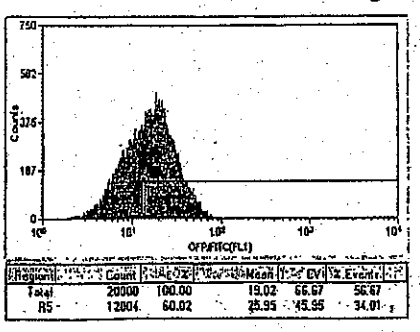
Clone 5H11



Region	Count		Mean	CV	
Total	20000	100.00	4.31	54.45	52.27
R5	170	0.85	16.91	18.60	0.44

Figure 17F

Clone 5H11 + FITC α–hIgG



Region	Count		Mean	CV	
Total	20000	100.00	19.02	66.67	56.67
R5	12004	60.02	25.95	45.95	34.01

Figure 18

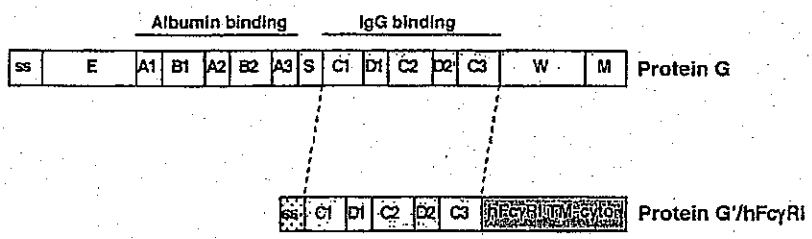

Albumin binding
IgG binding
ss
E
A1
B1
A2
B2
A3
S
C1
D1
C2
D2
C3
W
M
Protein G
ss
C1
D1
C2
D2
C3
hFcγRI
tM-cyto
Protein G'/hFcγRI

Figure 19

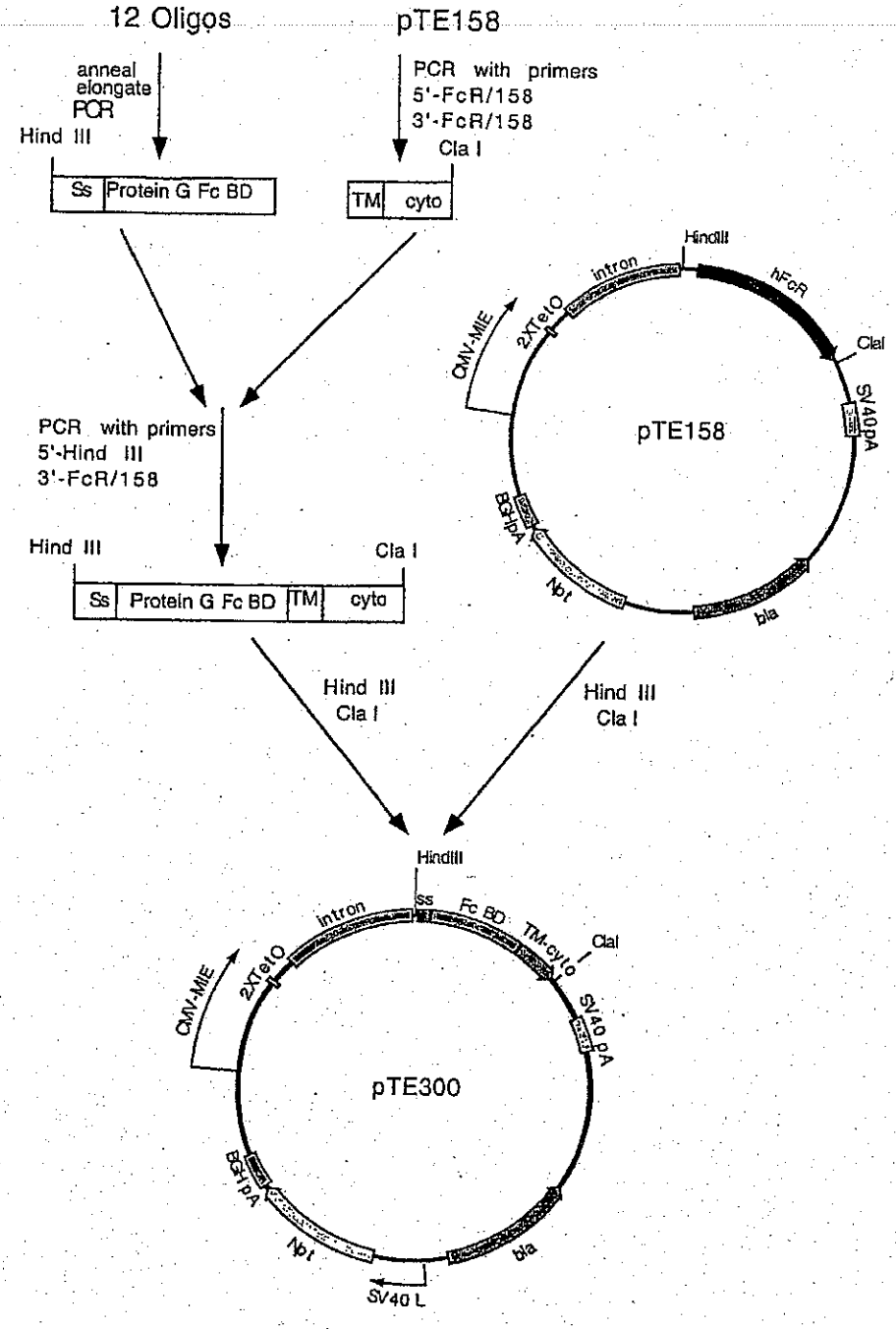

12 Oligos
pTE158
anneal
elongate
PCR
Hind III
PCR with primers
5'-FcR/158
3'-FcR/158
Cla I
Ss
Protein G Fc BD
TM
cyto
HindIII
intron
hFcR
ClaI
CMV-MIE
2XTetO
SV40pA
pTE158
BGHpA
Npt
bla
PCR with primers
5'-Hind III
3'-FcR/158
Hind III
Cla I
Ss
Protein G Fc BD
TM
cyto
Hind III
Cla I
Hind III
Cla I
HindIII
Ss
Fc BD
TM-cyto
ClaI
intron
CMV-MIE
2XTetO
SV40 pA
pTE300
BGHpA
Npt
bla
SV40 L

Figure 20A
RGC14

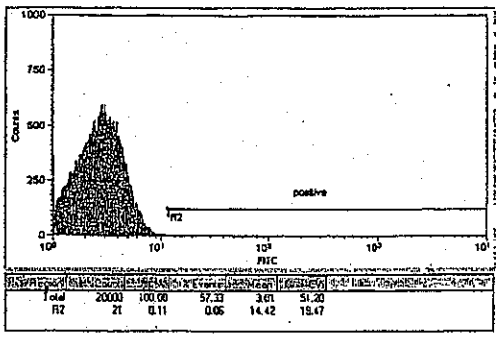


Figure 20B
RGC14 + FITC-hFc

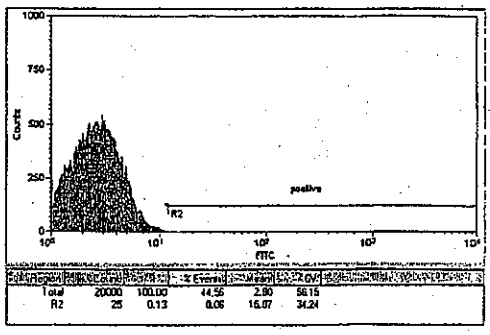


Figure 20C
RGC14/pTE300 pool

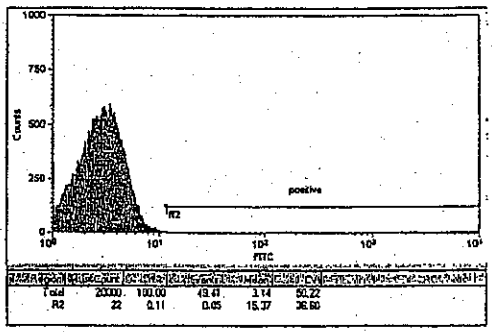


Figure 20D
RGC14/pTE300 pool + FITC-hFc

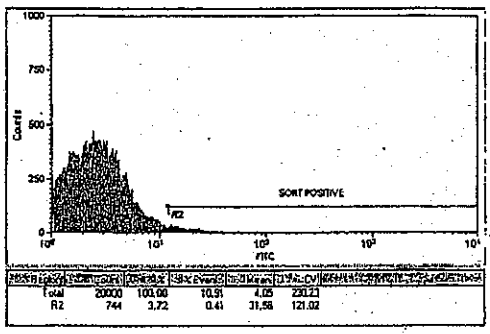
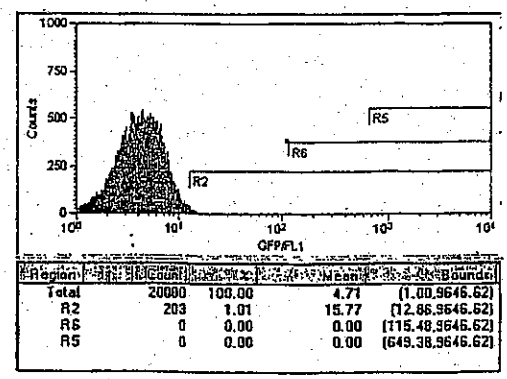
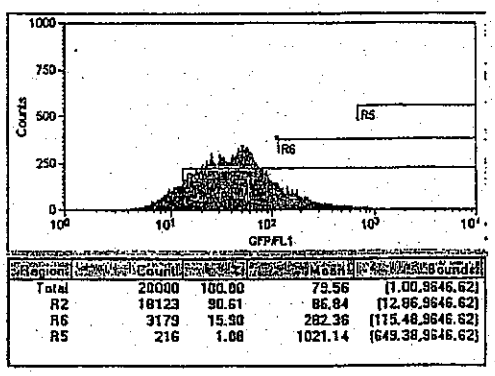


Figure 20E
RGC18



Region	Count	%	Mean	% Bounds
Total	20000	100.00	4.71	(1.00,9646.62)
R2	203	1.01	15.77	(12.86,9646.62)
R6	0	0.00	0.00	(115.48,9646.62)
R5	0	0.00	0.00	(649.38,9646.62)

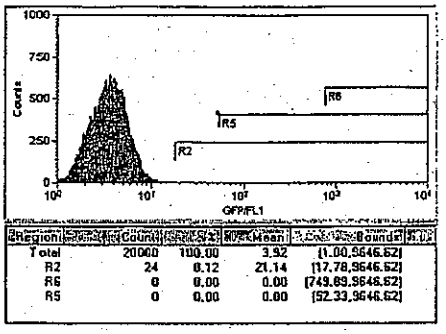
Figure 20F
RGC18 + FITC α–bIgG



Region	Count	%	Mean	% Bounds
Total	20000	100.00	79.56	(1.00,9646.62)
R2	18123	90.61	86.84	(12.86,9646.62)
R6	3179	15.90	282.36	(115.48,9646.62)
R5	216	1.08	1021.14	(649.38,9646.62)

Figure 21A

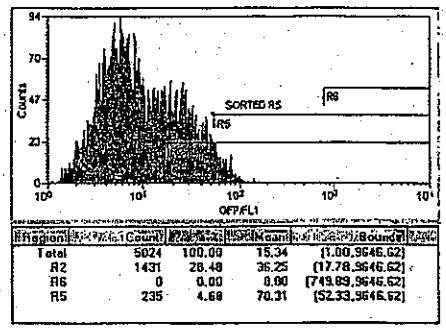
4SC622 pool



Region	Count	%	Mean	Bounds
Total	20000	100.00	3.92	[1.00,9646.62]
R2	24	0.12	21.14	[17.78,9646.62]
R6	0	0.00	0.00	[749.89,9646.62]
R5	0	0.00	0.00	[52.33,9646.62]

Figure 21B

4SC622 pool + FITC α–hIgG



Region	Count	%	Mean	Bounds
Total	5024	100.00	15.34	[1.00,9646.62]
R2	1431	28.48	36.25	[17.78,9646.62]
R6	0	0.00	0.00	[749.89,9646.62]
R5	235	4.68	70.31	[52.33,9646.62]

Figure 22A.

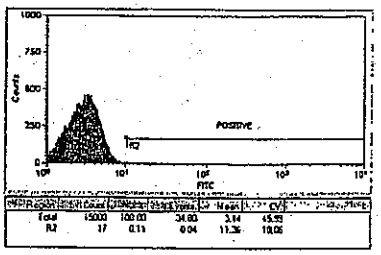


Figure 22B.

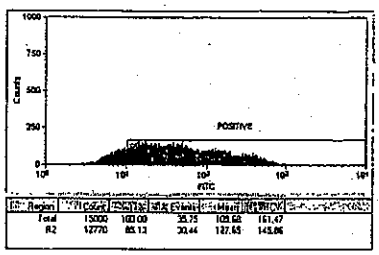


Figure 22C.

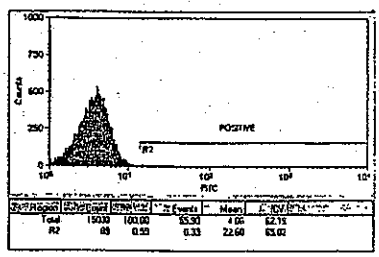


Figure 22D.

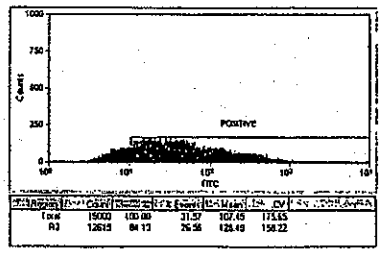


Figure 22E.

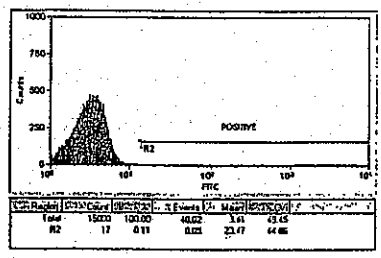


Figure 22F.

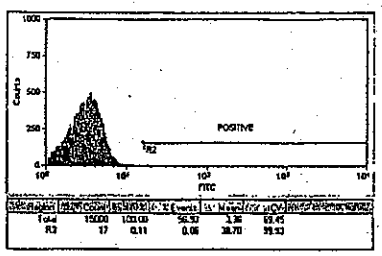


Figure 22G.

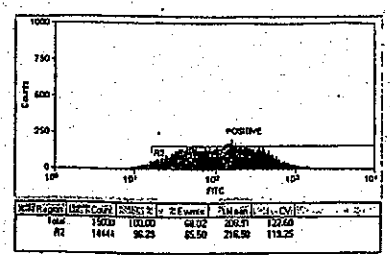


Figure 22H.

