## FIGURE LEGENDS

FIGURE 1. **Downregulation of DNMT1 leads to decreased apoptosis.** Malme3M (A and B) or SNU387 cells (C and D) were transiently transfected with control SiRNA (10 nM) or DNMT1 SiRNA (10 nM). At 48 h after transfection, cells were treated with celastrol (1 μM) or taxol (1 μM) for 16 h. Cell number (A and C) or caspase-3 activity (B and D) was measured as described. Each value represents an average of three independent experiments. S denotes drug-sensitive.

FIGURE 2. **Induction of CAGE leads to enhanced tumorigenic potential of HeLa cells.** (A) HeLa cells stably transfected with CAGE under Tet-on system was treated with or without doxycycline (1 μg/ml) for various time intervals. Cell lysates were prepared and subjected to Western blot analysis. (B) Cells were pretreated with or without doxycycline (1 μg/ml) for 16 h, followed by treatment with taxol at the indicated concentrations for 24 h. Cell number and capsase-3 activity were determined as described. Each value represents an average of three independent experiments. (C) Stable transfectants of HeLa cells ($10^6$/mouse) expressing CAGE under the control of doxycycine were injected into nude mice. Nude mice were treated with or without doxycycline (1 μg/ml) via tail vein twice a week. Tumor volume of each nude mouse injected with HeLa cells was measured (left panel). Each group consists of ten mice. Each value represents an average values obtained from tumor volume of each mouse. Comparison of tumor tissues derived from HeLa cells treated with or without doxycycline (right panel). (D) Conditioned medium of HeLa cells treated with doxycycline (1 μg/ml) enhances tube formation in HUVEC.

FIGURE 3. **CAGE exerts negative effect on casapse-3 activity in response to drugs.** (A) SNU387 and SNU387$^R$ cells were treated with or without drugs at the indicated concentrations for 16 h. Caspase-3 activity was measured. (B) is the same as (A) except that Malme3M and Malme3M$^R$ cells were treated with or without drugs. SNU387 (C) or Malme3M cells (D) were transiently tranfected with control vector (1 μg) or CAGE cDNA (1 μg). At 24 h after transfection, cells were treated with each drug for 16 h. Caspase-3 activity was measured. S denotes drug-sensitive and R denotes drug-resistant.

FIGURE 4. **Downregulation of CAGE enhances sensitivity to drugs.** (A) SNU387$^{R}$ cells were transiently transfected with control SiRNA (10 nM), scrambled CAGE SiRNA (10 nM) or CAGE SiRNA (10 nM). At 48 h after transfection, cells were treated with celastrol (1 μM) or taxol (1 μM) for 16 h. Cell lysates were subjected to Western blot analysis. (B) is the same as (A) except that cell number was measured. (C) is the same as (A) except that caspase-3 activity assay was performed.

FIGURE 5. **Downregulation of CAGE exerts negative effect on cellular invasion.** (A) SNU387$^{R}$ or Malme3M$^{R}$ cells were transiently transfected with control SiRNA (10 nM), scrambled CAGE SiRNA (10 nM) or CAGE siRNA (10 nM). At 48 h after transfection, cells were treated with celastrol (1 μM) or taxol (1 μM) for 16 h. Cell lysates were subjected to Western blot analysis. (B) is the same as (A) except that invasion potential was measured. **P<0.005.

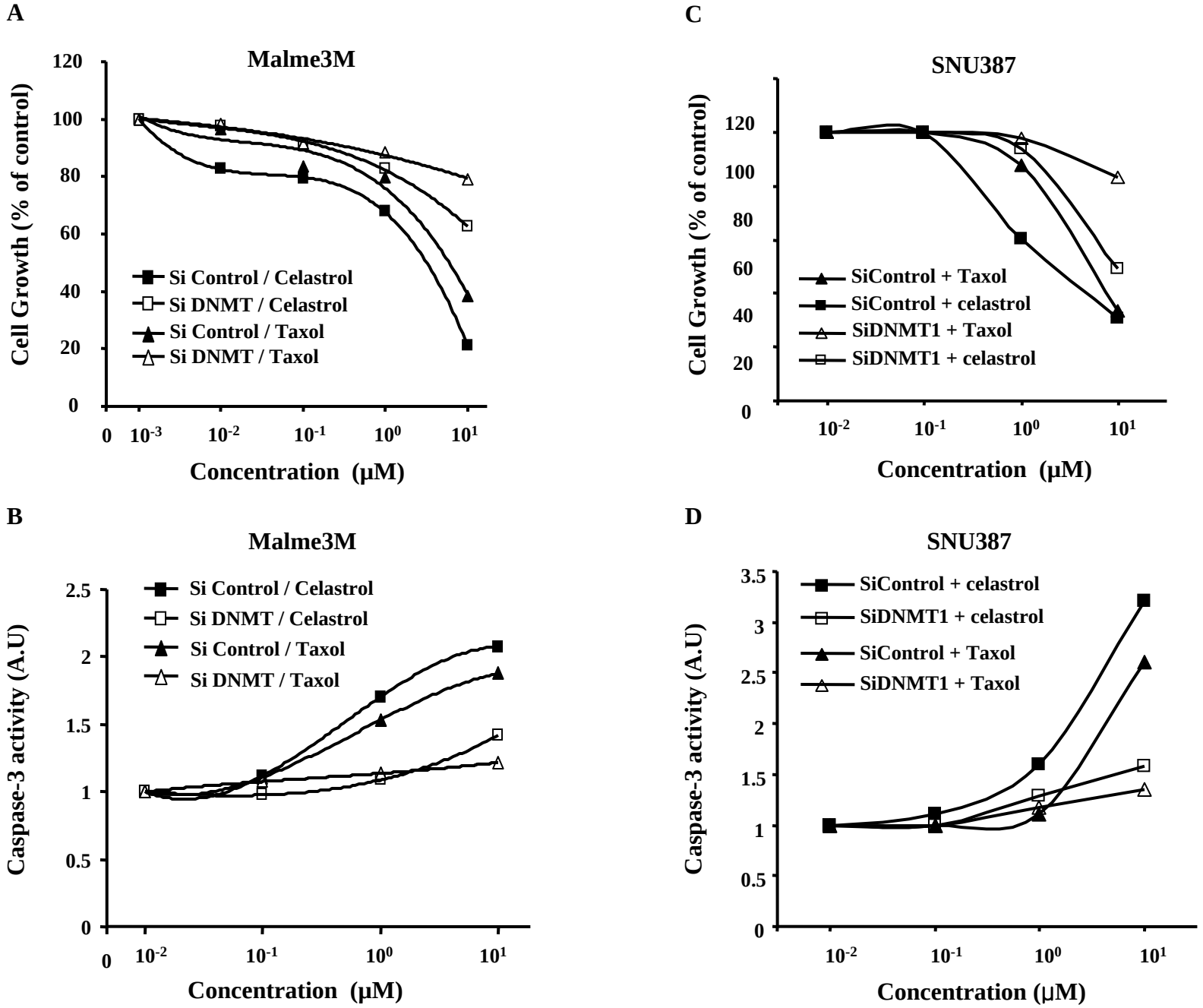
FIGURE 6. **CAGE regulates angiogenic potential of cancer cells.** Supernatants from SNU387 (A, upper panel) and Malme3M (B, upper panel) cells transiently transfected with CAGE (1 μg) enhance tube formation in HUVEC. Supernatants from SNU387$^{R}$ (*A*, lower panel) and Malme3M$^{R}$ cells (B, lower panel), that are transiently tranfected with CAGE SiRNA (10 nM), show lower angiogenic potential than supernatant from transiently transfected with control siRNA. (C) Downregulation of CAGE leads to decreased expression of VEGFR1 in drug-resistant cancer cell lines. .

FIGURE 7. **CAGE confers resistance to cytotoxic effect of CD8$^{+}$ T cells.** (A) Downregulation of CAGE leads to decreased expression of B7-H1, an activator of CD8$^{+}$ T cells, in SNU387$^{R}$ (left panel) and Malme3M$^{R}$ cells (right panel). (B) SNU387$^{R}$ and Malme3M$^{R}$ cells show resistance to cytotoxic effect of CD8$^{+}$ T cells based on MTT assays (upper panel). CD8$^{+}$ T cells (effector cells, $10^{4}$, $10^{5}$ cells /well) activated by CD3 ligation for 48 h were incubated with target cells ($10^{4}$cells/well) for 48 h. MTT assays were performed to count viable cells. *, p<0.05; **, p<0.005. (C), Expression level of CAGE determines sensitivity to CD8$^{+}$ T cells. Cells were transiently transfected with various constructs as indicated. At 48 h after transfection, activated CD8$^{+}$ T cells were incubated with target cells for 48 h, followed by MTT assays.

FIGURE 8. **HDAC2-snail complex represses p53 expression by CAGE.** (A) SNU387 cells were transiently transfected with control vector (1 μg) or CAGE cDNA (1 μg). Over expression of CAGE induces expression of snail. (B) Cellular fractionation shows that HDAC2 interacts

with snail in SNU387$^{R}$ cells. (C) Western blot shows that downregulation of snail restores expression of p53 in SNU387$^{R}$ cells and Malme3M$^{R}$ cells.

**FIGURE 1**


A
Malme3M
Cell Growth (% of control)
120
100
80
60
40
20
0
Si Control / Celastrol
Si DNMT / Celastrol
Si Control / Taxol
Si DNMT / Taxol
0
10-3
10-2
10-1
100
101
Concentration (µM)
C
SNU387
Cell Growth (% of control)
120
100
80
60
40
20
0
SiControl + Taxol
SiControl + celastrol
SiDNMT1 + Taxol
SiDNMT1 + celastrol
10-2
10-1
100
101
Concentration (µM)
B
Malme3M
Caspase-3 activity (A.U)
2.5
2
1.5
1
0.5
0
Si Control / Celastrol
Si DNMT / Celastrol
Si Control / Taxol
Si DNMT / Taxol
0
10-2
10-1
100
101
Concentration (µM)
D
SNU387
Caspase-3 activity (A.U)
3.5
3
2.5
2
1.5
1
0.5
0
SiControl + celastrol
SiDNMT1 + celastrol
SiControl + Taxol
SiDNMT1 + Taxol
10-2
10-1
100
101
Concentration (µM)

# FIGURE 2

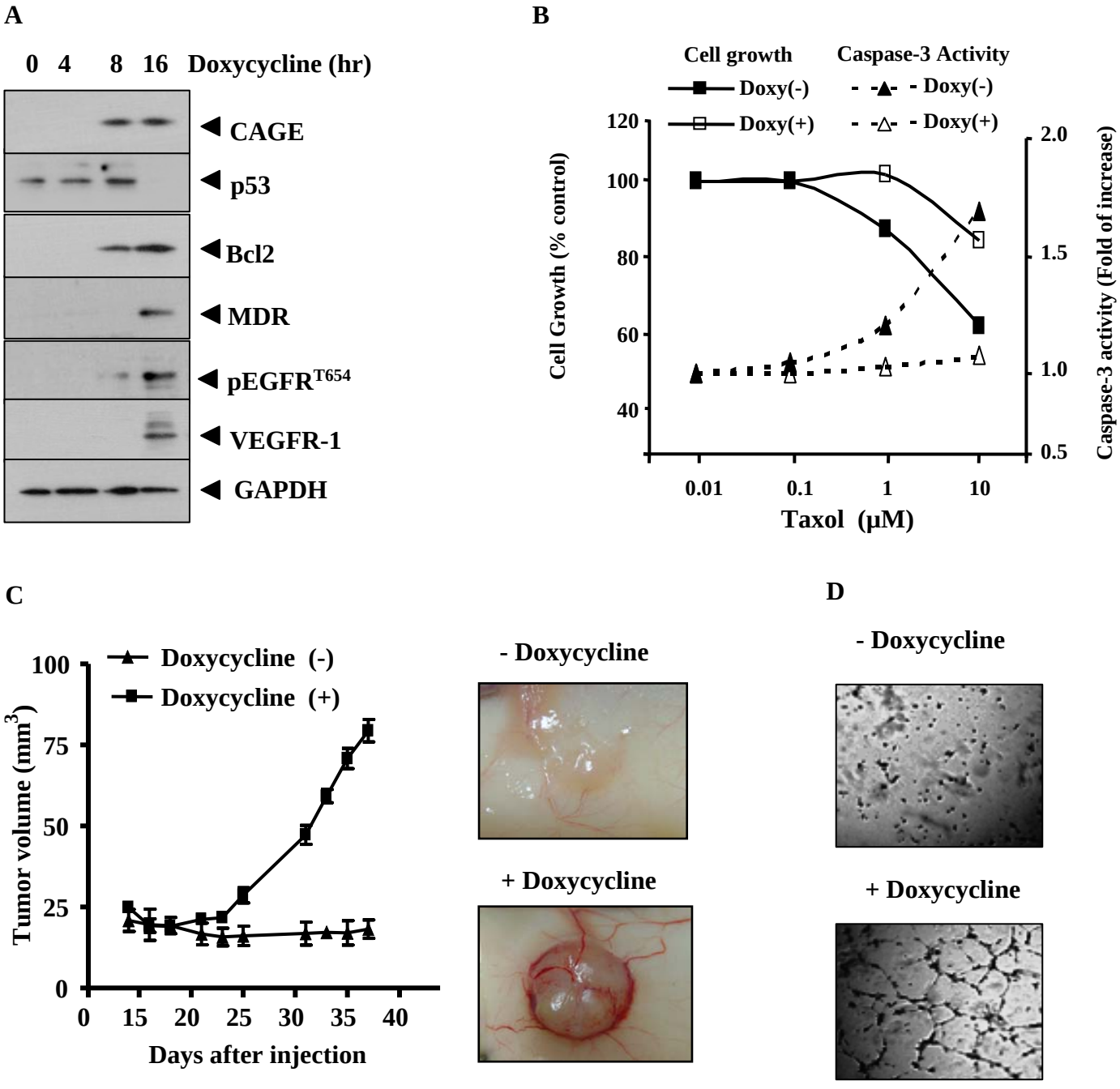
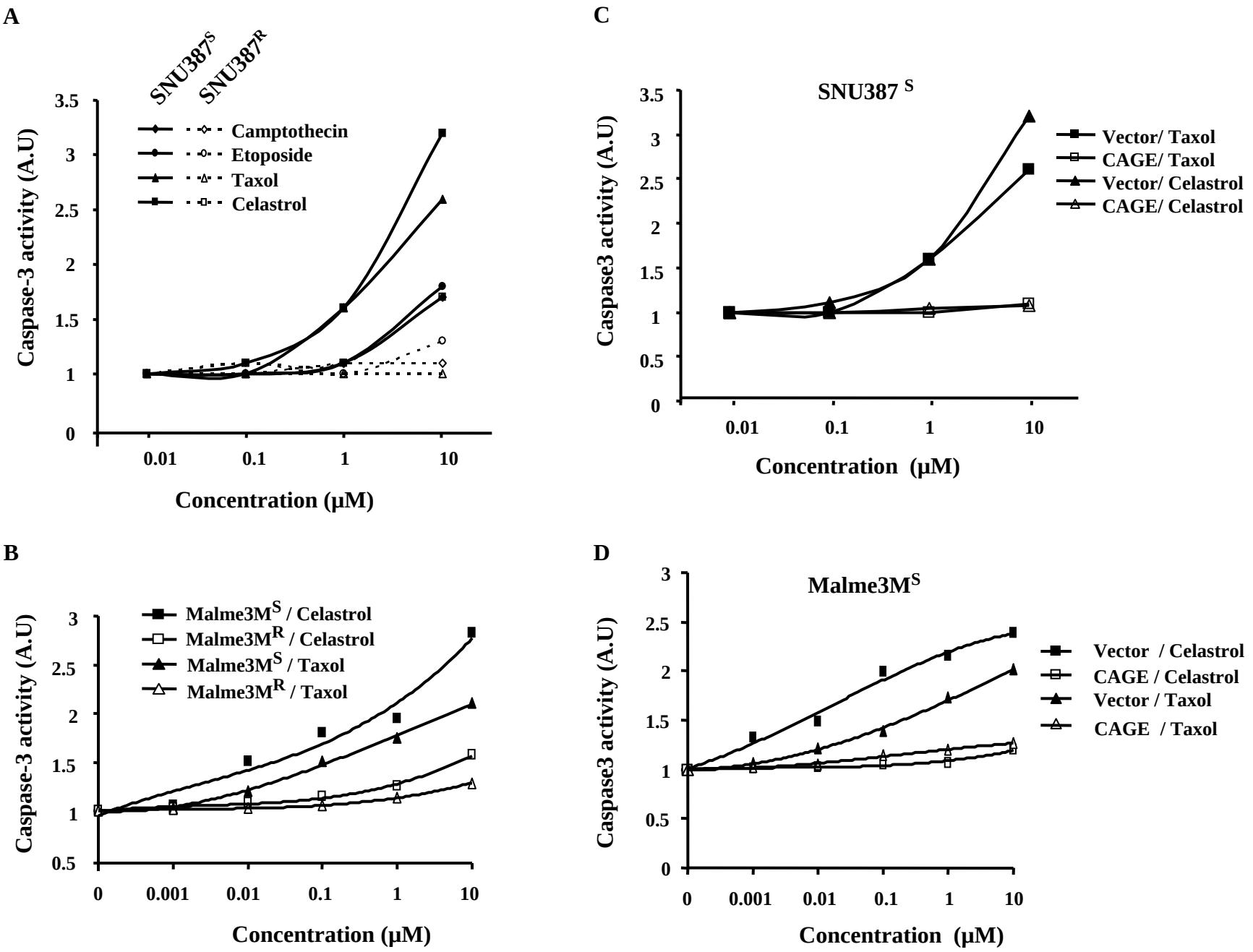
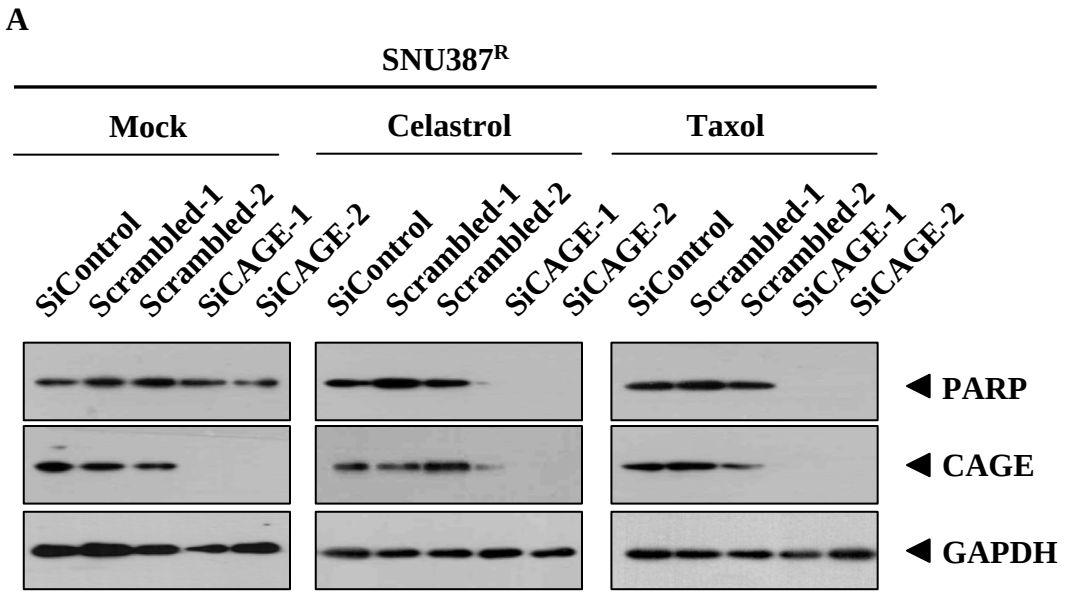
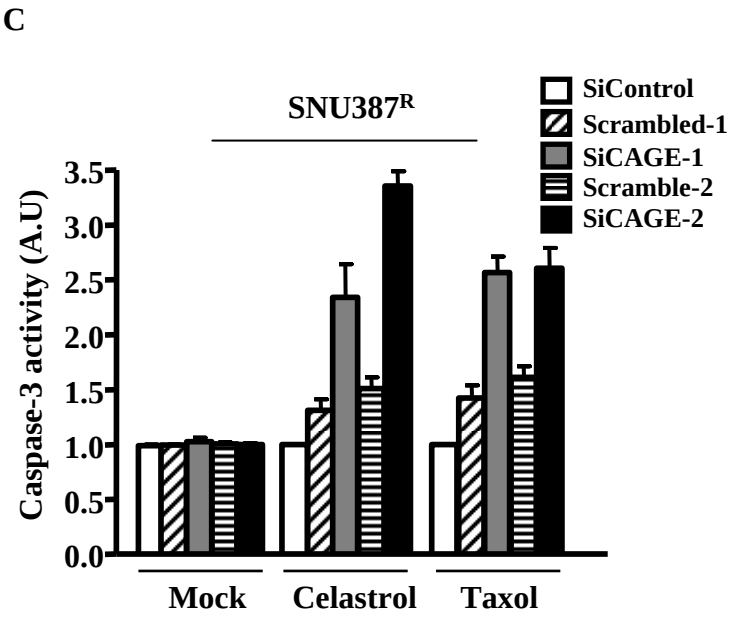

A
0 4 8 16 Doxycycline (hr)
CAGE
p53
Bcl2
MDR
pEGFR$^{T654}$
VEGFR-1
GAPDH
B
Cell growth
Caspase-3 Activity
Doxy(-)
Doxy(+)
Doxy(-)
Doxy(+)
Cell Growth (% control)
Caspase-3 activity (Fold of increase)
120
100
80
60
40
2.0
1.5
1.0
0.5
0.01
0.1
1
10
Taxol (μM)
C
Doxycycline (-)
Doxycycline (+)
Tumor volume (mm$^3$)
100
75
50
25
0
0 15 20 25 30 35 40
Days after injection
- Doxycycline
+ Doxycycline
D
- Doxycycline
+ Doxycycline

**FIGURE 3**


A
SNU387S
SNU387R
Camptothecin
Etoposide
Taxol
Celastrol
Caspase-3 activity (A.U)
Concentration (μM)
B
Malme3MS / Celastrol
Malme3MR / Celastrol
Malme3MS / Taxol
Malme3MR / Taxol
Caspase-3 activity (A.U)
Concentration (μM)
C
SNU387 S
Vector/ Taxol
CAGE/ Taxol
Vector/ Celastrol
CAGE/ Celastrol
Caspase3 activity (A.U)
Concentration (μM)
D
Malme3MS
Vector / Celastrol
CAGE / Celastrol
Vector / Taxol
CAGE / Taxol
Caspase3 activity (A.U)
Concentration (μM)

# FIGURE 4


A
SNU387R
Mock
Celastrol
Taxol
SiControl
Scrambled-1
Scrambled-2
SiCAGE-1
SiCAGE-2
PARP
CAGE
GAPDH


C
SNU387R
SiControl
Scrambled-1
SiCAGE-1
Scramble-2
SiCAGE-2
Caspase-3 activity (A.U)
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
Mock
Celastrol
Taxol

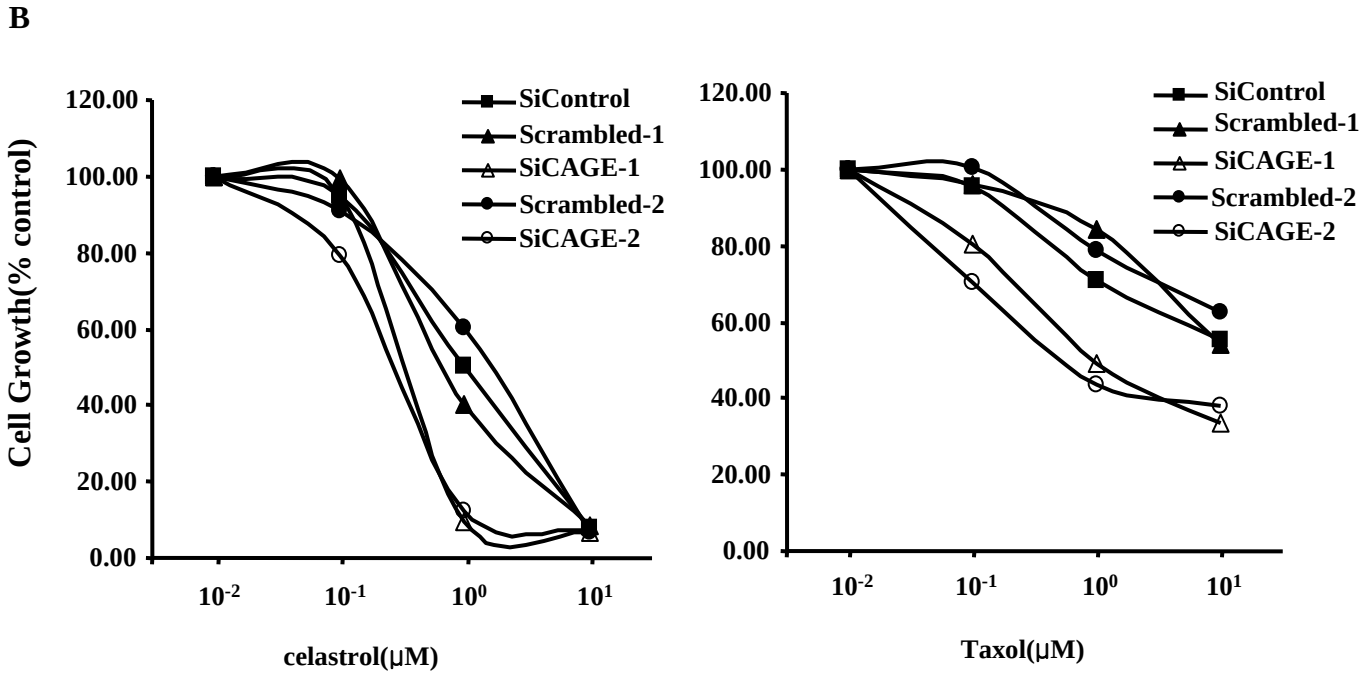
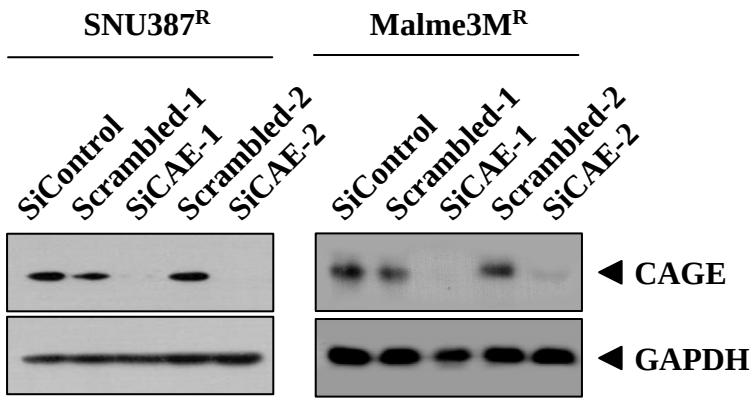
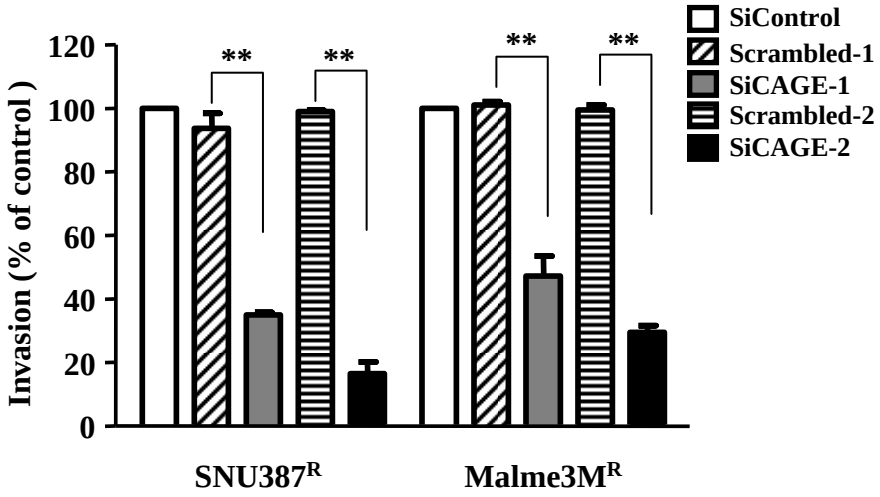

B
Cell Growth(% control)
120.00
100.00
80.00
60.00
40.00
20.00
0.00
SiControl
Scrambled-1
SiCAGE-1
Scrambled-2
SiCAGE-2
10-2
10-1
100
101
celastrol(μM)
Taxol(μM)

**FIGURE 5**

**A**


SNU387R
Malme3MR
SiControl
Scrambled-1
SiCAE-1
Scrambled-2
SiCAE-2
SiControl
Scrambled-1
SiCAE-1
Scrambled-2
SiCAE-2
CAGE
GAPDH

**B**


SiControl
Scrambled-1
SiCAGE-1
Scrambled-2
SiCAGE-2
Invasion (% of control )
120
100
80
60
40
20
0
**
**
**
**
SNU387R
Malme3MR

# FIGURE 6

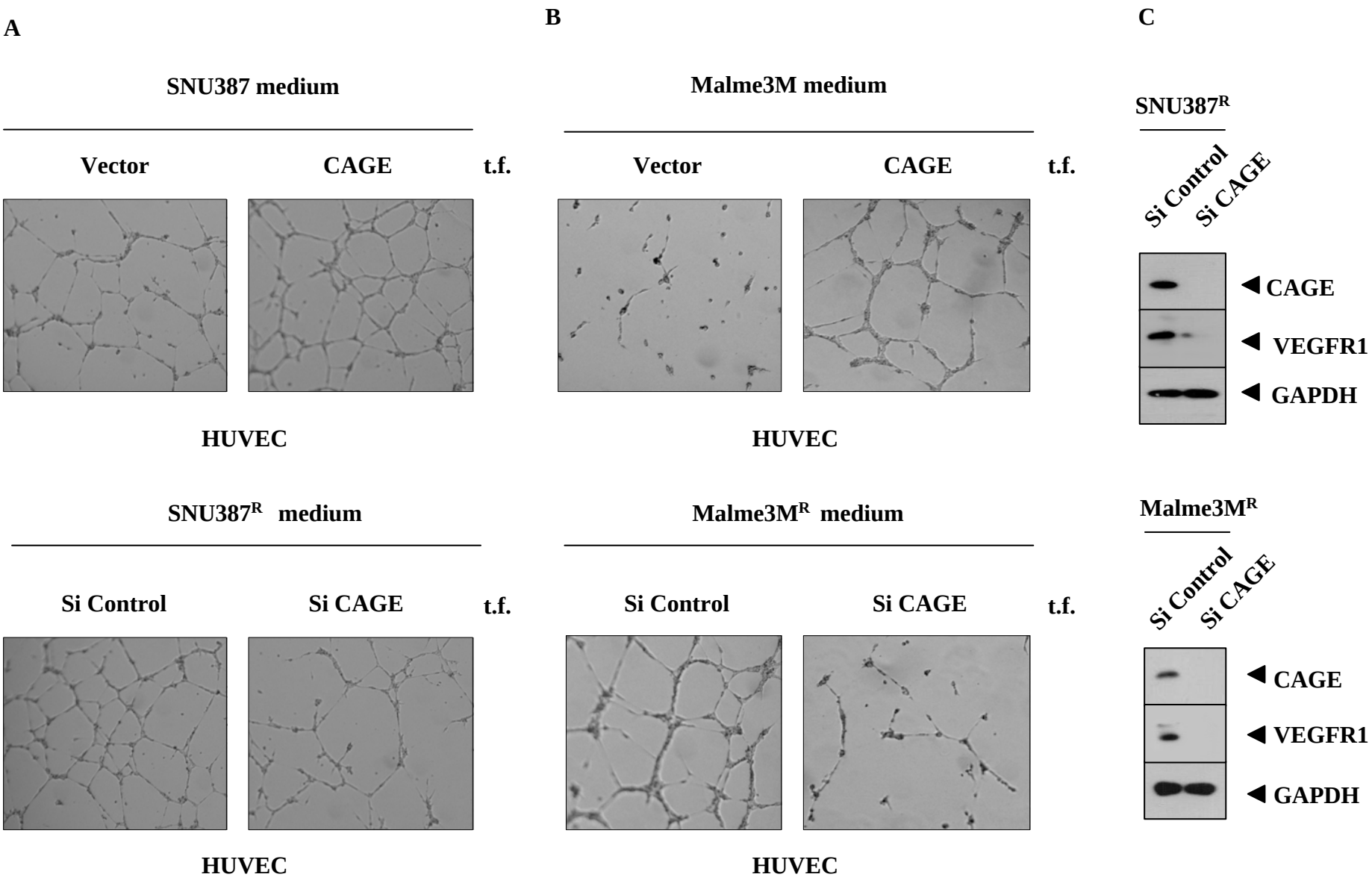

A
SNU387 medium
Vector
CAGE
t.f.
HUVEC
SNU387R medium
Si Control
Si CAGE
t.f.
HUVEC
B
Malme3M medium
Vector
CAGE
t.f.
HUVEC
Malme3MR medium
Si Control
Si CAGE
t.f.
HUVEC
C
SNU387R
Si Control
Si CAGE
CAGE
VEGFR1
GAPDH
Malme3MR
Si Control
Si CAGE
CAGE
VEGFR1
GAPDH

**FIGURE 7**

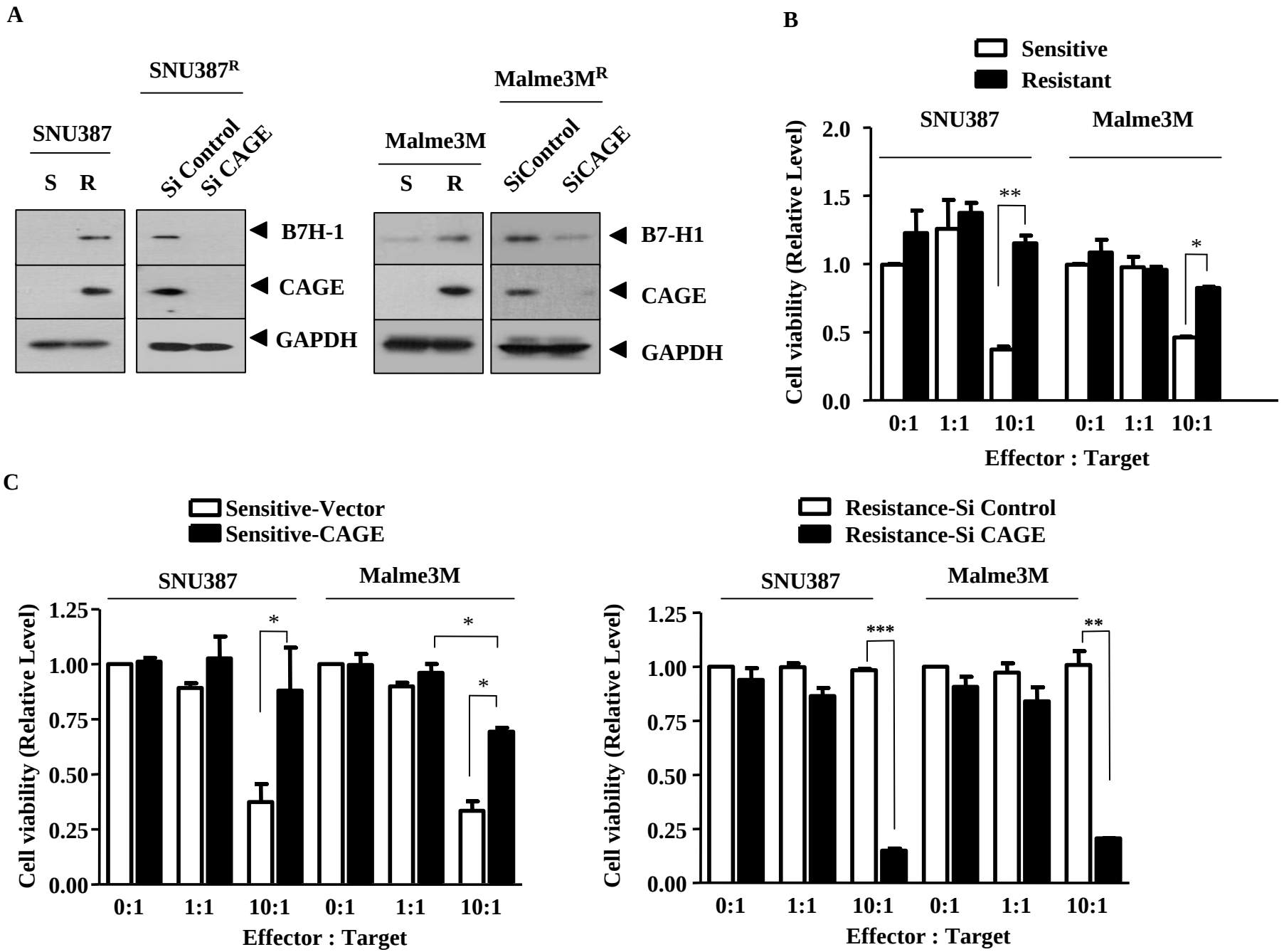
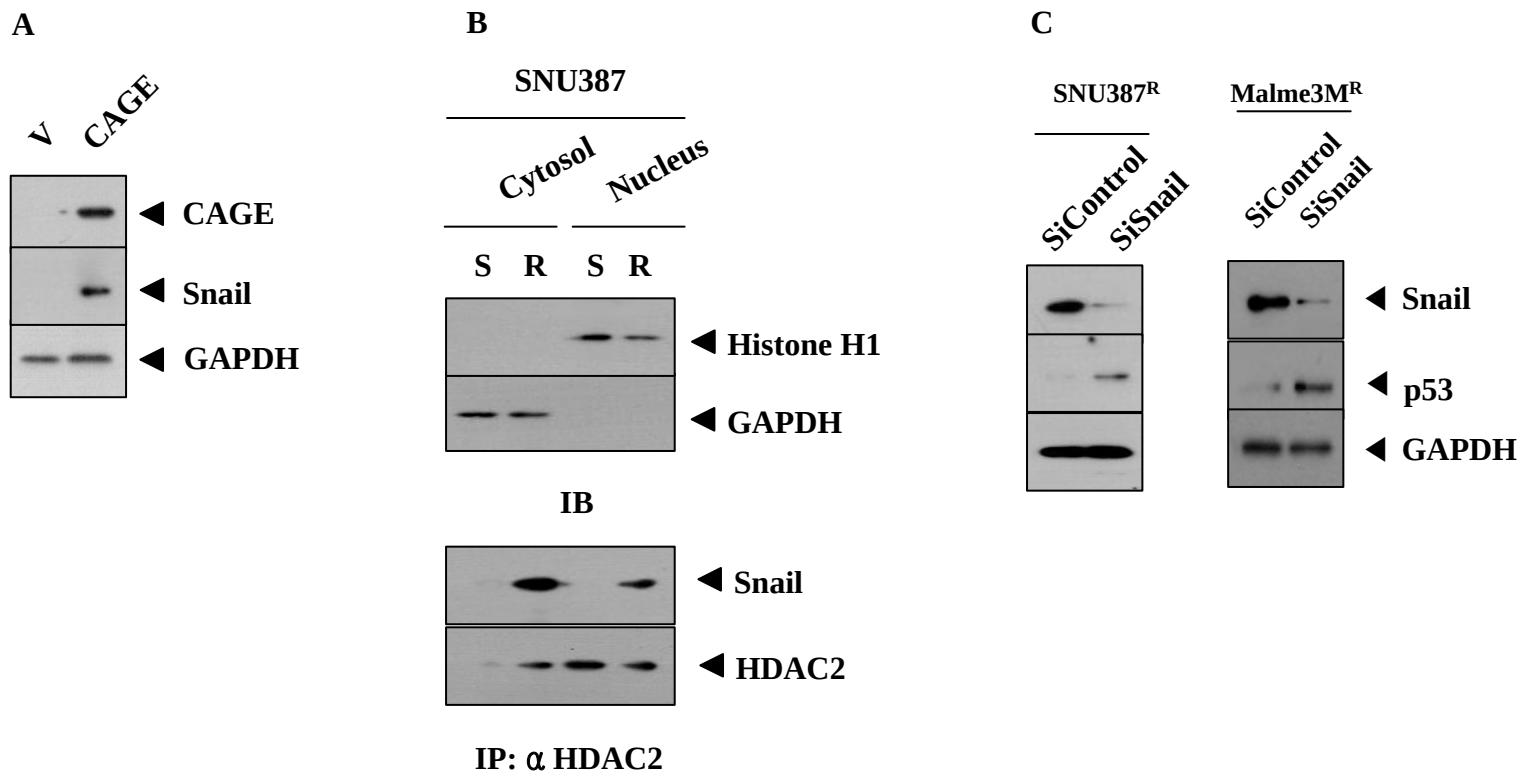

A
SNU387R
Malme3MR
SNU387
Si Control
Si CAGE
Malme3M
SiControl
SiCAGE
S
R
S
R
B7H-1
CAGE
GAPDH
B7-H1
CAGE
GAPDH
B
Sensitive
Resistant
SNU387
Malme3M
Cell viability (Relative Level)
2.0
1.5
1.0
0.5
0.0
**
*
0:1
1:1
10:1
0:1
1:1
10:1
Effector : Target
C
Sensitive-Vector
Sensitive-CAGE
SNU387
Malme3M
Cell viability (Relative Level)
1.25
1.00
0.75
0.50
0.25
0.00
*
*
*
0:1
1:1
10:1
0:1
1:1
10:1
Effector : Target
Resistance-Si Control
Resistance-Si CAGE
SNU387
Malme3M
Cell viability (Relative Level)
1.25
1.00
0.75
0.50
0.25
0.00
***
**
0:1
1:1
10:1
0:1
1:1
10:1
Effector : Target

**FIGURE 8**


A
V
CAGE
CAGE
Snail
GAPDH
B
SNU387
Cytosol
Nucleus
S
R
S
R
Histone H1
GAPDH
IB
Snail
HDAC2
IP: α HDAC2
C
SNU387R
Malme3MR
SiControl
SiSnail
SiControl
SiSnail
Snail
p53
GAPDH