

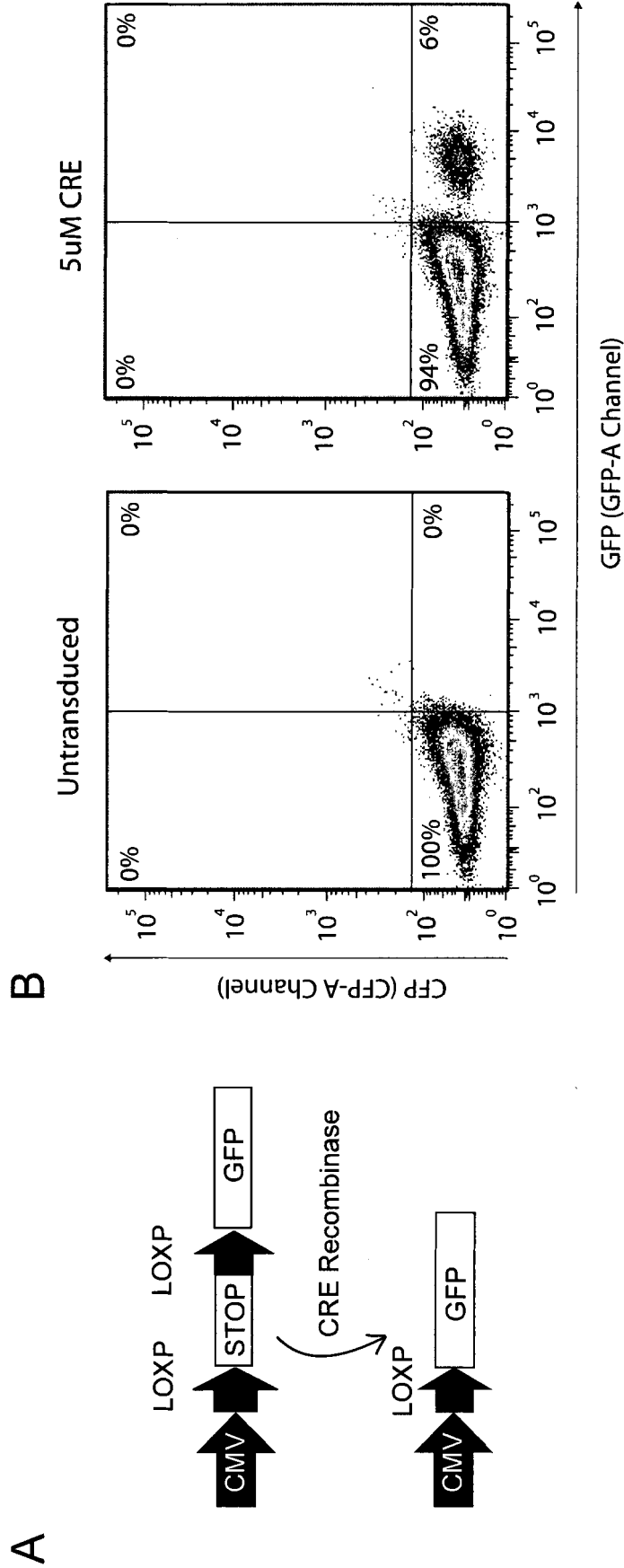


Figure 1

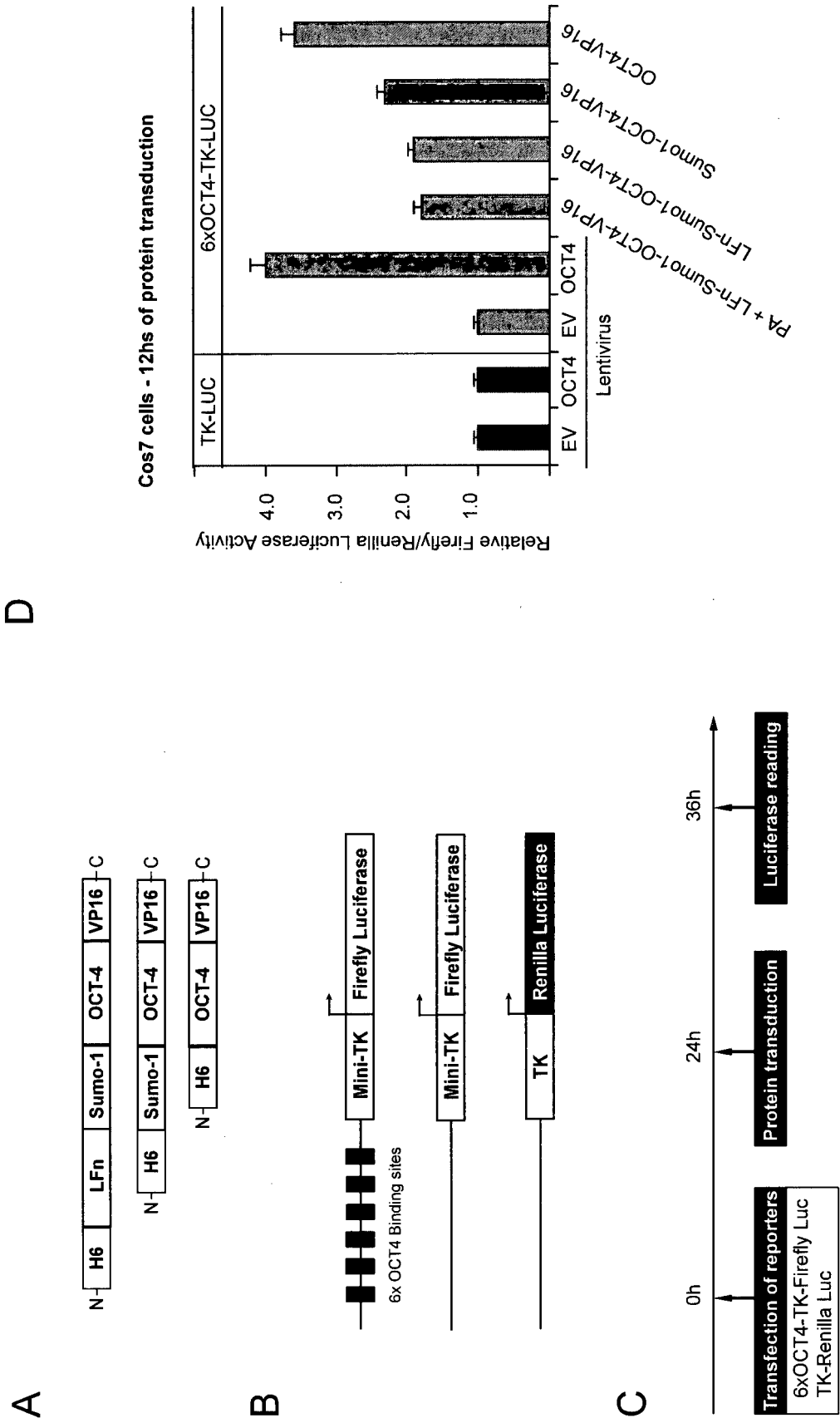
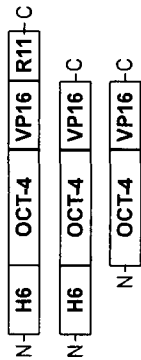
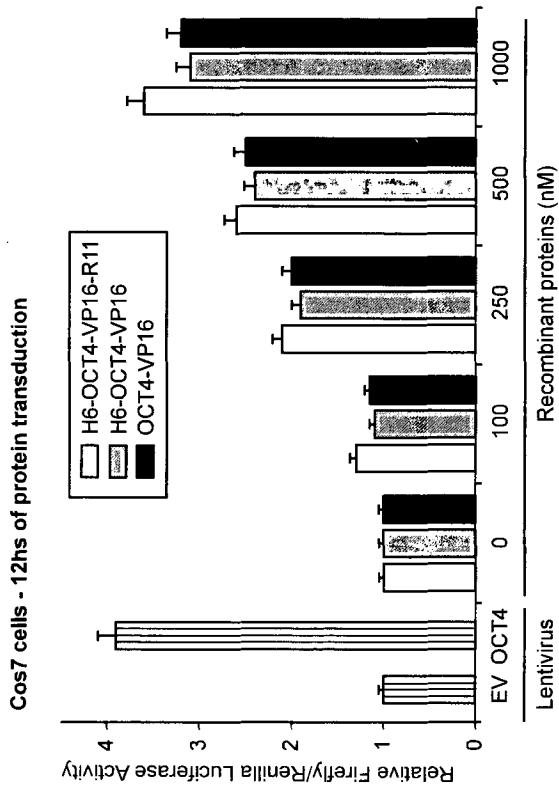


Figure 2

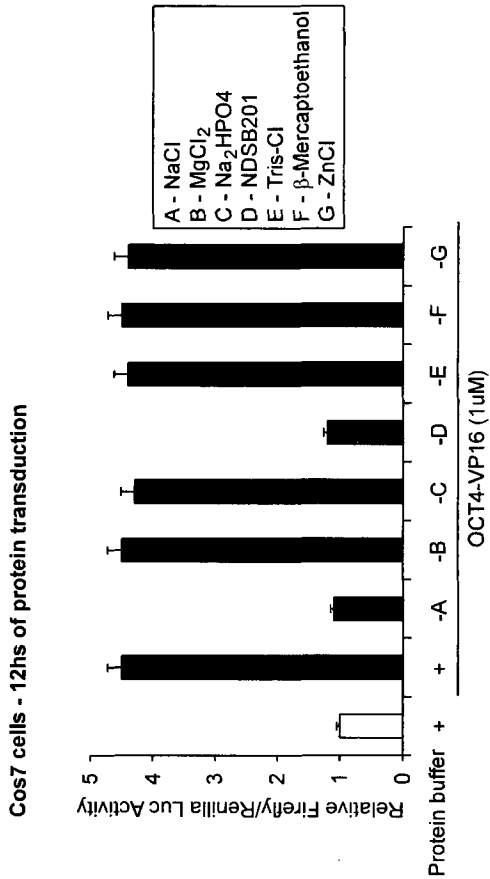
E



F



G



H

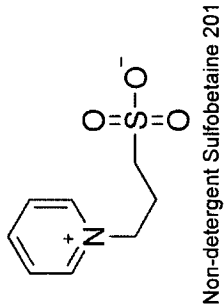


Figure 2 (CONT.)

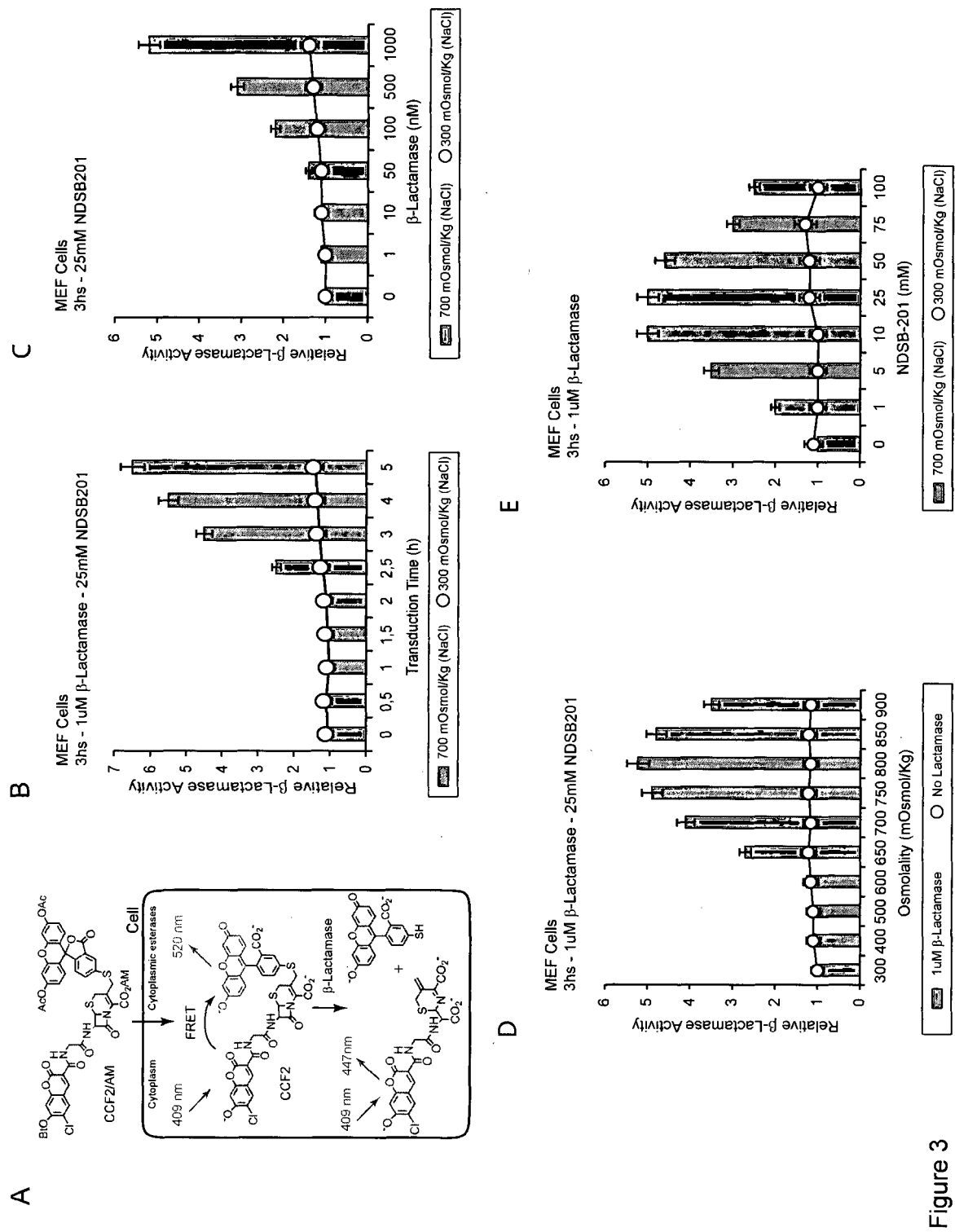
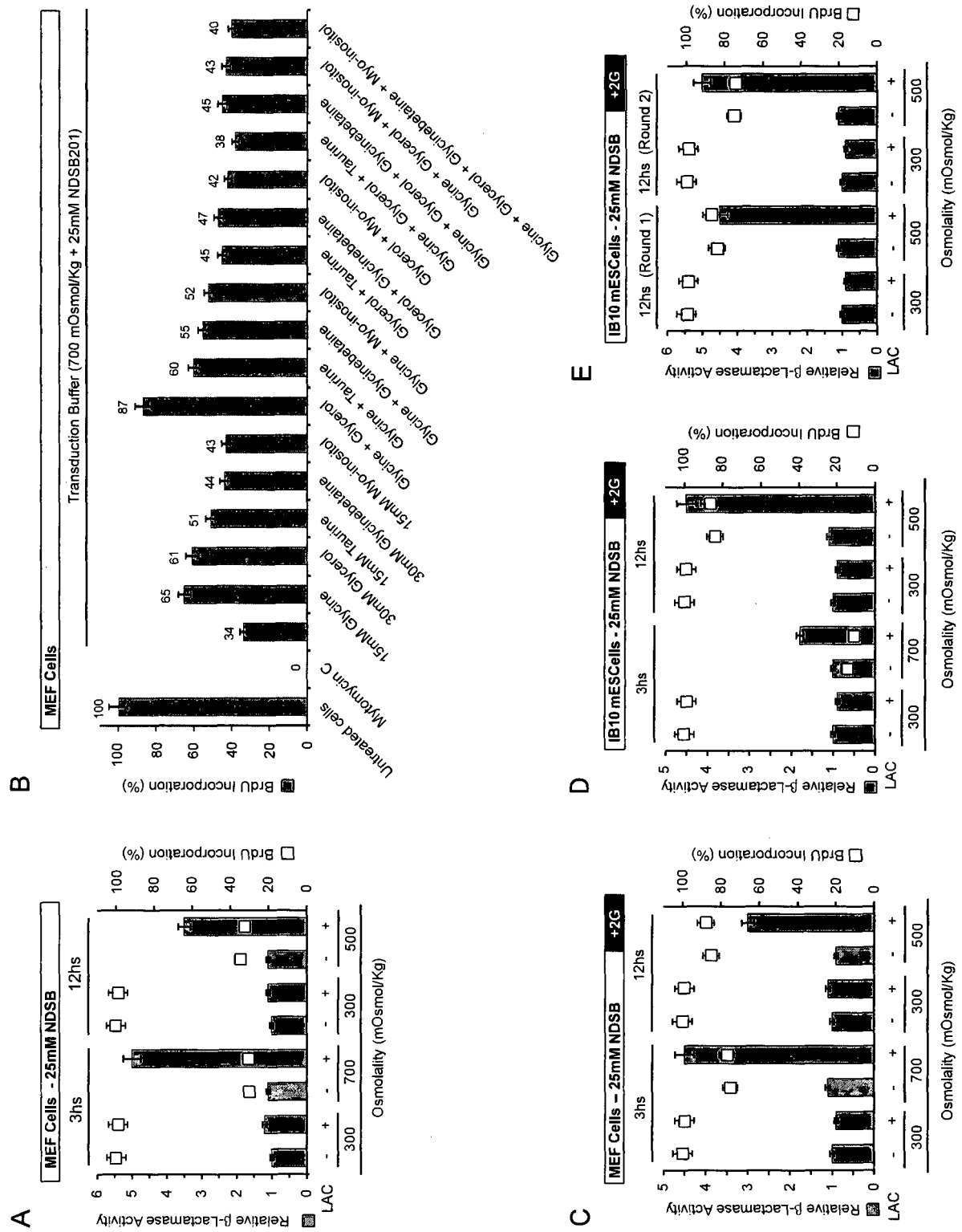


Figure 3



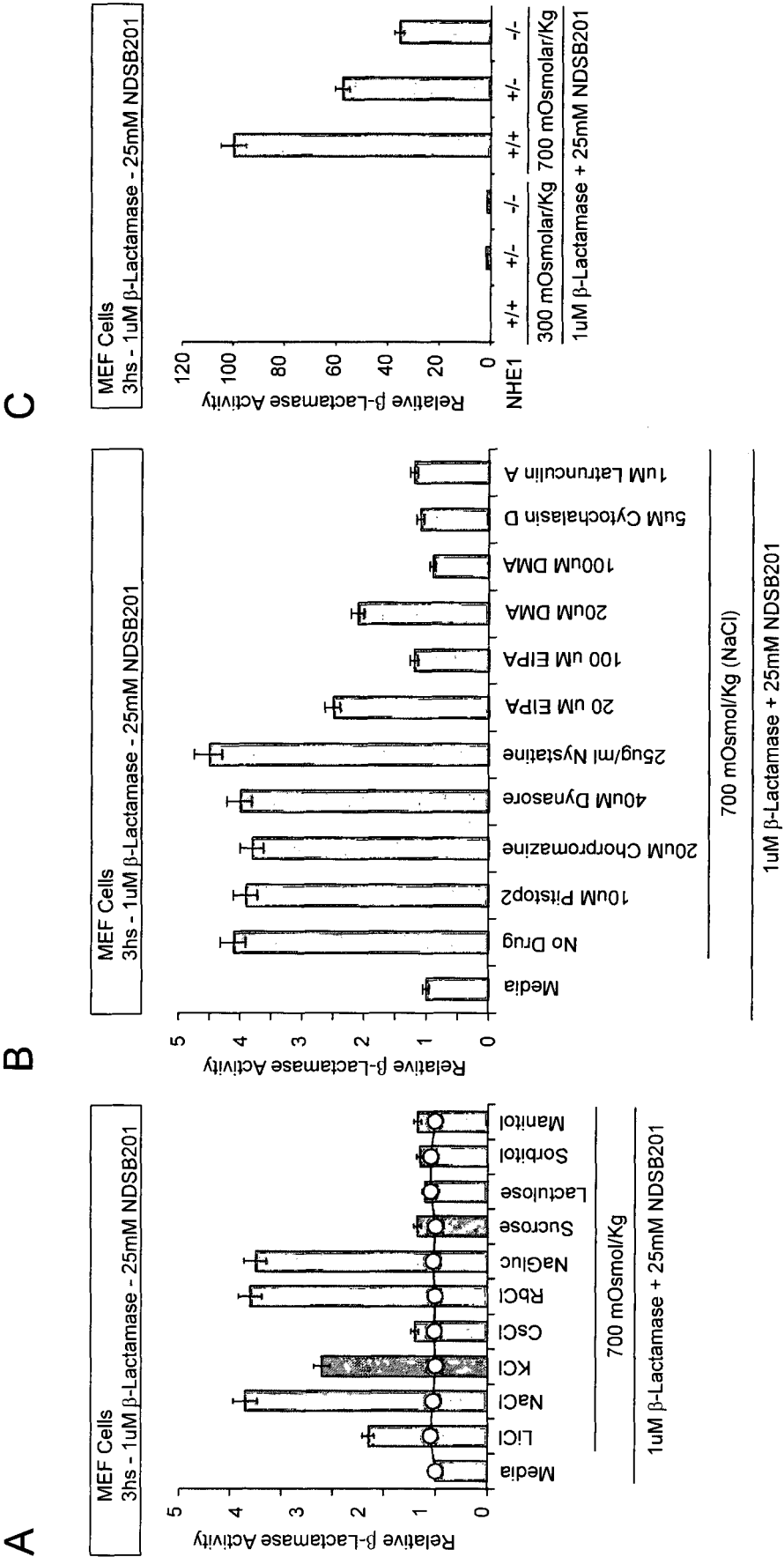
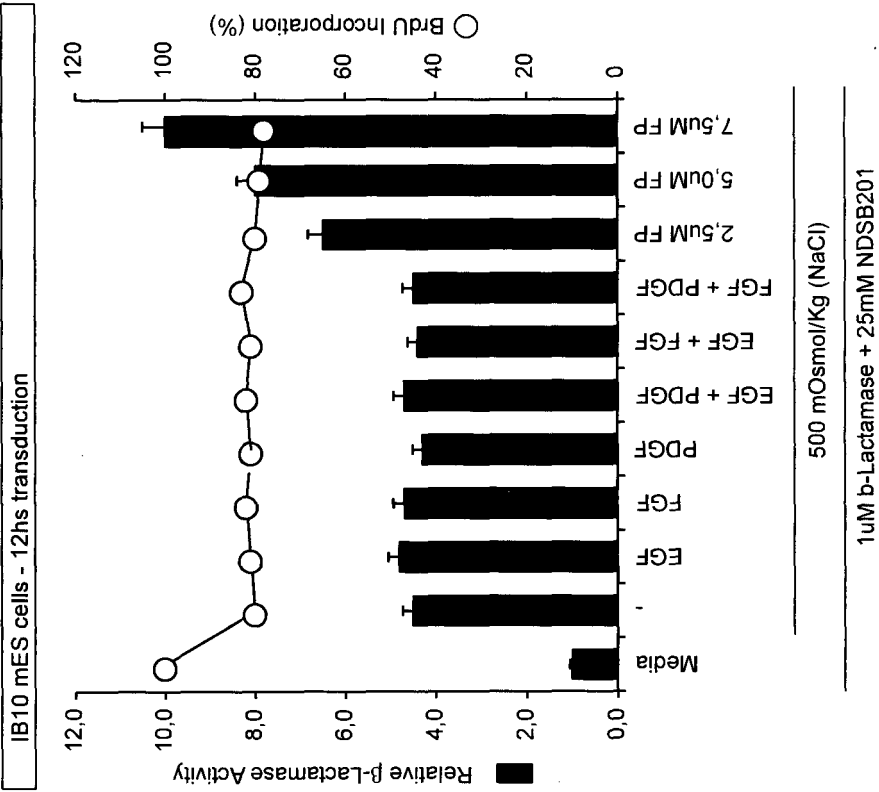


Figure 5

E



D

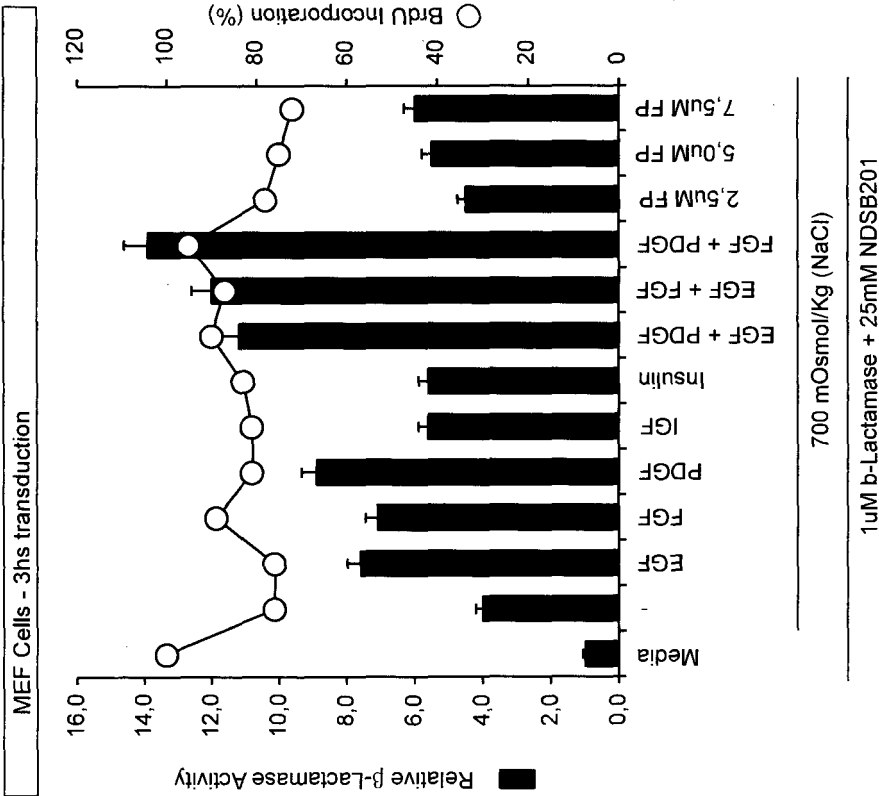


Figure 5 (CONT.)

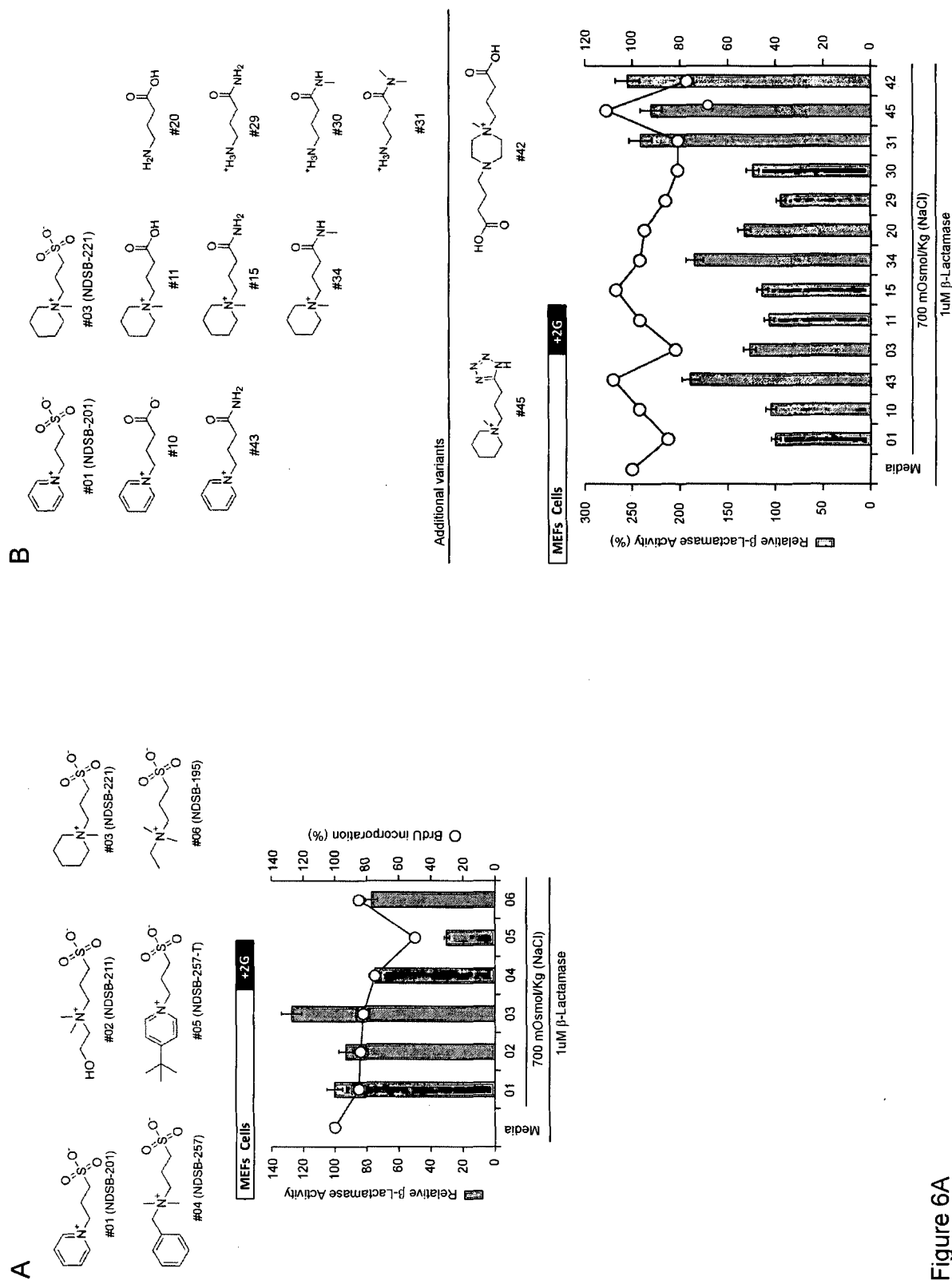
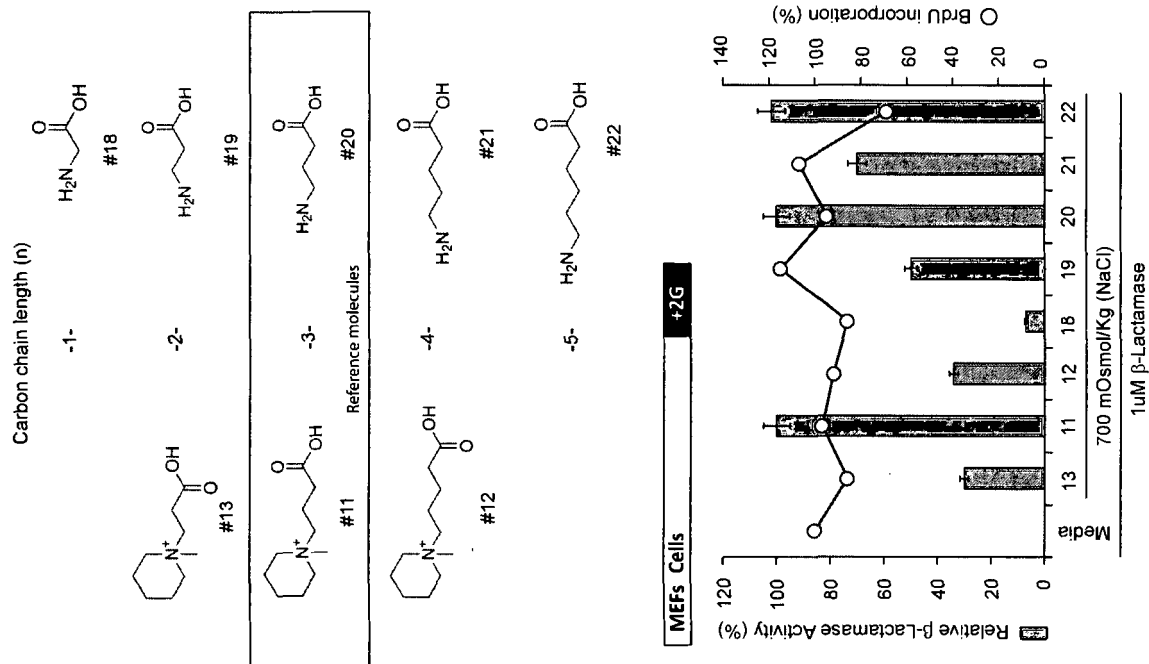


Figure 6A

D



C

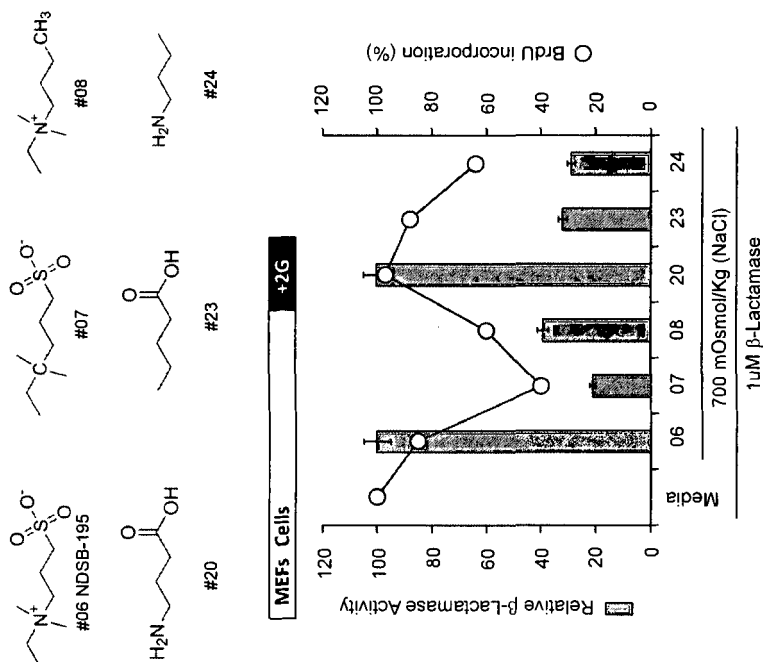


Figure 6A (CONT.)

E

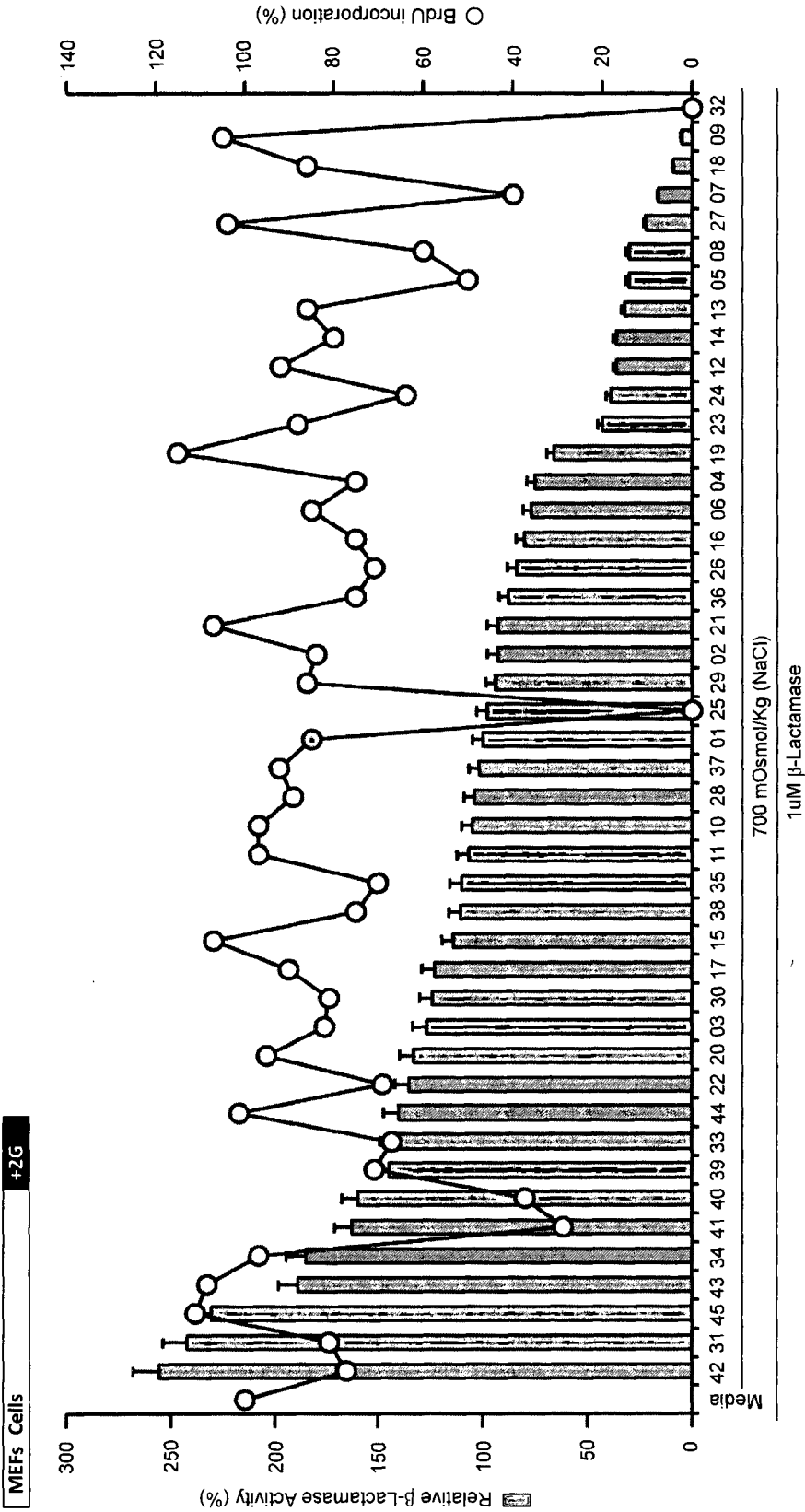
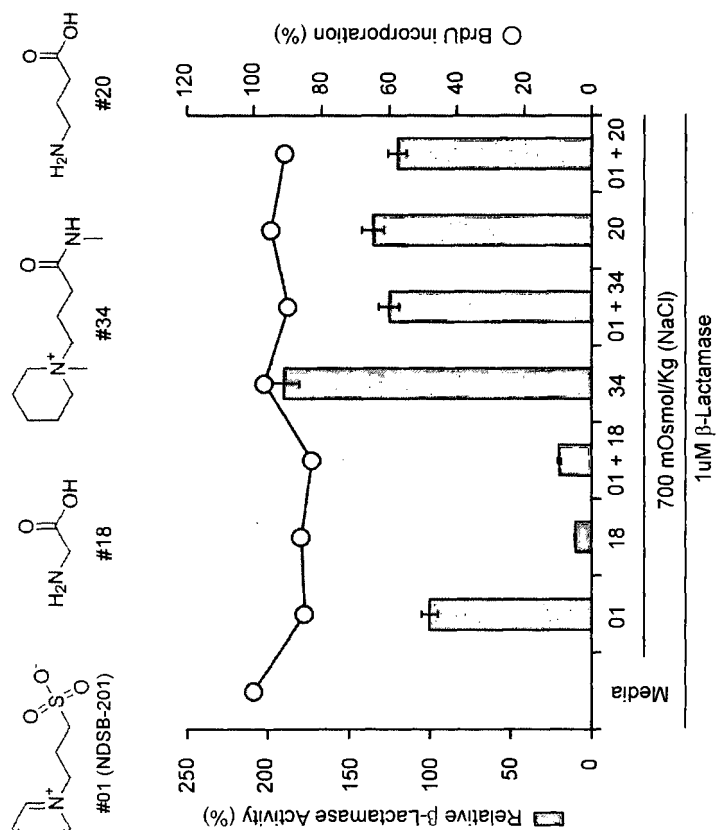


Figure 6B

F



G

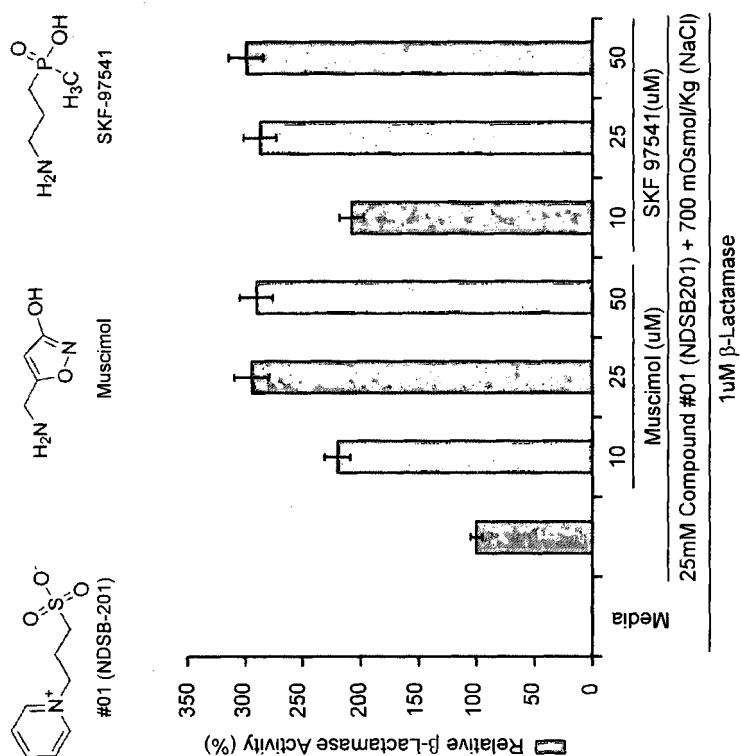
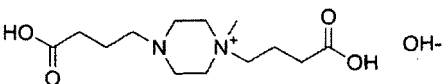
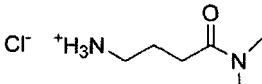
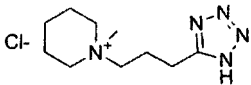
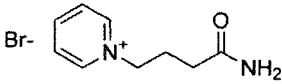
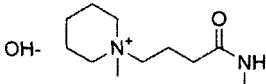
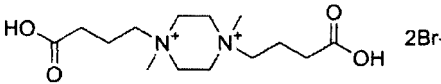
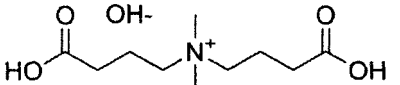
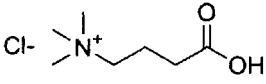
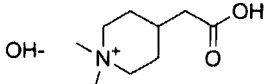
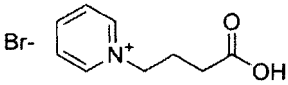
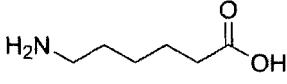
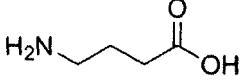
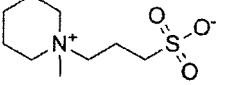


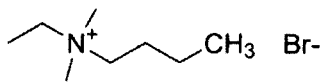
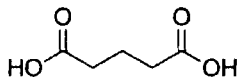
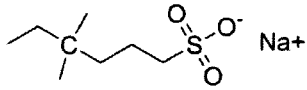
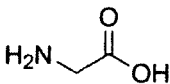
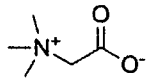
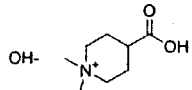
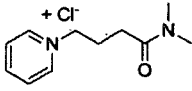
Figure 6B (CONT.)

Table1.

Code #	Structure	Transduction (%)	Cell proliferation (%)
42		255	77
31		242	81
45		230	111
43		189	108
34		185	97
41		163	29
40		160	37
39		144	71
33		142	67
44		140	101
22		135	69
20		133	95
03		127	82

Code #	Structure	Transduction (%)	Cell proliferation (%)
30	<chem>Cl- [NH3+]CCCC(=O)N</chem>	124	81
17	<chem>NCCCCC(=O)O</chem>	123	90
15	<chem>[OH-] [N+]1CCCCC1CCCC(=O)N</chem>	114	107
38	<chem>Cl- CC(C)[N+]1CCCC1C(O)CC(=O)O</chem>	111	75
35	<chem>Br- c1ccc[n+](c1)CCCC(=O)(=O)O</chem>	110	70
11	<chem>[OH-] [N+]1CCCCC1CCCC(=O)O</chem>	107	97
10	<chem>c1ccc[n+](c1)CCCC(=O)[O-]</chem>	105	97
28	<chem>OCCCCO</chem>	104	89
37	<chem>CC(=O)NCCCC(=O)O</chem>	102	92
01	<chem>c1ccc[n+](c1)CCCC(=O)(=O)[O-]</chem>	100	85
25	<chem>NCCCCN</chem>	98	0
29	<chem>Cl- [NH3+]CCCCC(=O)N</chem>	94	86
02	<chem>CC(C)[N+]1CCCC1C(O)CCCC(=O)(=O)[O-]</chem>	93	84

Code #	Structure	Transduction (%)	Cell proliferation (%)
21		93	107
36		88	75
26		84	71
16		80	75
06		77	85
04		75	75
19		66	115
23		43	88
24		39	64
12		36	92
14		36	80
13		32	86
05		30	50

Code #	Structure	Transduction (%)	Cell proliferation (%)
08		30	60
27		22	104
07		16	40
18		9	86
09		5	105
32		0	0
46			

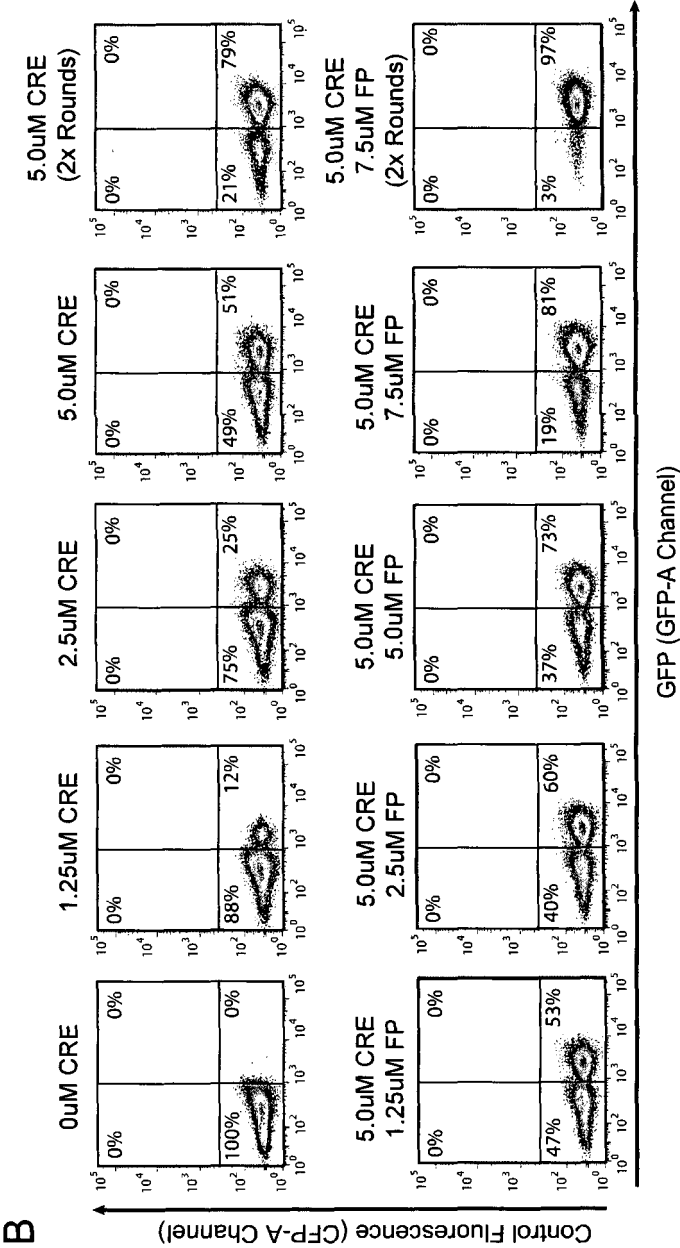


Figure 7

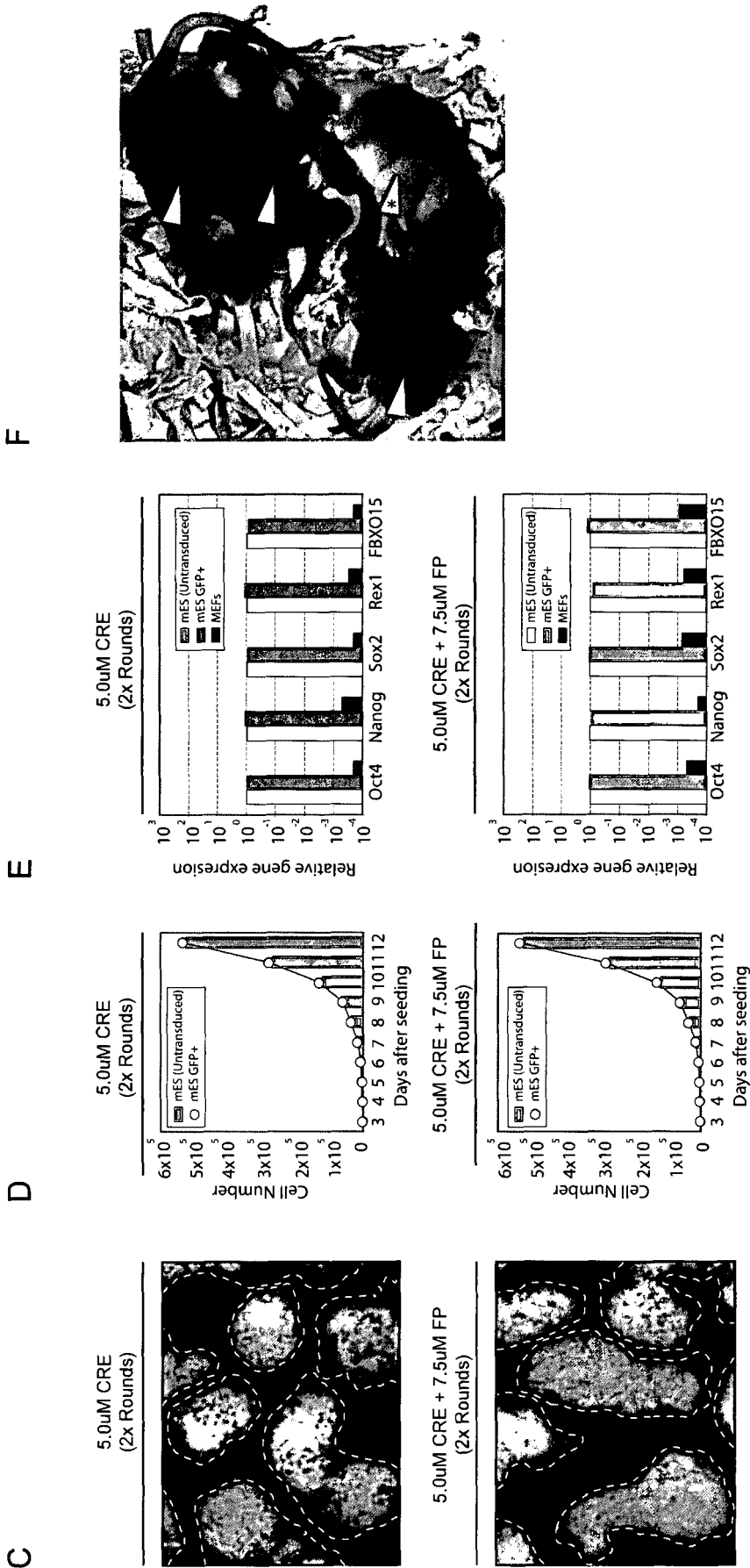


Figure 7 (CONT.)

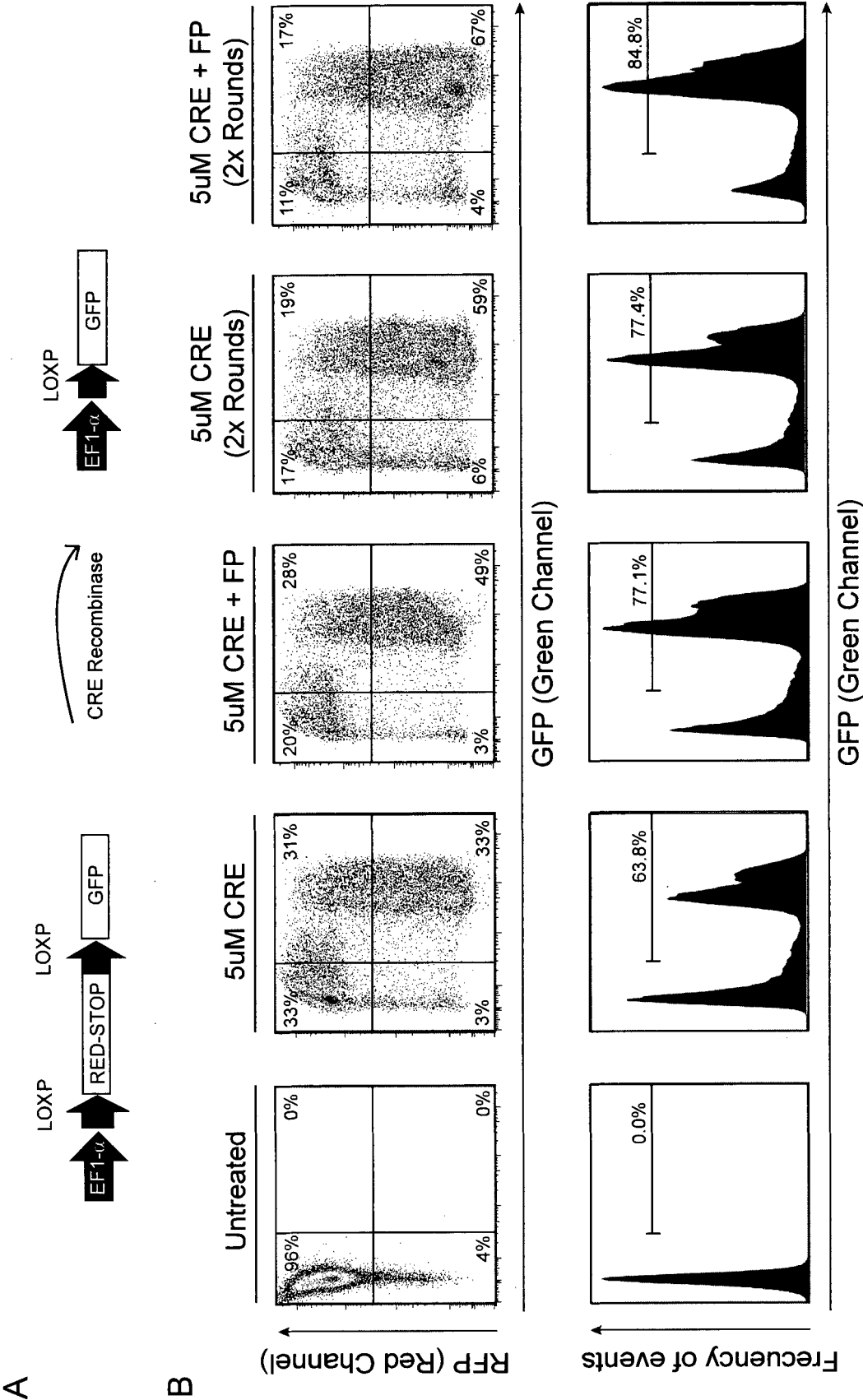


Figure 8

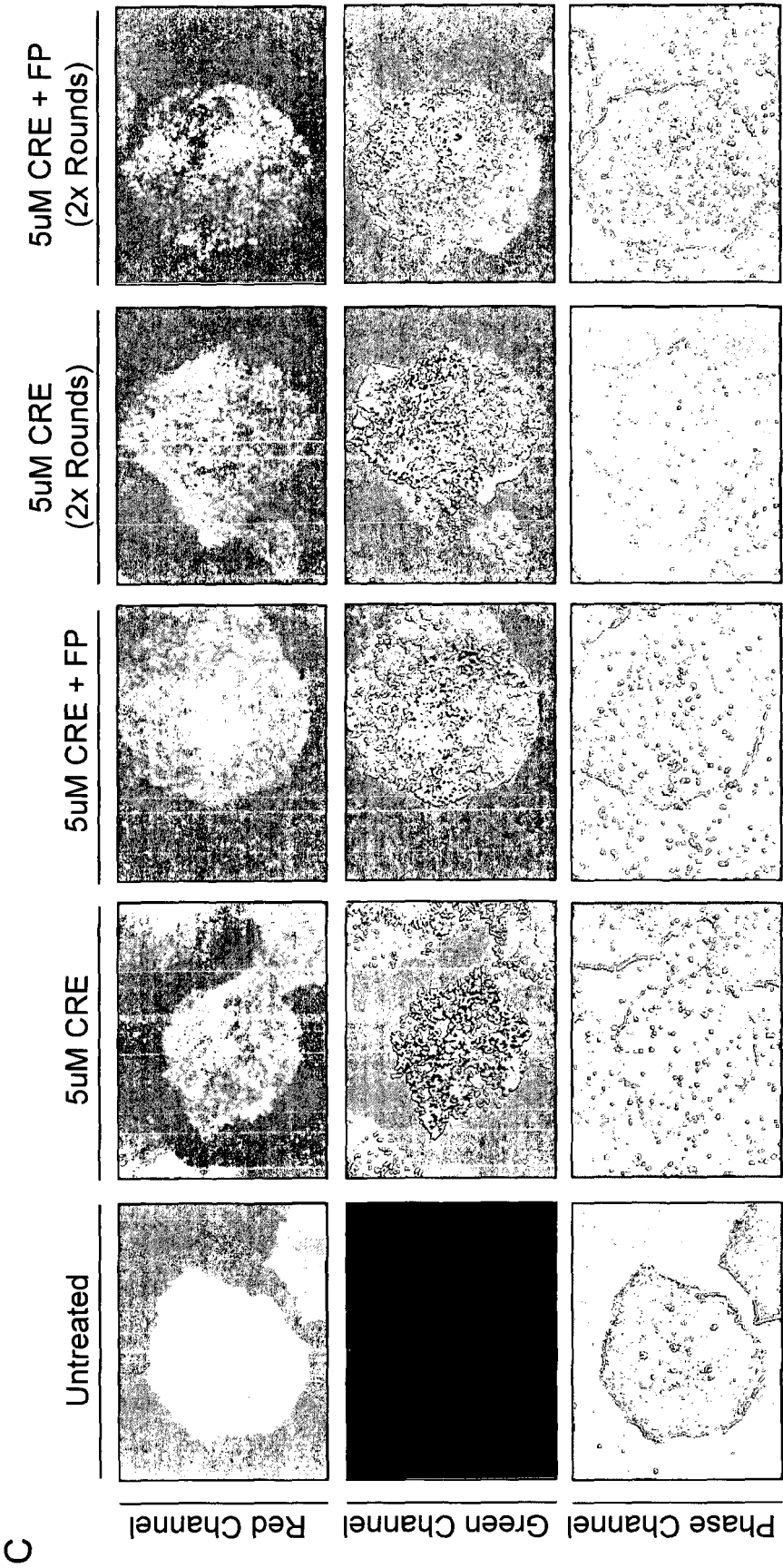
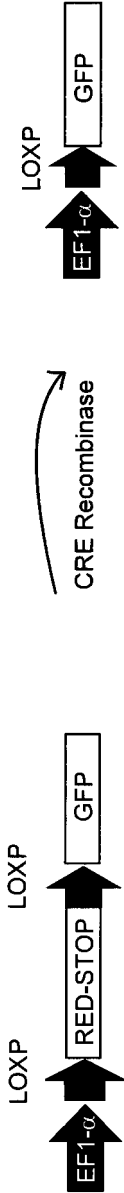


Figure 8 (CONT.)

A



B

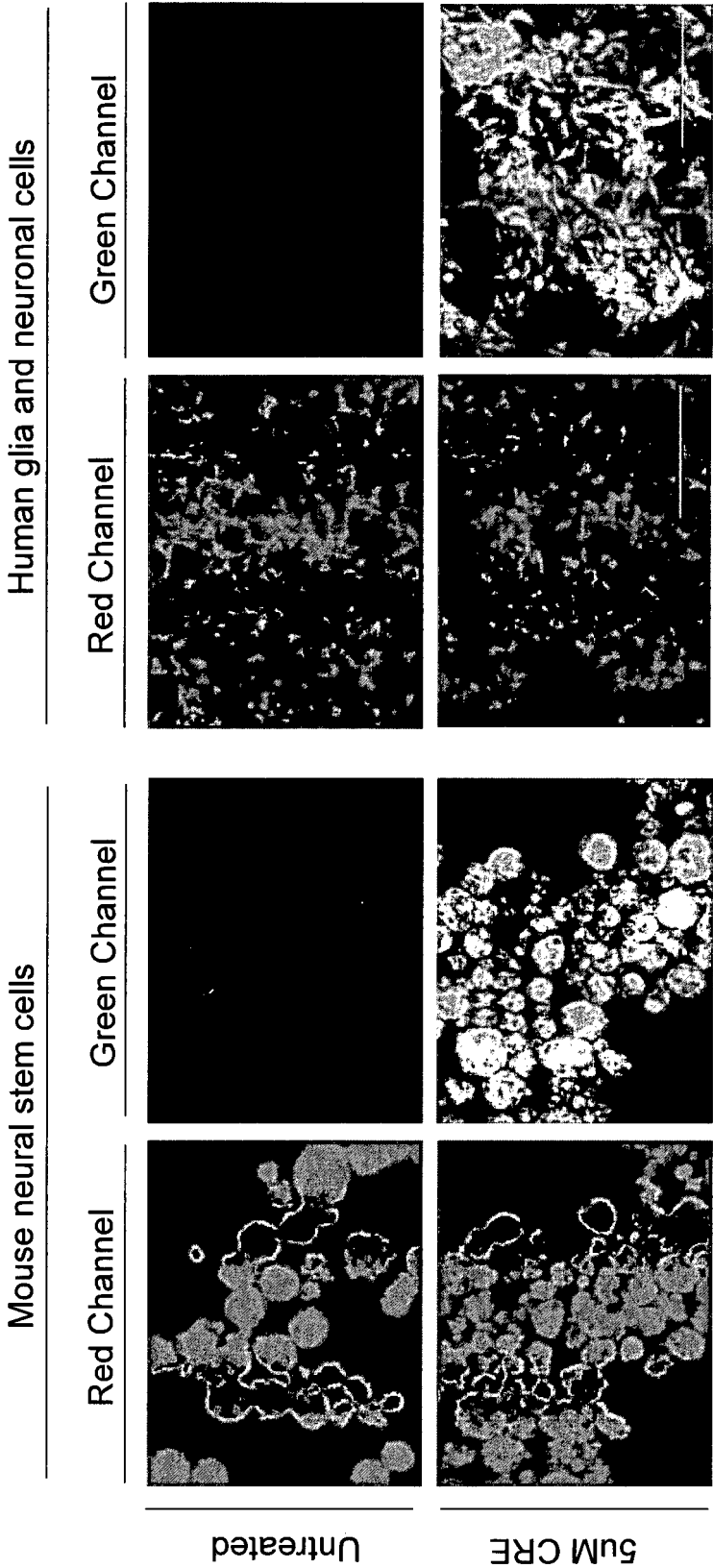


Figure 9

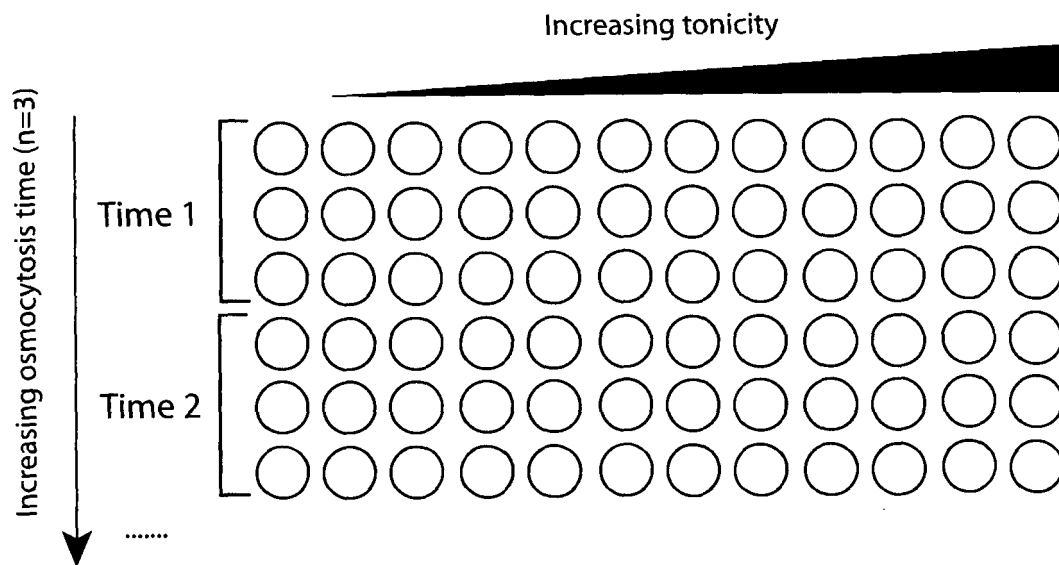


Figure 10

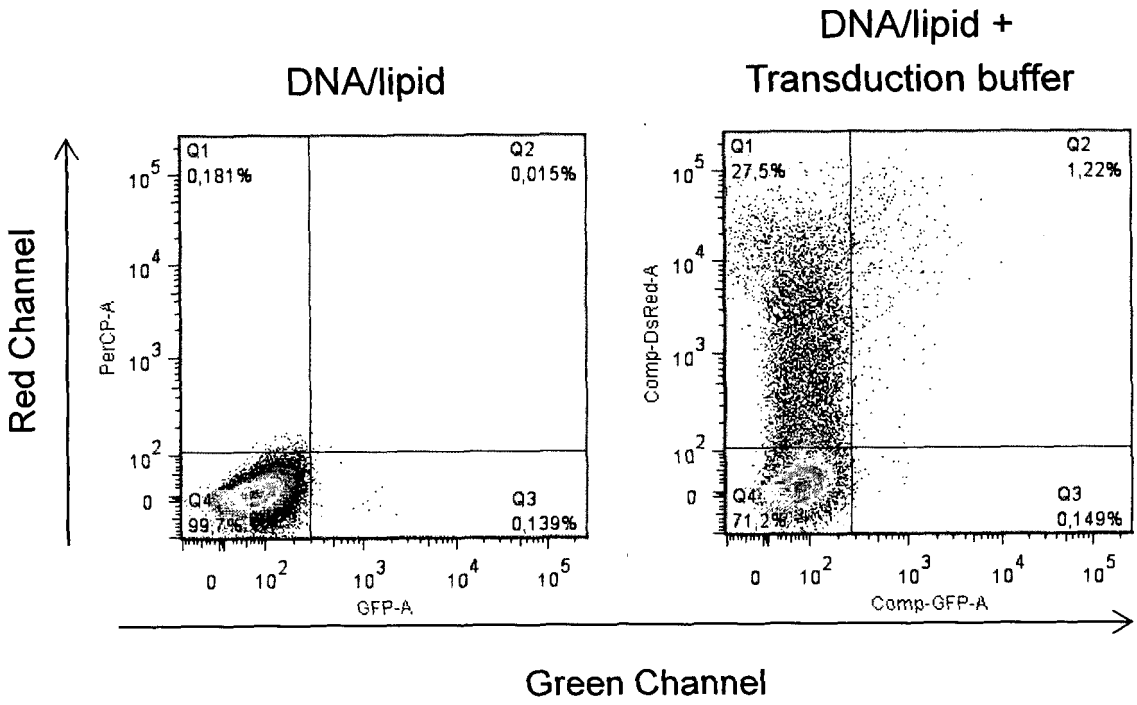


Figure 11

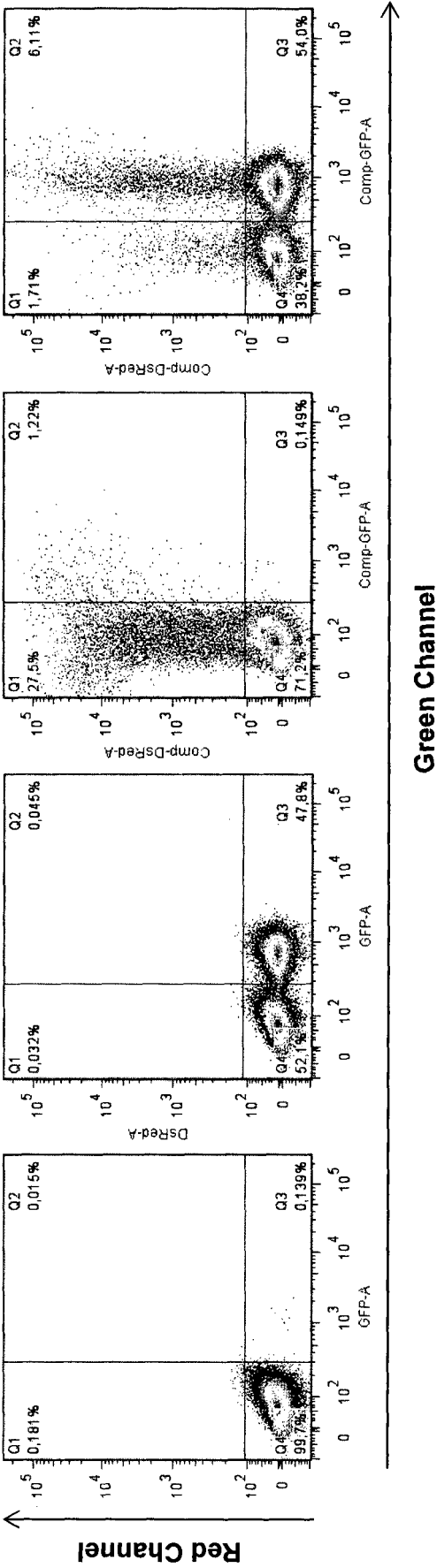


Figure 12

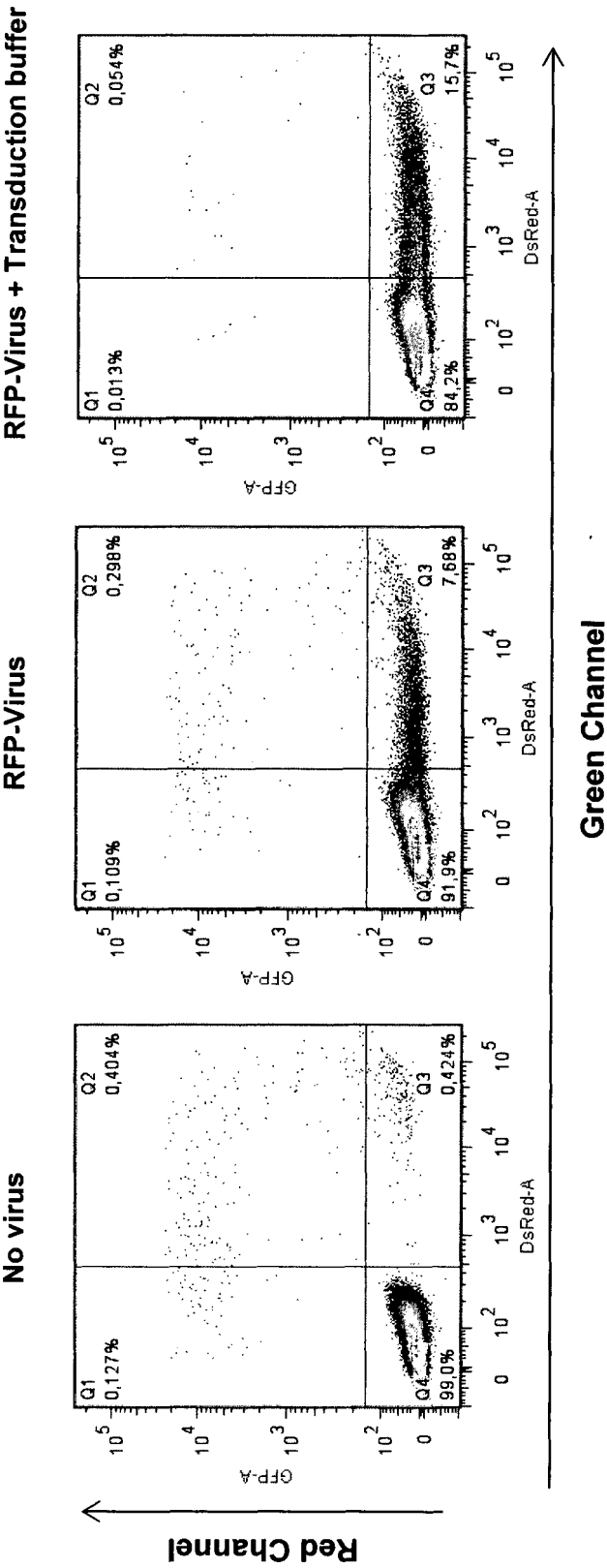


Figure 13

Mutations in 8 of 10 sequences (80%)

```

WT  GTATCCTGTAATGCTCTCATTTGAAACAGCTATATTTCTTTTTCAGATTAGTGATGCA TGAACCCAGGTTATGACCTTGCATTATTTTTCGATA CCTAATCATTTATGCTGAGGATTTGGAAAGGG
      TTTTCGATA
Δ10  GTATCCTGTAATGCTCTCATTTGAAACAGCTATATTTCTTTTTCAGATTAGTGATGCA -----TATGACCTTGCATTATTTTTCGATA CCTAATCATTTATGCTGAGGATTTGGAAAGGG
Δ20  GTATCCTGTAATGCTCTCATTTGAAACAGCTATATTTCTTTTTCAGATTAGTGATGCA -----GACCTTGCATTATTTTTCGATA CCTAATCATTTATGCTGAGGATTTGGAAAGGG
Δ14  GTATCCTGTAATGCTCTCATTTGAAACAGCTATATTTCTTTTTCAGATTAGTGATGCA -----TTGATTATTTTTCGATA CCTAATCATTTATGCTGAGGATTTGGAAAGGG
Δ50  GTATCCTGTAATGCTCTCATTTGAAACAGCTA -----TTTTCGATA CCTAATCATTTATGCTGAGGATTTGGAAAGGG
Δ88  GTATCCTGTAATGCTCTCA -----GGATTTCGAAAGGG
Δ112 GTATC -----AGGG
Δ177 TTCTCTGTAGACTAAG -----//-----TATGCACAGGTAAGTAAG
Δ269 TAATTCATTATCATACCT -----//-----TTTACTTTTCTCTGTT

```

Figure 14

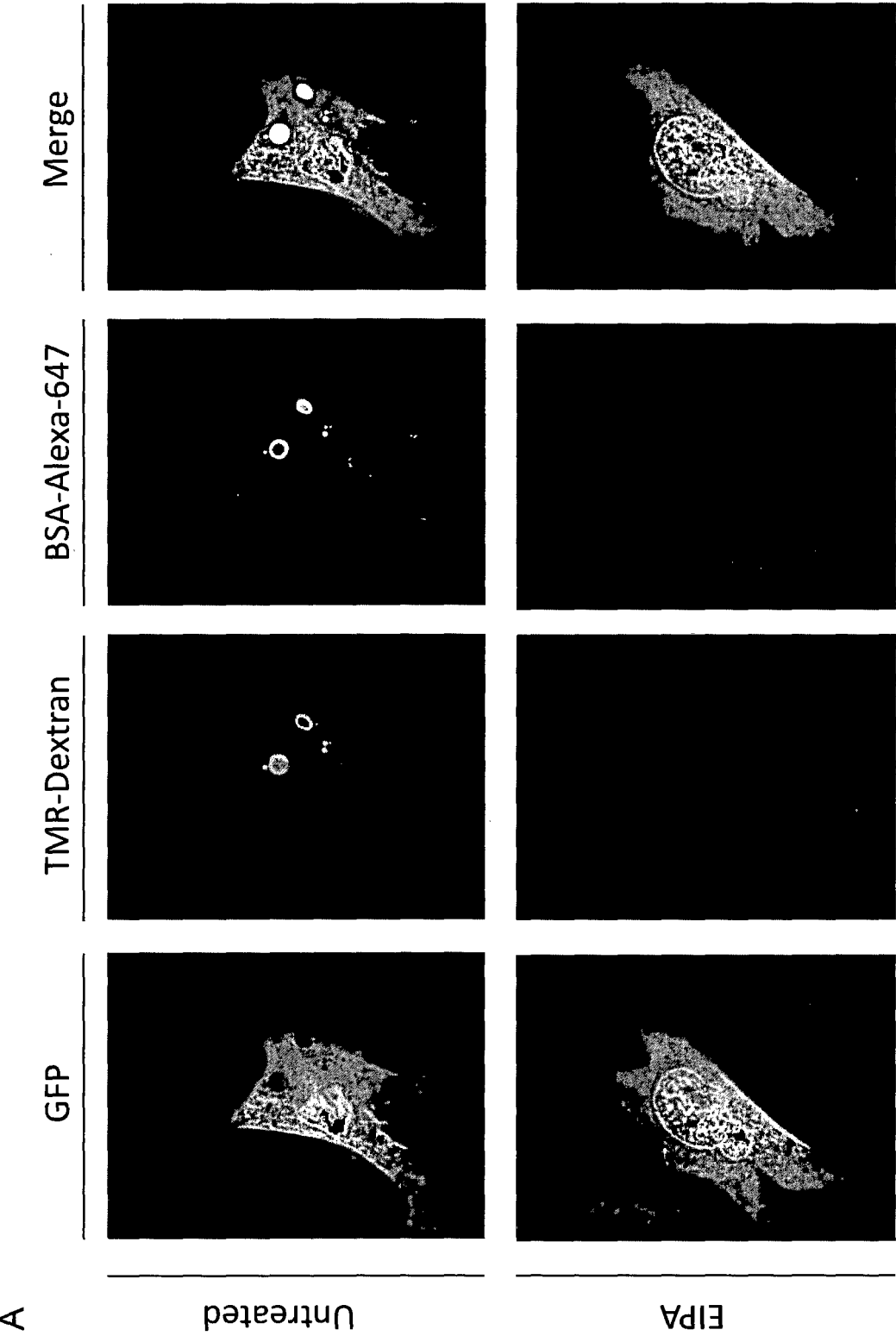


Figure 15

B

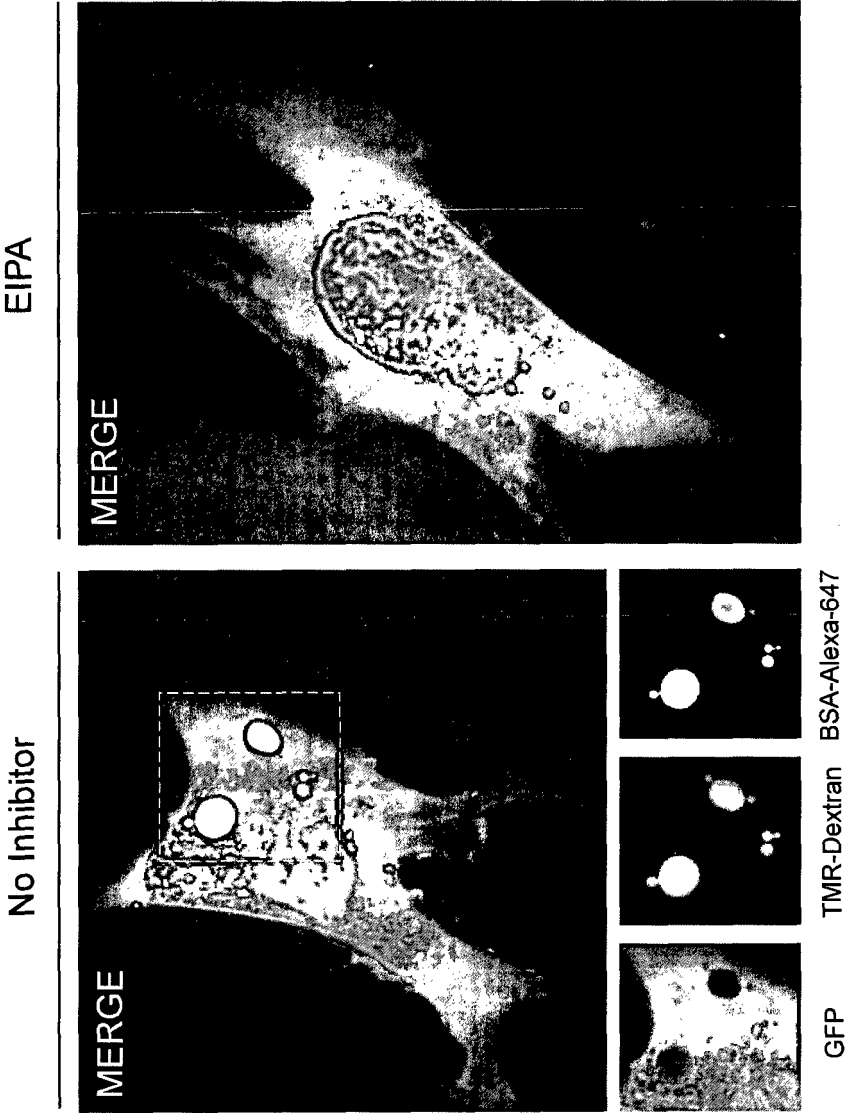


Figure 15 (CONT.)

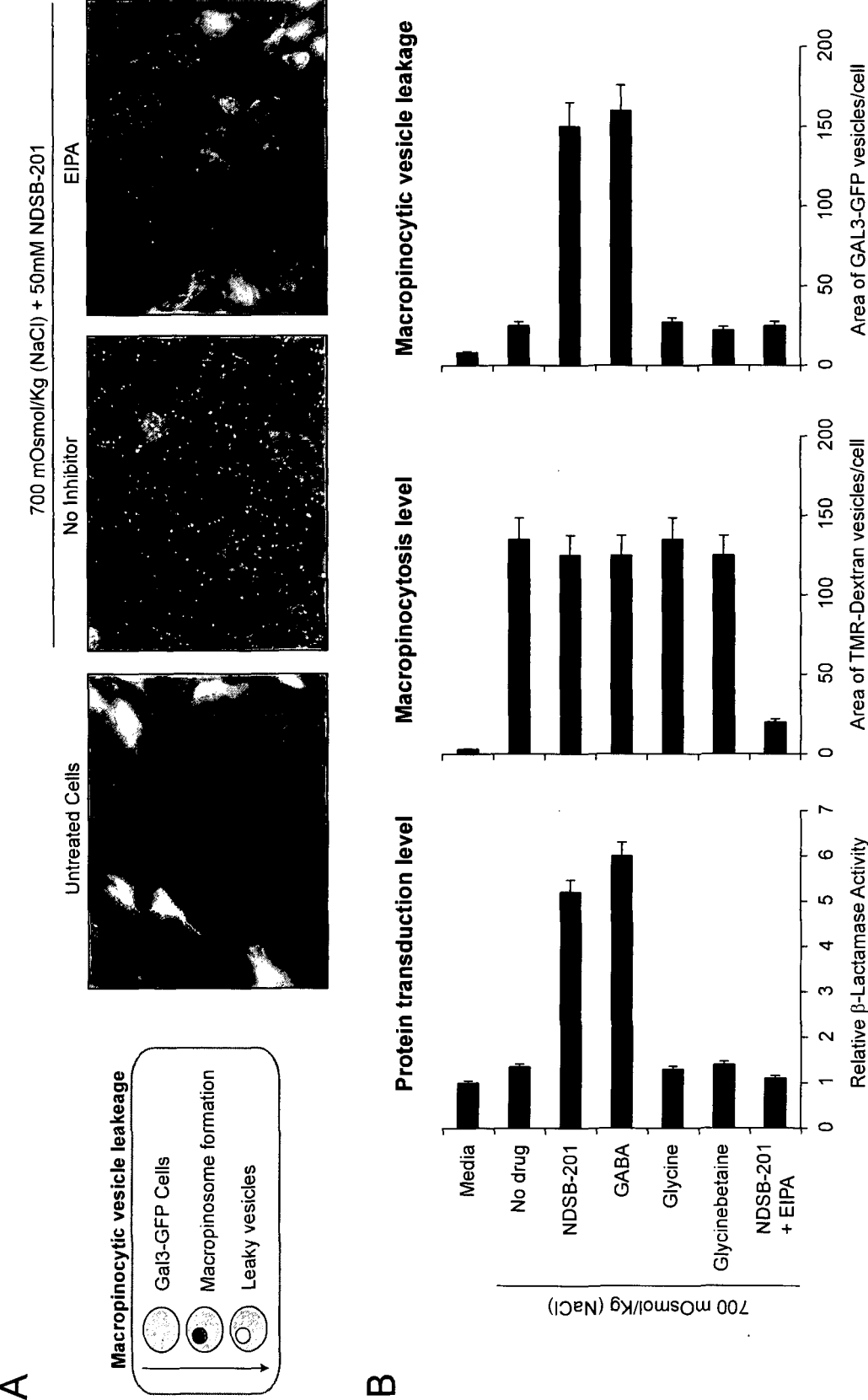


Figure 16

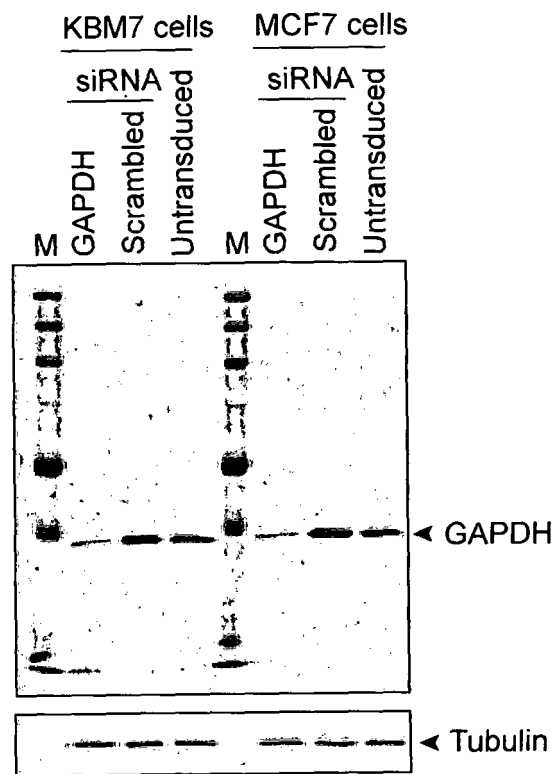


Figure 17

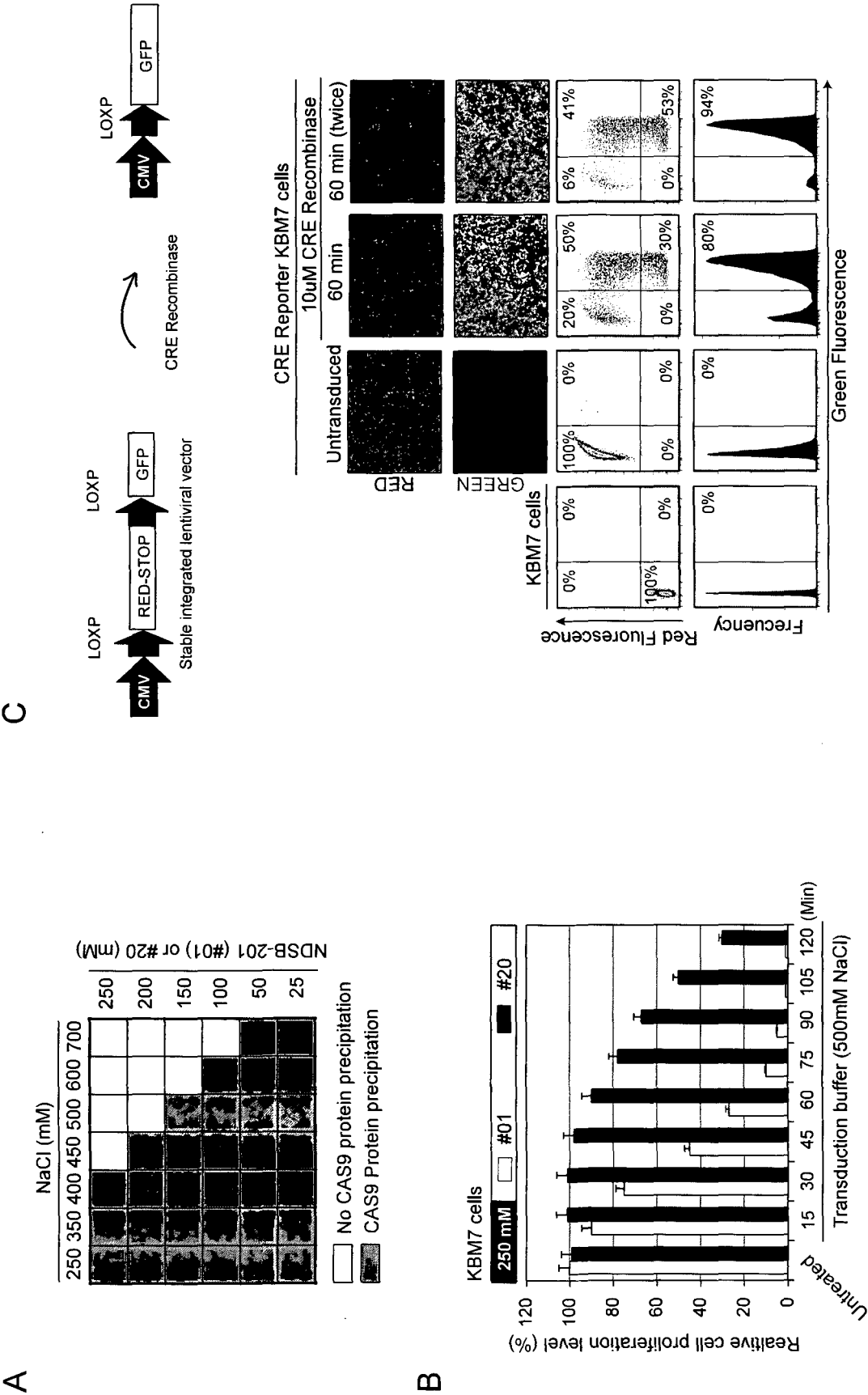
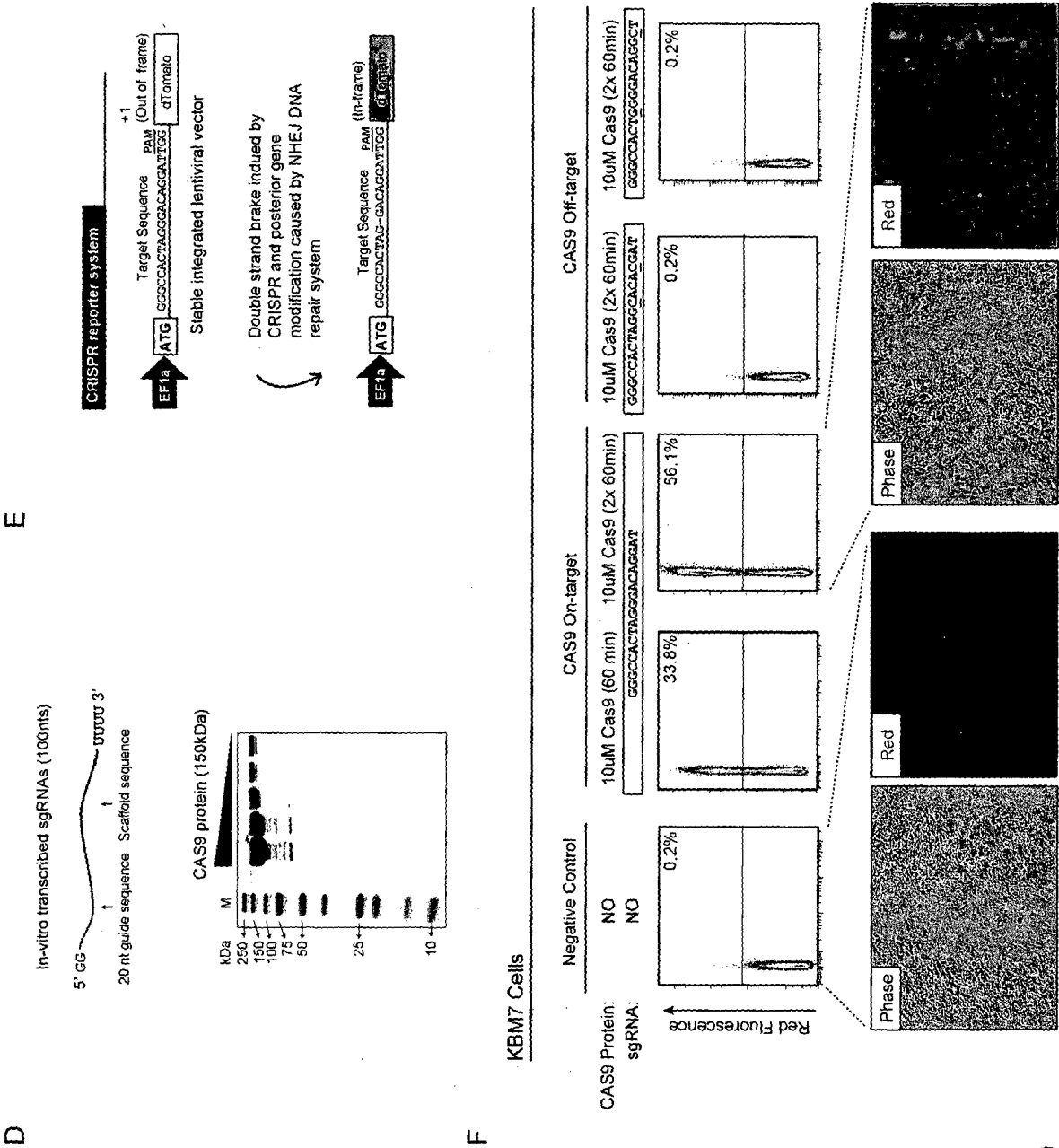


Figure 18



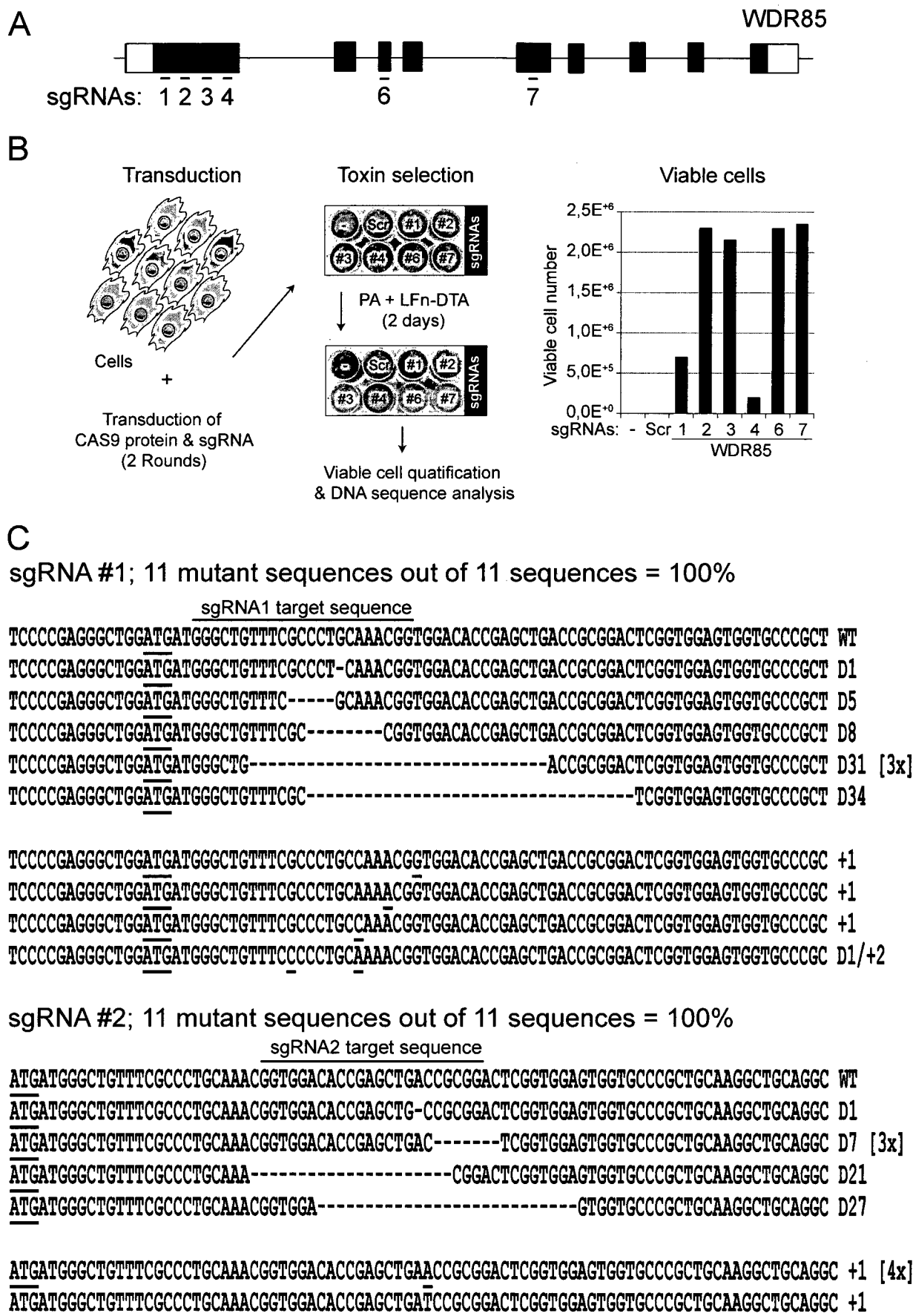


Figure 19

SUBSTITUTE SHEET (RULE 26)

sgRNA #3; 10 mutant sequences out of 10 sequences = 100%

sgRNA3 target sequence

CTGACCGCGGACTCGGTGGAGTGGTGCCCGCTGCAAGGCTGCAGGCACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCGGCCGG WT
 CTGACCGCGGACTCGGTGGAGTGGTGCCCGCTGCA-----AGGCACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCGGCCGG D7 [3x]
 CTGACCGCGGACTCGGTGGAGTGGTGCCCGCTGCA-----GGCACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCGGCCGG D8
 CTGACCGCGGACTCGGTGGAGTGGTGCC-----CTGCTGGCGTGCGGGACCTACCAGCTGCGGCGGCCGG D19

 CTGACCGCGGACTCGGTGGAGTGGTGCCCGCTGCAAGGCTTGCAGGCACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCGGCCGG D1/+1
 CTGACCGCGGACTCGGTGGAGTGGTGCCCGCTGCAAGGCATGCAGGCACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCGGCCGG D1/+1
 GGGCTCTGCGCTCGTCCGGCCG--/173/--ACCTGCTGGCGTGCGGGACCTACCAGCTGCGG---/14/--GCCTGCGGCCCC D187/+32
 CTGACCGCGG-----GCACCACTC-----CACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCGGCCGG D35/+9
 CTGACCGCGGACTCGGTGGAGTGGTGC-----ACGCACG-----TGCTGGCGTGCGGGACCTACCAGCTGCGGCGGCCGG D22/+7

sgRNA #4; 10 mutant sequences out of 10 sequences = 100%

sgRNA4 target sequence

CTGCAGGCACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCGGCCGGAGGACCGGCCTGCCGGCCCCCAGAACAAGGTGCCG WT
 CTGCAGGCACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCGGCCGGAG-ACCGGCCTGCCGGCCCCCAGAACAAGGTGCCG D1
 CTGCAGGCACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCGGC-----GACCGGCCTGCCGGCCCCCAGAACAAGGTGCCG D5
 CTGCAGGCACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCGGCCGGAG-----CTGCCGGCCCCCAGAACAAGGTGCCG D7 [2x]
 CTGCAGGCACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCG-----CCTGCCGGCCCCCAGAACAAGGTGCCG D13
 CTGCAGGCACCTGCTGGCGTGCGGGACCTACCAGCTGCGGC-----TGCGGCCCCCAGAACAAGGTGCCG D16
 CTGCAGGCACCTGCTGGCGTGCGGGACCTACCAGCTGC-----TGCGGCCCCCAGAACAAGGTGCCG D22/+2 [2x]
 CTGCAGGCACCTGCTGGCGTGCGGGACCTACC-----TGCGGTTCCCCAGAACAAGGTGCCG D25
 CTGCAGGC-----CCCCAGAACAAGGTGCCG D56

sgRNA #6; 10 mutant sequences out of 10 sequences = 100%

sgRNA6 target sequence

TGGGCCCTCTCCTCAGGTGTACATCCCGGTGGCTGGACATGCCCTCTTGGGCTTGGCAGATGCCAGTGGATCCATACAAGTGT WT
 TGGGCCCTCTCCTCAGGTGTACATCCCGGTGGCTGGACATGCCCTC-TGGGCTTGGCAGATGCCAGTGGATCCATACAAGTGT D1
 TGGGCCCTCTCCTCAGGTGTACATCCCGGTGGCTGGACATG-----GGCTTGGCAGATGCCAGTGGATCCATACAAGTGT D8
 TGGGCCCTCTCCTCAGGTGTACATCCCGGTGGCTGGACATGCCCTCT-----ATGCCAGTGGATCCATACAAGTGT D12
 TGGGCCCTCTCCTCAGGTGTACATCCCGGTGGCTGGACATGC-----AGTGGATCCATACAAGTGT D22
 TGGGCCCTCTCCTCAGGTGTACATCCA-----TTGGCAGATGCCAGTGGATCCATACAAGTGT D26/+1
 TGGGCCCTCTCCTCAGGTGTAC-----TGCATGTCCA-----CTTGGCAGATGCCAGTGGATCCATACAAGTGT D29/+10

 TGGGCCCTCTCCTCAGGTGTACATCCCGGTGGCTGGACATGCCCTCTTGGGCTTGGCAGATGCCAGTGGATCCATACAAGTGC +1
 TGGGCCCTCTCCTCAGGTGTACATCCCGGTGGCTGGACATGCCCTCTTGTGGGCTTGGCAGATGCCAGTGGATCCATACAAGTGC +2
 TGGGCCCTCTCCTCAGGTGTACATCCCGGTGGCTGGACATGCCCTCTATTGGGCTTGGCAGATGCCAGTGGATCCATACAAGTGC +2
 CATAGGAGCAGTGGCCATACCATAT-----//-----GGCTTGGCAGATGCCAGTGGATCCATACAAGTGT D182

Figure 19 (CONT.)

sgRNA #7; 12 mutant sequences out of 12 sequences = 100%

sgRNA7 target sequence

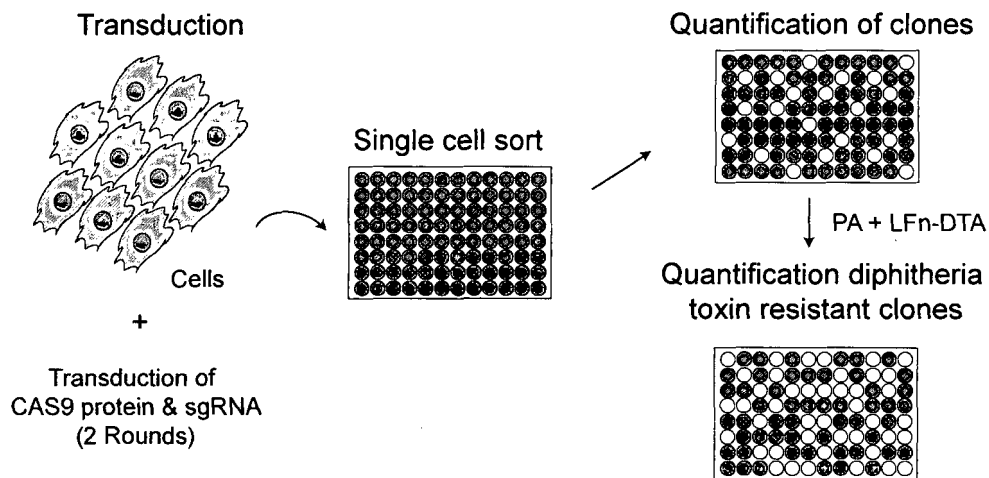
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AGGGCAGCTCCACCTCCTGATGGTGAATGAGACGAGGCCAGGCTGCAGAAAGTGGCCTCATGGCAGGCACATCAATTCGAGGCC WT
AGGGCAGCTCCACCTCCTGATGGTGAATGAGACGAGGCCAGGCTGCAGAAAG-----TGGCAGGCACATCAATTCGAGGCC D8 [3x]
AGGGCAGCTCCACCTCCTGATGGTGAATGAGACGAGGCCAGGCTGCAGAAAGC-----GCACATCAATTCGAGGCC D14/+1
AGGGCAGCTCCACCTCCTGATGGTGAATGAGACGAGGCCAGGCTG-----CATCAATTCGAGGCC D24 [2x]
AGGGCAGCTCCACCTCCTGATGGTGAATGAGACGAGGCCAGGCT-----CATCAATTCGAGGCC D25
AGGGCAGCTACACGTCCTG-----ATGGCAGGCACATCAATTCGAGGCC D41
AG-----GGCACATCAATTCGAGGCC D64

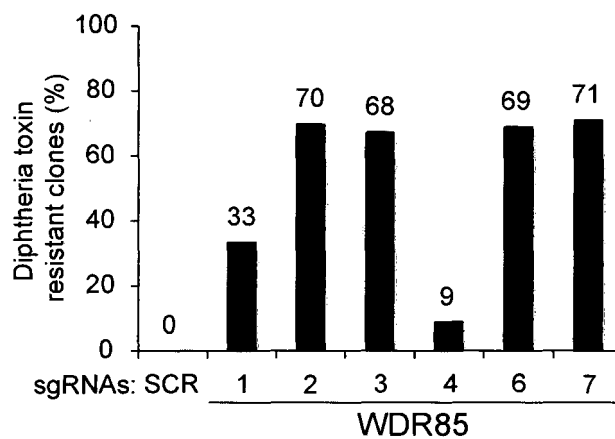
AGGGCAGCTCCACCTCCTGATGGTGAATGAGACGAGGCCAGGCTGCAGAAAGTGGCCTCATGGCAGGCACATCAATTCGAGGCC +1
AGGGCAGCTCCACCTCCTGATGGTGAATGAGACGAGGCCAGGCTGCAGAAAGTGGCCTCATGGCAGGCACATCAATTCGAGGCC +1
TGTTTGGGAACCAGAAG-----//-----GGCAGGCACATCAATTCGAGGCC D155
  
```

Figure 19 (CONT.)

D



Biallelic gene Knockout efficiency



WDR85 Knockout clones - sgRNA #2

	<u>sgRNA2 target sequence</u>	
	GCTGTTTCGCCCTGCAAACGGTGGACACCGAGCTGACCGCGGACTCGGTGGAGTGGTGCCCGCTGCAAGGCTGCAGGCACC WT	
Clone		
#202	A1-GCTGTTTCGCCCTGCAAACGGTGGACACCGAGC-----GGACTCGGTGGAGTGGTGCCCGCTGCAAGGCTGCAGGCACC	D7
	A2-GCTGTTTCGCCCTGCAAACGGTGGACACCG-----CGGACTCGGTGGAGTGGTGCCCGCTGCAAGGCTGCAGGCACCD9	
#205	A1-GCTGTTTCGCCCTGCAAACGGTGGACACCGAGCTGAACCGCGGACTCGGTGGAGTGGTGCCCGCTGCAAGGCTGCAGGCAC	+1
	A2-GCTGTTTCGCCCTGCAAACGGTGGACACCGAGCTGA [↑] CCGCGGACTCGGTGGAGTGGTGCCCGCTGCAAGGCTGCAGGCACC	+75
	<u>AACTAGCCCTGAAAATGGATGGCGCTGGAGCGTCGGGGCCCATACCCGGCCGTCGCCGGCAGTCGAGAGTGGACGG</u>	
#208	A1-GCTGTTTCGCCCTGCAAACGGTGGACAC-----GGACTCGGTGGAGTGGTGCCCGCTGCAAGGCTGCAGGCACC	D12
	A2-GCTGTTTCGCCCTGCAAACGGTGGACACCG-----CGGACTCGGTGGAGTGGTGCCCGCTGCAAGGCTGCAGGCACC	D9

Figure 19 (CONT.)

WDR85 Knockout clones - sgRNA #3

		<u>sgRNA3 target sequence</u>	
		ACCGCGGACTCGGTGGAGTGGTGTGCCCCTGCAAGGCTGCAGGCACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCGG	WT
Clone			
#301	A1-	ACCGCGGACTCGGTGGAGTGGTGTGCCCCTGCAAGGCC-CAGGCACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCGG	D2/+1
	A2-	ACCGCGGACTCGGTGGAGTGGTGTGCCCCTGCA-----GGCACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCGG	D8
#304	A1-	ACCGCGGACTCGGTGGAGTGGTGTGCCCCTGCA-----AGGCACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCGG	D7
	A2-	ACCGCGGACTCGGTGGAGTGGTGTGCCCCTGCAAGGC-----GTGCGGGACCTACCAGCTGCGGCGG	D17
#305	A1-	ACCGCGGACTCGGTGGAGTGGTGTGCCCCTGCAAGGC-----ACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCGG	D7
	A2-	ACCGCGGACTCGGTGGAGTGGTGTGCCCCTGCA-----GGCACCTGCTGGCGTGCGGGACCTACCAGCTGCGGCGG	D8

WDR85 Knockout clones - sgRNA #6

		<u>sgRNA6 target sequence</u>	
		TCCTCAGGTGTACATCCCGGTGGCTGGACATGCCCTCTTGGGCTTGGCAGATGCCAGTGGATCCATACAACCTGCTCC	WT
Clone			
#605	A1-	TCCTCAGGTGTACATCCCGGTGGCT-----TGGCAGATGCCAGTGGATCCATACAACCTGCTCC	D19
	A2-	TCCTCAGGTGTACATCCCGGTGGCTGGACATGCCCTCT-----//-----GAAGTGACAGGATGTTTCATTG	D276
#606	A1-	TCCTCAGGTGTACATCCCGGTGGCTGGACATGCCCTCT-TGGCTTGGCAGATGCCAGTGGATCCATACAACCTGCTCC	D1
	A2-	TCCTCAGGTGTACATCCCGGTGGCTGGACATGCCCTCTTGGGCTTGGCAGATGCCAGTGGATCCATACAACCTGCTCC	+59
		<u>CTGGCATGCACCTGTAATTACAGCTACTGTCTGTGCATCTAACCATTTTGTCAATCCAC</u>	
#609	A1-	TCCTCAGGTGTACATCCCGGTGGCTGGACATGCCCT-----TGGCAGATGCCAGTGGATCCATACAACCTGCTCC	D8
	A2-	CTAAGAGCTC-----/606/-----GGGCTTGGCAGATGCCAGTGGATCCATACAACCTGCTCC	D606

WDR85 Knockout clones - sgRNA #7

		<u>sgRNA7 target sequence</u>	
		CCACCTCCTGATGGTGAATGAGACGAGGCCAGGCTGCAGAAAGTGGCCTCATGGCAGGCACATCAATTCGAGGCC	WT
Clone			
#703	A1-	CCACCTCCTGATGGTGAATGAGACGAGGCCAGGCTGCAGAAAGTGGC-----AGGCACATCAATTCGAGGCC	D8
	A2-	CCACCTCCTGATGGTGAATGAGACGAGGCCAGGCTGCAGAAAGTGGC-----CAGGCACATCAATTCGAGGCC	D7
#706	A1-	CCACCTCCTGATGGTGAATGAGACGAGGCCAGGCTGCAGAAAGTGGCA-----GCACATCAATTCGAGGCC	D10/+1
	A2-	CCACCTCCTGATGGTGAATGAGACGAGGCC-----CAGGCACATCAATTCGAGGCC	D25
#708	A1-	CCACCTCCTGATGGTGAATGAGACGAGGCCAGGCTGCAG-----CAGGCACATCAATTCGAGGCC	D15
	A2-	CCACCTCCTGATGGTGAATGAGACGAGGCCAGGCTGCAGAAAGTGGCCAATCATGGCAGGCACATCAATTCGAGG	+2

Figure 19 (CONT.)