Fig. 1

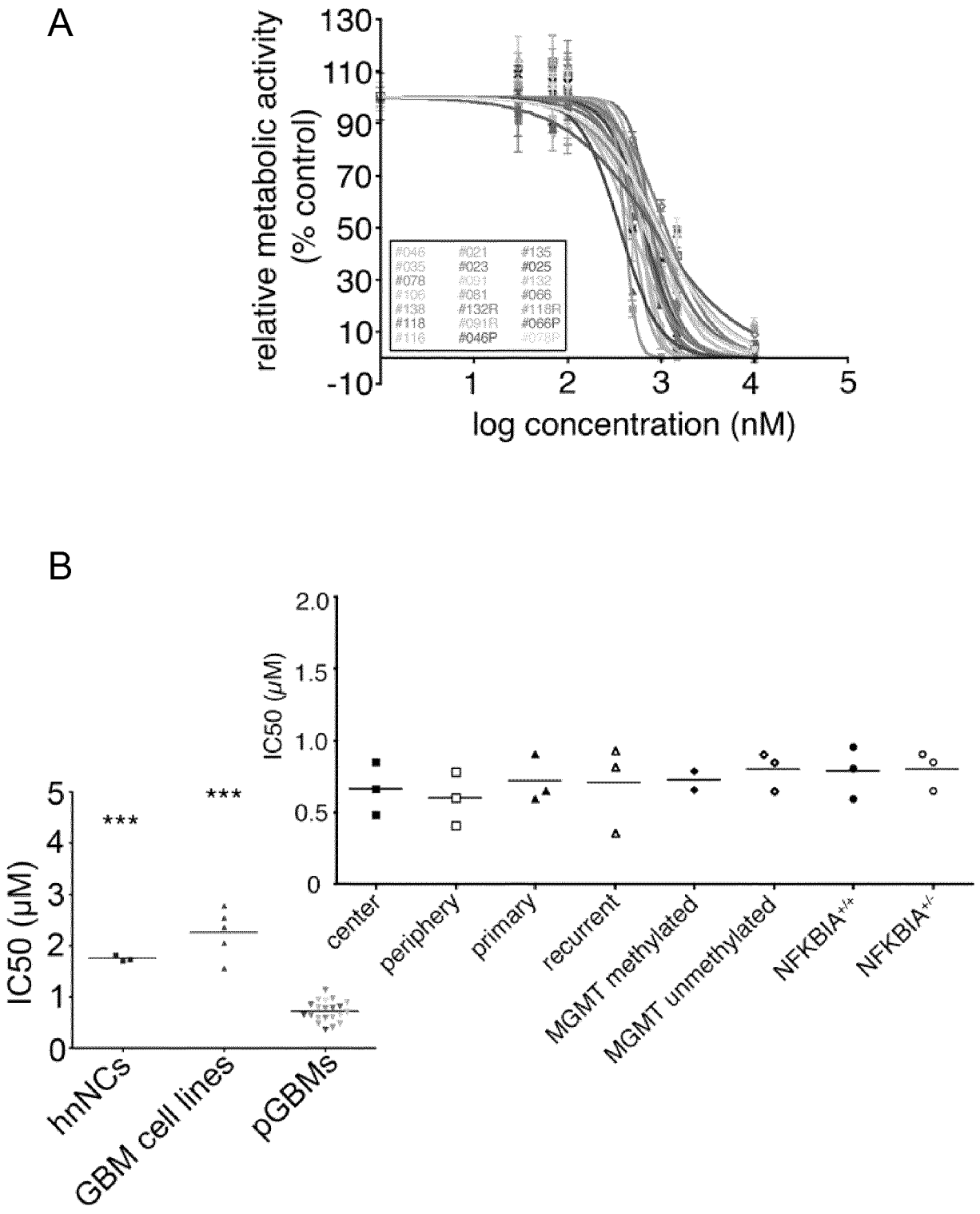

A
relative metabolic activity (% control)
130
110
90
70
50
30
10
-10
1
2
3
4
5
log concentration (nM)
#046
#021
#135
#035
#023
#025
#078
#091
#132
#106
#081
#066
#138
#132R
#118R
#118
#091R
#066P
#116
#046P
#078P
B
IC50 (µM)
5
4
3
2
1
0
***
***
hnNCs
GBM cell lines
pGBMs
IC50 (µM)
2.0
1.5
1.0
0.5
0
center
periphery
primary
recurrent
MGMT methylated
MGMT unmethylated
NFKBIA+/+
NFKBIA+/-

Fig. 2

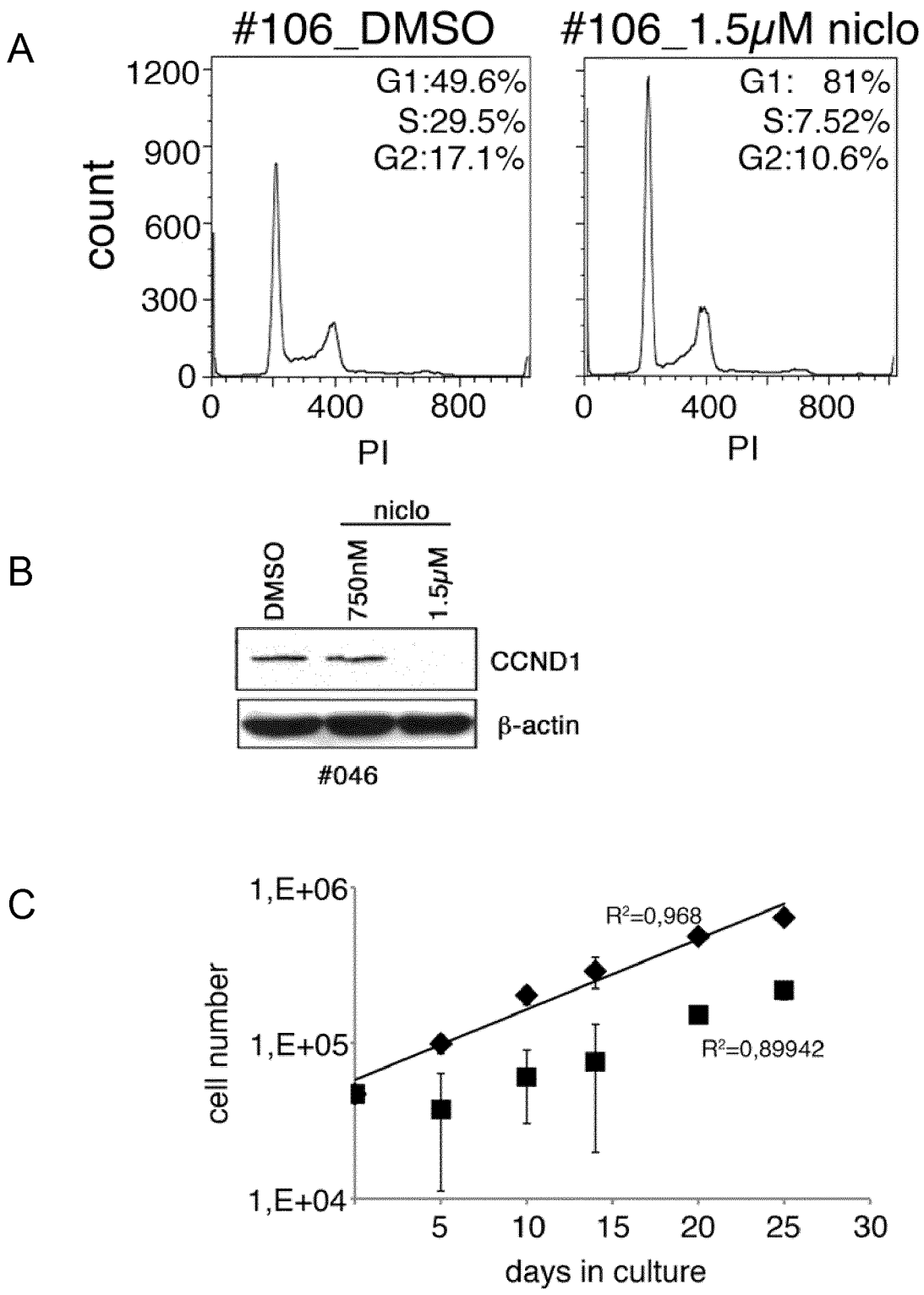
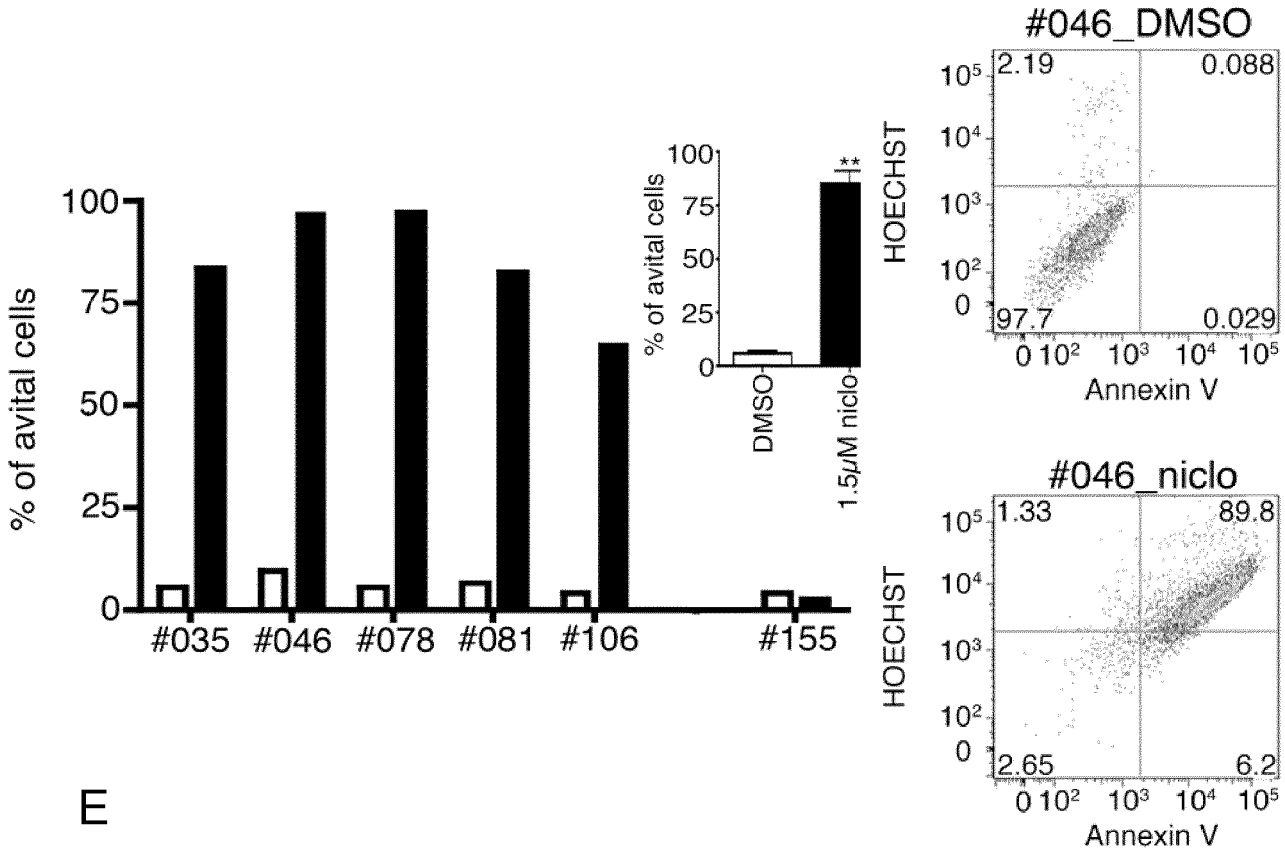

A
#106_DMSO
#106_1.5μM niclo
G1:49.6%
S:29.5%
G2:17.1%
G1: 81%
S:7.52%
G2:10.6%
count
1200
900
600
300
0
0
400
800
PI
B
niclo
DMSO
750nM
1.5μM
CCND1
β-actin
#046
C
1,E+06
1,E+05
1,E+04
cell number
R²=0,968
R²=0,89942
5
10
15
20
25
30
days in culture

Fig. 2 (continued)

D



E

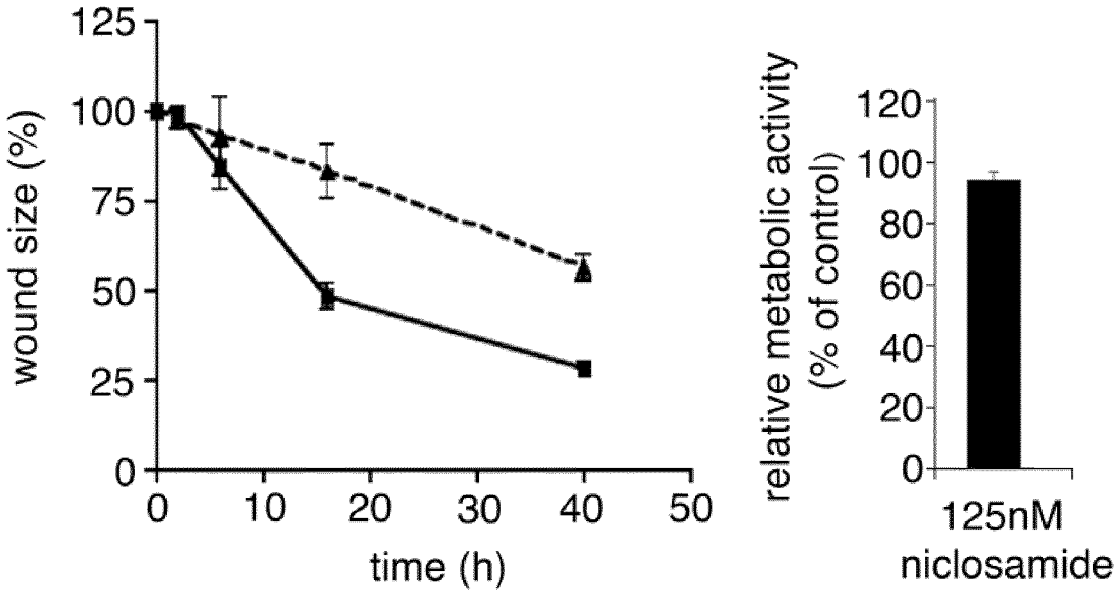


Fig. 3

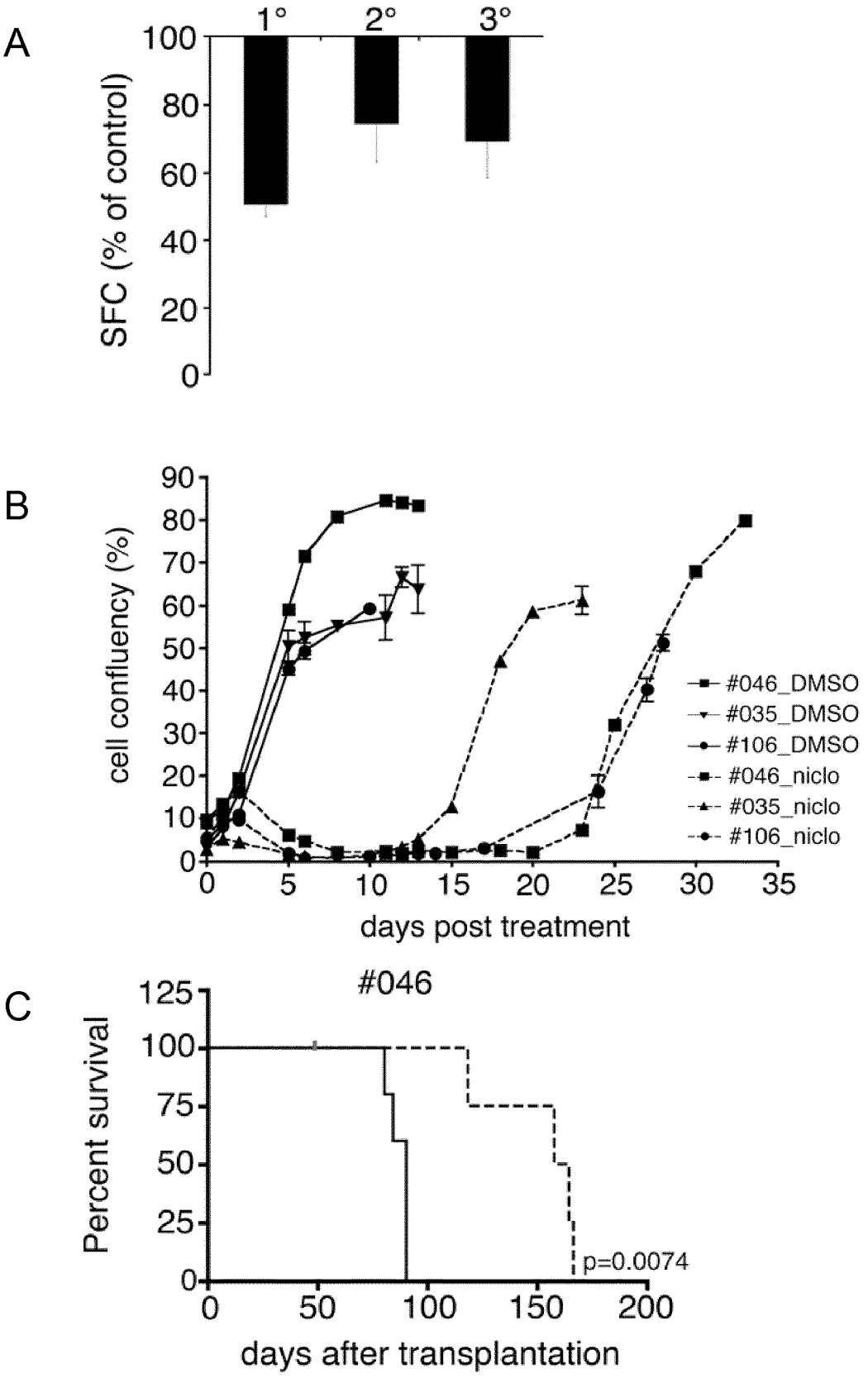

A
1°
2°
3°
SFC (% of control)
100
80
60
40
20
0
B
cell confluency (%)
90
80
70
60
50
40
30
20
10
0
0
5
10
15
20
25
30
35
days post treatment
#046_DMSO
#035_DMSO
#106_DMSO
#046_niclo
#035_niclo
#106_niclo
C
#046
Percent survival
125
100
75
50
25
0
p=0.0074
0
50
100
150
200
days after transplantation

Fig. 4

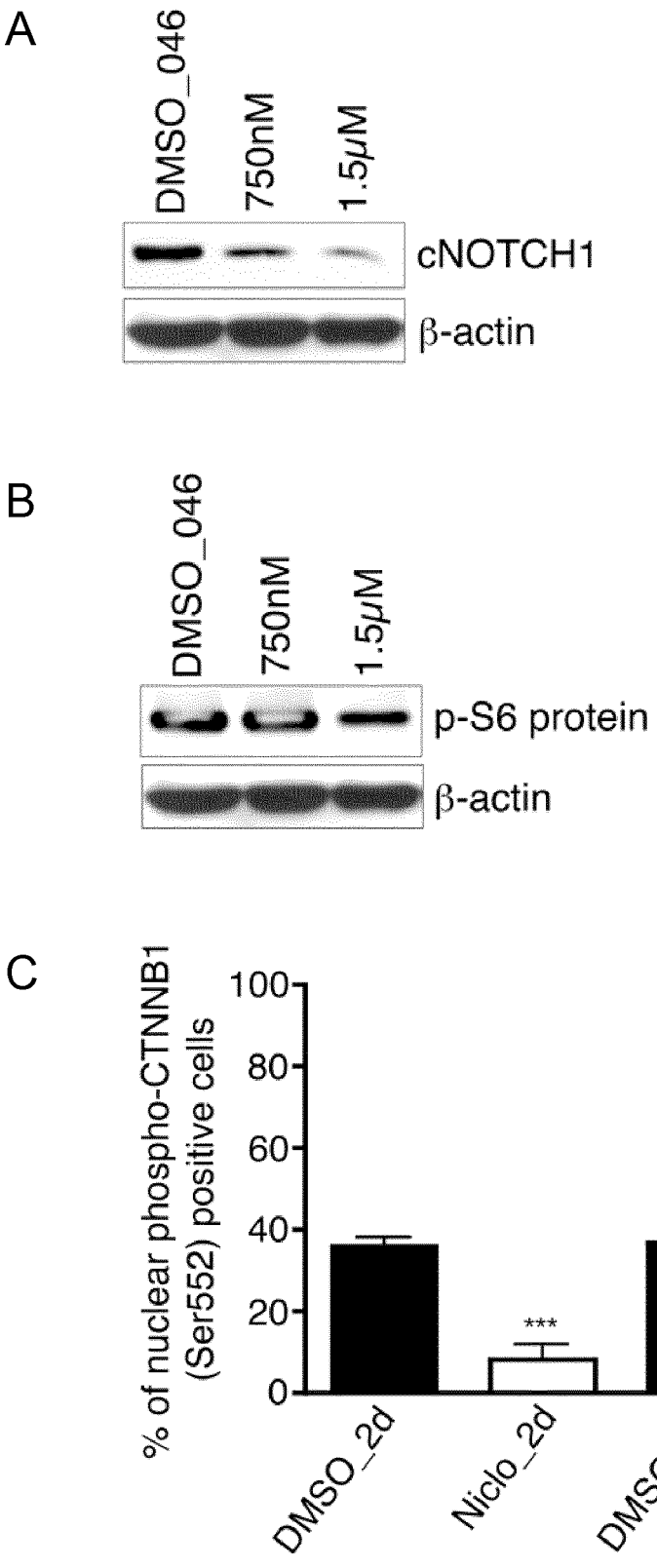

A
DMSO_046
750nM
1.5μM
cNOTCH1
β-actin
B
DMSO_046
750nM
1.5μM
p-S6 protein
β-actin
C
% of nuclear phospho-CTNNB1 (Ser552) positive cells
100
80
60
40
20
0
***
***
DMSO_2d
Niclo_2d
DMSO_4d
Niclo_4d

Fig. 4 (continued)

D

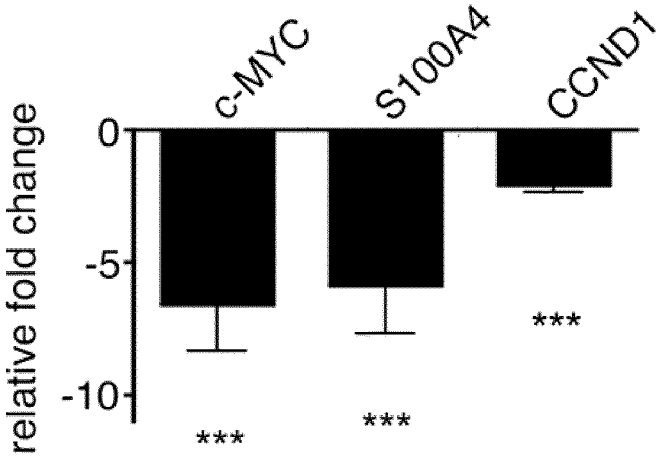


Fig. 5

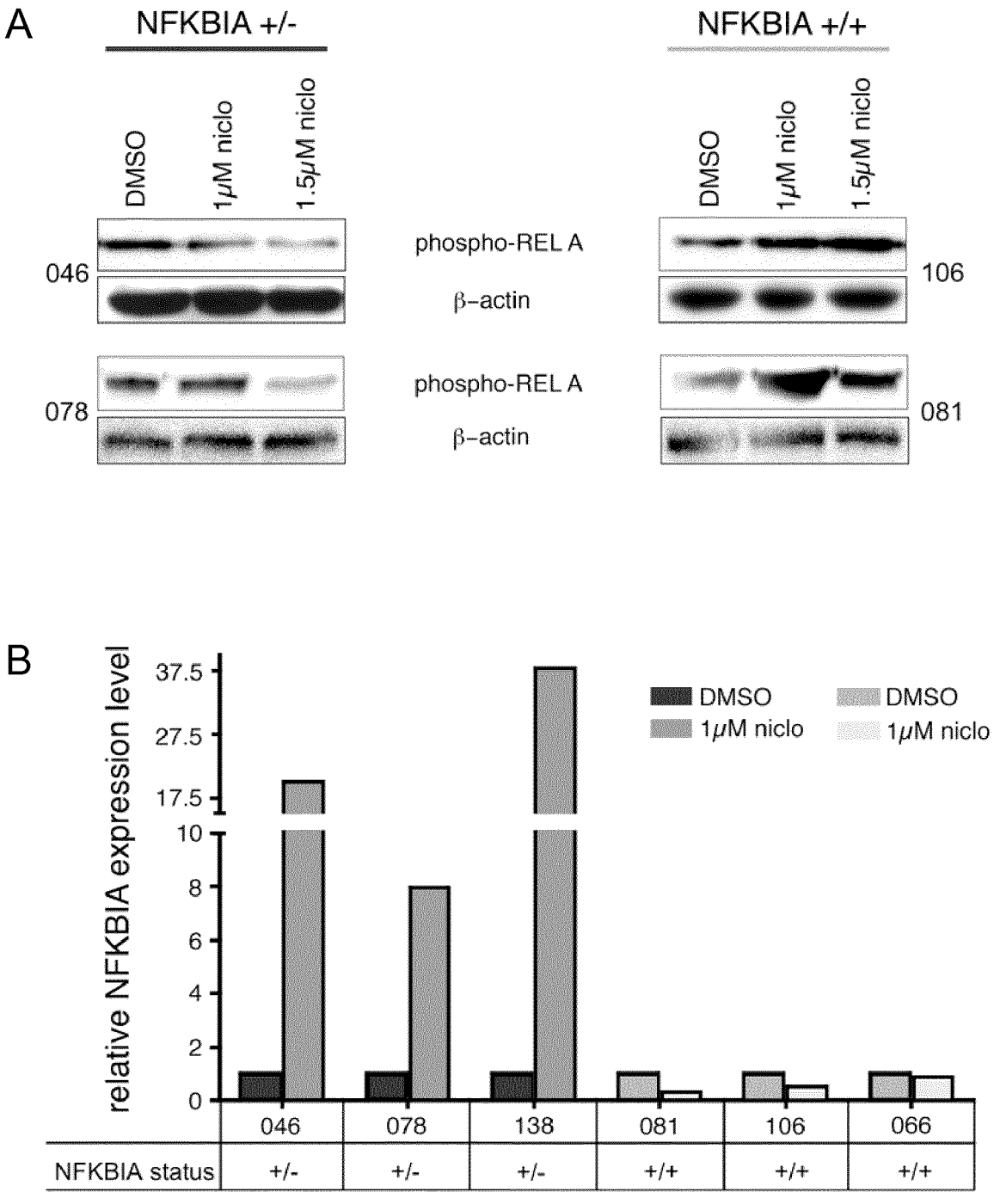

A
NFKBIA +/-
NFKBIA +/+
DMSO
1μM niclo
1.5μM niclo
DMSO
1μM niclo
1.5μM niclo
046
phospho-REL A
β-actin
106
078
phospho-REL A
β-actin
081
B
relative NFKBIA expression level
37.5
27.5
17.5
10
8
6
4
2
0
DMSO
1μM niclo
DMSO
1μM niclo
046
078
138
081
106
066
NFKBIA status
+/-
+/-
+/-
+/+
+/+
+/+

Fig. 5 (continued)

C

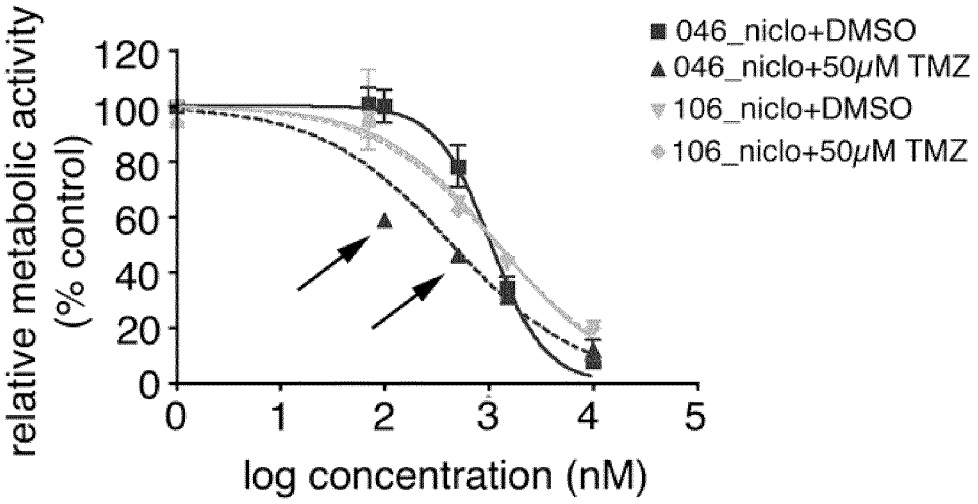
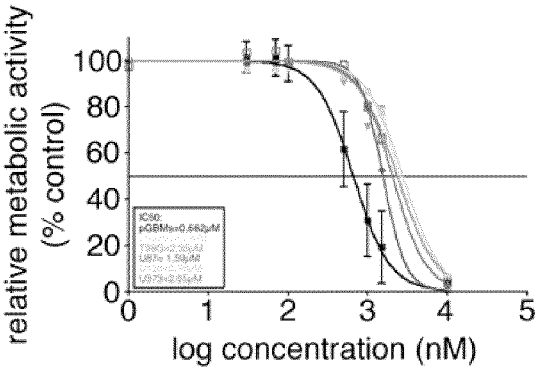
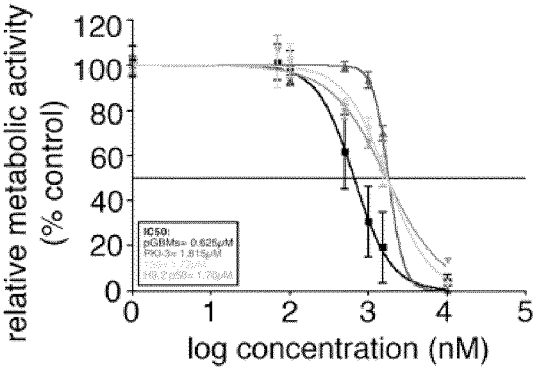


Fig. 6

A


relative metabolic activity
(% control)
100
80
60
40
20
0
0
1
2
3
4
5
log concentration (nM)

B


relative metabolic activity
(% control)
120
100
80
60
40
20
0
0
1
2
3
4
5
log concentration (nM)

C

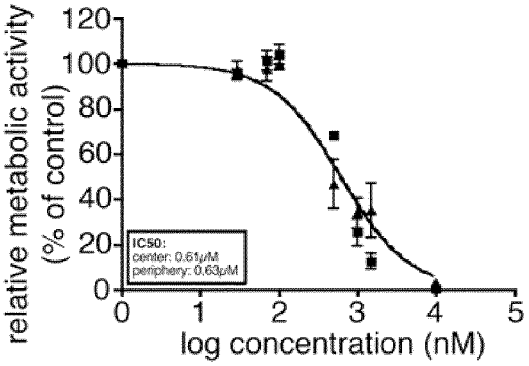
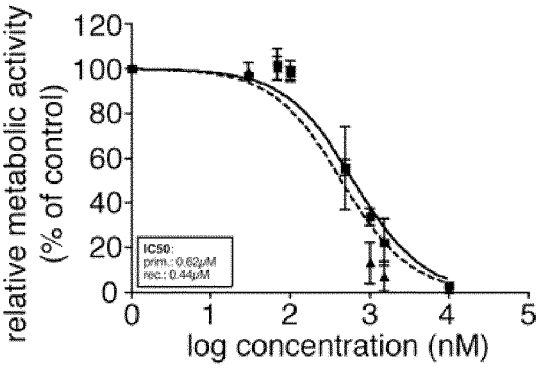
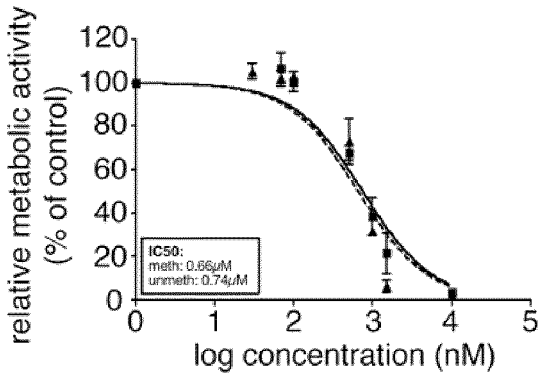

relative metabolic activity
(% of control)
120
100
80
60
40
20
0
0
1
2
3
4
5
log concentration (nM)
IC50:
center: 0.61µM
periphery: 0.63µM

Fig. 6 (continued)

D



E



F

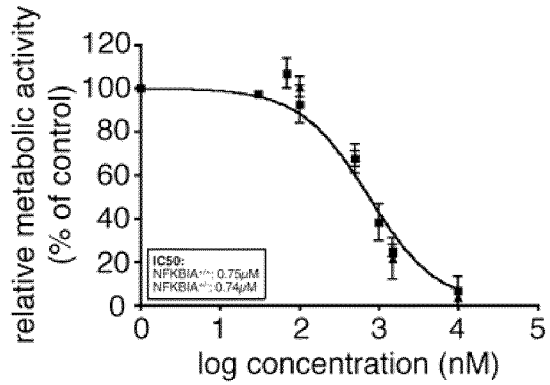


Fig. 7

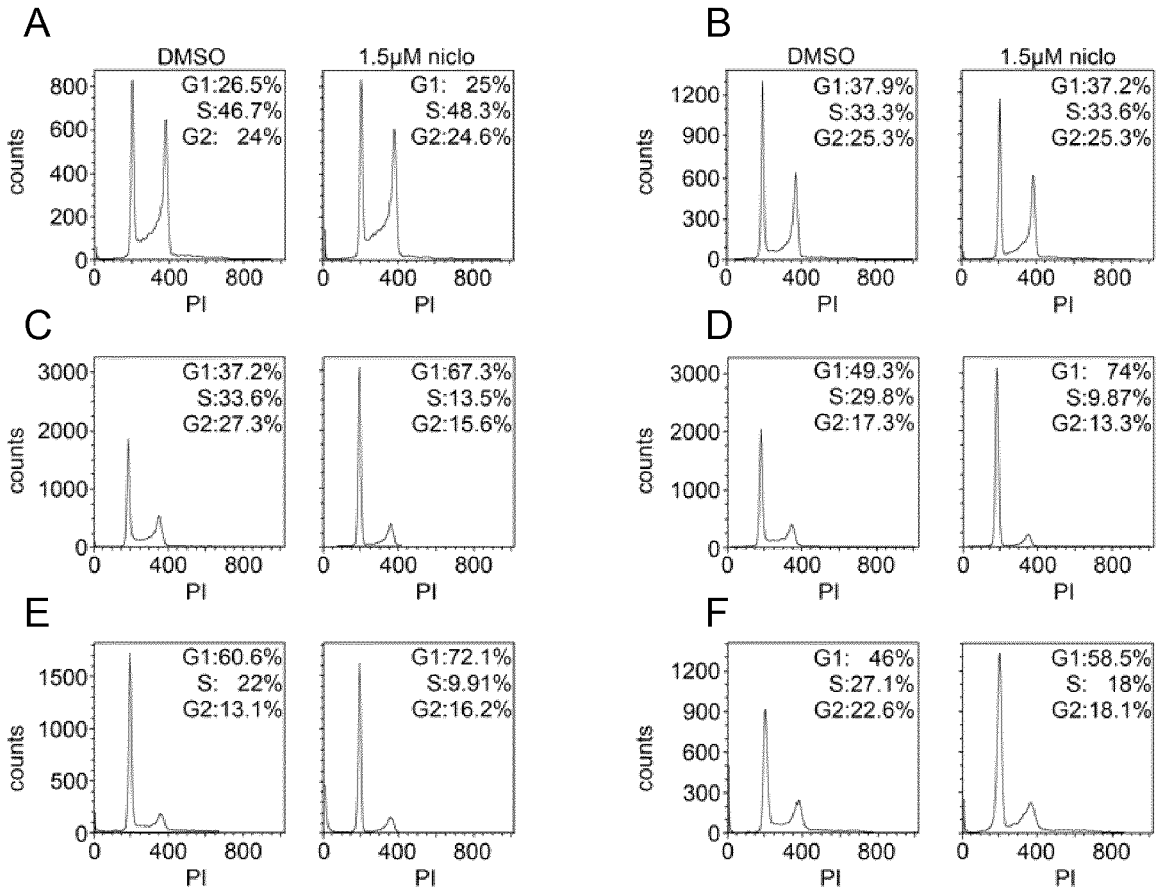
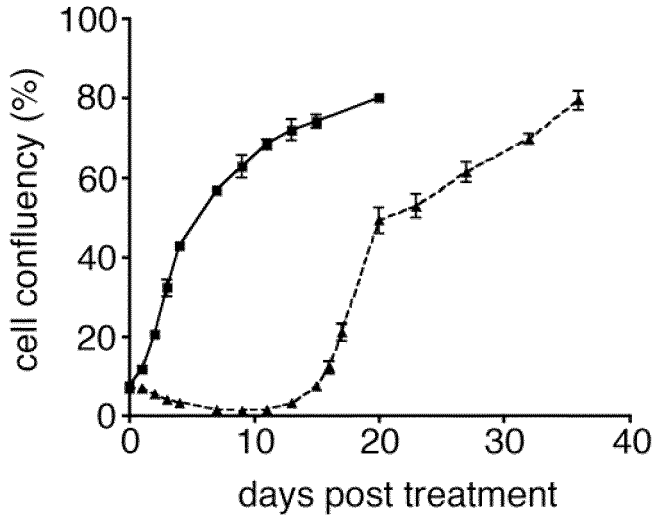
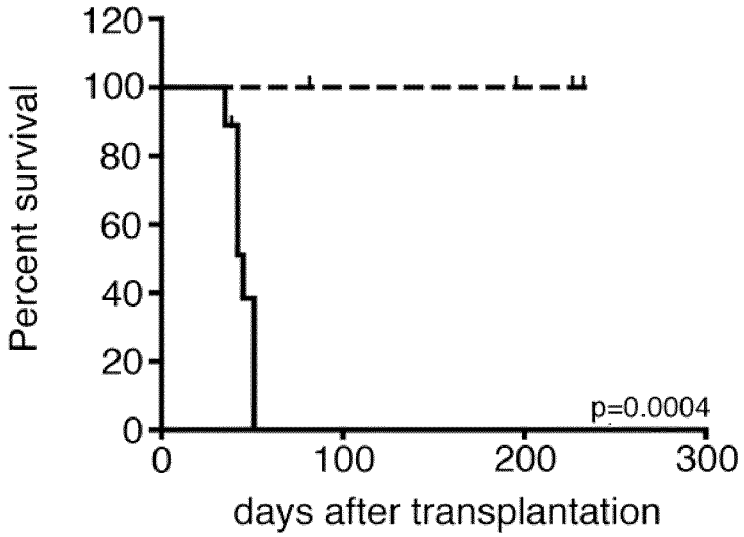

A
DMSO
1.5μM niclo
G1:26.5%
S:46.7%
G2: 24%
G1: 25%
S:48.3%
G2:24.6%
B
DMSO
1.5μM niclo
G1:37.9%
S:33.3%
G2:25.3%
G1:37.2%
S:33.6%
G2:25.3%
C
G1:37.2%
S:33.6%
G2:27.3%
G1:67.3%
S:13.5%
G2:15.6%
D
G1:49.3%
S:29.8%
G2:17.3%
G1: 74%
S:9.87%
G2:13.3%
E
G1:60.6%
S: 22%
G2:13.1%
G1:72.1%
S:9.91%
G2:16.2%
F
G1: 46%
S:27.1%
G2:22.6%
G1:58.5%
S: 18%
G2:18.1%
counts
PI

Fig. 8

A


100
80
60
40
20
0
cell confluency (%)
0
10
20
30
40
days post treatment

B


120
100
80
60
40
20
0
Percent survival
p=0.0004
0
100
200
300
days after transplantation

C

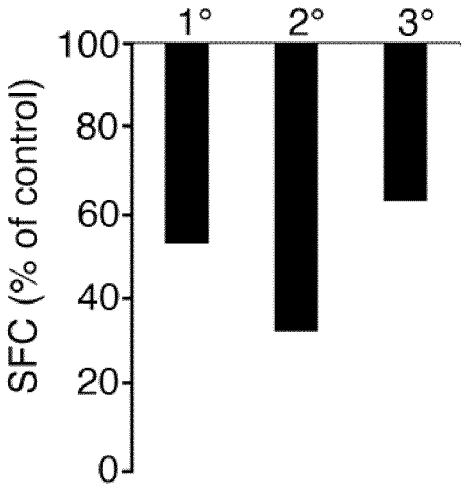

1°
2°
3°
100
80
60
40
20
0
SFC (% of control)

Fig. 9

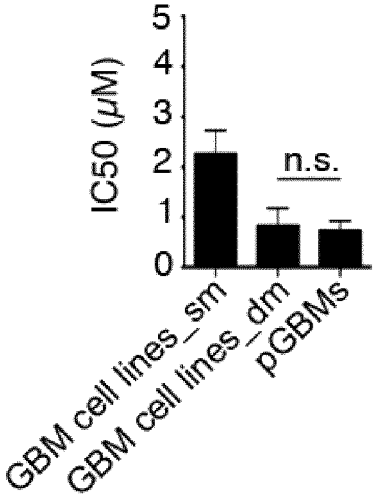
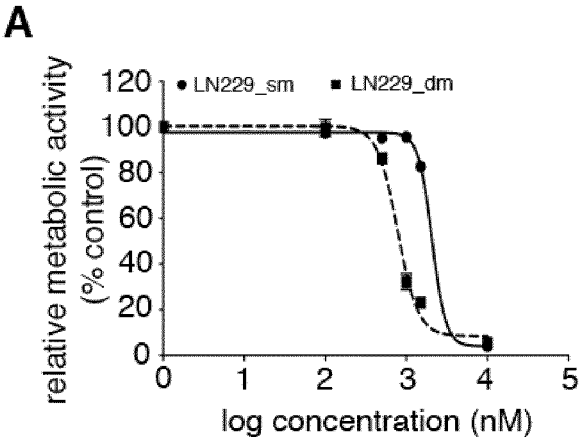
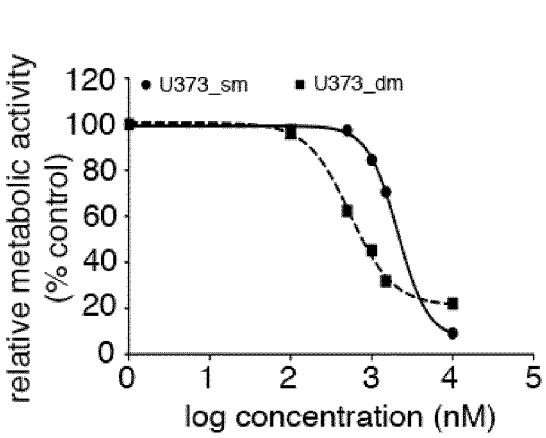
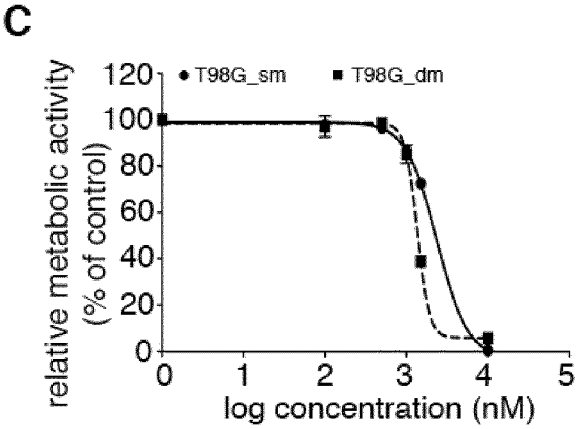
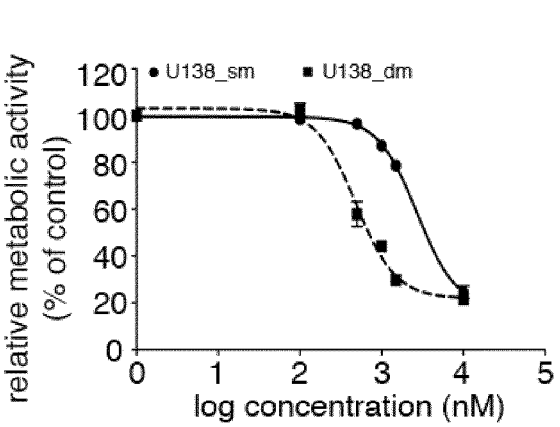


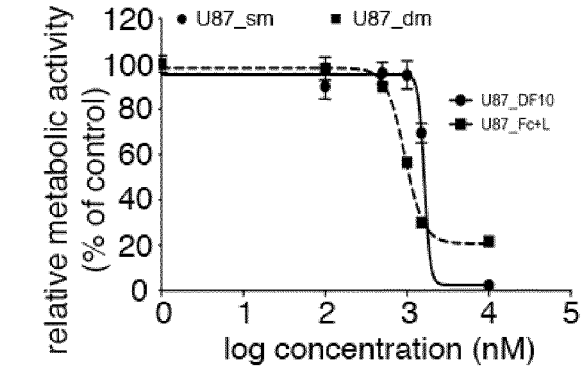
Fig. 10











F

	U373	U138	U87	LN229	T98G
sm	2100nM	2716nM	1602nM	2073nM	2369nM
dm	543nM	484nM	940nM	782nM	1355nM

Fig. 11

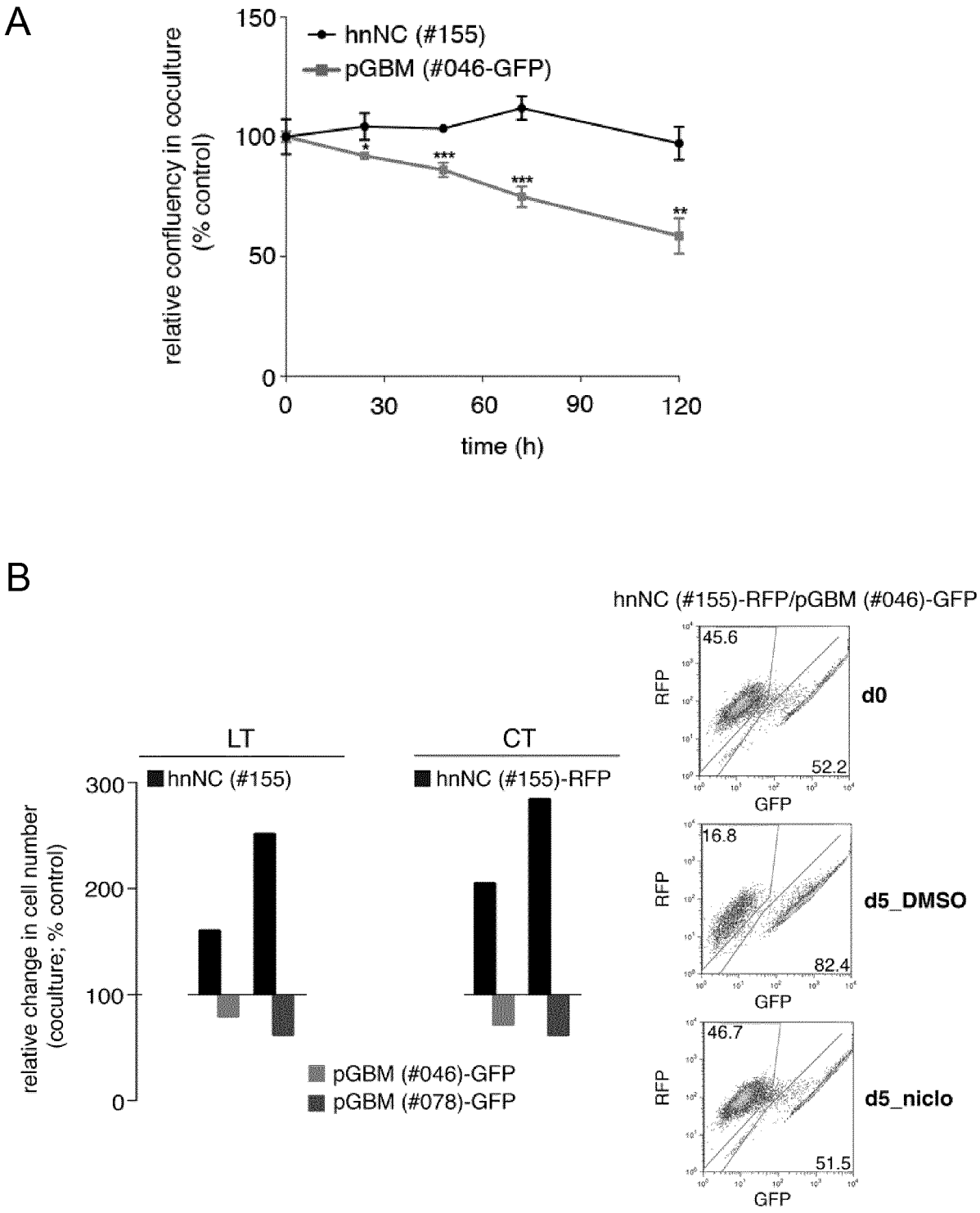

A
150
100
50
0
hnNC (#155)
pGBM (#046-GFP)
relative confluency in coculture (% control)
*
***
***
**
0
30
60
90
120
time (h)
B
hnNC (#155)-RFP/pGBM (#046)-GFP
LT
CT
hnNC (#155)
hnNC (#155)-RFP
300
200
100
0
relative change in cell number (coculture; % control)
pGBM (#046)-GFP
pGBM (#078)-GFP
45.6
52.2
RFP
GFP
d0
16.8
82.4
d5_DMSO
46.7
51.5
d5_niclo

Fig. 12

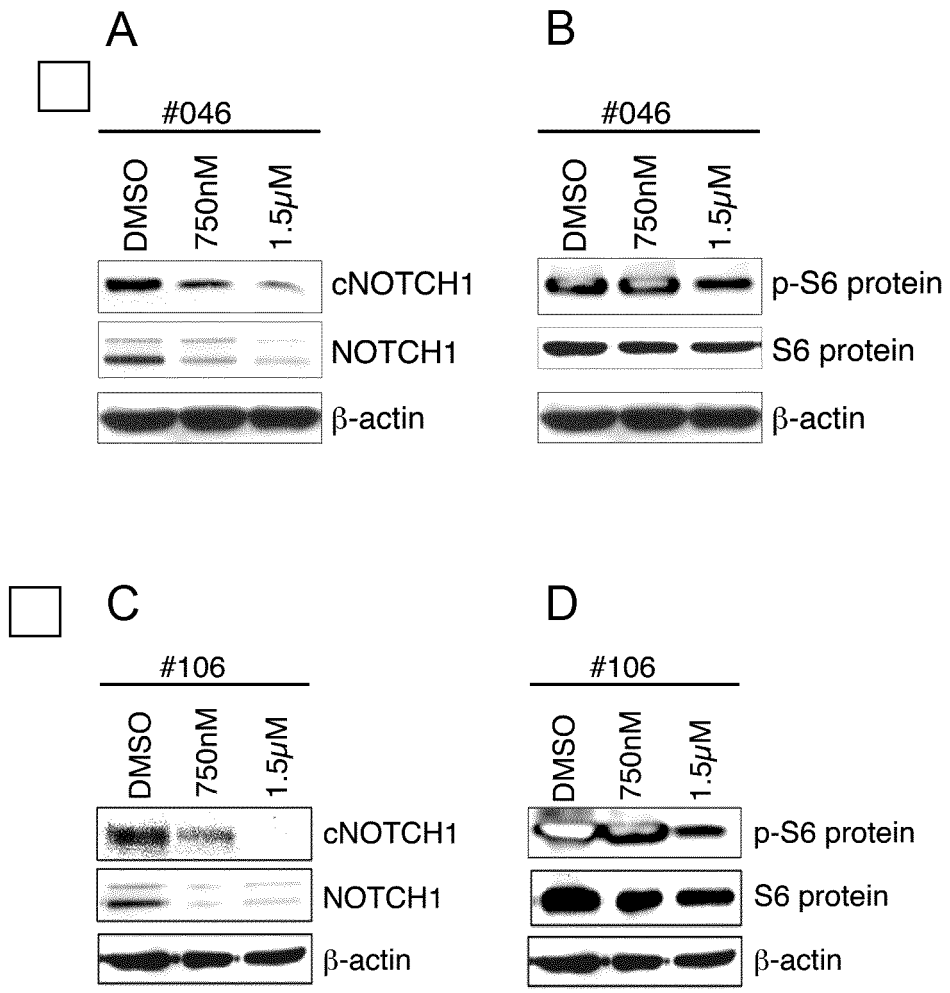

A
#046
DMSO
750nM
1.5μM
cNOTCH1
NOTCH1
β-actin
B
#046
DMSO
750nM
1.5μM
p-S6 protein
S6 protein
β-actin
C
#106
DMSO
750nM
1.5μM
cNOTCH1
NOTCH1
β-actin
D
#106
DMSO
750nM
1.5μM
p-S6 protein
S6 protein
β-actin

Fig. 13

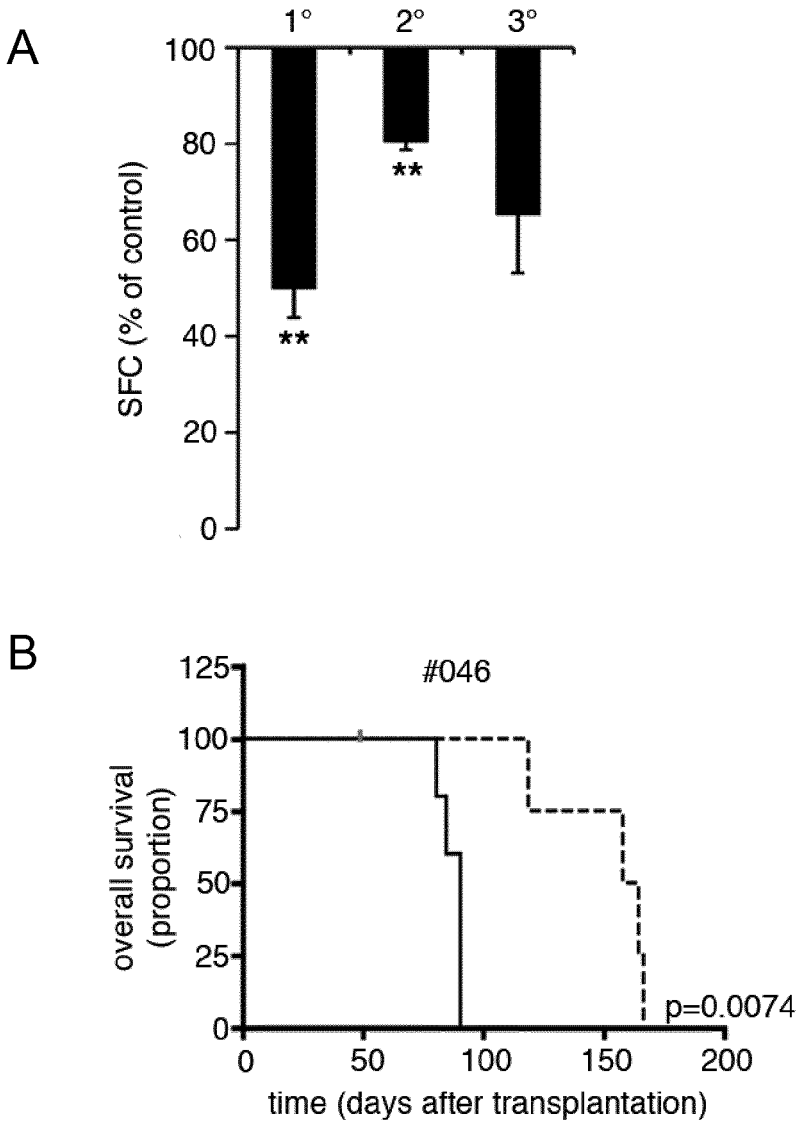
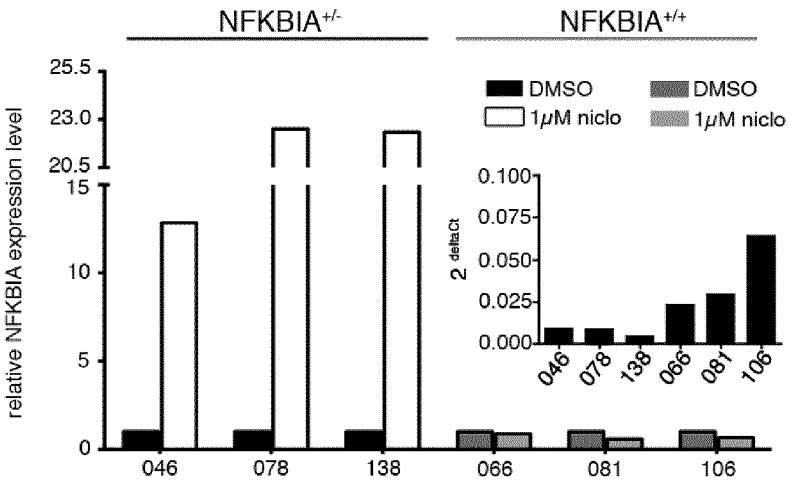

A
1°
2°
3°
100
80
60
40
20
0
SFC (% of control)
**
**
B
#046
125
100
75
50
25
0
overall survival (proportion)
p=0.0074
0
50
100
150
200
time (days after transplantation)

Fig. 14

A



B

pGBM #	*NFKBIA* status	Expected survival propotion	Observed survival proportion	Combinatorial index
046	+/-	0.667	0.444	**0.665**
078	+/-	0.574	0.426	**0.742**
118	+/-	0.831	0.496	**0.596**
138	+/-	0.551	0.396	**0.718**
066	+/+	0.705	0.720	**0.979**
081	+/+	0.496	0.447	**0.901**
106	+/+	0.785	0.752	**0.957**

C

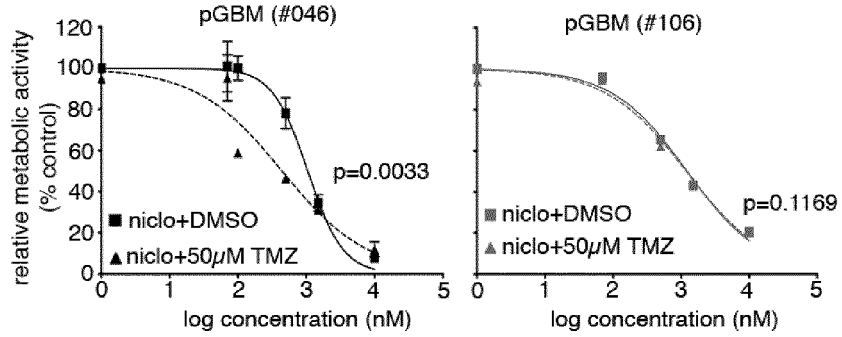
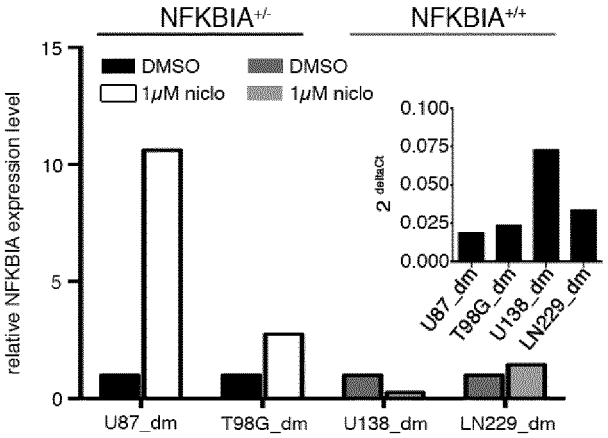


Fig. 15

A



B

GBM cell line	*NFKBIA* status	Expected survival propotion	Observed survival proportion	Combinatorial index
U87	+/-	0.522	0.382	**0.731**
T98G	+/-	0.868	0.684	**0.788**
U138	+/+	0.403	0.402	**0.996**
LN229	+/+	0.261	0.238	**0.912**

C

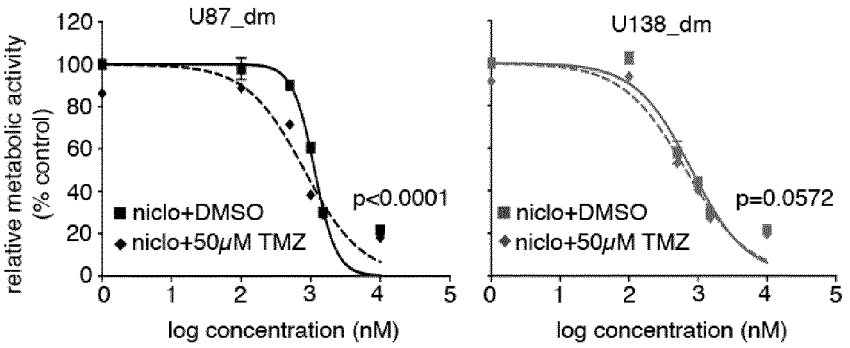


Fig. 16

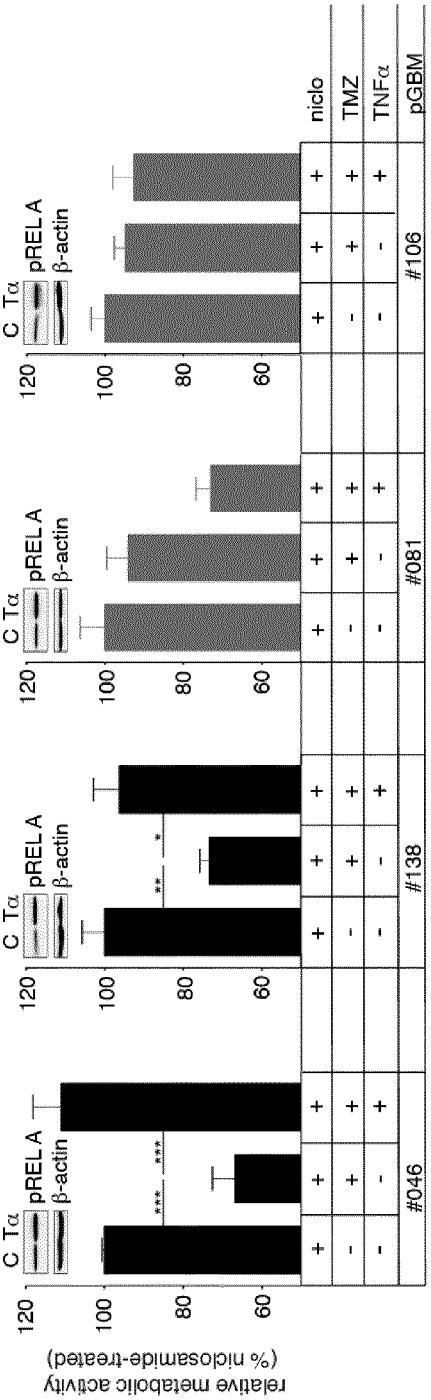

relative metabolic activity
(% niclosamide-treated)
120
100
80
60
C Tα
pREL A
β-actin
***
***
**
*
niclo
TMZ
TNFα
pGBM
#046
#138
#081
#106

Fig. 17

pGBM	NFKBIA	screening	Pharma	CT/LT	FACS	NSA	kinetics	WB	qPCR	transplantation	SNP
021	+/+	-	8	-	-	-	-	-	-	-	6
023	+/+	10	9	-	-	-	-	-	-	-	7
025	+/+	-	8	-	-	-	-	-	-	-	7
035	+/+	10	8-12	10	9-11	-	8	10/11	9/10	-	5
046	+/-	10	8-12	10	9-11	9	8	10/11	9/10	12	5
066	+/+	-	8-12	-	-	-	8	10/11	9/10	-	5
078	+/-	-	8-12	10	9-11	9	-	10/11	9/10	-	5
081	+/+	-	9-12	-	11	-	-	10/11	9/10	-	5
91	n.d.	-	11	-	-	-	-	-	-	-	-
106	+/+	10	8-12	10	9-11	9	8	10/11	9/10	-	6
116	n.d.	-	11	-	-	-	-	-	-	-	-
118	+/-	-	9-12	-	-	-	-	10/11	9/10	-	7/6
132	n.d.	-	10	-	-	-	-	-	-	-	-
135	+/-	-	8	-	-	-	-	-	-	-	7
138	+/-	-	9-12	-	-	-	-	10/11	9/10	-	5
GNV019	+/++	-	11	-	-	11	-	-	-	11	3/10