FIG. 1

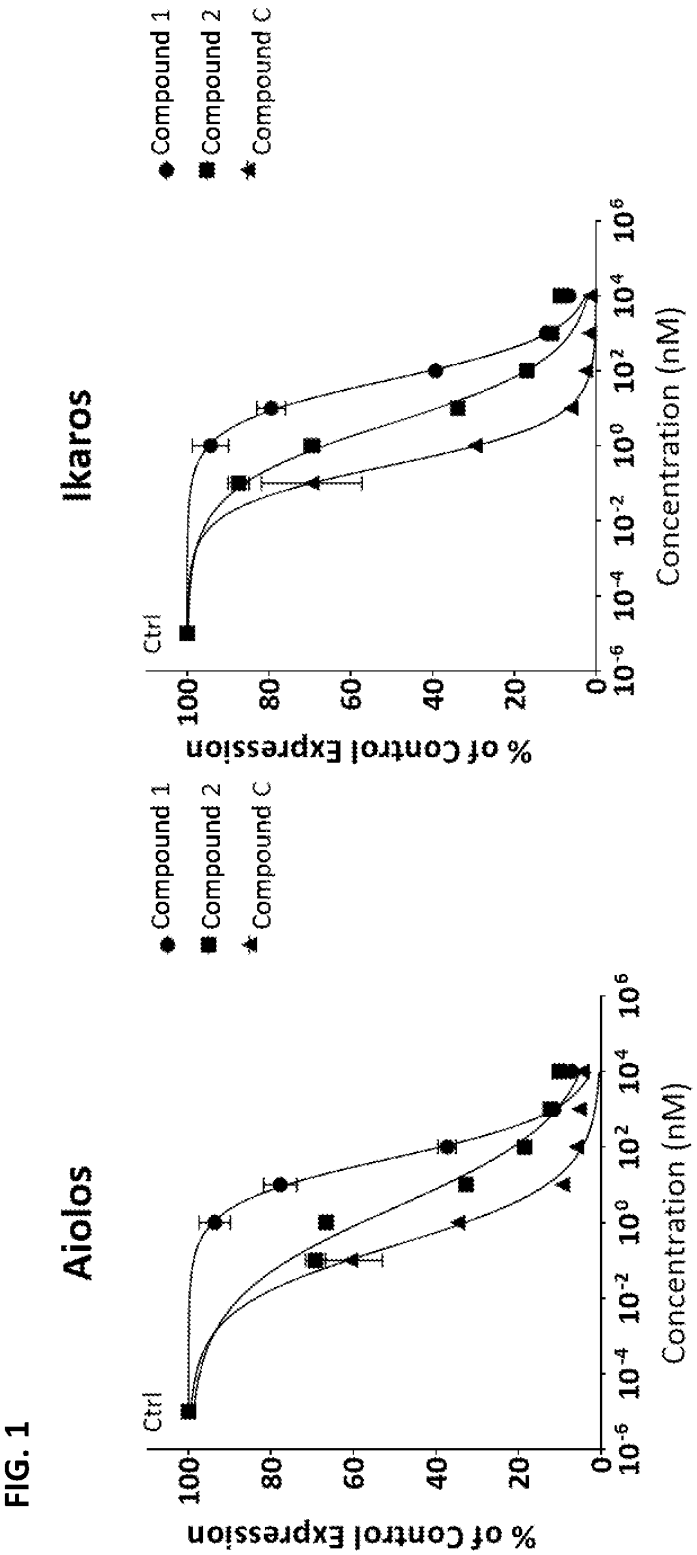

Aiolos
Ikaros
Compound 1
Compound 2
Compound C
Ctrl
% of Control Expression
Concentration (nM)
0
20
40
60
80
100
10^{-6}
10^{-4}
10^{-2}
10^{0}
10^{2}
10^{4}
10^{6}

FIG. 2A

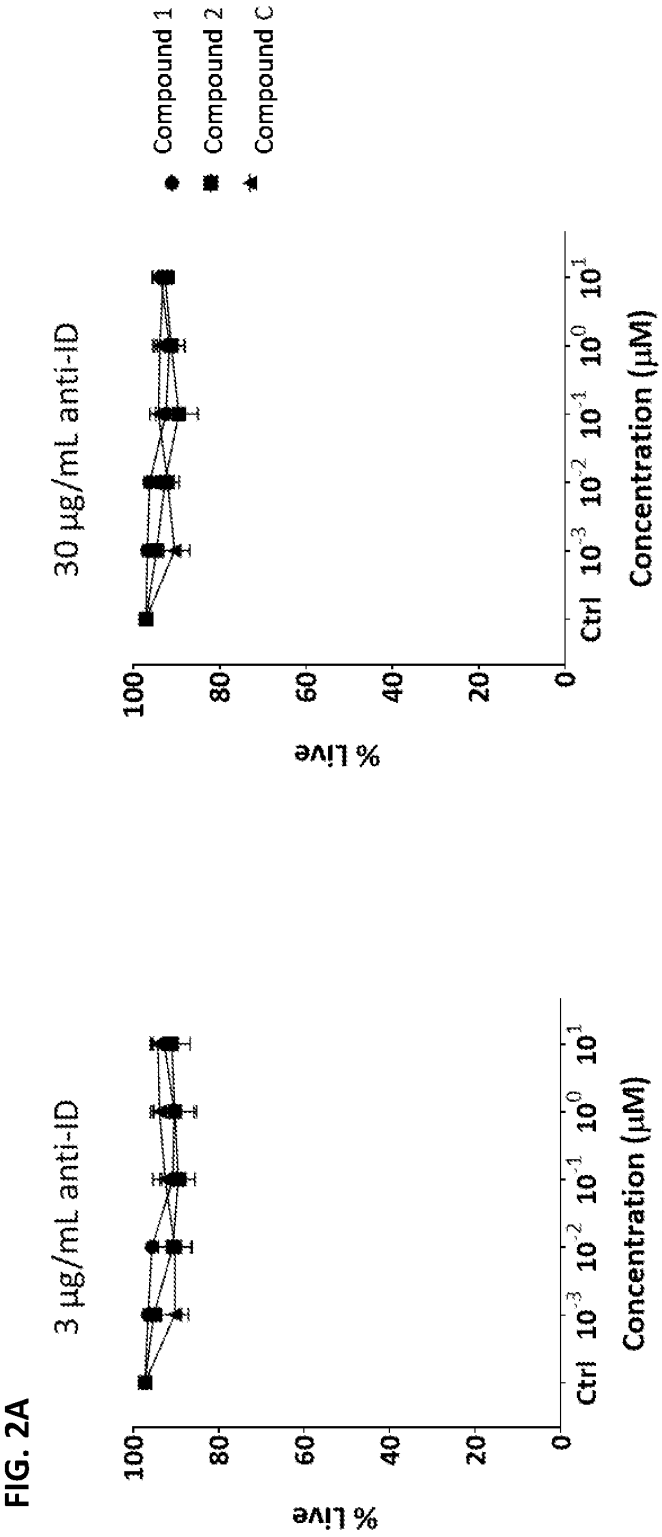

3 µg/mL anti-ID
30 µg/mL anti-ID
Compound 1
Compound 2
Compound C
% Live
100
80
60
40
20
0
Ctrl
10^{-3}
10^{-2}
10^{-1}
10^{0}
10^{1}
Concentration (µM)

FIG. 2C

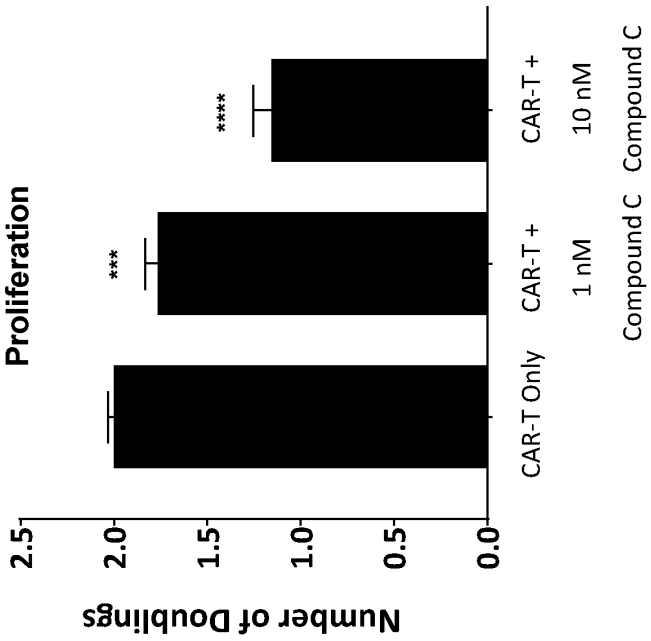
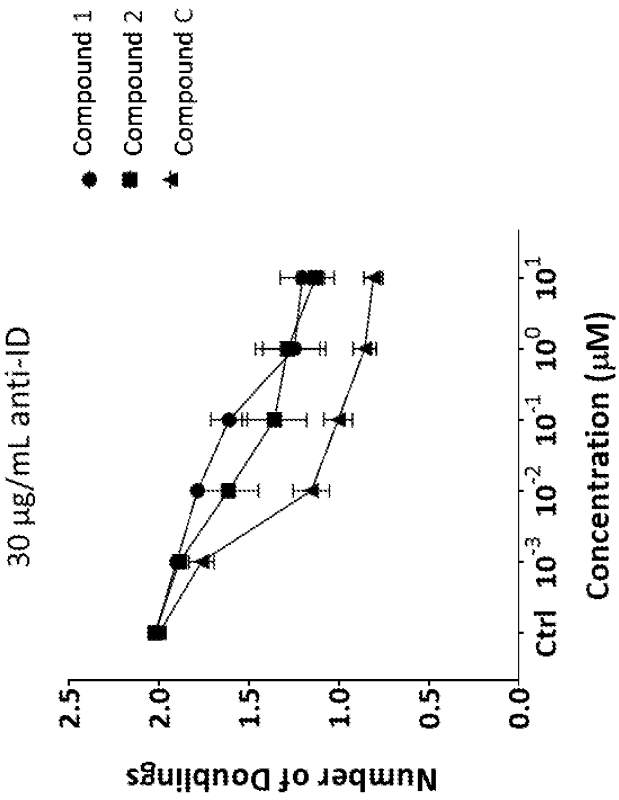

Proliferation
Number of Doublings
2.5
2.0
1.5
1.0
0.5
0.0
***
****
CAR-T Only
CAR-T + 1 nM Compound C
CAR-T + 10 nM Compound C

FIG. 2B


30 µg/mL anti-ID
Compound 1
Compound 2
Compound C
Number of Doublings
2.5
2.0
1.5
1.0
0.5
0.0
Ctrl
10-3
10-2
10-1
100
101
Concentration (µM)

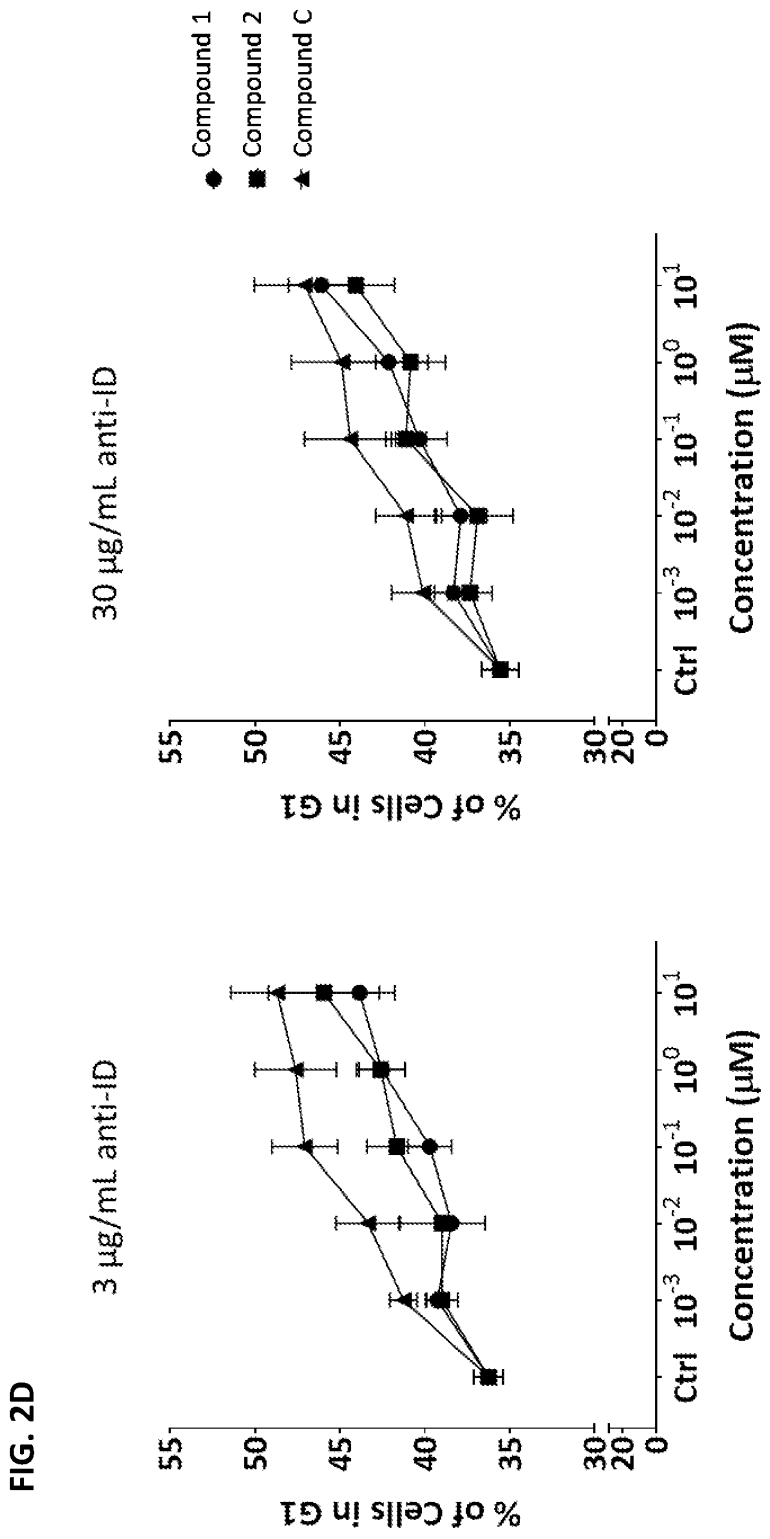

Compound 1
Compound 2
Compound C
30 µg/mL anti-ID
3 µg/mL anti-ID
% of Cells in G1
Concentration (µM)
Ctrl
0
20
30
35
40
45
50
55
10^{-3}
10^{-2}
10^{-1}
10^{0}
10^{1}

FIG. 2D

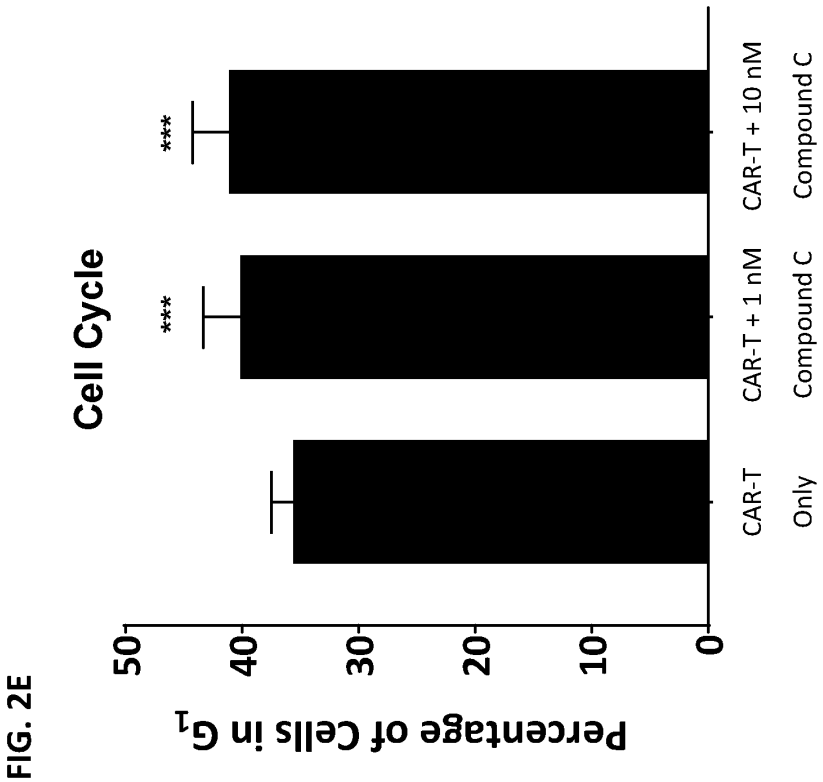

Cell Cycle
Percentage of Cells in G1
0
10
20
30
40
50
***
***
CAR-T Only
CAR-T + 1 nM Compound C
CAR-T + 10 nM Compound C

FIG. 2E

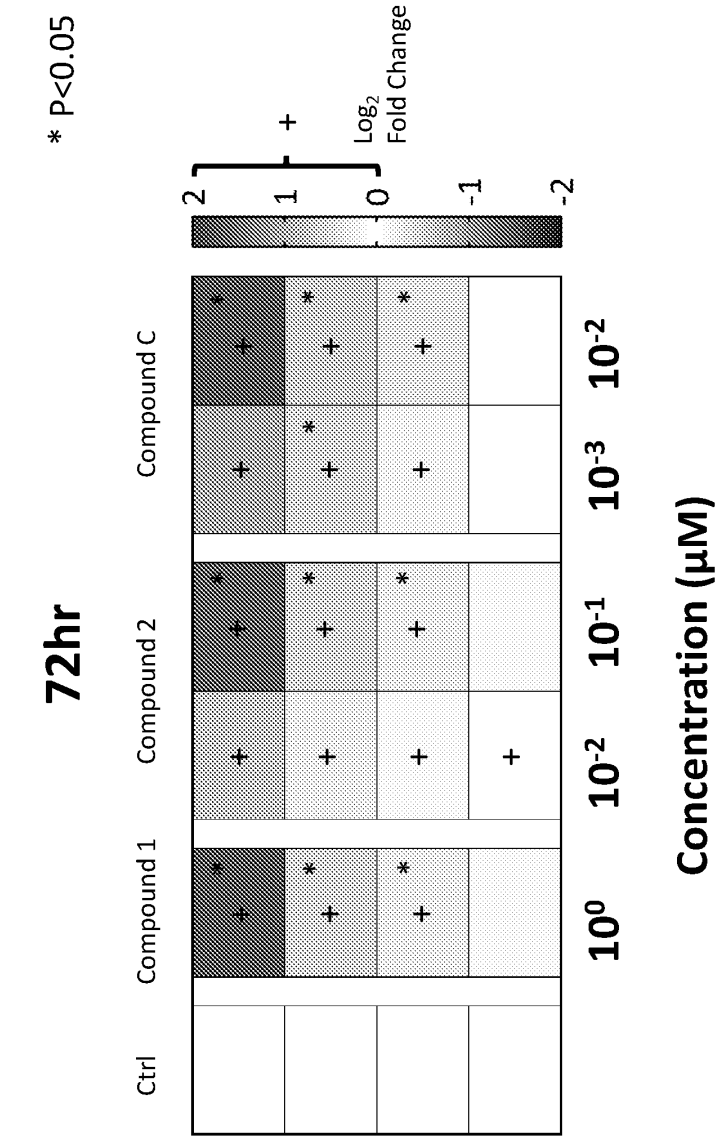
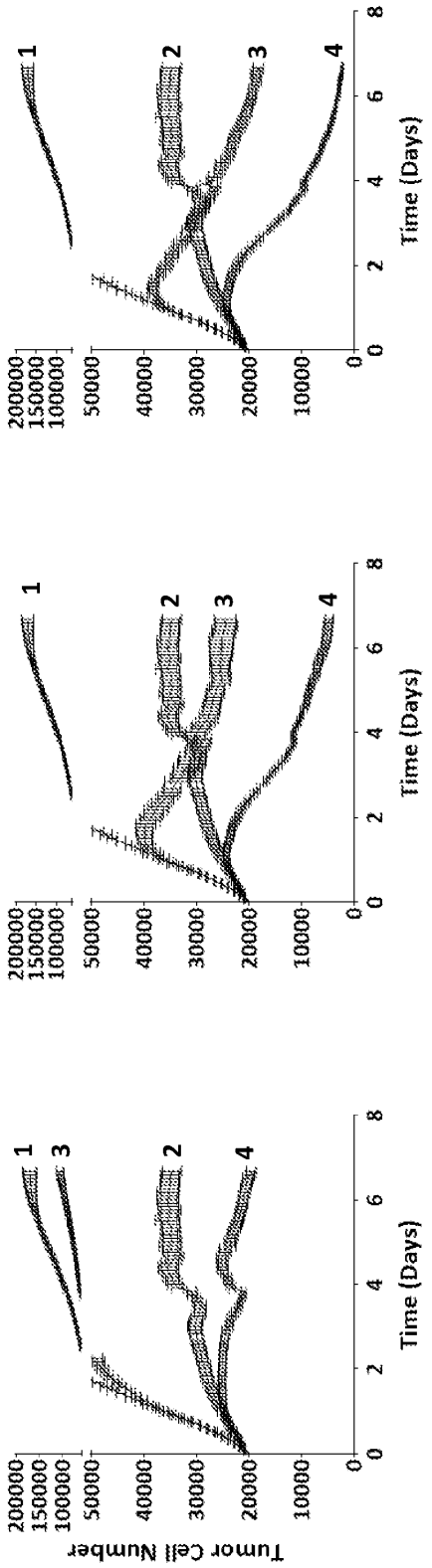

* P<0.05
72hr
Ctrl
Compound 1
Compound 2
Compound C
MFI
IFNg
Perforin
Granzyme B
IL-2
2
1
0
-1
-2
Log2 Fold Change
10^0
10^-2
10^-1
10^-3
10^-2
Concentration (µM)

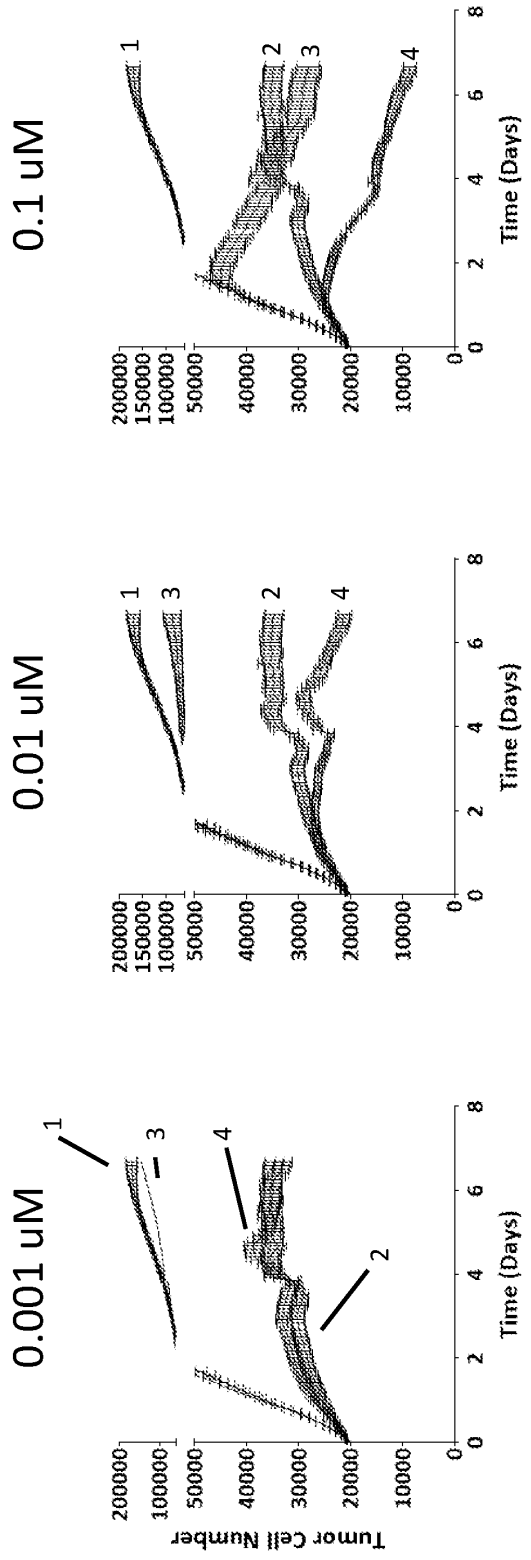
FIG. 2F

FIG. 2G



1 – Tumor Only, 2 – CAR T Only, 3 – Compound C Only, 4 – CAR T + Compound C

FIG. 2H



1 – Tumor Only, 2 – CAR T Only, 3 – Compound 2 Only, 4 – CAR T + Compound 2

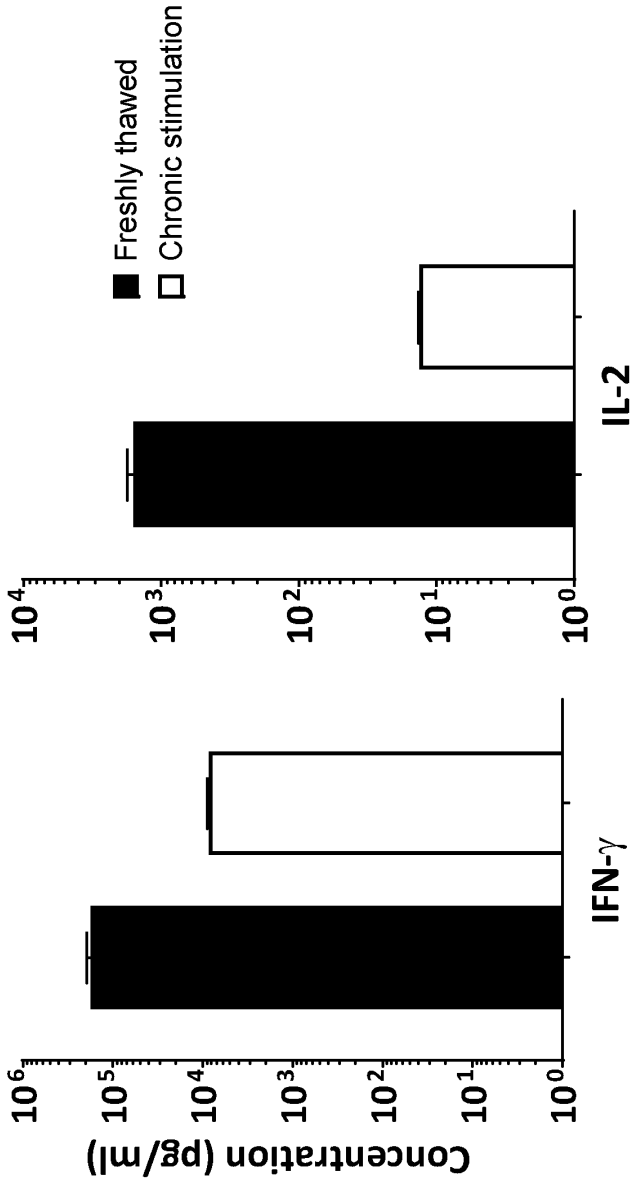

Freshly thawed
Chronic stimulation
Concentration (pg/ml)
10^6
10^5
10^4
10^3
10^2
10^1
10^0
IFN-γ
IL-2

FIG. 3A

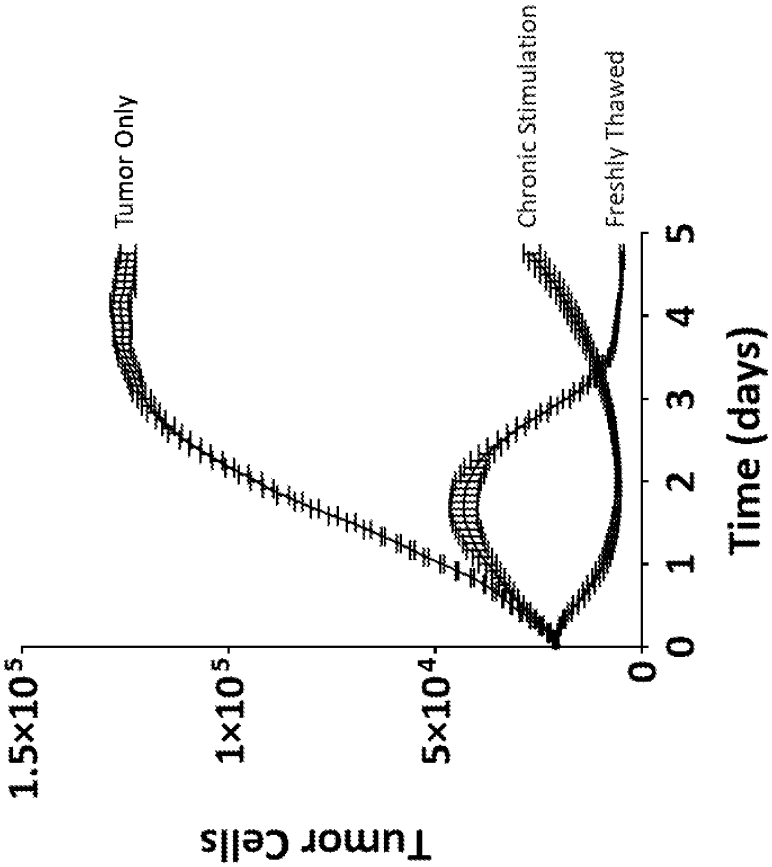

Tumor Only
Chronic Stimulation
Freshly Thawed
Tumor Cells
1.5×10^5
1×10^5
5×10^4
0
0 1 2 3 4 5
Time (days)

FIG. 3B

FIG. 3C

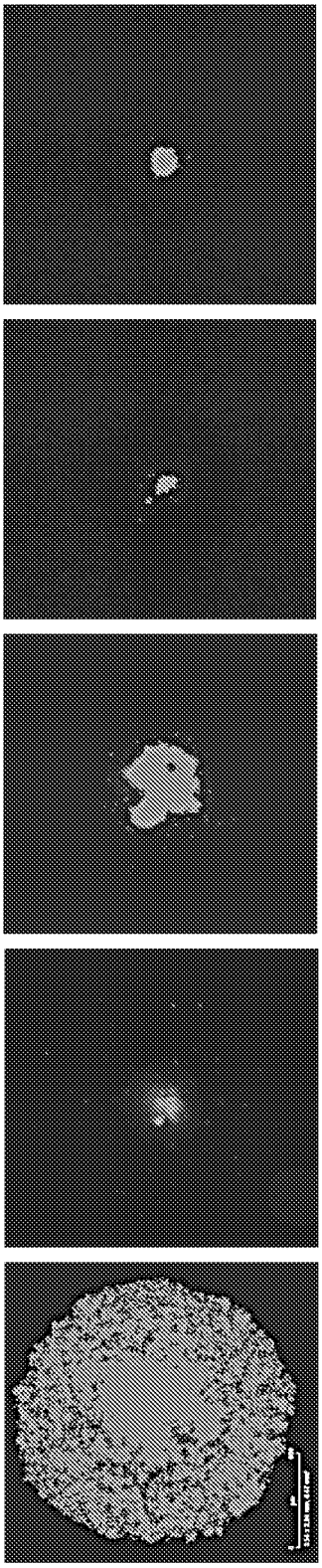

Granta-519
Tumor Only
+ Freshly thawed CAR T
+ Chronic stim CAR T
+ Chronic stim CAR T
+ Compound C 1 nM
+ Chronic stim CAR T
+ Compound C 10 nM

FIG. 3D

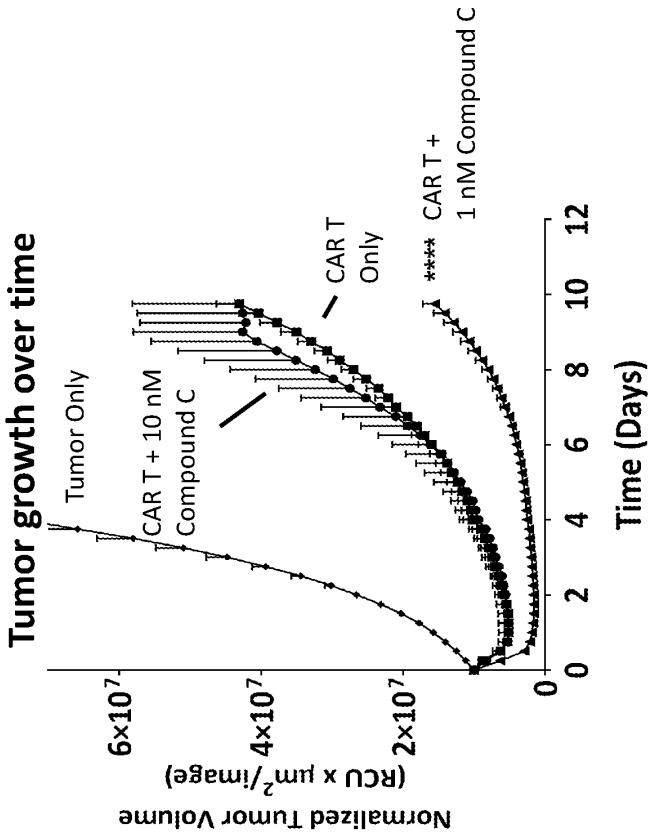
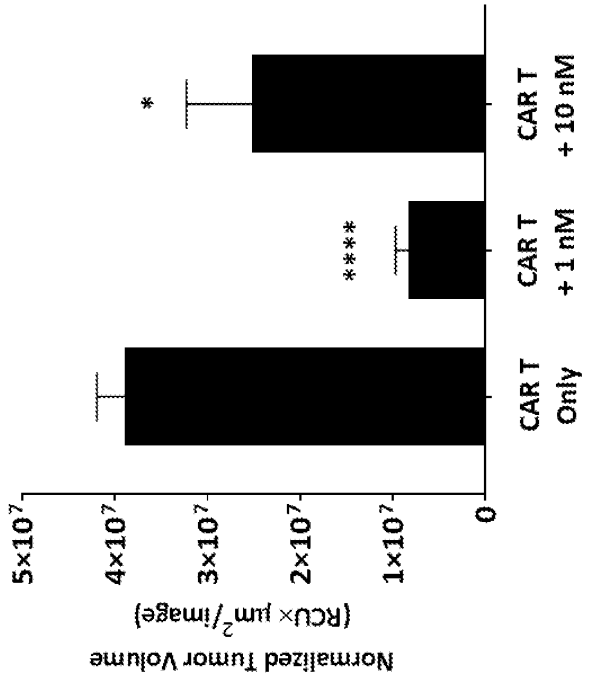

Tumor growth over time
Normalized Tumor Volume
(RCU x μm²/image)
6×10⁷
4×10⁷
2×10⁷
0
0
2
4
6
8
10
12
Time (Days)
Tumor Only
CAR T + 10 nM Compound C
CAR T Only
**** CAR T + 1 nM Compound C

FIG. 3E


Day 9 Tumor Size
Normalized Tumor Volume
(RCU× μm²/image)
5×10⁷
4×10⁷
3×10⁷
2×10⁷
1×10⁷
0
****
*
CAR T Only
CAR T + 1 nM
CAR T + 10 nM

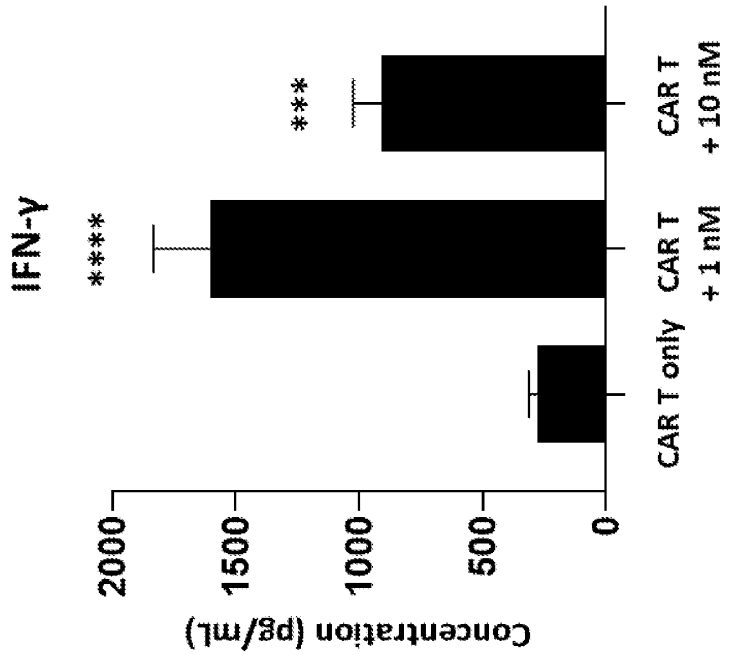

IFN-γ
****
***
Concentration (pg/mL)
2000
1500
1000
500
0
CAR T only
CAR T + 1 nM
CAR T + 10 nM

FIG. 3F

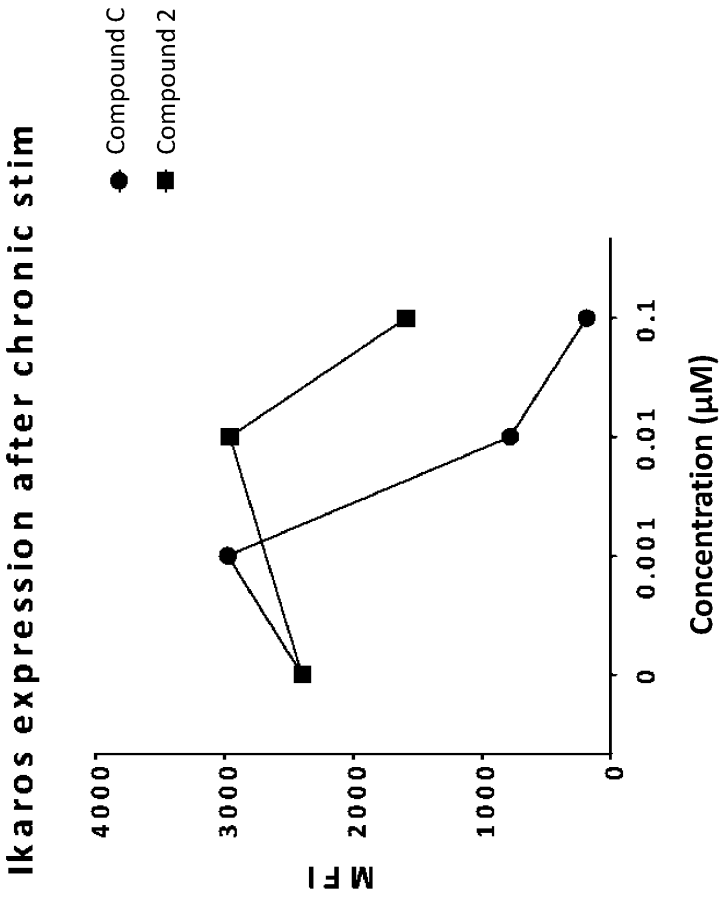

Ikaros expression after chronic stim
Compound C
Compound 2
MFI
4000
3000
2000
1000
0
0
0.001
0.01
0.1
Concentration (μM)

FIG. 3G

FIG. 3H

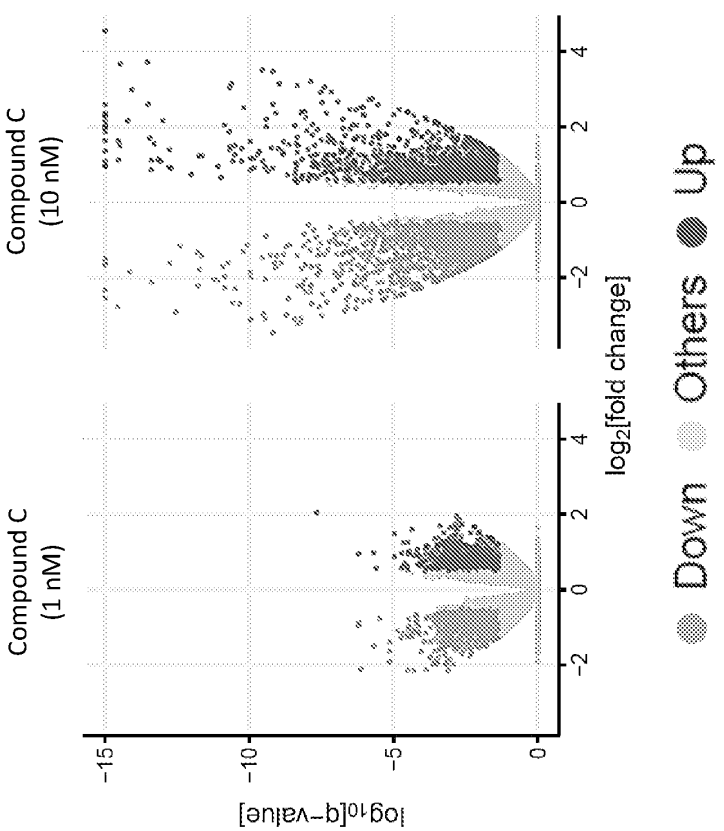
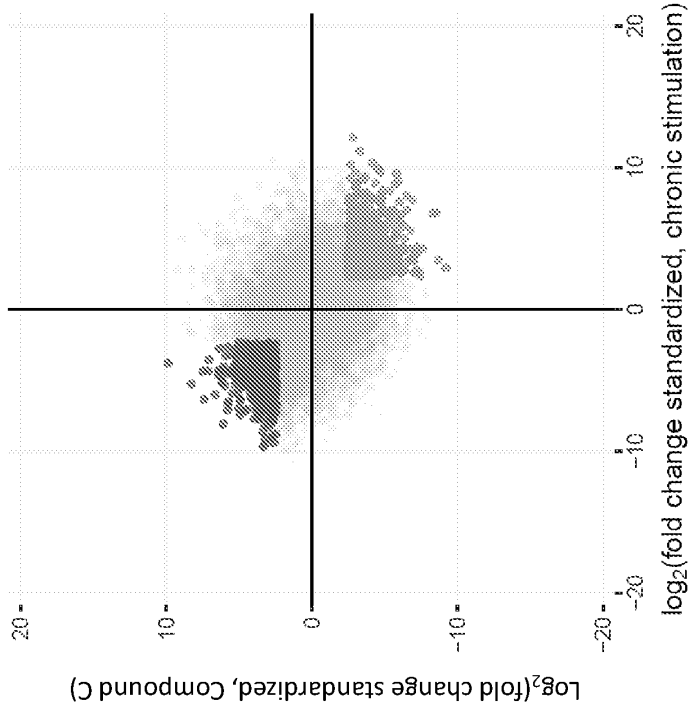

Compound C
(1 nM)
Compound C
(10 nM)
log10[q-value]
log2[fold change]
Down
Others
Up

FIG. 3I


Log2(fold change standardized, Compound C)
log2(fold change standardized, chronic stimulation)

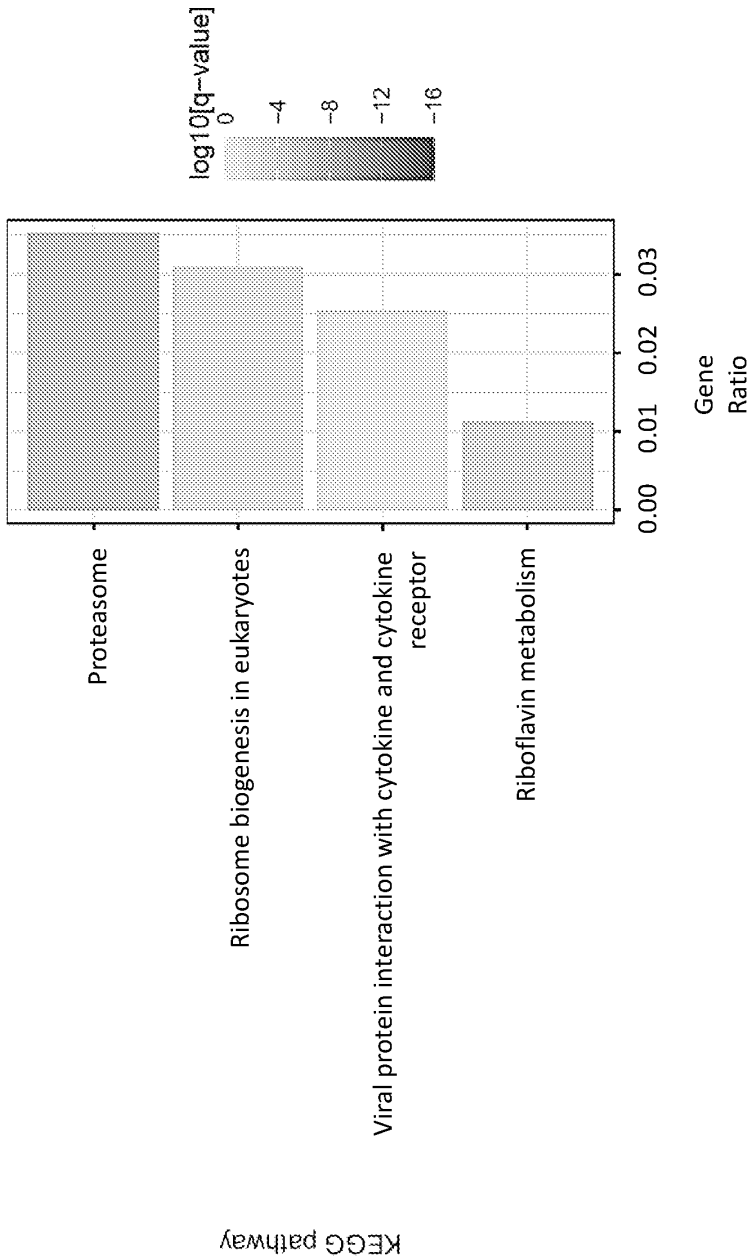

log10[q-value]
0
-4
-8
-12
-16
Proteasome
Ribosome biogenesis in eukaryotes
Viral protein interaction with cytokine and cytokine receptor
Riboflavin metabolism
0.00
0.01
0.02
0.03
Gene Ratio
KEGG pathway

FIG. 3J

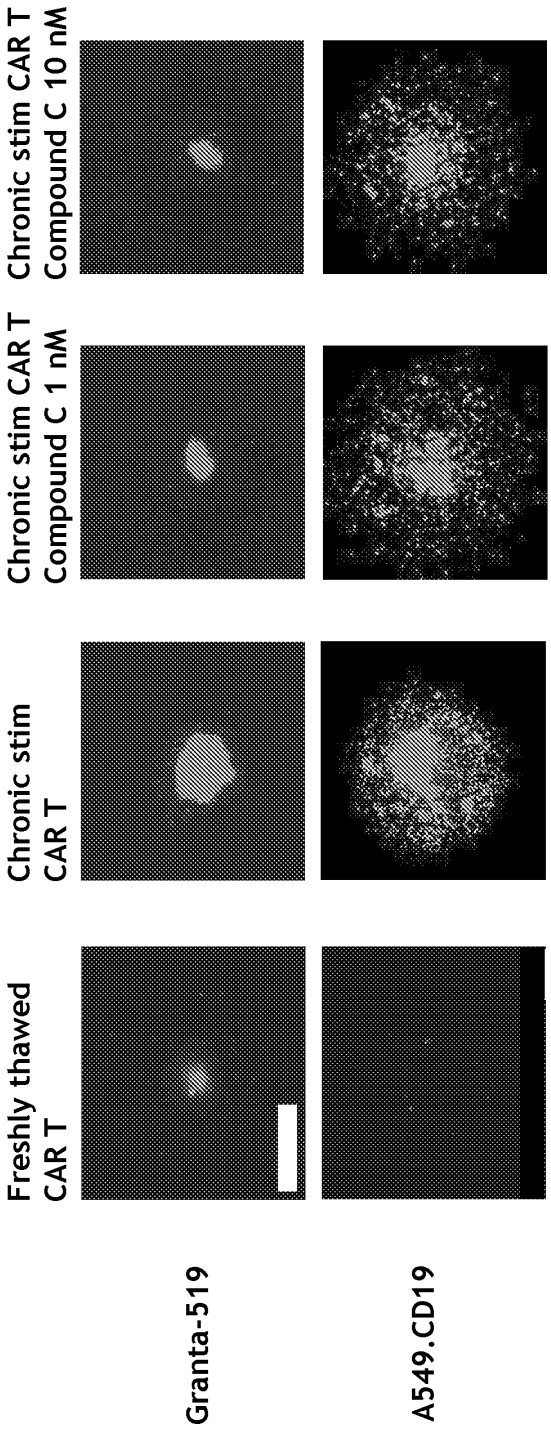

Freshly thawed CAR T
Chronic stim CAR T
Chronic stim CAR T Compound C 1 nM
Chronic stim CAR T Compound C 10 nM
Granta-519
A549.CD19

FIG. 4A

FIG. 4B

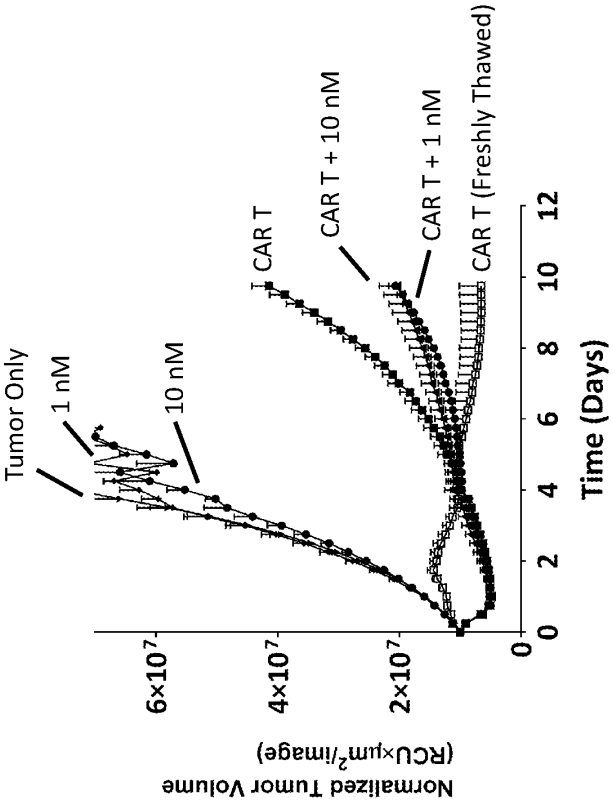
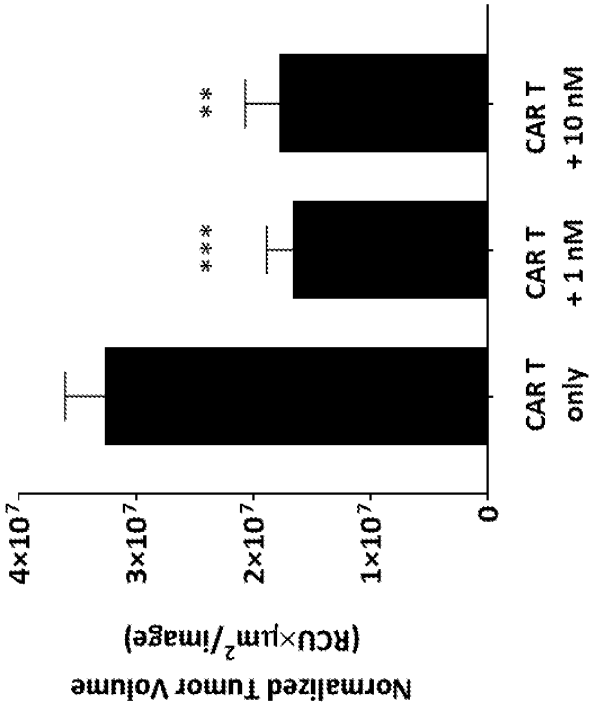

Normalized Tumor Volume
(RCU×μm²/image)
6×10⁷
4×10⁷
2×10⁷
0
0
2
4
6
8
10
12
Time (Days)
Tumor Only
1 nM
10 nM
CAR T
CAR T + 10 nM
CAR T + 1 nM
CAR T (Freshly Thawed)

FIG. 4C


Normalized Tumor Volume
(RCU×μm²/image)
4×10⁷
3×10⁷
2×10⁷
1×10⁷
0
***
**
CAR T only
CAR T + 1 nM
CAR T + 10 nM

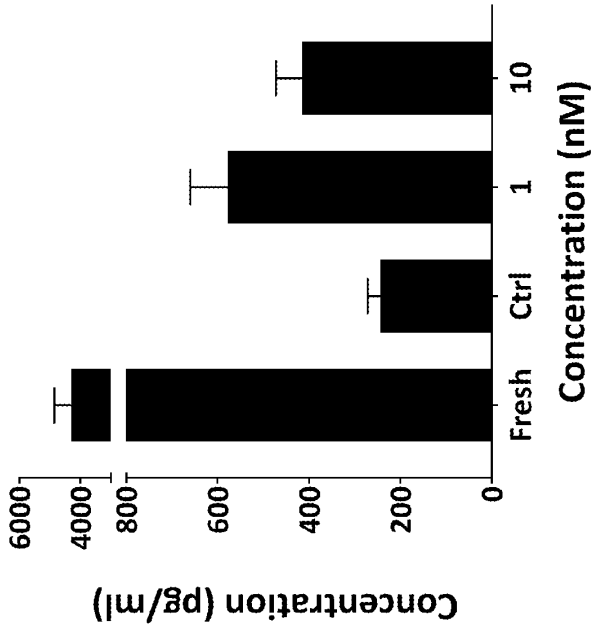

Concentration (pg/ml)
6000
4000
800
600
400
200
0
Fresh
Ctrl
1
10
Concentration (nM)

FIG. 4D

FIG. 4F

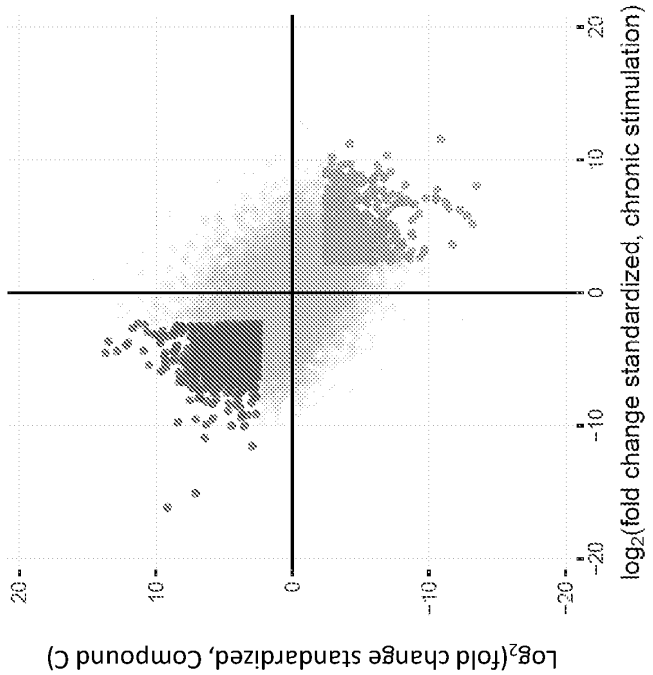
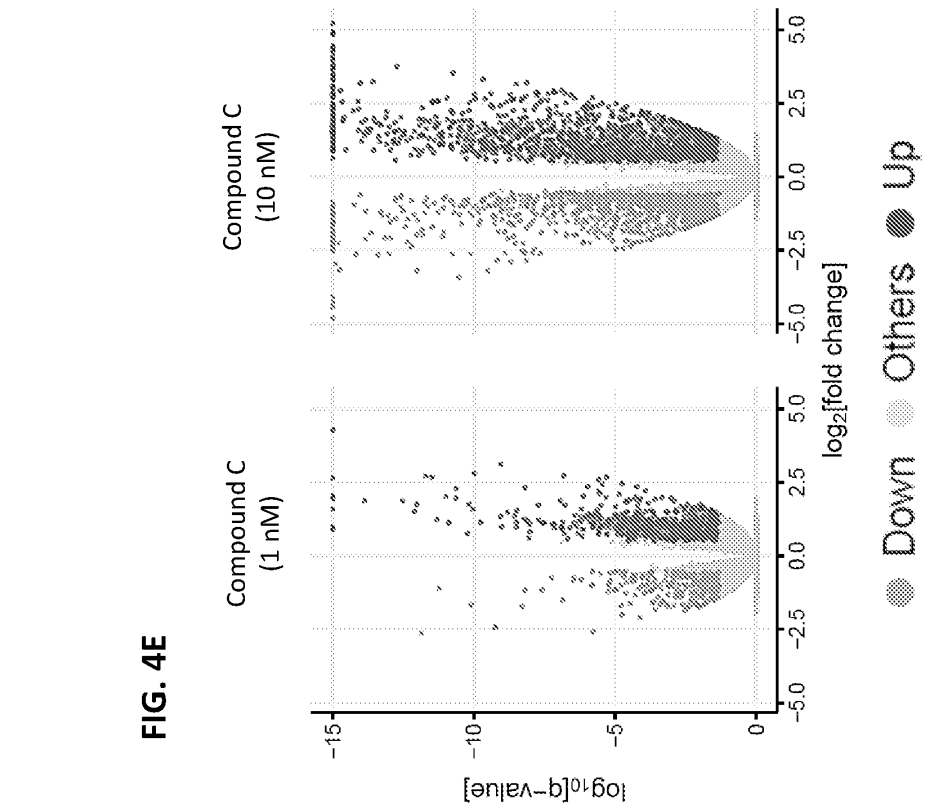

Log2(fold change standardized, Compound C)
log2(fold change standardized, chronic stimulation)
20
10
0
-10
-20

FIG. 4E


Compound C
(1 nM)
Compound C
(10 nM)
log10[q-value]
log2[fold change]
Down
Others
Up

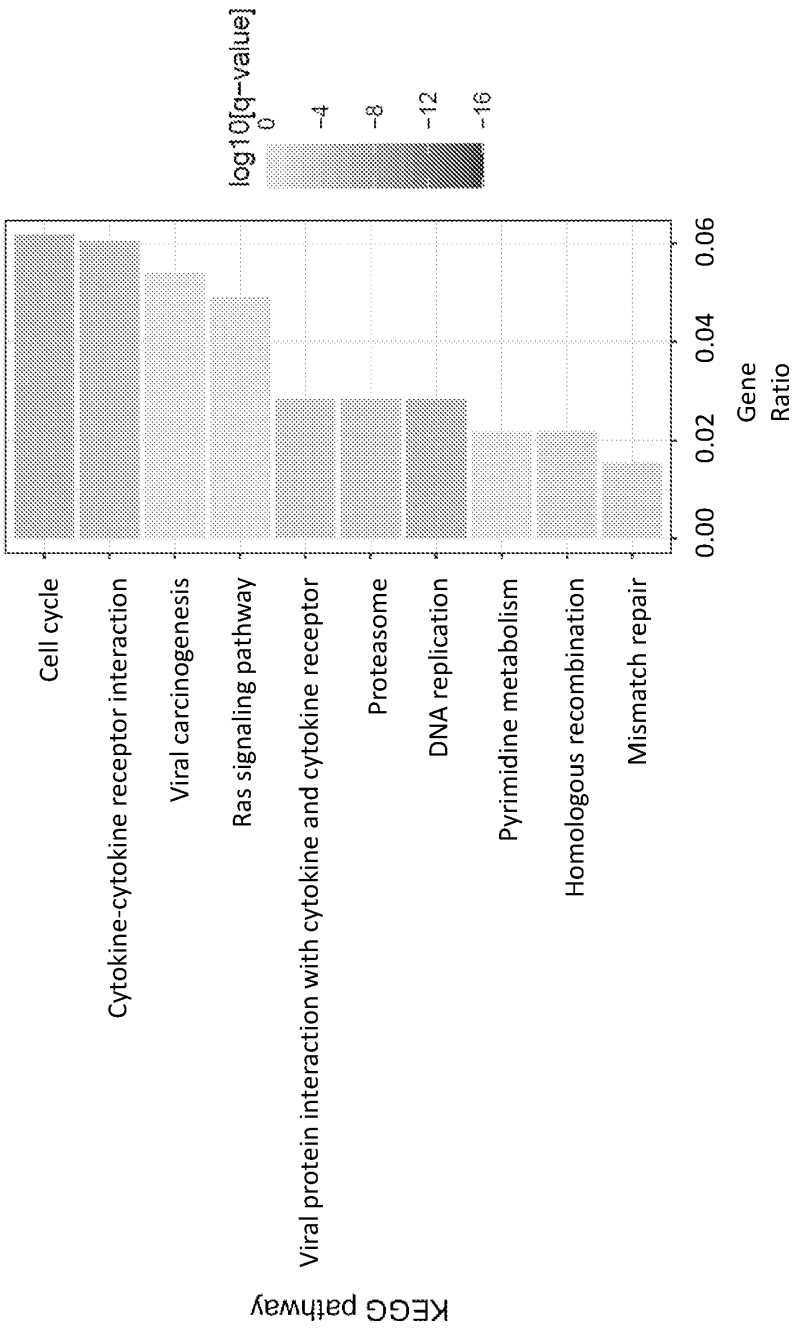

log10[q-value]
0
-4
-8
-12
-16
Cell cycle
Cytokine-cytokine receptor interaction
Viral carcinogenesis
Ras signaling pathway
Viral protein interaction with cytokine and cytokine receptor
Proteasome
DNA replication
Pyrimidine metabolism
Homologous recombination
Mismatch repair
KEGG pathway
0.00
0.02
0.04
0.06
Gene Ratio

FIG. 4G

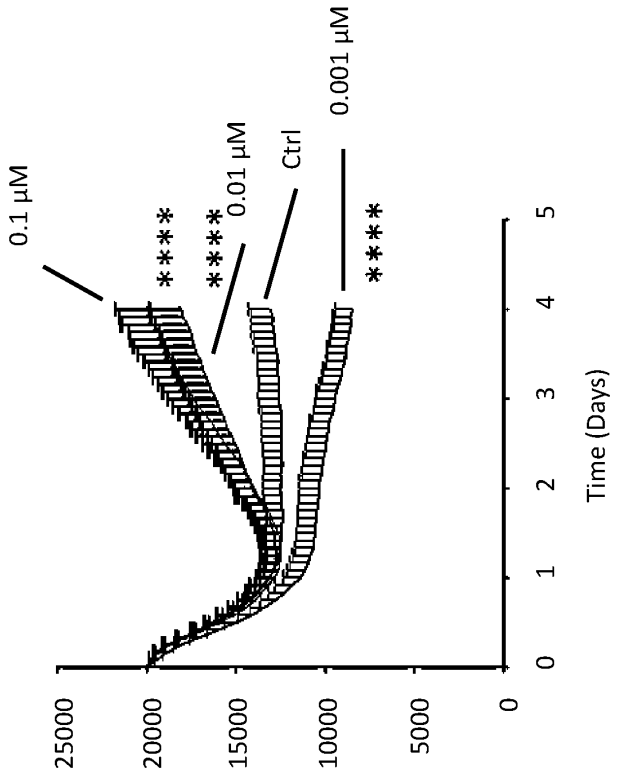

0.1 µM
****
****
0.01 µM
Ctrl
0.001 µM
****
25000
20000
15000
10000
5000
0
0
1
2
3
4
5
Time (Days)

FIG. 5B

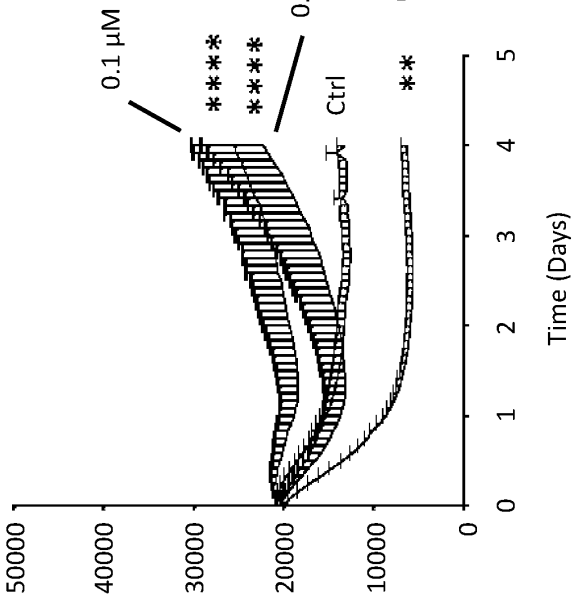

0.1 µM
****
****
0.01 µM
Ctrl
**
0.001 µM
50000
40000
30000
20000
10000
0
0
1
2
3
4
5
Time (Days)

FIG. 5A

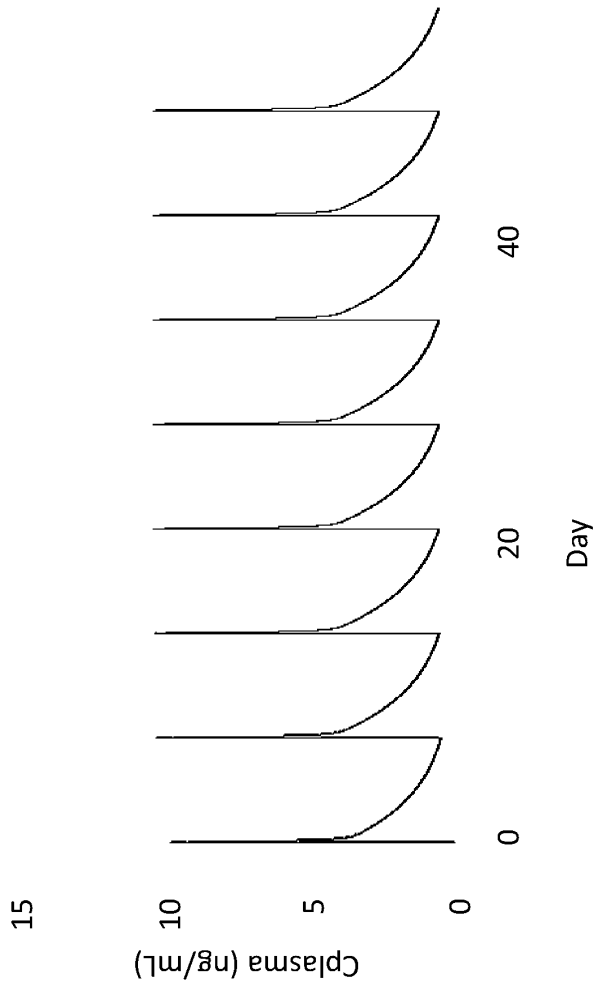


FIG. 6A

FIG. 6B

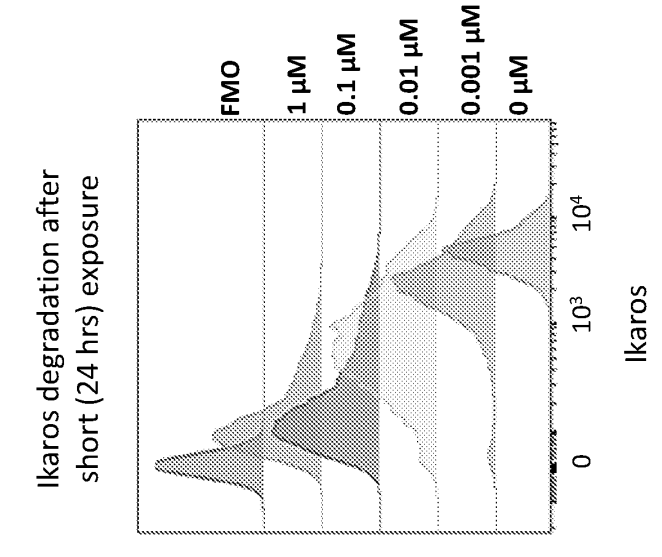
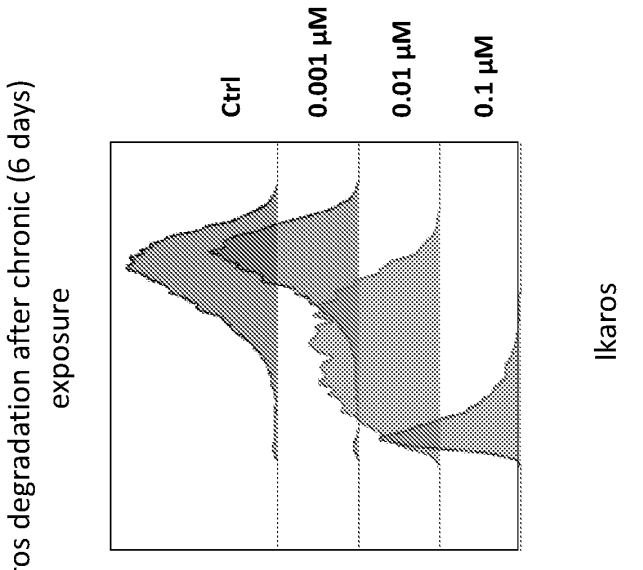

Ikaros degradation after short (24 hrs) exposure
FMO
1 µM
0.1 µM
0.01 µM
0.001 µM
0 µM
0
10^3
10^4
Ikaros

FIG. 6C


Ikaros degradation after chronic (6 days) exposure
Ctrl
0.001 µM
0.01 µM
0.1 µM
Ikaros

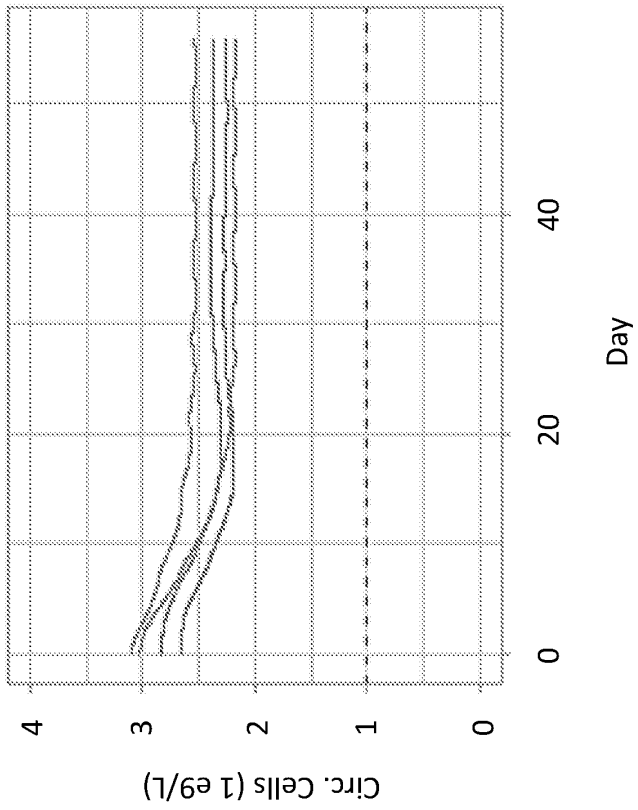


FIG. 6D

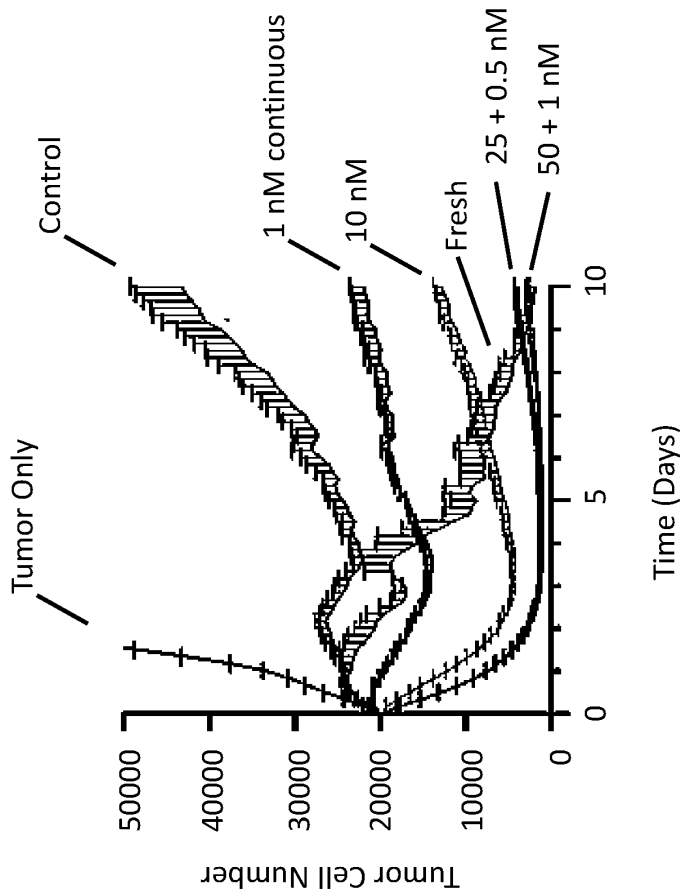

Tumor Only
Control
1 nM continuous
10 nM
Fresh
25 + 0.5 nM
50 + 1 nM
Tumor Cell Number
50000
40000
30000
20000
10000
0
0
5
10
Time (Days)

FIG. 7A

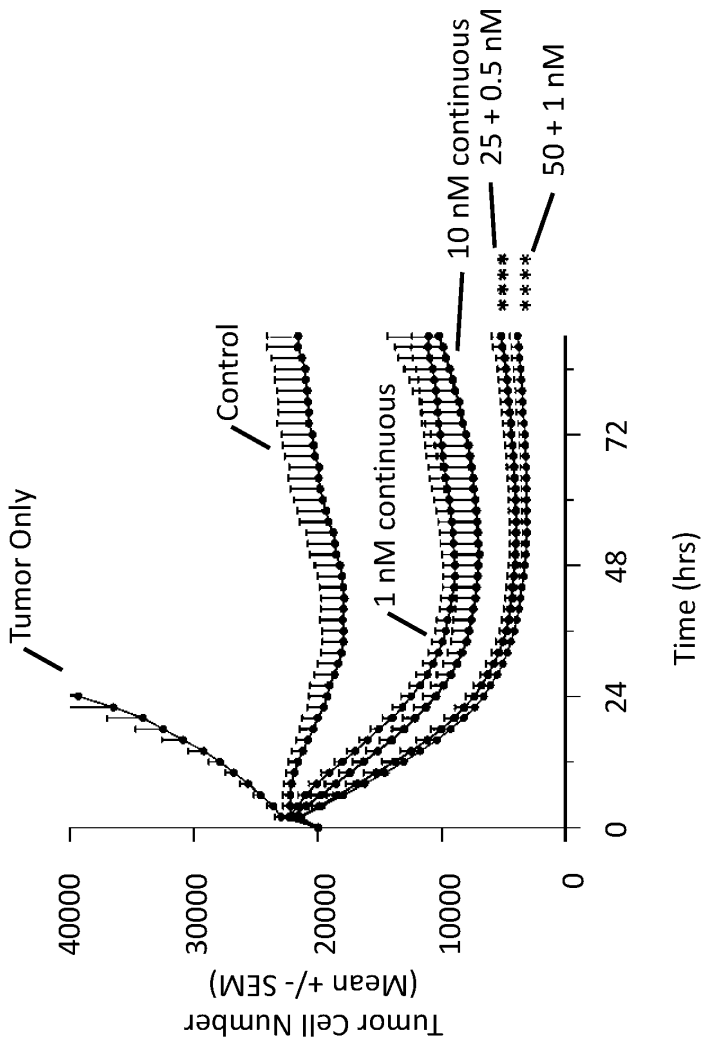

Tumor Only
Control
1 nM continuous
10 nM continuous
25 + 0.5 nM
50 + 1 nM
****
****
Tumor Cell Number
(Mean +/- SEM)
40000
30000
20000
10000
0
0
24
48
72
Time (hrs)

FIG. 7B

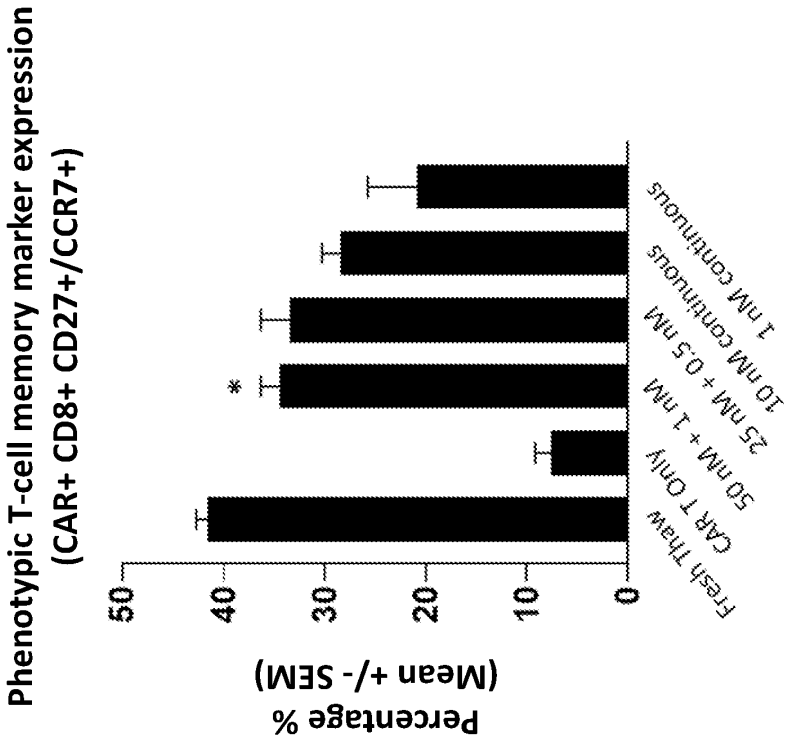

Phenotypic T-cell memory marker expression
(CAR+ CD8+ CD27+/CCR7+)
Percentage %
(Mean +/- SEM)
0
10
20
30
40
50
*
Fresh Thaw
CAR T Only
50 nM + 1 nM
25 nM + 1 nM
10 nM + 0.5 nM
1 nM continuous

FIG. 7C