Figure 1

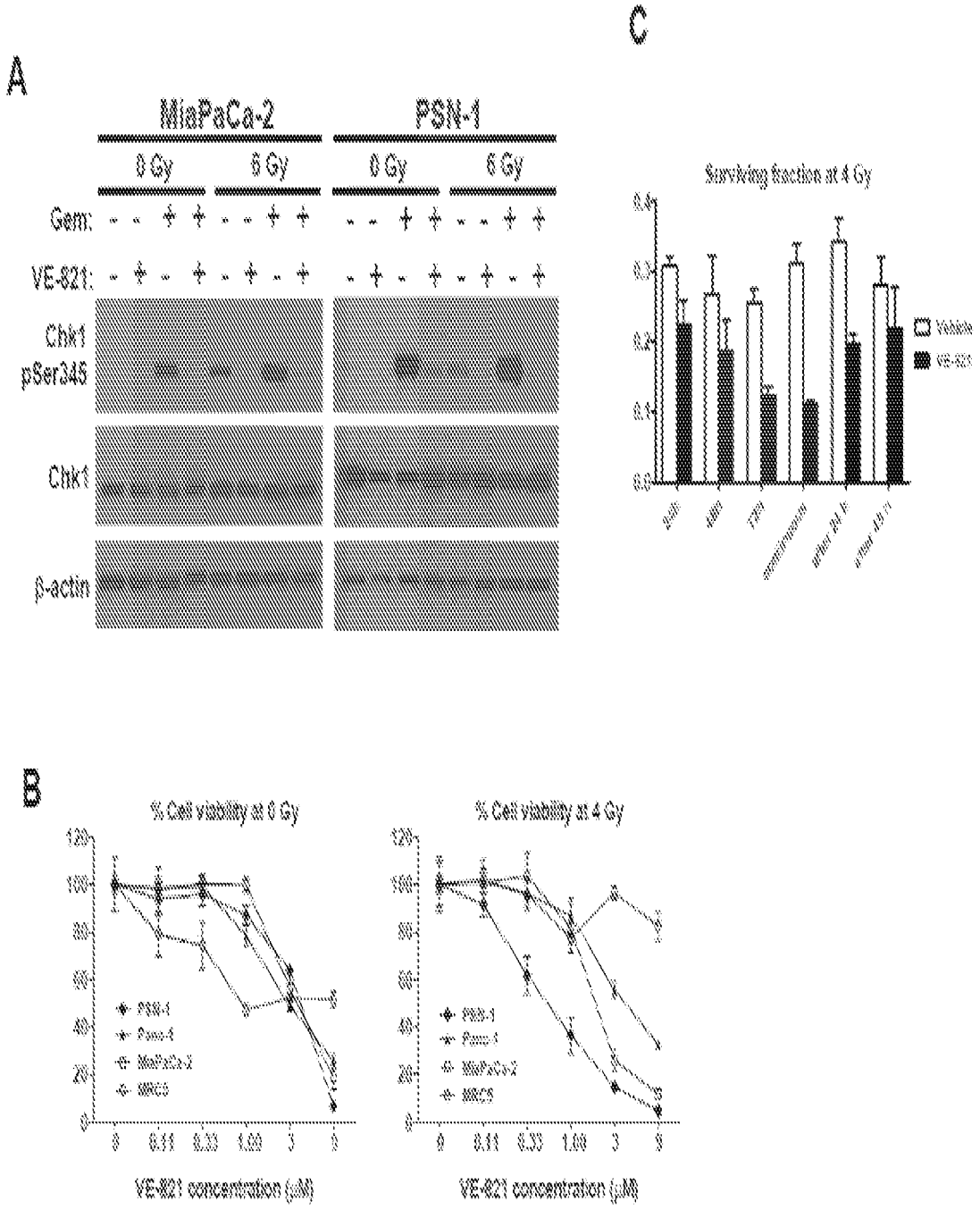

A
MiaPaCa-2
PSN-1
0 Gy
6 Gy
0 Gy
6 Gy
Gem:
VE-821:
Chk1 pSer345
Chk1
β-actin
C
Surviving fraction at 4 Gy
Vehicle
VE-821
B
% Cell viability at 0 Gy
% Cell viability at 4 Gy
VE-821 concentration (μM)
VE-821 concentration (μM)

Figure 1

D

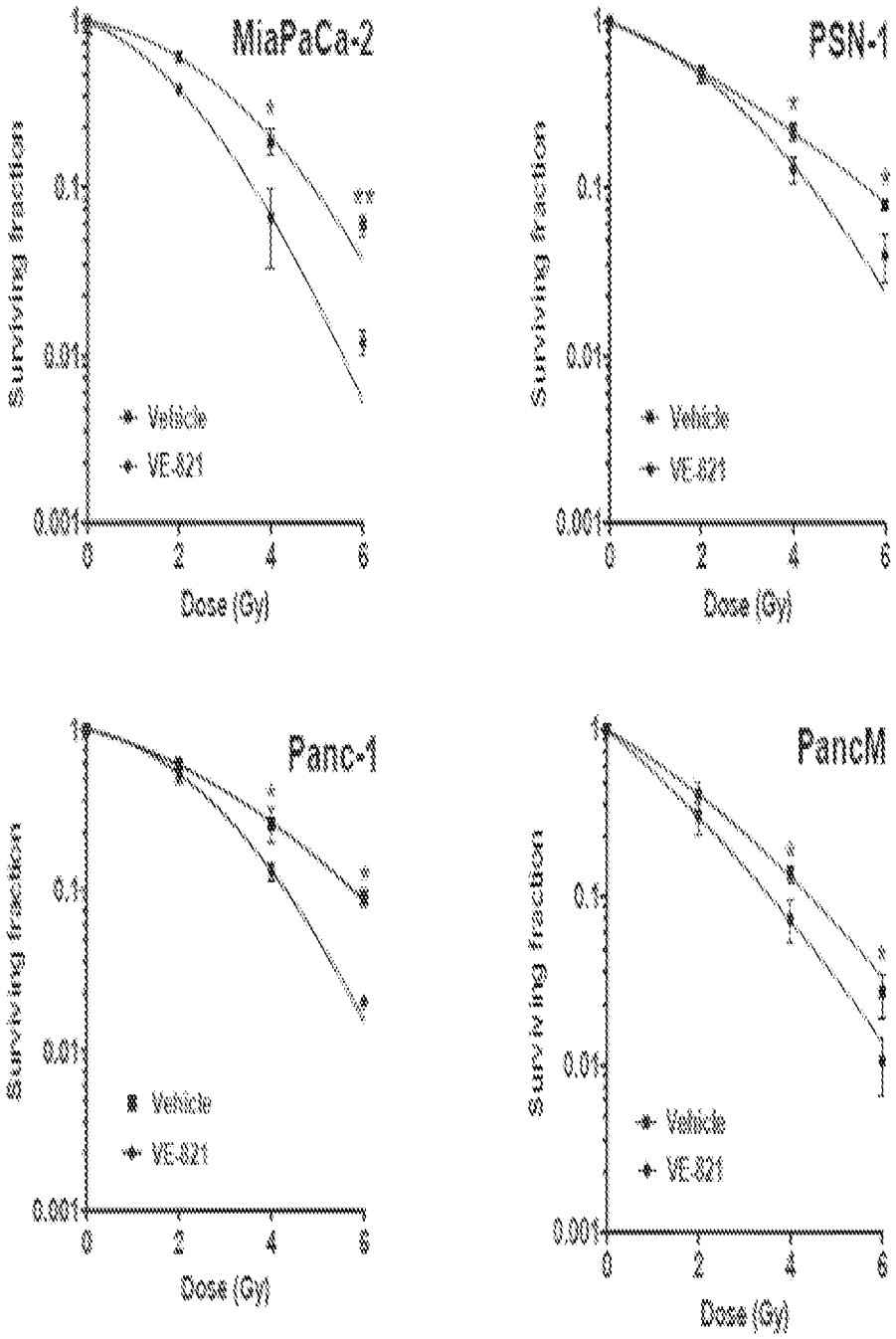

MiaPaCa-2
PSN-1
Panc-1
PancM
Surviving fraction
Dose (Gy)
Vehicle
VE-821
1
0.1
0.01
0.001
0
2
4
6

Figure 2

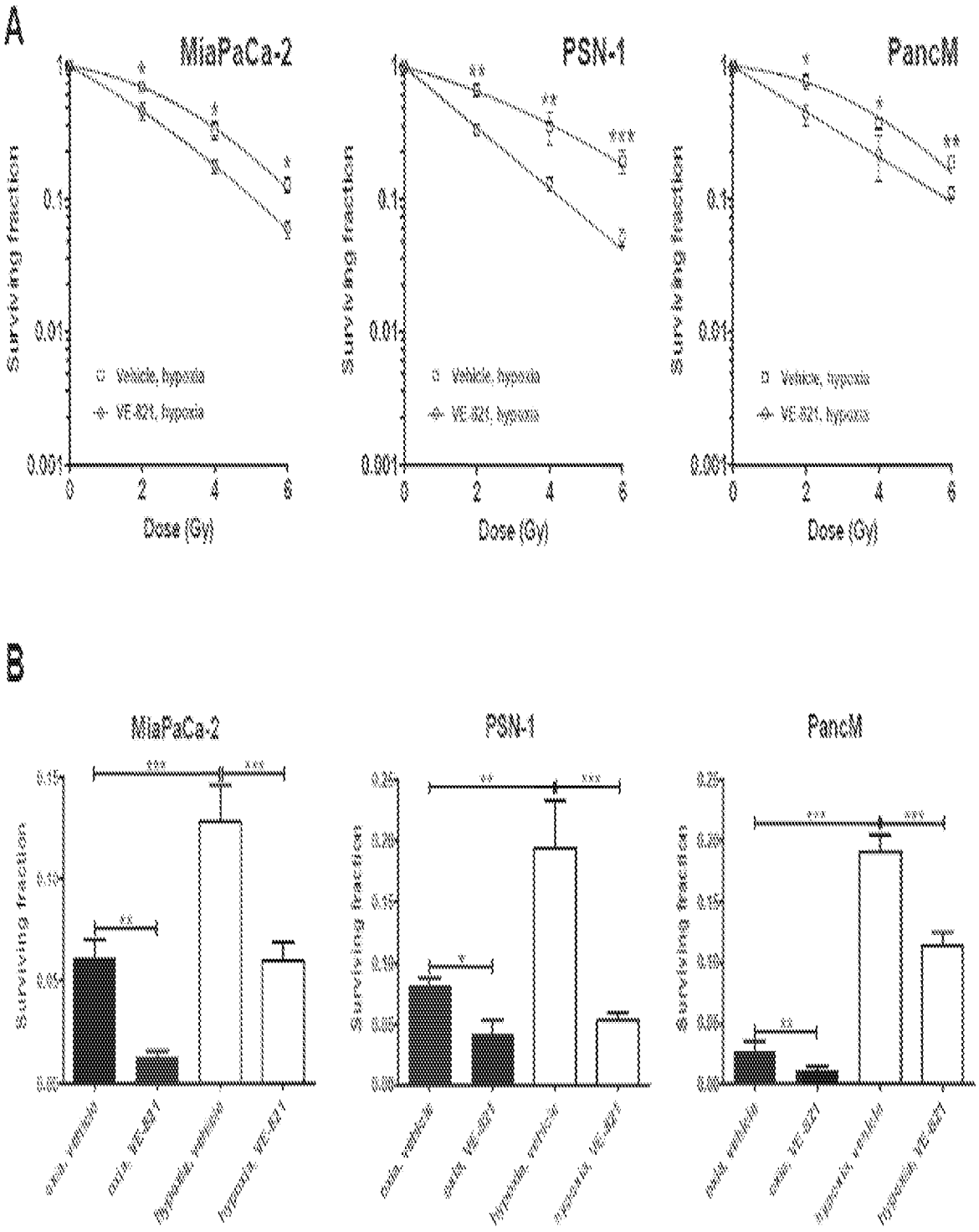

A
MiaPaCa-2
PSN-1
PancM
Surviving fraction
Dose (Gy)
Vehicle, hypoxia
VE-821, hypoxia
B
MiaPaCa-2
PSN-1
PancM
Surviving fraction

Figure 3

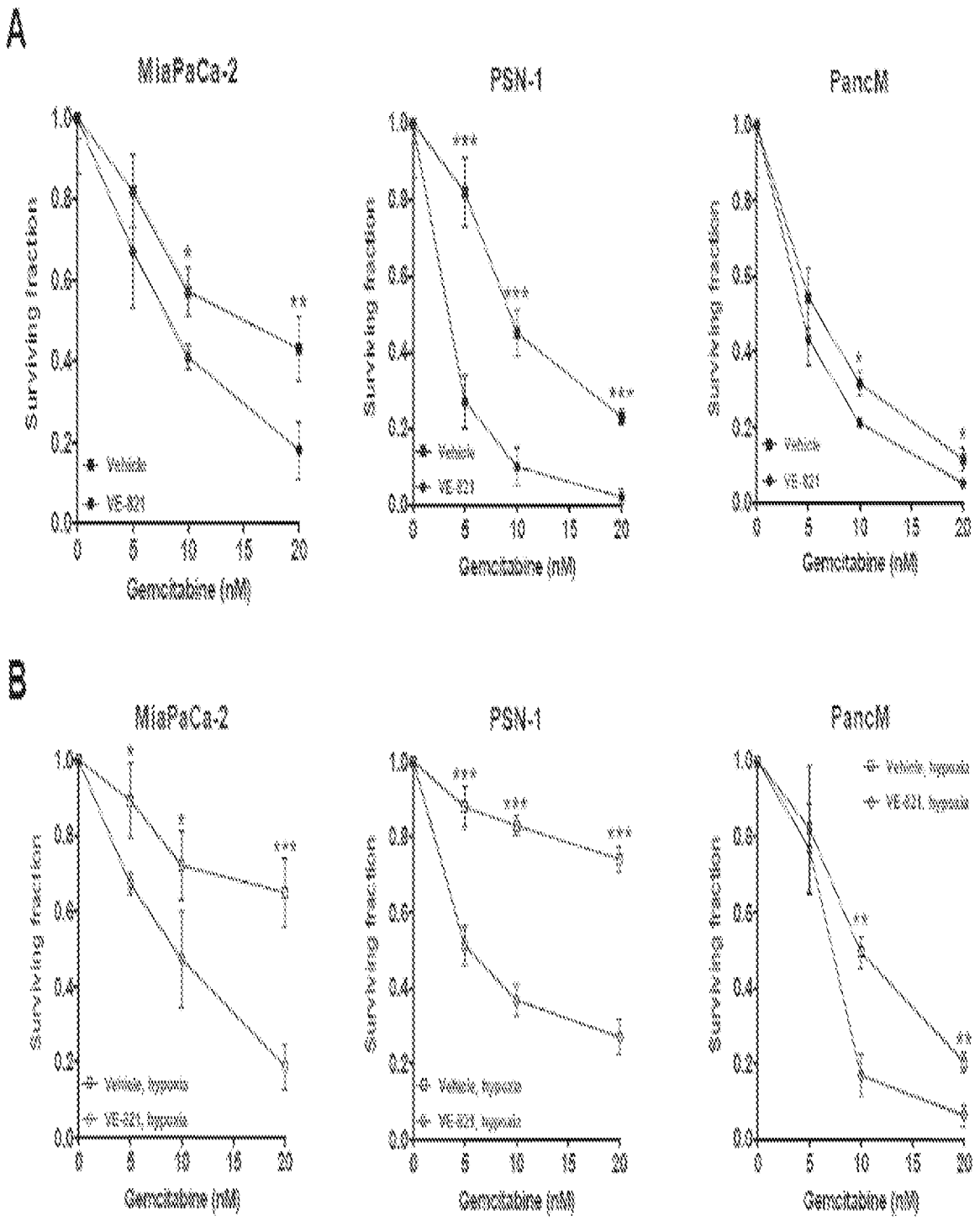
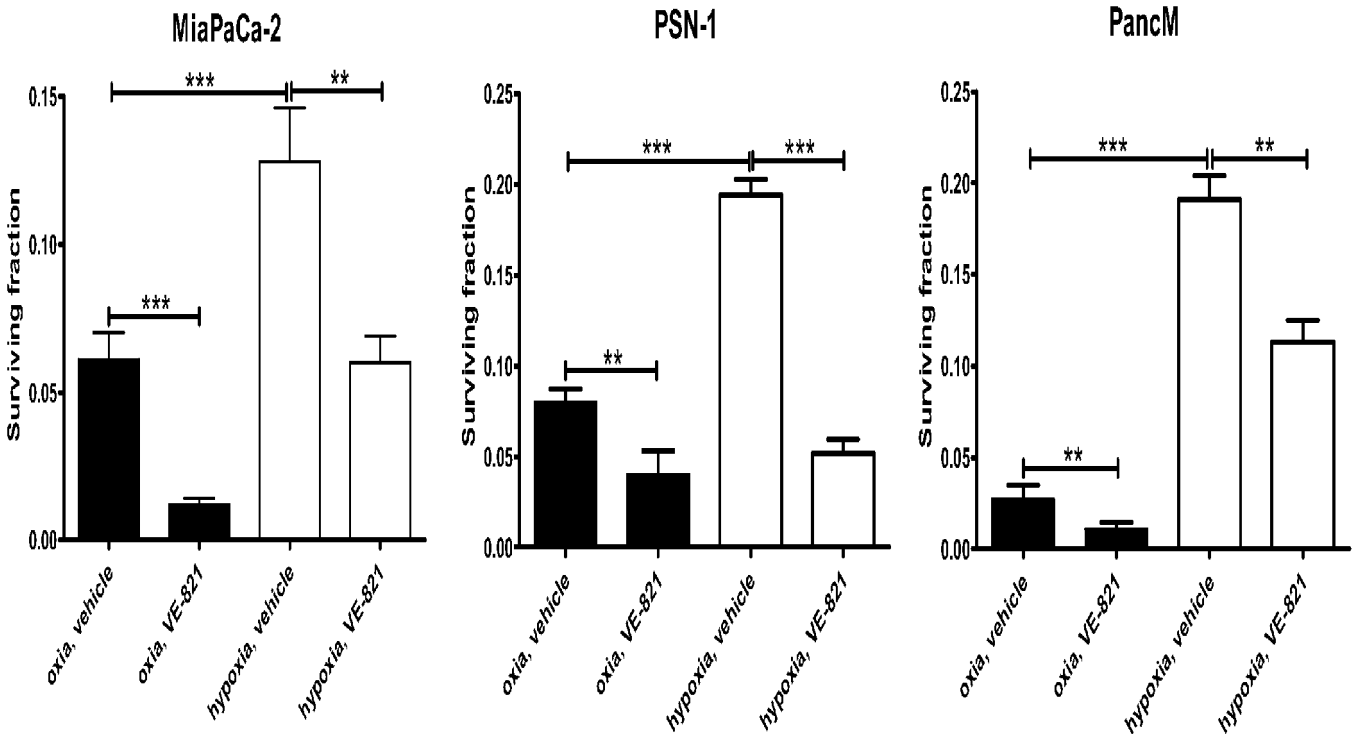

A
MiaPaCa-2
PSN-1
PancM
Surviving fraction
Gemcitabine (nM)
Vehicle
VE-821
B
MiaPaCa-2
PSN-1
PancM
Surviving fraction
Gemcitabine (nM)
Vehicle, hypoxia
VE-821, hypoxia

Figure 3

C


MiaPaCa-2
PSN-1
PancM
Surviving fraction
0.15
0.10
0.05
0.00
0.25
0.20
0.15
0.10
0.05
0.00
***
**
oxia, vehicle
oxia, VE-821
hypoxia, vehicle
hypoxia, VE-821

D

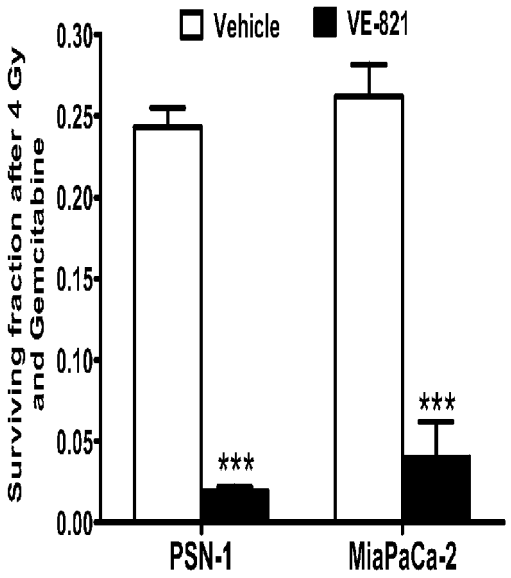

Vehicle
VE-821
Surviving fraction after 4 Gy and Gemcitabine
0.30
0.25
0.20
0.15
0.10
0.05
0.00
***
PSN-1
MiaPaCa-2

Figure 4

A

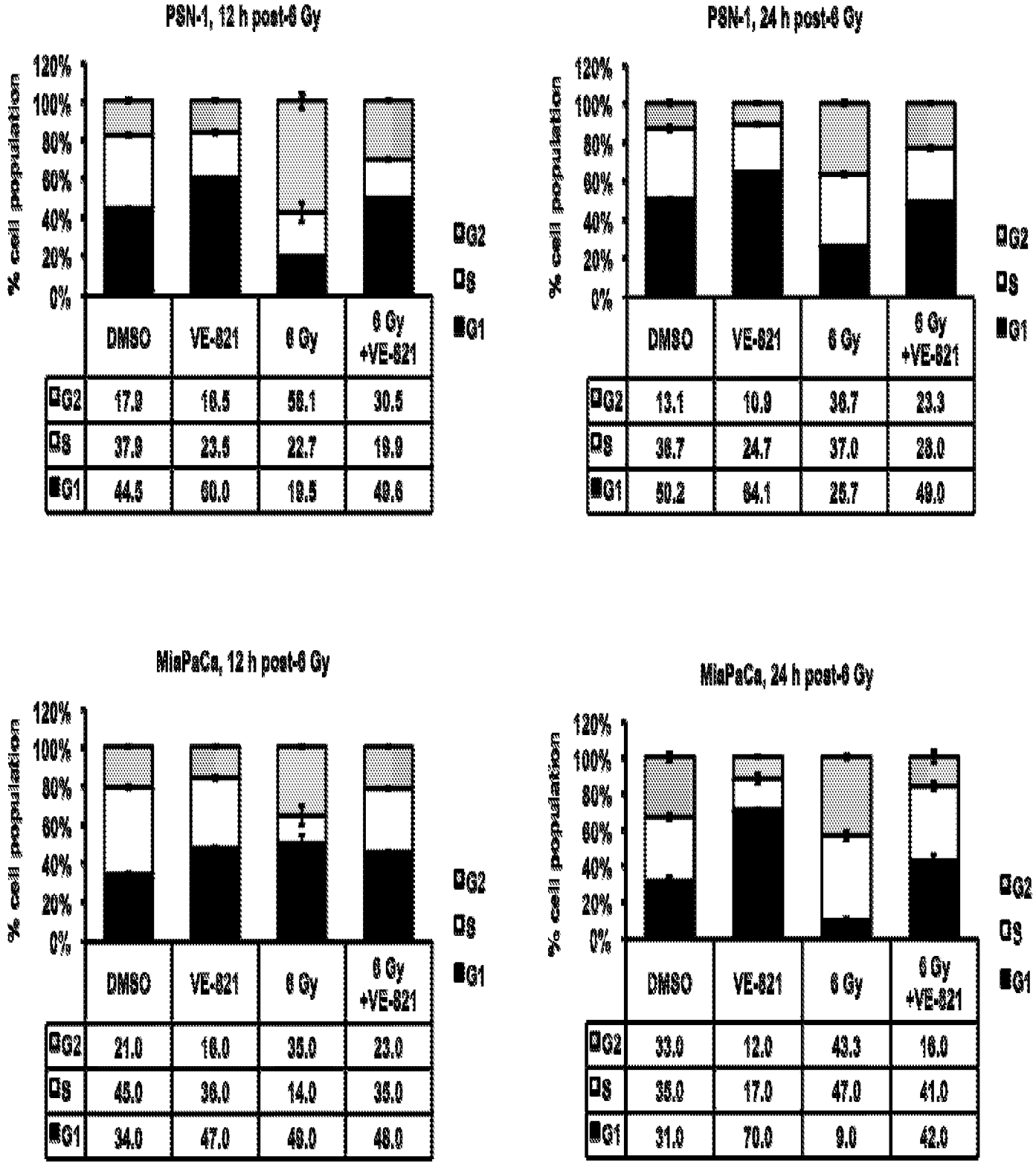
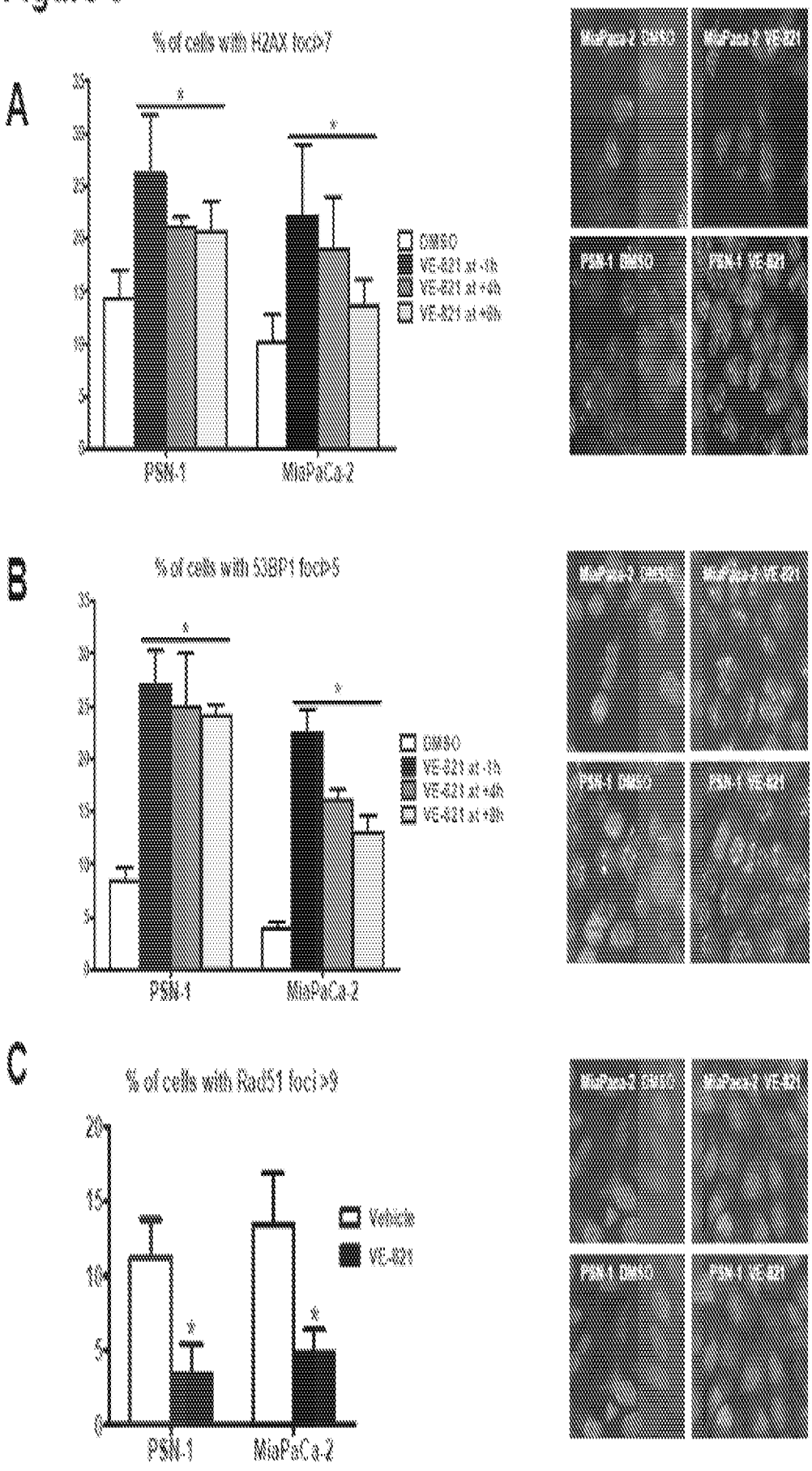
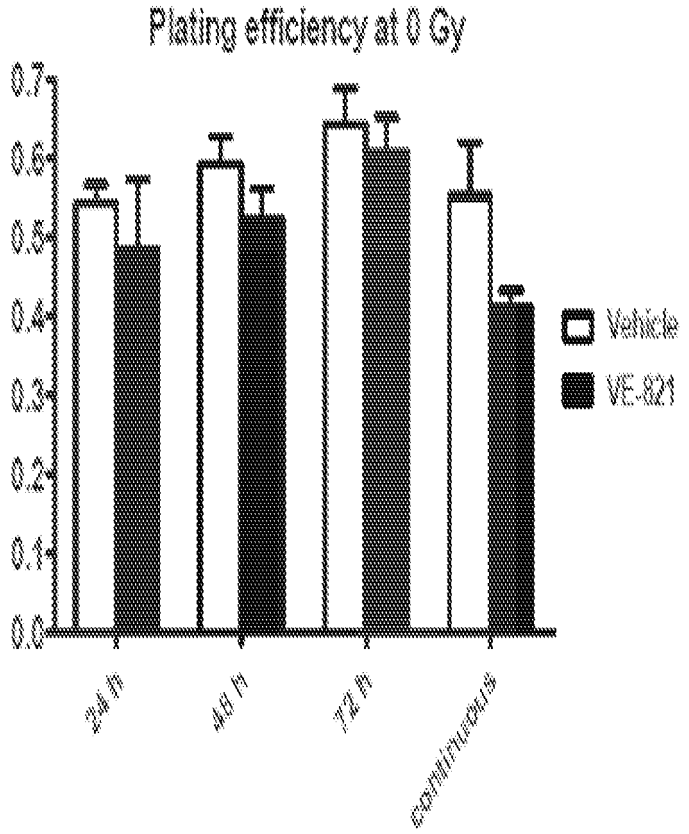

PSN-1, 12 h post-6 Gy
% cell population
120%
100%
80%
60%
40%
20%
0%
G2
S
G1
DMSO
VE-821
6 Gy
6 Gy +VE-821
G2 17.9 18.5 58.1 30.5
S 37.9 23.5 22.7 19.9
G1 44.5 60.0 19.5 49.6
PSN-1, 24 h post-6 Gy
G2 13.1 10.9 36.7 23.3
S 36.7 24.7 37.0 28.0
G1 50.2 64.1 25.7 49.0
MiaPaCa, 12 h post-6 Gy
G2 21.0 16.0 35.0 23.0
S 45.0 36.0 14.0 35.0
G1 34.0 47.0 49.0 48.0
MiaPaCa, 24 h post-6 Gy
G2 33.0 12.0 43.3 16.0
S 35.0 17.0 47.0 41.0
G1 31.0 70.0 9.0 42.0

Figure 5


A
% of cells with H2AX foci>7
PSN-1
MiaPaCa-2
DMSO
VE-821 at -1h
VE-821 at +4h
VE-821 at +8h
B
% of cells with 53BP1 foci>5
PSN-1
MiaPaCa-2
C
% of cells with Rad51 foci>5
Vehicle
VE-821
PSN-1
MiaPaCa-2

Supplementary Fig. 1



Supplementary Fig. 2

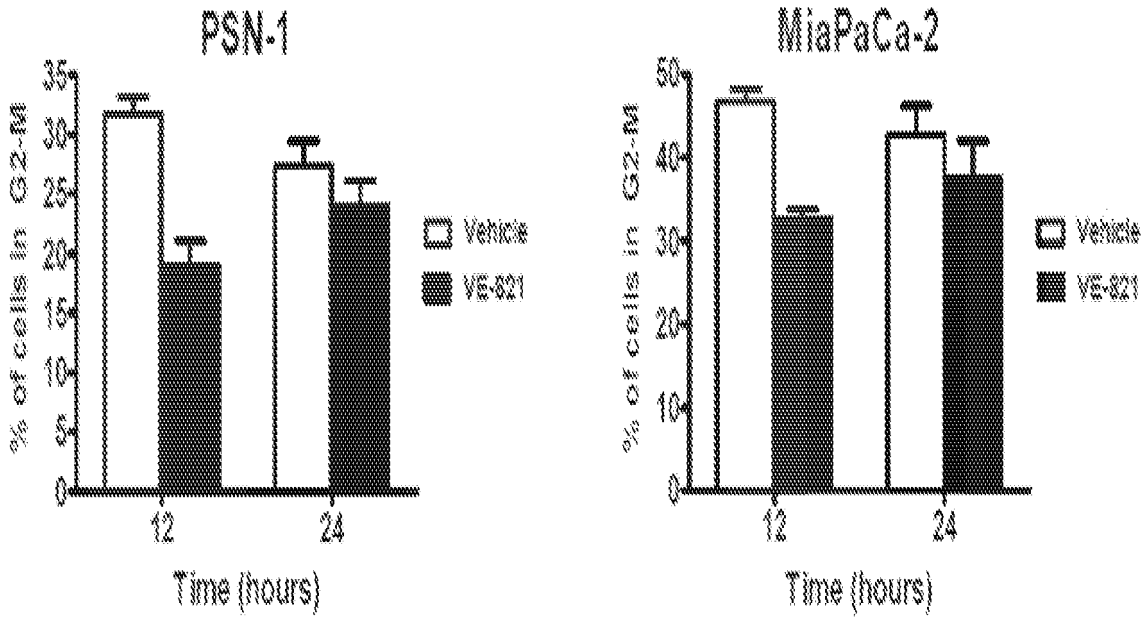


Figure 1X

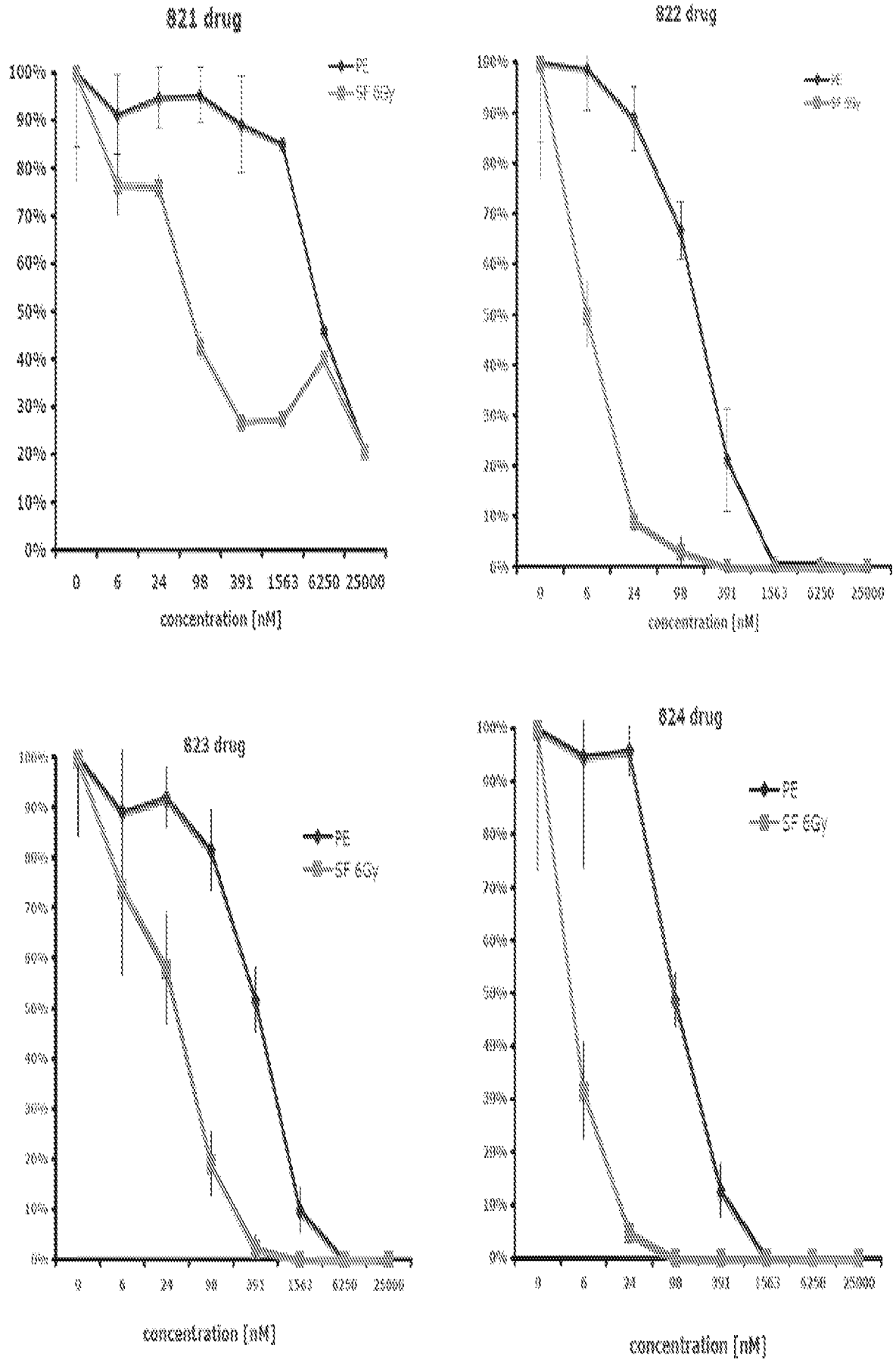

821 drug
822 drug
823 drug
824 drug
PE
SF 6Gy
100%
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
0
6
24
98
391
1563
6250
25000
concentration [nM]

Figure 2X

A

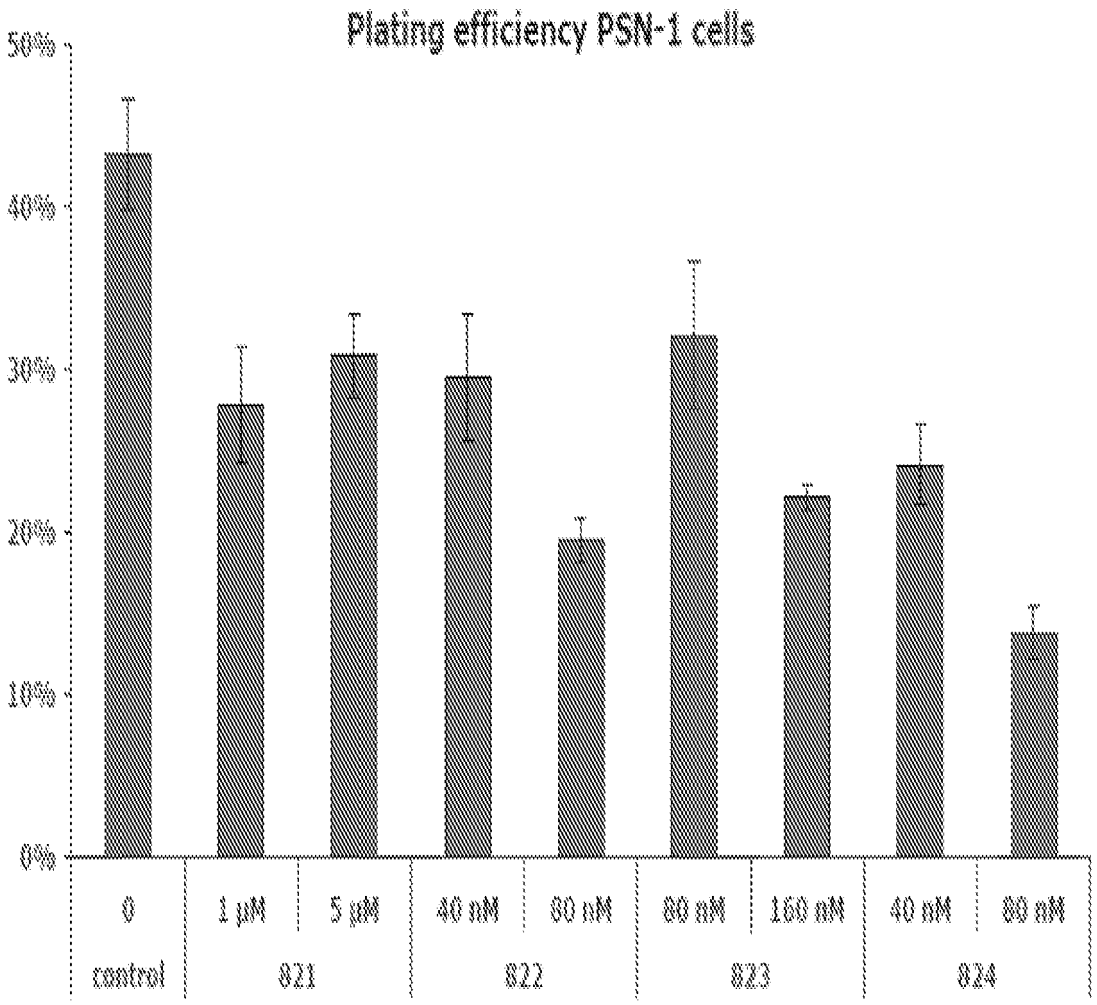

Plating efficiency PSN-1 cells
50%
40%
30%
20%
10%
0%
0
1 μM
5 μM
40 nM
80 nM
80 nM
160 nM
40 nM
80 nM
control
821
822
823
824

Figure 2X

A

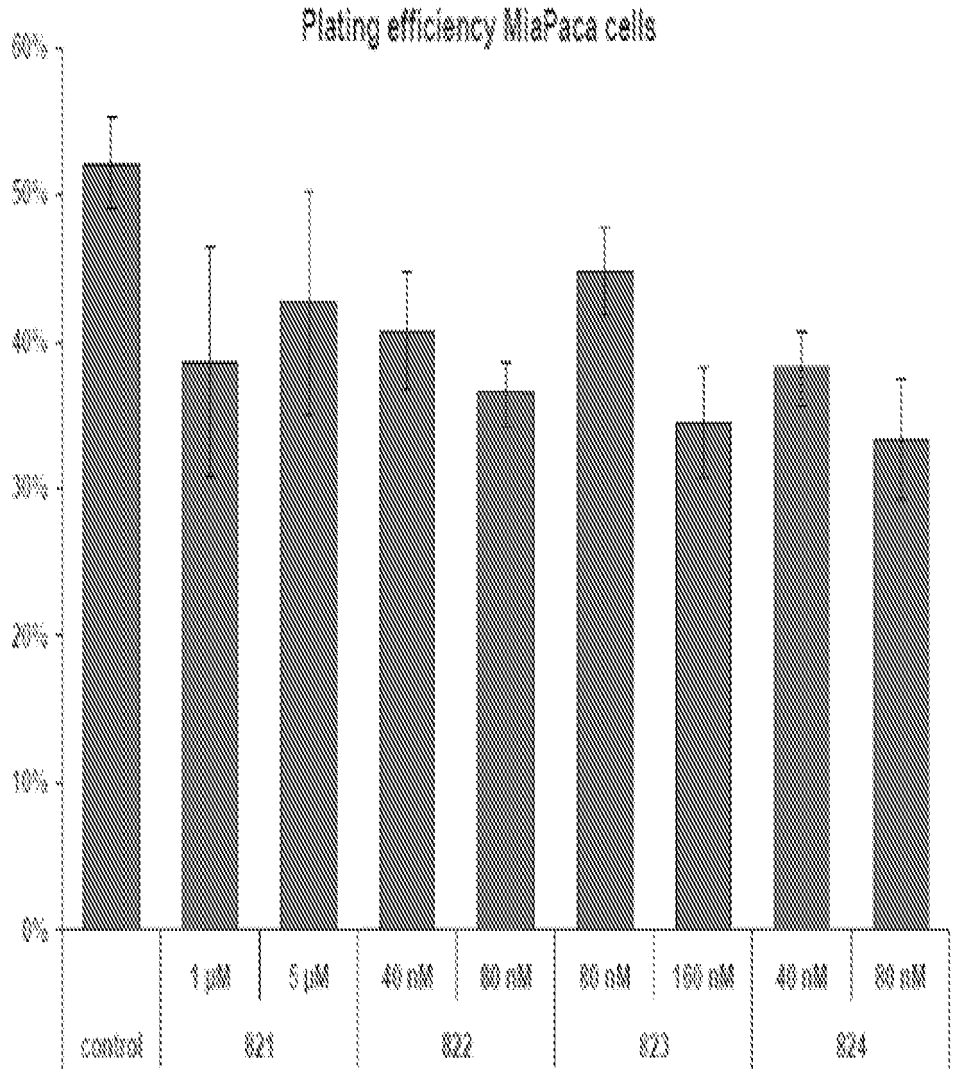

Plating efficiency MiaPaca cells
60%
50%
40%
30%
20%
10%
0%
control
1 µM
5 µM
40 nM
80 nM
80 nM
160 nM
40 nM
80 nM
821
822
823
824

Figure 2X

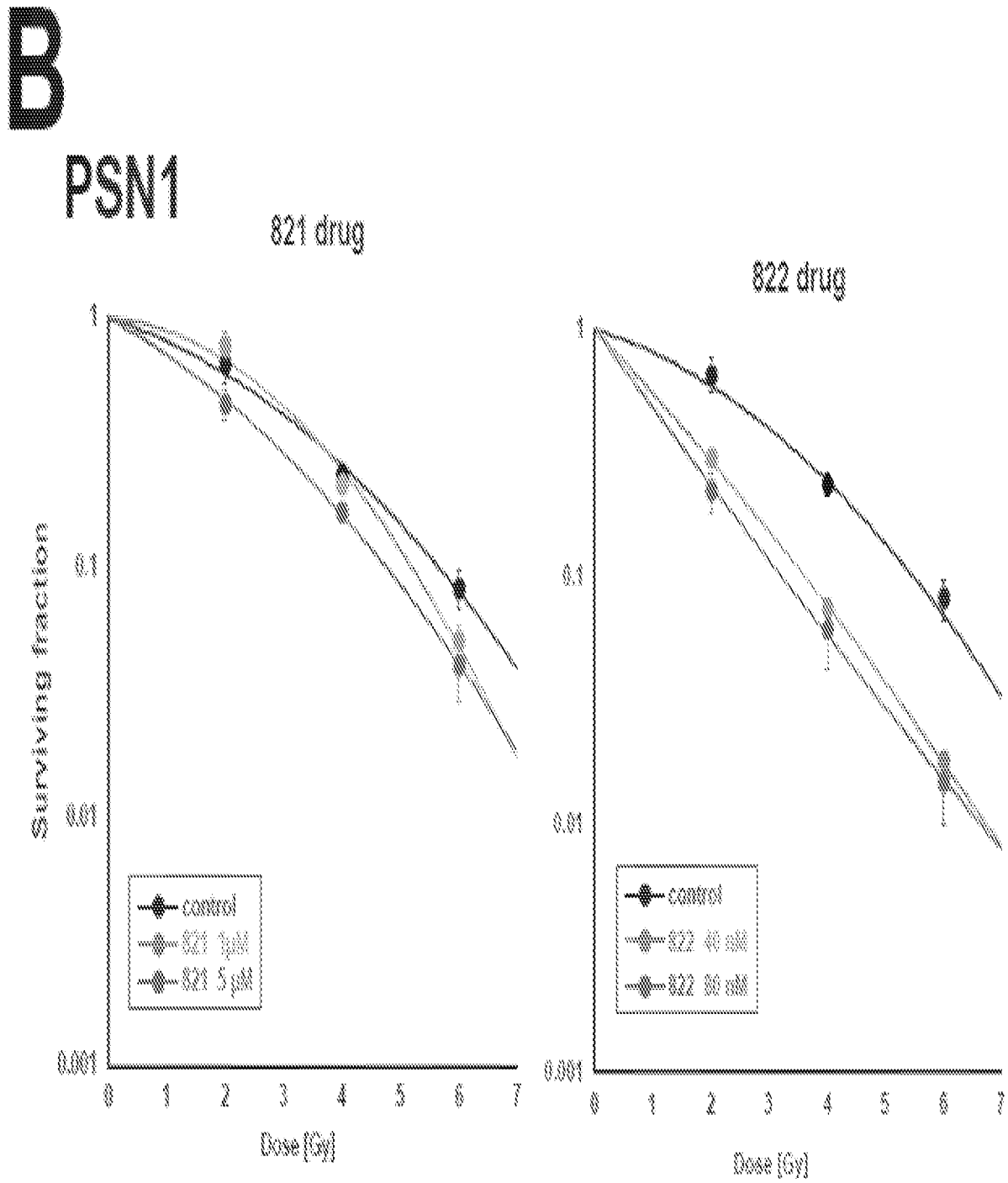

B
PSN1
821 drug
822 drug
surviving fraction
1
0.1
0.01
0.001
control
821 1 μM
821 5 μM
822 40 nM
822 80 nM
0
1
2
3
4
5
6
7
Dose [Gy]

Figure 2X

B

PSN1

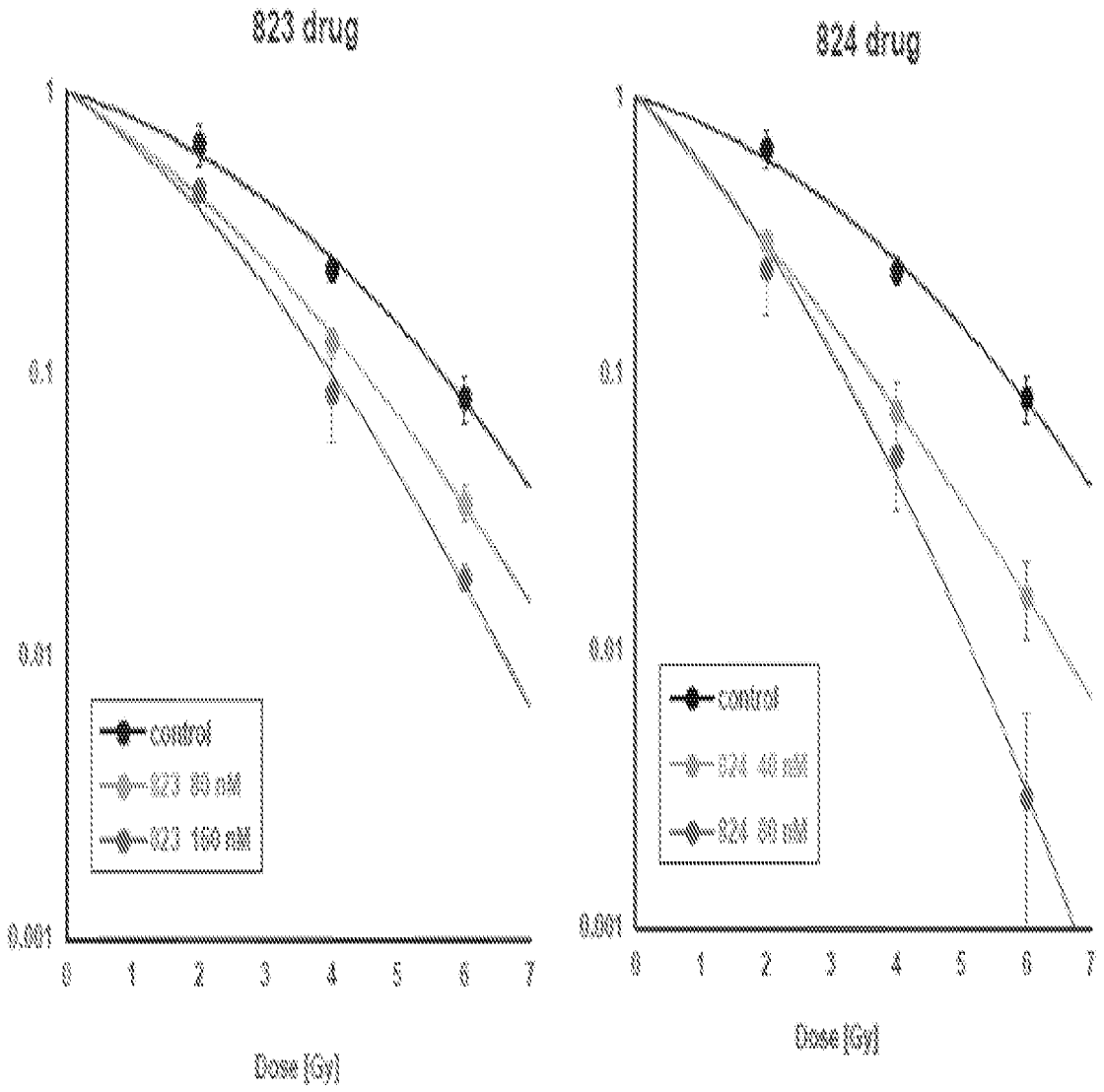


Figure 2X

B

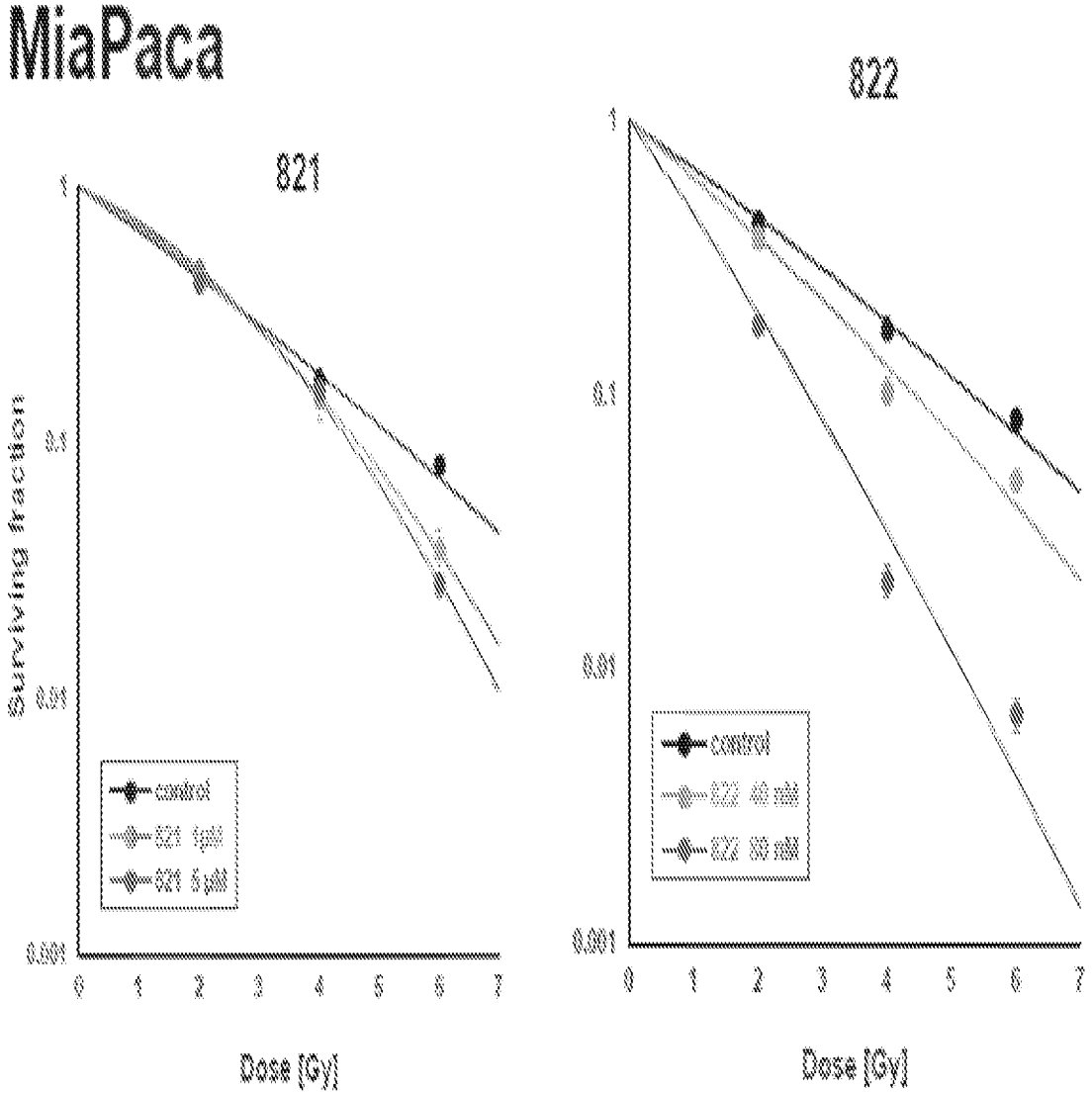


Figure 2X

B

MiaPaca

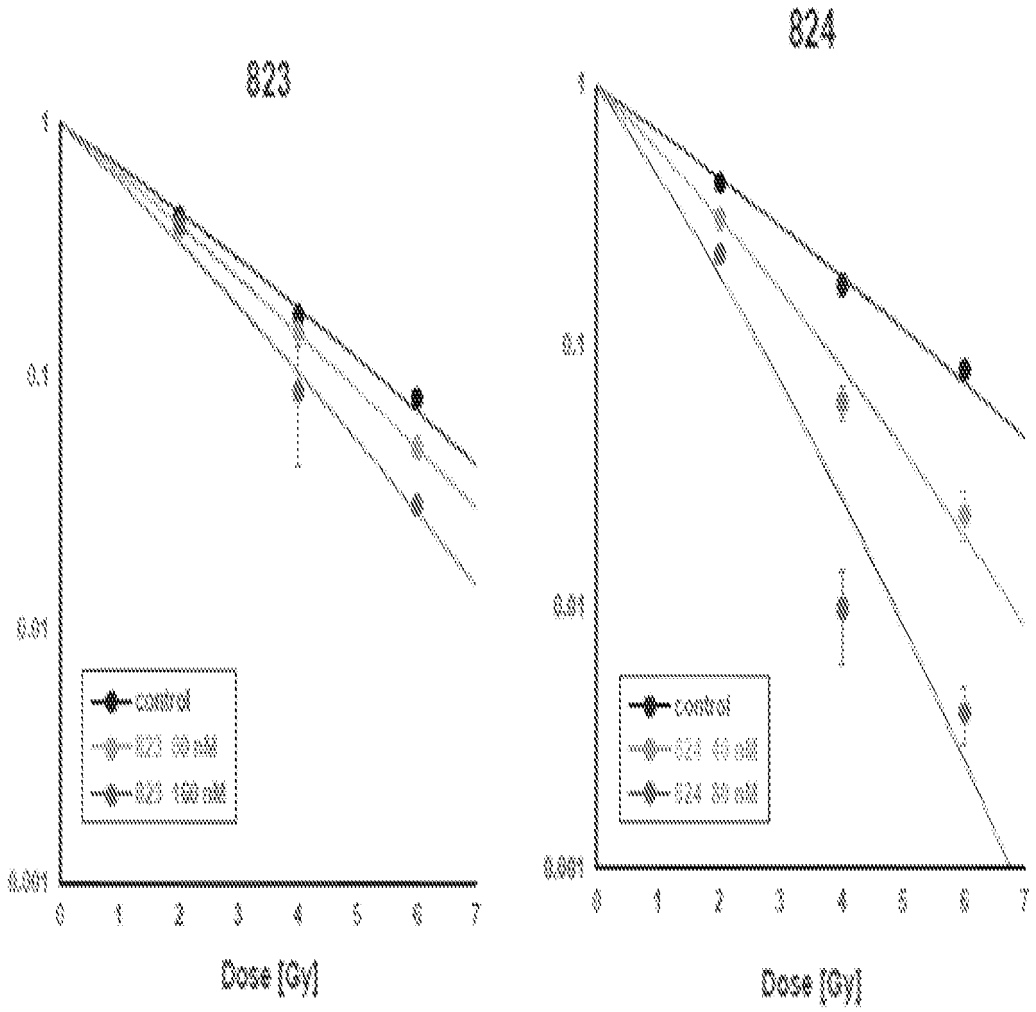


Figure 2X

B

MRC5

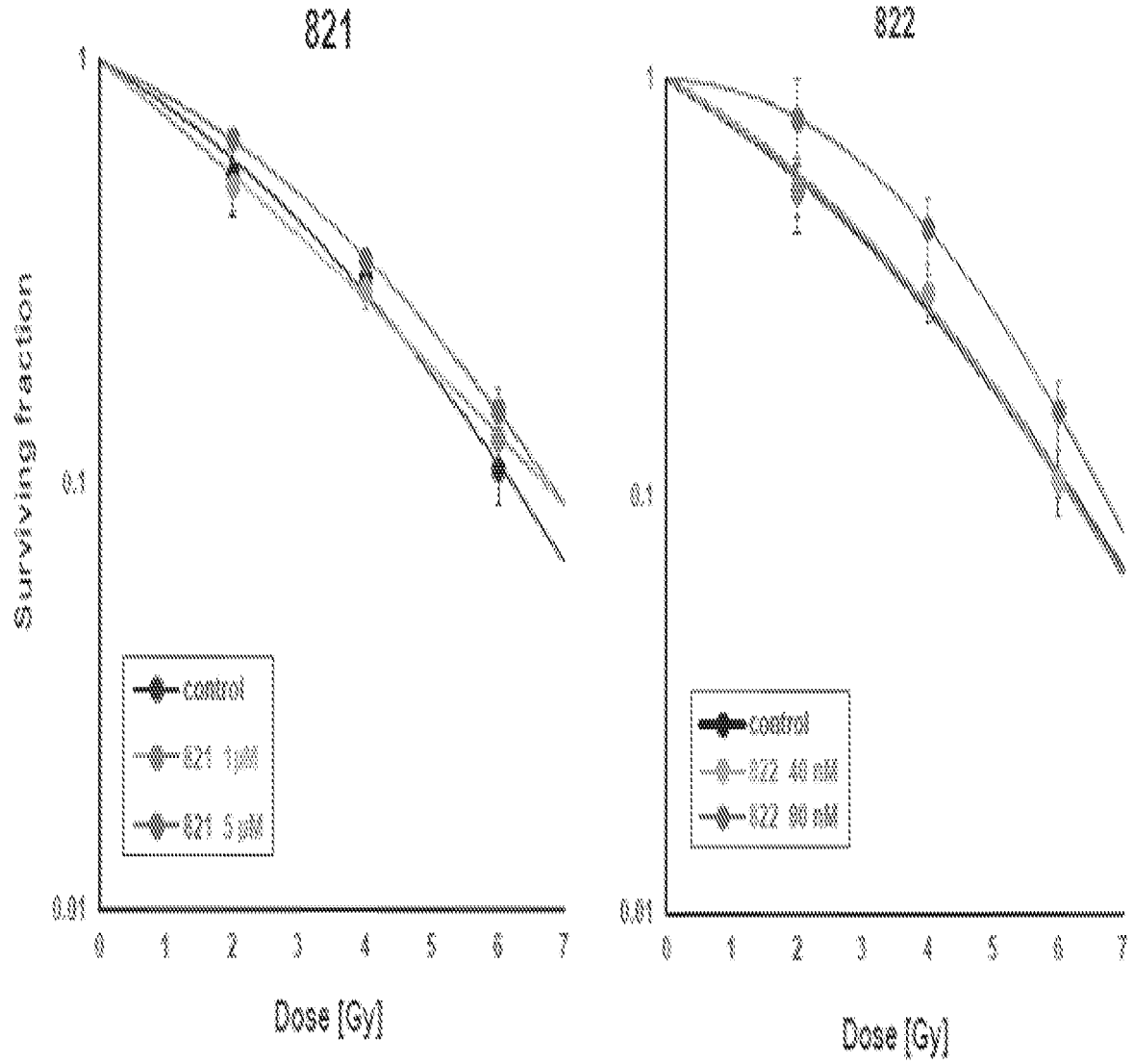


Figure 2X

B

MRC5

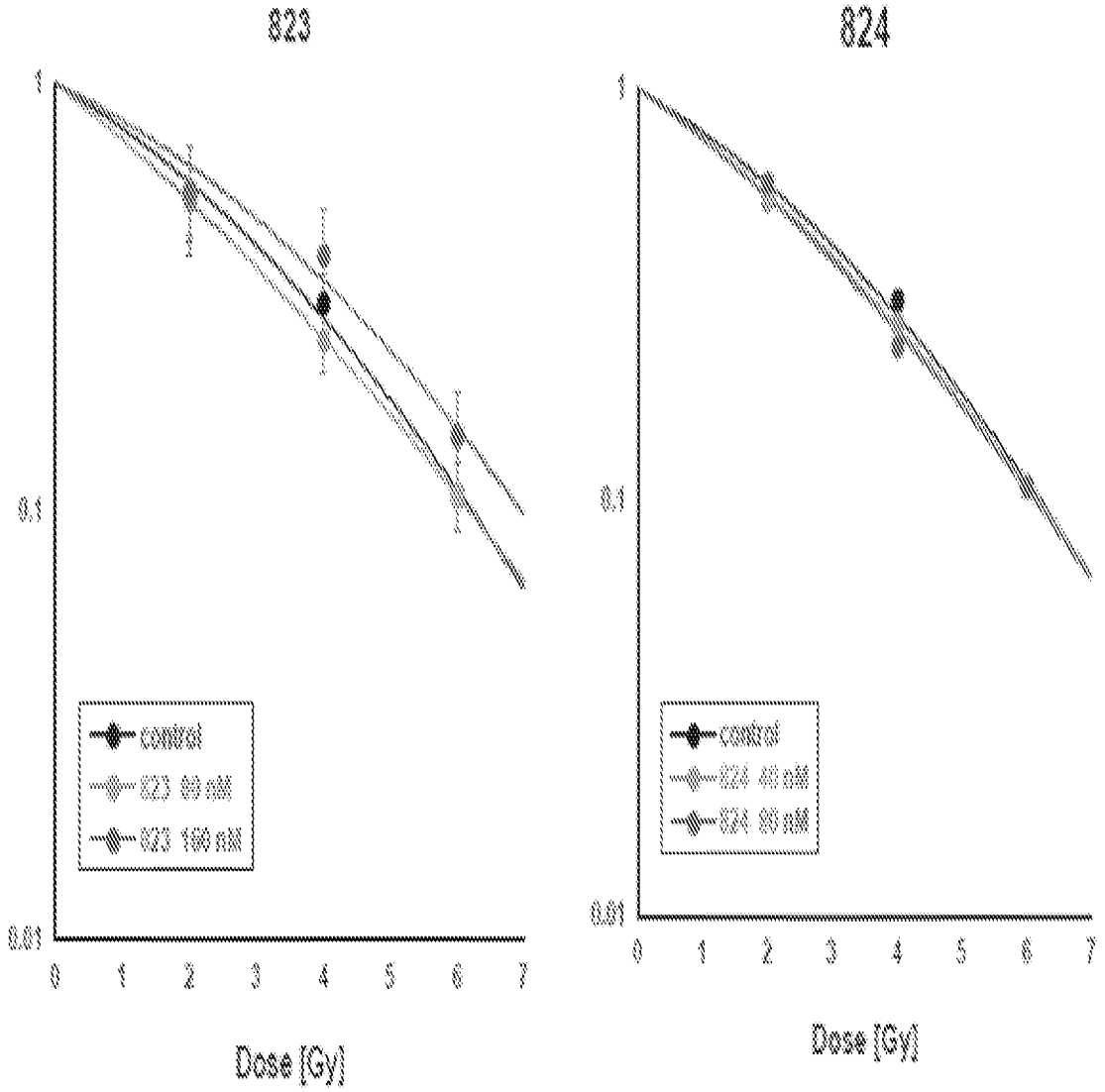


Figure 3X

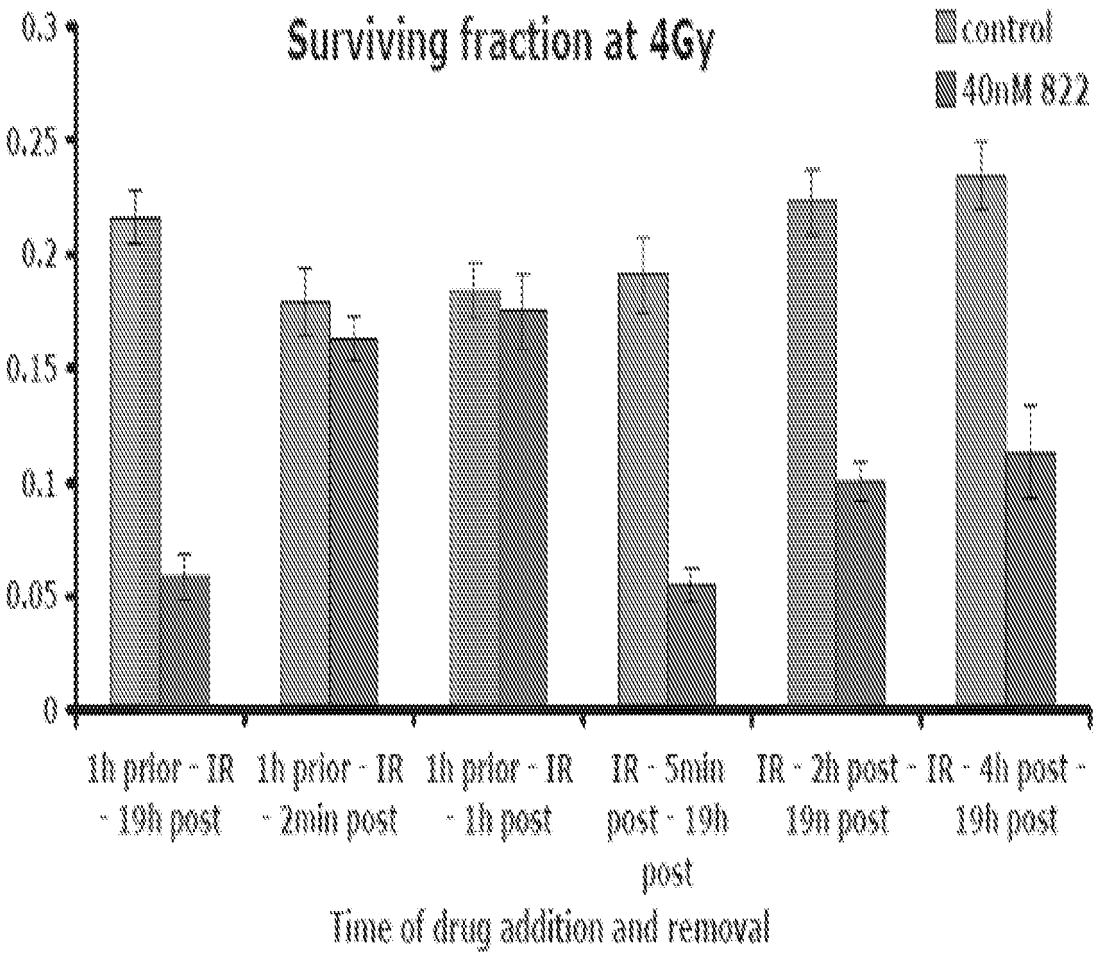

Surviving fraction at 4Gy
control
40nM 822
0.3
0.25
0.2
0.15
0.1
0.05
0
1h prior - IR - 19h post
1h prior - IR - 2min post
1h prior - IR - 1h post
IR - 5min post - 19h post
IR - 2h post - 19n post
IR - 4h post - 19h post
Time of drug addition and removal

Figure 3X

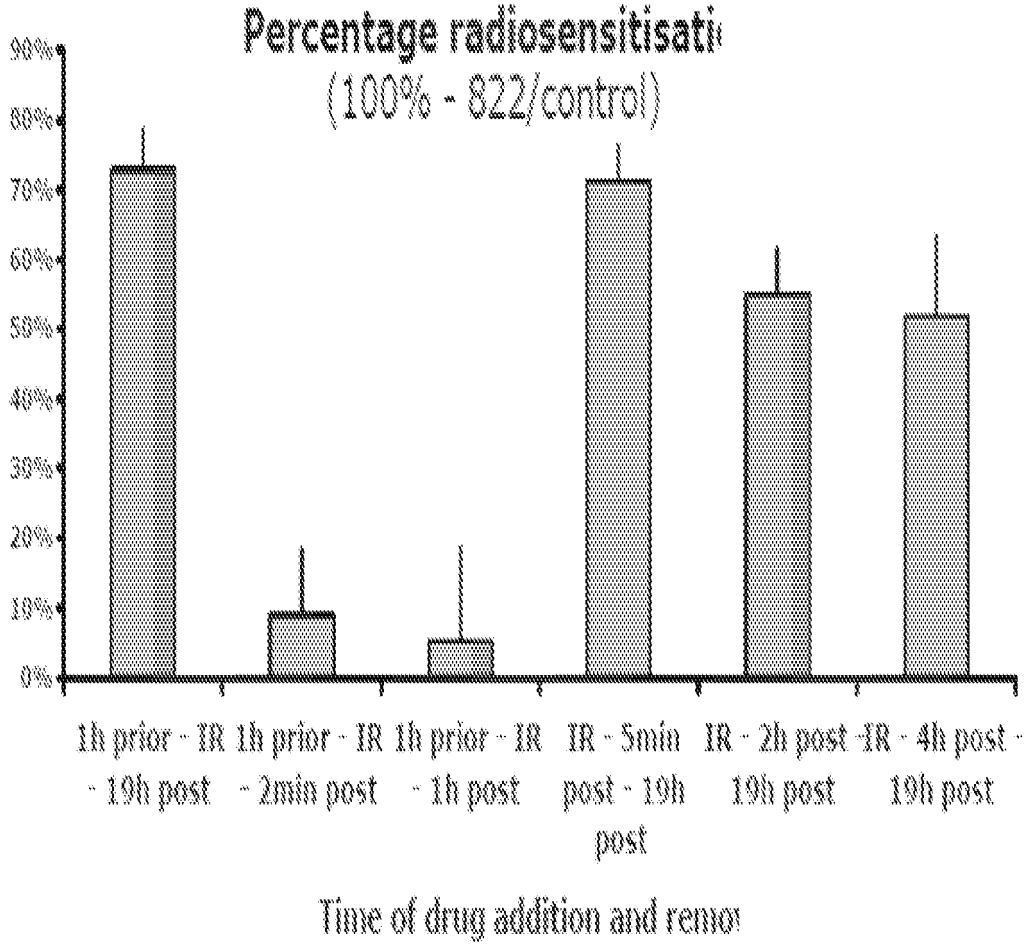

Percentage radiosensitisati
(100% - 822/control)
90%
80%
70%
60%
50%
40%
30%
20%
10%
0%
1h prior - IR - 19h post
1h prior - IR - 2min post
1h prior - IR - 1h post
IR - 5min post - 19h post
IR - 2h post - 19h post
IR - 4h post - 19h post
Time of drug addition and remo

Figure 3X

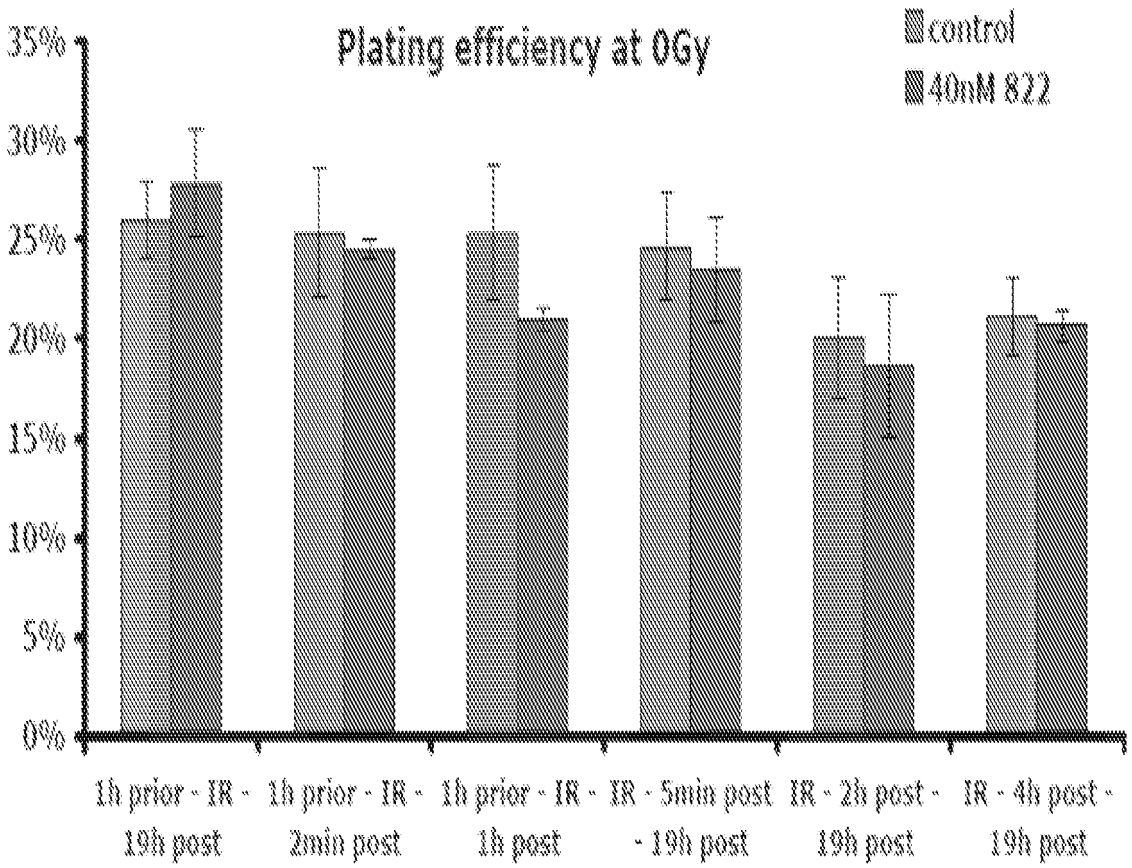

Plating efficiency at 0Gy
control
40nM 822
35%
30%
25%
20%
15%
10%
5%
0%
1h prior - IR - 19h post
1h prior - IR - 2min post
1h prior - IR - 1h post
IR - 5min post - 19h post
IR - 2h post - 19h post
IR - 4h post - 19h post

Figure 4X

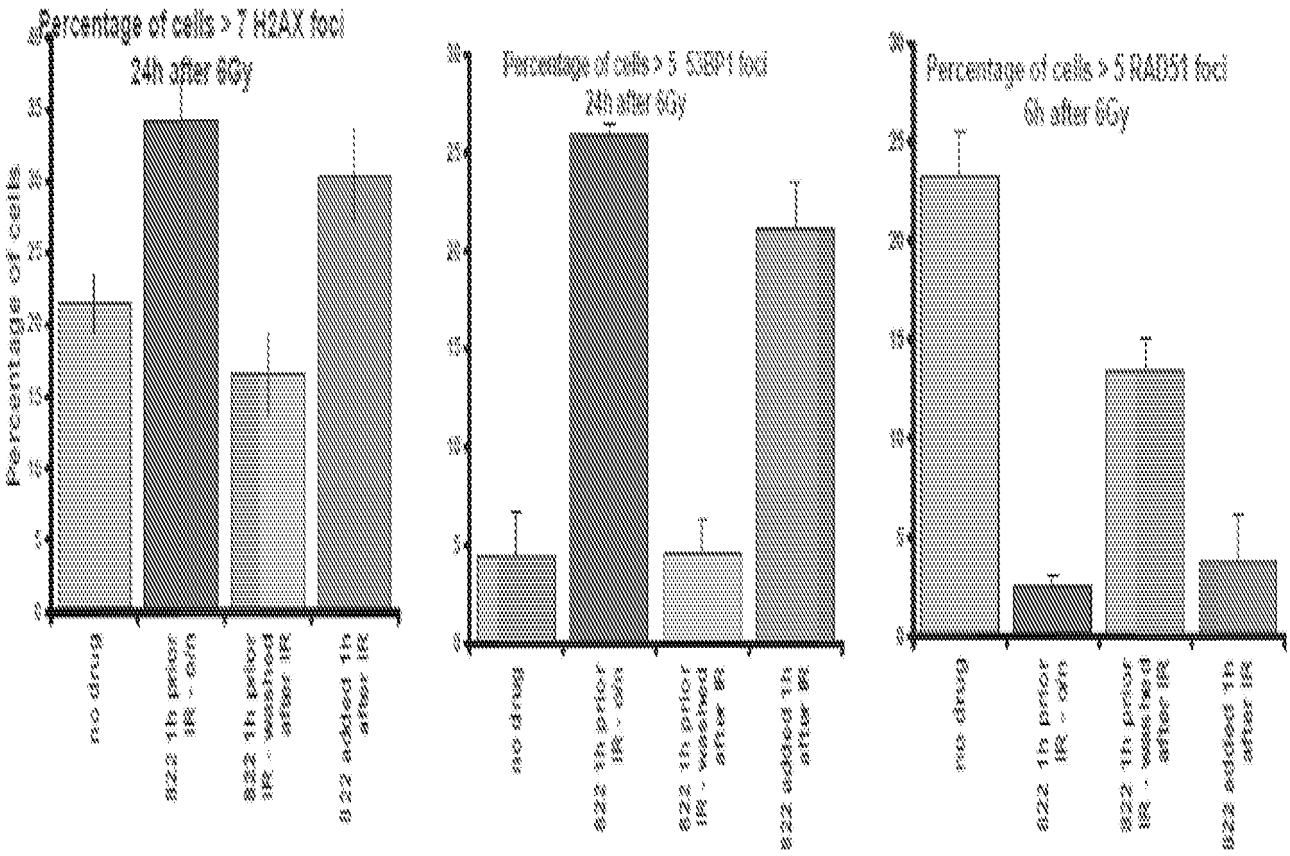

Percentage of cells > 7 H2AX foci
24h after 6Gy
Percentage of cells
Percentage of cells > 5 53BP1 foci
24h after 6Gy
Percentage of cells > 5 RAD51 foci
6h after 6Gy

Figure 4X

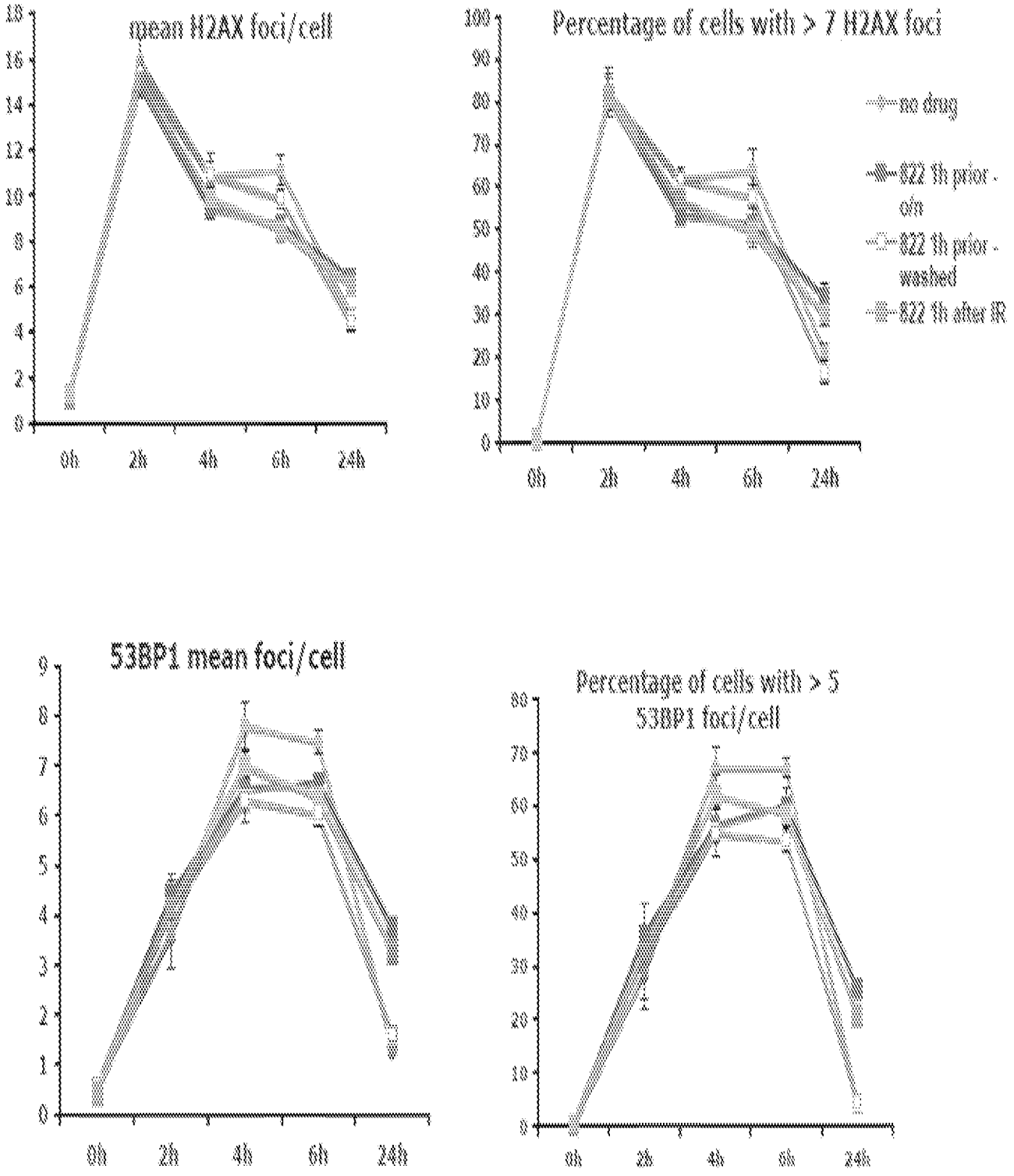
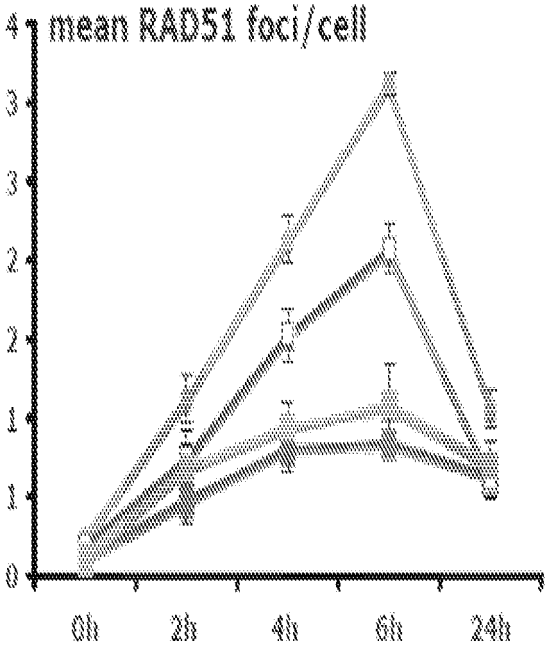

mean H2AX foci/cell
Percentage of cells with > 7 H2AX foci
no drug
822 1h prior - o/n
822 1h prior - washed
822 1h after IR
53BP1 mean foci/cell
Percentage of cells with > 5 53BP1 foci/cell
0h
2h
4h
6h
24h

Figure 4X


mean RAD51 foci/cell
4
3
3
2
2
1
1
0
0h
2h
4h
6h
24h

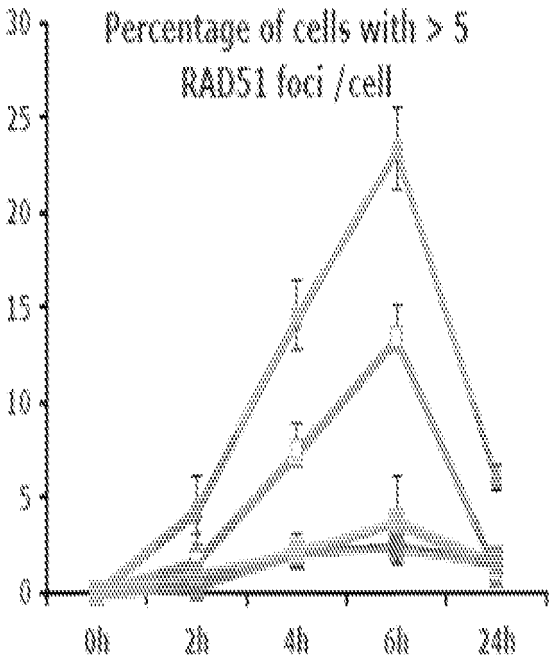

Percentage of cells with > 5
RAD51 foci /cell
30
25
20
15
10
5
0
0h
2h
4h
6h
24h

Figure 5X

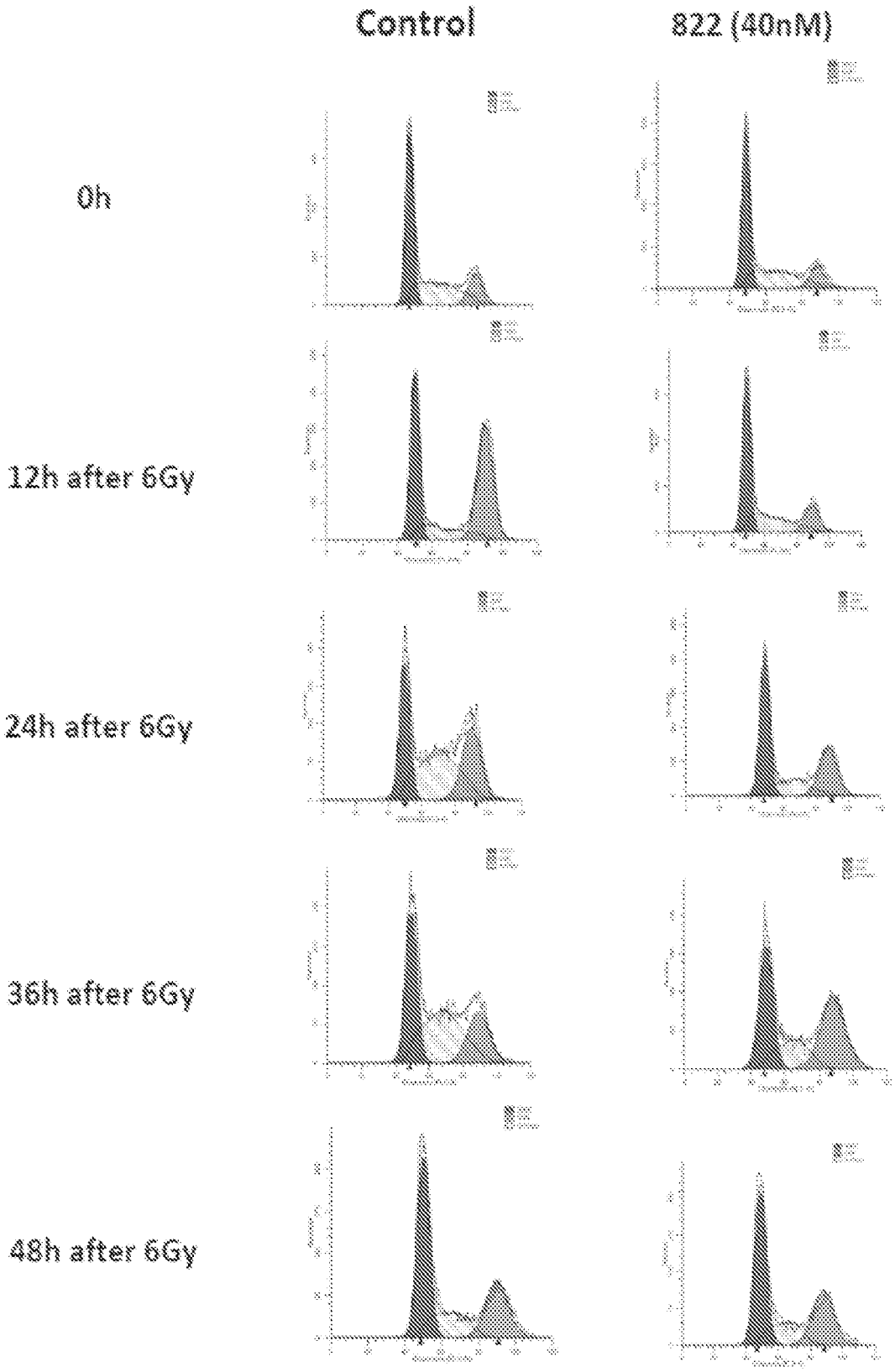

Control
822 (40nM)
0h
12h after 6Gy
24h after 6Gy
36h after 6Gy
48h after 6Gy

Figure 5X

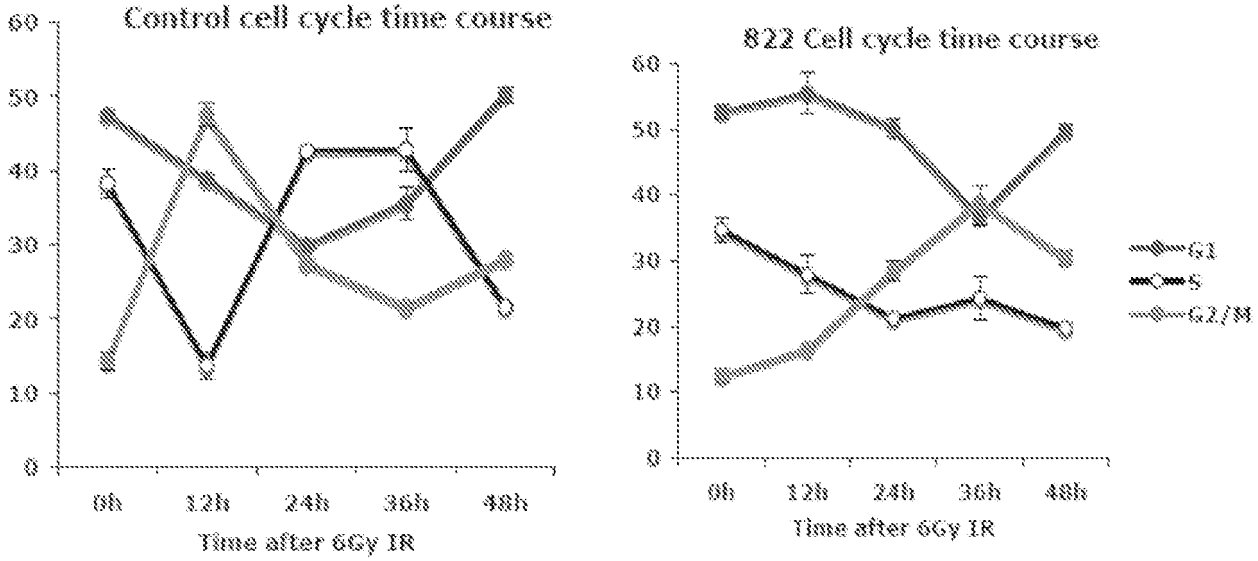

Control cell cycle time course
60
50
40
30
20
10
0
0h
12h
24h
36h
48h
Time after 6Gy IR
822 Cell cycle time course
60
50
40
30
20
10
0
0h
12h
24h
36h
48h
Time after 6Gy IR
G1
S
G2/M

Figure 1Y

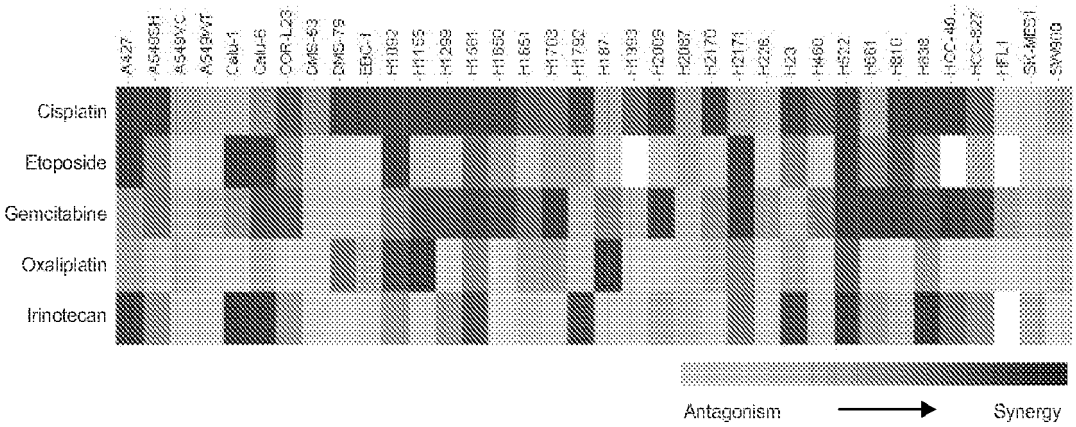


Figure 2Y

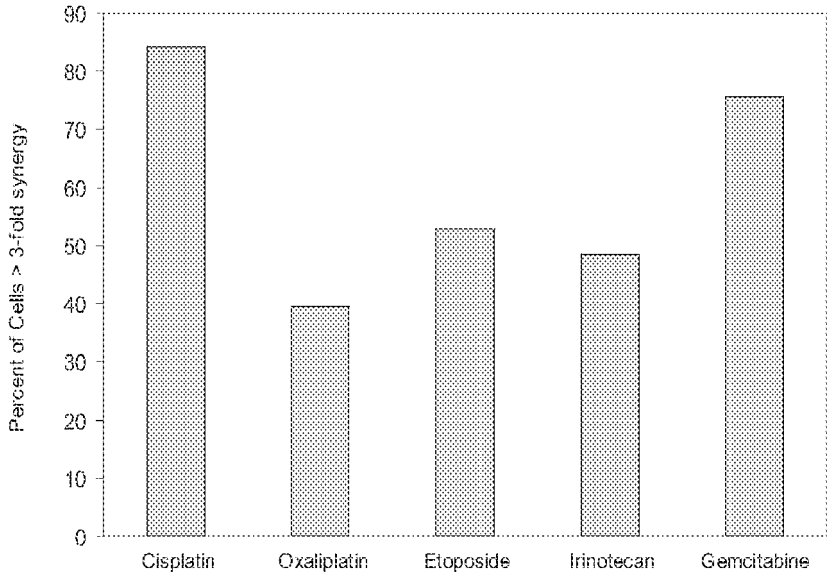


Figure 3Y

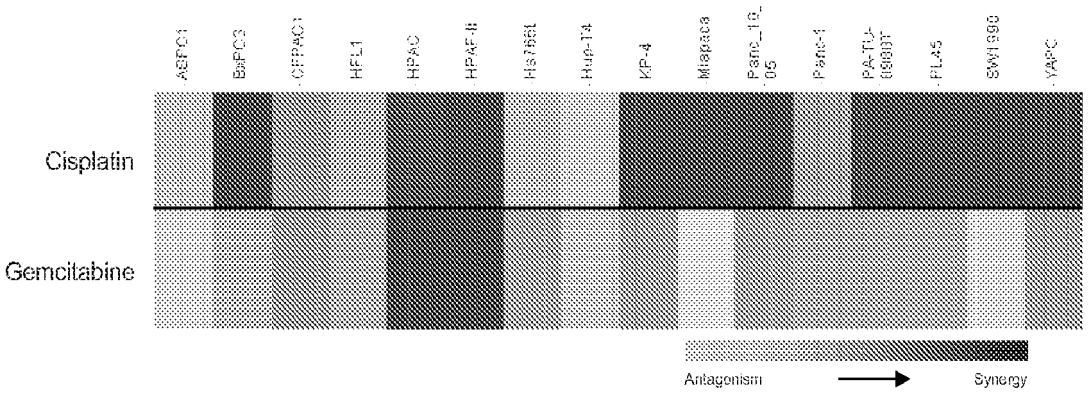


Figure 4Y

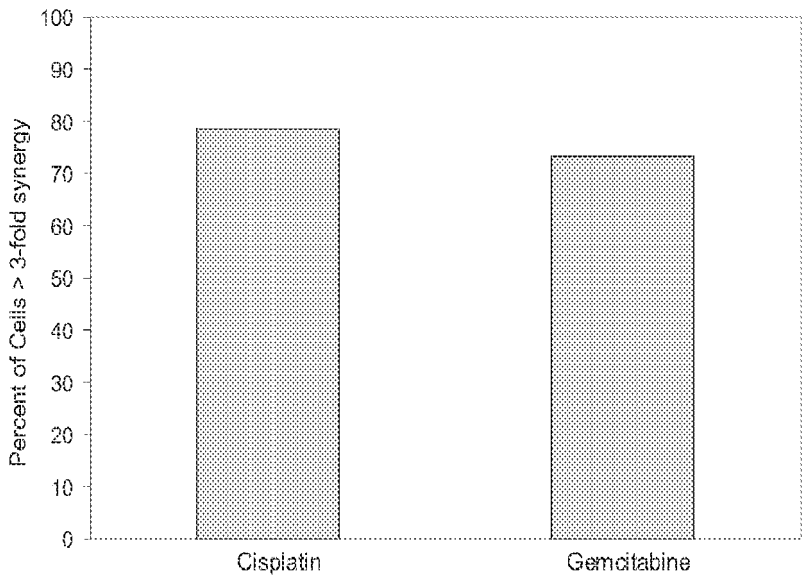


Fig 5Y

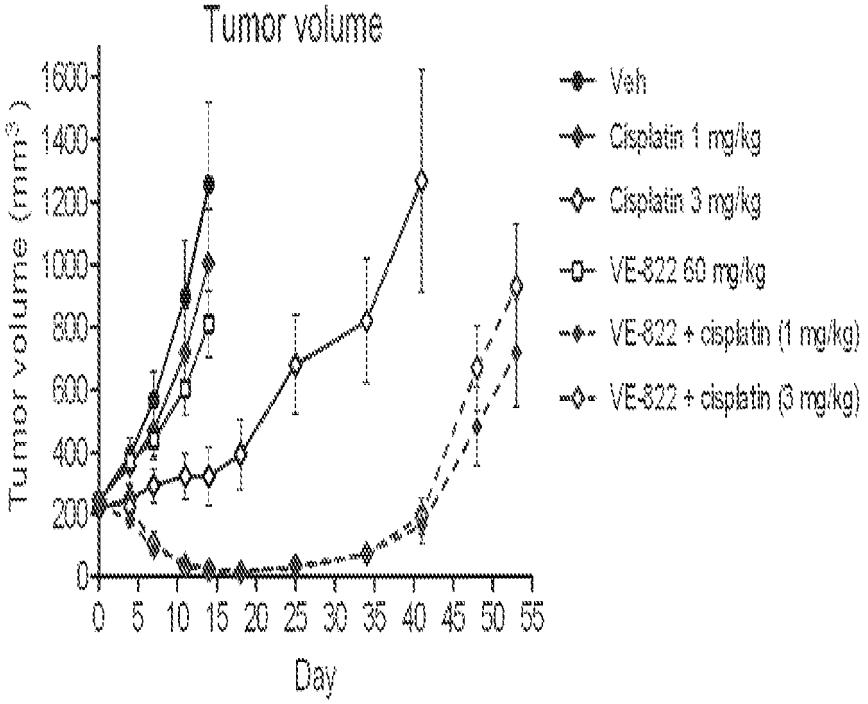
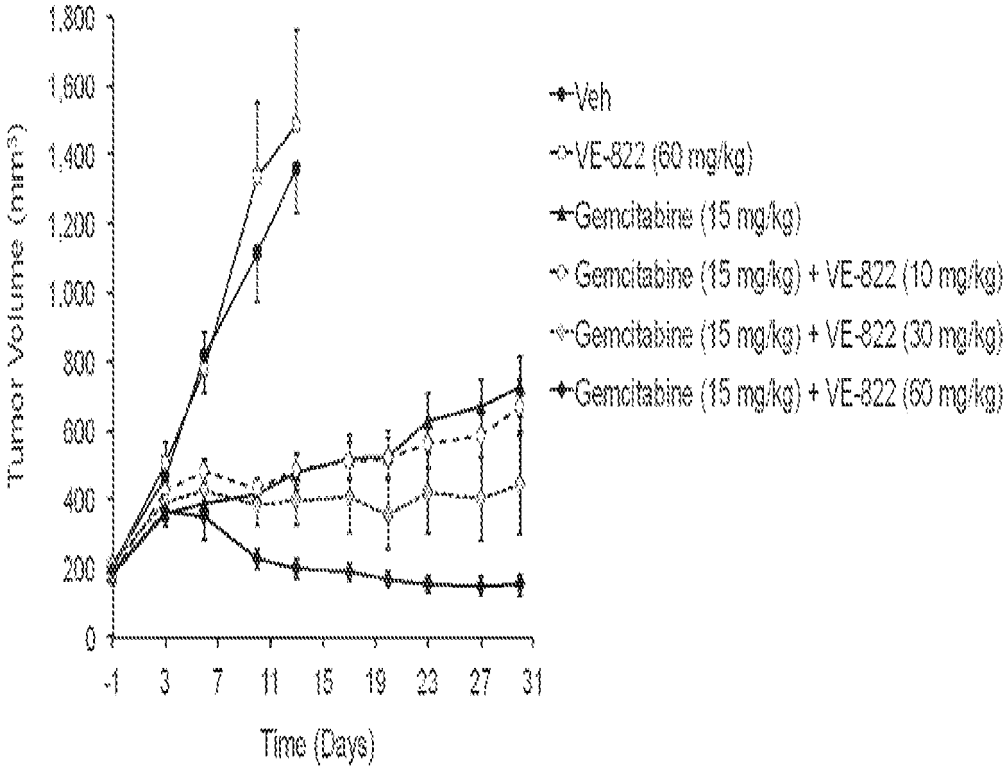

Tumor volume
Tumor volume (mm³)
1600
1400
1200
1000
800
600
400
200
0
0 5 10 15 20 25 30 35 40 45 50 55
Day
Veh
Cisplatin 1 mg/kg
Cisplatin 3 mg/kg
VE-822 60 mg/kg
VE-822 + cisplatin (1 mg/kg)
VE-822 + cisplatin (3 mg/kg)

Fig 6Y


Tumor Volume (mm³)
1,800
1,600
1,400
1,200
1,000
800
600
400
200
0
-1 3 7 11 15 19 23 27 31
Time (Days)
Veh
VE-822 (60 mg/kg)
Gemcitabine (15 mg/kg)
Gemcitabine (15 mg/kg) + VE-822 (10 mg/kg)
Gemcitabine (15 mg/kg) + VE-822 (30 mg/kg)
Gemcitabine (15 mg/kg) + VE-822 (60 mg/kg)