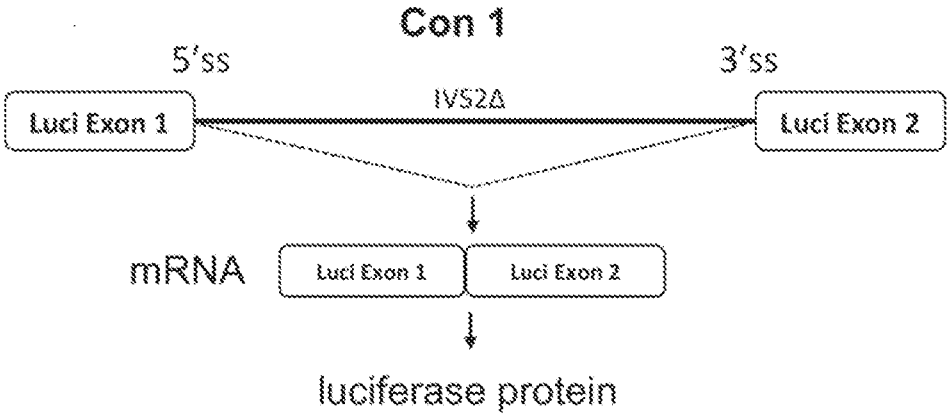

Con 1
5'ss
3'ss
IVS2Δ
Luci Exon 1
Luci Exon 2
mRNA
Luci Exon 1
Luci Exon 2
luciferase protein

Fig. 1a

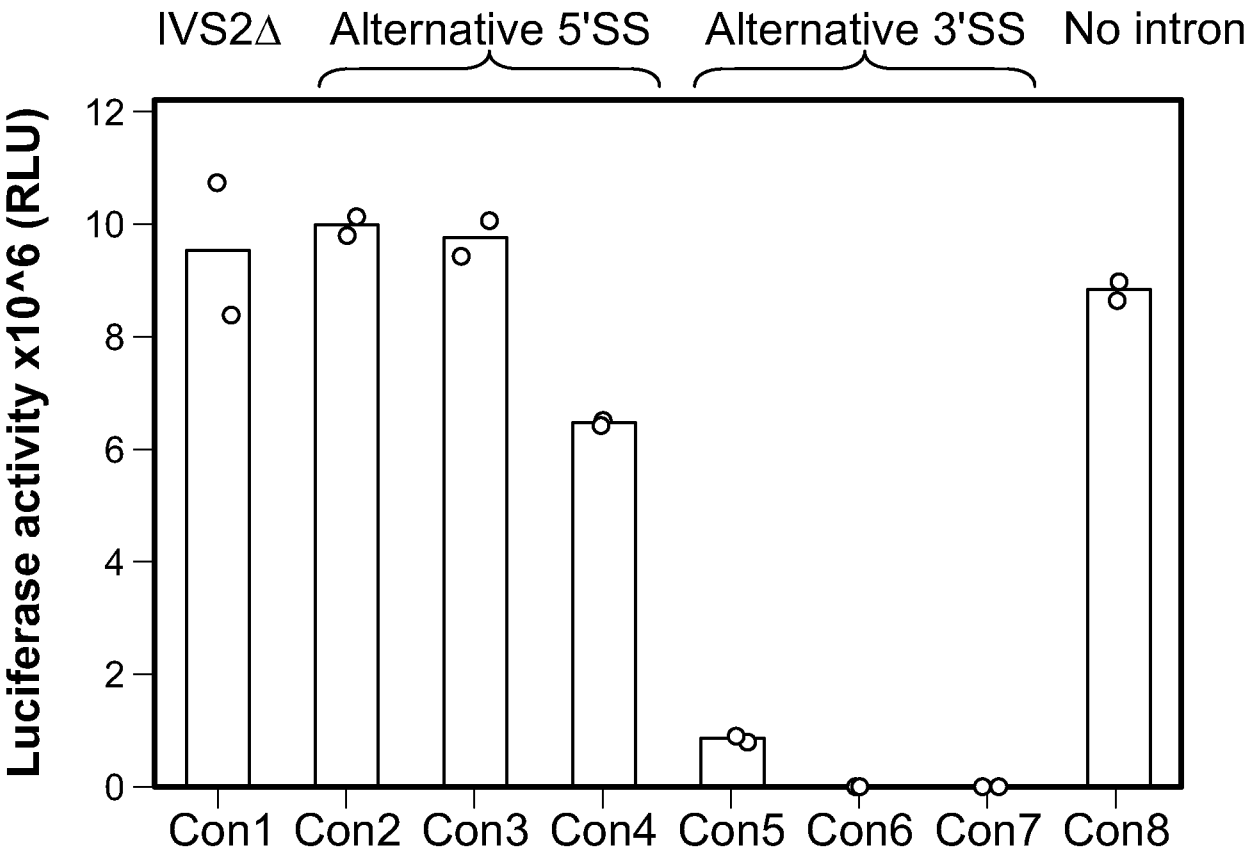

IVS2Δ
Alternative 5'SS
Alternative 3'SS
No intron
Luciferase activity x10^6 (RLU)
12
10
8
6
4
2
0
Con1
Con2
Con3
Con4
Con5
Con6
Con7
Con8

Fig. 1b

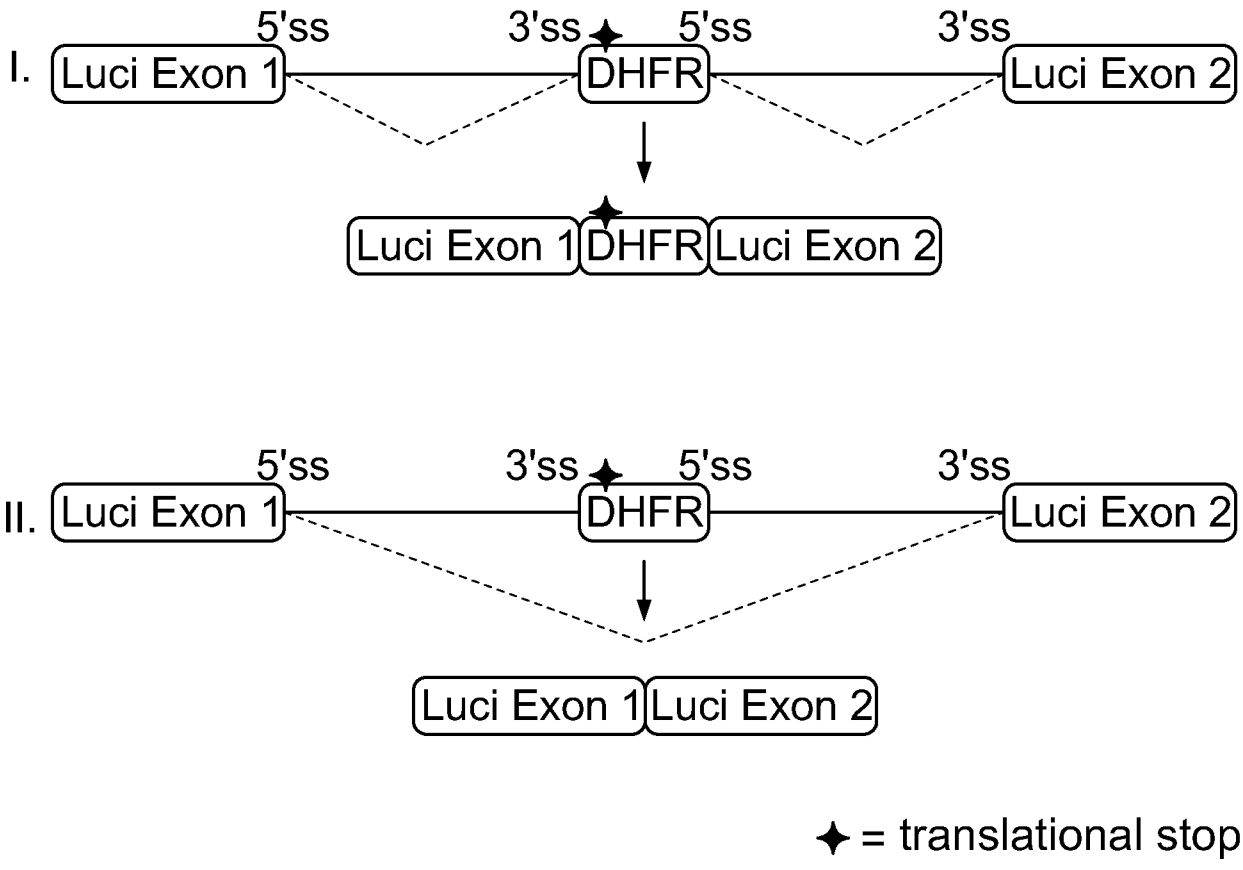

I.
5'ss
3'ss
5'ss
3'ss
Luci Exon 1
DHFR
Luci Exon 2
Luci Exon 1
DHFR
Luci Exon 2
II.
5'ss
3'ss
5'ss
3'ss
Luci Exon 1
DHFR
Luci Exon 2
Luci Exon 1
Luci Exon 2
= translational stop

Fig. 2a

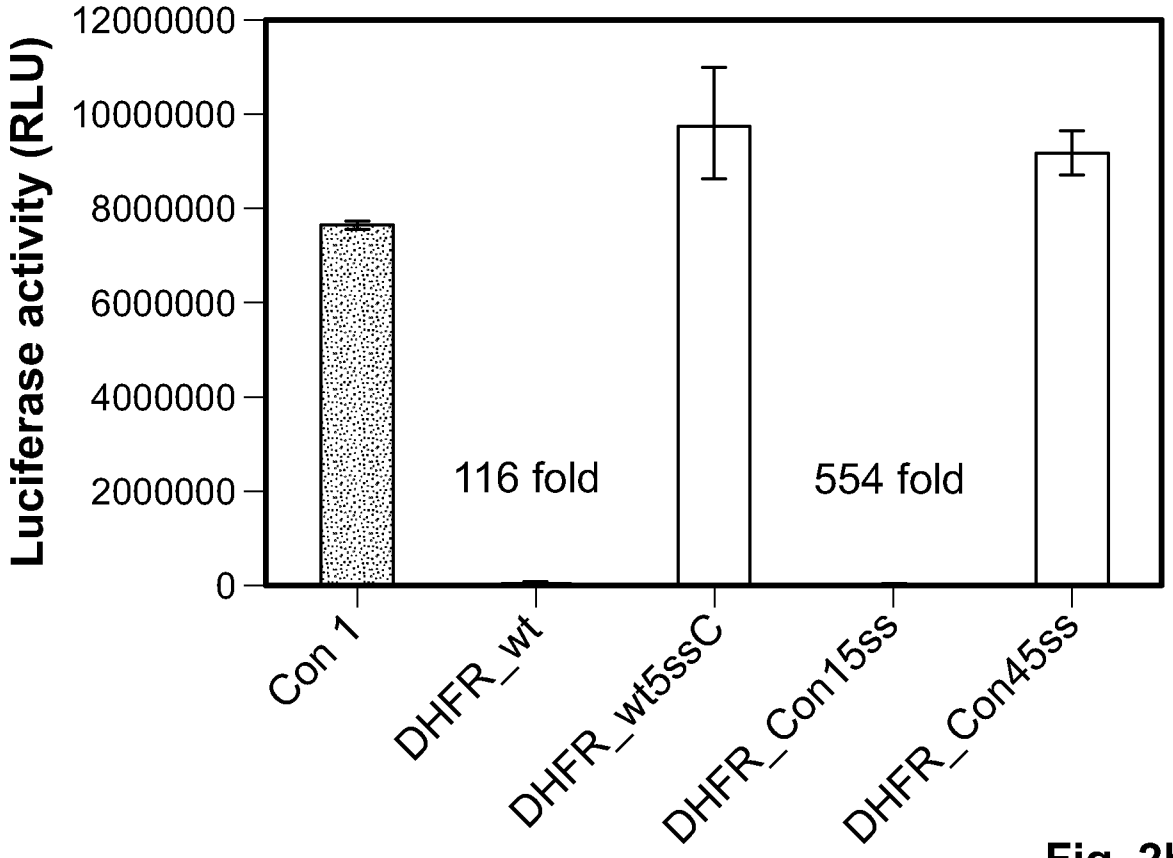

Luciferase activity (RLU)
12000000
10000000
8000000
6000000
4000000
2000000
0
116 fold
554 fold
Con 1
DHFR_wt
DHFR_wt5ssC
DHFR_Con15ss
DHFR_Con45ss

Fig. 2b

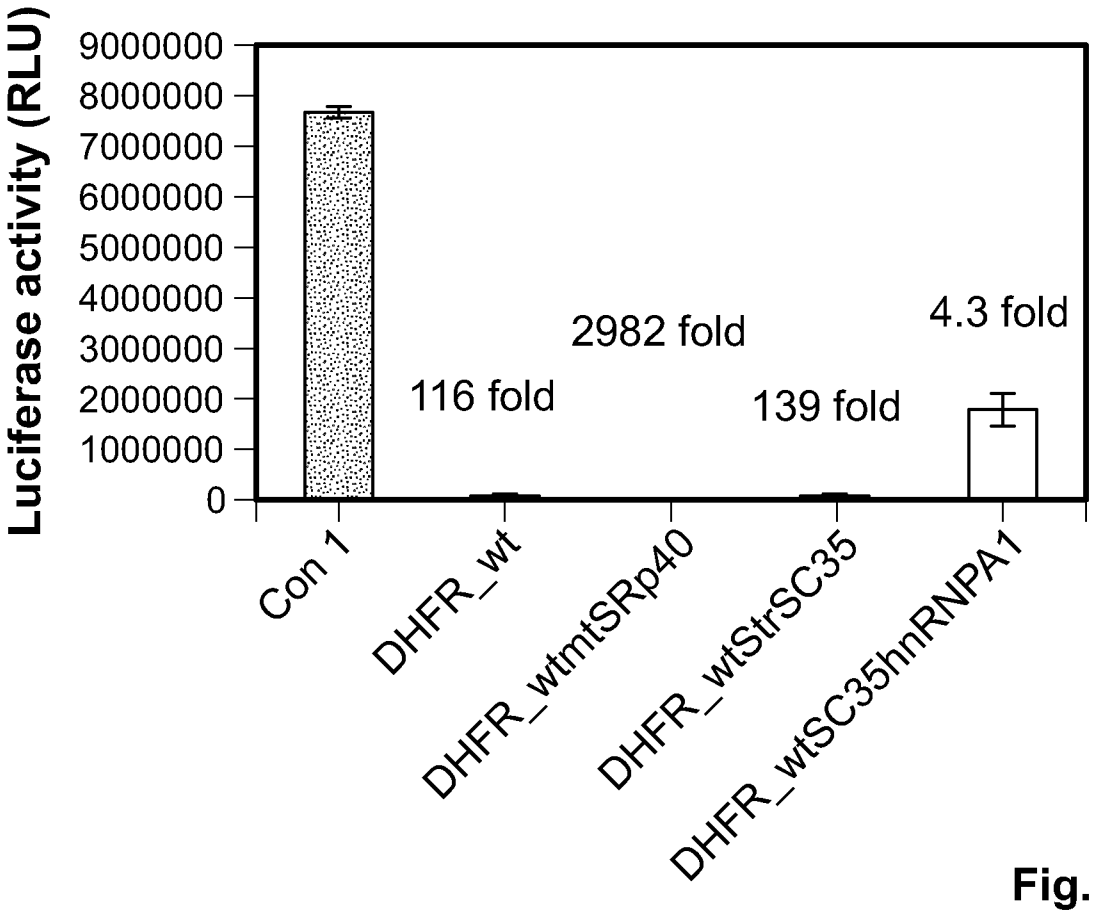

Luciferase activity (RLU)
9000000
8000000
7000000
6000000
5000000
4000000
3000000
2000000
1000000
0
116 fold
2982 fold
139 fold
4.3 fold
Con 1
DHFR_wt
DHFR_wtmtSRp40
DHFR_wtStrSC35
DHFR_wtSC35hnRNPA1

Fig. 2c

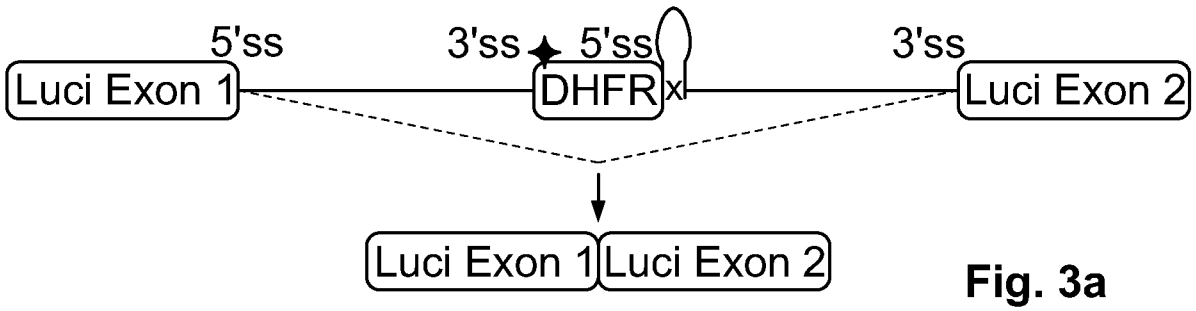


Fig. 3a

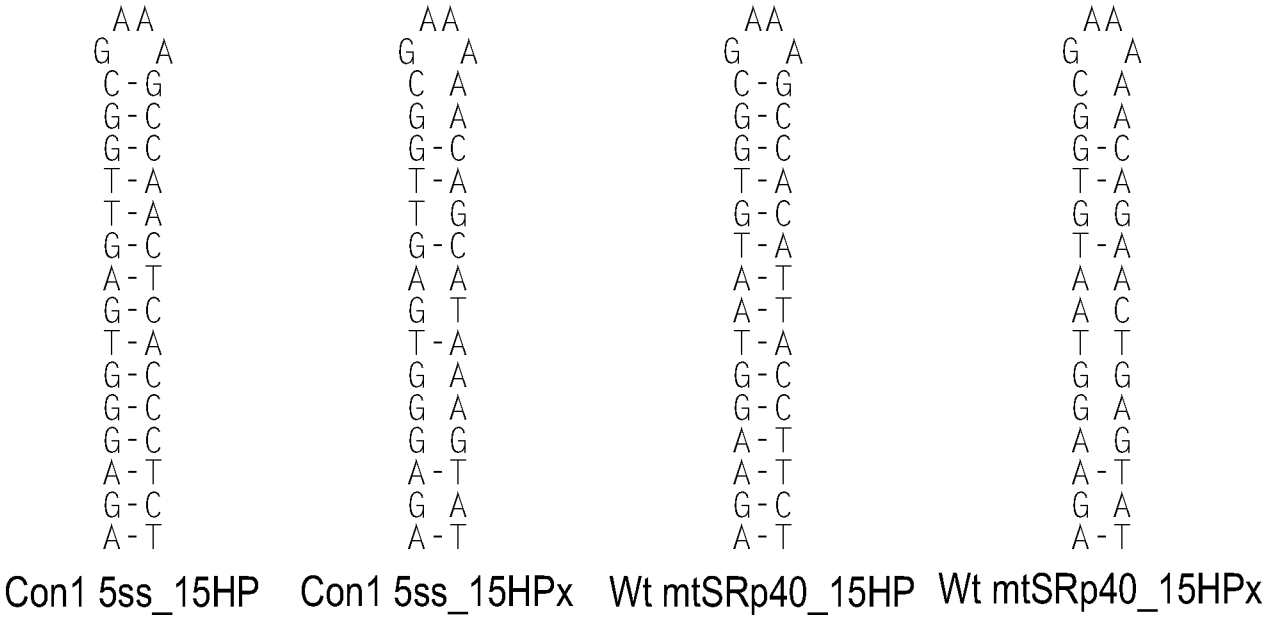


Fig. 3b

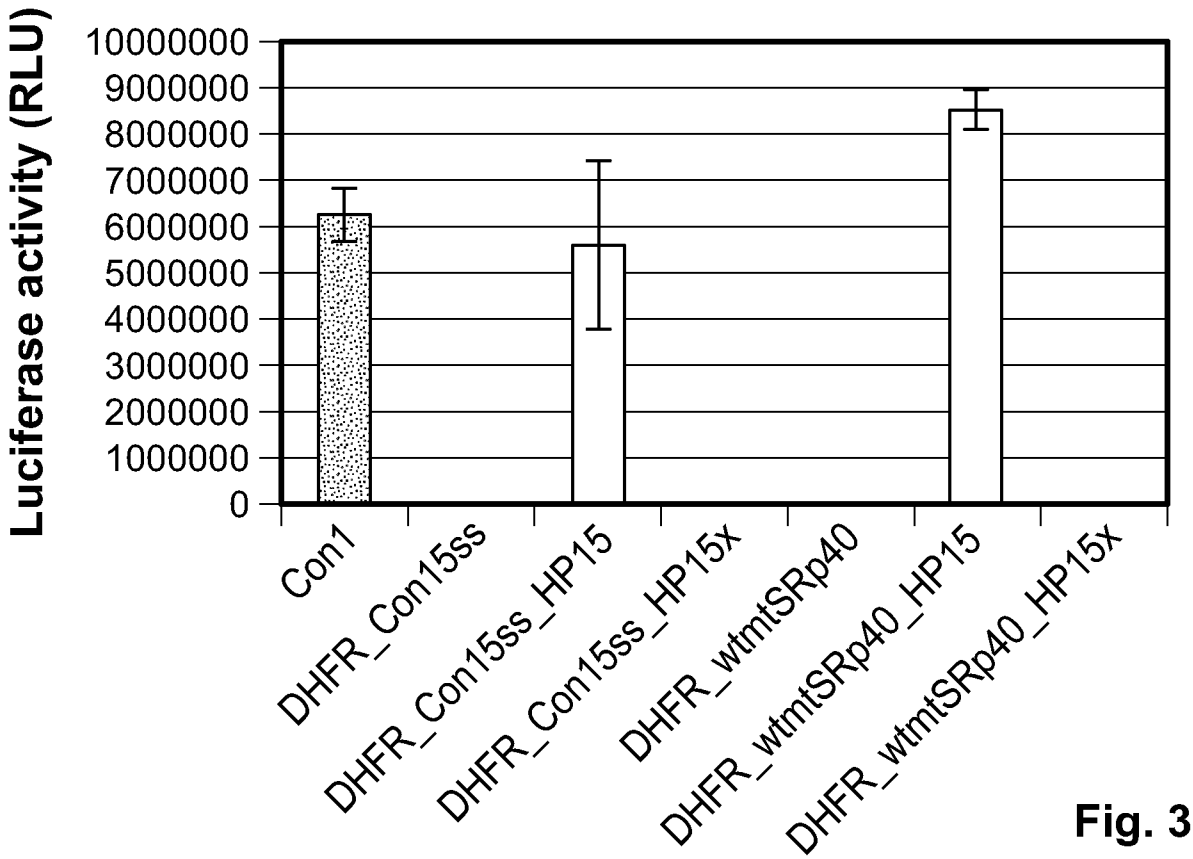


Fig. 3c

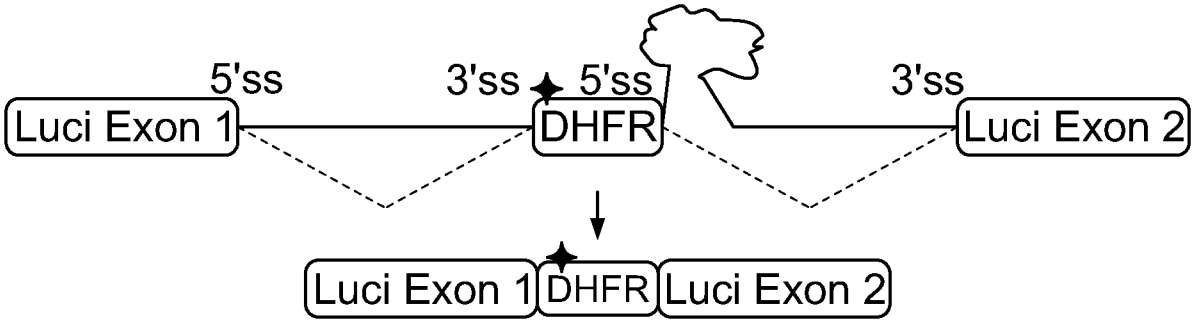

5'ss
3'ss
5'ss
3'ss
Luci Exon 1
DHFR
Luci Exon 2
Luci Exon 1
DHFR
Luci Exon 2

Fig. 4a

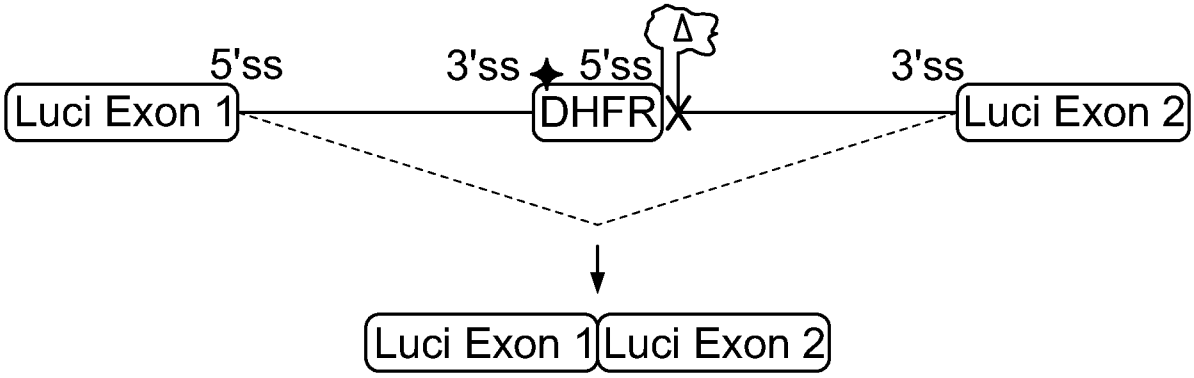
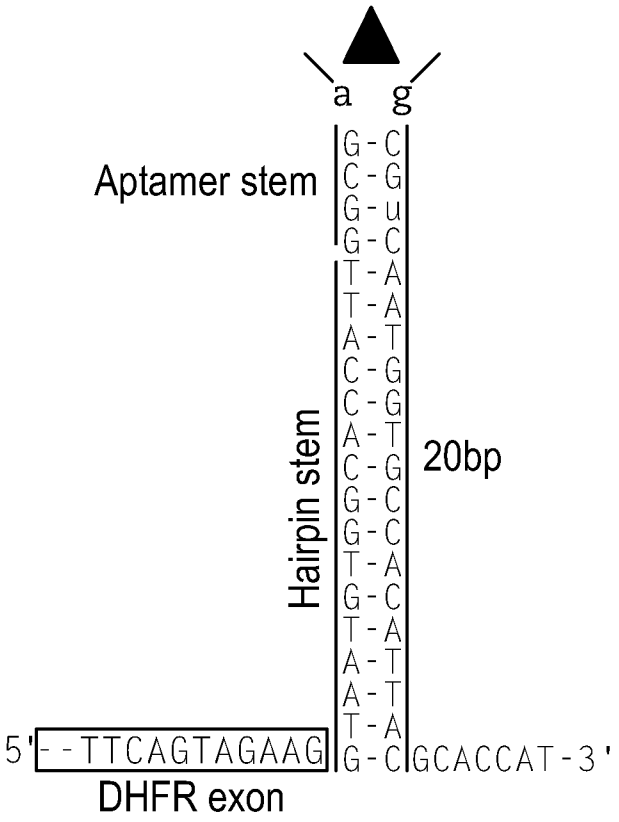
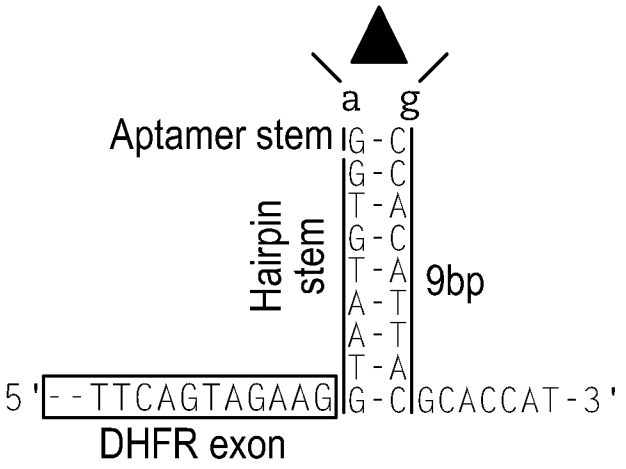

5'ss
3'ss
5'ss
3'ss
Luci Exon 1
DHFR
Luci Exon 2
Luci Exon 1
Luci Exon 2

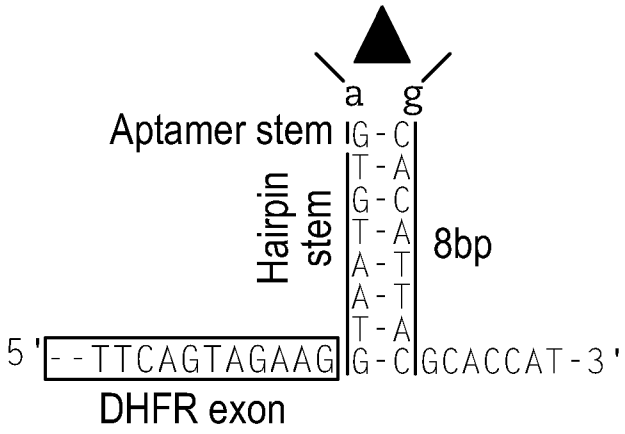
Fig. 4b



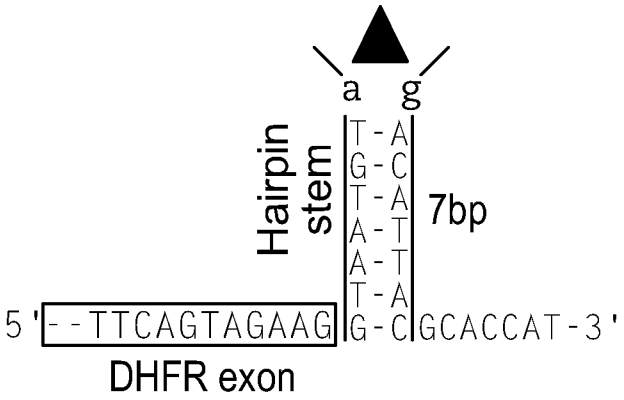
DHFR_Theo1



DHFR_Theo12



DHFR_Theo13



DHFR_Theo14

Fig. 4c

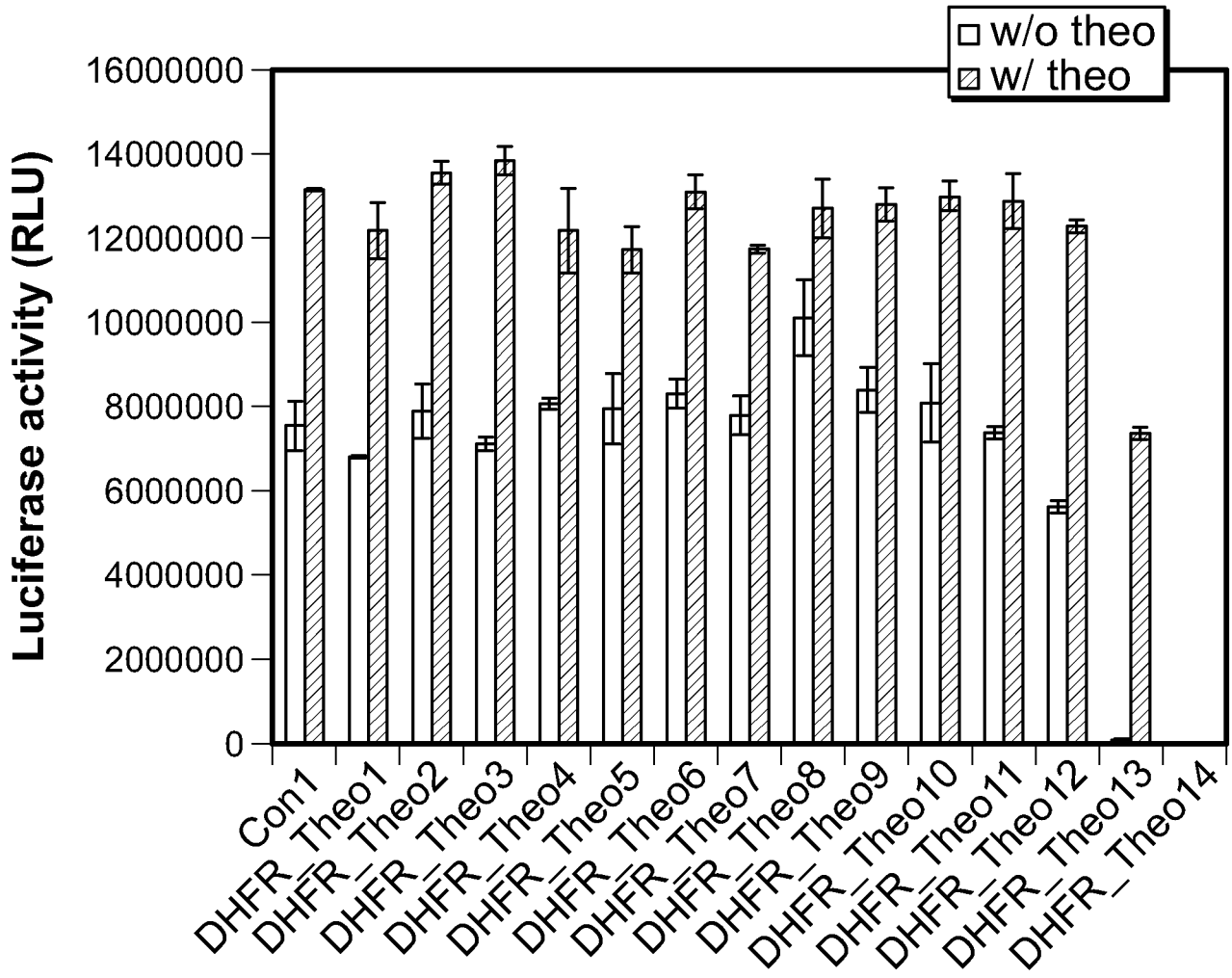
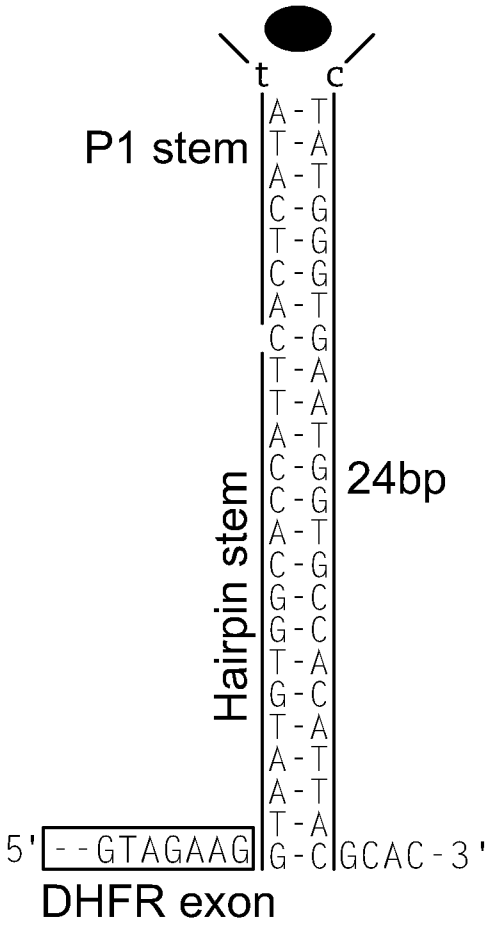
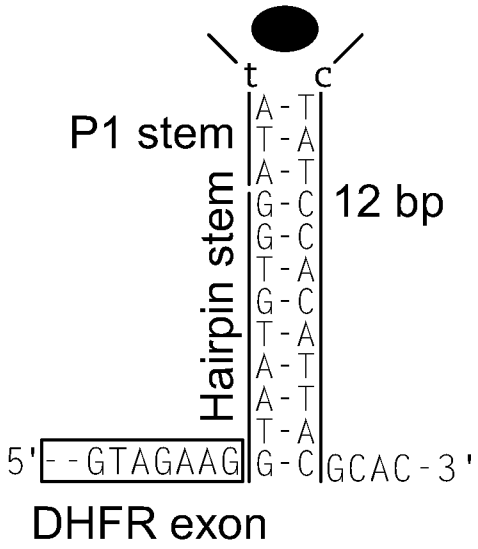

w/o theo
w/ theo
Luciferase activity (RLU)
16000000
14000000
12000000
10000000
8000000
6000000
4000000
2000000
0
Con1
DHFR_Theo1
DHFR_Theo2
DHFR_Theo3
DHFR_Theo4
DHFR_Theo5
DHFR_Theo6
DHFR_Theo7
DHFR_Theo8
DHFR_Theo9
DHFR_Theo10
DHFR_Theo11
DHFR_Theo12
DHFR_Theo13
DHFR_Theo14

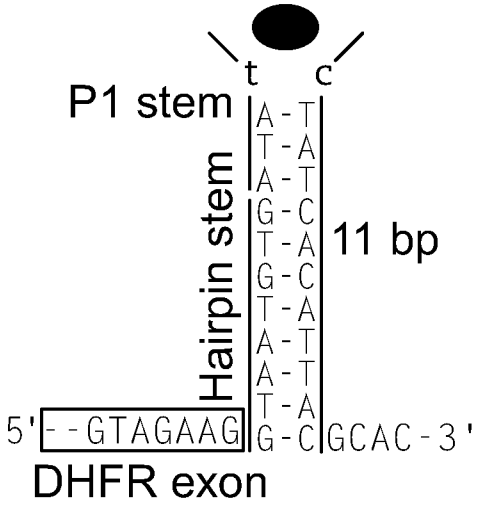
Fig. 4d



DHFR _G1

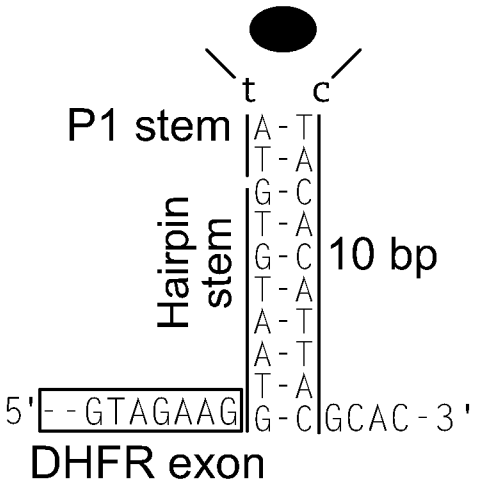


DHFR _G13

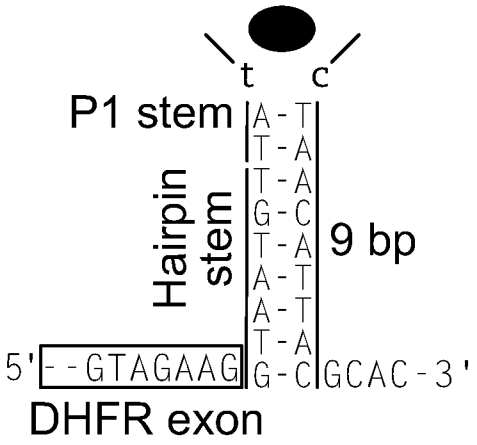


DHFR _G14

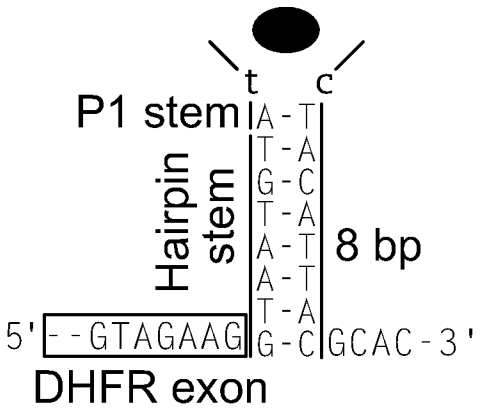
Fig. 5a



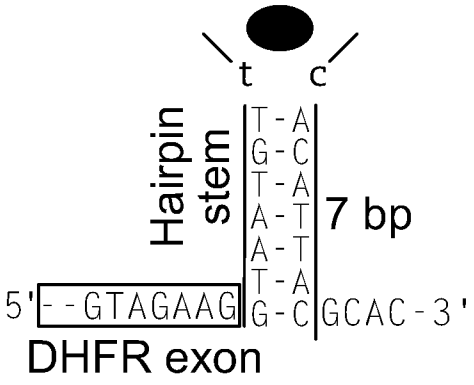
DHFR _G15



DHFR _G16



DHFR _G17



DHFR _G18

Fig. 5a (Cont.)

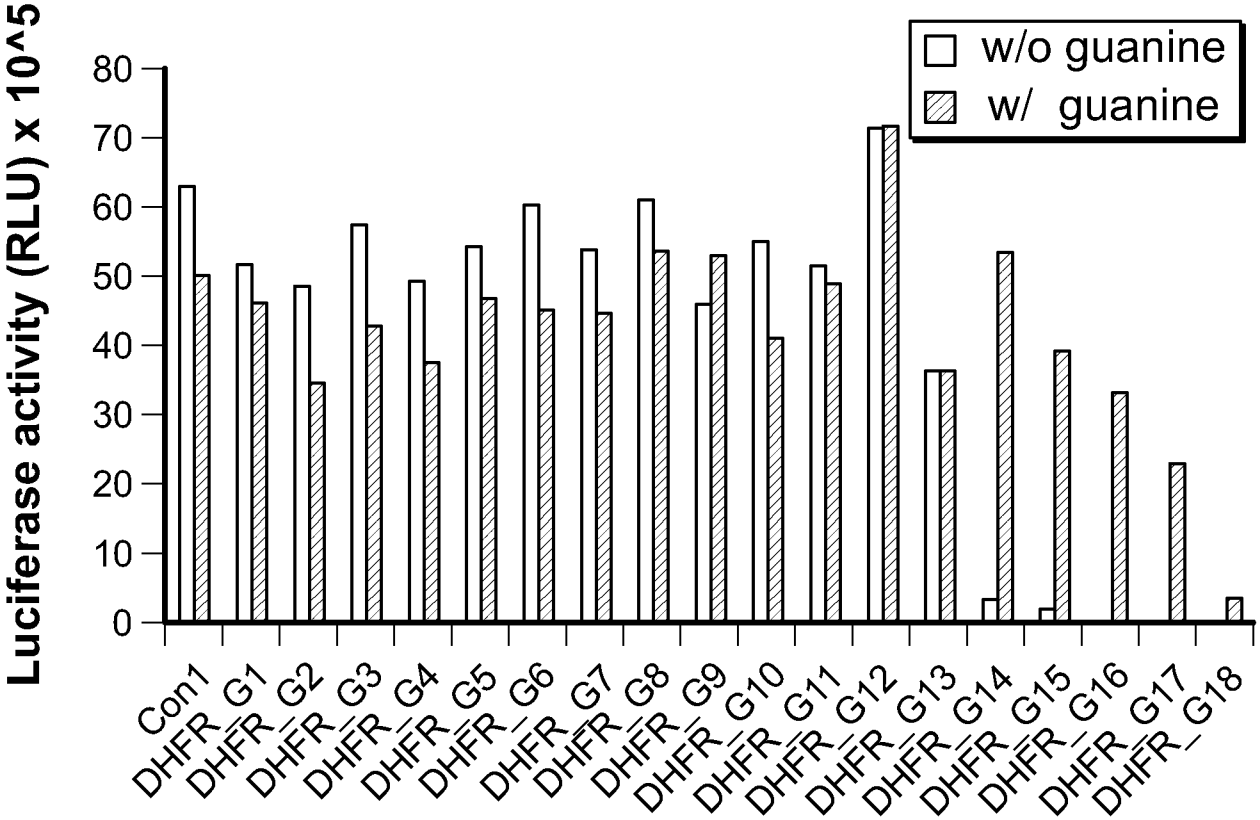

Luciferase activity (RLU) x 10^5
80
70
60
50
40
30
20
10
0
w/o guanine
w/ guanine
Con1
DHFR_G1
DHFR_G2
DHFR_G3
DHFR_G4
DHFR_G5
DHFR_G6
DHFR_G7
DHFR_G8
DHFR_G9
DHFR_G10
DHFR_G11
DHFR_G12
DHFR_G13
DHFR_G14
DHFR_G15
DHFR_G16
DHFR_G17
DHFR_G18

FIG. 5b

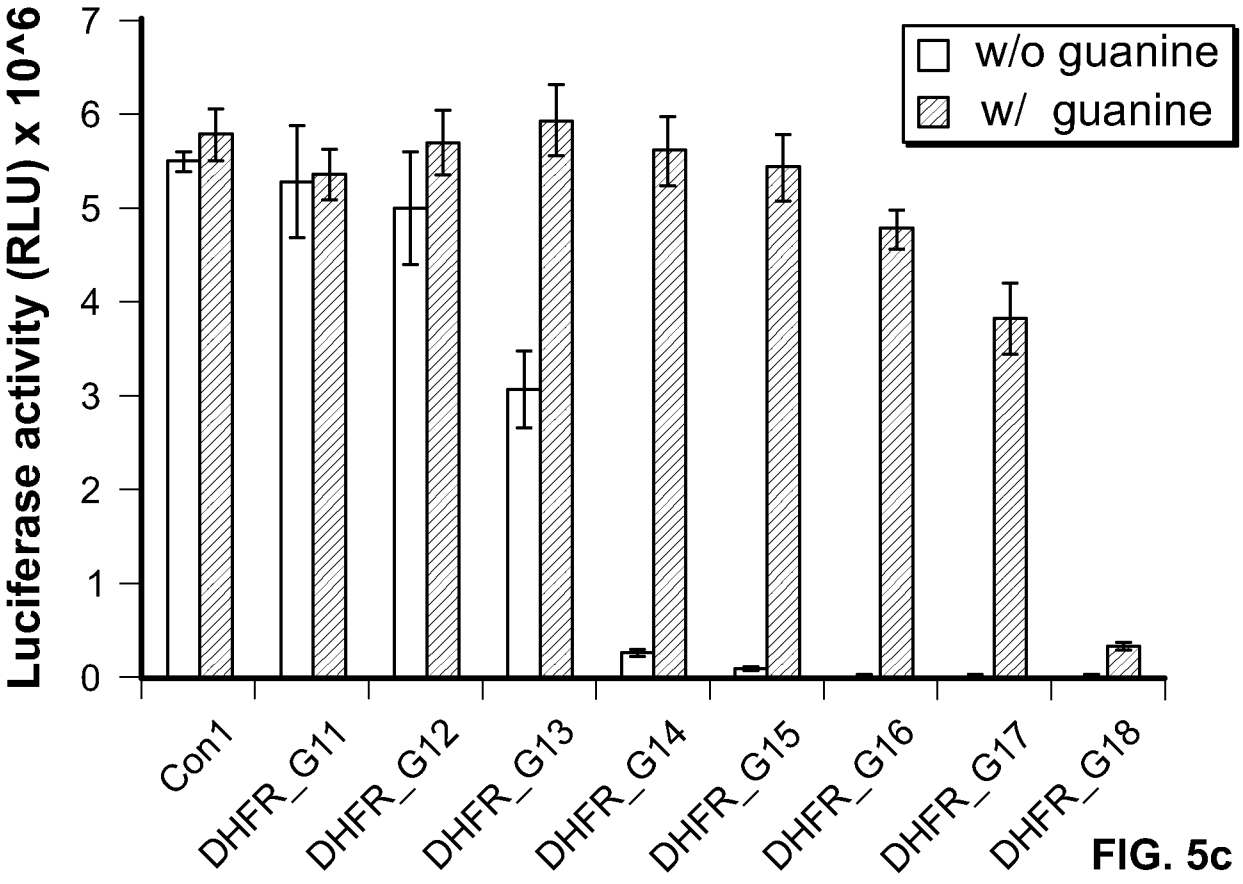

Luciferase activity (RLU) x 10^6
7
6
5
4
3
2
1
0
w/o guanine
w/ guanine
Con1
DHFR_G11
DHFR_G12
DHFR_G13
DHFR_G14
DHFR_G15
DHFR_G16
DHFR_G17
DHFR_G18

FIG. 5c

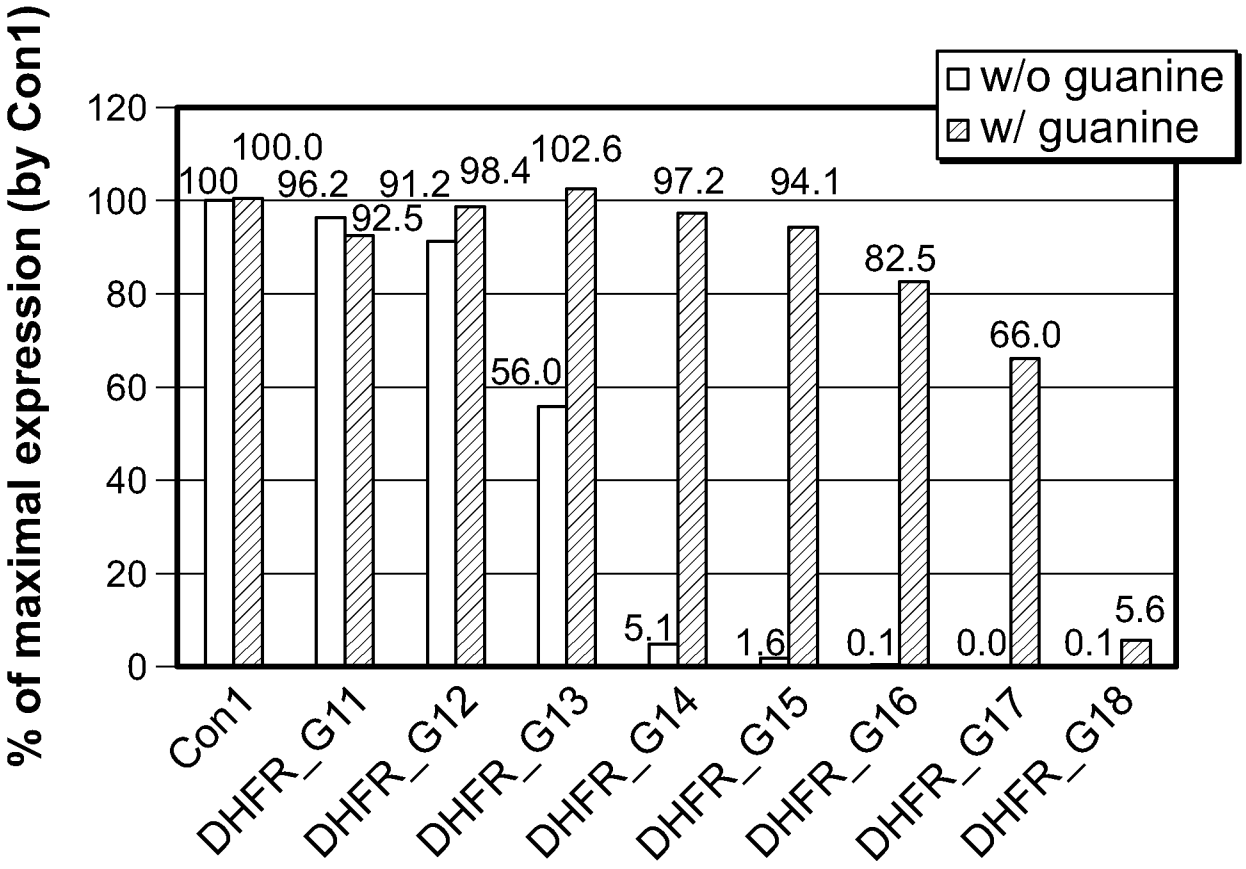


Fig. 5d

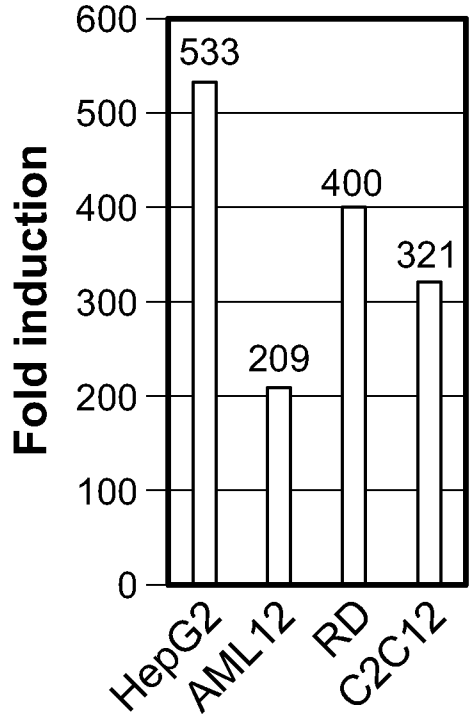


Fig. 5e

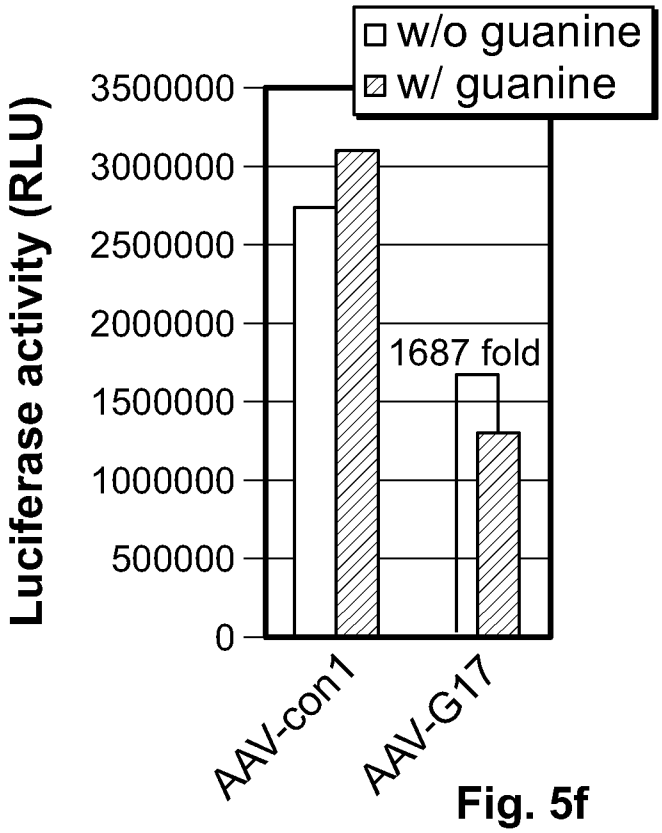


Fig. 5f

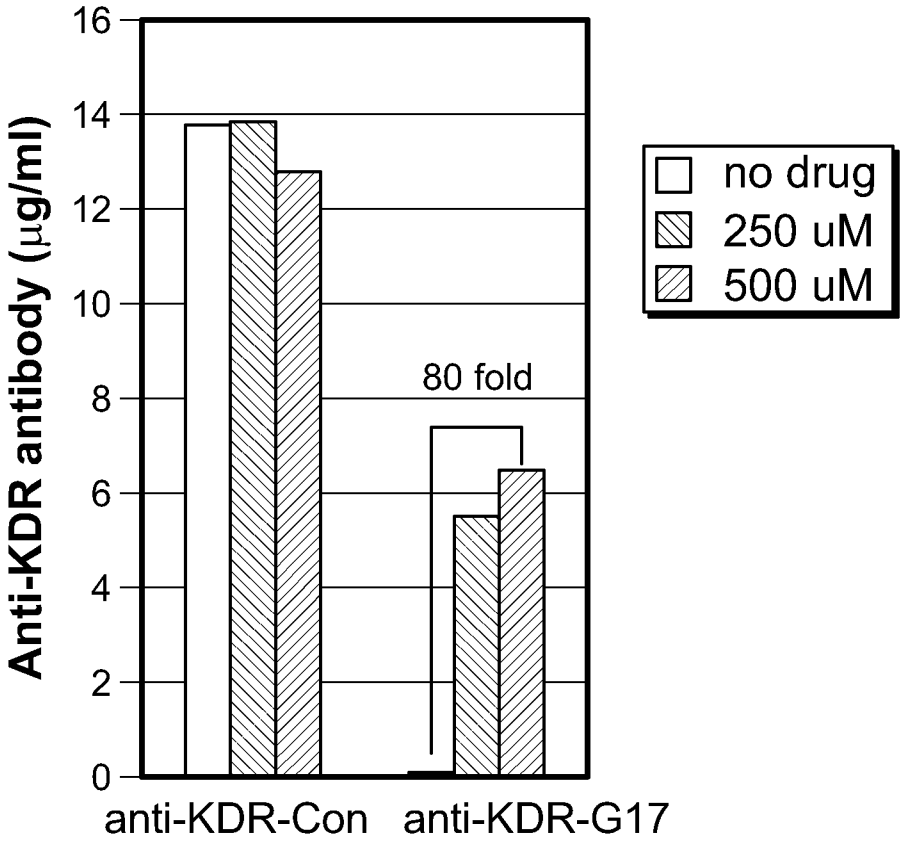

Anti-KDR antibody (μg/ml)
16
14
12
10
8
6
4
2
0
80 fold
no drug
250 uM
500 uM
anti-KDR-Con
anti-KDR-G17

Fig. 5g

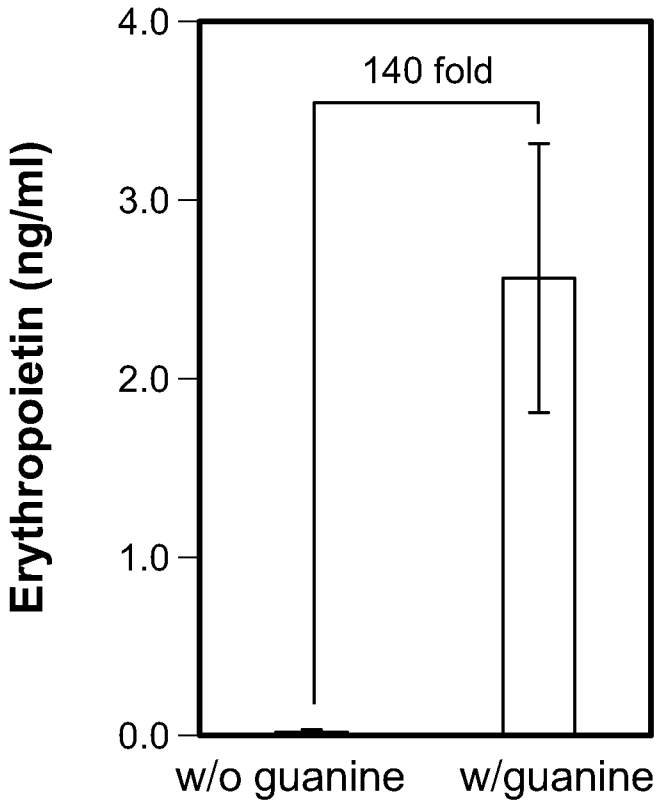

Erythropoietin (ng/ml)
4.0
3.0
2.0
1.0
0.0
140 fold
w/o guanine
w/guanine

Fig. 5h

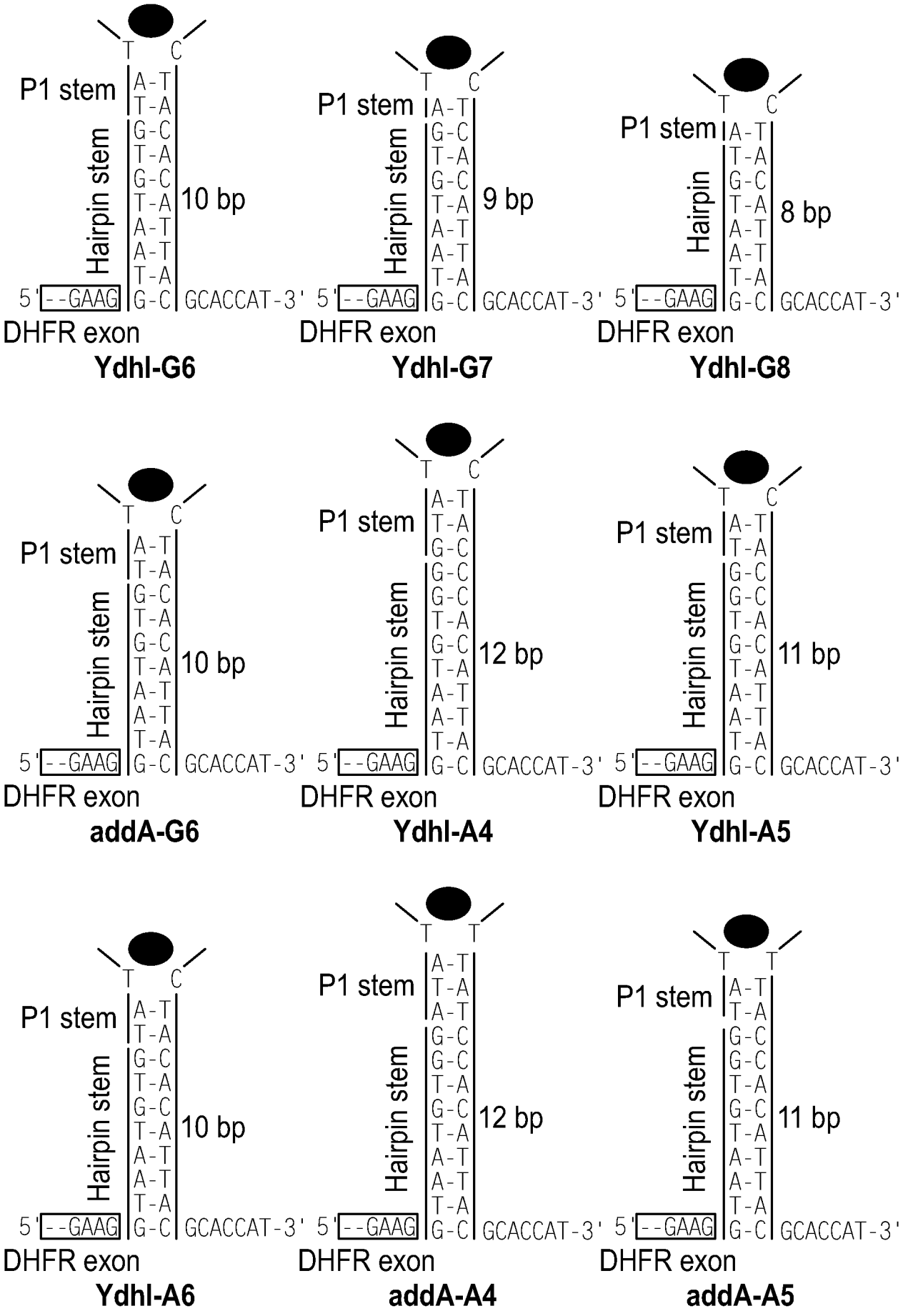

T C
P1 stem
A-T
T-A
Hairpin stem
G-C
T-A
G-C
T-A
A-T
A-T
T-A
10 bp
5'--GAAG
G-C
GCACCAT-3'
DHFR exon
YdhI-G6
9 bp
YdhI-G7
8 bp
YdhI-G8
addA-G6
12 bp
YdhI-A4
11 bp
YdhI-A5
T T
YdhI-A6
addA-A4
addA-A5

FIG. 6a

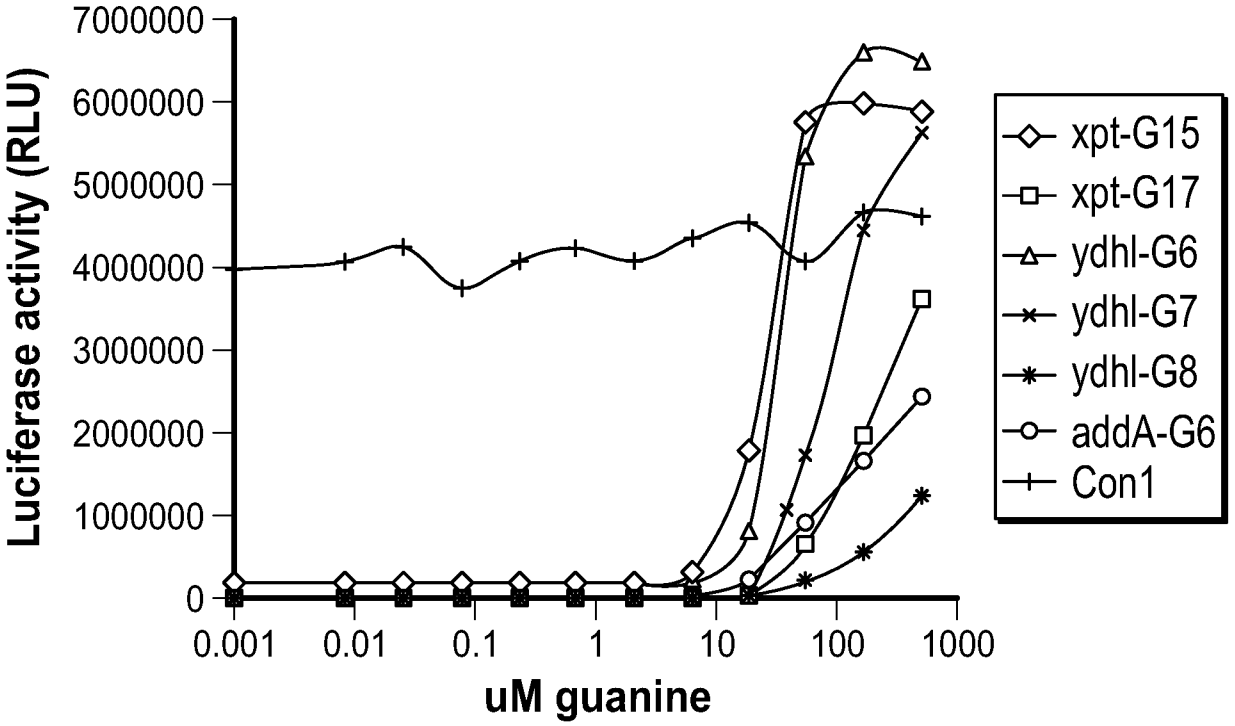

Luciferase activity (RLU)
7000000
6000000
5000000
4000000
3000000
2000000
1000000
0
0.001
0.01
0.1
1
10
100
1000
uM guanine
xpt-G15
xpt-G17
ydhl-G6
ydhl-G7
ydhl-G8
addA-G6
Con1

Fig. 6b

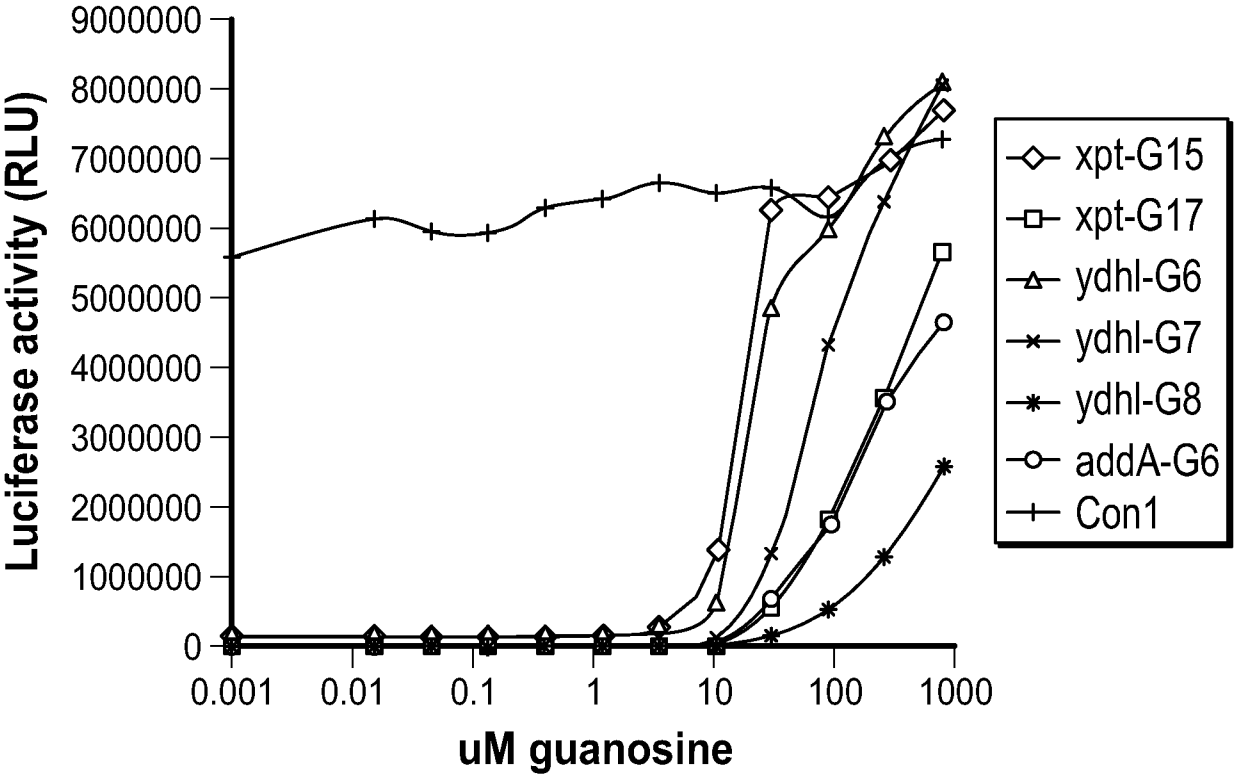

Luciferase activity (RLU)
9000000
8000000
7000000
6000000
5000000
4000000
3000000
2000000
1000000
0
0.001
0.01
0.1
1
10
100
1000
uM guanosine
xpt-G15
xpt-G17
ydhl-G6
ydhl-G7
ydhl-G8
addA-G6
Con1

Fig. 6c

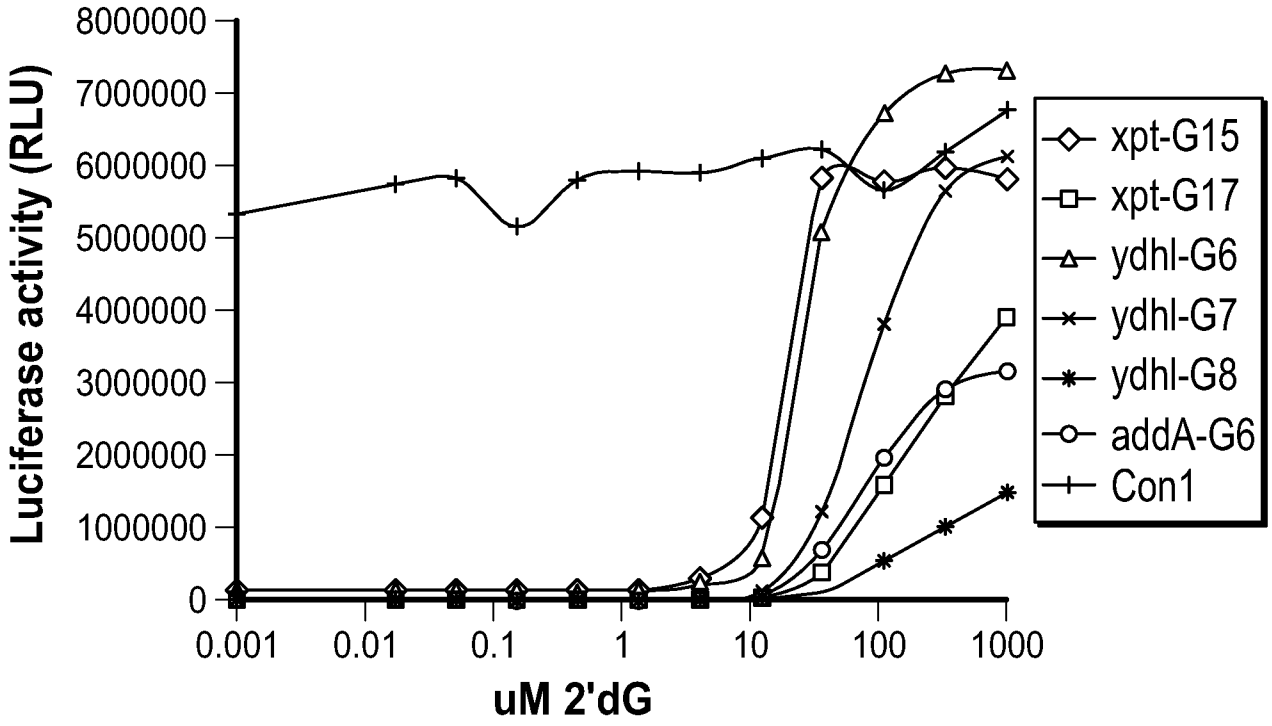

8000000
7000000
6000000
5000000
4000000
3000000
2000000
1000000
0
Luciferase activity (RLU)
0.001
0.01
0.1
1
10
100
1000
uM 2'dG
xpt-G15
xpt-G17
ydhl-G6
ydhl-G7
ydhl-G8
addA-G6
Con1

Fig. 6d

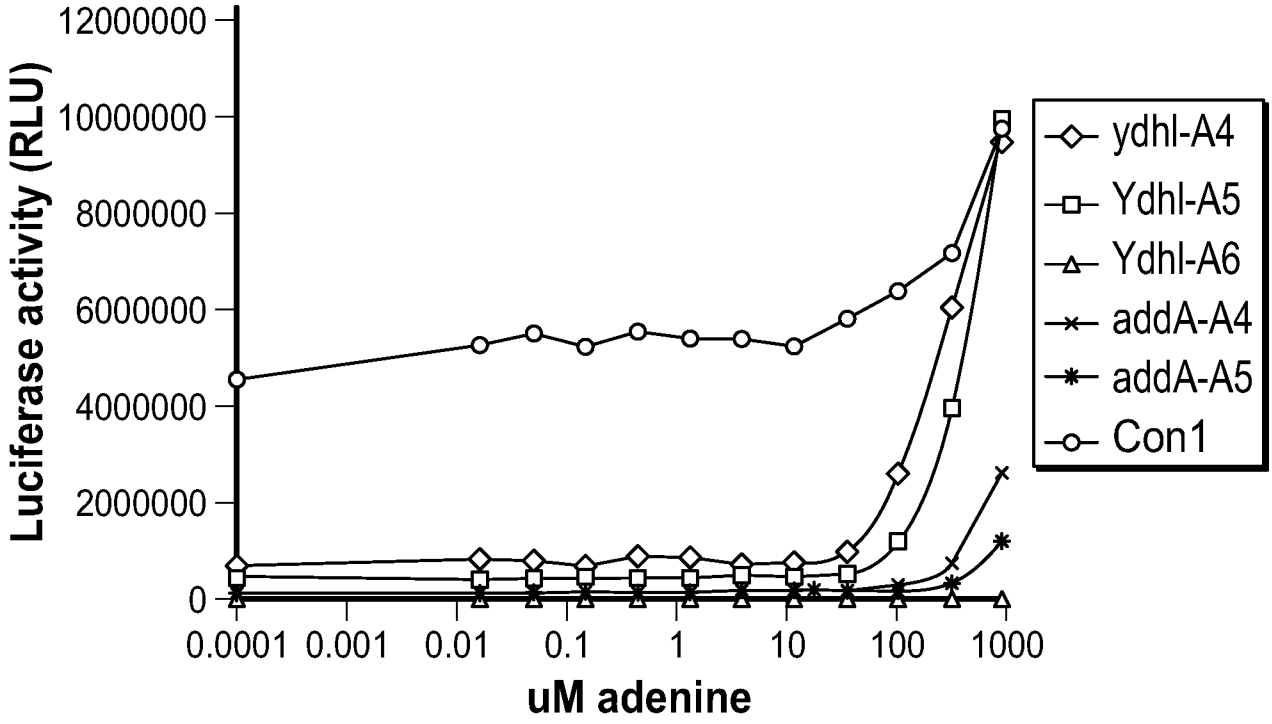

12000000
10000000
8000000
6000000
4000000
2000000
0
Luciferase activity (RLU)
0.0001
0.001
0.01
0.1
1
10
100
1000
uM adenine
ydhl-A4
Ydhl-A5
Ydhl-A6
addA-A4
addA-A5
Con1

Fig. 6e

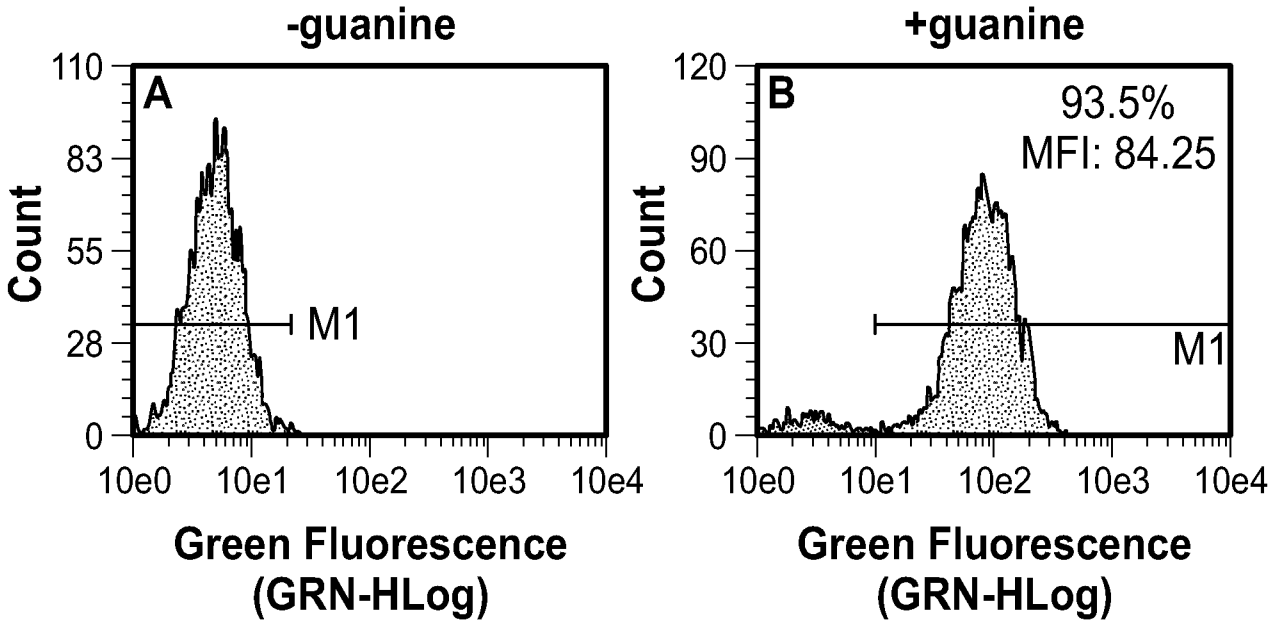

-guanine
+guanine
A
B
93.5%
MFI: 84.25
M1
M1
Count
Count
110
83
55
28
0
120
90
60
30
0
10e0
10e1
10e2
10e3
10e4
Green Fluorescence
(GRN-HLog)
Green Fluorescence
(GRN-HLog)

Fig. 7a

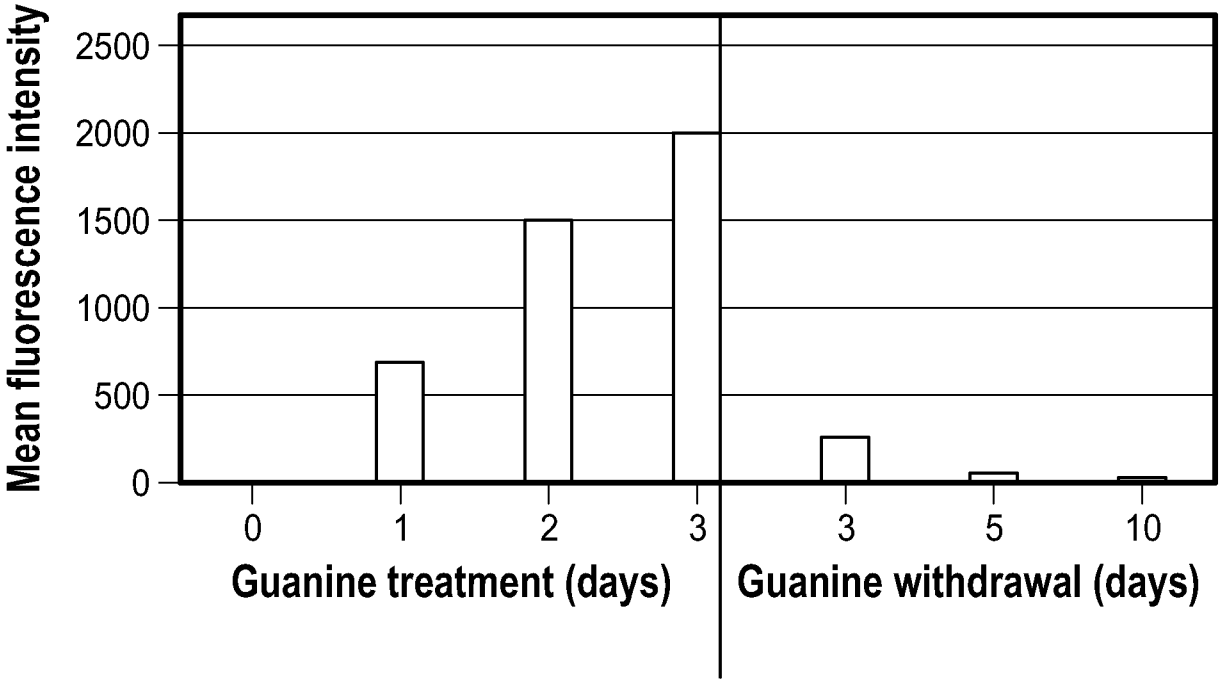

Mean fluorescence intensity
2500
2000
1500
1000
500
0
0
1
2
3
3
5
10
Guanine treatment (days)
Guanine withdrawal (days)

Fig. 7b

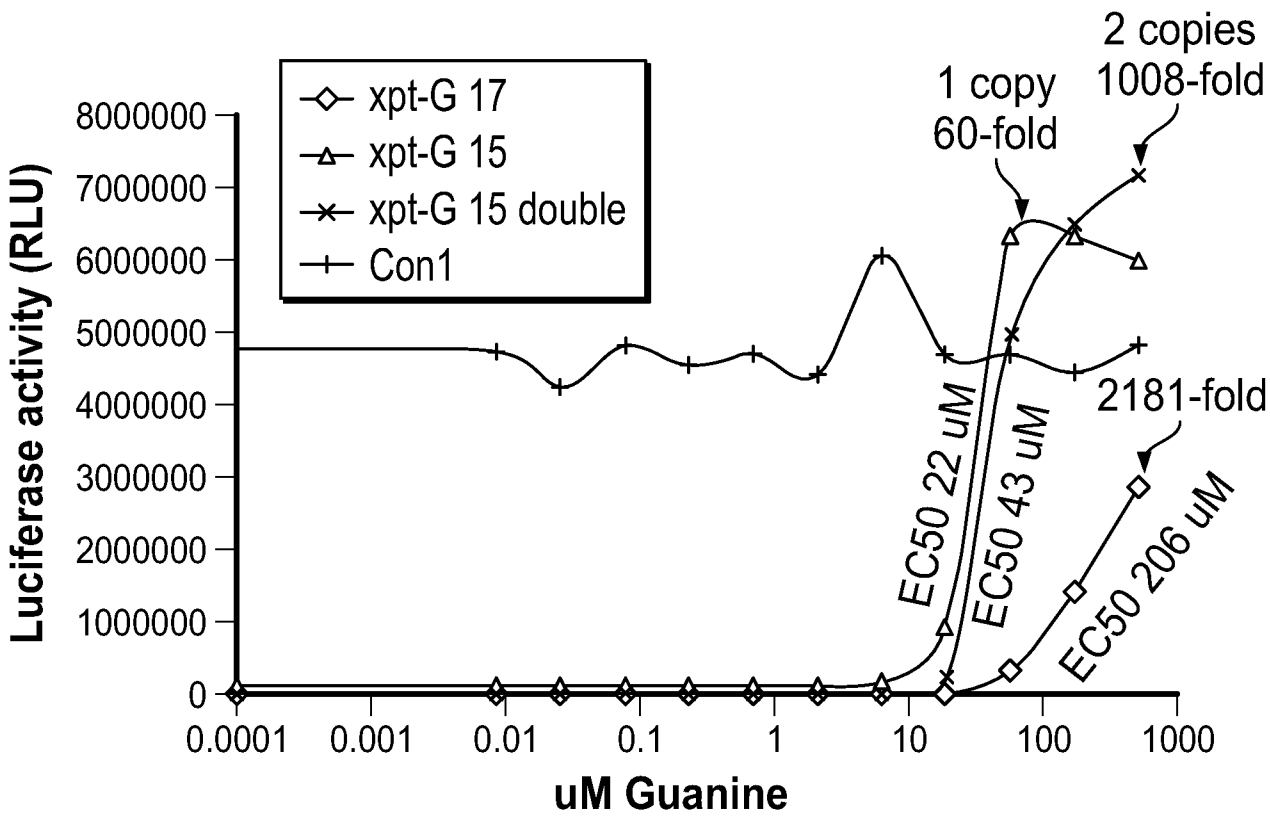

xpt-G 17
xpt-G 15
xpt-G 15 double
Con1
1 copy
60-fold
2 copies
1008-fold
2181-fold
EC50 22 uM
EC50 43 uM
EC50 206 uM
Luciferase activity (RLU)
8000000
7000000
6000000
5000000
4000000
3000000
2000000
1000000
0
0.0001
0.001
0.01
0.1
1
10
100
1000
uM Guanine

Fig. 8a

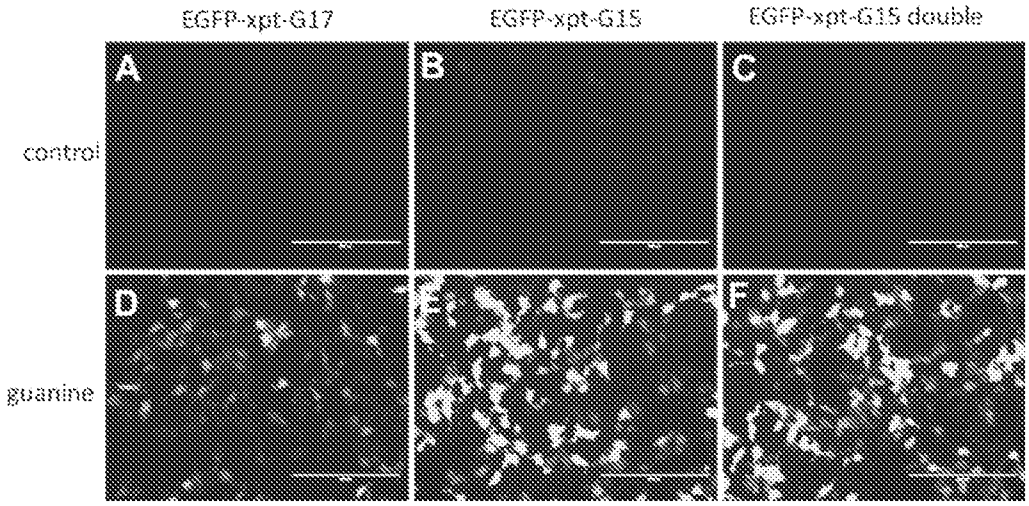

EGFP-xpt-G17
EGFP-xpt-G15
EGFP-xpt-G15 double
A
B
C
control
D
E
F
guanine

Fig. 8b

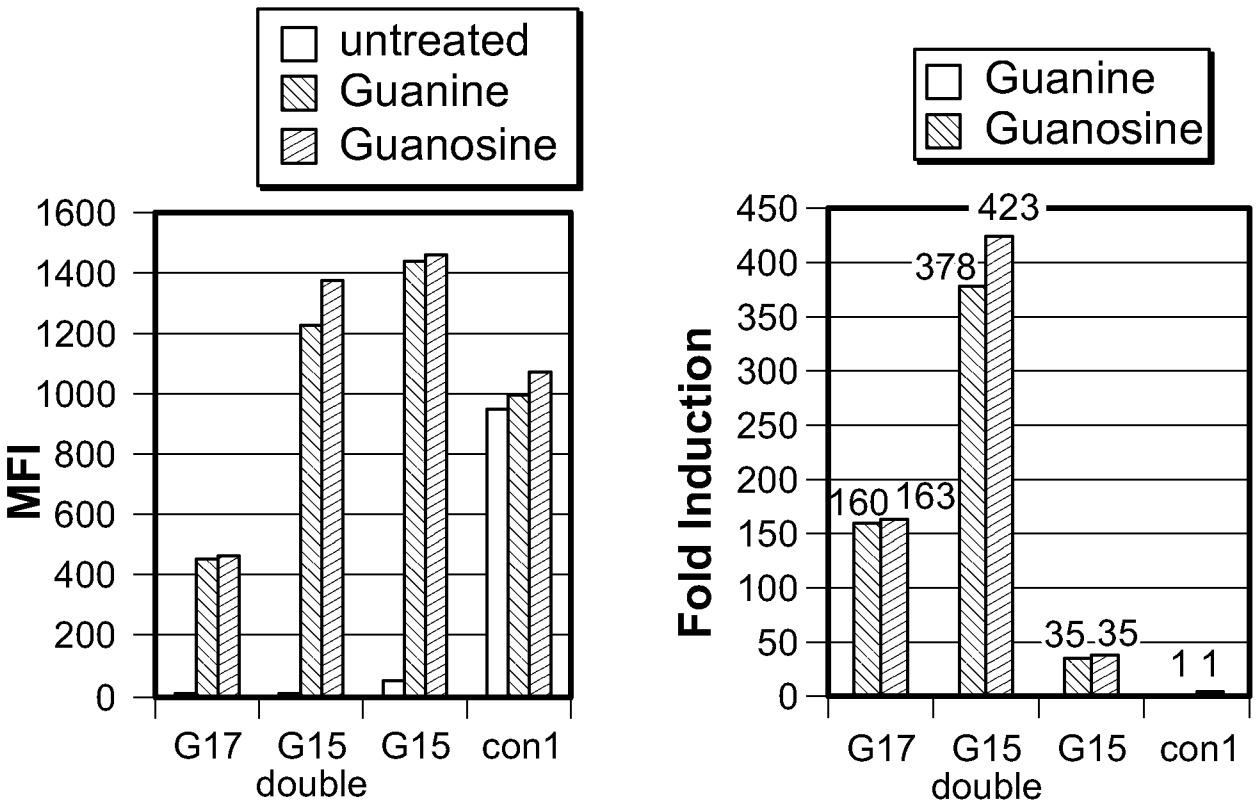

untreated
Guanine
Guanosine
MFI
1600
1400
1200
1000
800
600
400
200
0
G17
G15 double
G15
con1
Guanine
Guanosine
Fold Induction
450
400
350
300
250
200
150
100
50
0
423
378
160
163
35
35
1
1
G17
G15 double
G15
con1

Fig. 8c

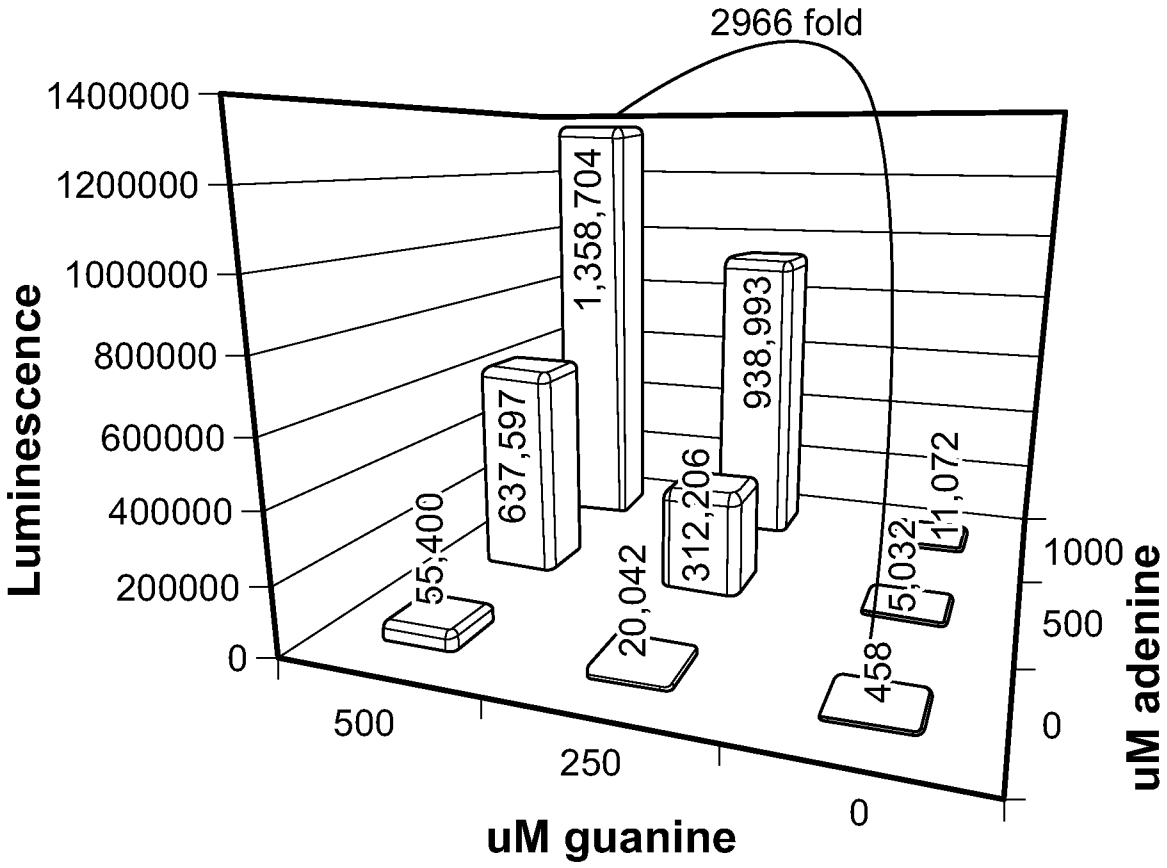

2966 fold
Luminescence
1400000
1200000
1000000
800000
600000
400000
200000
0
1,358,704
938,993
637,597
312,206
55,400
20,042
11,072
5,032
458
500
250
0
uM guanine
1000
500
0
uM adenine

Fig. 8d

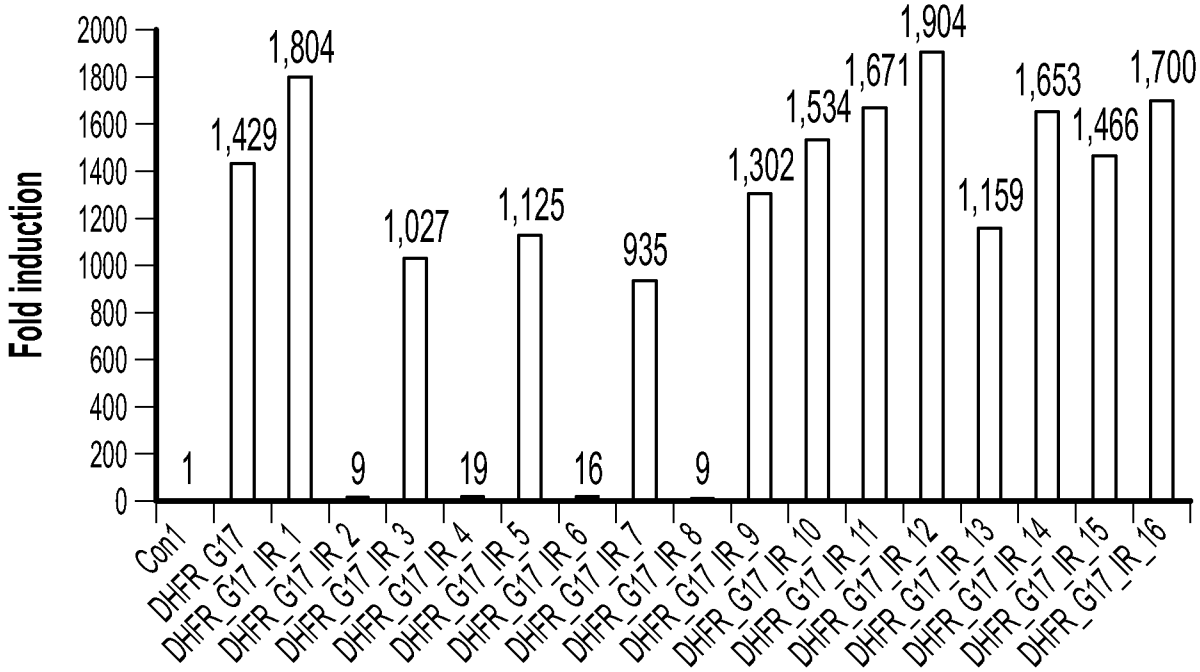

Fold induction
2000
1800
1600
1400
1200
1000
800
600
400
200
0
1
1,429
1,804
9
1,027
19
1,125
16
935
9
1,302
1,534
1,671
1,904
1,159
1,653
1,466
1,700
Con1
DHFR G17
DHFR G17 IR 1
DHFR G17 IR 2
DHFR G17 IR 3
DHFR G17 IR 4
DHFR G17 IR 5
DHFR G17 IR 6
DHFR G17 IR 7
DHFR G17 IR 8
DHFR G17 IR 9
DHFR G17 IR 10
DHFR G17 IR 11
DHFR G17 IR 12
DHFR G17 IR 13
DHFR G17 IR 14
DHFR G17 IR 15
DHFR G17 IR 16

Fig. 9a

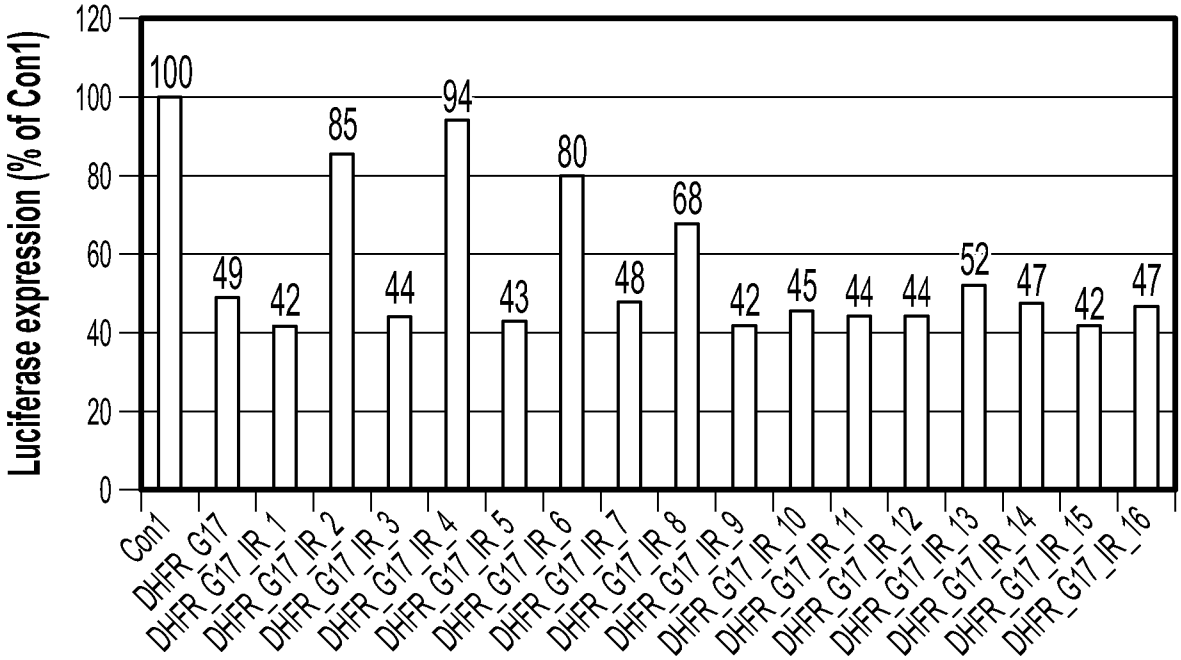

Luciferase expression (% of Con1)
120
100
80
60
40
20
0
100
49
42
85
44
94
43
80
48
68
42
45
44
44
52
47
42
47
Con1
DHFR G17
DHFR G17 IR 1
DHFR G17 IR 2
DHFR G17 IR 3
DHFR G17 IR 4
DHFR G17 IR 5
DHFR G17 IR 6
DHFR G17 IR 7
DHFR G17 IR 8
DHFR G17 IR 9
DHFR G17 IR 10
DHFR G17 IR 11
DHFR G17 IR 12
DHFR G17 IR 13
DHFR G17 IR 14
DHFR G17 IR 15
DHFR G17 IR 16

Fig. 9b

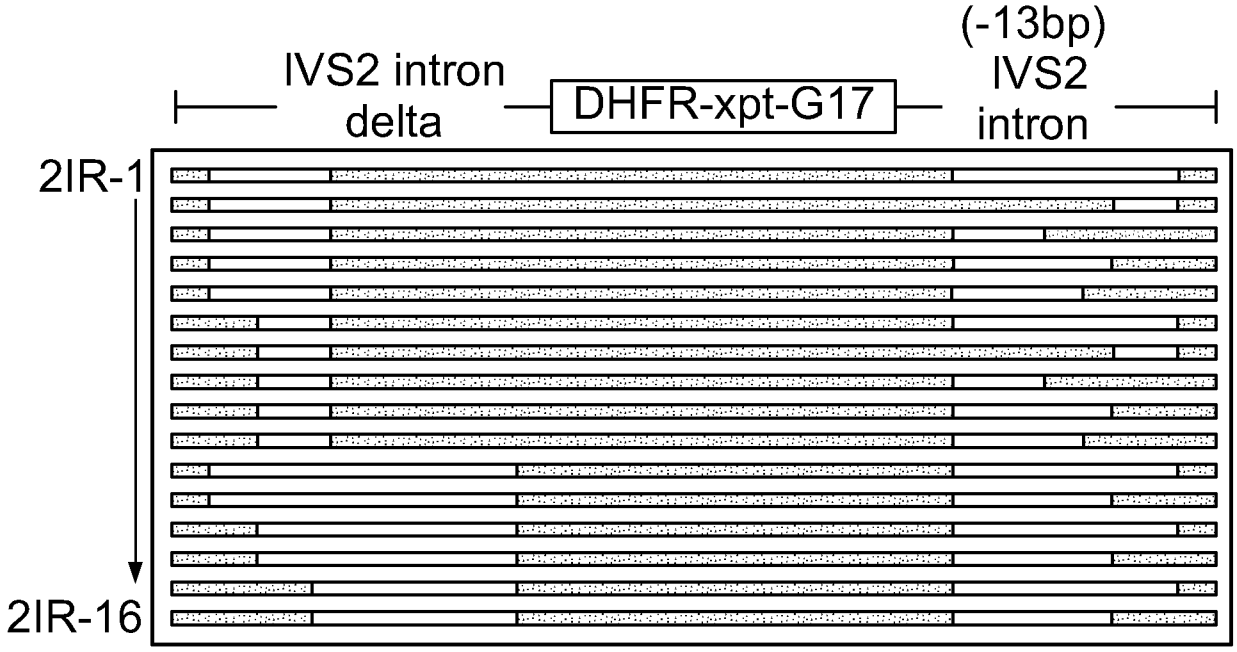

IVS2 intron delta
DHFR-xpt-G17
(-13bp) IVS2 intron
2IR-1
2IR-16

Fig. 9c

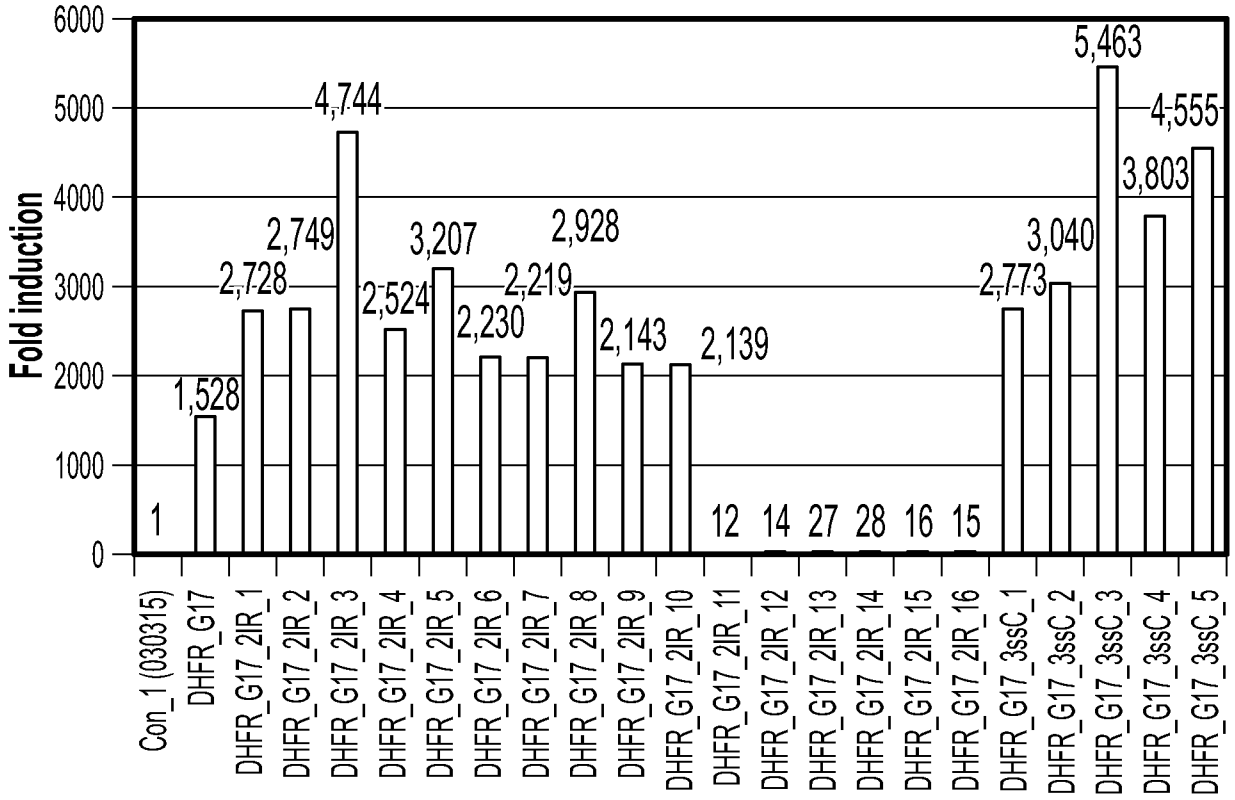

Fold induction
6000
5000
4000
3000
2000
1000
0
1
1,528
2,728
2,749
4,744
2,524
3,207
2,230
2,219
2,928
2,143
2,139
12
14
27
28
16
15
2,773
3,040
5,463
4,555
3,803
Con_1 (030315)
DHFR_G17
DHFR_G17_2IR_1
DHFR_G17_2IR_2
DHFR_G17_2IR_3
DHFR_G17_2IR_4
DHFR_G17_2IR_5
DHFR_G17_2IR_6
DHFR_G17_2IR_7
DHFR_G17_2IR_8
DHFR_G17_2IR_9
DHFR_G17_2IR_10
DHFR_G17_2IR_11
DHFR_G17_2IR_12
DHFR_G17_2IR_13
DHFR_G17_2IR_14
DHFR_G17_2IR_15
DHFR_G17_2IR_16
DHFR_G17_3ssC_1
DHFR_G17_3ssC_2
DHFR_G17_3ssC_3
DHFR_G17_3ssC_4
DHFR_G17_3ssC_5

Fig. 9d

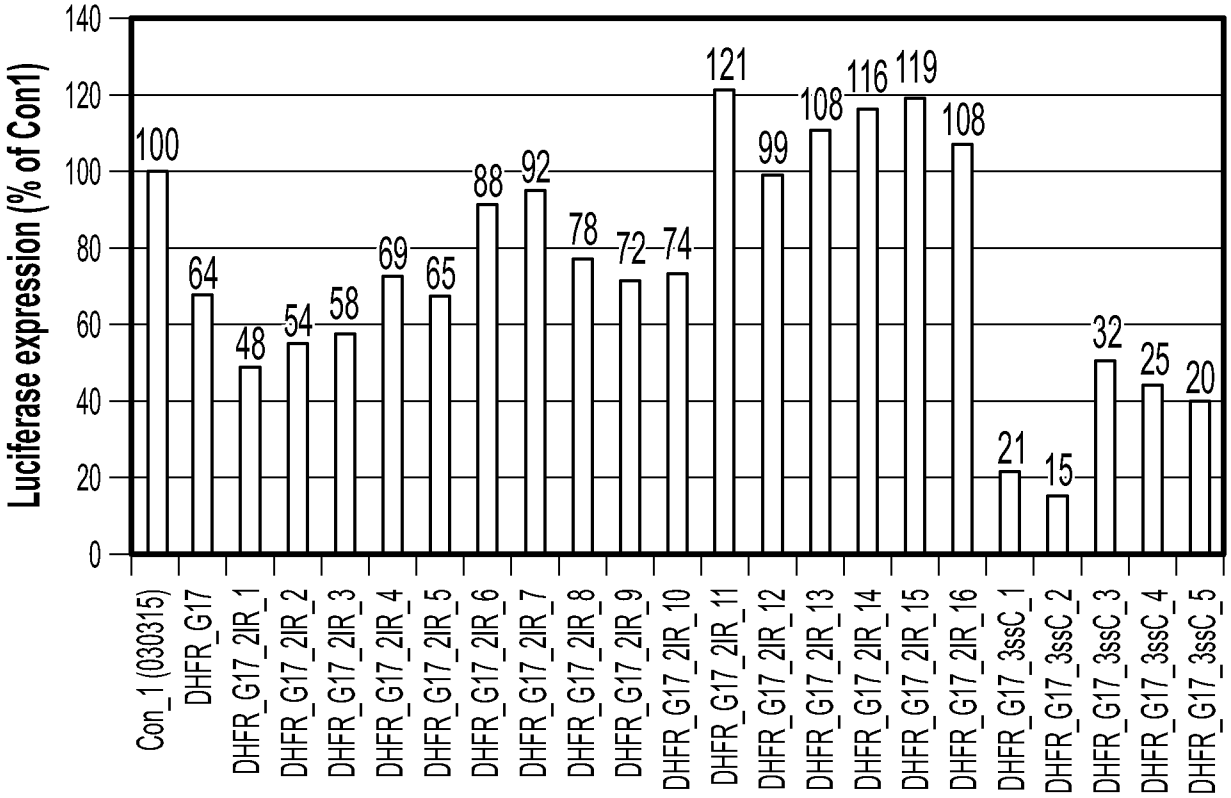

Luciferase expression (% of Con1)
140
120
100
80
60
40
20
0
100
64
48
54
58
69
65
88
92
78
72
74
121
99
108
116
119
108
21
15
32
25
20
Con_1 (030315)
DHFR_G17
DHFR_G17_2IR_1
DHFR_G17_2IR_2
DHFR_G17_2IR_3
DHFR_G17_2IR_4
DHFR_G17_2IR_5
DHFR_G17_2IR_6
DHFR_G17_2IR_7
DHFR_G17_2IR_8
DHFR_G17_2IR_9
DHFR_G17_2IR_10
DHFR_G17_2IR_11
DHFR_G17_2IR_12
DHFR_G17_2IR_13
DHFR_G17_2IR_14
DHFR_G17_2IR_15
DHFR_G17_2IR_16
DHFR_G17_3ssC_1
DHFR_G17_3ssC_2
DHFR_G17_3ssC_3
DHFR_G17_3ssC_4
DHFR_G17_3ssC_5

Fig. 9e

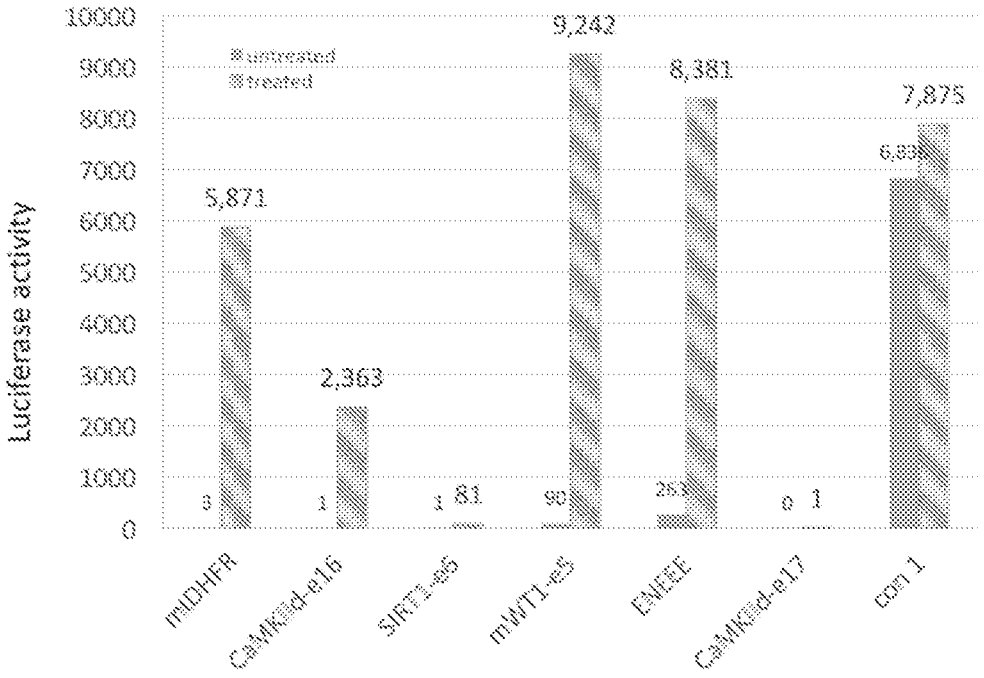


Fig. 10a

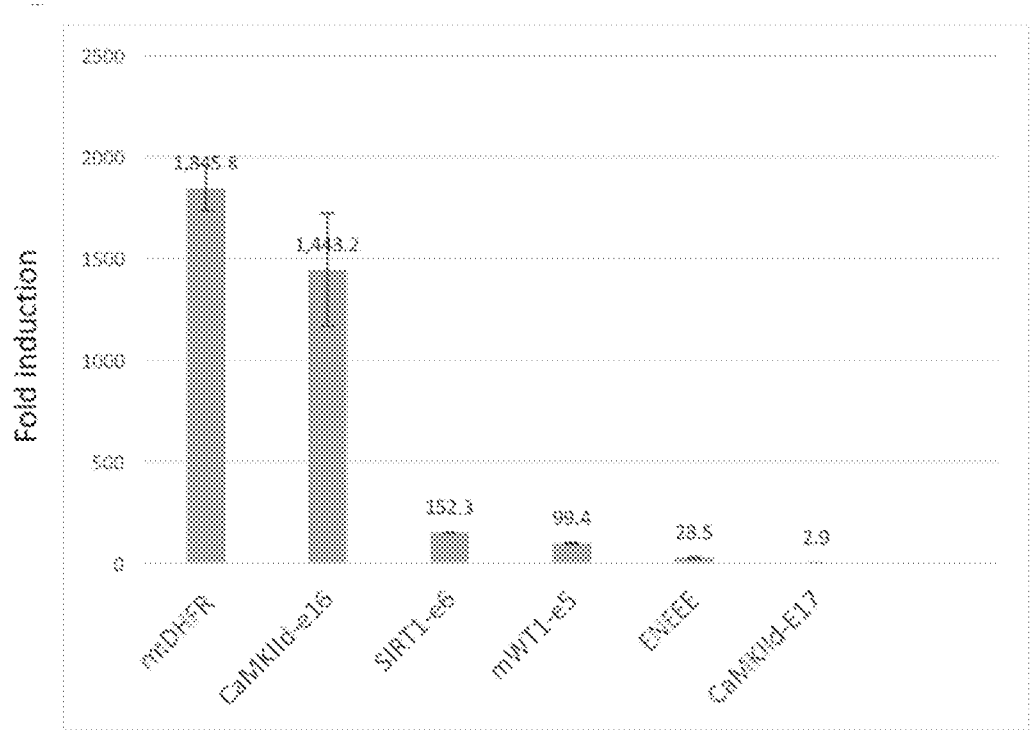


Fig. 10b

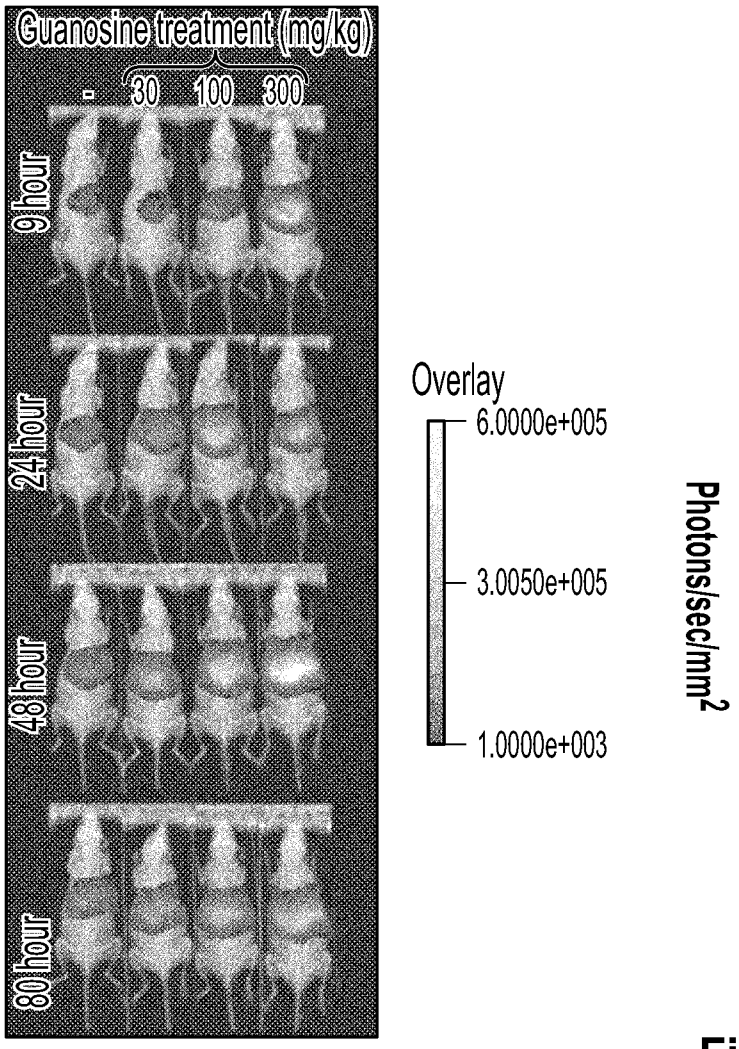


Fig. 11a

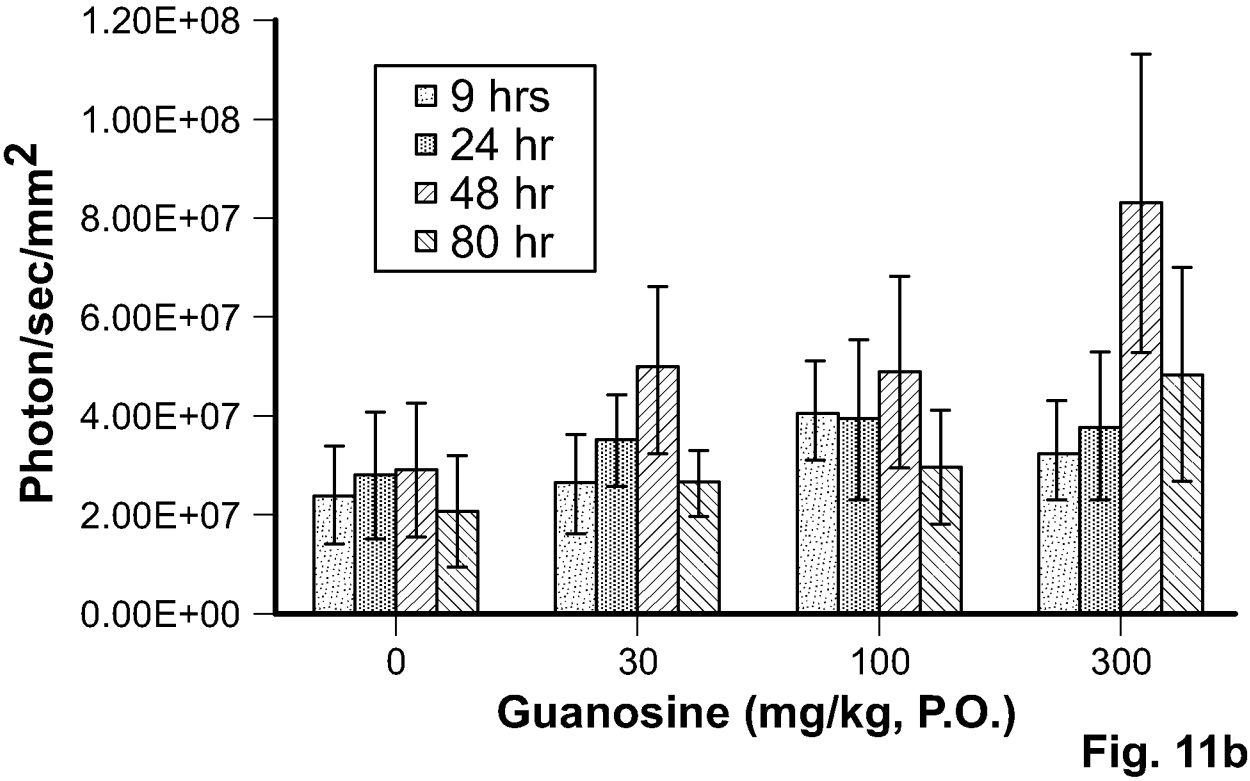


Fig. 11b

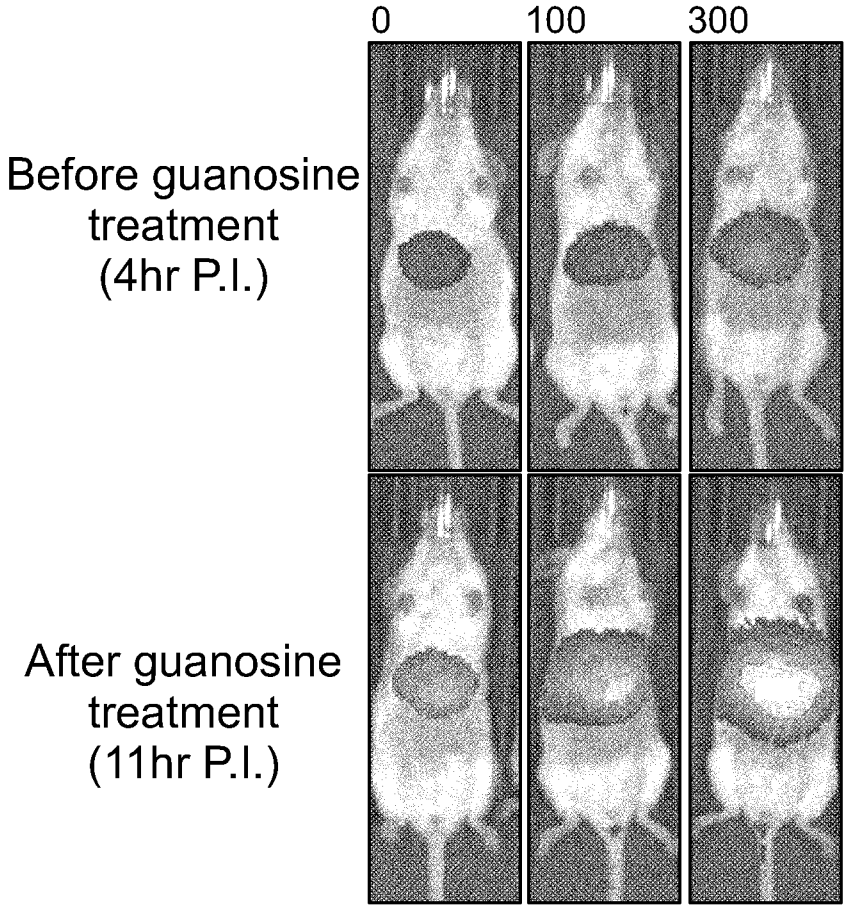


Fig. 11c

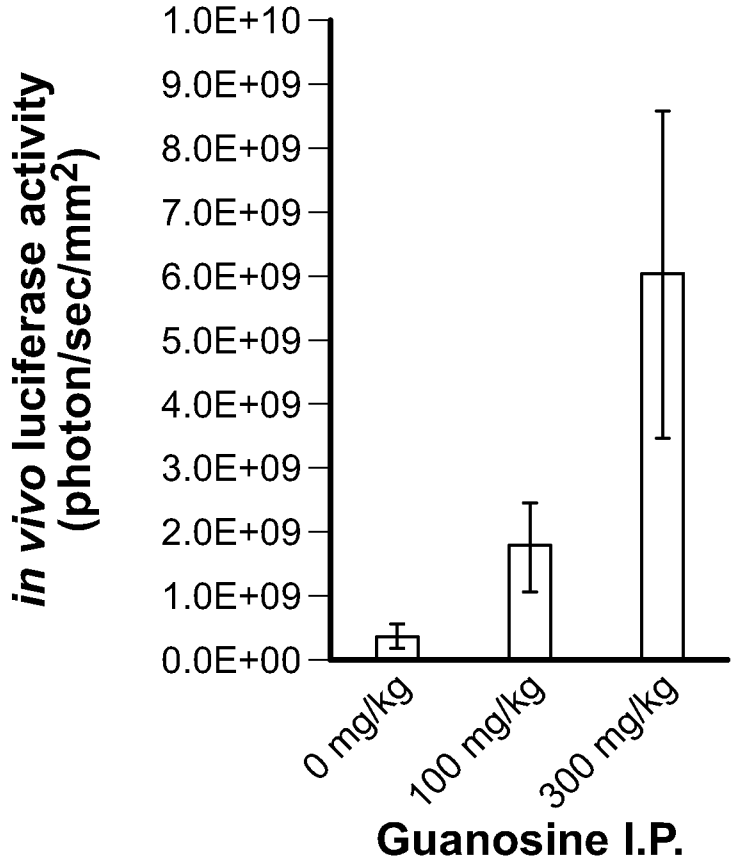


Fig. 11d

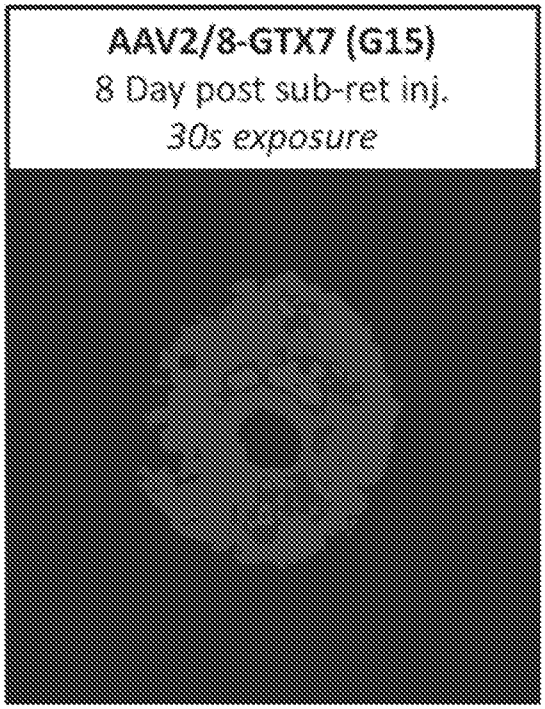


Fig. 12

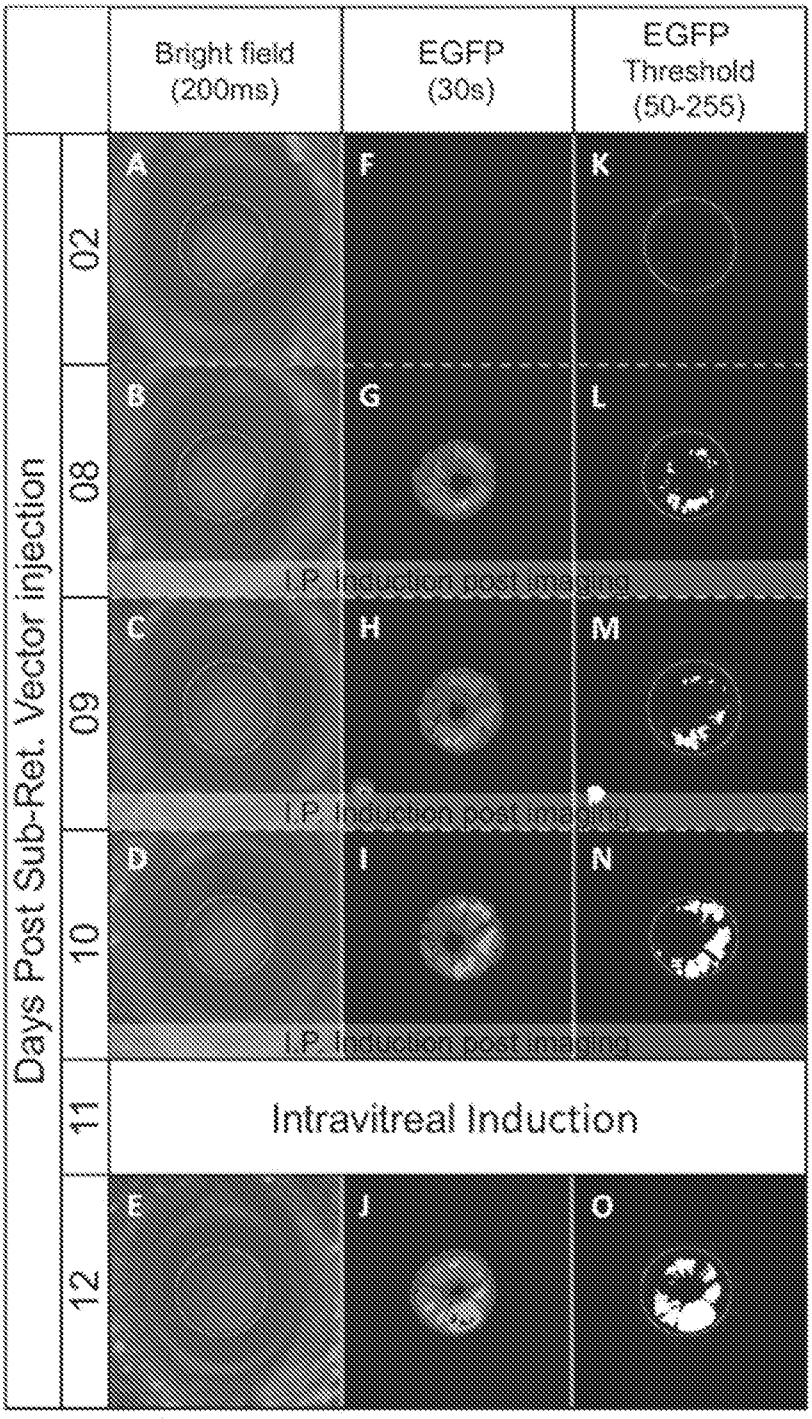

Bright field (200ms)
EGFP (30s)
EGFP Threshold (50-255)
Days Post Sub-Ret. Vector injection
02
08
09
10
11
12
A
B
C
D
E
F
G
H
I
J
K
L
M
N
O
Intravitreal Induction

Fig. 13a

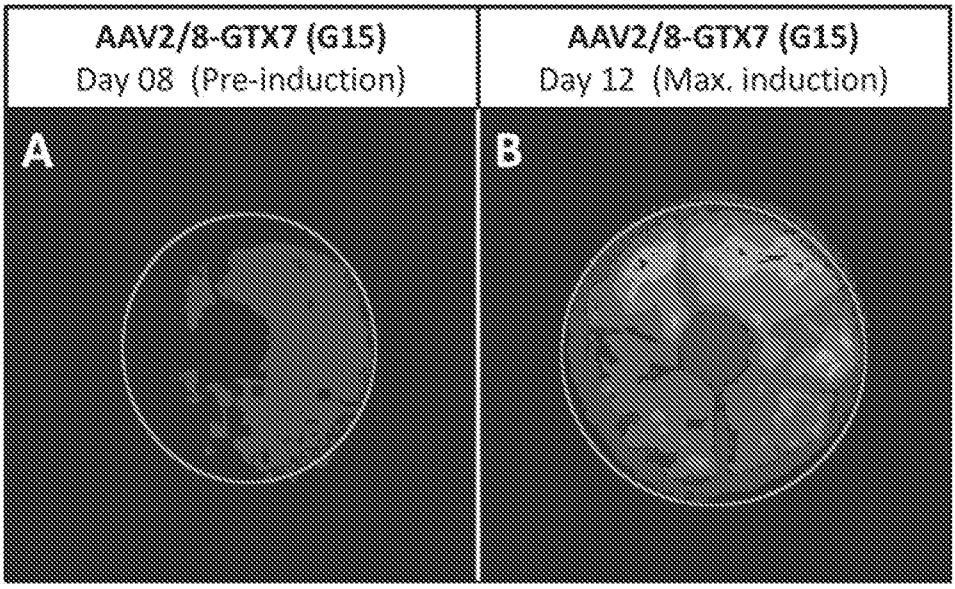

AAV2/8-GTX7 (G15)
Day 08 (Pre-induction)
AAV2/8-GTX7 (G15)
Day 12 (Max. induction)
A
B
30s exposure

Fig. 13b

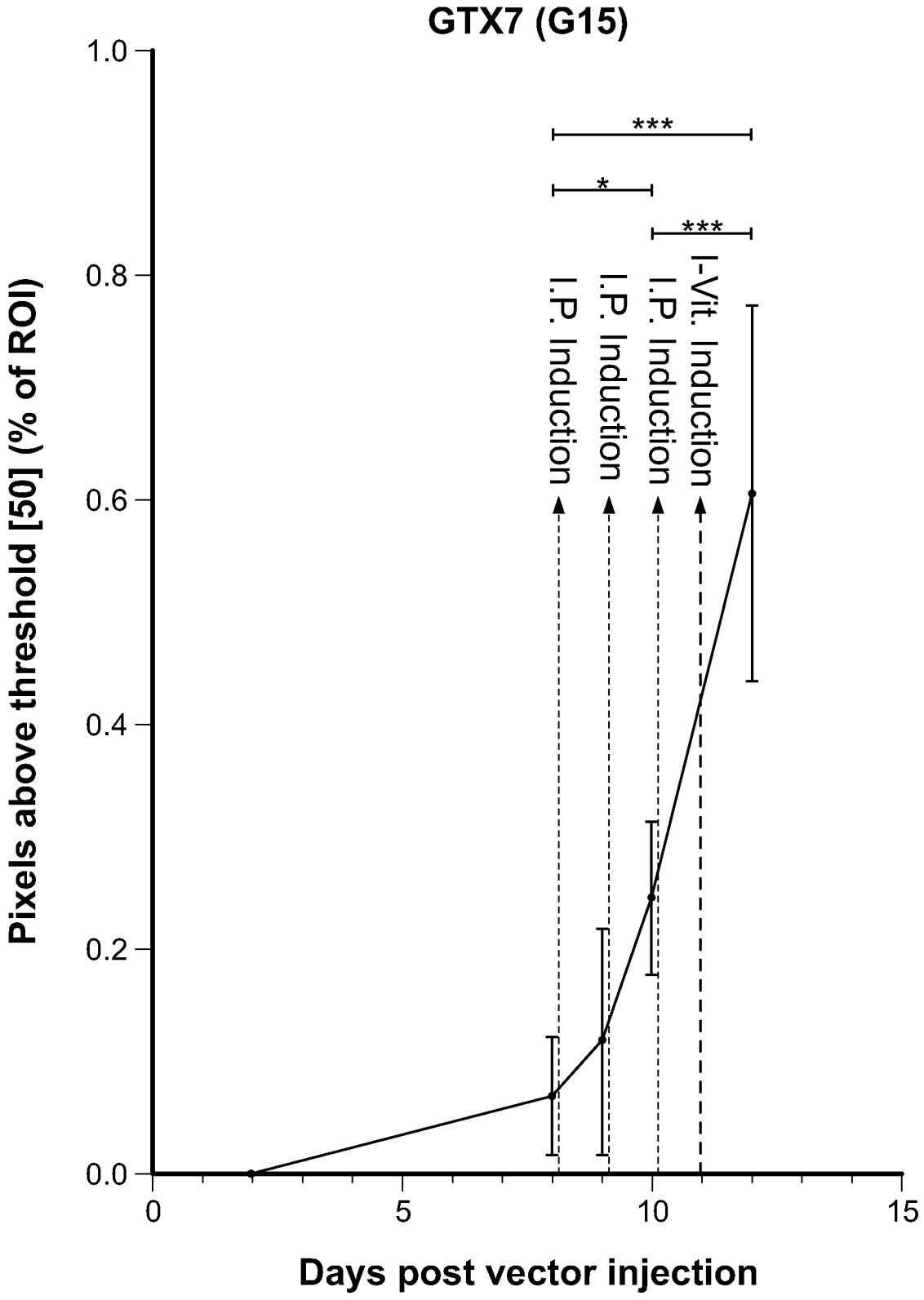

GTX7 (G15)
Pixels above threshold [50] (% of ROI)
1.0
0.8
0.6
0.4
0.2
0.0
0
5
10
15
Days post vector injection
***
*
***
I.P. Induction
I.P. Induction
I.P. Induction
I-Vit. Induction

Fig. 13c

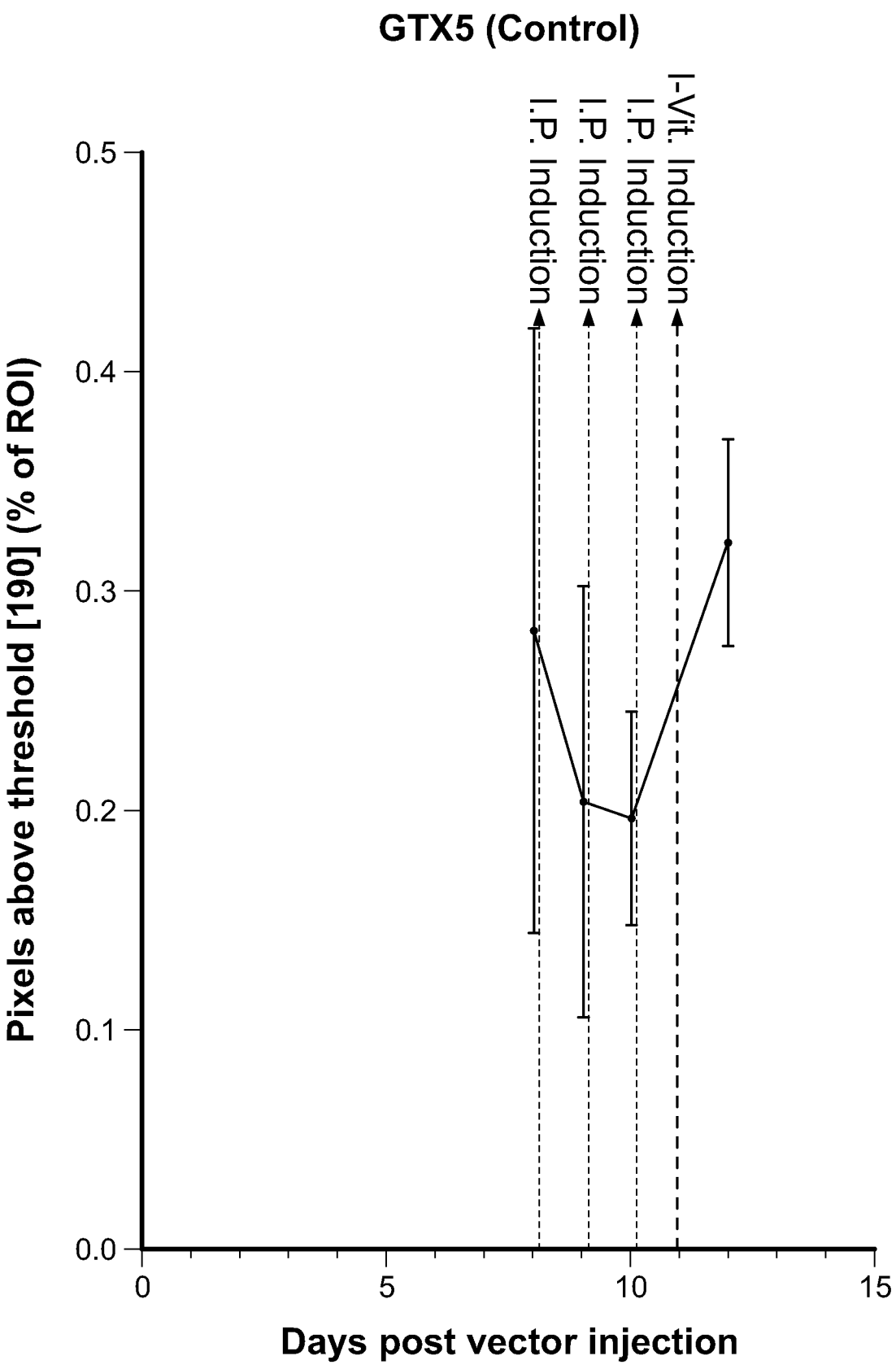

GTX5 (Control)
I.P. Induction
I.P. Induction
I.P. Induction
I-Vit. Induction
Pixels above threshold [190] (% of ROI)
0.5
0.4
0.3
0.2
0.1
0.0
0
5
10
15
Days post vector injection

Fig. 13d