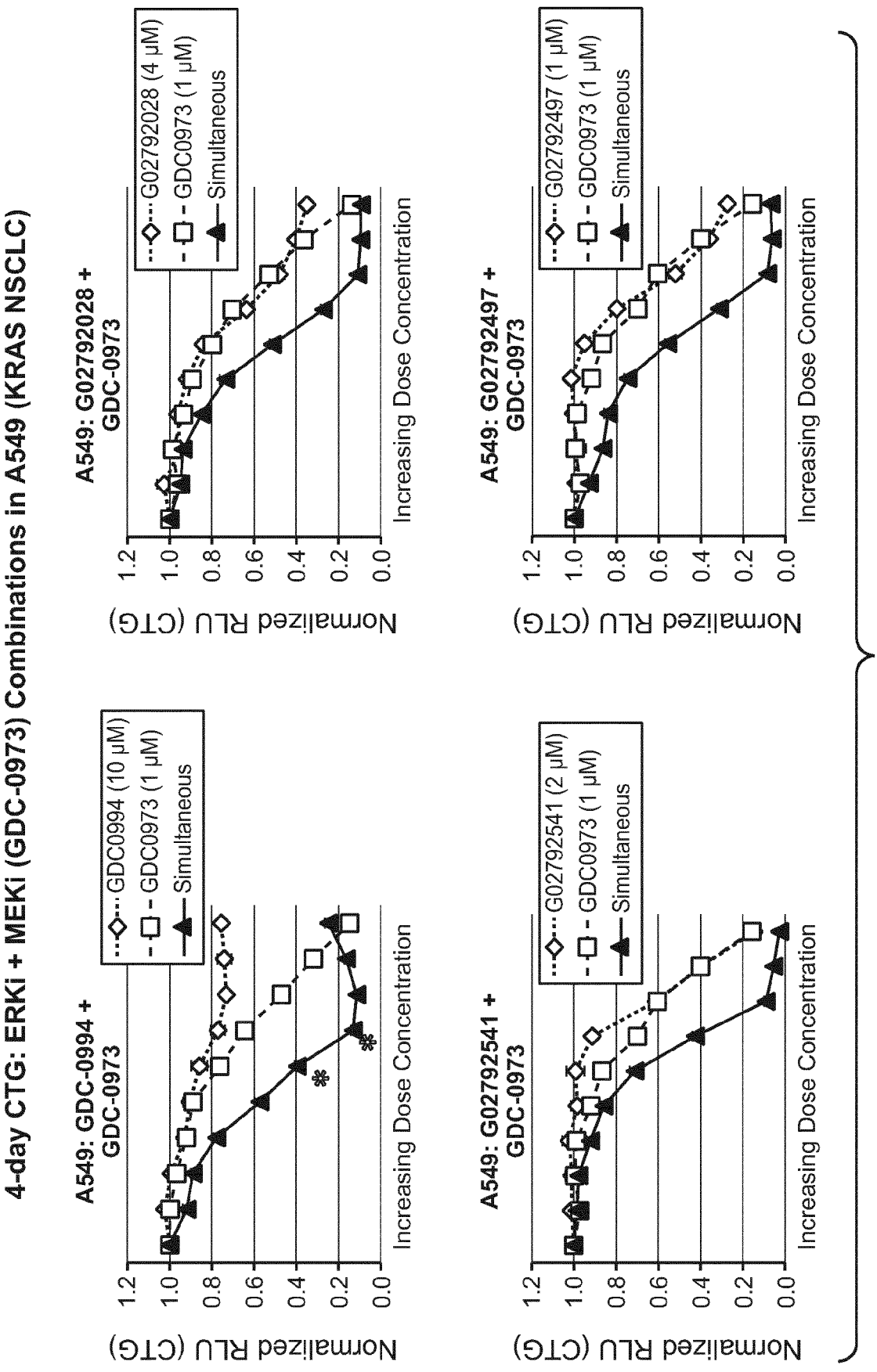

4-day CTG: ERKi + MEKi (GDC-0973) Combinations in A549 (KRAS NSCLC)
A549: GDC-0994 + GDC-0973
A549: G02792028 + GDC-0973
A549: G02792541 + GDC-0973
A549: G02792497 + GDC-0973
GDC0994 (10 µM) - GDC0973 (1 µM) Simultaneous
G02792028 (4 µM) - GDC0973 (1 µM) Simultaneous
G02792541 (2 µM) - GDC0973 (1 µM) Simultaneous
G02792497 (1 µM) - GDC0973 (1 µM) Simultaneous
Normalized RLU (CTG)
Increasing Dose Concentration
0.0
0.2
0.4
0.6
0.8
1.0
1.2

FIG. 1

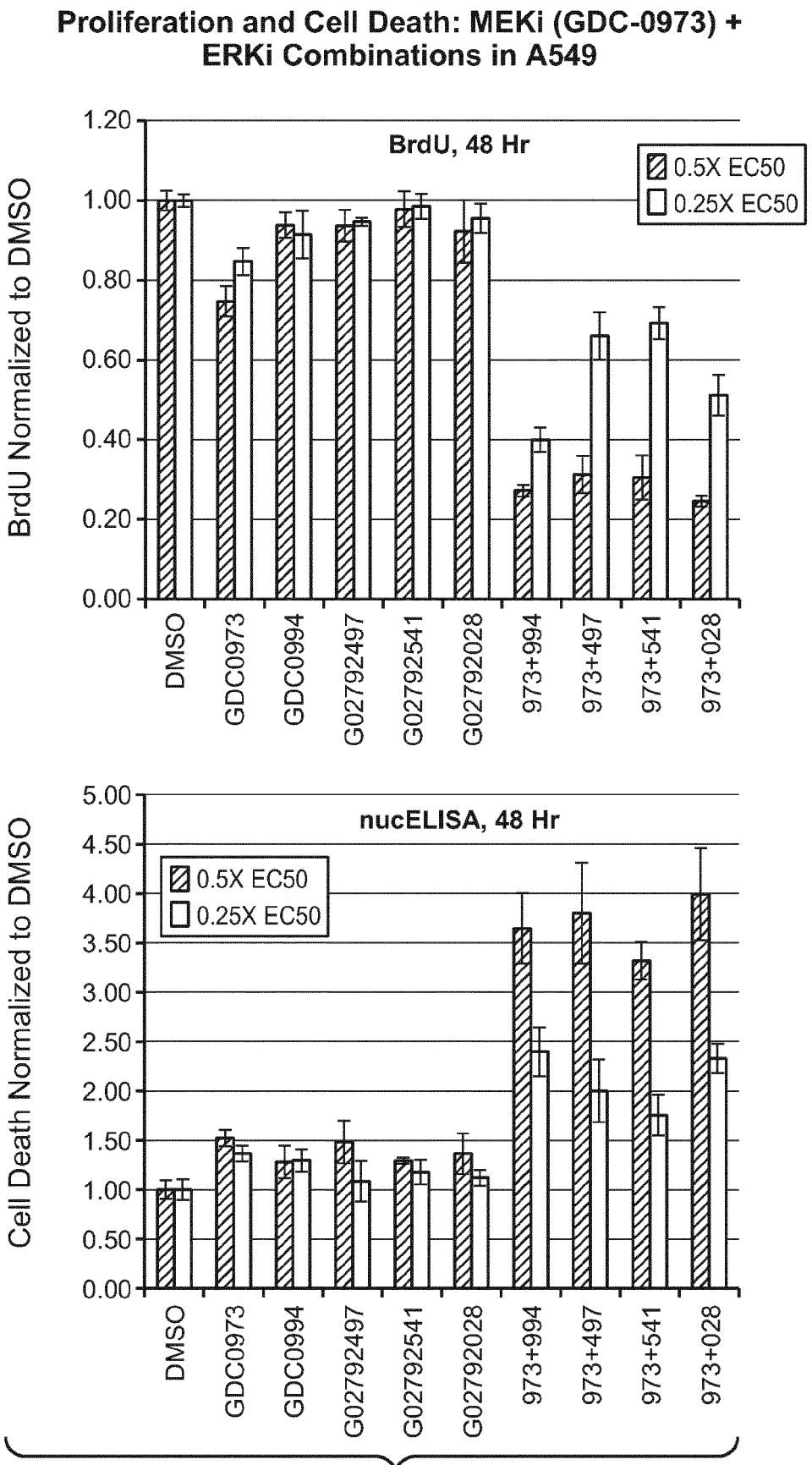

Proliferation and Cell Death: MEKi (GDC-0973) + ERKi Combinations in A549
BrdU, 48 Hr
BrdU Normalized to DMSO
0.5X EC50
0.25X EC50
1.20
1.00
0.80
0.60
0.40
0.20
0.00
DMSO
GDC0973
GDC0994
G02792497
G02792541
G02792028
973+994
973+497
973+541
973+028
nucELISA, 48 Hr
Cell Death Normalized to DMSO
5.00
4.50
4.00
3.50
3.00
2.50
2.00
1.50
1.00
0.50
0.00

FIG. 2A

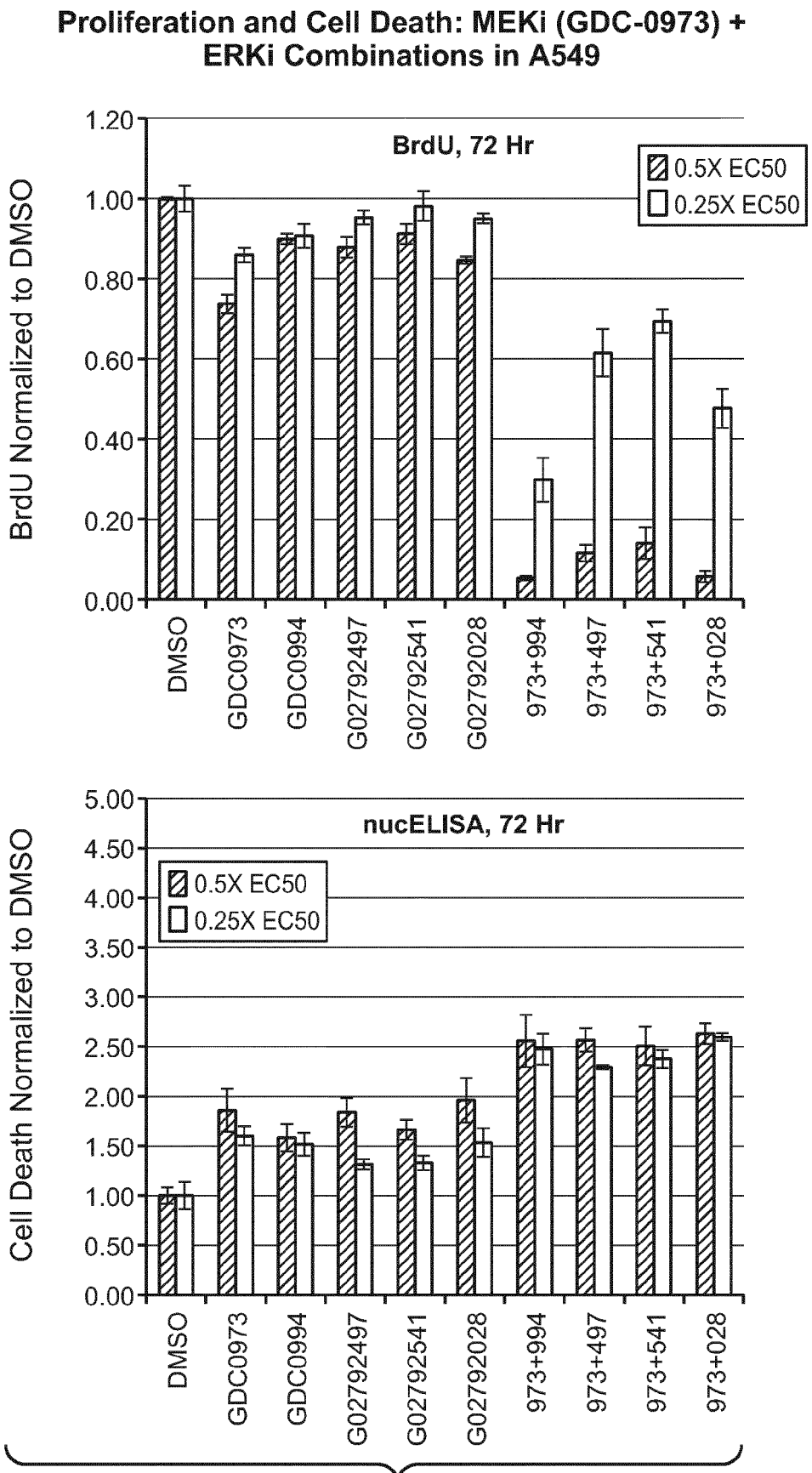

Proliferation and Cell Death: MEKi (GDC-0973) + ERKi Combinations in A549
BrdU, 72 Hr
0.5X EC50
0.25X EC50
BrdU Normalized to DMSO
1.20
1.00
0.80
0.60
0.40
0.20
0.00
DMSO
GDC0973
GDC0994
G02792497
G02792541
G02792028
973+994
973+497
973+541
973+028
nucELISA, 72 Hr
0.5X EC50
0.25X EC50
Cell Death Normalized to DMSO
5.00
4.50
4.00
3.50
3.00
2.50
2.00
1.50
1.00
0.50
0.00
DMSO
GDC0973
GDC0994
G02792497
G02792541
G02792028
973+994
973+497
973+541
973+028

FIG. 2B

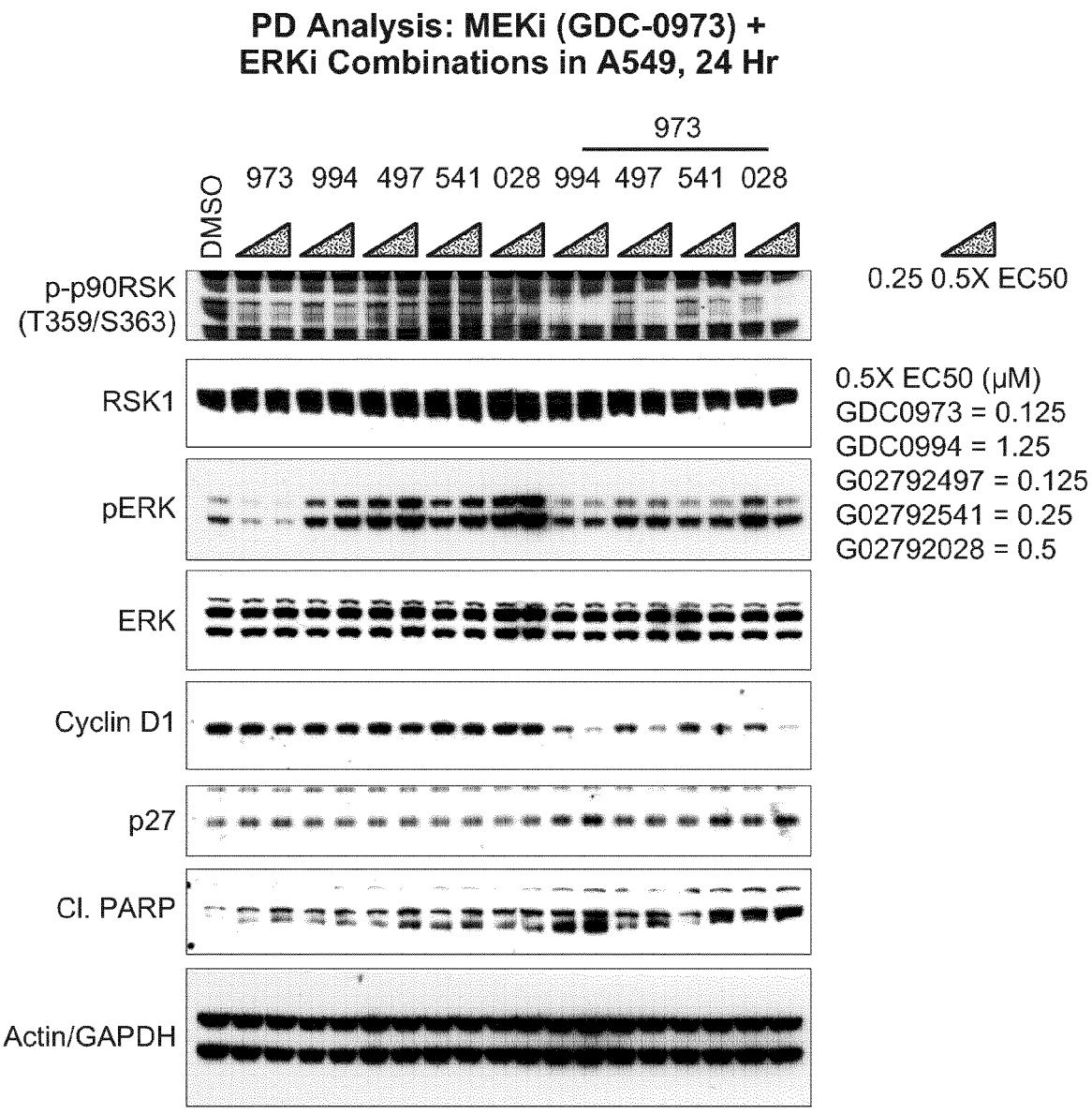

PD Analysis: MEKi (GDC-0973) +
ERKi Combinations in A549, 24 Hr
973
DMSO
973 994 497 541 028 994 497 541 028
0.25 0.5X EC50
p-p90RSK
(T359/S363)
RSK1
pERK
ERK
Cyclin D1
p27
Cl. PARP
Actin/GAPDH
0.5X EC50 (μM)
GDC0973 = 0.125
GDC0994 = 1.25
G02792497 = 0.125
G02792541 = 0.25
G02792028 = 0.5

FIG. 3

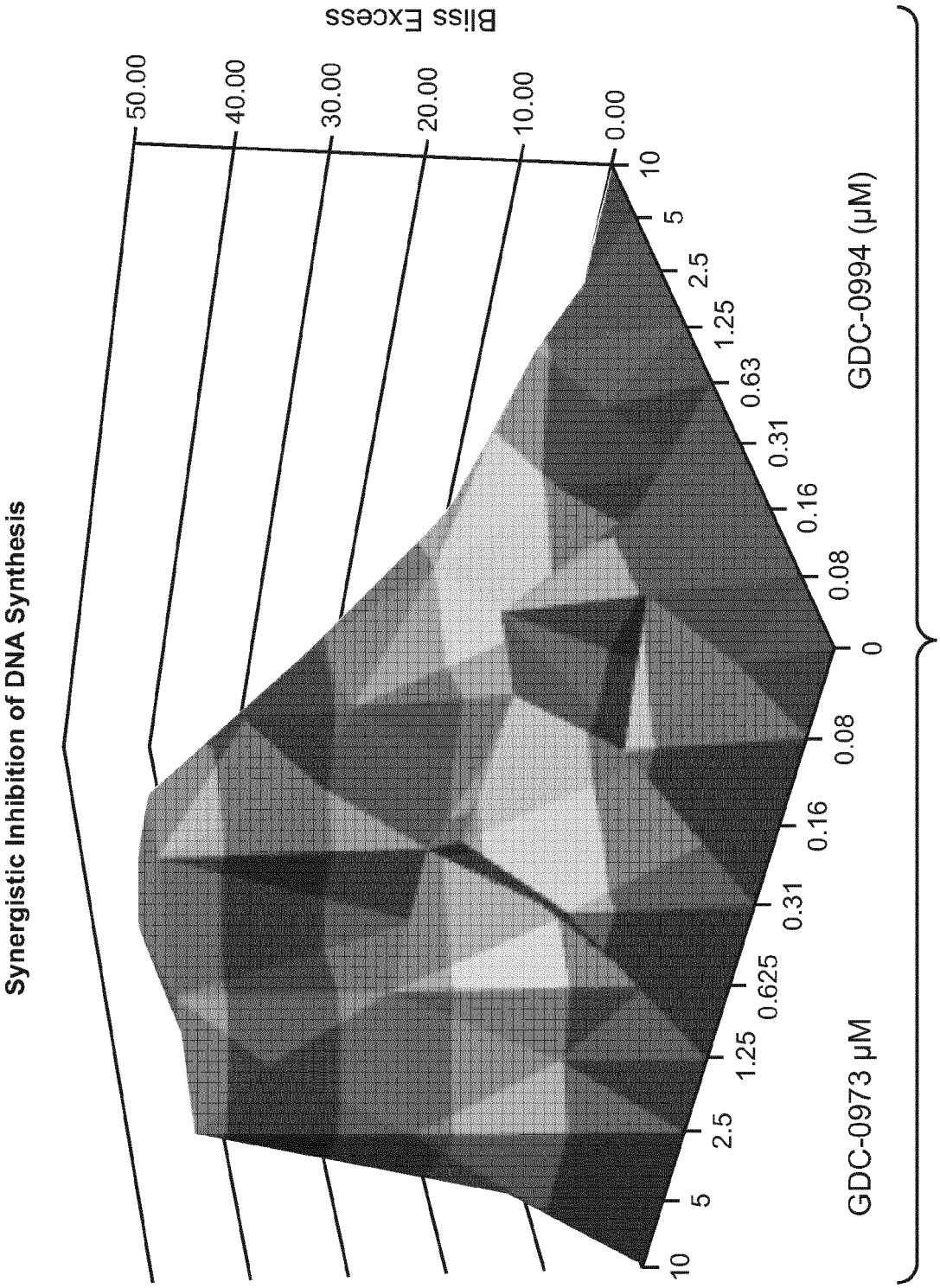

Synergistic Inhibition of DNA Synthesis
Bliss Excess
50.00
40.00
30.00
20.00
10.00
0.00
GDC-0973 µM
10
5
2.5
1.25
0.625
0.31
0.16
0.08
0
GDC-0994 (µM)
0.08
0.16
0.31
0.63
1.25
2.5
5
10

FIG. 4

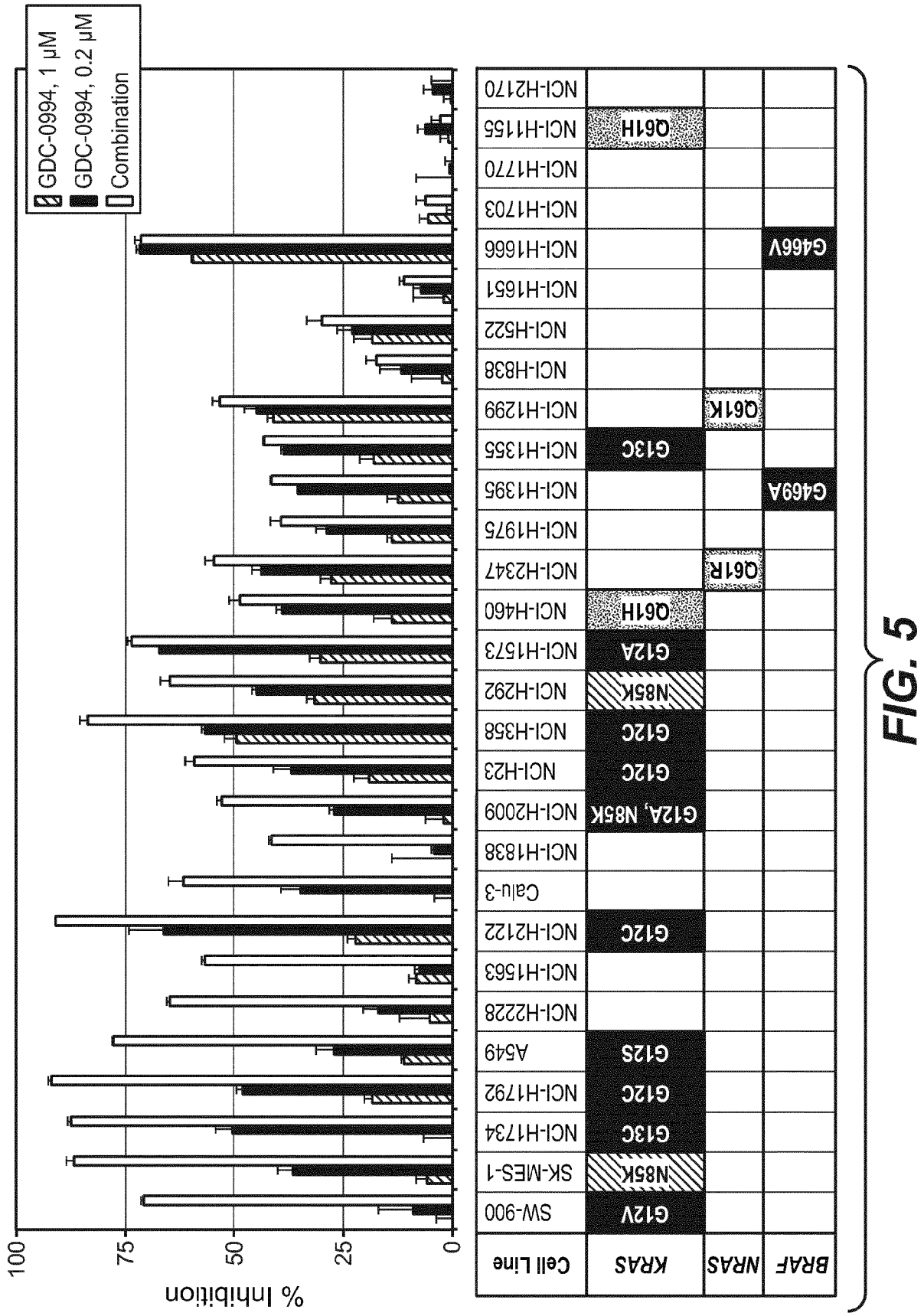

GDC-0994, 1 µM
GDC-0994, 0.2 µM
Combination
% Inhibition
0
25
50
75
100
Cell Line
KRAS
NRAS
BRAF
SW-900 G12V
SK-MES-1 N85K
NCI-H1734 G13C
NCI-H1792 G12C
A549 G12S
NCI-H2228
NCI-H1563
NCI-H2122 G12C
Calu-3
NCI-H1838
NCI-H2009 G12A, N85K
NCI-H23 G12C
NCI-H358 G12C
NCI-H292 N85K
NCI-H1573 G12A
NCI-H460 Q61H
NCI-H2347 Q61R
NCI-H1975
NCI-H1395 G469A
NCI-H1355 G13C
NCI-H1299 Q61K
NCI-H838
NCI-H522
NCI-H1651
NCI-H1666 G466V
NCI-H1703
NCI-H1770
NCI-H1155 Q61H
NCI-H2170

FIG. 5

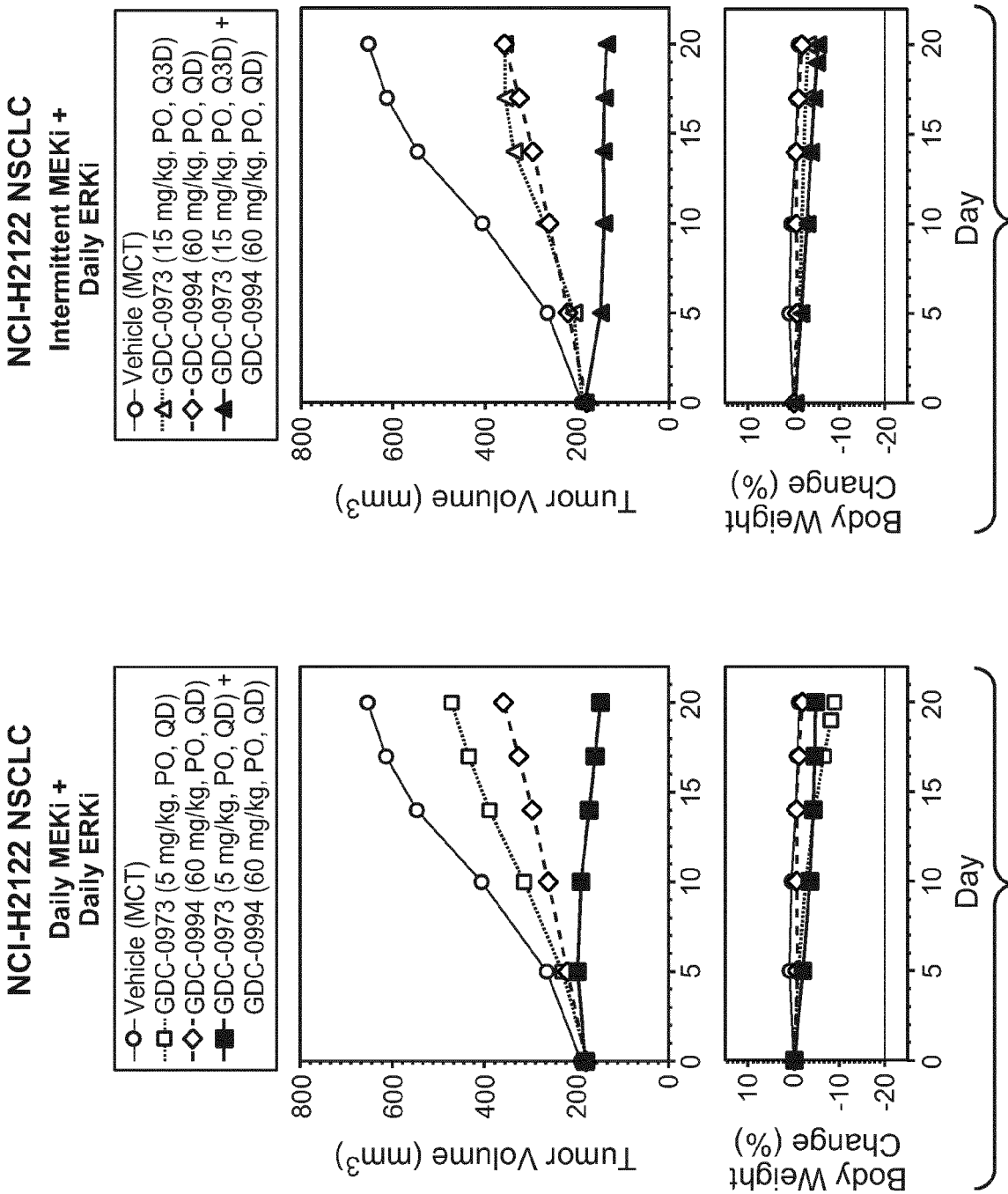

NCI-H2122 NSCLC
Daily MEKi +
Daily ERKi
Vehicle (MCT)
GDC-0973 (5 mg/kg, PO, QD)
GDC-0994 (60 mg/kg, PO, QD)
GDC-0973 (5 mg/kg, PO, QD) +
GDC-0994 (60 mg/kg, PO, QD)
Tumor Volume (mm3)
Body Weight
Change (%)
Day
NCI-H2122 NSCLC
Intermittent MEKi +
Daily ERKi
Vehicle (MCT)
GDC-0973 (15 mg/kg, PO, Q3D)
GDC-0994 (60 mg/kg, PO, QD)
GDC-0973 (15 mg/kg, PO, Q3D) +
GDC-0994 (60 mg/kg, PO, QD)
Tumor Volume (mm3)
Body Weight
Change (%)
Day

FIG. 6A

FIG. 6B

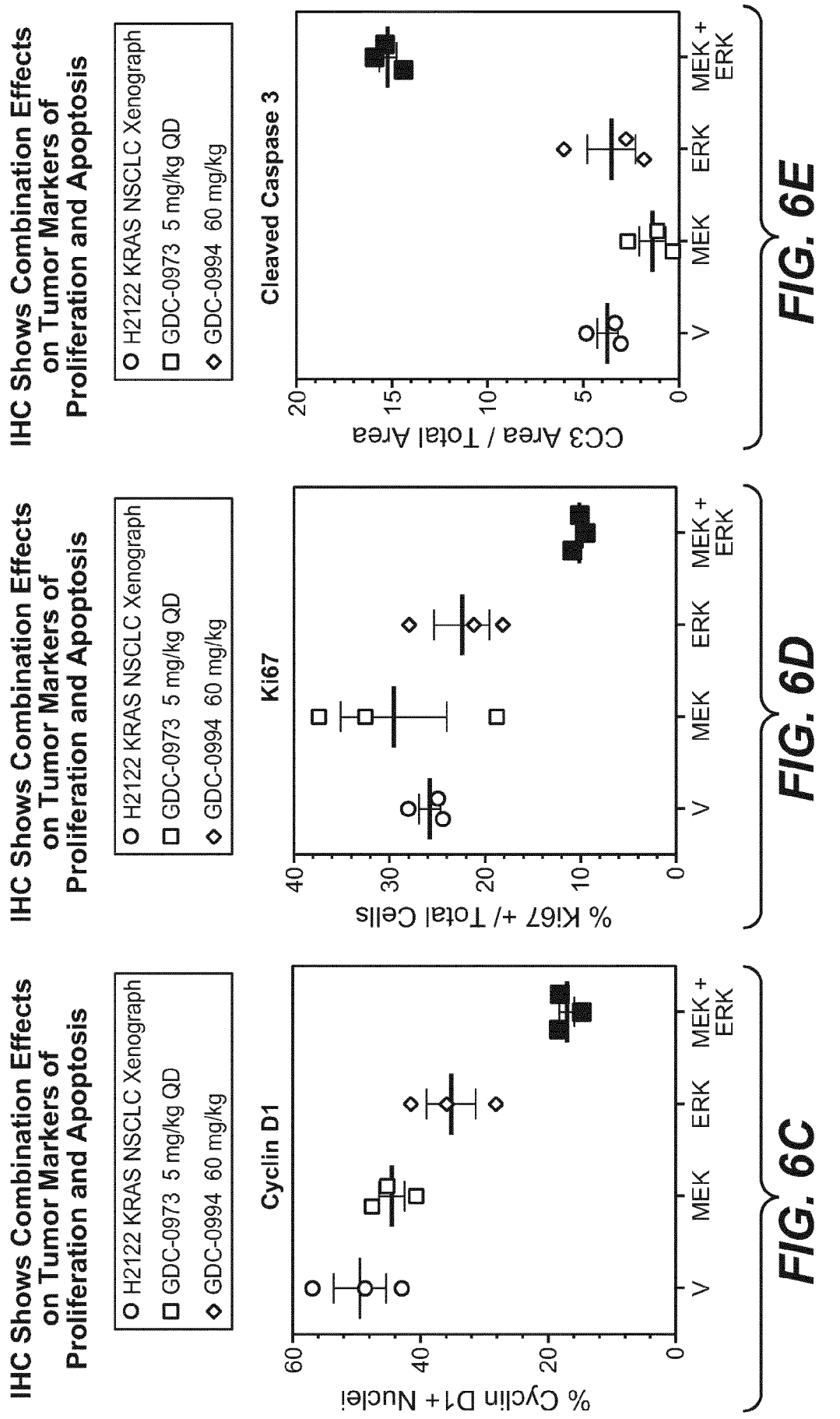
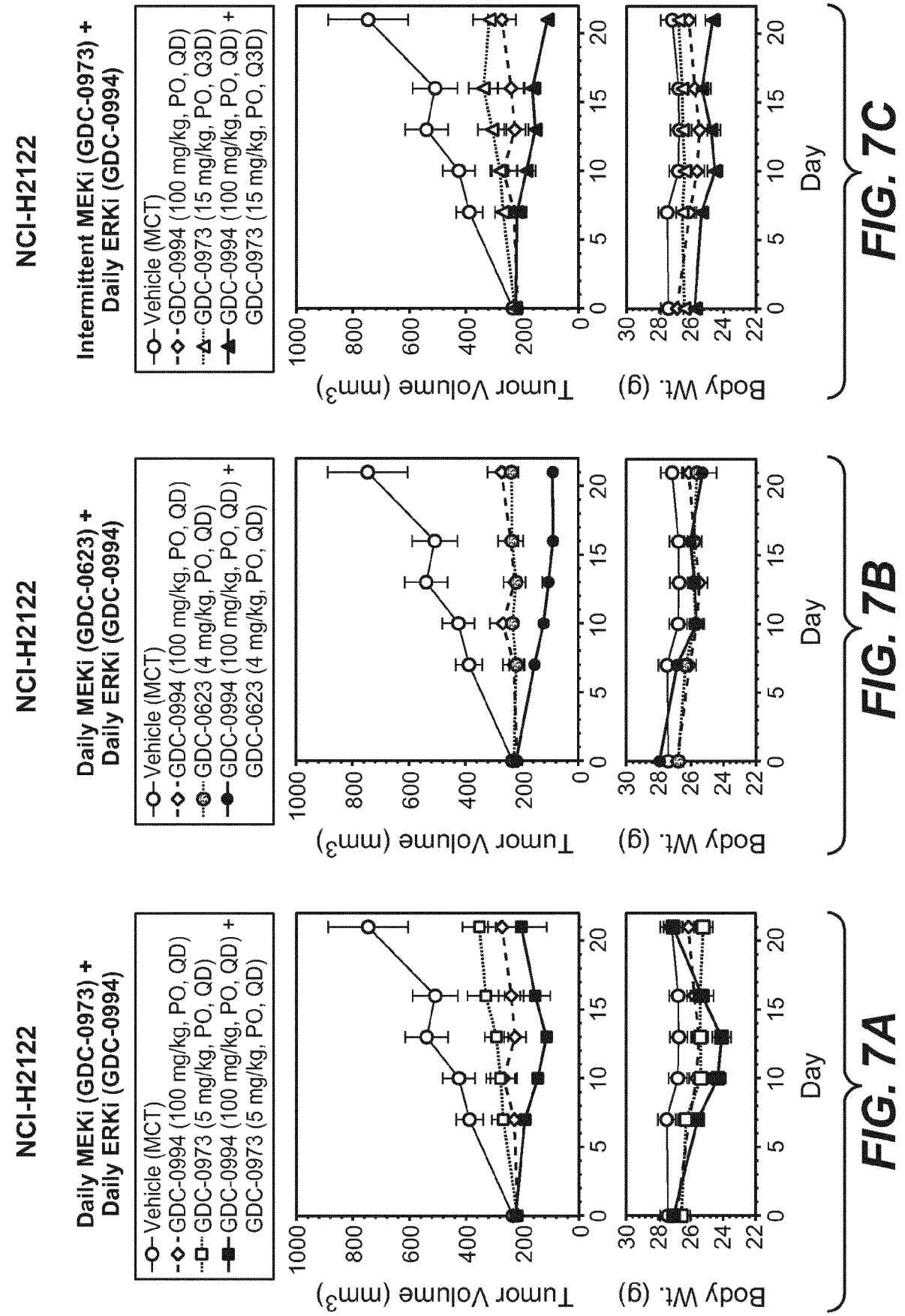


FIG. 6C

FIG. 6D

FIG. 6E


NCI-H2122
Daily MEKi (GDC-0973) + Daily ERKi (GDC-0994)
Vehicle (MCT)
GDC-0994 (100 mg/kg, PO, QD)
GDC-0973 (5 mg/kg, PO, QD)
GDC-0994 (100 mg/kg, PO, QD) + GDC-0973 (5 mg/kg, PO, QD)
Tumor Volume (mm3)
Body Wt. (g)
Day
FIG. 7A
NCI-H2122
Daily MEKi (GDC-0623) + Daily ERKi (GDC-0994)
Vehicle (MCT)
GDC-0994 (100 mg/kg, PO, QD)
GDC-0623 (4 mg/kg, PO, QD)
GDC-0994 (100 mg/kg, PO, QD) + GDC-0623 (4 mg/kg, PO, QD)
Tumor Volume (mm3)
Body Wt. (g)
Day
FIG. 7B
NCI-H2122
Intermittent MEKi (GDC-0973) + Daily ERKi (GDC-0994)
Vehicle (MCT)
GDC-0994 (100 mg/kg, PO, QD)
GDC-0973 (15 mg/kg, PO, Q3D)
GDC-0994 (100 mg/kg, PO, QD) + GDC-0973 (15 mg/kg, PO, Q3D)
Tumor Volume (mm3)
Body Wt. (g)
Day
FIG. 7C

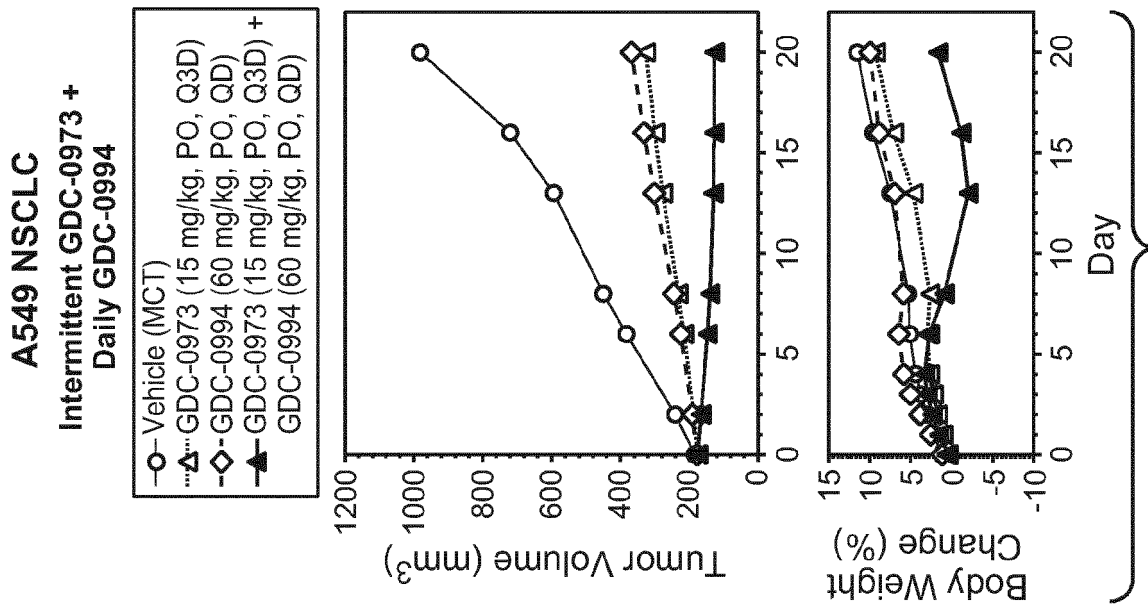

A549 NSCLC
Intermittent GDC-0973 +
Daily GDC-0994
Vehicle (MCT)
GDC-0973 (15 mg/kg, PO, Q3D)
GDC-0994 (60 mg/kg, PO, QD)
GDC-0973 (15 mg/kg, PO, Q3D) +
GDC-0994 (60 mg/kg, PO, QD)
Tumor Volume (mm3)
Body Weight
Change (%)
Day

FIG. 8B

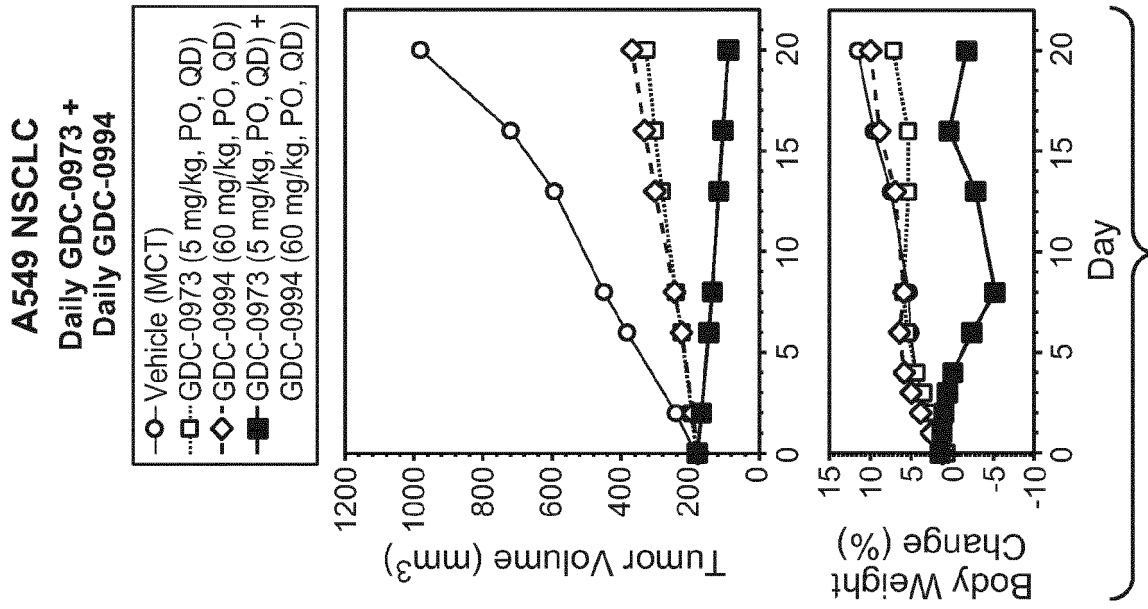

A549 NSCLC
Daily GDC-0973 +
Daily GDC-0994
Vehicle (MCT)
GDC-0973 (5 mg/kg, PO, QD)
GDC-0994 (60 mg/kg, PO, QD)
GDC-0973 (5 mg/kg, PO, QD) +
GDC-0994 (60 mg/kg, PO, QD)
Tumor Volume (mm3)
Body Weight
Change (%)
Day

FIG. 8A

Combination Efficacy at Tolerated Doses is Observed with GDC-0973 and GDC-0994 Combinations in HCT116 Cells.

%TGI (Lower, Upper CI)		GDC-0994 (mg/kg)						
		0	25	37.5	50	62.5	75	150
GDC-0973 (mg/kg)	0	0 (0,0)	38 (-49,80)	n.d.	79 (52,99)	n.d.	66 (22,87)	104 (90,119)
	1	48 (-5,77)	57 (11,81)	n.d.	73 (39,90)	n.d.	74 (42,95)	n.d.
	2	n.d.	n.d.	81 (53,97)	n.d.	93 (72,110)	0 (0,0)	n.d.
	3	41 (-18,72)	73 (43,91)	n.d.	68 (35,89)	n.d.	97 (79,113)	n.d.
	5	62 (19,86)	85 (61,102)	80 (55,98)	n.d.	98 (80,113)	112 (100,131)	n.d.
	7.5	72 (39,92)	100 (86,116)	n.d.	111 (99,126)	n.d.	117 (106,138)	n.d.
	10	87 (68,101)	n.d.	n.d.	n.d.	n.d.	n.d.	n.d.

- Grey cells indicate the best combination efficacy for a tolerated regimen
- Black cells indicates non-tolerated combinations
- Dosing was daily (QD) oral gavage (PO)
- Numbers represent % Tumor Growth Inhibition (%TGI) relative to vehicle (0,0) +/- 95th percent confidence intervals (CI).

n.d. = not determined

FIG. 9

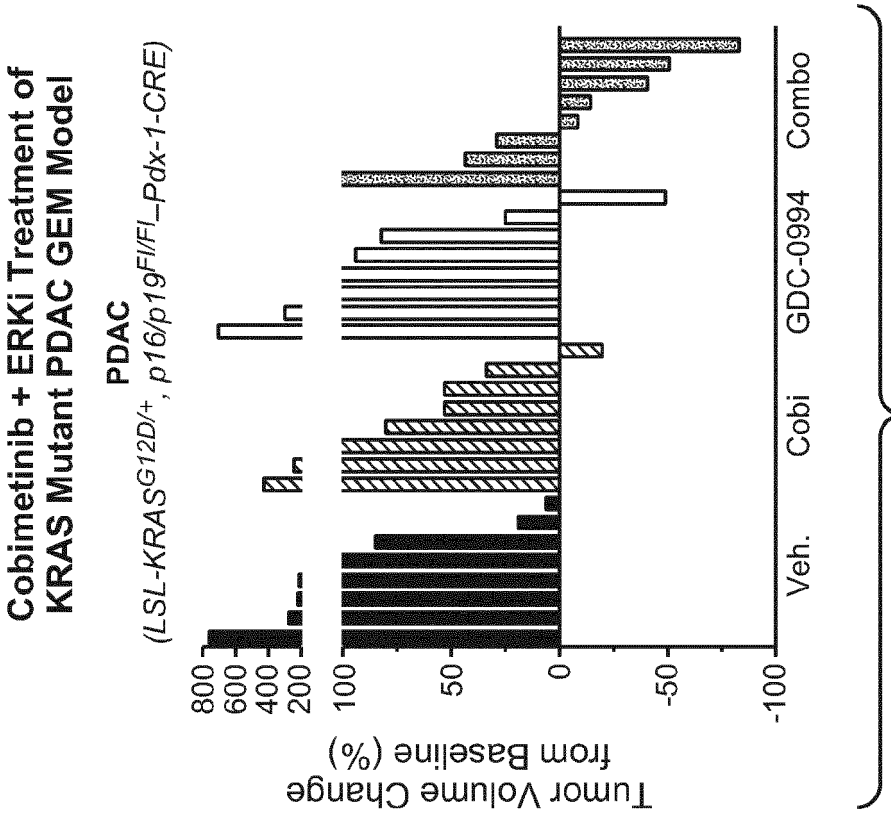
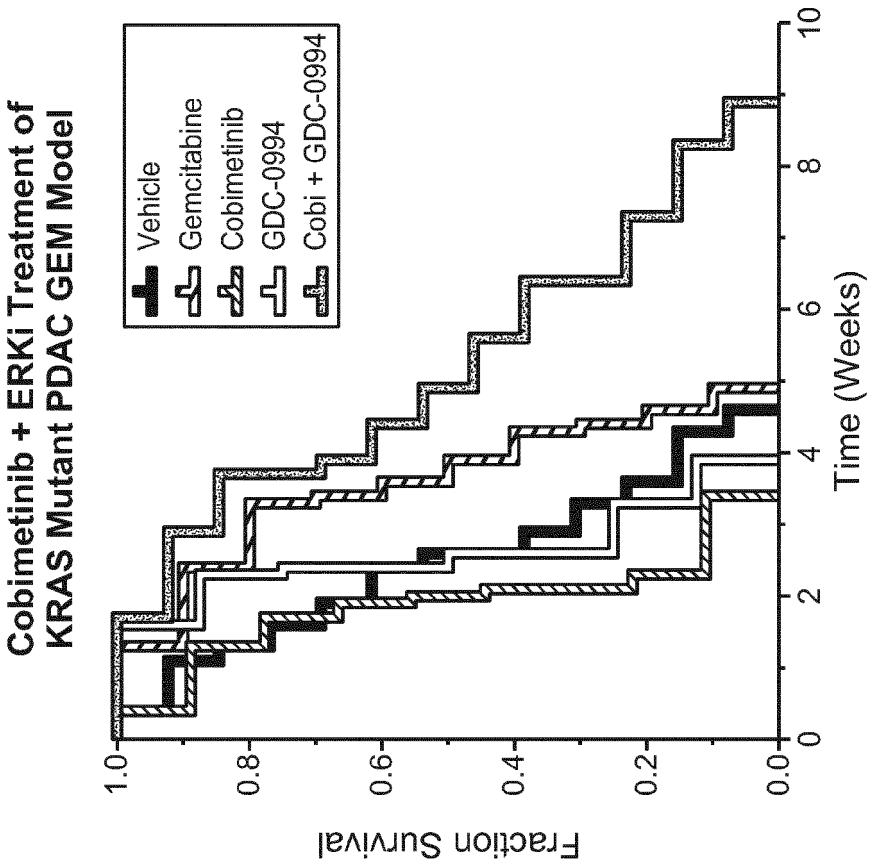


FIG. 10A



Treatment	Median Survival
Vehicle (n = 13)	2.6
MEKi (Cobimetinib) (n = 9)	2
ERKi (GDC-0994) (n = 8)	2.5
MEKi + ERKi (n = 13)	4.9
Gemcitabine (n = 10)	3.7

FIG. 10B

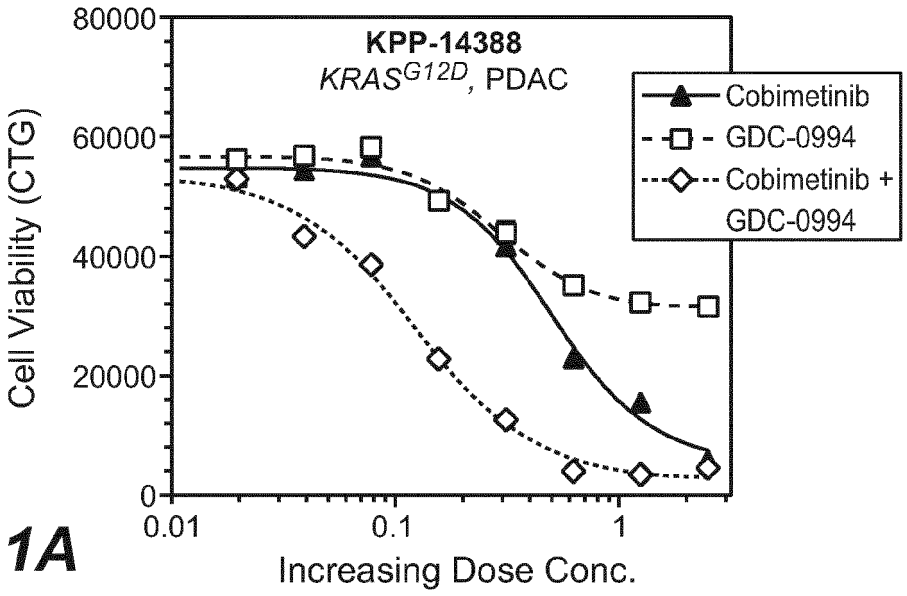


FIG. 11A

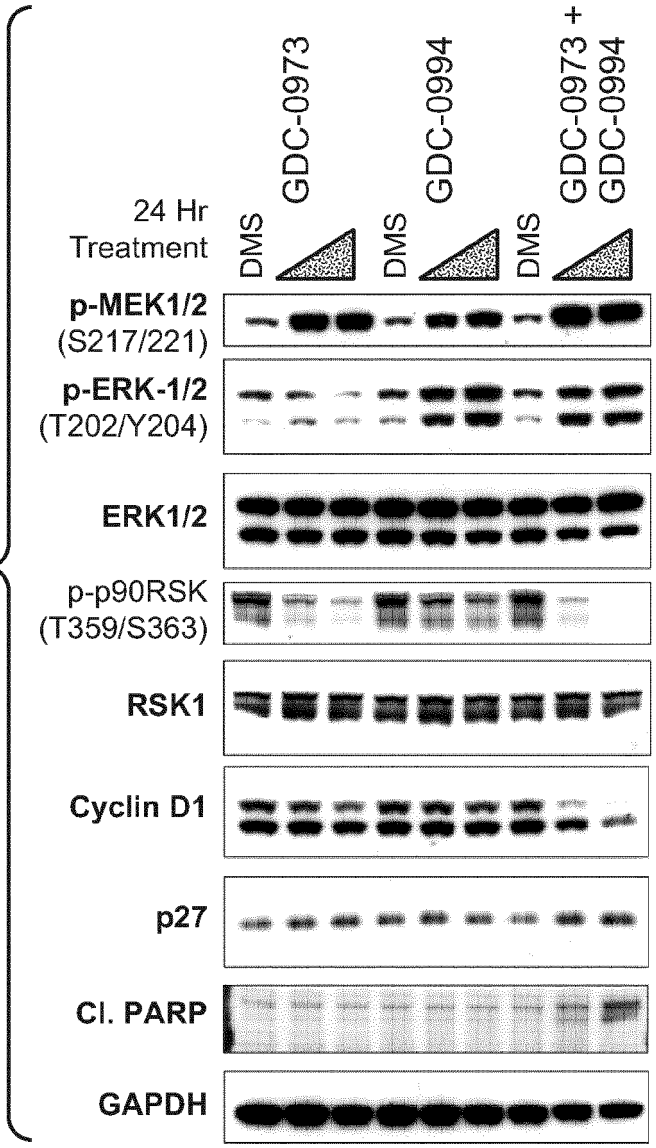


FIG. 11B

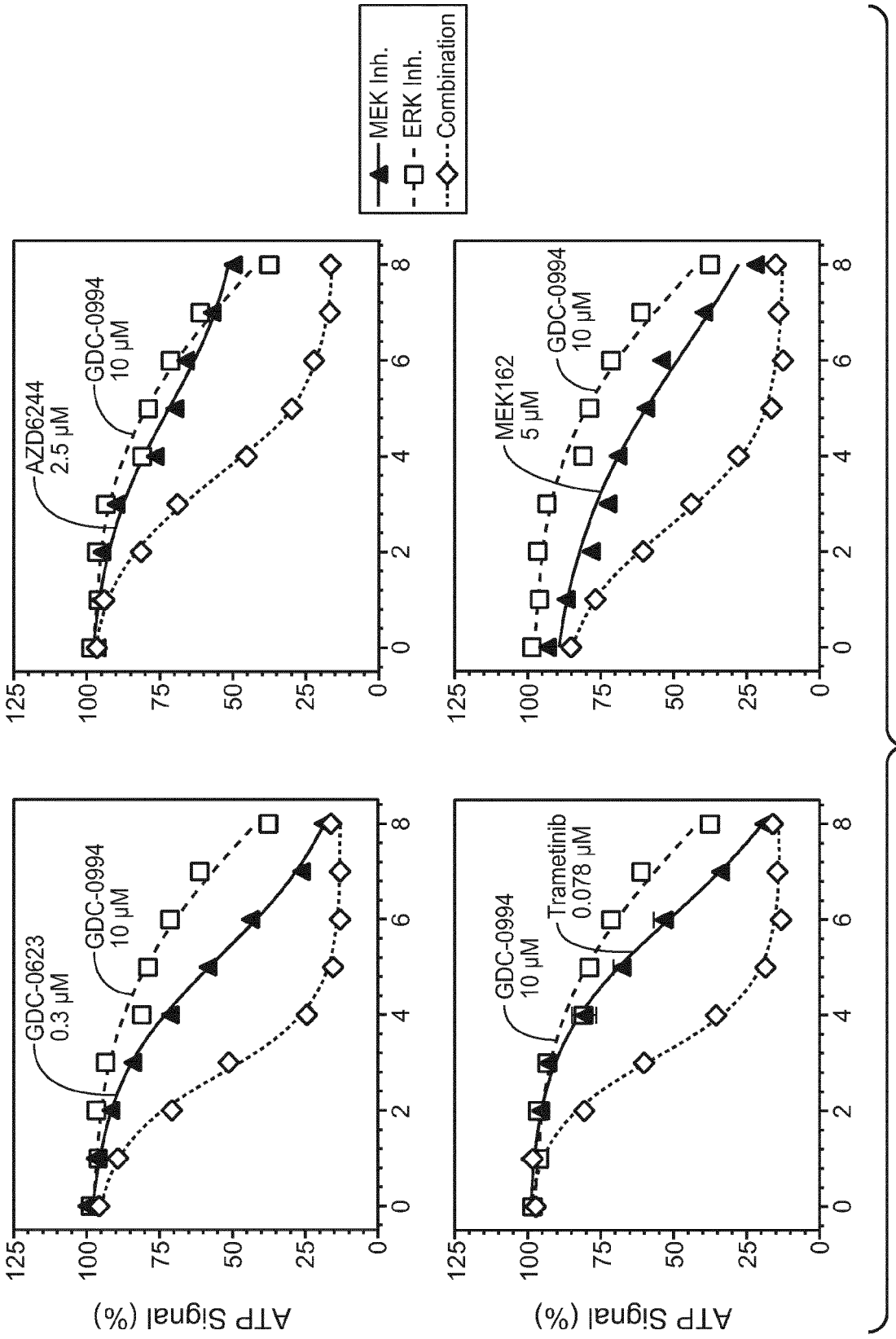

MEK Inh.
ERK Inh.
Combination
ATP Signal (%)
GDC-0623
0.3 µM
GDC-0994
10 µM
AZD6244
2.5 µM
GDC-0994
10 µM
GDC-0994
10 µM
Trametinib
0.078 µM
MEK162
5 µM
GDC-0994
10 µM
0
25
50
75
100
125
0
2
4
6
8

FIG. 12

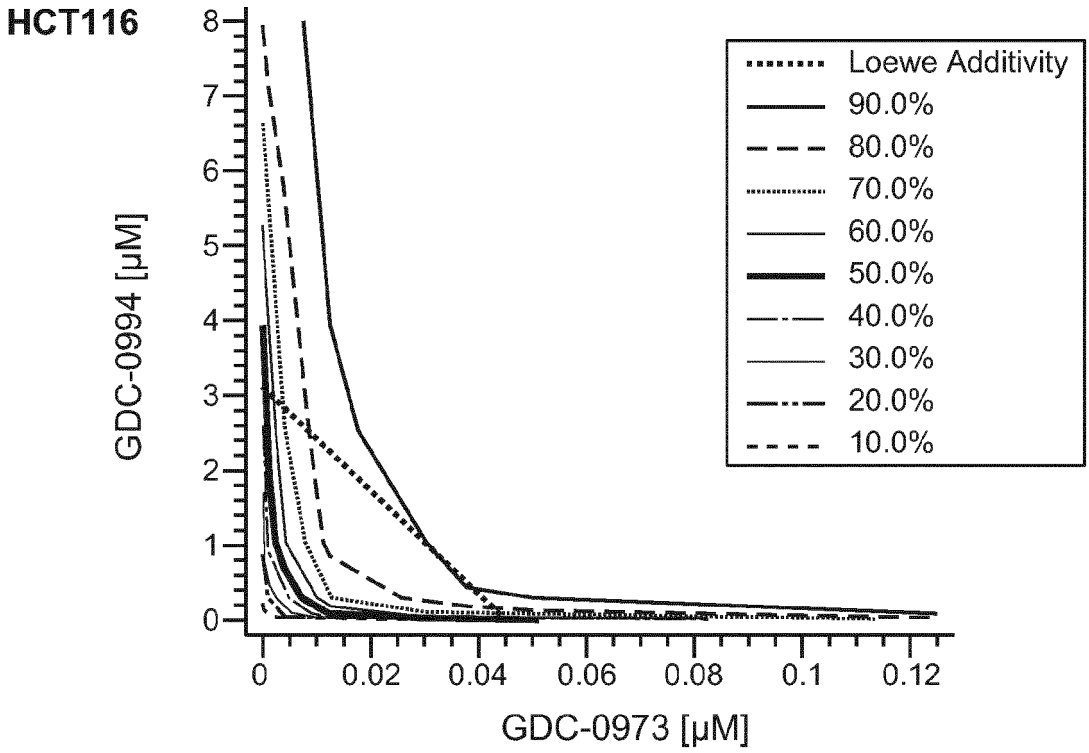


FIG. 13A

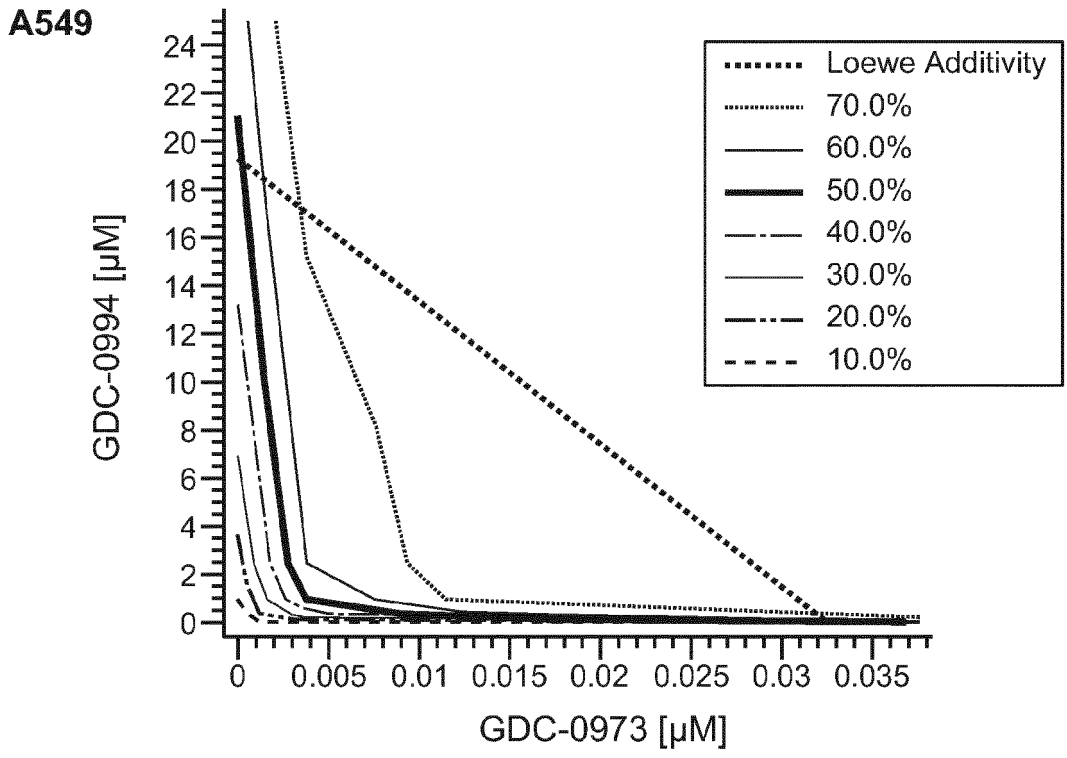


FIG. 13C

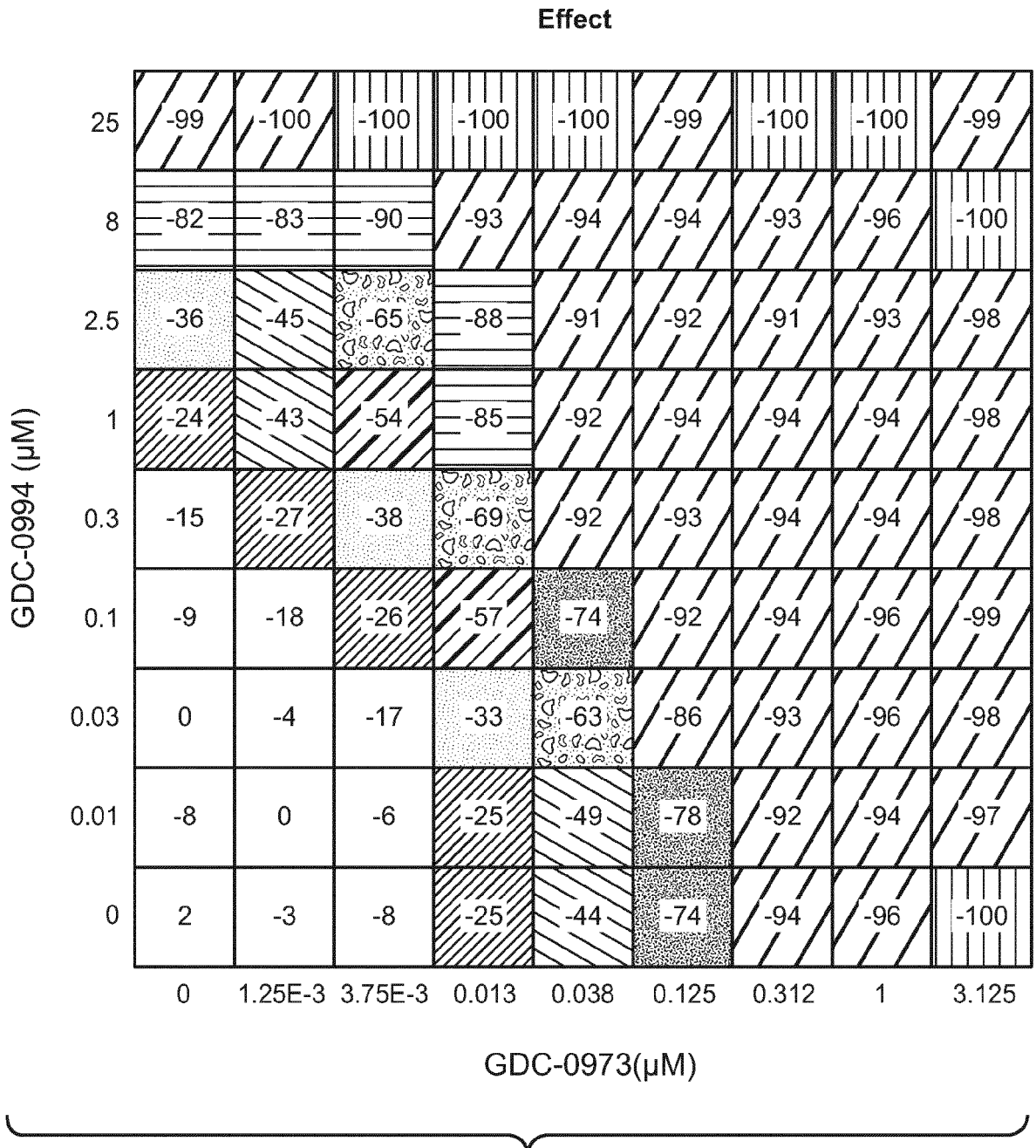

Effect
GDC-0994 (μM)
25
8
2.5
1
0.3
0.1
0.03
0.01
0
-99 -100 -100 -100 -100 -99 -100 -100 -99
-82 -83 -90 -93 -94 -94 -93 -96 -100
-36 -45 -65 -88 -91 -92 -91 -93 -98
-24 -43 -54 -85 -92 -94 -94 -94 -98
-15 -27 -38 -69 -92 -93 -94 -94 -98
-9 -18 -26 -57 -74 -92 -94 -96 -99
0 -4 -17 -33 -63 -86 -93 -96 -98
-8 0 -6 -25 -49 -78 -92 -94 -97
2 -3 -8 -25 -44 -74 -94 -96 -100
0 1.25E-3 3.75E-3 0.013 0.038 0.125 0.312 1 3.125
GDC-0973(μM)

FIG. 13B-1

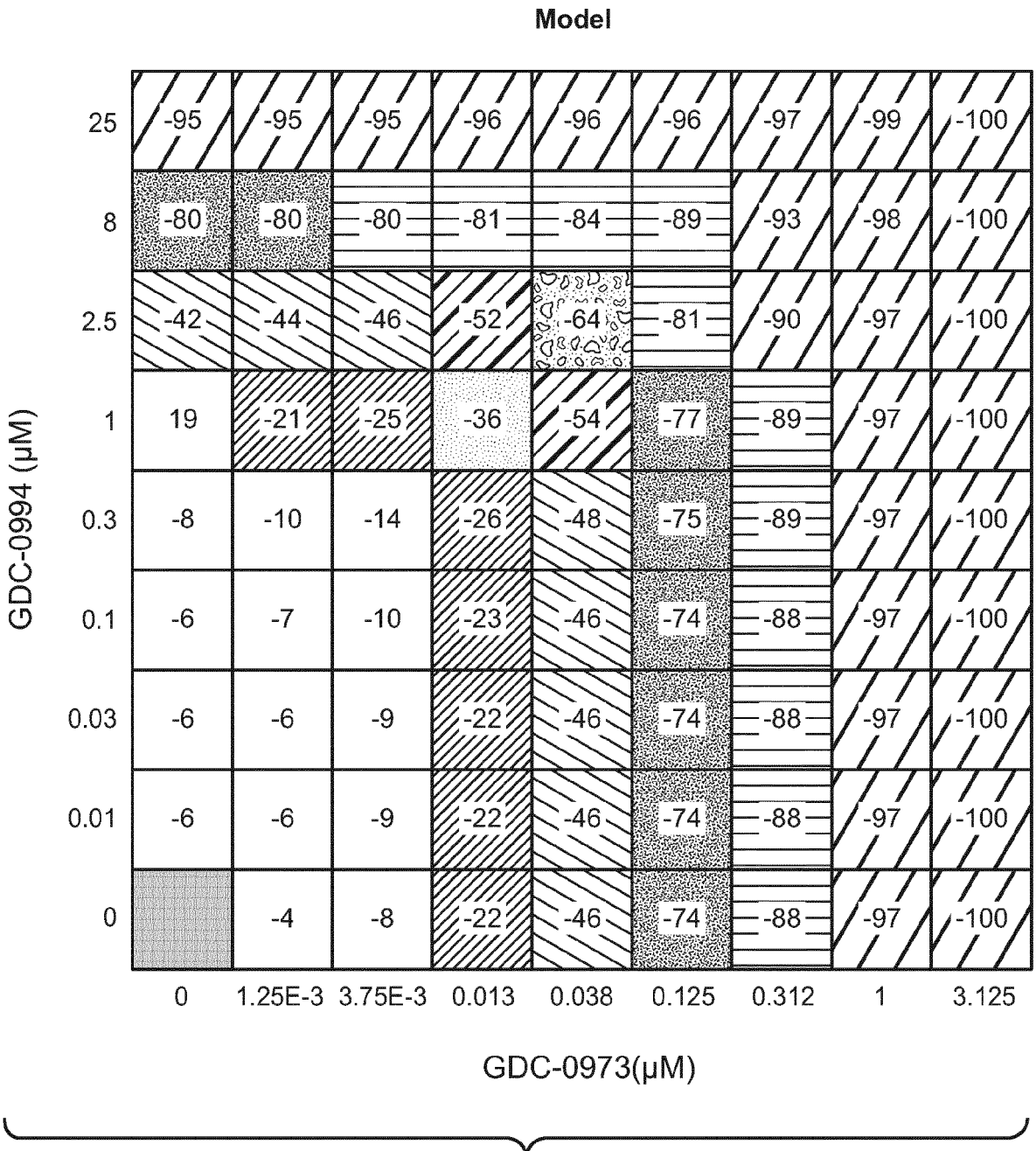

Model
GDC-0994 (μM)
25
8
2.5
1
0.3
0.1
0.03
0.01
0
-95 -95 -95 -96 -96 -96 -97 -99 -100
-80 -80 -80 -81 -84 -89 -93 -98 -100
-42 -44 -46 -52 -64 -81 -90 -97 -100
19 -21 -25 -36 -54 -77 -89 -97 -100
-8 -10 -14 -26 -48 -75 -89 -97 -100
-6 -7 -10 -23 -46 -74 -88 -97 -100
-6 -6 -9 -22 -46 -74 -88 -97 -100
-6 -6 -9 -22 -46 -74 -88 -97 -100
-4 -8 -22 -46 -74 -88 -97 -100
0 1.25E-3 3.75E-3 0.013 0.038 0.125 0.312 1 3.125
GDC-0973(μM)

FIG. 13B-2

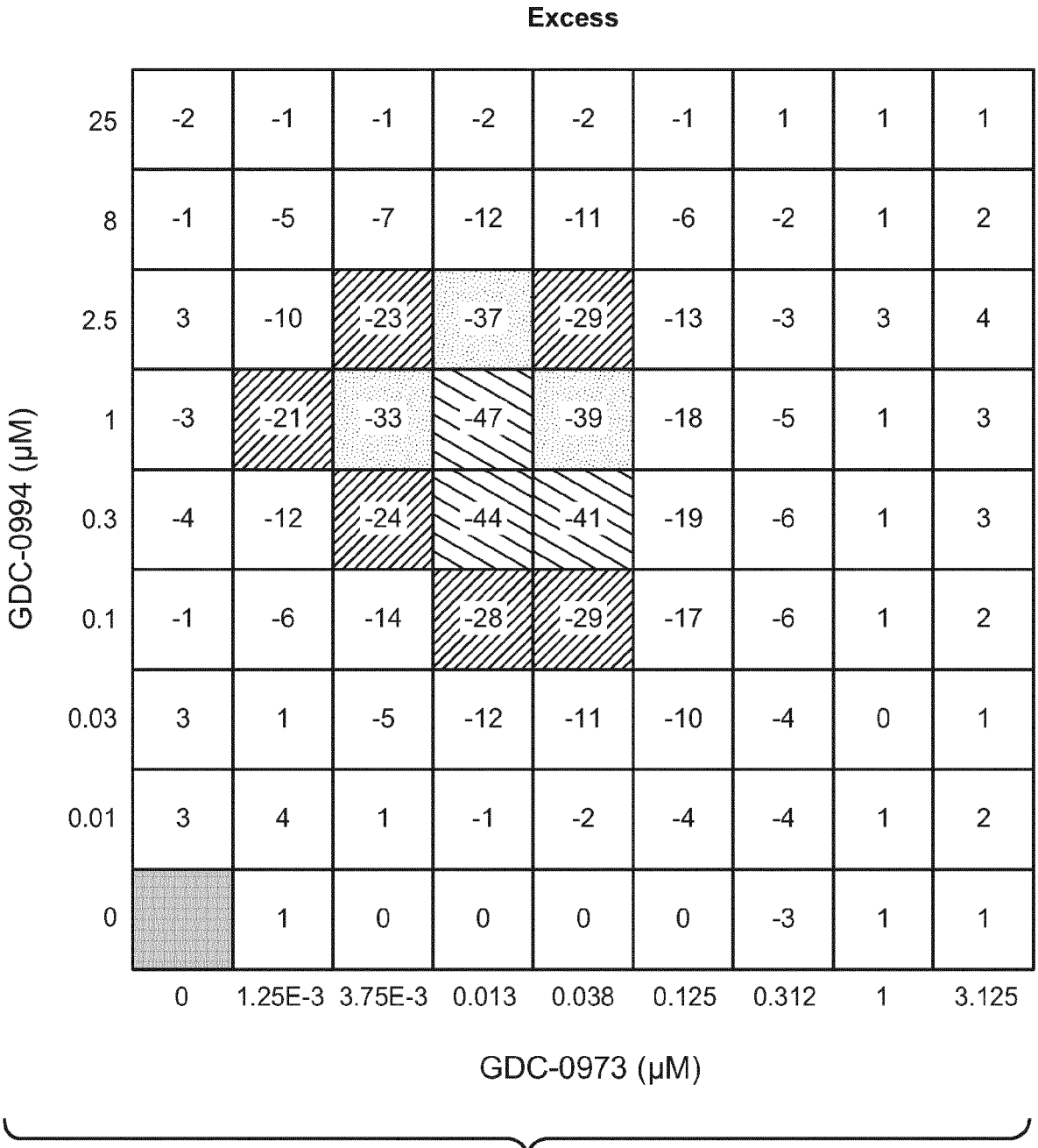

Excess
GDC-0994 (µM)
25 -2 -1 -1 -2 -2 -1 1 1 1
8 -1 -5 -7 -12 -11 -6 -2 1 2
2.5 3 -10 -23 -37 -29 -13 -3 3 4
1 -3 -21 -33 -47 -39 -18 -5 1 3
0.3 -4 -12 -24 -44 -41 -19 -6 1 3
0.1 -1 -6 -14 -28 -29 -17 -6 1 2
0.03 3 1 -5 -12 -11 -10 -4 0 1
0.01 3 4 1 -1 -2 -4 -4 1 2
0 1 0 0 0 0 -3 1 1
0 1.25E-3 3.75E-3 0.013 0.038 0.125 0.312 1 3.125
GDC-0973 (µM)

FIG. 13B-3

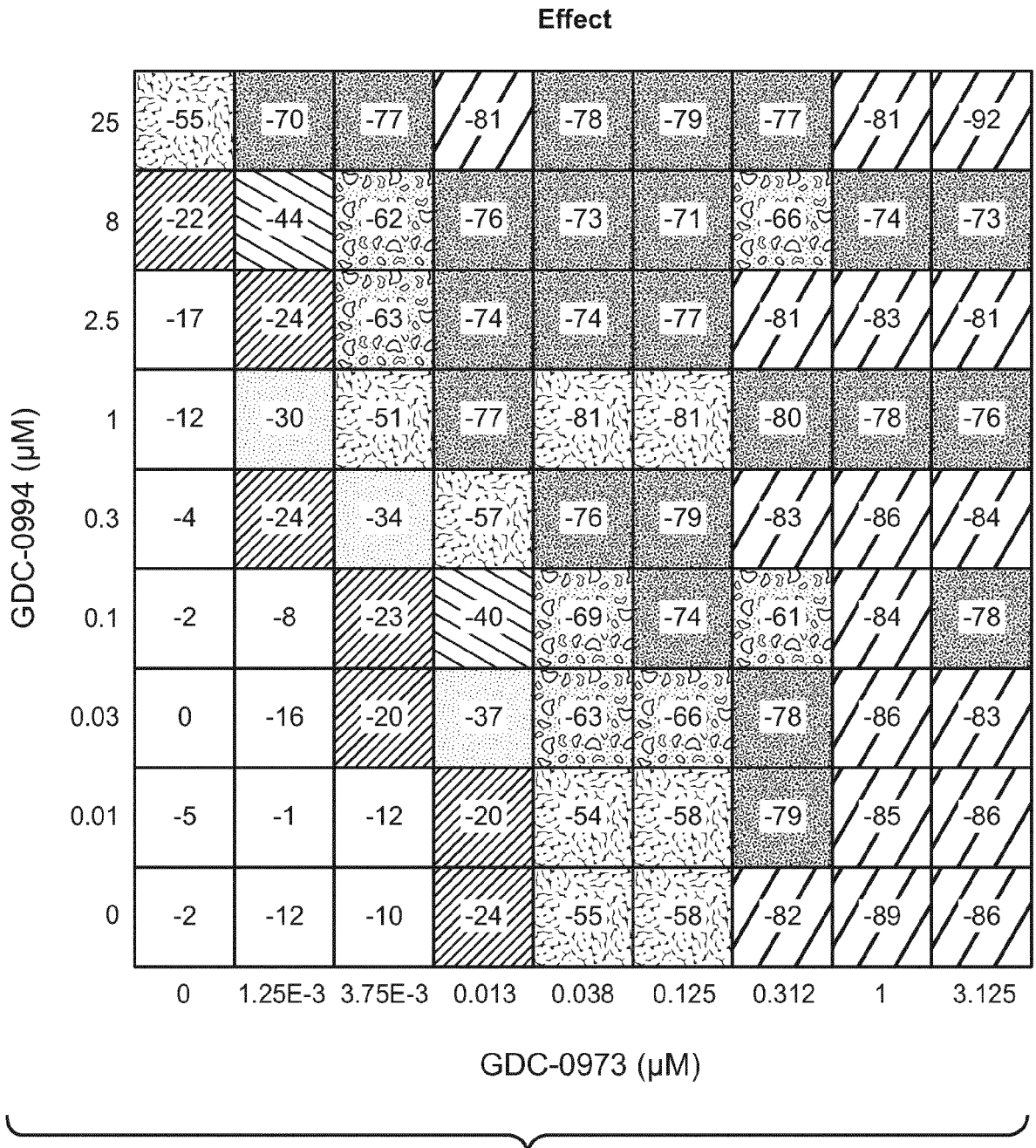

Effect
GDC-0994 (µM)
25
8
2.5
1
0.3
0.1
0.03
0.01
0
-55 -70 -77 -81 -78 -79 -77 -81 -92
-22 -44 -62 -76 -73 -71 -66 -74 -73
-17 -24 -63 -74 -74 -77 -81 -83 -81
-12 -30 -51 -77 -81 -81 -80 -78 -76
-4 -24 -34 -57 -76 -79 -83 -86 -84
-2 -8 -23 -40 -69 -74 -61 -84 -78
0 -16 -20 -37 -63 -66 -78 -86 -83
-5 -1 -12 -20 -54 -58 -79 -85 -86
-2 -12 -10 -24 -55 -58 -82 -89 -86
0 1.25E-3 3.75E-3 0.013 0.038 0.125 0.312 1 3.125
GDC-0973 (µM)

FIG. 13D-1

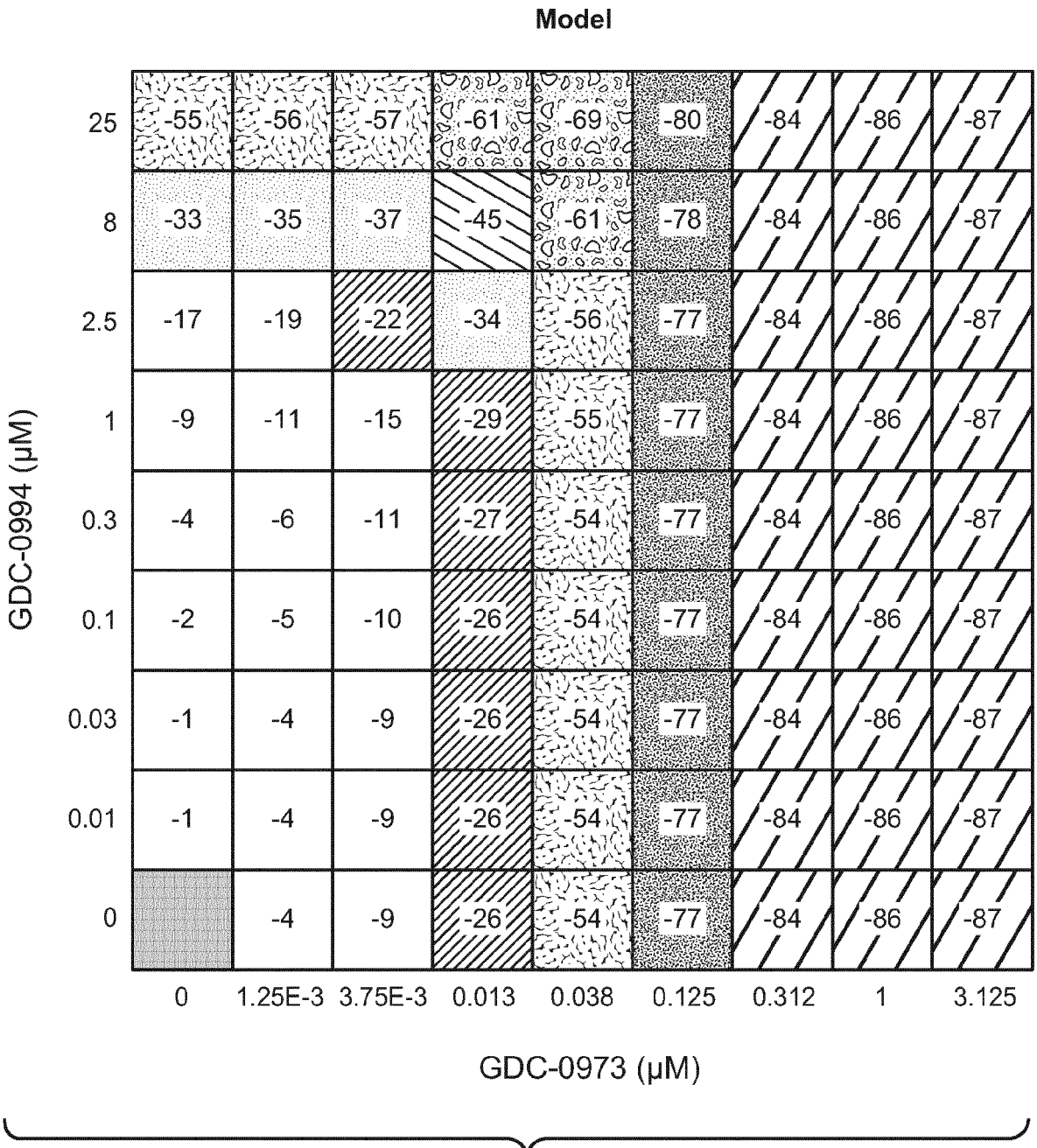

Model
GDC-0994 (μM)
25
8
2.5
1
0.3
0.1
0.03
0.01
0
-55 -56 -57 -61 -69 -80 -84 -86 -87
-33 -35 -37 -45 -61 -78 -84 -86 -87
-17 -19 -22 -34 -56 -77 -84 -86 -87
-9 -11 -15 -29 -55 -77 -84 -86 -87
-4 -6 -11 -27 -54 -77 -84 -86 -87
-2 -5 -10 -26 -54 -77 -84 -86 -87
-1 -4 -9 -26 -54 -77 -84 -86 -87
-1 -4 -9 -26 -54 -77 -84 -86 -87
-4 -9 -26 -54 -77 -84 -86 -87
0 1.25E-3 3.75E-3 0.013 0.038 0.125 0.312 1 3.125
GDC-0973 (μM)

FIG. 13D-2

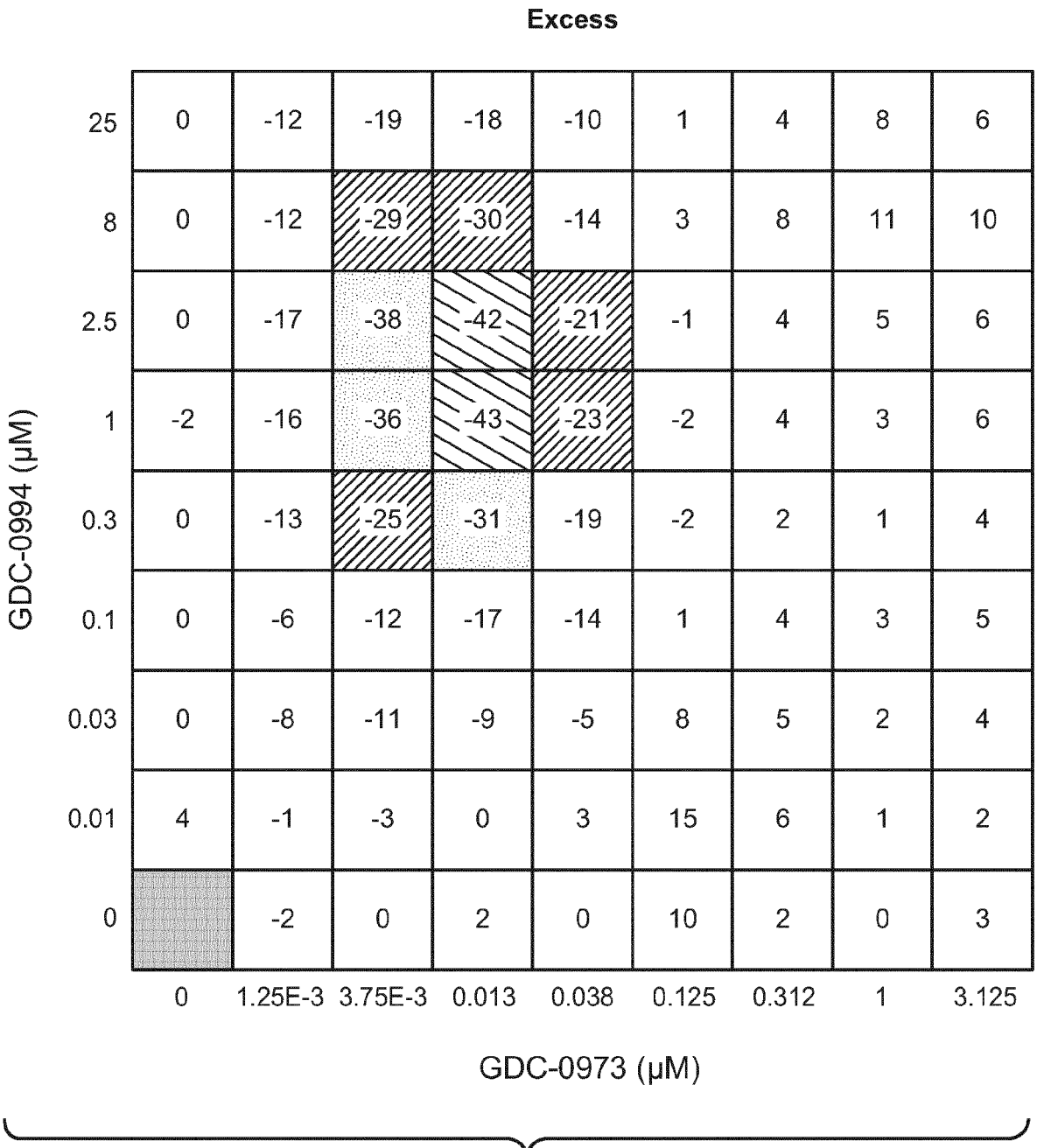

Excess
GDC-0994 (μM)
25 0 -12 -19 -18 -10 1 4 8 6
8 0 -12 -29 -30 -14 3 8 11 10
2.5 0 -17 -38 -42 -21 -1 4 5 6
1 -2 -16 -36 -43 -23 -2 4 3 6
0.3 0 -13 -25 -31 -19 -2 2 1 4
0.1 0 -6 -12 -17 -14 1 4 3 5
0.03 0 -8 -11 -9 -5 8 5 2 4
0.01 4 -1 -3 0 3 15 6 1 2
0 -2 0 2 0 10 2 0 3
0 1.25E-3 3.75E-3 0.013 0.038 0.125 0.312 1 3.125
GDC-0973 (μM)

FIG. 13D-3