Fig. 1


Extracellular Loop 1
CLDN6 (SEQ ID NO: 2) MASAGMQILGVVLTLLGWVNGLVSCALPMWKVTAFIGNSIVVAQVVWEGLWMSCVVQSTG 60
CLDN9 (SEQ ID NO: 9) MASTGLELLGMTLAVLGWLGTLVSCALPLWKVTAFIGNSIVVAQVVWEGLWMSCVVQSTG 60
CLDN4 (SEQ ID NO: 10) MASMGLQVMGIALAVLGWLAVMLCCALPMWRVTAFIGSNIVTSQTIWEGLWMNCVVQSTG 60
CLDN3 (SEQ ID NO: 11) -MSMGLEITGTALAVLGWLGTIVCCALPMWRVSAFIGSNIITSQNIWEGLWMNCVVQSTG 59
CLDN6 (SEQ ID NO: 2) QMQCKVYDSLLALPQDLQAARALCVIALLVALFGLLVYLAGAKCTTCVEEKDSKARLVLT 120
CLDN9 (SEQ ID NO: 9) QMQCKVYDSLLALPQDLQAARALCVIALLLALLGLLVAITGAQCTTCVEDEGAKARIVLT 120
CLDN4 (SEQ ID NO: 10) QMQCKVYDSLLALPQDLQAARALVIISIIVAALGVLLSVVGGKCTNCLEDESAKAKTMIV 120
CLDN3 (SEQ ID NO: 11) QMQCKVYDSLLALPQDLQAARALIVVAILLAAFGLLVALVGAQCTNCVQDDTAKAKITIV 119
Extracellular Loop 2
CLDN6 (SEQ ID NO: 2) SGIVFVISGVLTLIPVCWTAHAIIRDFYNPLVAEAQKRELGASLYLGWAASGLLLLGGGL 180
CLDN9 (SEQ ID NO: 9) AGVILLLAGILVLIPVCWTAHAIIQDFYNPLVAEALKRELGASLYLGWAAAALLMLGGGL 180
CLDN4 (SEQ ID NO: 10) AGVVFLLAGLMVIVPVSWTAHNIIQDFYNPLVASGQKREMGASLYVGWAASGLLLLGGGL 180
CLDN3 (SEQ ID NO: 11) AGVLFLLAALLTLVPVSWSANTIIRDFYNPVVPEAQKREMGAGLYVGWAAAALQLLGGAL 179
CLDN6 (SEQ ID NO: 2) LCCTCPSGGSQGPSHYMARYSTSAPAISRGPSEYP-----TKNYV 220
CLDN9 (SEQ ID NO: 9) LCCTCPPPQVERPRG--PRLGYSIPSRS-GASGLD-----KRDYV 217
CLDN4 (SEQ ID NO: 10) LCCNCPPRT---DKPYSAK---YSAARSAAASN----------YV 209
CLDN3 (SEQ ID NO: 11) LCCSCPPR----EKKYTATKVVYSAPRSTGPGASLGTGYDRKDYV 220

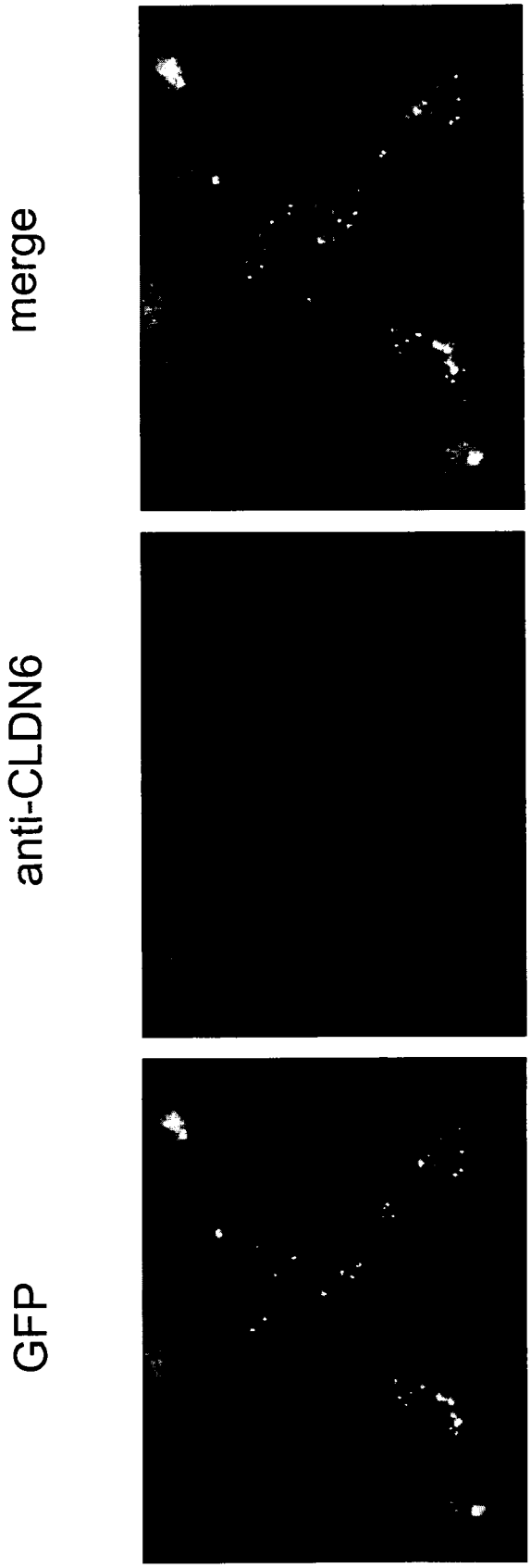


Fig. 2A



Fig. 2B

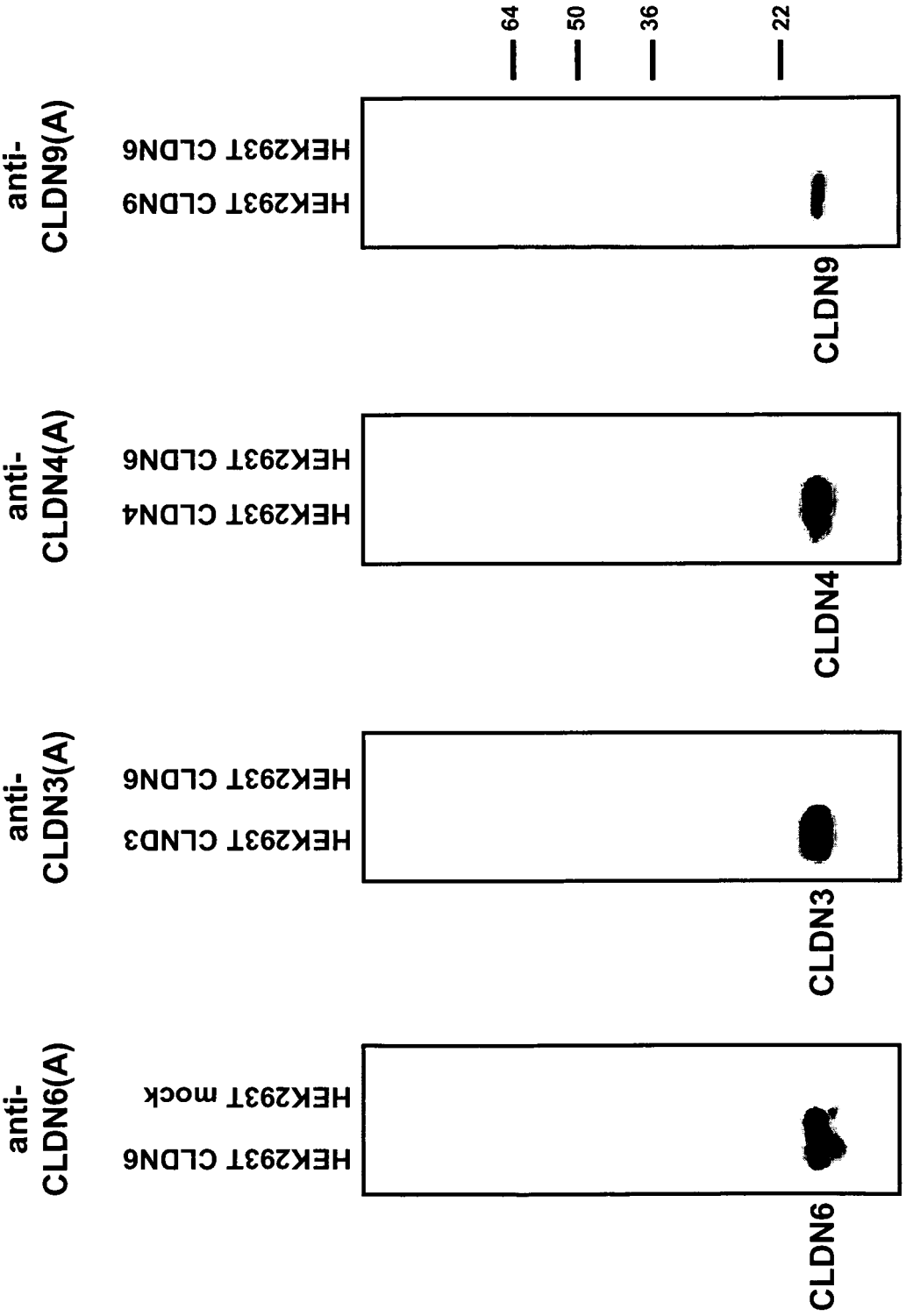

64
50
36
22
anti-CLDN9(A)
HEK293T CLDN6
HEK293T CLDN9
CLDN9
anti-CLDN4(A)
HEK293T CLDN6
HEK293T CLDN4
CLDN4
anti-CLDN3(A)
HEK293T CLDN6
HEK293T CLND3
CLDN3
anti-CLDN6(A)
HEK293T mock
HEK293T CLDN6
CLDN6

Fig. 3

Fig. 4

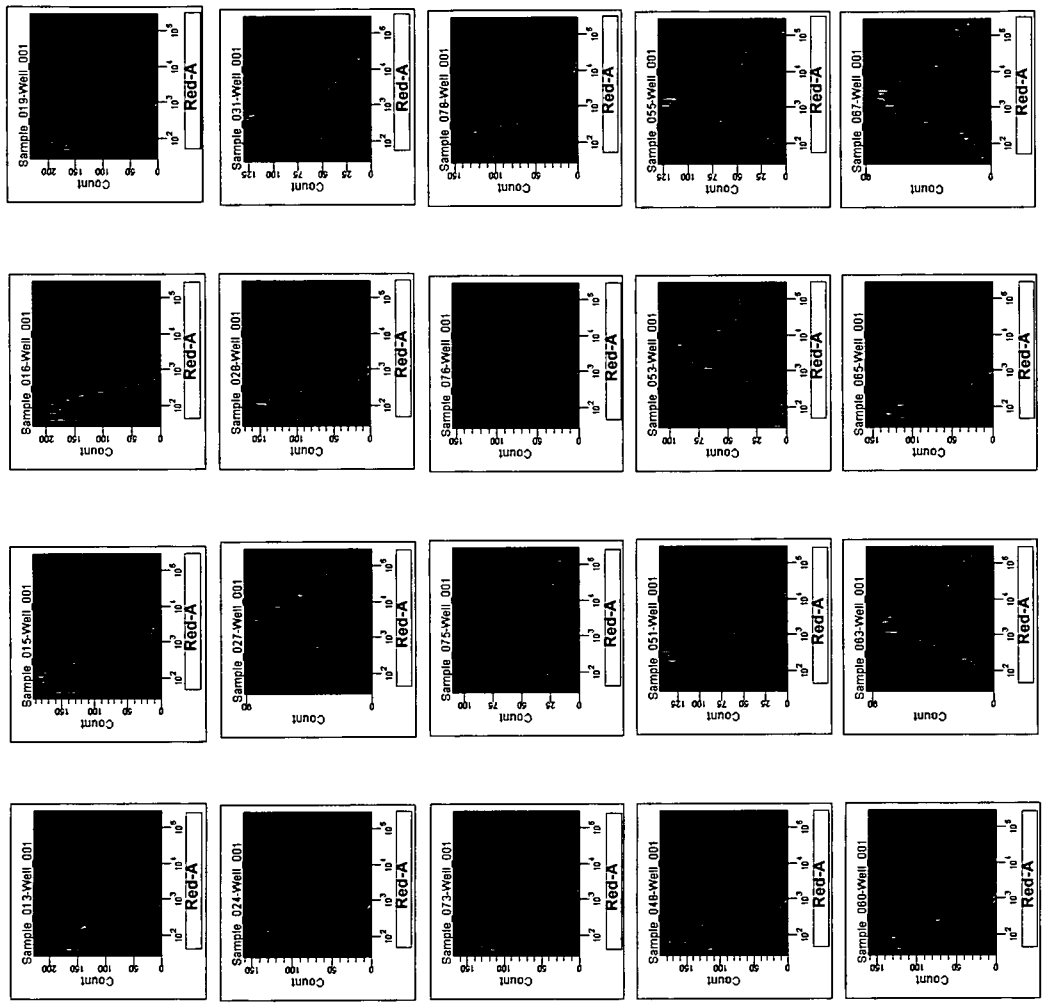

isotype
anti-CLDN6
anti-CLDN3
anti-CLDN4
non-transfected
CLDN6-transfected
CLDN9-transfected
CLDN3-transfected
CLDN4-transfected
Count
Red-A

Fig. 5A

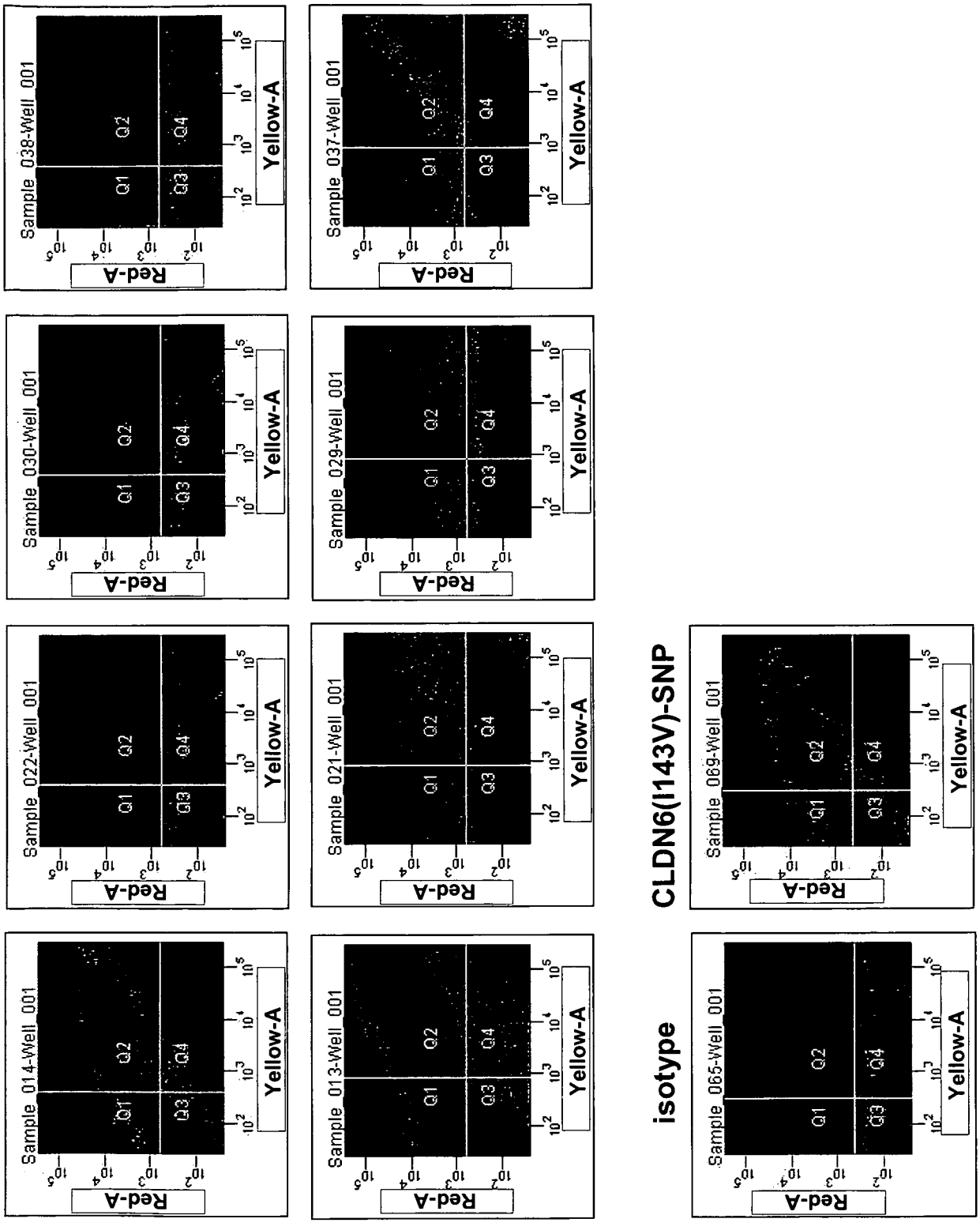

CLDN6
CLDN3
CLDN4
CLDN9
CLDN6(I143V)-SNP
isotype
F3-6C3-H8
F4-4F7-F2
F3-6C3-H8
Red-A
Yellow-A
Q1
Q2
Q3
Q4

Fig. 5B

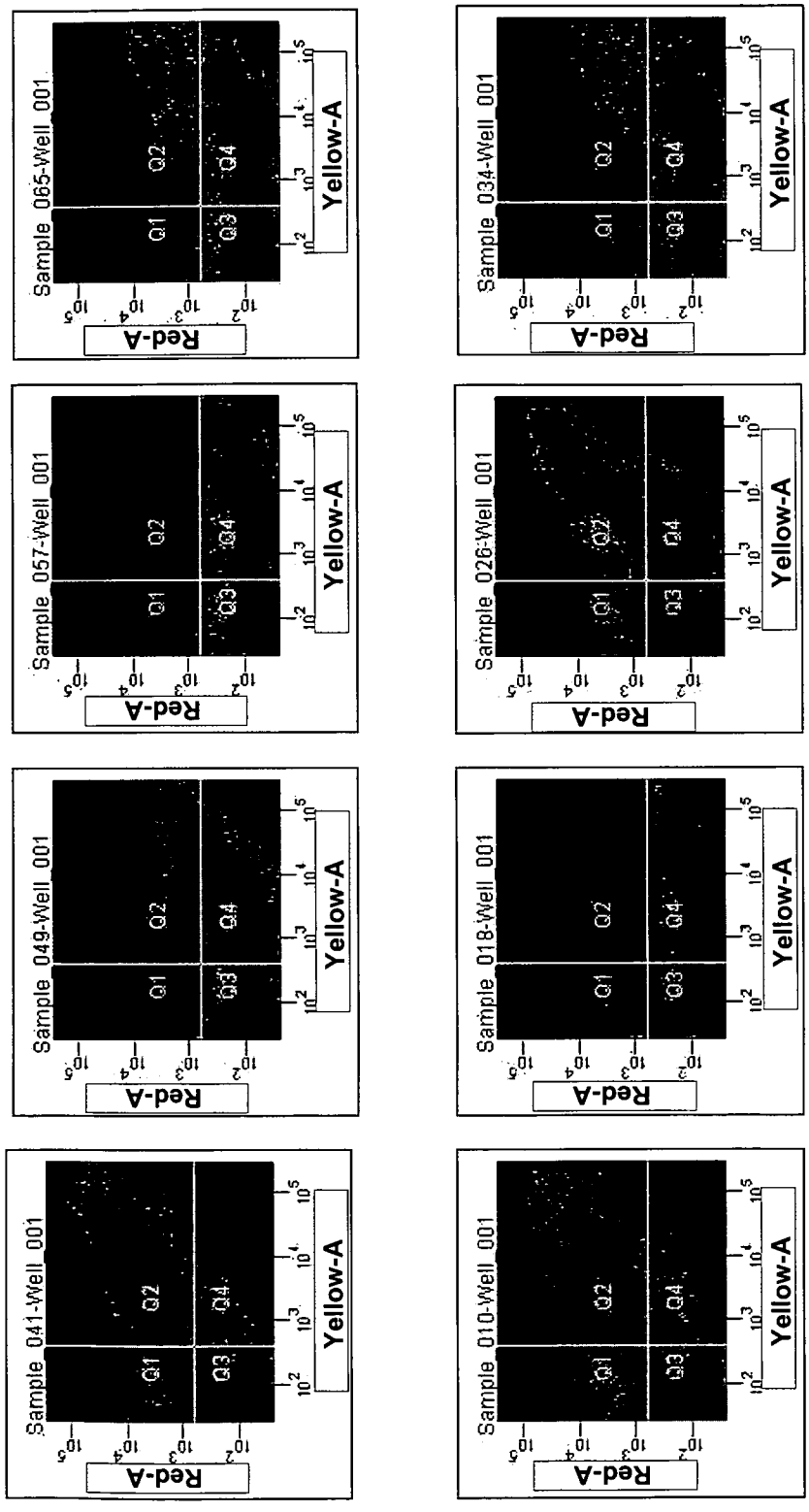
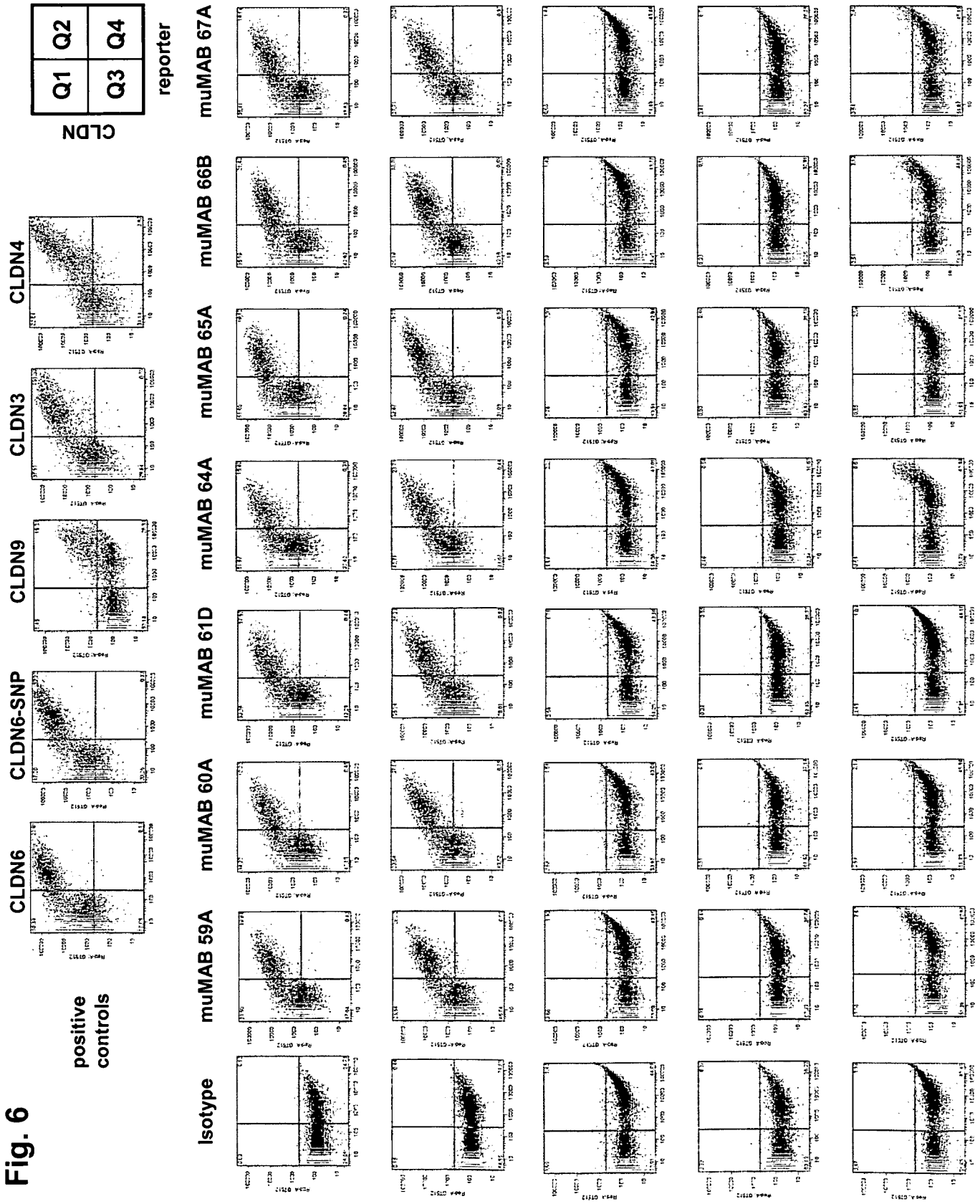

CLDN6
CLDN3
CLDN4
CLDN9
F3-7B3-B4
F3-3F7-A5
Sample 041-Well 001
Sample 049-Well 001
Sample 057-Well 001
Sample 065-Well 001
Sample 010-Well 001
Sample 018-Well 001
Sample 026-Well 001
Sample 034-Well 001
Red-A
Yellow-A
Q1
Q2
Q3
Q4

Fig. 6


positive controls
CLDN6
CLDN6-SNP
CLDN9
CLDN3
CLDN4
CLDN
Q1
Q2
Q3
Q4
reporter
Isotype
muMAB 59A
muMAB 60A
muMAB 61D
muMAB 64A
muMAB 65A
muMAB 66B
muMAB 67A
CLDN6
CLDN6-SNP(I143V)
CLDN9
CLDN3
CLDN4

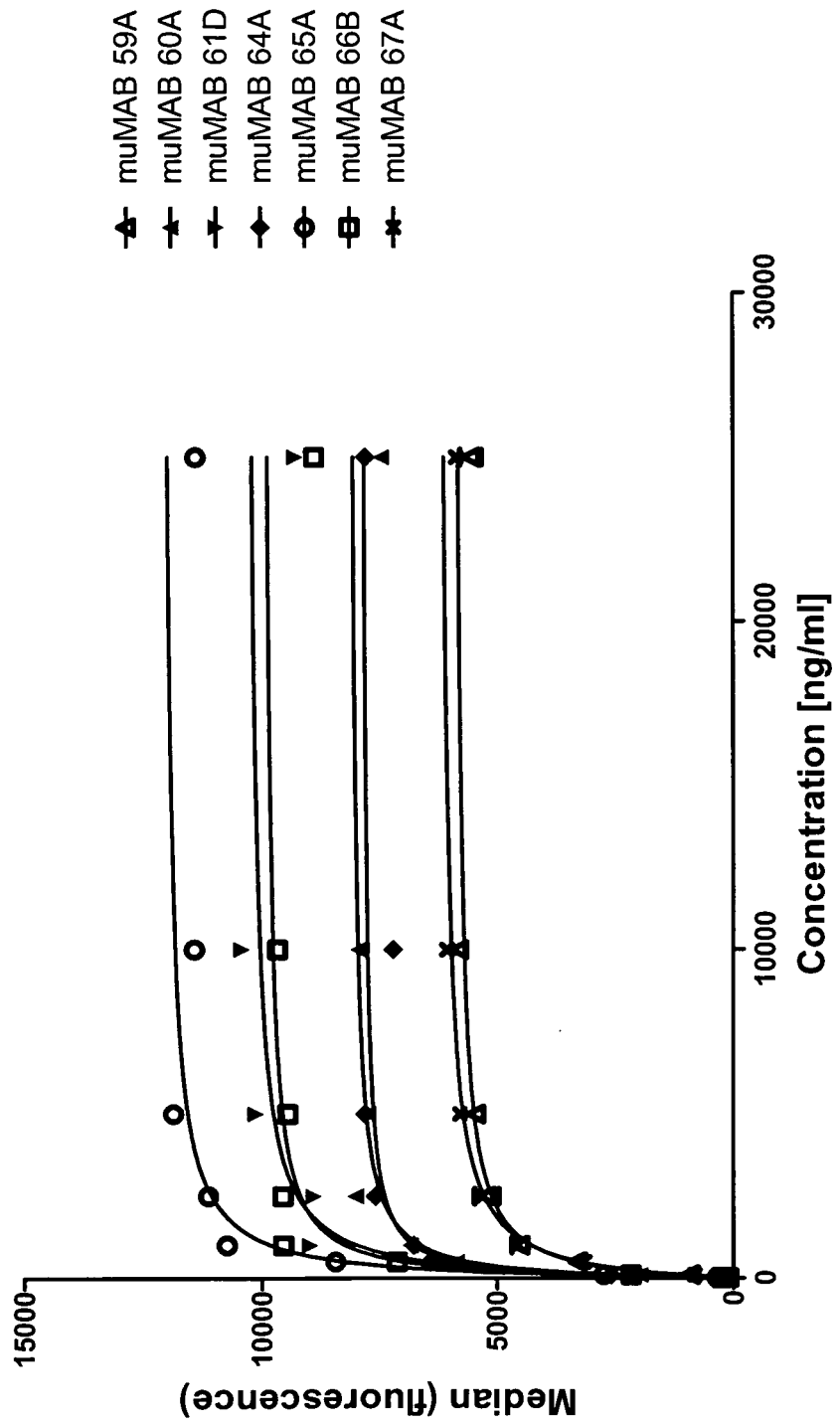

muMAB 59A
muMAB 60A
muMAB 61D
muMAB 64A
muMAB 65A
muMAB 66B
muMAB 67A
Median (fluorescence)
15000
10000
5000
0
0
10000
20000
30000
Concentration [ng/ml]

Fig. 7

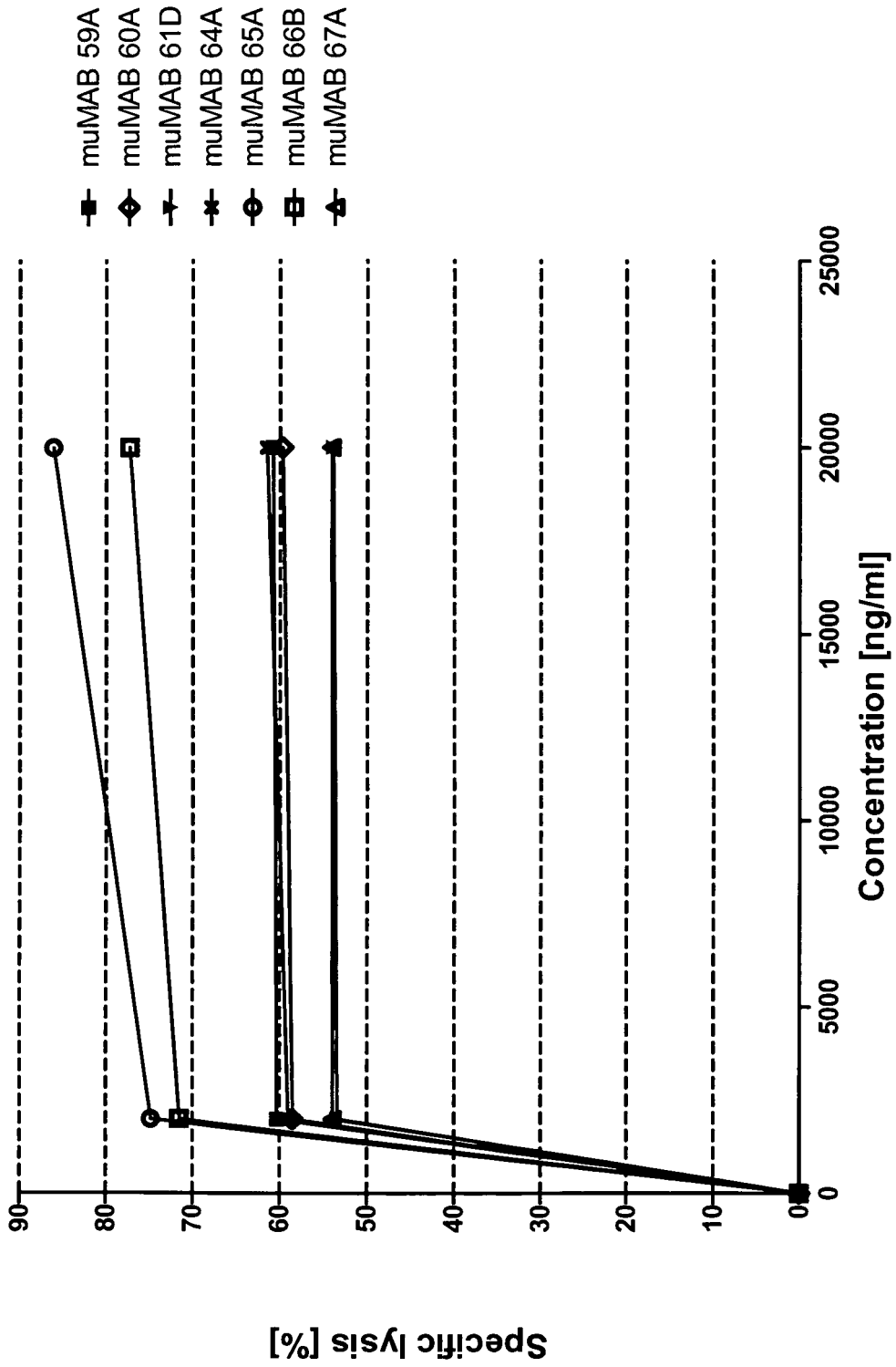
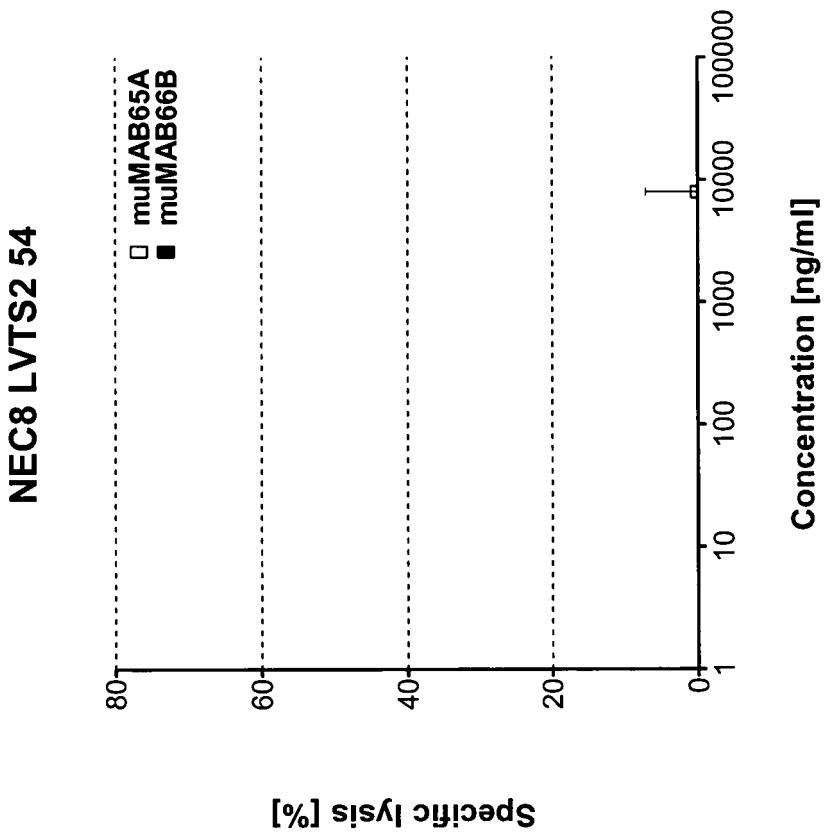
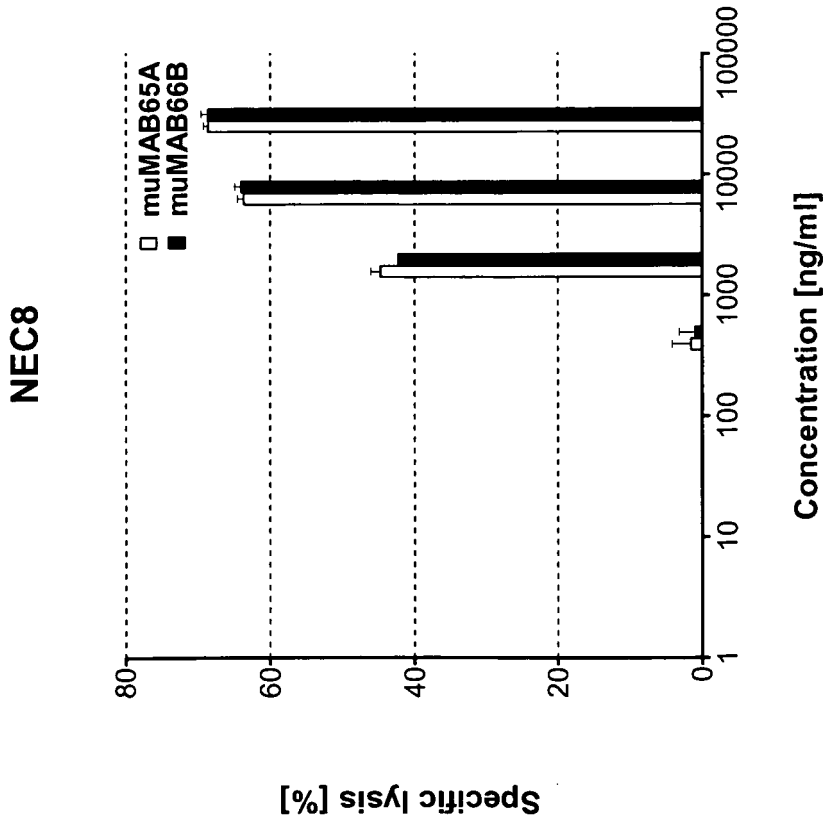

muMAB 59A
muMAB 60A
muMAB 61D
muMAB 64A
muMAB 65A
muMAB 66B
muMAB 67A
Specific lysis [%]
0
10
20
30
40
50
60
70
80
90
0
5000
10000
15000
20000
25000
Concentration [ng/ml]

Fig. 8

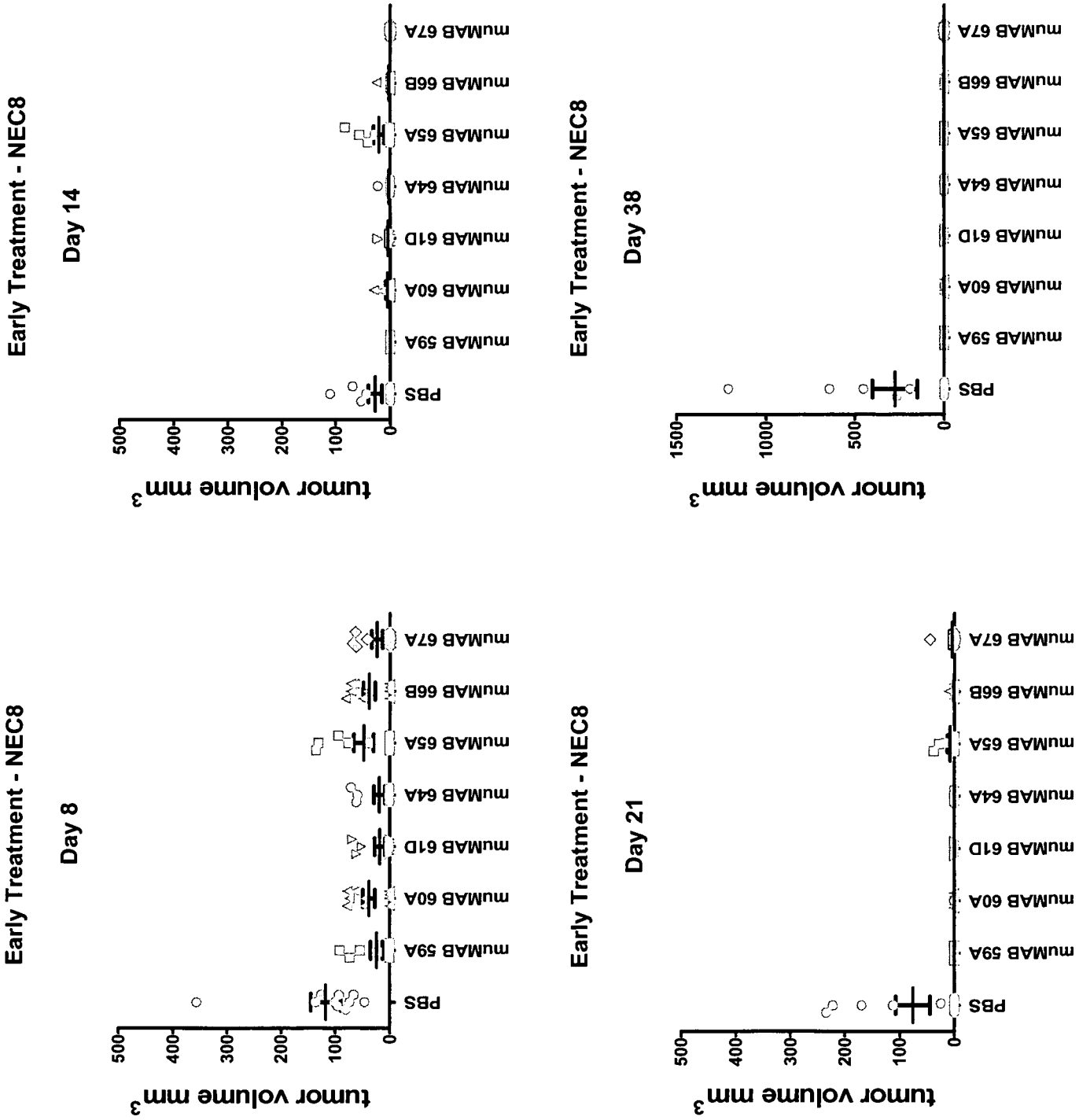




error bars: SEM

Fig. 9

Fig. 10


Early Treatment - NEC8
Day 8
tumor volume mm3
0
100
200
300
400
500
PBS
muMAB 59A
muMAB 60A
muMAB 61D
muMAB 64A
muMAB 65A
muMAB 66B
muMAB 67A
Early Treatment - NEC8
Day 14
tumor volume mm3
0
100
200
300
400
500
PBS
muMAB 59A
muMAB 60A
muMAB 61D
muMAB 64A
muMAB 65A
muMAB 66B
muMAB 67A
Early Treatment - NEC8
Day 21
tumor volume mm3
0
100
200
300
400
500
PBS
muMAB 59A
muMAB 60A
muMAB 61D
muMAB 64A
muMAB 65A
muMAB 66B
muMAB 67A
Early Treatment - NEC8
Day 38
tumor volume mm3
0
500
1000
1500
PBS
muMAB 59A
muMAB 60A
muMAB 61D
muMAB 64A
muMAB 65A
muMAB 66B
muMAB 67A

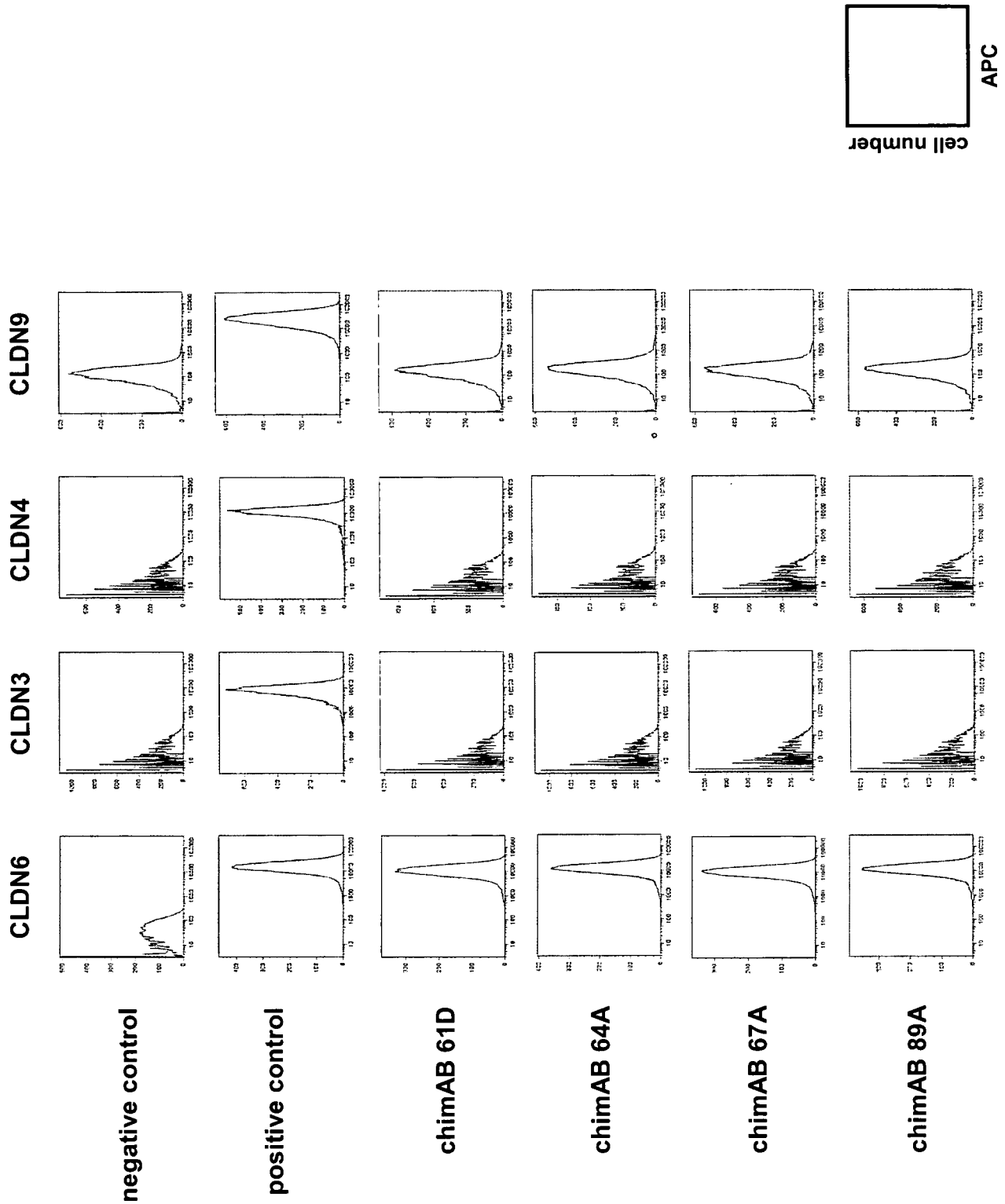

CLDN6
CLDN3
CLDN4
CLDN9
negative control
positive control
chimAB 61D
chimAB 64A
chimAB 67A
chimAB 89A
cell number
APC

Fig. 11

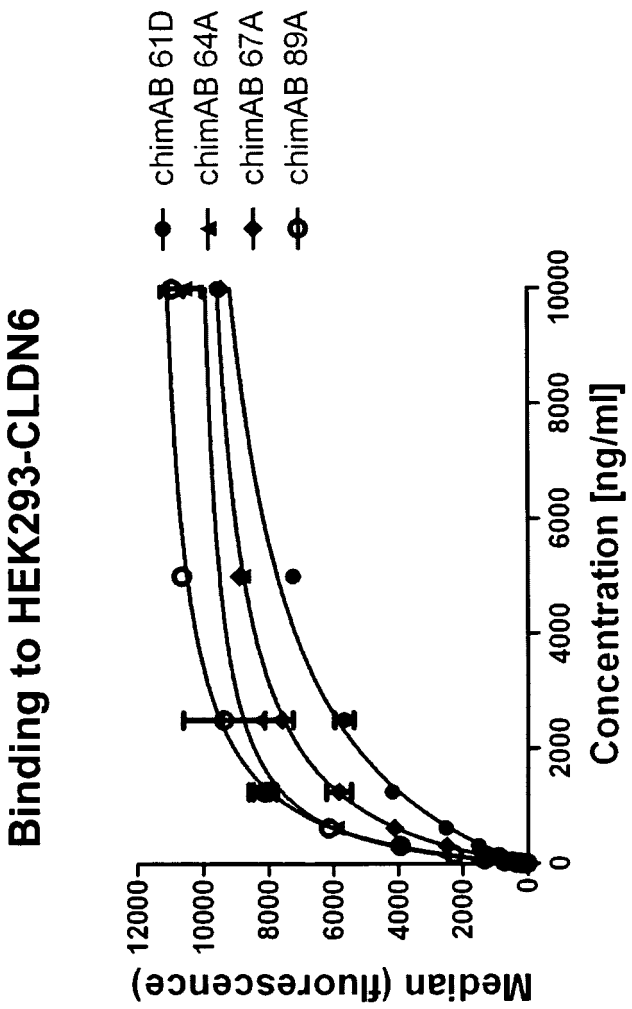

Binding to HEK293-CLDN6
chimAB 61D
chimAB 64A
chimAB 67A
chimAB 89A
Median (fluorescence)
0
2000
4000
6000
8000
10000
12000
Concentration [ng/ml]
0
2000
4000
6000
8000
10000

Fig. 12

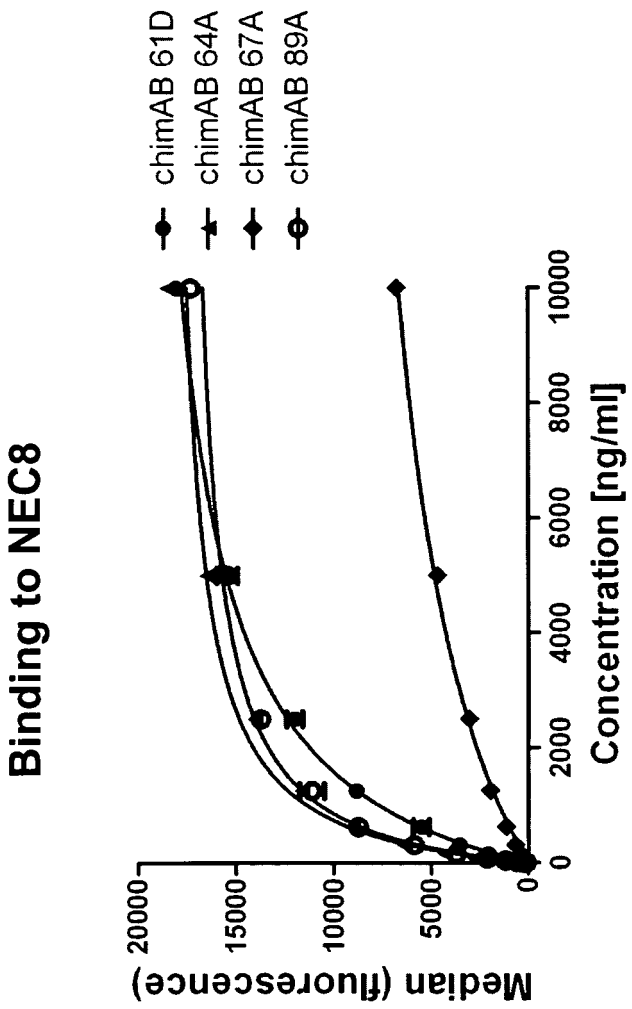

Binding to NEC8
chimAB 61D
chimAB 64A
chimAB 67A
chimAB 89A
Median (fluorescence)
20000
15000
10000
5000
0
0
2000
4000
6000
8000
10000
Concentration [ng/ml]

Fig. 13

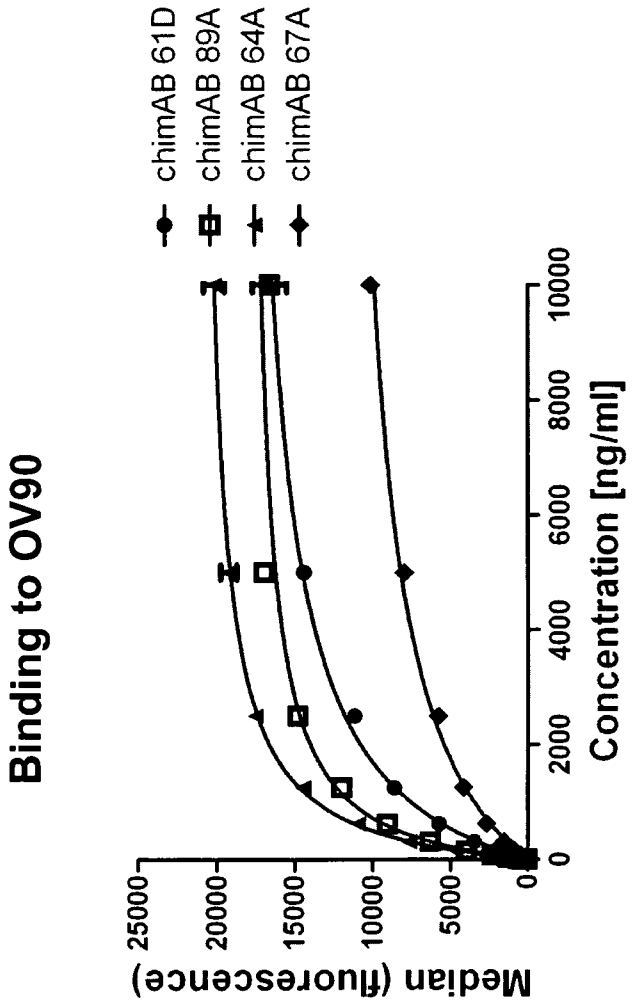

Binding to OV90
chimAB 61D
chimAB 89A
chimAB 64A
chimAB 67A
Median (fluorescence)
0
5000
10000
15000
20000
25000
Concentration [ng/ml]
0
2000
4000
6000
8000
10000

Fig. 14

Fig. 15

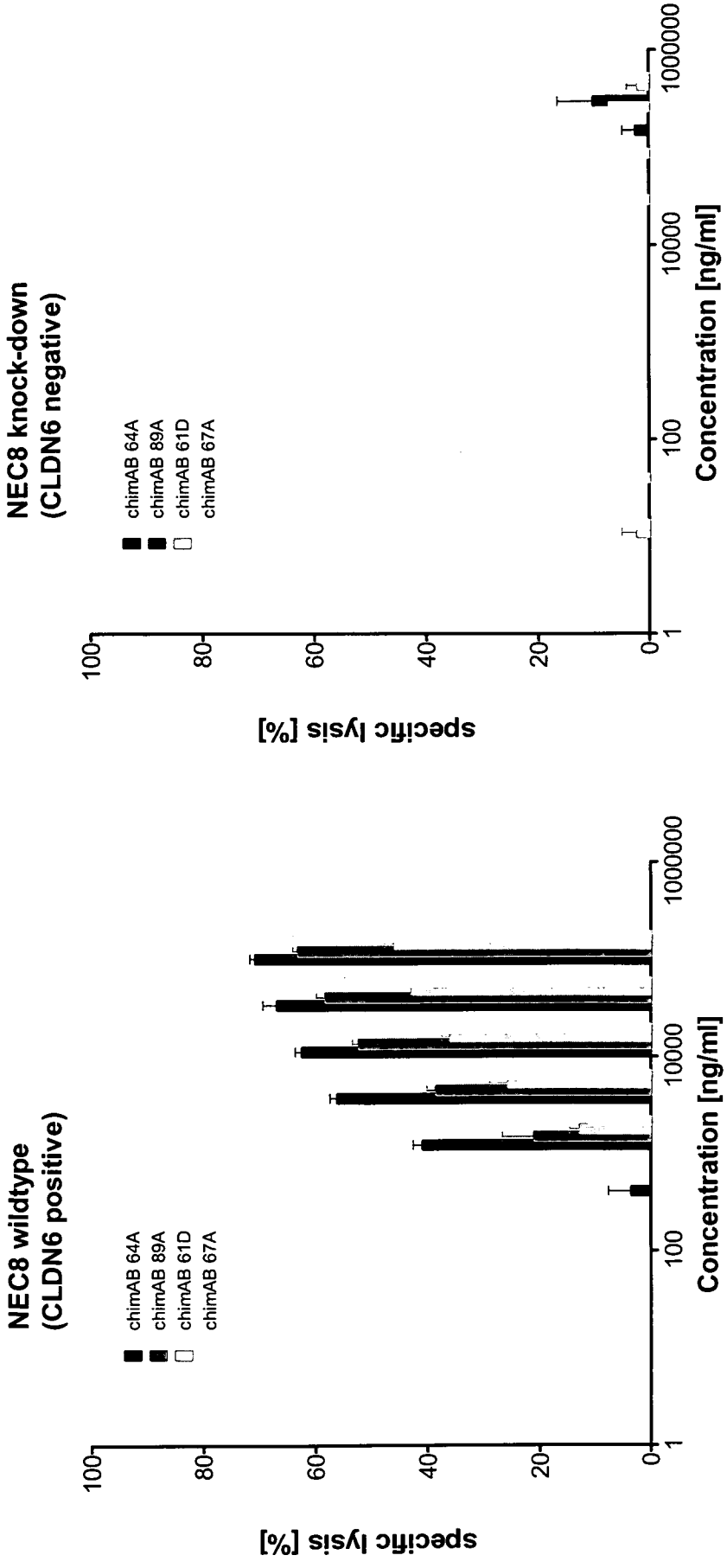

NEC8 wildtype
(CLDN6 positive)
chimAB 64A
chimAB 89A
chimAB 61D
chimAB 67A
specific lysis [%]
0
20
40
60
80
100
1
100
10000
1000000
Concentration [ng/ml]
NEC8 knock-down
(CLDN6 negative)
chimAB 64A
chimAB 89A
chimAB 61D
chimAB 67A
specific lysis [%]
0
20
40
60
80
100
1
100
10000
1000000
Concentration [ng/ml]

Fig. 16

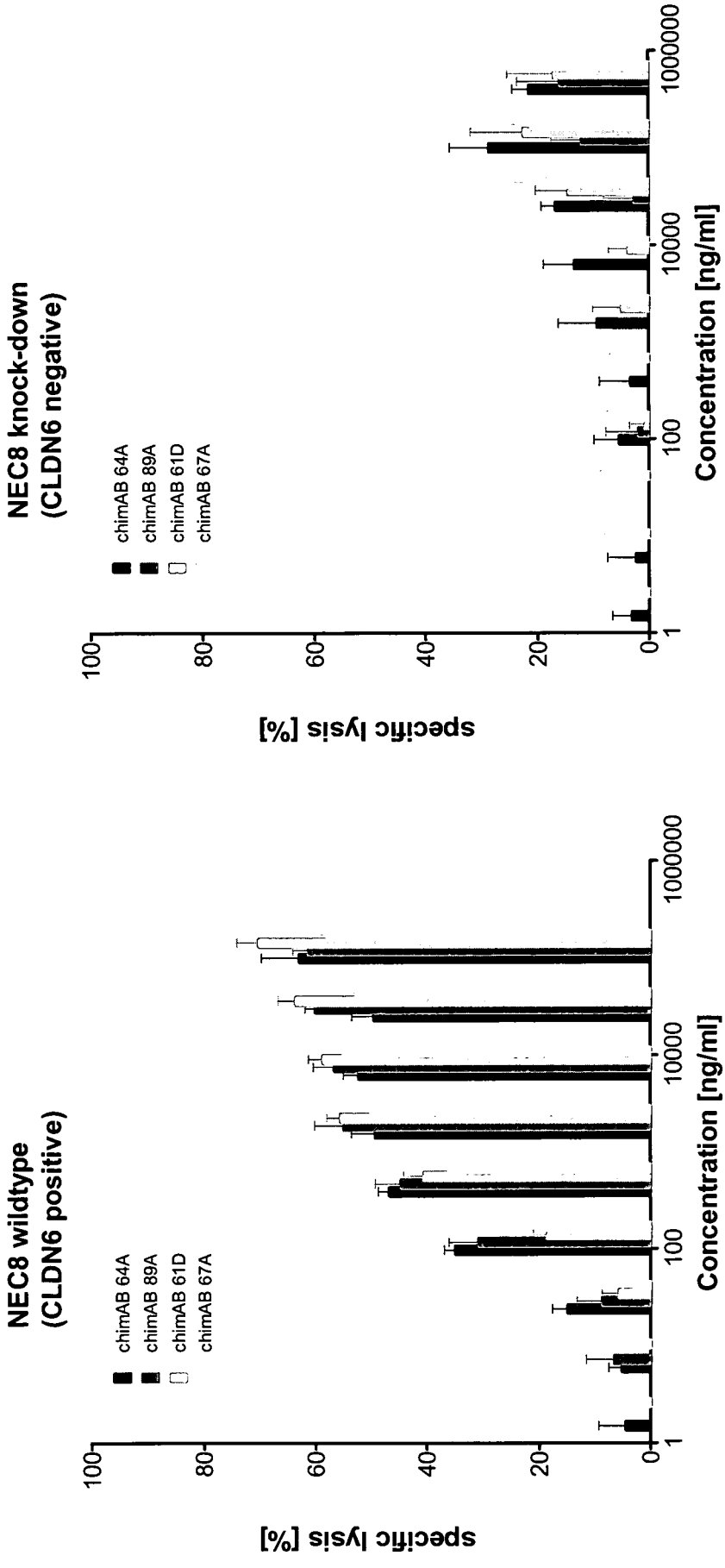

NEC8 knock-down
(CLDN6 negative)
chimAB 64A
chimAB 89A
chimAB 61D
chimAB 67A
specific lysis [%]
0
20
40
60
80
100
1
100
10000
1000000
Concentration [ng/ml]
NEC8 wildtype
(CLDN6 positive)
chimAB 64A
chimAB 89A
chimAB 61D
chimAB 67A
specific lysis [%]
0
20
40
60
80
100
1
100
10000
1000000
Concentration [ng/ml]

Fig. 17

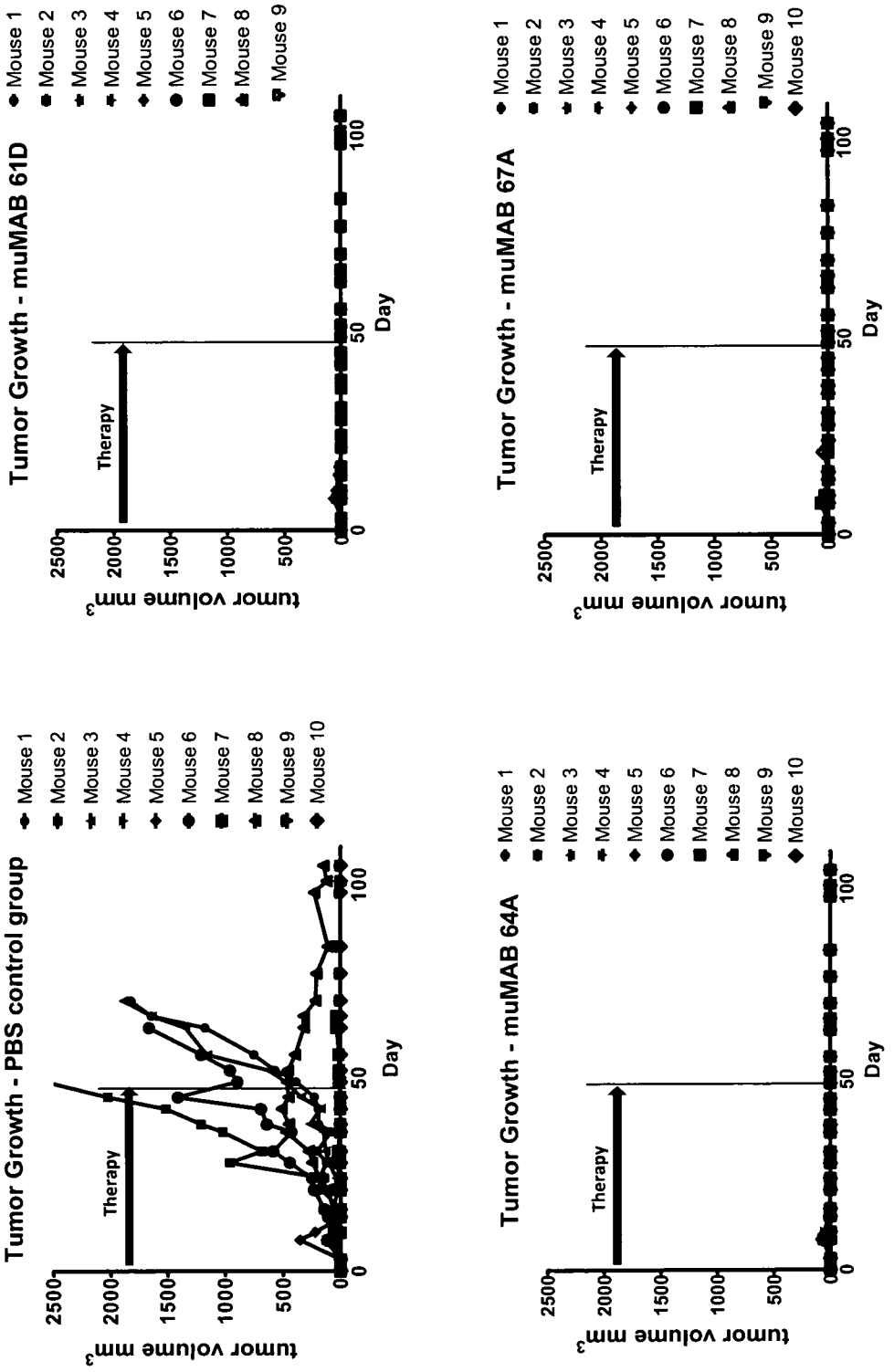

Tumor Growth - muMAB 61D
Mouse 1
Mouse 2
Mouse 3
Mouse 4
Mouse 5
Mouse 6
Mouse 7
Mouse 8
Mouse 9
Therapy
tumor volume mm³
2500
2000
1500
1000
500
0
0
50
100
Day
Tumor Growth - muMAB 67A
Mouse 1
Mouse 2
Mouse 3
Mouse 4
Mouse 5
Mouse 6
Mouse 7
Mouse 8
Mouse 9
Mouse 10
Therapy
tumor volume mm³
2500
2000
1500
1000
500
0
0
50
100
Day
Tumor Growth - PBS control group
Mouse 1
Mouse 2
Mouse 3
Mouse 4
Mouse 5
Mouse 6
Mouse 7
Mouse 8
Mouse 9
Mouse 10
Therapy
tumor volume mm³
2500
2000
1500
1000
500
0
0
50
100
Day
Tumor Growth - muMAB 64A
Mouse 1
Mouse 2
Mouse 3
Mouse 4
Mouse 5
Mouse 6
Mouse 7
Mouse 8
Mouse 9
Mouse 10
Therapy
tumor volume mm³
2500
2000
1500
1000
500
0
0
50
100
Day

Fig. 18

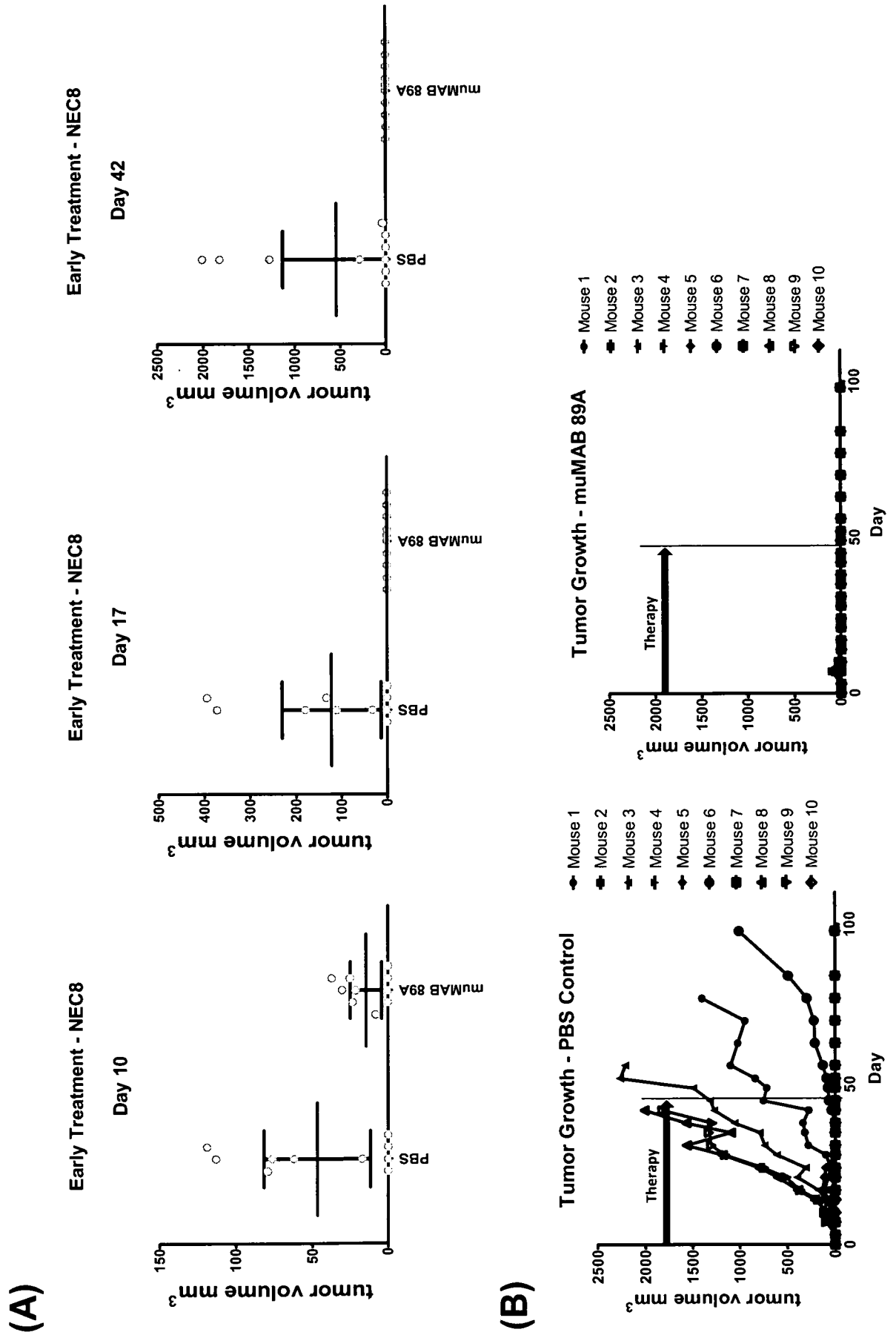

(A)
Early Treatment - NEC8
Day 10
tumor volume mm3
PBS
muMAB 89A
Early Treatment - NEC8
Day 17
tumor volume mm3
PBS
muMAB 89A
Early Treatment - NEC8
Day 42
tumor volume mm3
PBS
muMAB 89A
(B)
Tumor Growth - PBS Control
Therapy
tumor volume mm3
Day
Mouse 1
Mouse 2
Mouse 3
Mouse 4
Mouse 5
Mouse 6
Mouse 7
Mouse 8
Mouse 9
Mouse 10
Tumor Growth - muMAB 89A
Therapy
tumor volume mm3
Day
Mouse 1
Mouse 2
Mouse 3
Mouse 4
Mouse 5
Mouse 6
Mouse 7
Mouse 8
Mouse 9
Mouse 10

Fig. 19

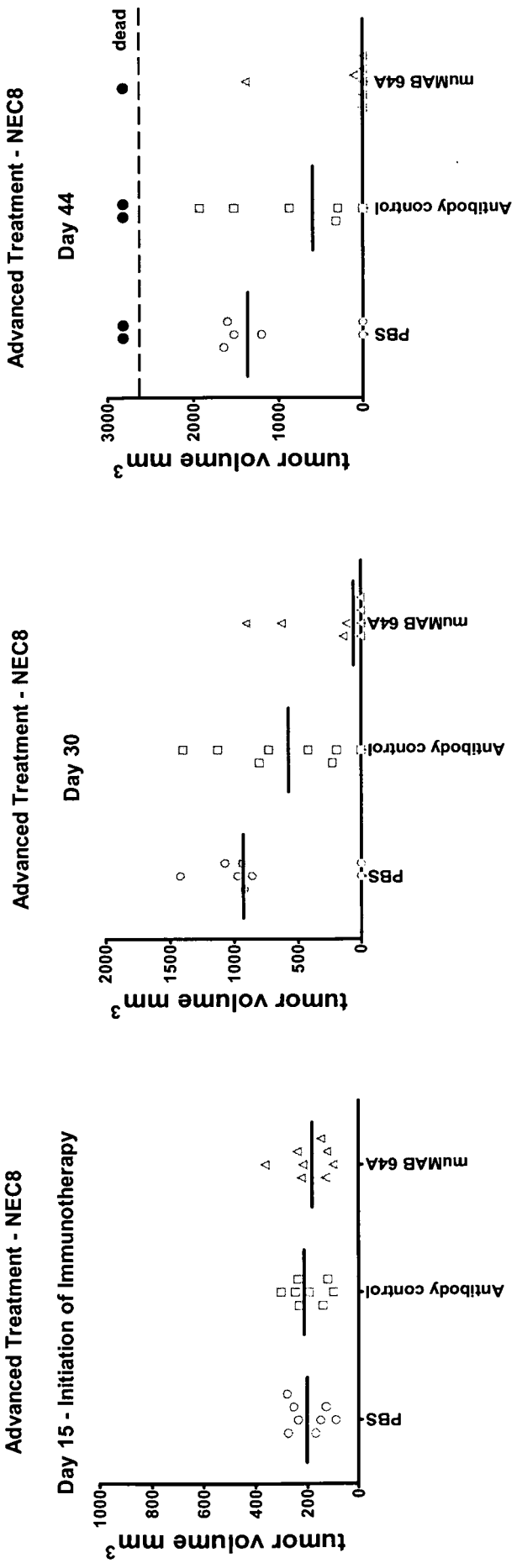
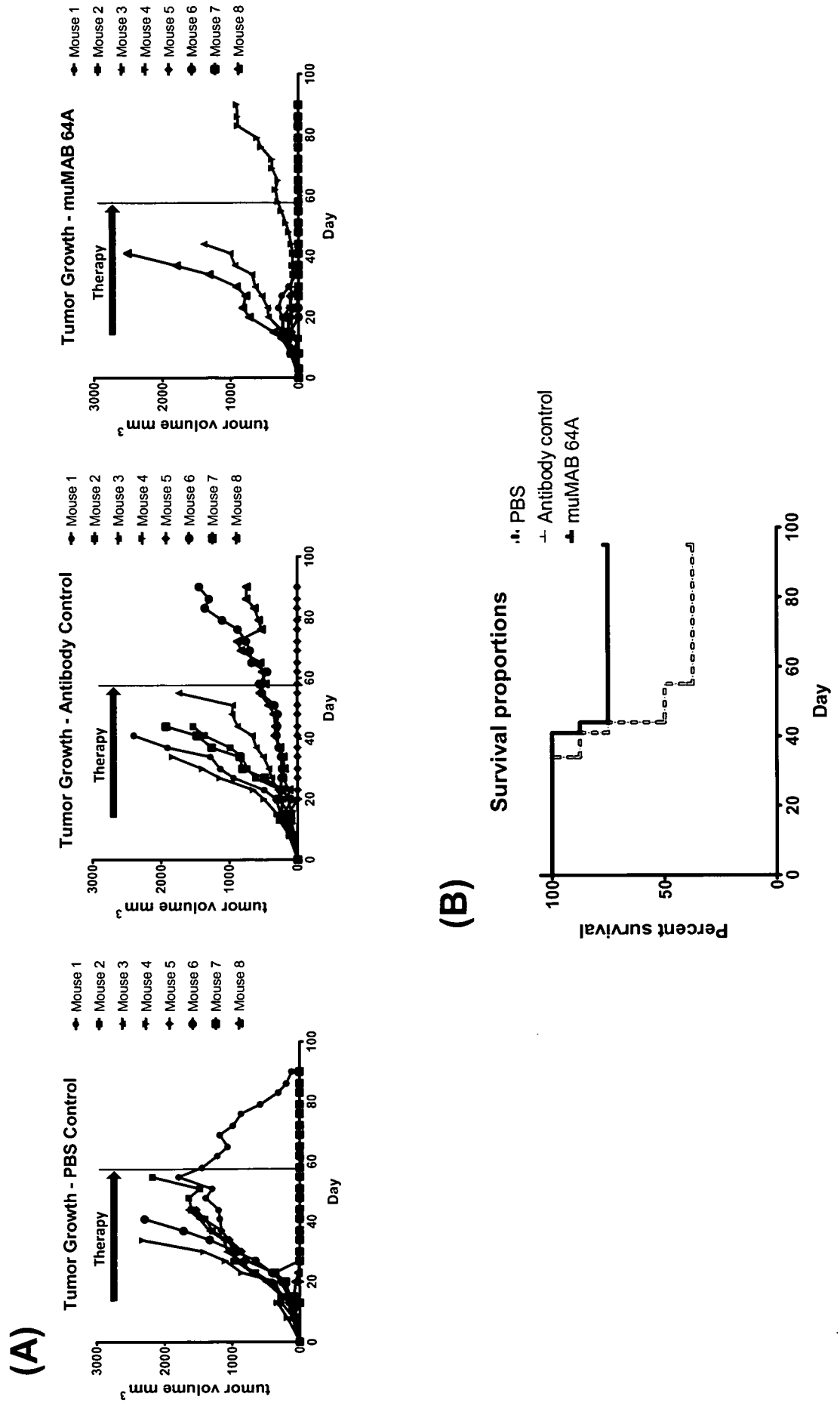

Advanced Treatment - NEC8
Day 15 - Initiation of Immunotherapy
tumor volume mm3
1000
800
600
400
200
0
PBS
Antibody control
muMAB 64A
Advanced Treatment - NEC8
Day 30
tumor volume mm3
2000
1500
1000
500
0
PBS
Antibody control
muMAB 64A
Advanced Treatment - NEC8
Day 44
tumor volume mm3
3000
2000
1000
0
dead
PBS
Antibody control
muMAB 64A

Fig. 20


(A)
Tumor Growth - PBS Control
Therapy
tumor volume mm3
Day
Mouse 1
Mouse 2
Mouse 3
Mouse 4
Mouse 5
Mouse 6
Mouse 7
Mouse 8
Tumor Growth - Antibody Control
Tumor Growth - muMAB 64A
(B)
Survival proportions
PBS
Antibody control
muMAB 64A
Percent survival

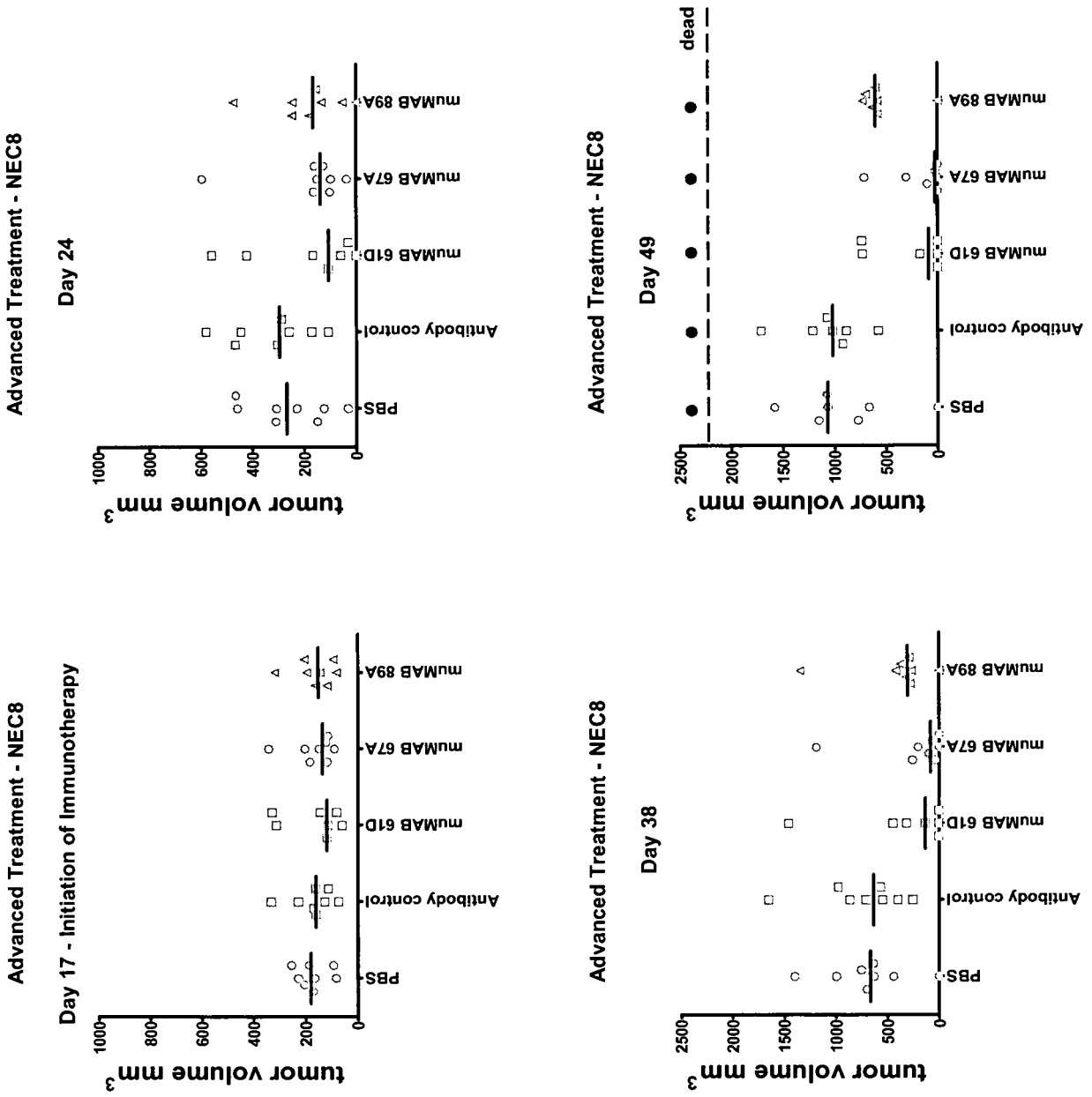

Advanced Treatment - NEC8
Day 17 - Initiation of Immunotherapy
Day 24
Day 38
Day 49
dead
tumor volume mm3
0
200
400
600
800
1000
500
1000
1500
2000
2500
PBS
Antibody control
muMAB 61D
muMAB 67A
muMAB 89A

Fig. 21

Fig. 22

(A)

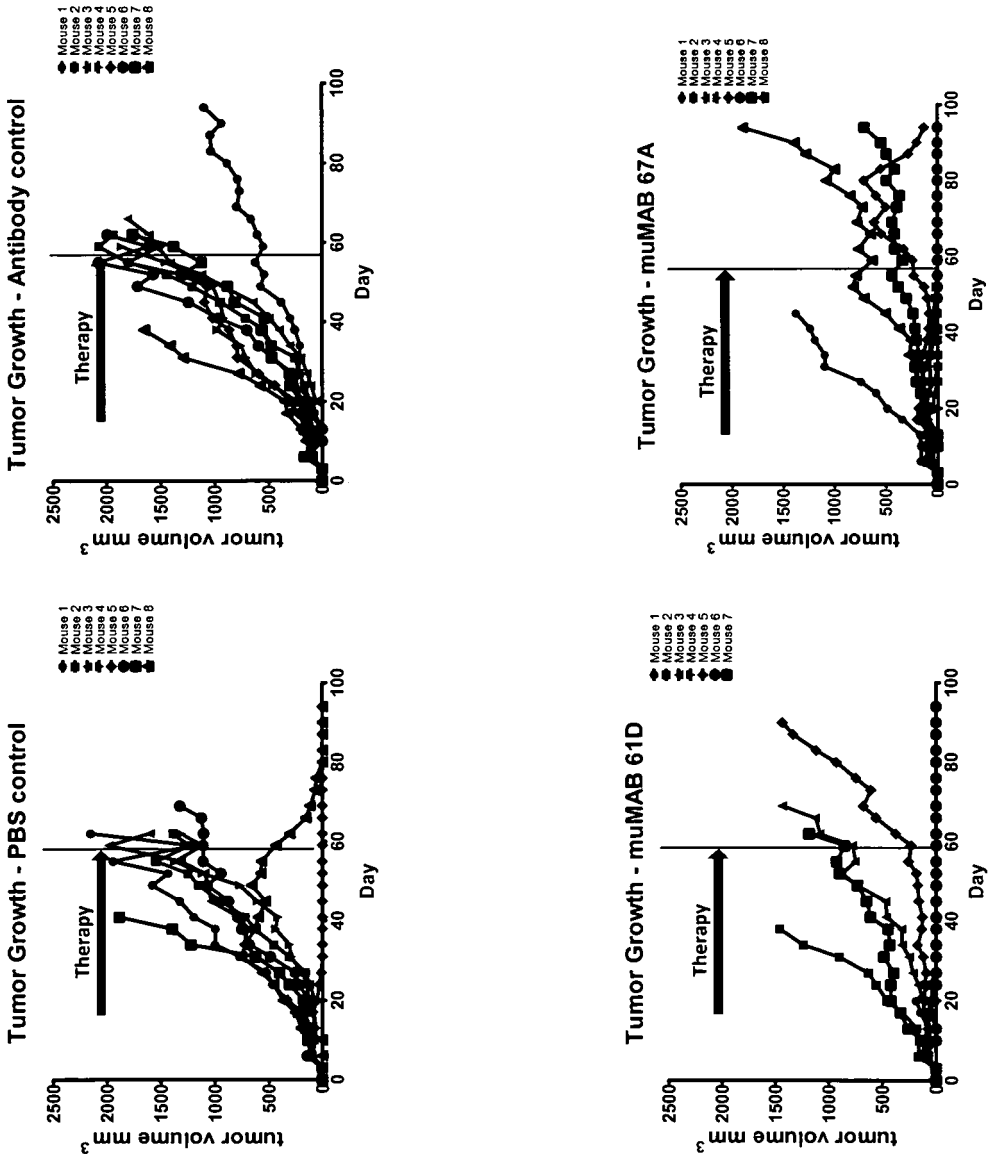

Tumor Growth - PBS control
Tumor Growth - Antibody control
Tumor Growth - muMAB 61D
Tumor Growth - muMAB 67A
Tumor Growth - muMAB 89A
Therapy
Day
tumor volume mm 3
Mouse 1
Mouse 2
Mouse 3
Mouse 4
Mouse 5
Mouse 6
Mouse 7
Mouse 8

Fig. 22

(B)

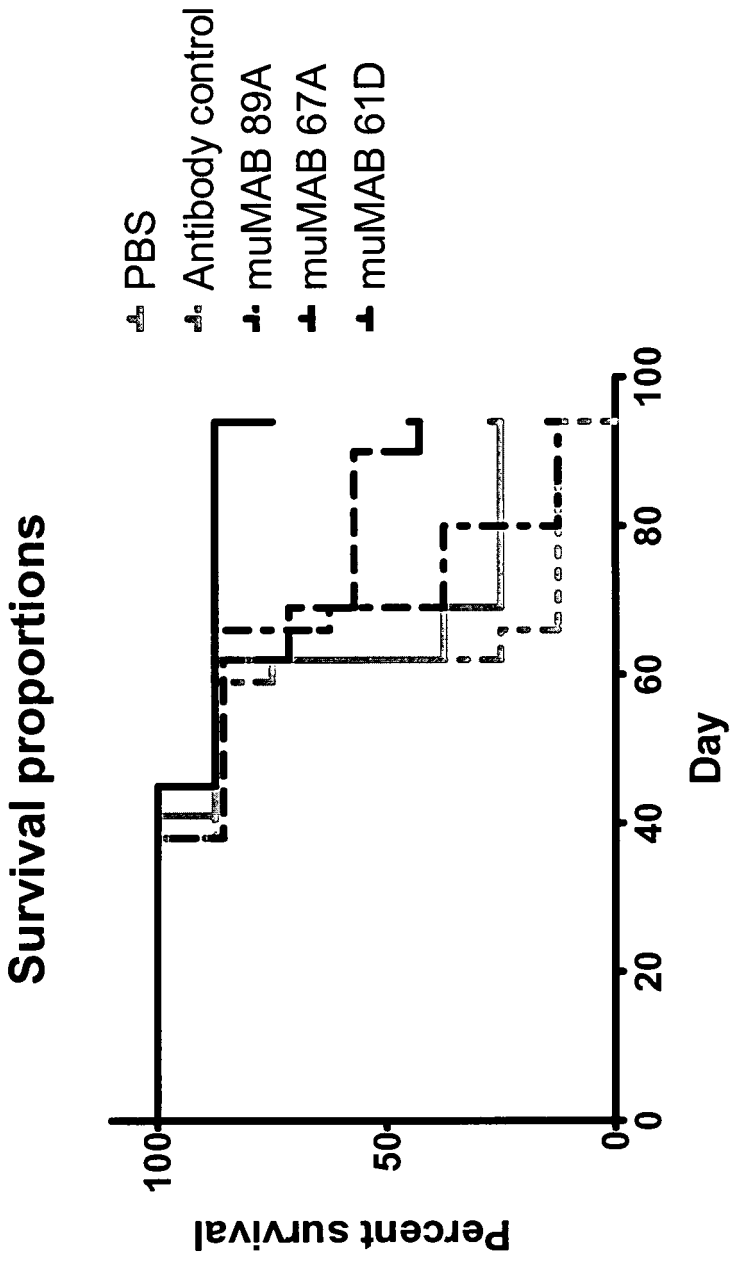

Survival proportions
PBS
Antibody control
muMAB 89A
muMAB 67A
muMAB 61D
Percent survival
100
50
0
0
20
40
60
80
100
Day

Fig. 23

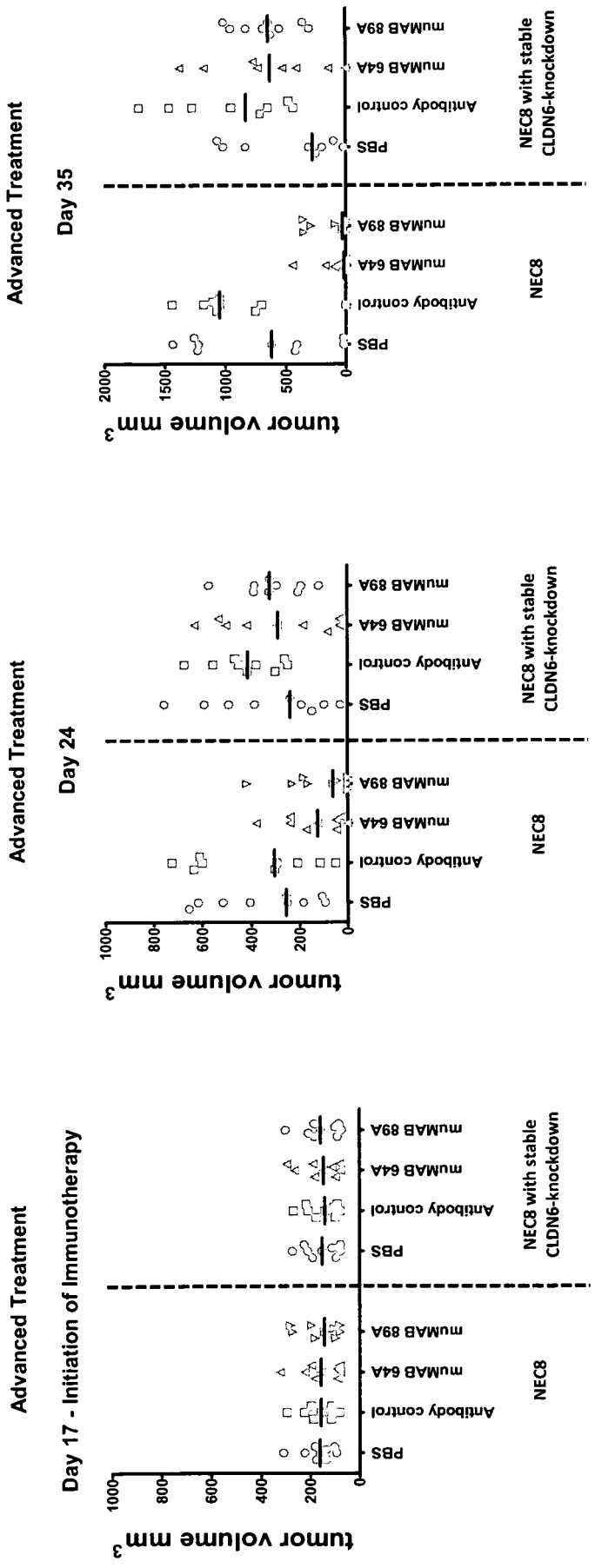
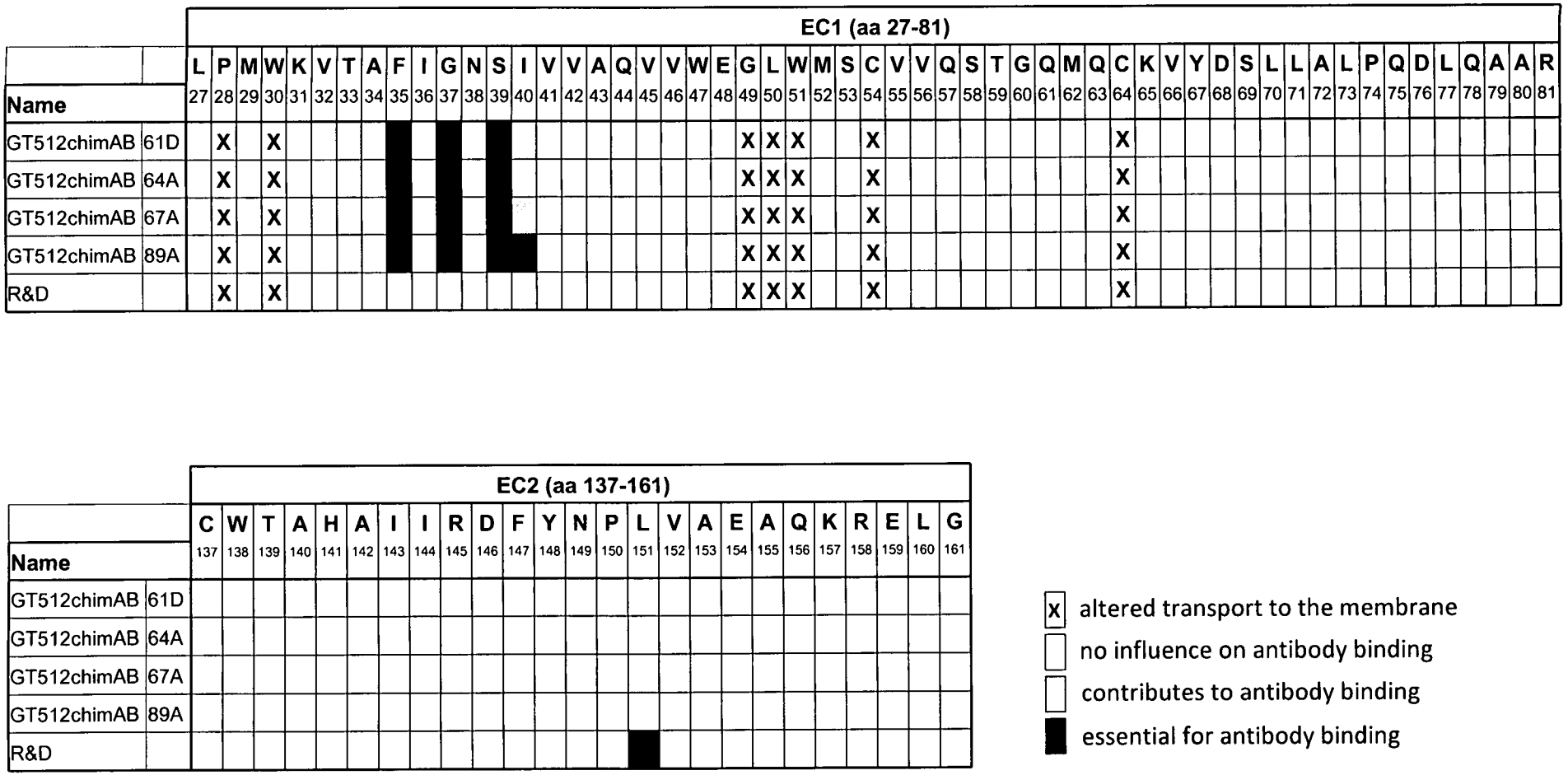

Advanced Treatment
Day 17 - Initiation of Immunotherapy
Day 24
Day 35
tumor volume mm3
0
200
400
600
800
1000
500
1000
1500
2000
PBS
Antibody control
muMAB 64A
muMAB 89A
NEC8
NEC8 with stable CLDN6-knockdown

Fig. 24



WO 2012/156018 PCT/EP2012/001721

Fig. 25

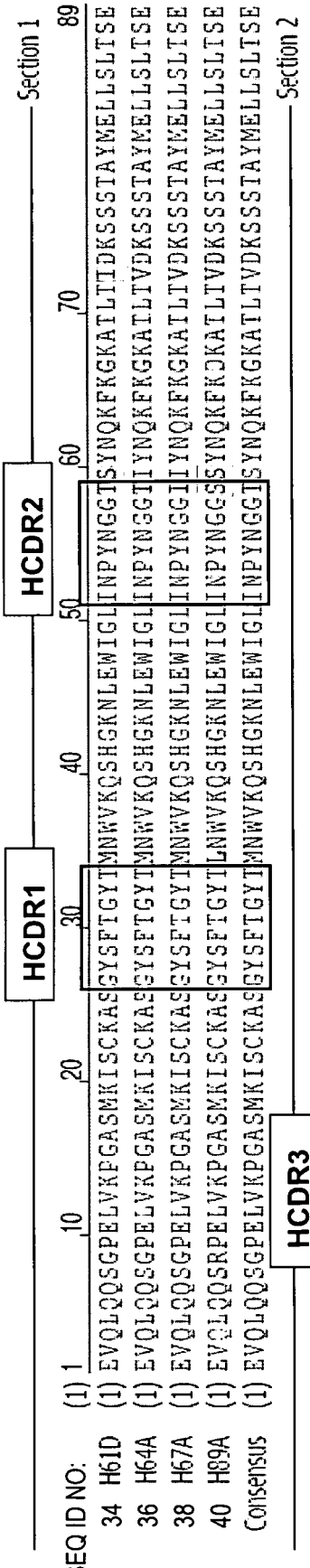

HCDR1
HCDR2
HCDR3
Section 1
Section 2
SEQ ID NO:
34 H61D
36 H64A
38 H67A
40 H89A
Consensus
(1) EVQLQQSGPELVKPGASMKISCKASGYSFTGYTMNWVKQSHGKNLEWIGLINPYNGGTSYNQKFKGKATLTVDKSSSTAYMELLSLTSE
(911) DSAVYYCARDYGYVLDYWGQGTTLTVSS

Fig. 26

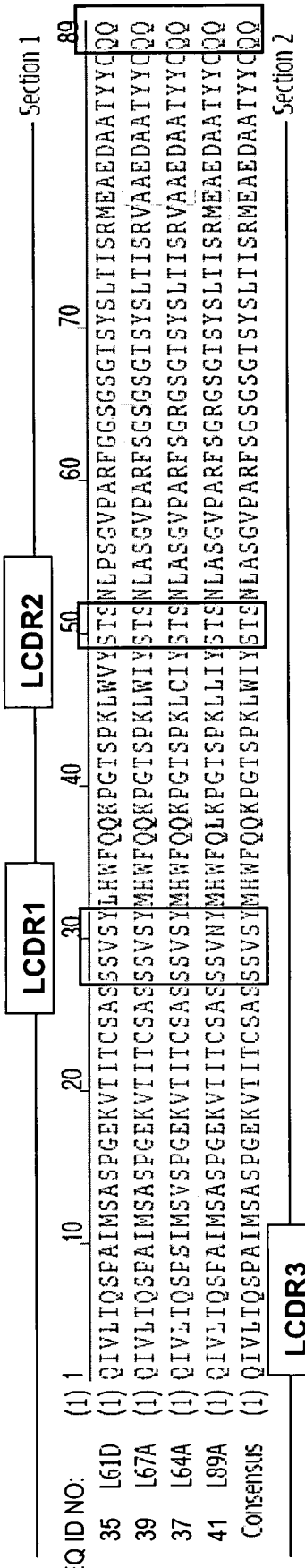
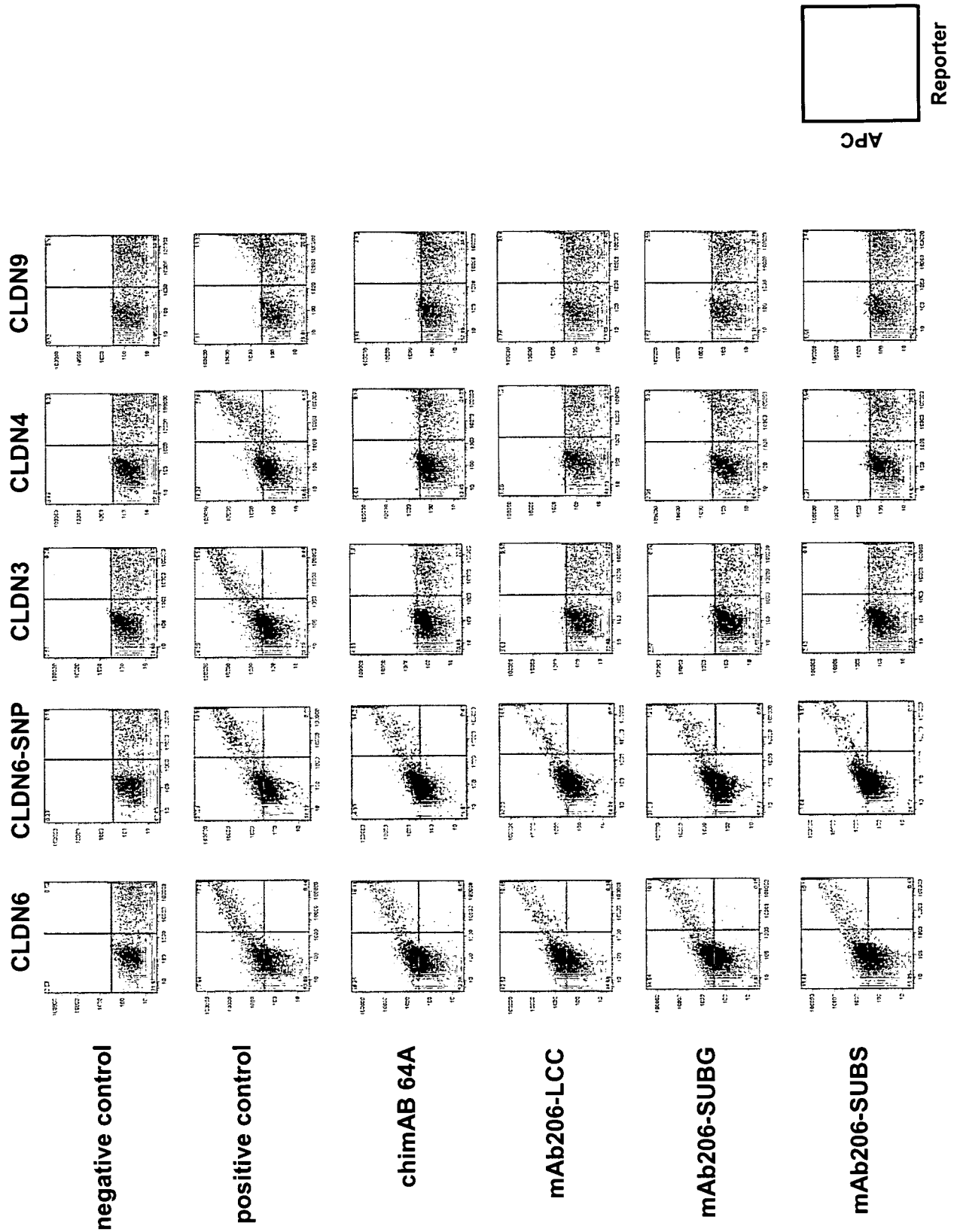

Section 1
Section 2
LCDR1
LCDR2
LCDR3
SEQ ID NO:
35 L61D
39 L67A
37 L64A
41 L89A
Consensus
(1) QIVLTQSPAIMSASPGEKVTITCSAS SSVSY LHWFQQKPGTSPKLWVY STS NLPSGVPARFGGSGSGTSYSLTISRMEAEDAATYYC QQ
(1) QIVLTQSPAIMSASPGEKVTITCSAS SSVSY MHWFQQKPGTSPKLWIY STS NLASGVPARFSGSGSGTSYSLTISRVAAEDAATYYC QQ
(1) QIVLTQSPSIMSVSPGEKVTITCSAS SSVSY MHWFQQKPGTSPKLCIY STS NLASGVPARFSGRGSGTSYSLTISRVAAEDAATYYC QQ
(1) QIVLTQSPAIMSASPGEKVTITCSAS SSVNY MHWFQLKPGTSPKLLIY STS NLASGVPARFSGRGSGTSYSLTISRMEAEDAATYYC QQ
(1) QIVLTQSPAIMSASPGEKVTITCSAS SSVSY MHWFQQKPGTSPKLWIY STS NLASGVPARFSGSGSGTSYSLTISRMEAEDAATYYC QQ
(90) RSIYPPWT FGGGTKLEIK
(90) RSTYPPWT FGGGTKLEIK
(90) RSNYPPWT FGGGTKLEIK
(90) RNNYPPWT FGGGTKLEIK
(90) RSNYPPWT FGGGTKLEIK

Fig. 27


CLDN6
CLDN6-SNP
CLDN3
CLDN4
CLDN9
negative control
positive control
chimAB 64A
mAb206-LCC
mAb206-SUBG
mAb206-SUBS
APC
Reporter

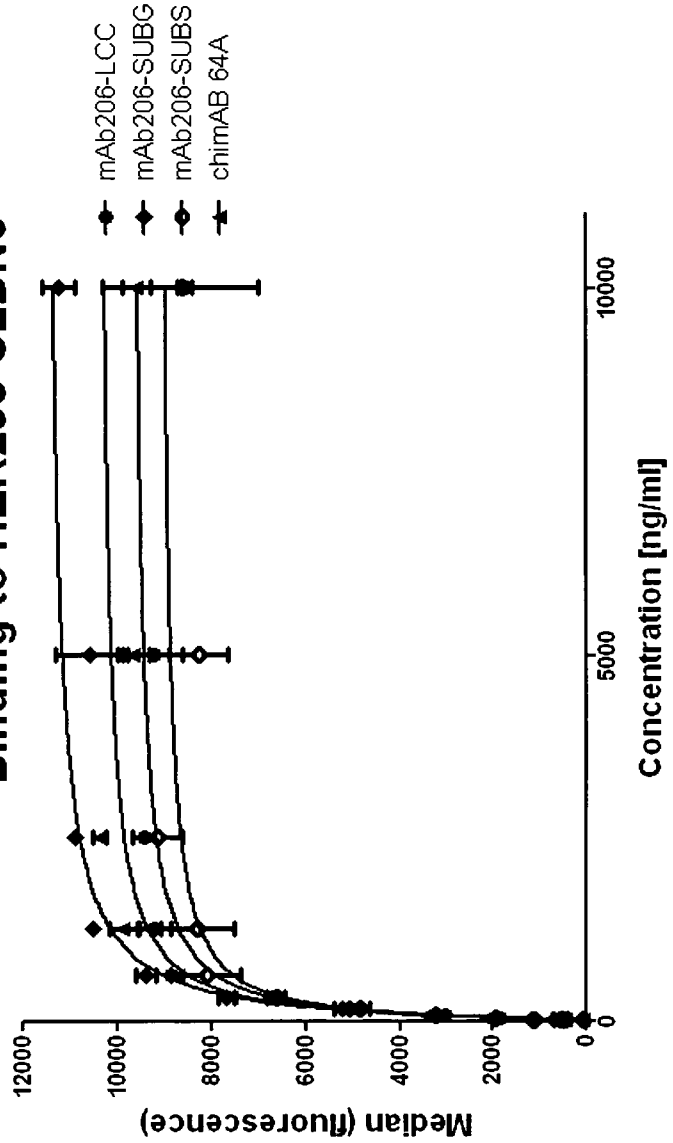

Binding to HEK293-CLDN6
mAb206-LCC
mAb206-SUBG
mAb206-SUBS
chimAB 64A
Median (fluorescence)
12000
10000
8000
6000
4000
2000
0
0
5000
10000
Concentration [ng/ml]

Fig. 28

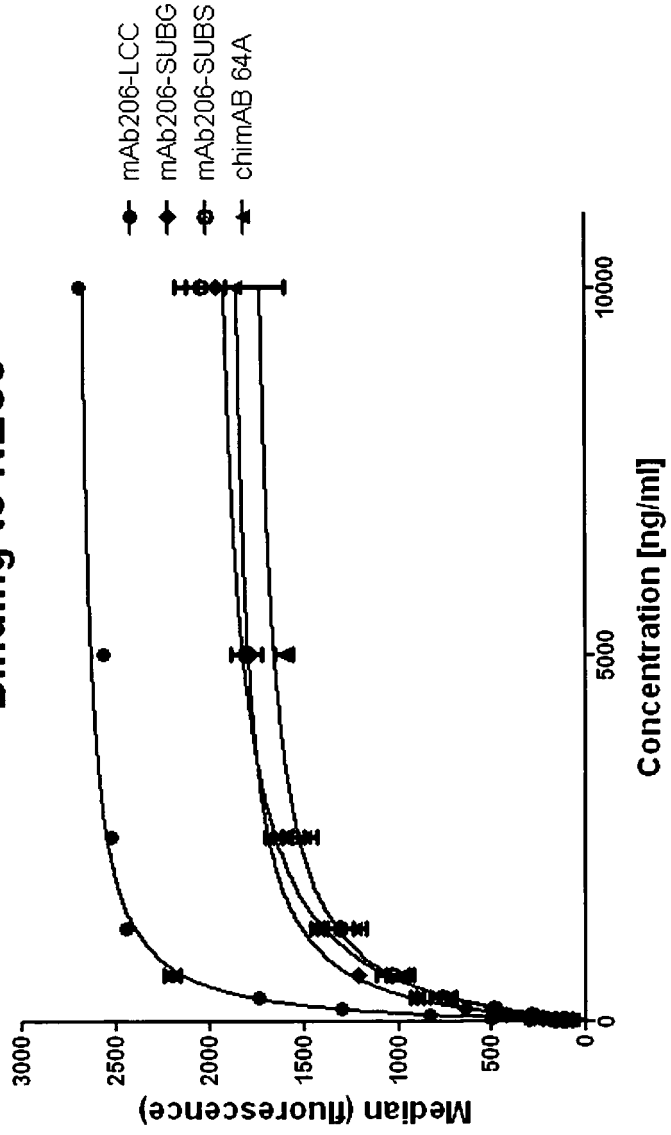

Binding to NEC8
mAb206-LCC
mAb206-SUBG
mAb206-SUBS
chimAB 64A
Median (fluorescence)
3000
2500
2000
1500
1000
500
0
0
5000
10000
Concentration [ng/ml]

Fig. 29

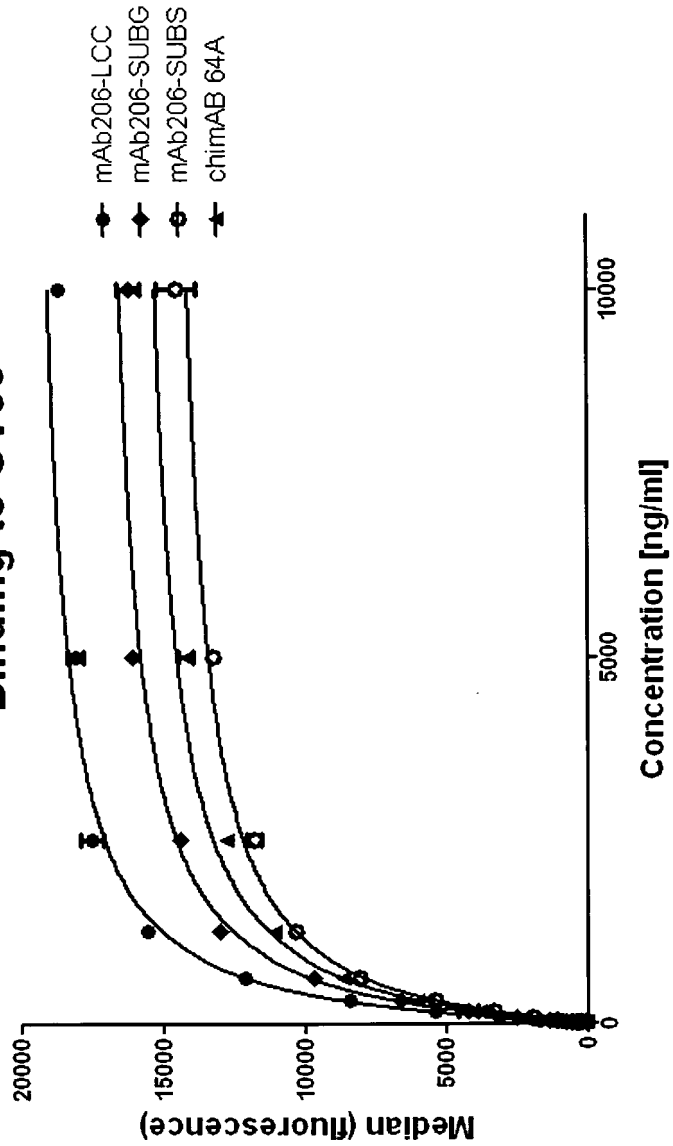
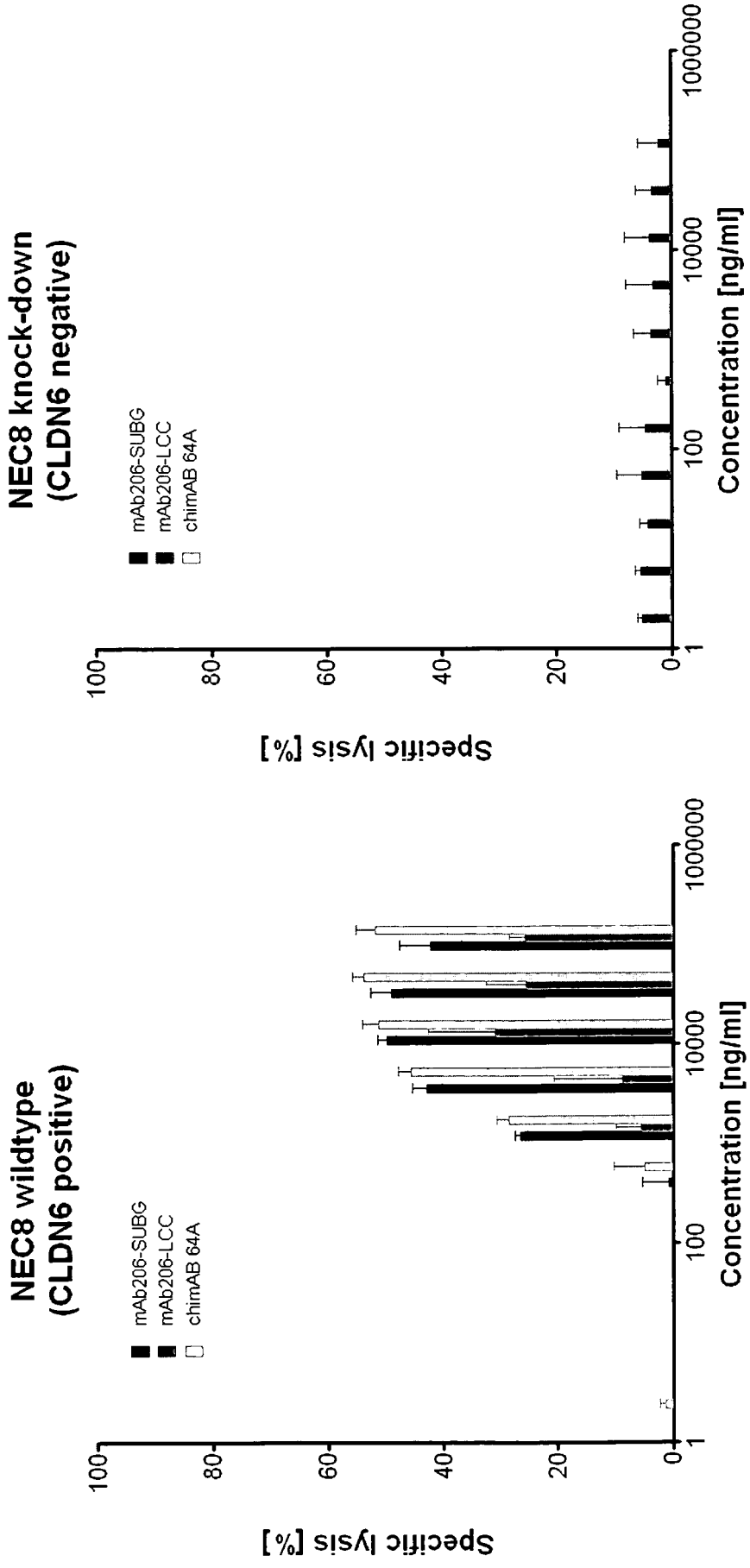

Binding to OV90
mAb206-LCC
mAb206-SUBG
mAb206-SUBS
chimAB 64A
Median (fluorescence)
20000
15000
10000
5000
0
0
5000
10000
Concentration [ng/ml]

Fig. 30

Fig. 31a


NEC8 knock-down
(CLDN6 negative)
mAb206-SUBG
mAb206-LCC
chimAB 64A
Specific lysis [%]
Concentration [ng/ml]
NEC8 wildtype
(CLDN6 positive)
mAb206-SUBG
mAb206-LCC
chimAB 64A
Specific lysis [%]
Concentration [ng/ml]

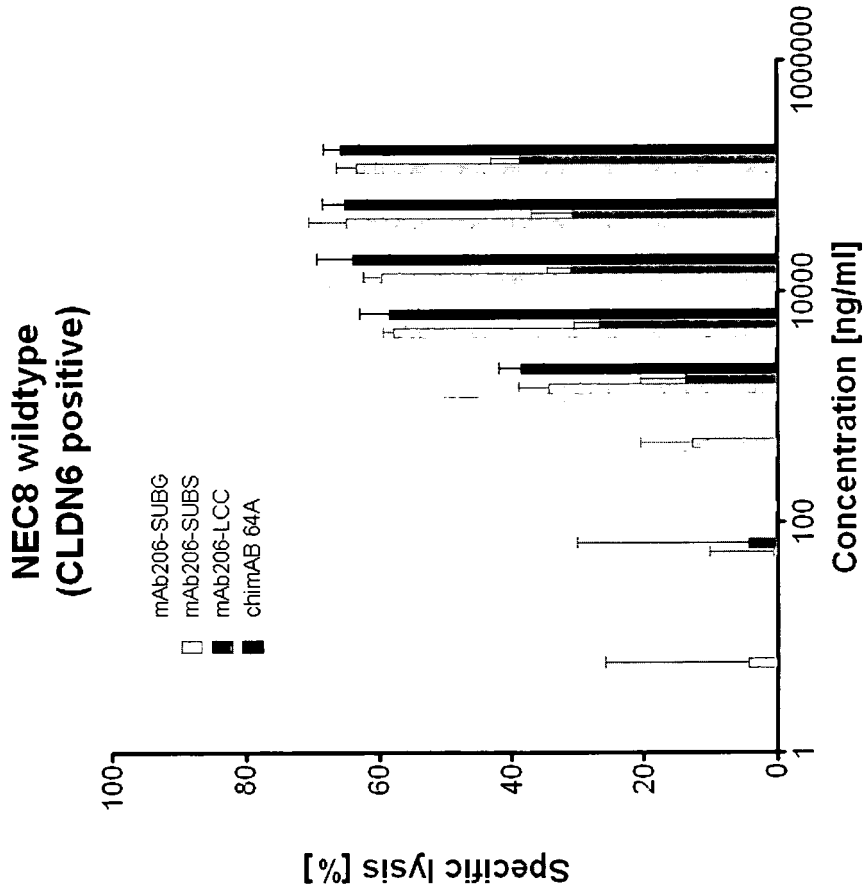

NEC8 wildtype
(CLDN6 positive)
mAb206-SUBG
mAb206-SUBS
mAb206-LCC
chimAB 64A
Specific lysis [%]
0
20
40
60
80
100
1
100
10000
1000000
Concentration [ng/ml]

Fig. 31b

Fig. 32a

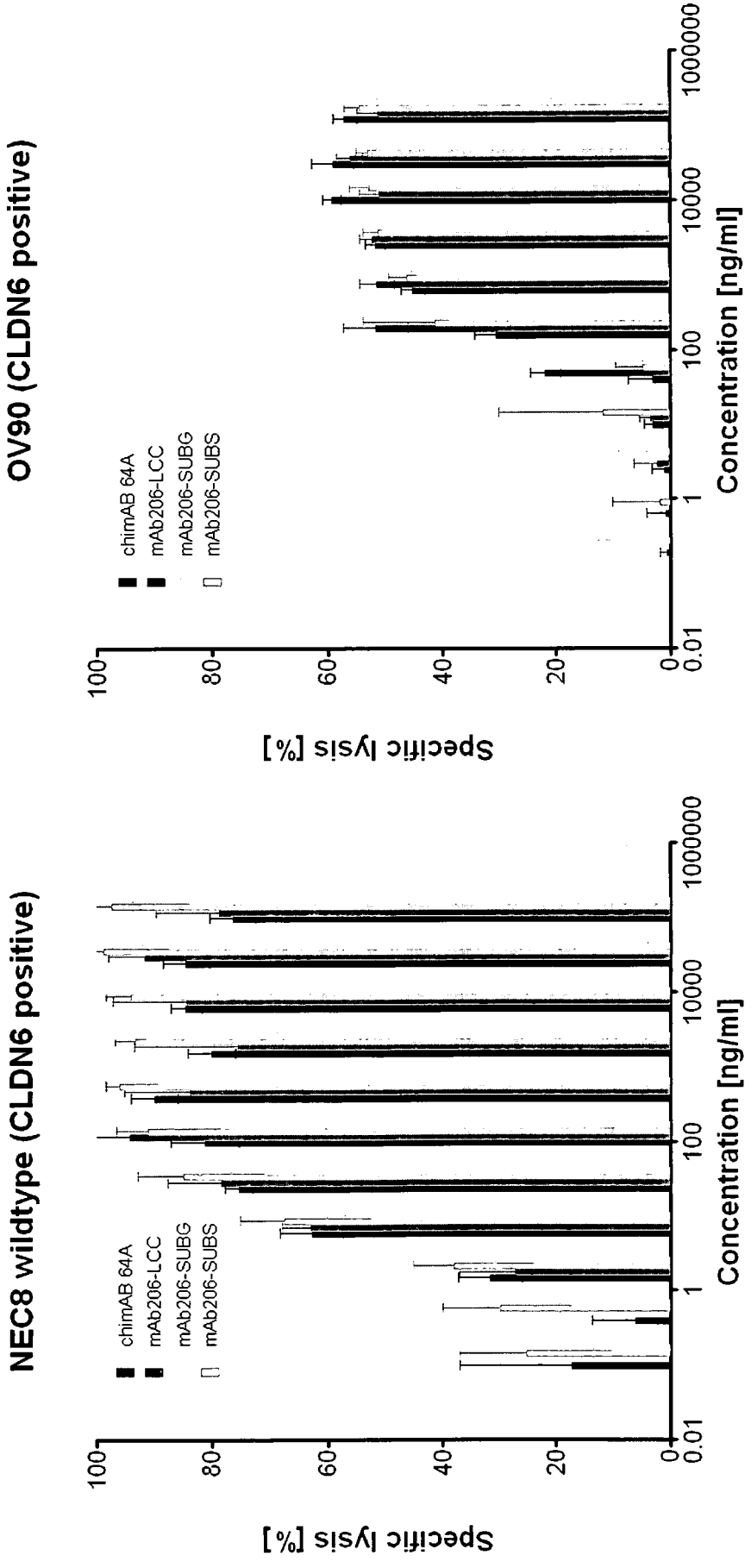

NEC8 wildtype (CLDN6 positive)
OV90 (CLDN6 positive)
chimAB 64A
mAb206-LCC
mAb206-SUBG
mAb206-SUBS
Specific lysis [%]
Concentration [ng/ml]
0
20
40
60
80
100
0.01
1
100
10000
1000000

Fig. 32b

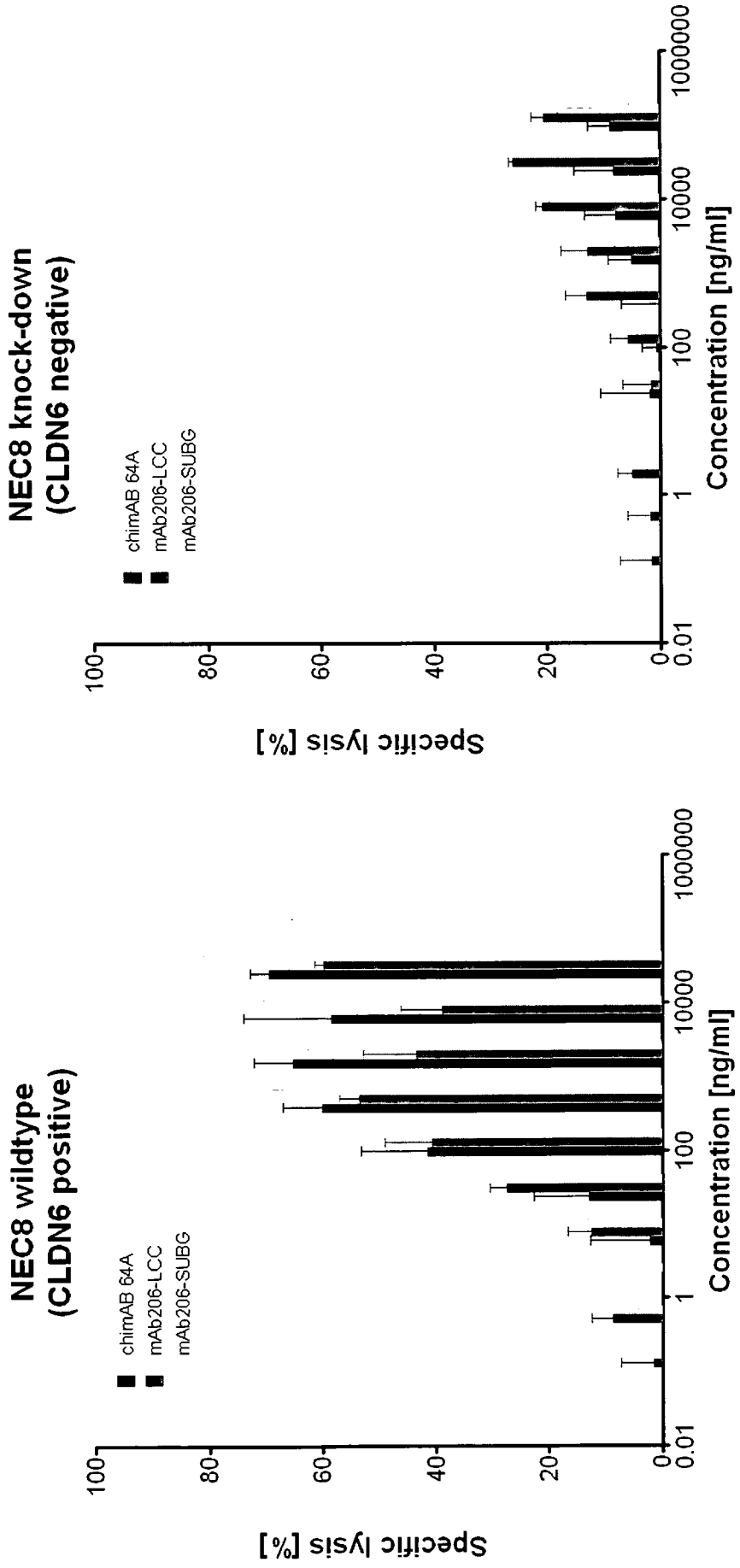

NEC8 knock-down
(CLDN6 negative)
chimAB 64A
mAb206-LCC
mAb206-SUBG
Specific lysis [%]
Concentration [ng/ml]
NEC8 wildtype
(CLDN6 positive)
chimAB 64A
mAb206-LCC
mAb206-SUBG
Specific lysis [%]
Concentration [ng/ml]

Fig. 33

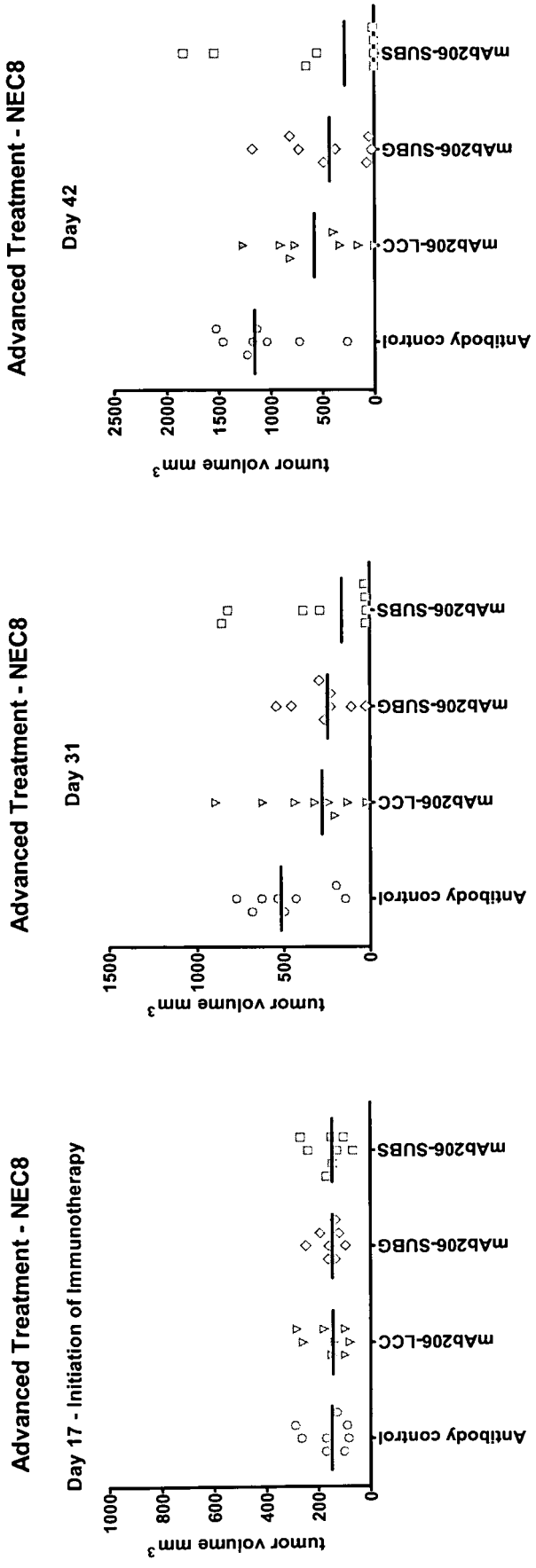
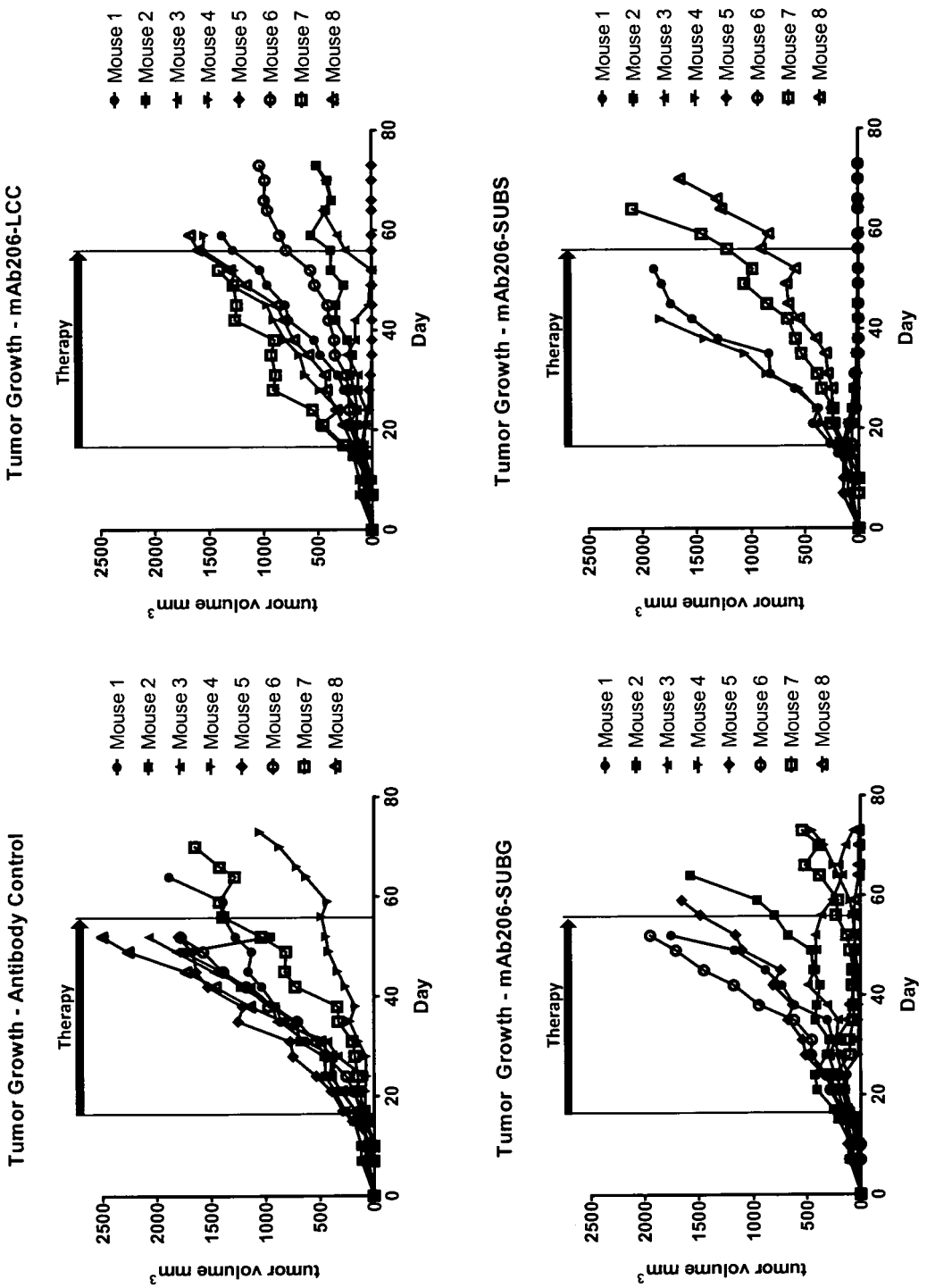

Advanced Treatment - NEC8
Day 17 - Initiation of Immunotherapy
tumor volume mm3
1000
800
600
400
200
0
Antibody control
mAb206-LCC
mAb206-SUBG
mAb206-SUBS
Advanced Treatment - NEC8
Day 31
tumor volume mm3
1500
1000
500
0
Antibody control
mAb206-LCC
mAb206-SUBG
mAb206-SUBS
Advanced Treatment - NEC8
Day 42
tumor volume mm3
2500
2000
1500
1000
500
0
Antibody control
mAb206-LCC
mAb206-SUBG
mAb206-SUBS

Fig. 34a


Tumor Growth - Antibody Control
Tumor Growth - mAb206-LCC
Tumor Growth - mAb206-SUBG
Tumor Growth - mAb206-SUBS
Therapy
Day
tumor volume mm3
Mouse 1
Mouse 2
Mouse 3
Mouse 4
Mouse 5
Mouse 6
Mouse 7
Mouse 8
0
500
1000
1500
2000
2500
0
20
40
60
80

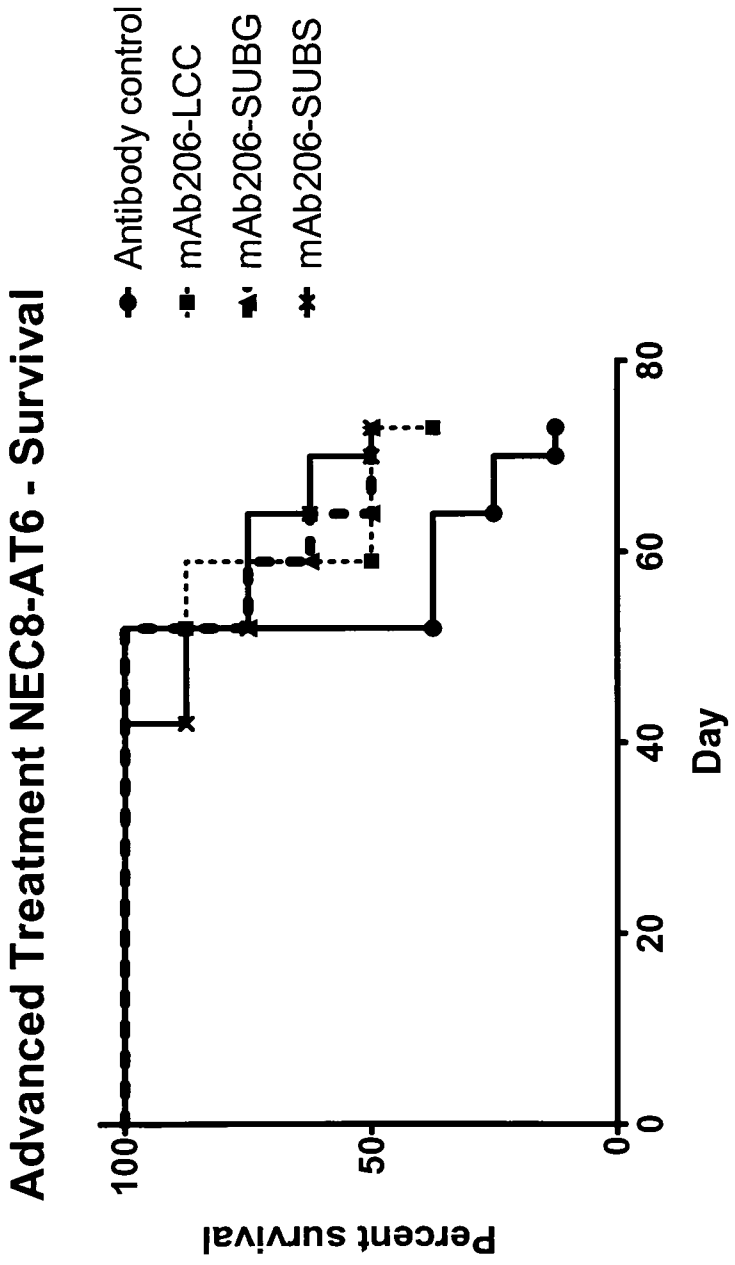
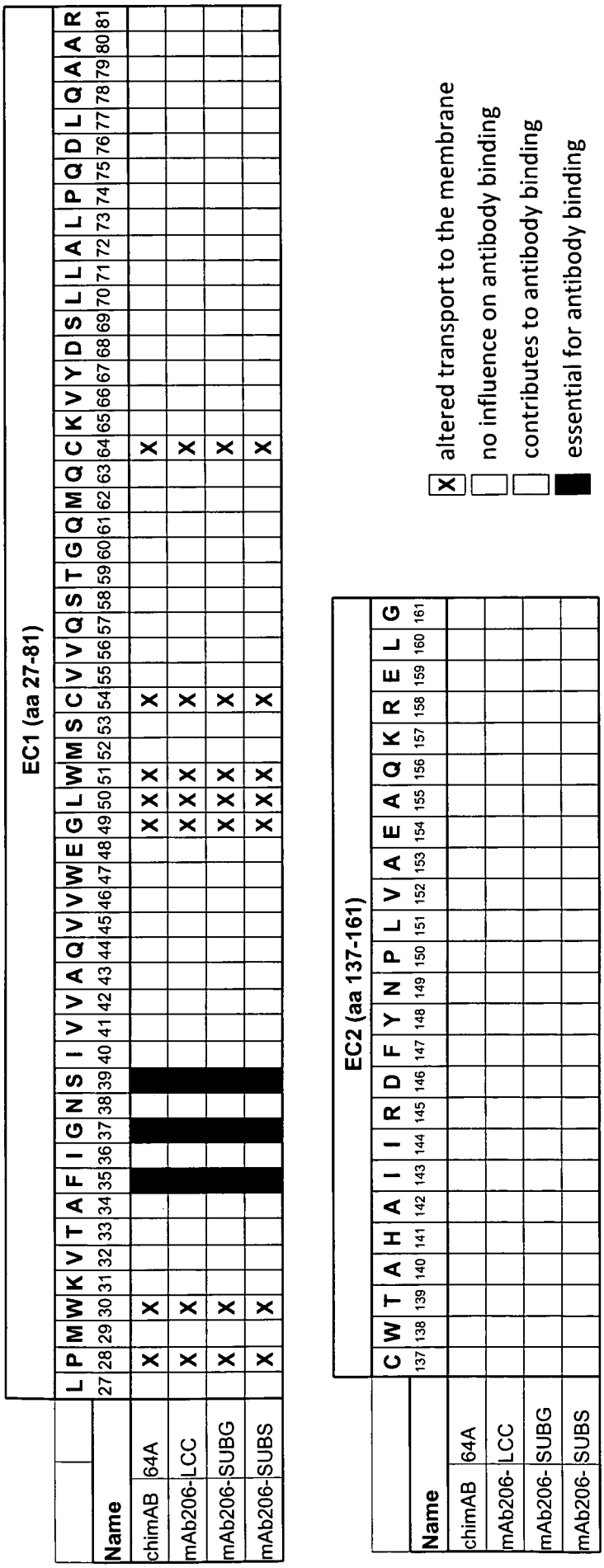

Advanced Treatment NEC8-AT6 - Survival
Antibody control
mAb206-LCC
mAb206-SUBG
mAb206-SUBS
Percent survival
100
50
0
0
20
40
60
80
Day

Fig. 34b

Fig. 35


EC1 (aa 27-81)
EC2 (aa 137-161)
Name
chimAB 64A
mAb206-LCC
mAb206-SUBG
mAb206-SUBS
altered transport to the membrane
no influence on antibody binding
contributes to antibody binding
essential for antibody binding

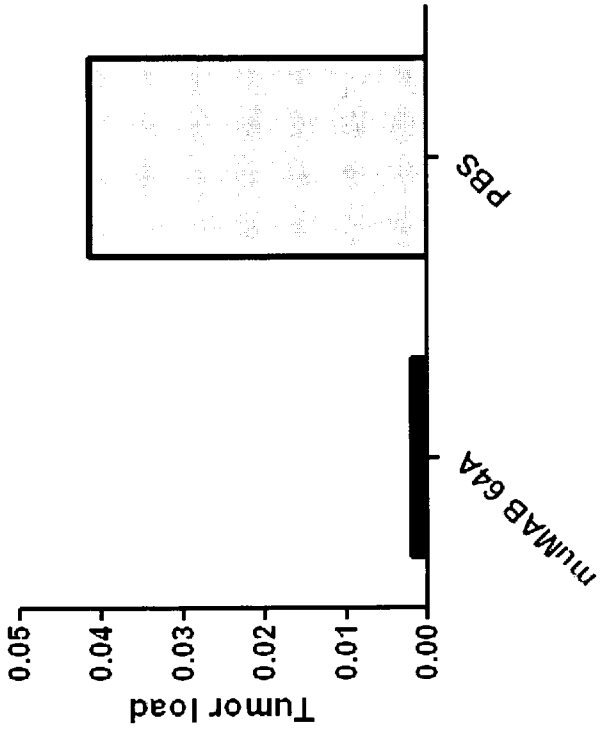


Fig. 36

Fig. 37

	mAb206-LCC	mAb206-SUBG	muMAB 64A
Cancer tissues			
OvarCA	2/2	2/2	2/2
TestisCA	1/1	1/1	4/4
Normal tissues			
Breast	0/1	0/1	-
Colon	0/1	0/1	0/4
Duodenum	0/1	0/1	-
Gall bladder	0/1	0/1	-
Heart	0/1	0/1	0/1
Kidney	0/1	0/1	0/4
Liver	0/1	0/1	0/2
Lung	0/1	0/1	-
Lymphnode	0/1	0/1	-
Oesophagus	0/1	0/1	0/1
Ovar	0/1	0/1	-
Pancreas	0/1	0/1	0/2
Placenta	0/1	0/1	-
Prostate	0/1	0/1	0/2
Sceletal muscle	0/1	0/1	-
Spleen	0/1	0/1	-
Stomach	0/1	0/1	0/2
Testis	0/4	0/4	0/2
Tube	0/1	0/1	-
Thyroidea	0/1	0/1	-
Uterus	0/2	0/2	-