FIG. 1

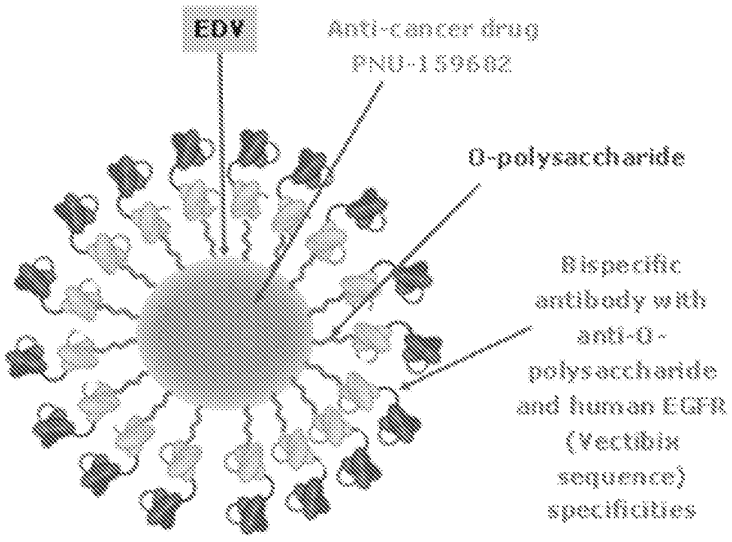

EDV
Anti-cancer drug
PNU-159682
O-polysaccharide
Bispecific
antibody with
anti-O-
polysaccharide
and human EGFR
(Vectibix
sequence)
specificities

FIG. 2

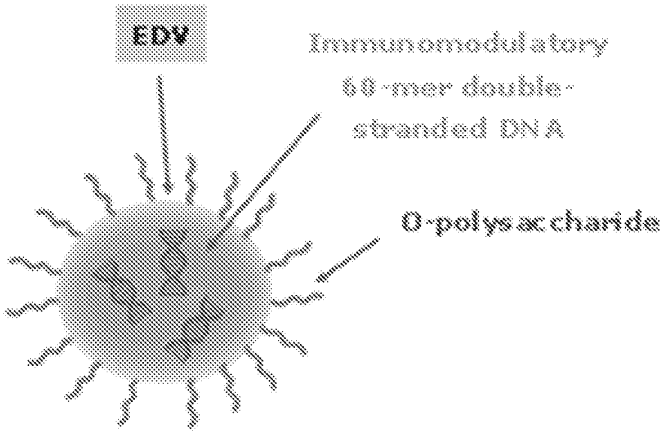

EDV
Immunomodulatory
60-mer double-
stranded DNA
O-polysaccharide

FIG. 3

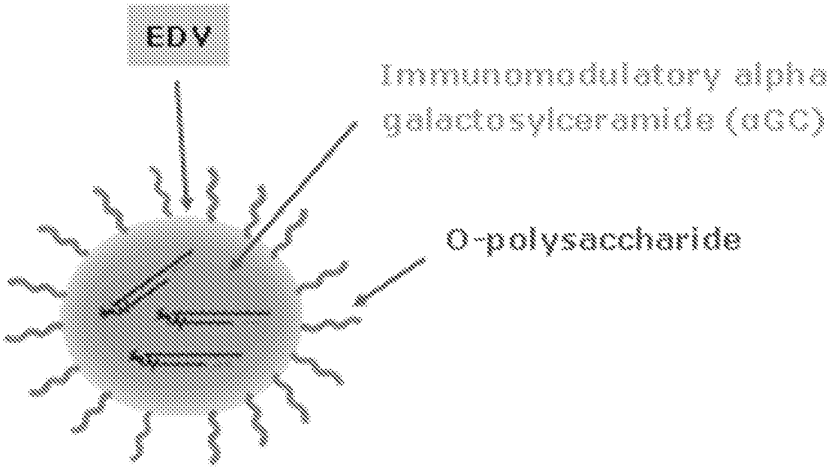

EDV
Immunomodulatory alpha galactosylceramide (αGC)
O-polysaccharide

FIG. 4

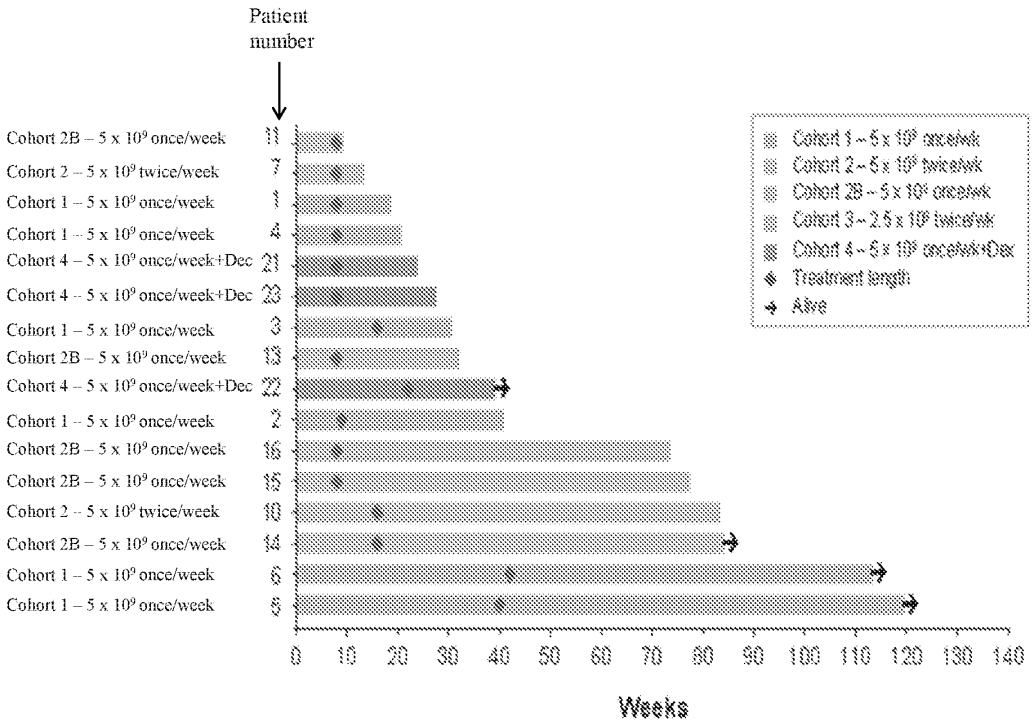

Patient number
Cohort 2B – 5 x 10⁹ once/week 11
Cohort 2 – 5 x 10⁹ twice/week 7
Cohort 1 – 5 x 10⁹ once/week 1
Cohort 1 – 5 x 10⁹ once/week 4
Cohort 4 – 5 x 10⁹ once/week+Dex 21
Cohort 4 – 5 x 10⁹ once/week+Dex 23
Cohort 1 – 5 x 10⁹ once/week 3
Cohort 2B – 5 x 10⁹ once/week 13
Cohort 4 – 5 x 10⁹ once/week+Dex 22
Cohort 1 – 5 x 10⁹ once/week 2
Cohort 2B – 5 x 10⁹ once/week 16
Cohort 2B – 5 x 10⁹ once/week 15
Cohort 2 – 5 x 10⁹ twice/week 10
Cohort 2B – 5 x 10⁹ once/week 14
Cohort 1 – 5 x 10⁹ once/week 6
Cohort 1 – 5 x 10⁹ once/week 5
Cohort 1 – 5 x 10⁹ once/wk
Cohort 2 – 5 x 10⁹ twice/wk
Cohort 2B – 5 x 10⁹ once/wk
Cohort 3 – 2.5 x 10⁹ twice/wk
Cohort 4 – 5 x 10⁹ once/wk+Dex
Treatment length
Alive
0 10 20 30 40 50 60 70 80 90 100 110 120 130 140
Weeks

FIG. 5

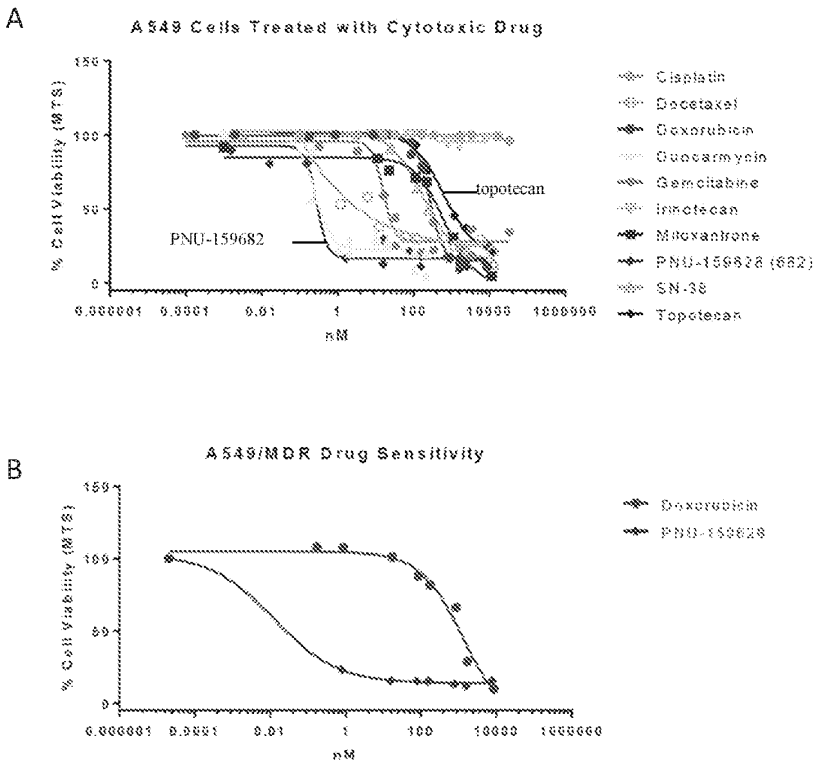

A
A549 Cells Treated with Cytotoxic Drug
% Cell Viability (MTS)
nM
topotecan
PNU-159682
Cisplatin
Docetaxel
Doxorubicin
Duocarmycin
Gemcitabine
Irinotecan
Mitoxantrone
PNU-159828 (682)
SN-38
Topotecan
B
A549/MDR Drug Sensitivity
% Cell Viability (MTS)
nM
Doxorubicin
PNU-159828

FIG. 6

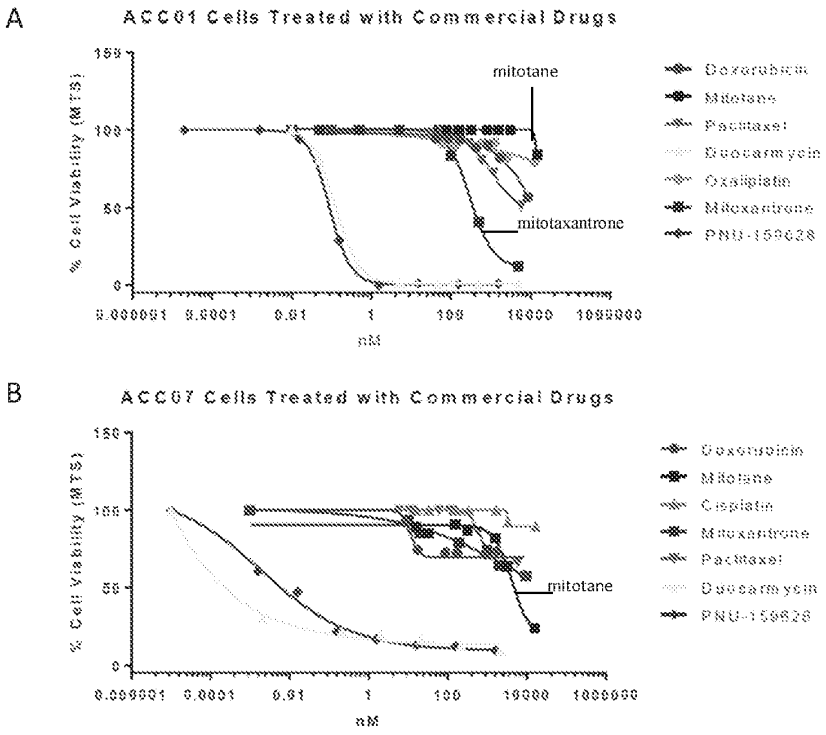

A
ACC01 Cells Treated with Commercial Drugs
% Cell Viability (MTS)
nM
mitotane
mitotaxantrone
Doxorubicin
Mitotane
Paclitaxel
Duocarmycin
Oxaliplatin
Mitoxantrone
PNU-159628
B
ACC07 Cells Treated with Commercial Drugs
% Cell Viability (MTS)
nM
mitotane
Doxorubicin
Mitotane
Cisplatin
Mitoxantrone
Paclitaxel
Duocarmycin
PNU-159628

FIG. 7

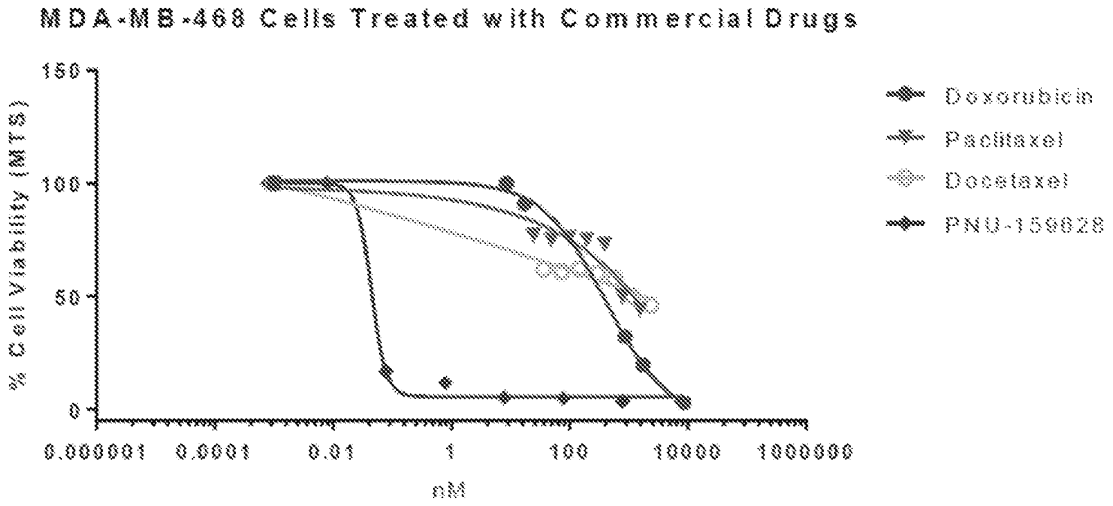

MDA-MB-468 Cells Treated with Commercial Drugs
% Cell Viability (MTS)
150
100
50
0
0.000001
0.0001
0.01
1
100
10000
1000000
nM
Doxorubicin
Paclitaxel
Docetaxel
PNU-159628

FIG. 8

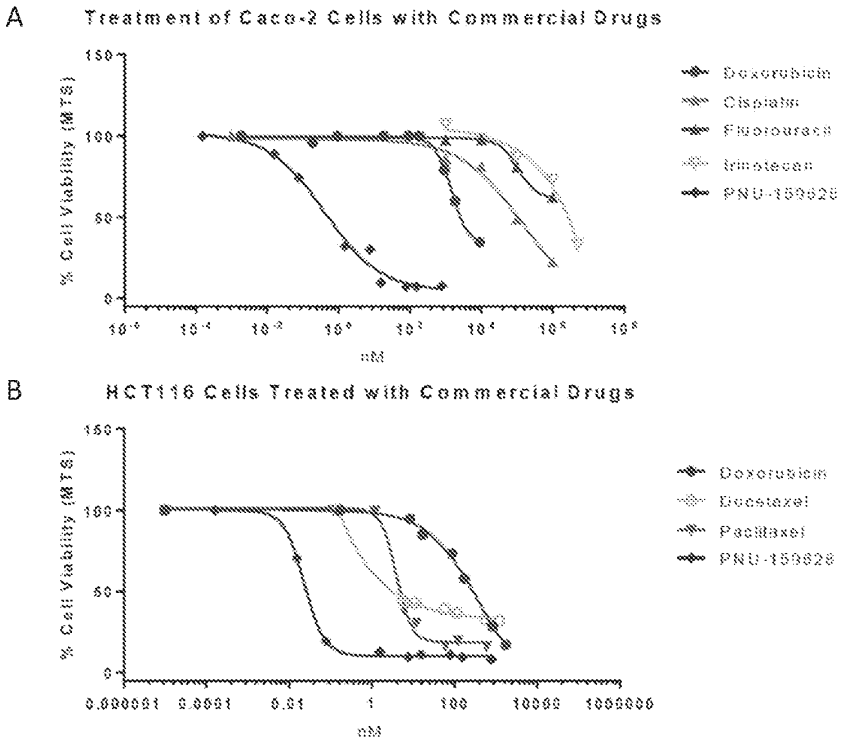

A
Treatment of Caco-2 Cells with Commercial Drugs
% Cell Viability (MTS)
150
100
50
0
nM
Doxorubicin
Cisplatin
Fluorouracil
Irinotecan
PNU-159626
B
HCT116 Cells Treated with Commercial Drugs
% Cell Viability (MTS)
150
100
50
0
0.000001
0.0001
0.01
1
100
10000
1000000
nM
Doxorubicin
Docetaxel
Paclitaxel
PNU-159626

FIG. 9

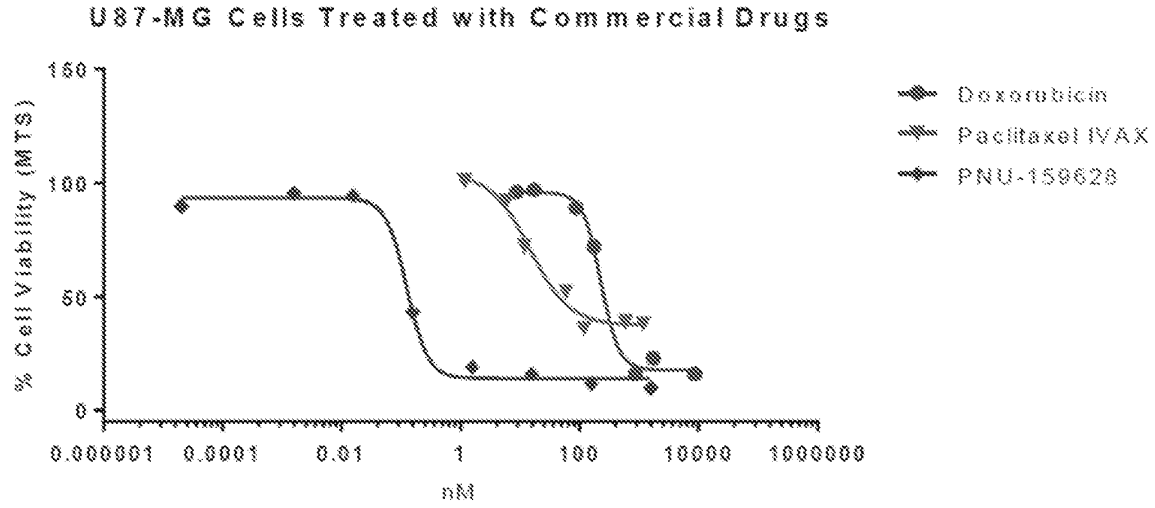

U87-MG Cells Treated with Commercial Drugs
% Cell Viability (MTS)
150
100
50
0
0.000001
0.0001
0.01
1
100
10000
1000000
nM
Doxorubicin
Paclitaxel IVAX
PNU-159628

FIG. 10

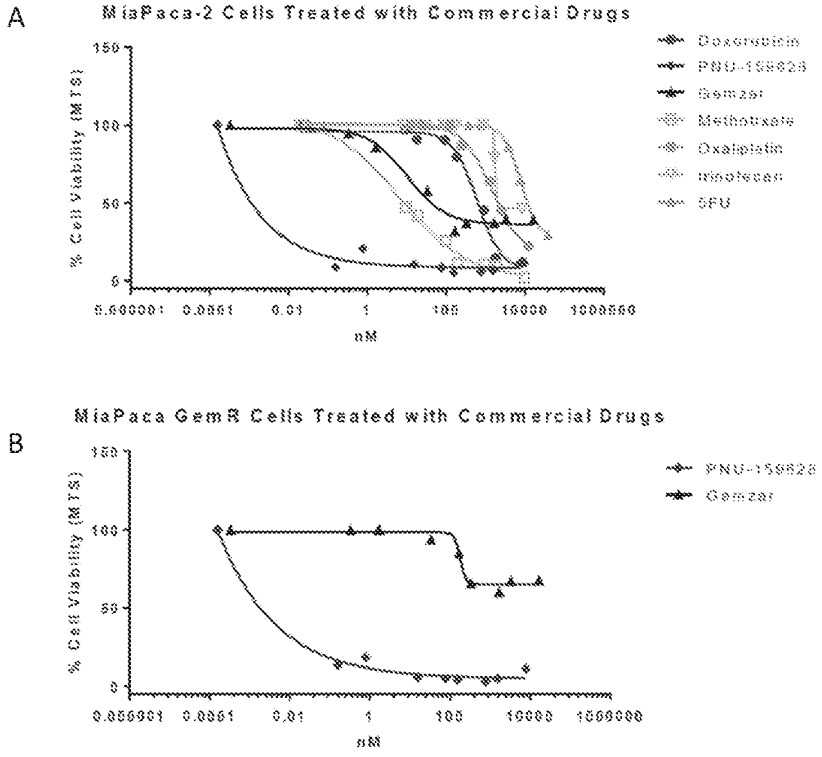

A
MiaPaca-2 Cells Treated with Commercial Drugs
% Cell Viability (MTS)
150
100
50
0
0.000001
0.0001
0.01
1
100
10000
1000000
nM
Doxorubicin
PNU-159682
Gemzar
Methotrexate
Oxaliplatin
Irinotecan
5FU
B
MiaPaca GemR Cells Treated with Commercial Drugs
% Cell Viability (MTS)
150
100
50
0
0.000001
0.0001
0.01
1
100
10000
1000000
nM
PNU-159682
Gemzar

FIG. 11

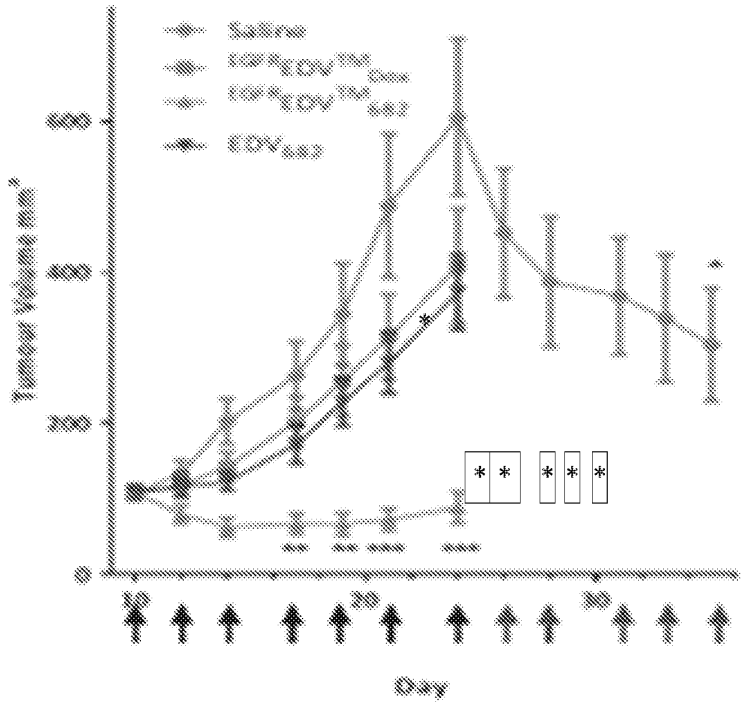


FIG. 12

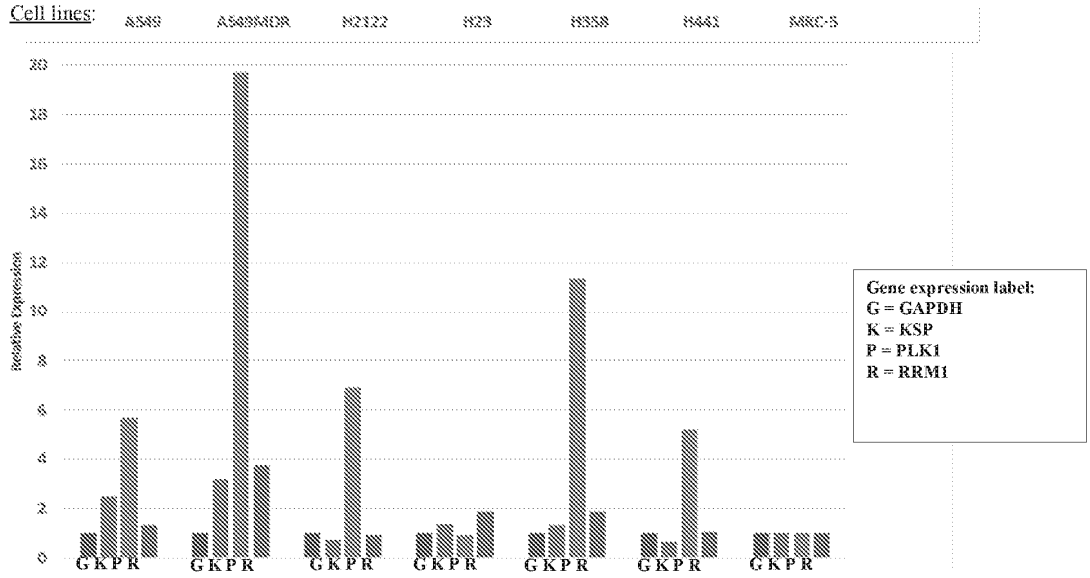

Cell lines:
A549
A549/DDP
H2122
H23
H358
H441
MRC-5
Relative Expression
0
2
4
6
8
10
12
14
16
18
20
G K P R
Gene expression label:
G = GAPDH
K = KSP
P = PLK1
R = RRM1

FIG. 13

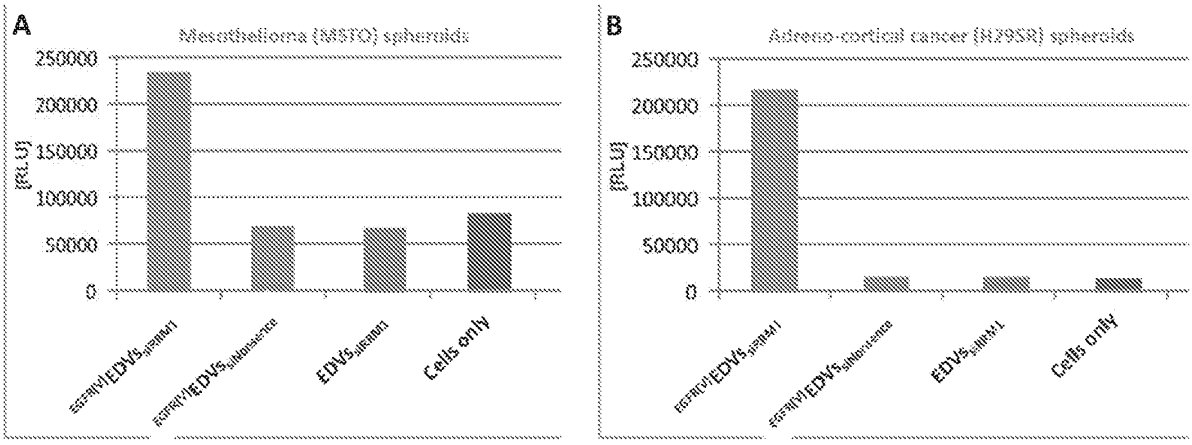
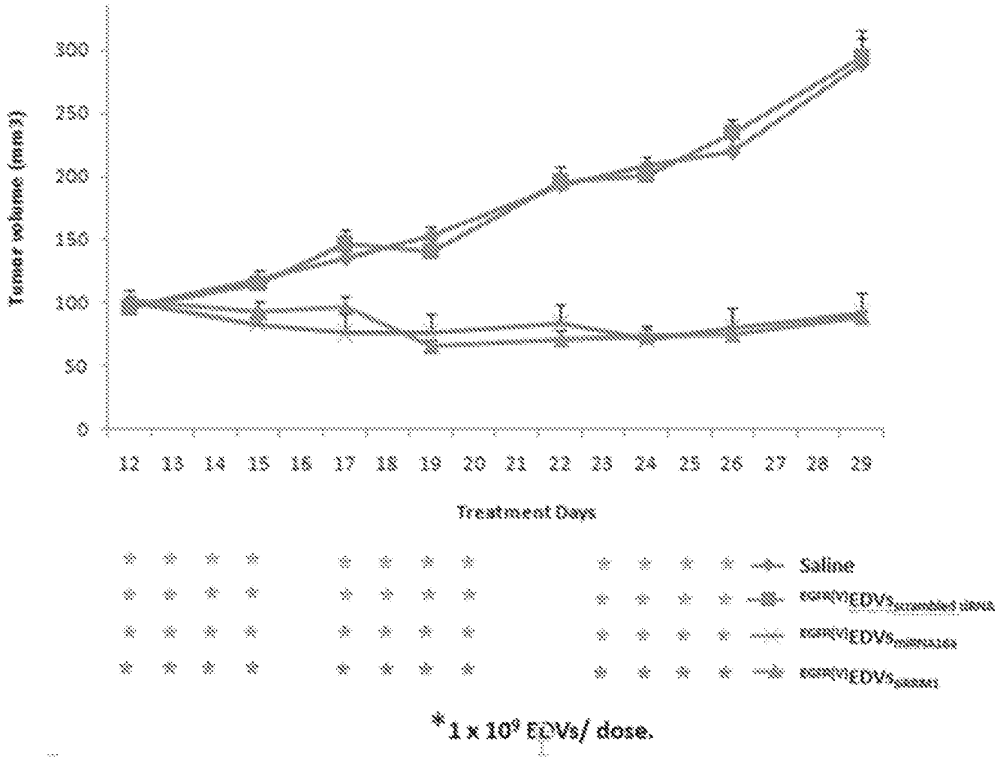

A
Mesothelioma (MSTO) spheroids
250000
200000
150000
100000
50000
0
[RLU]
Cells only
B
Adreno-cortical cancer (H295R) spheroids
250000
200000
150000
100000
50000
0
[RLU]
Cells only

FIG. 14



*1 x 10⁹ EDVs/ dose.

FIG. 15

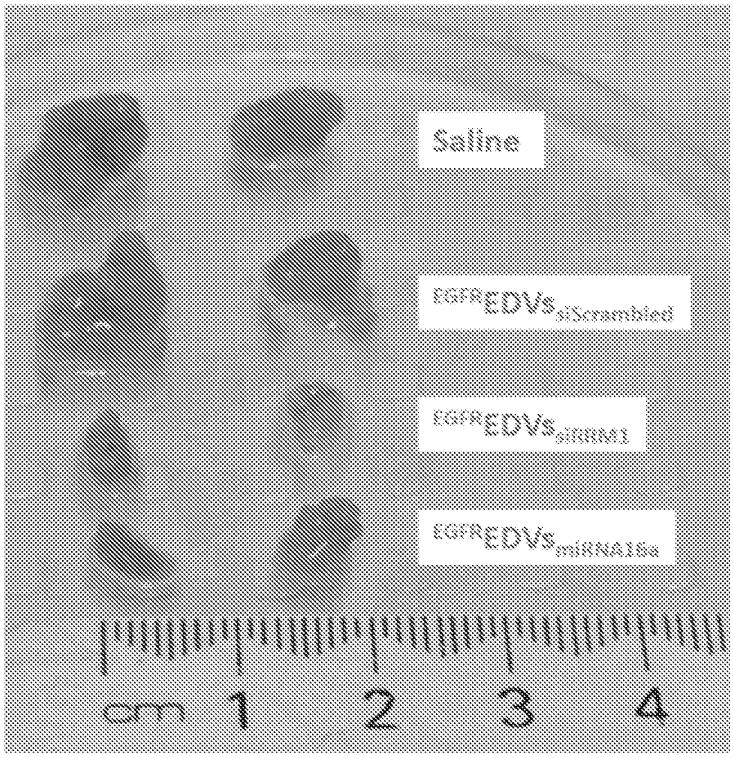

Saline
EGFREDVssiScrambled
EGFREDVssiRRM1
EGFREDVsmiRNA18a
cm 1 2 3 4

FIG. 16

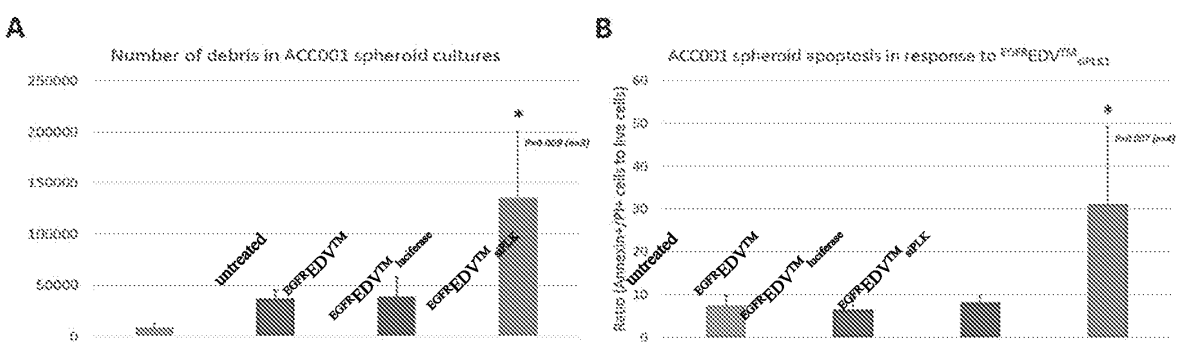

A
Number of debris in ACC001 spheroid cultures
250000
200000
150000
100000
50000
0
untreated
EGFREDVTM
EGFREDVTM luciferase
EGFREDVTM siPLK
*
B
ACC001 spheroid apoptosis in response to EGFREDVTM siPLK
Ratio (Annexin V/PI+ cells to live cells)
60
50
40
30
20
10
0
untreated
EGFREDVTM
EGFREDVTM luciferase
EGFREDVTM siPLK
*

FIG. 17

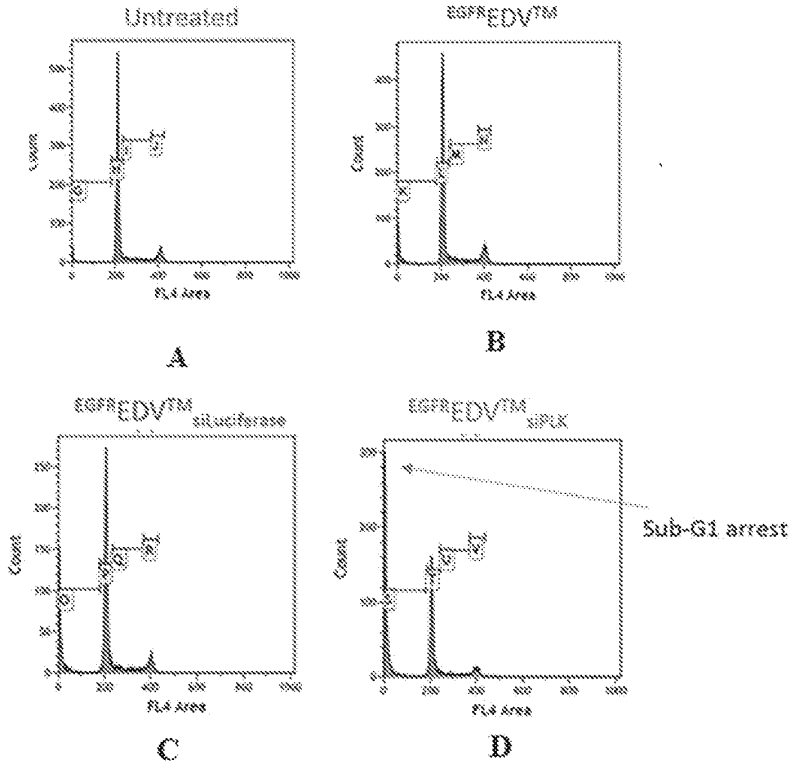

Untreated
EGFREDV™
A
B
EGFREDV™ siLuciferase
EGFREDV™ siPLK
Sub-G1 arrest
C
D

FIG. 18

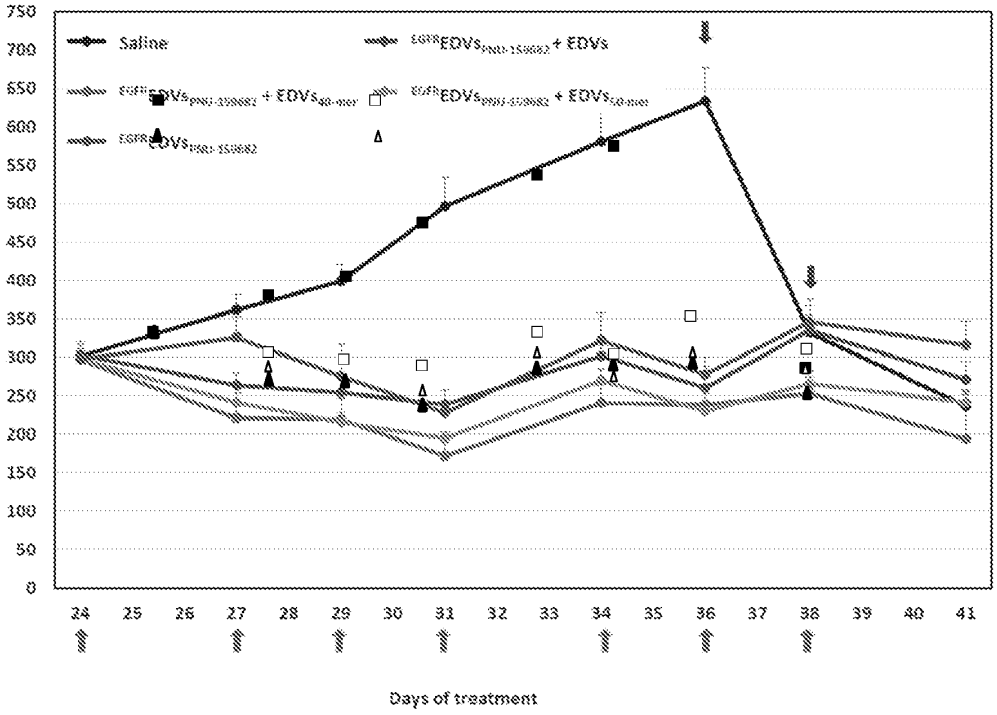

750
700
650
600
550
500
450
400
350
300
250
200
150
100
50
0
24 25 26 27 28 29 30 31 32 33 34 35 36 37 38 39 40 41
Saline
Days of treatment

FIG. 19

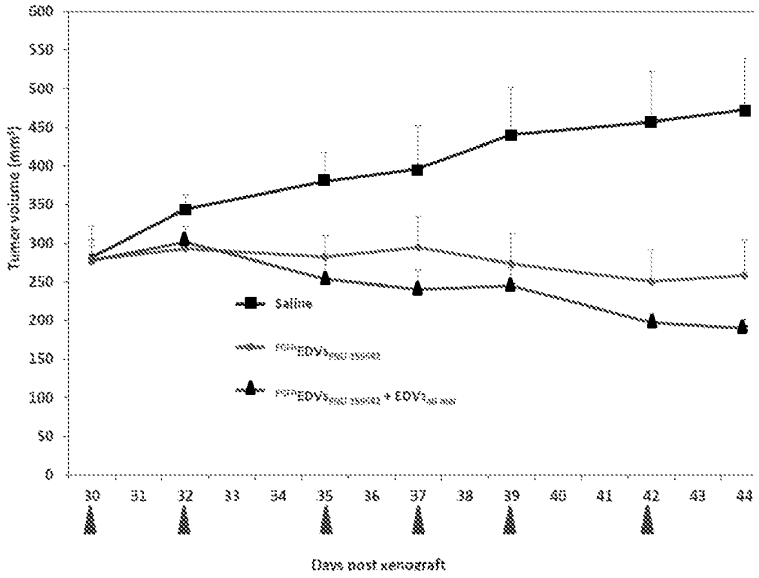

600
550
500
450
400
350
300
250
200
150
100
50
0
Tumor volume (mm³)
30
31
32
33
34
35
36
37
38
39
40
41
42
43
44
Days post xenograft
Saline

FIG. 20

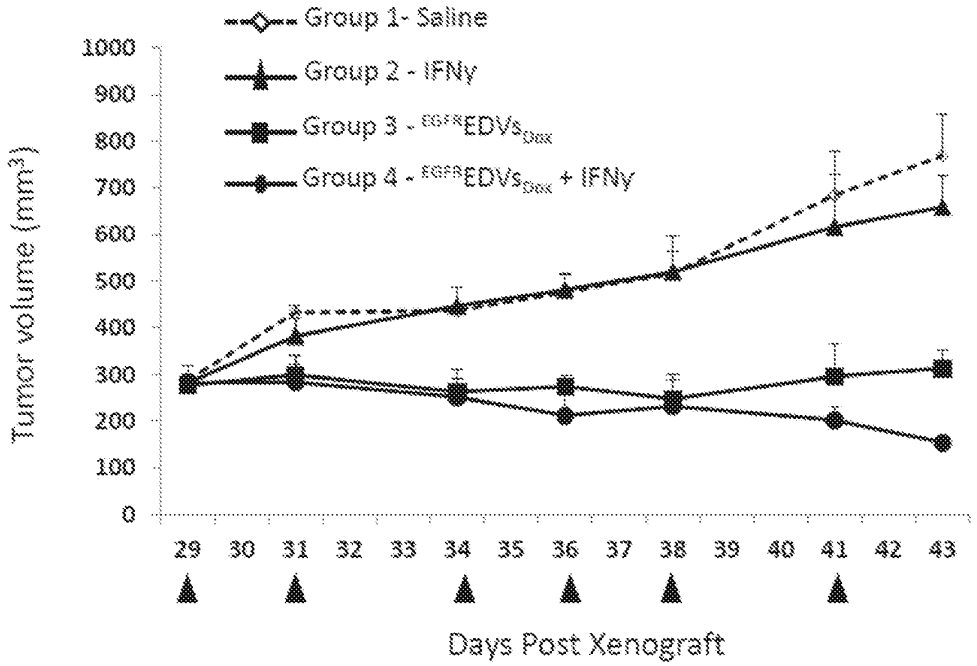

Group 1- Saline
Group 2 - IFNγ
Group 3 - EGFREDVsDox
Group 4 - EGFREDVsDox + IFNγ
Tumor volume (mm3)
1000
900
800
700
600
500
400
300
200
100
0
29 30 31 32 33 34 35 36 37 38 39 40 41 42 43
Days Post Xenograft

FIG. 21

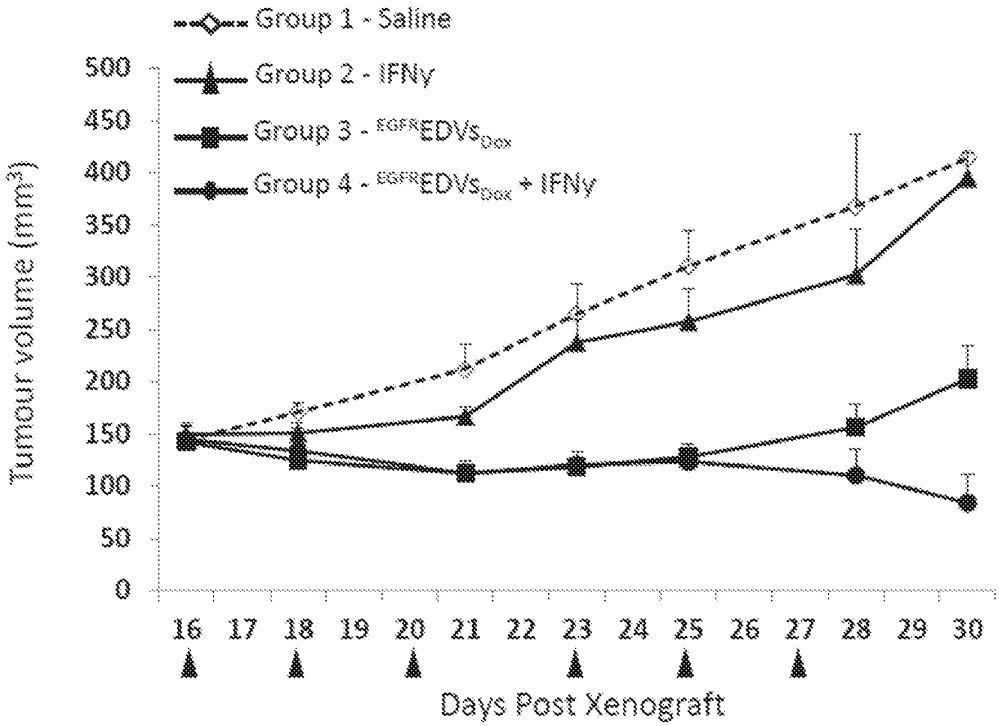

Group 1 - Saline
Group 2 - IFNγ
Group 3 - EGFREDVsDox
Group 4 - EGFREDVsDox + IFNγ
Tumour volume (mm3)
500
450
400
350
300
250
200
150
100
50
0
16 17 18 19 20 21 22 23 24 25 26 27 28 29 30
Days Post Xenograft

FIG. 22

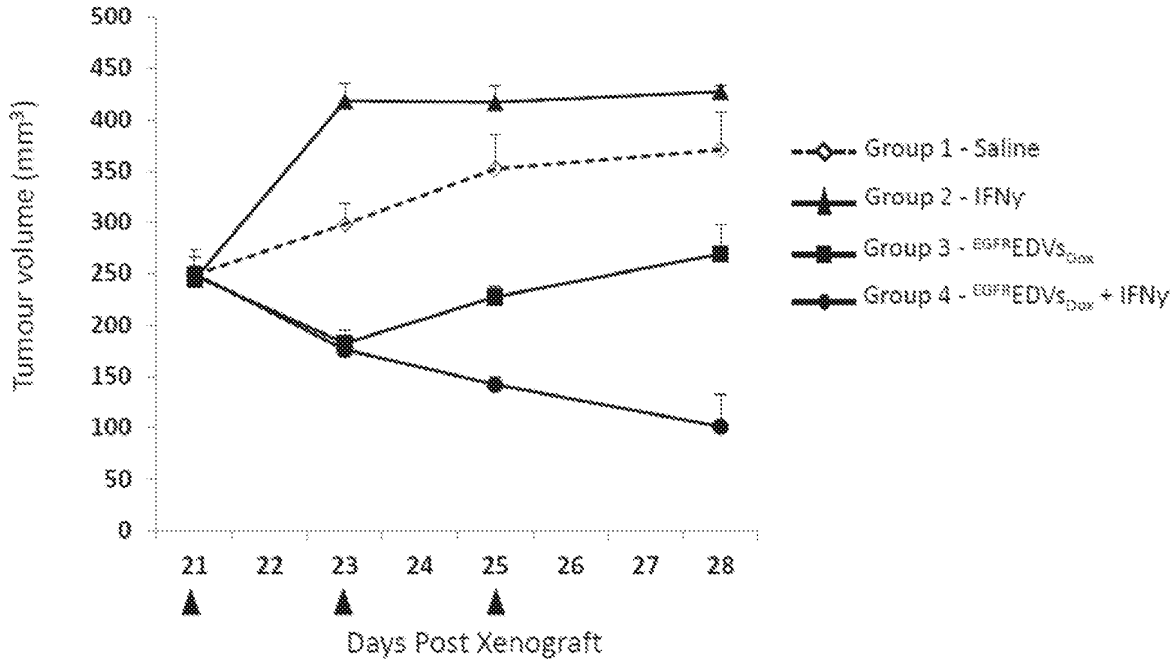

Tumour volume (mm³)
500
450
400
350
300
250
200
150
100
50
0
21
22
23
24
25
26
27
28
Days Post Xenograft
Group 1 - Saline
Group 2 - IFNγ
Group 3 - EGFREDVs Dox
Group 4 - EGFREDVs Dox + IFNγ

FIG. 23

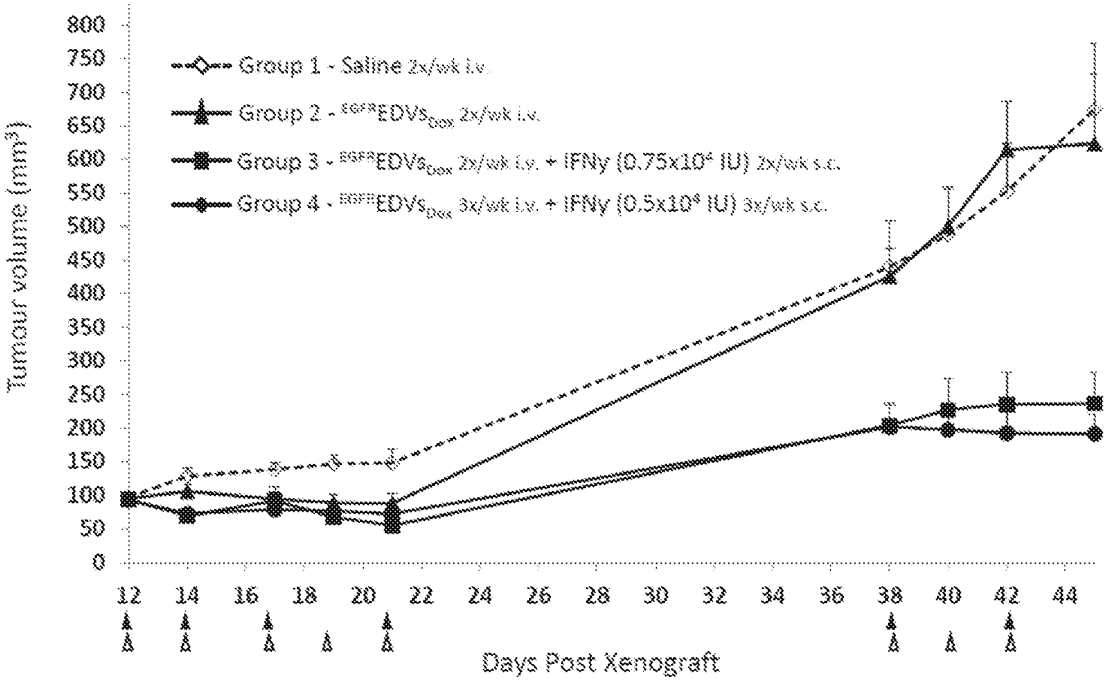

Tumour volume (mm³)
800
750
700
650
600
550
500
450
400
350
300
250
200
150
100
50
0
Group 1 - Saline 2x/wk i.v.
Group 2 - EGFREDVsDox 2x/wk i.v.
Group 3 - EGFREDVsDox 2x/wk i.v. + IFNγ (0.75x10⁴ IU) 2x/wk s.c.
Group 4 - EGFREDVsDox 3x/wk i.v. + IFNγ (0.5x10⁴ IU) 3x/wk s.c.
12 14 16 18 20 22 24 26 28 30 32 34 36 38 40 42 44
Days Post Xenograft

FIG. 24

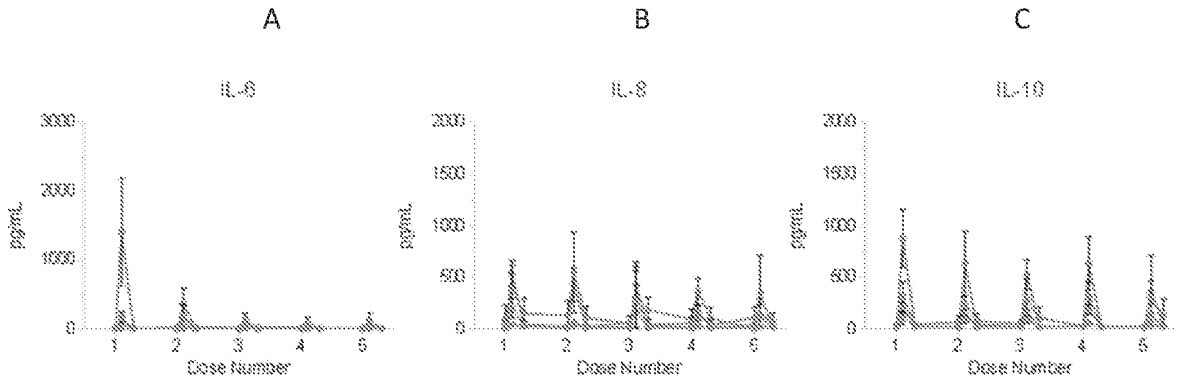

A
IL-6
pg/mL
3000
2000
1000
0
1
2
3
4
5
Dose Number
B
IL-8
pg/mL
2000
1500
1000
500
0
1
2
3
4
5
Dose Number
C
IL-10
pg/mL
2000
1500
1000
500
0
1
2
3
4
5
Dose Number

FIG. 24
(Continued)

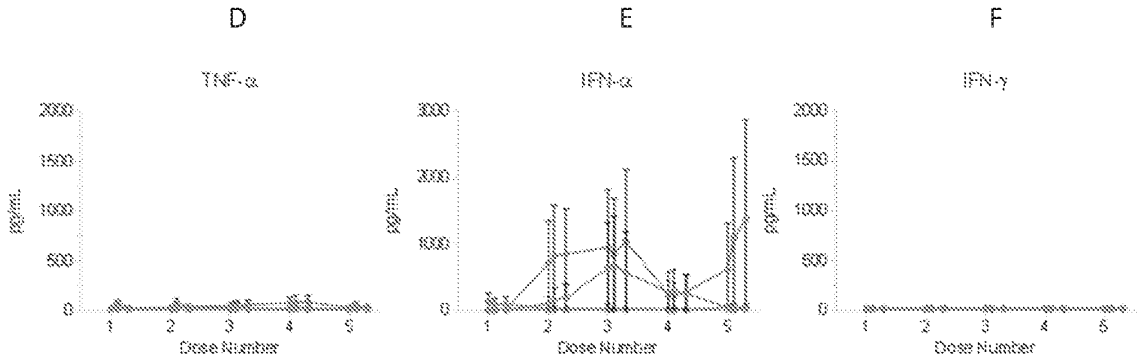


FIG. 24
(Continued)

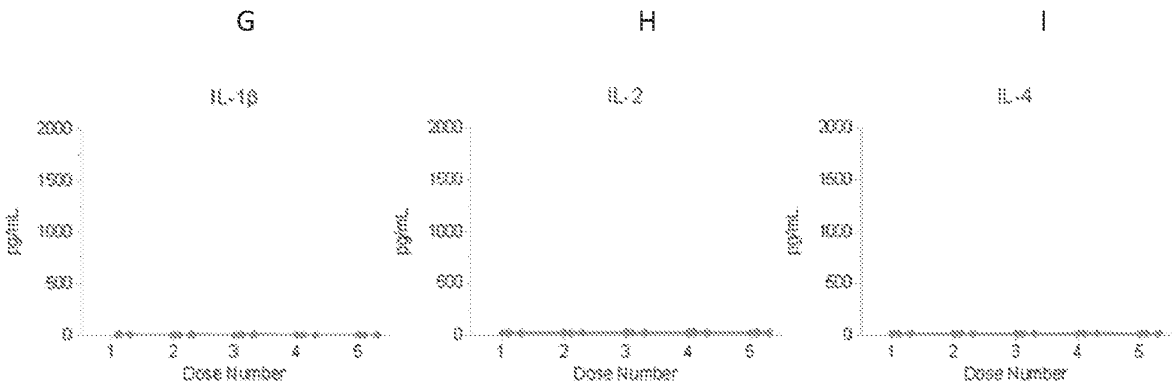


FIG. 24
(Continued)

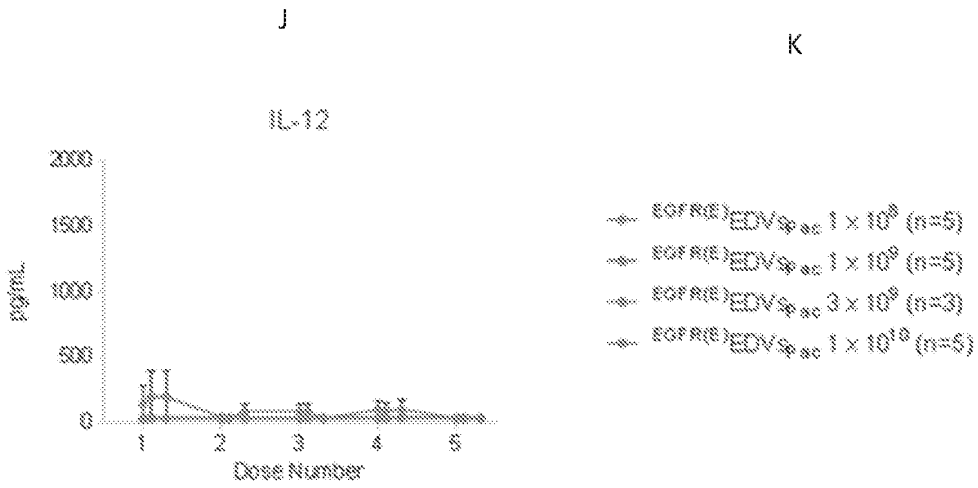


FIG. 25

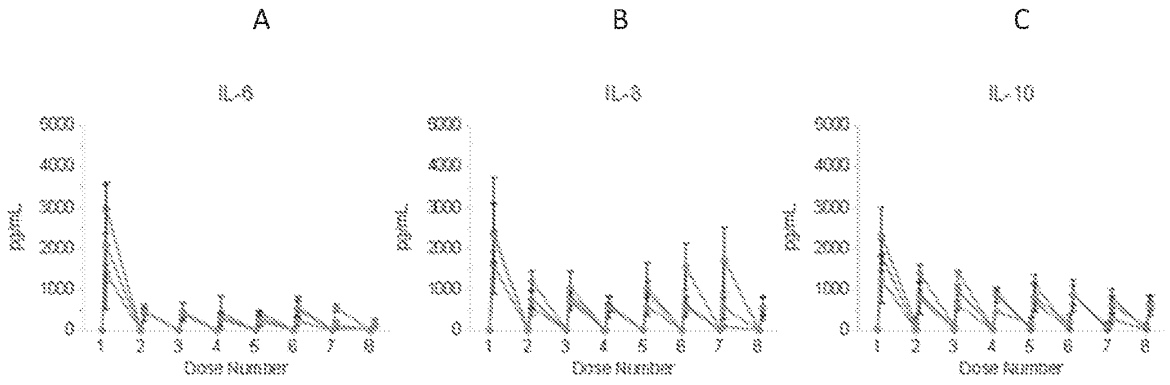

A
IL-6
pg/mL
5000
4000
3000
2000
1000
0
1 2 3 4 5 6 7 8
Dose Number
B
IL-8
pg/mL
5000
4000
3000
2000
1000
0
1 2 3 4 5 6 7 8
Dose Number
C
IL-10
pg/mL
5000
4000
3000
2000
1000
0
1 2 3 4 5 6 7 8
Dose Number

FIG. 25
(Continued)

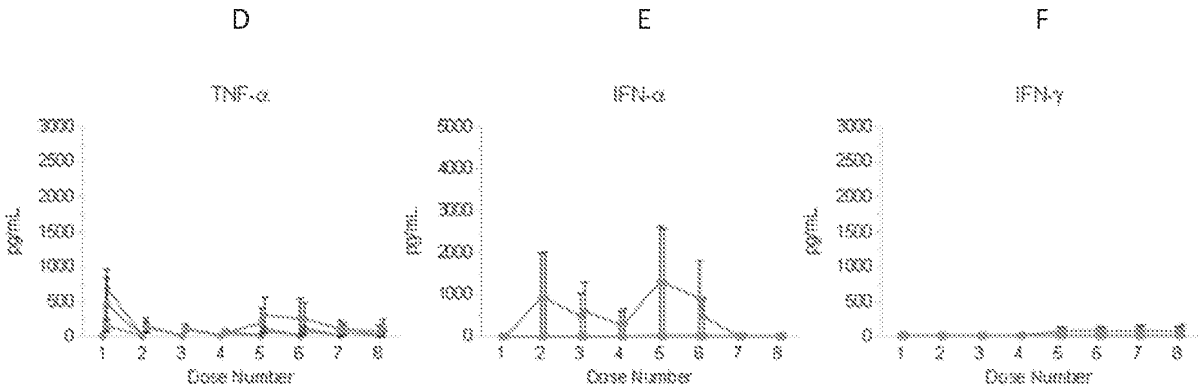


FIG. 25
(Continued)

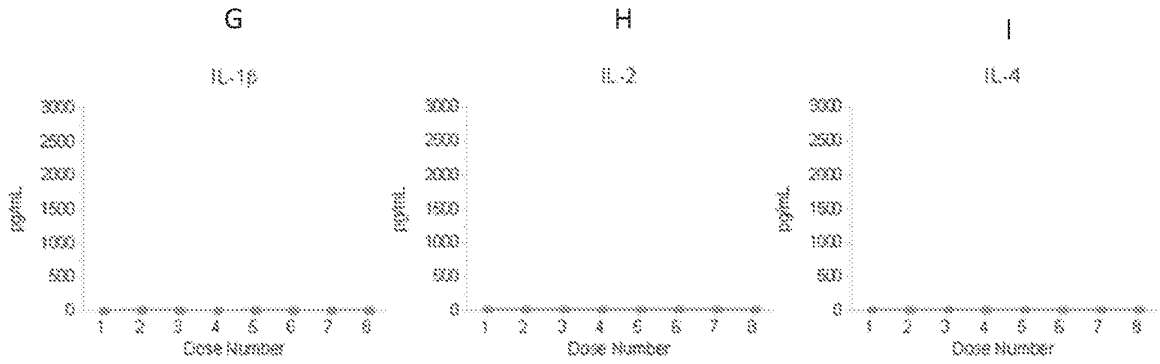


FIG. 25
(Continued)

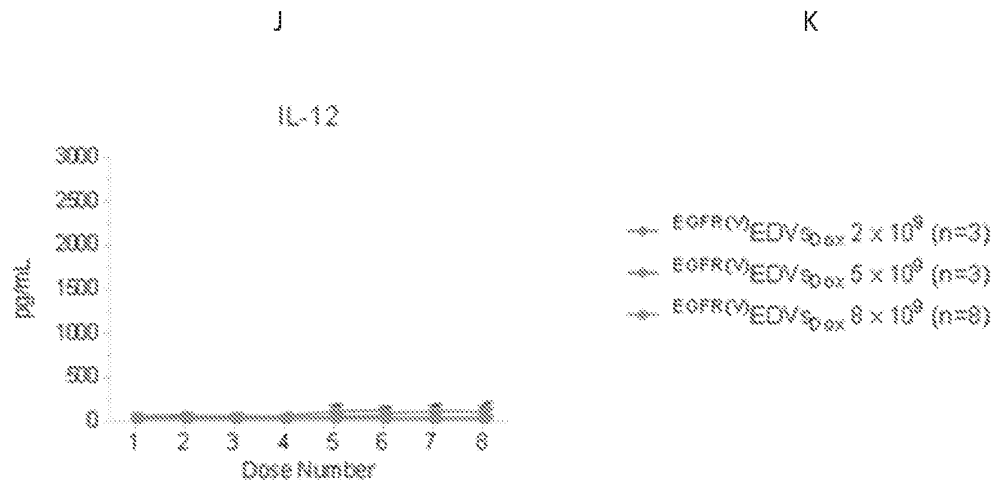


FIG. 26

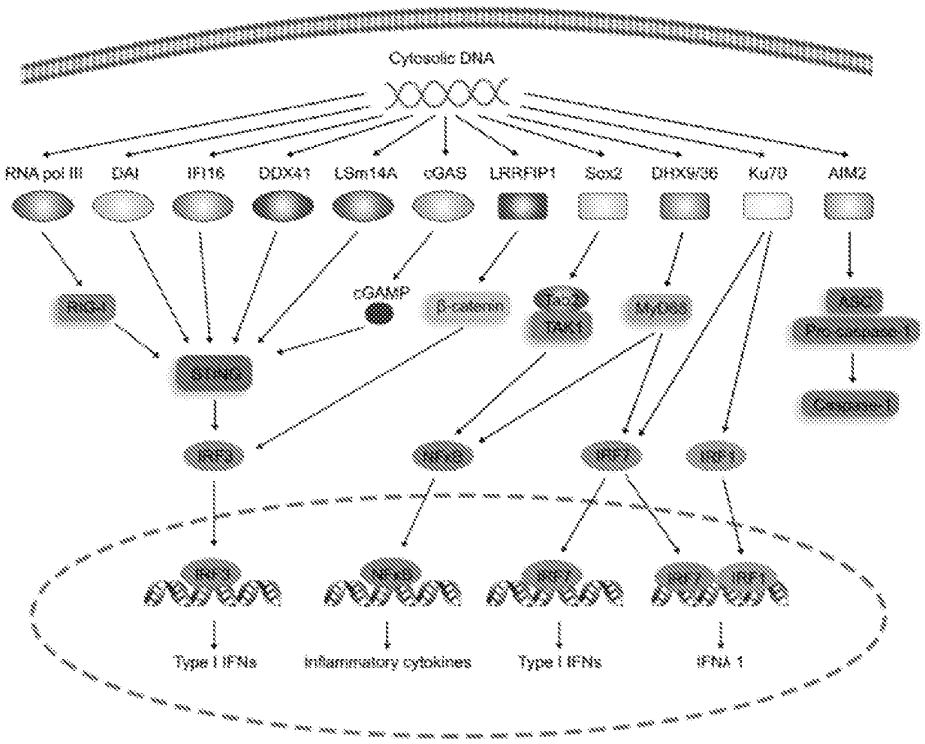

Cytosolic DNA
RNA pol III
DAI
IFI16
DDX41
LSm14A
cGAS
LRRFIP1
Sox2
DHX9/36
Ku70
AIM2
cGAMP
STING
IRF3
IRF7
IRF1
Type I IFNs
Inflammatory cytokines
Type I IFNs
IFNA 1

FIG. 27

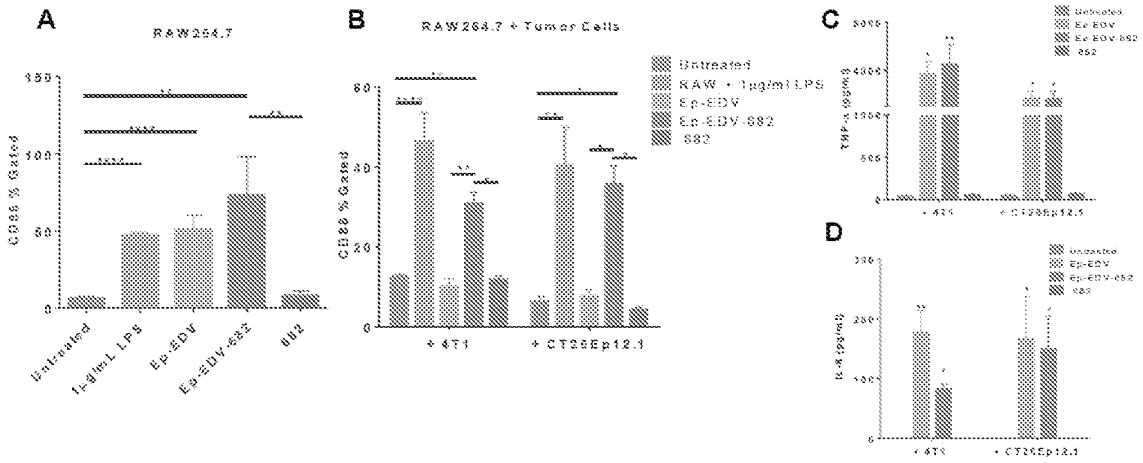

A
RAW264.7
CD86 % Gated
Untreated
1μg/ml LPS
Ep-EDV
Ep-EDV-882
882
B
RAW264.7 + Tumor Cells
CD80 % Gated
Untreated
RAW + 1μg/ml LPS
Ep-EDV
Ep-EDV-882
882
+ 4T1
+ CT26Ep12.1
C
TNF-α (pg/mL)
+ 4T1
+ CT26Ep12.1
D
IL-6 (pg/mL)
+ 4T1
+ CT26Ep12.1

FIG. 27
(Continued)

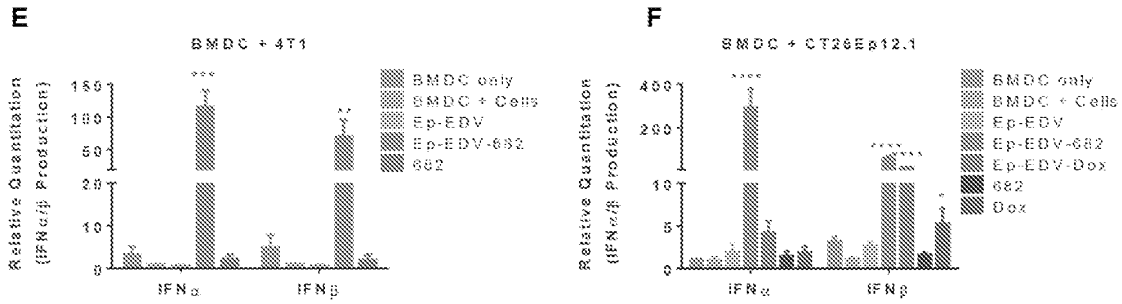


FIG. 27
(Continued)

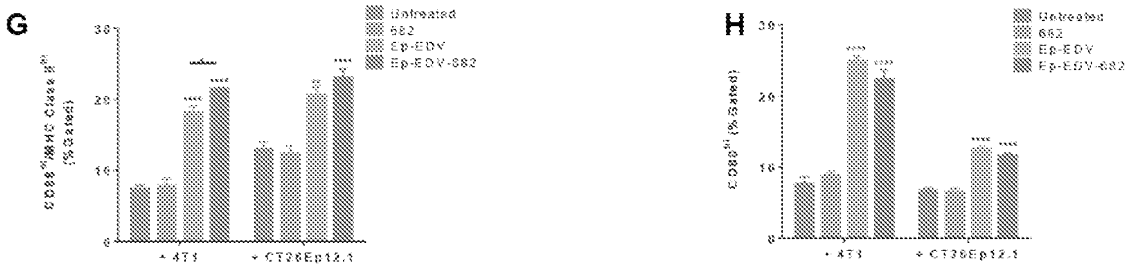


FIG. 27
(Continued)

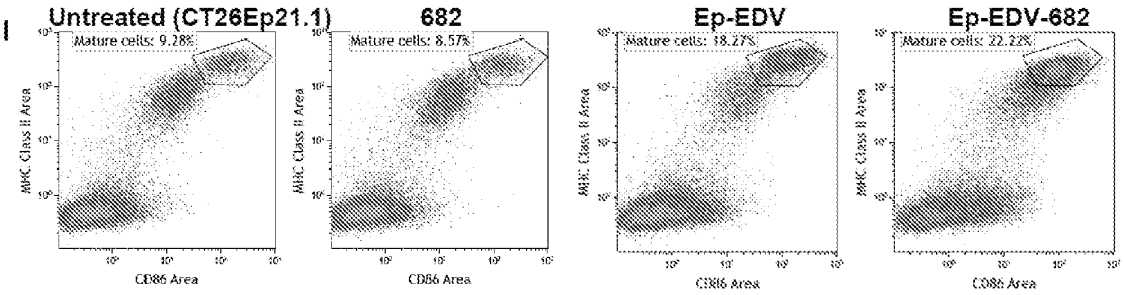


FIG. 27
(Continued)

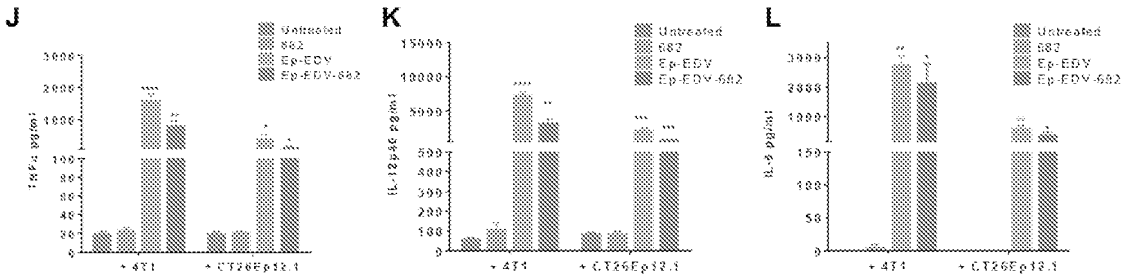


FIG. 28

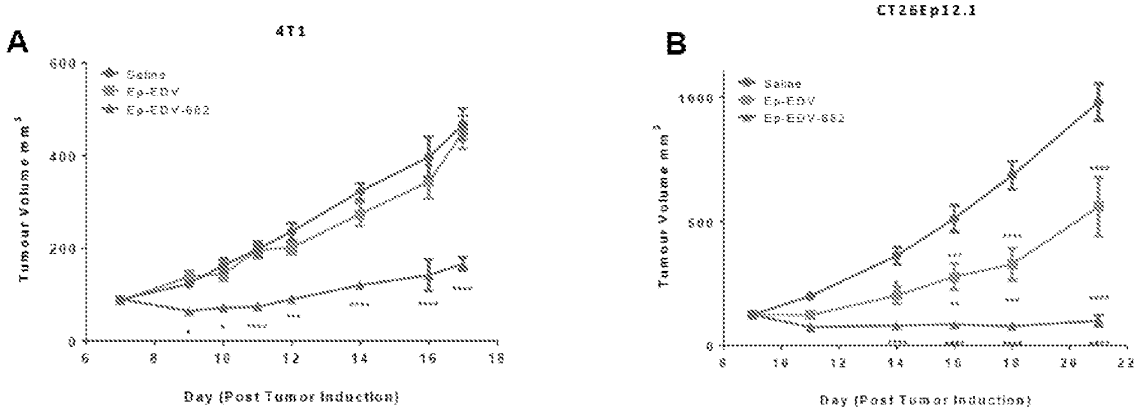

A
4T1
Saline
Ep-EDV
Ep-EDV-662
Tumour Volume mm³
Day (Post Tumor Induction)
B
CT26Ep12.1
Saline
Ep-EDV
Ep-EDV-662
Tumour Volume mm³
Day (Post Tumor Induction)

FIG. 28
(Continued)

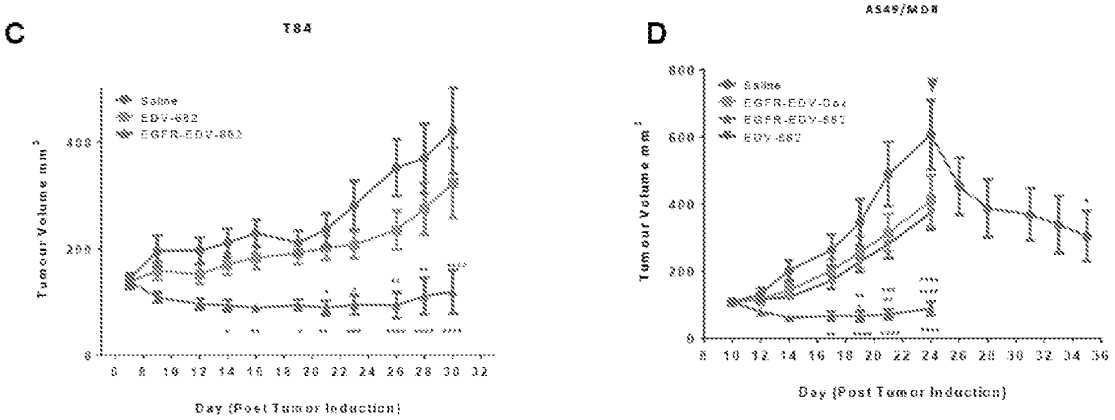


FIG. 28
(Continued)

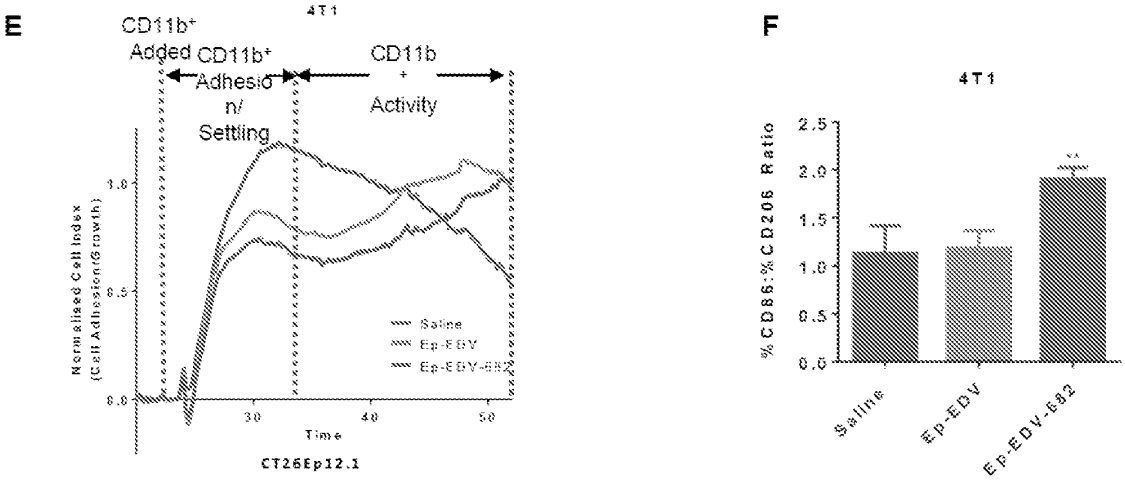
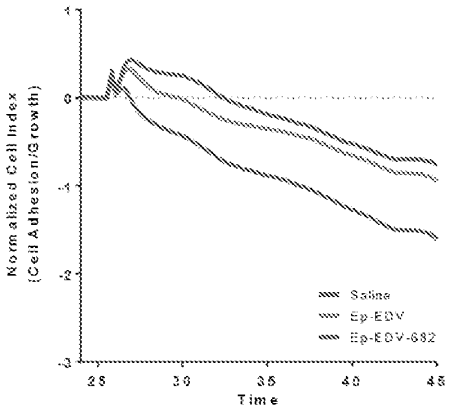


FIG. 28
(Continued)

G



H

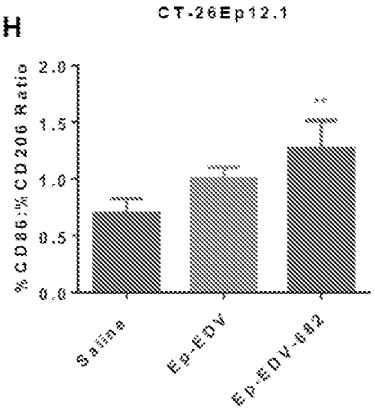


FIG. 29

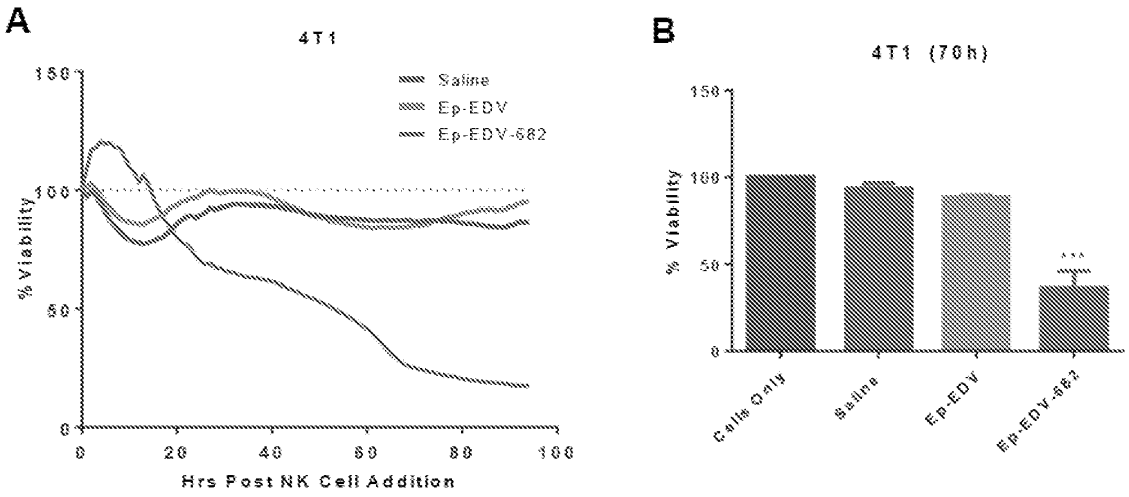

A
4T1
Saline
Ep-EDV
Ep-EDV-682
% Viability
150
100
50
0
0
20
40
60
80
100
Hrs Post NK Cell Addition
B
4T1 (70h)
% Viability
150
100
50
0
***
Cells Only
Saline
Ep-EDV
Ep-EDV-682

FIG. 29
(Continued)

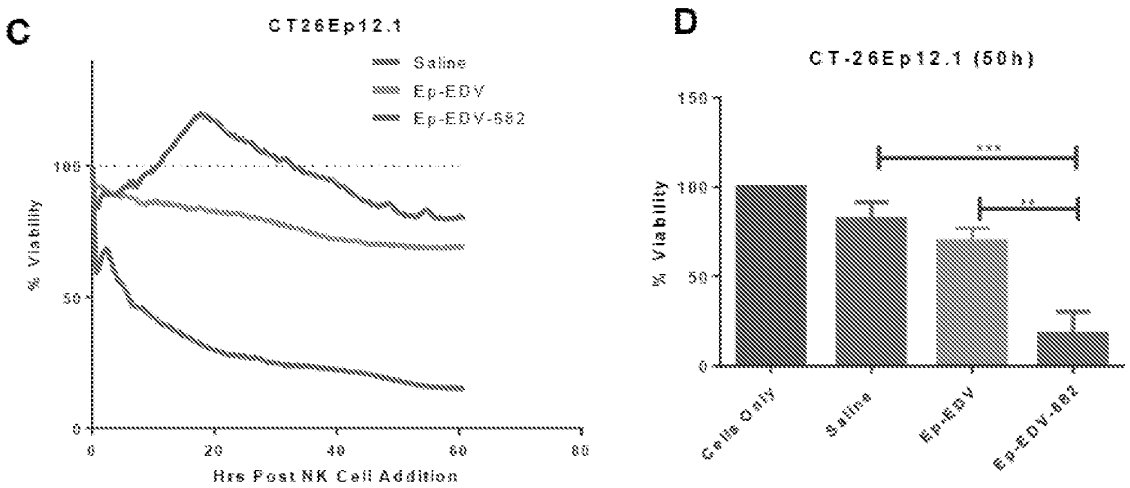


FIG. 29
(Continued)

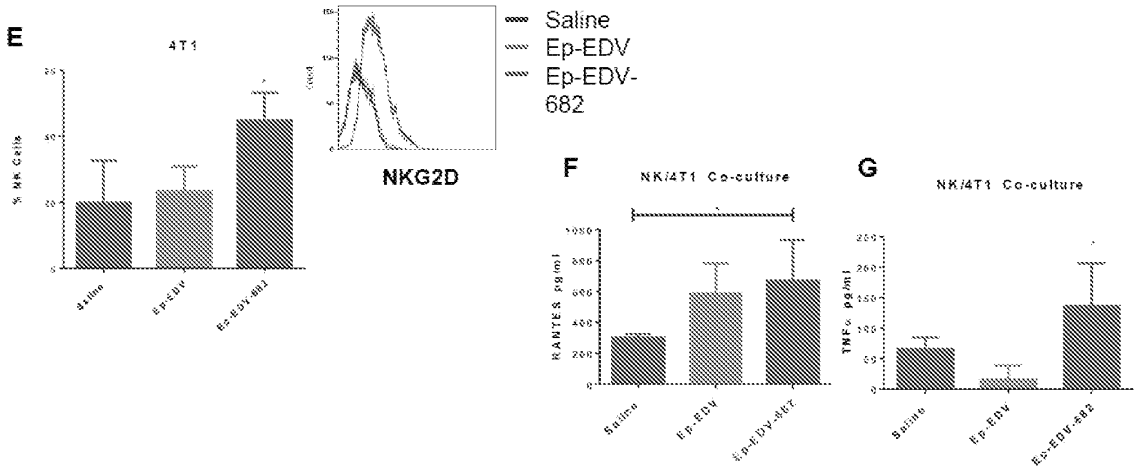


FIG. 29
(Continued)

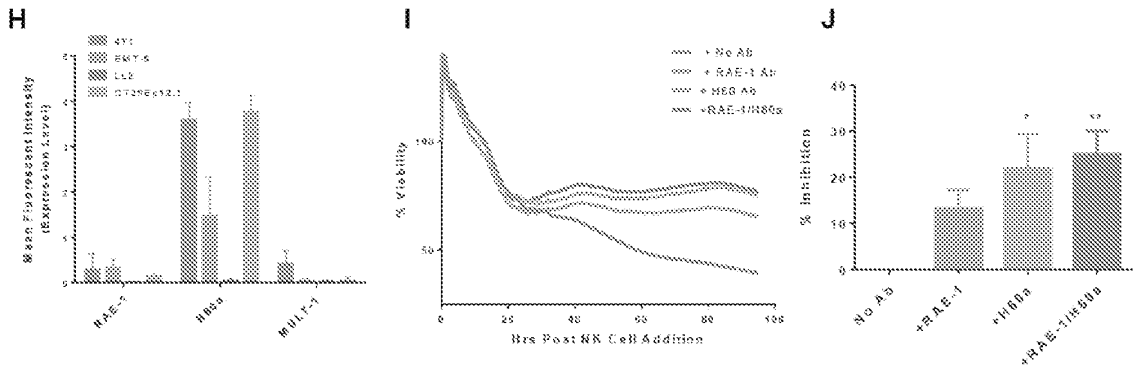


FIG. 30

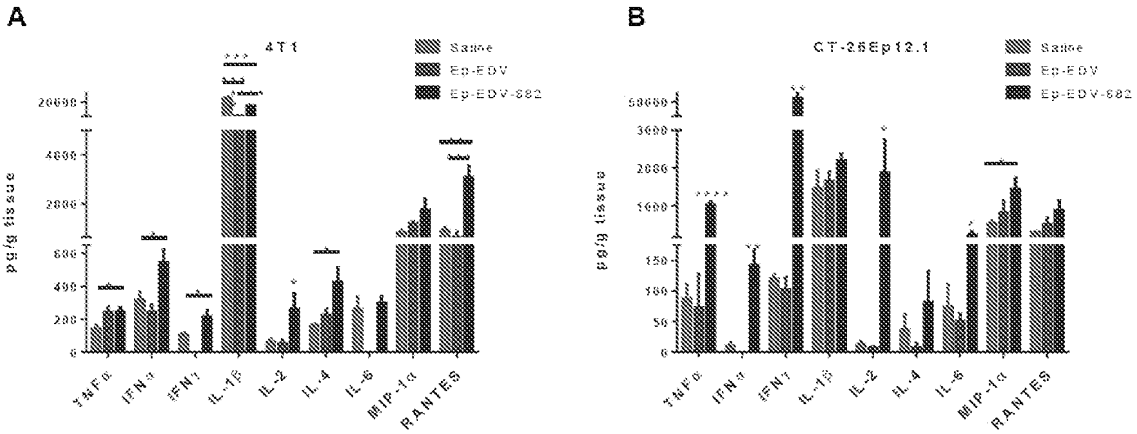

A
4T1
Same
Ep-EDV
Ep-EDV-882
pg/g tissue
20000
4000
2000
800
400
200
0
TNFα
IFNα
sFNT
IL-1β
IL-2
IL-4
IL-6
MIP-1α
RANTES
B
CT-26Ep12.1
Same
Ep-EDV
Ep-EDV-882
pg/g tissue
50000
3000
2000
1000
150
100
50
0
TNFα
IFNα
sFNT
IL-1β
IL-2
IL-4
IL-6
MIP-1α
RANTES

FIG. 30
(Continued)

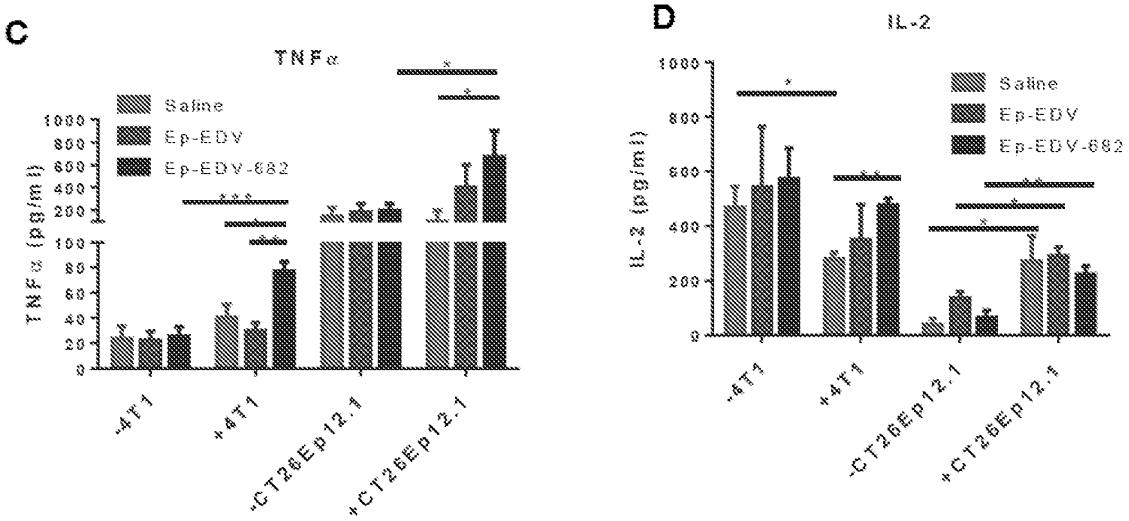


FIG. 30
(Continued)

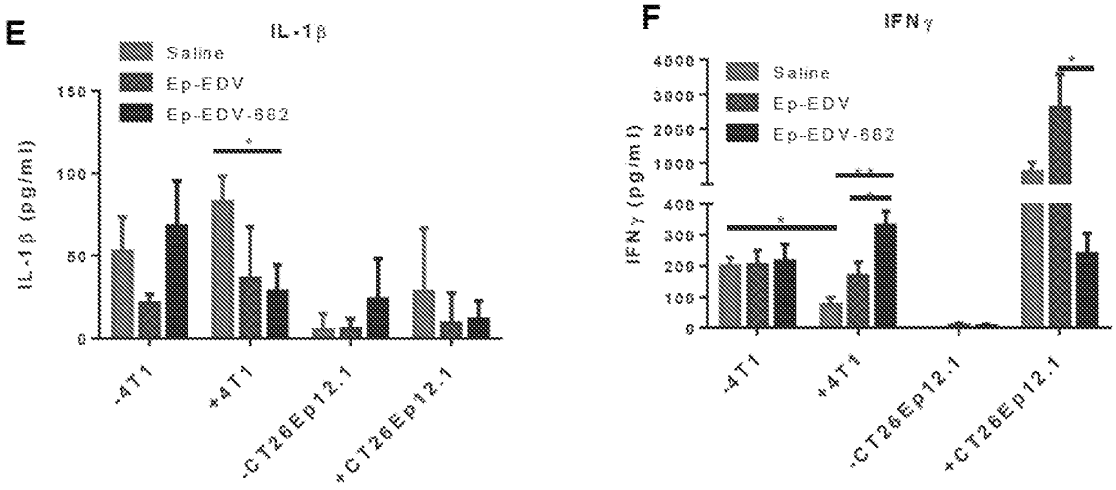


FIG. 30
(Continued)

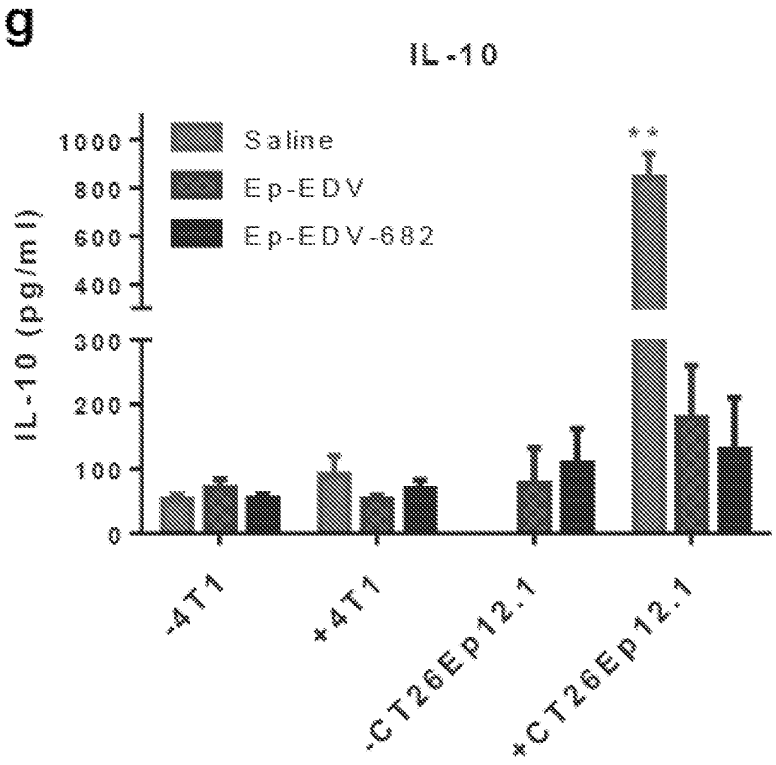


FIG. 31

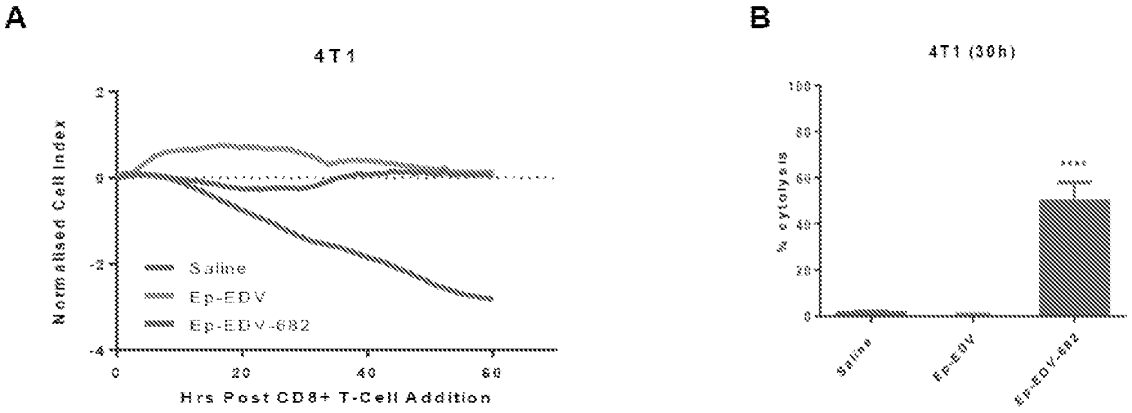

A
4T1
Normalised Cell Index
Saline
Ep-EDV
Ep-EDV-682
Hrs Post CD8+ T-Cell Addition
B
4T1 (30h)
% cytolysis
Saline
Ep-EDV
Ep-EDV-682

FIG. 31
(Continued)

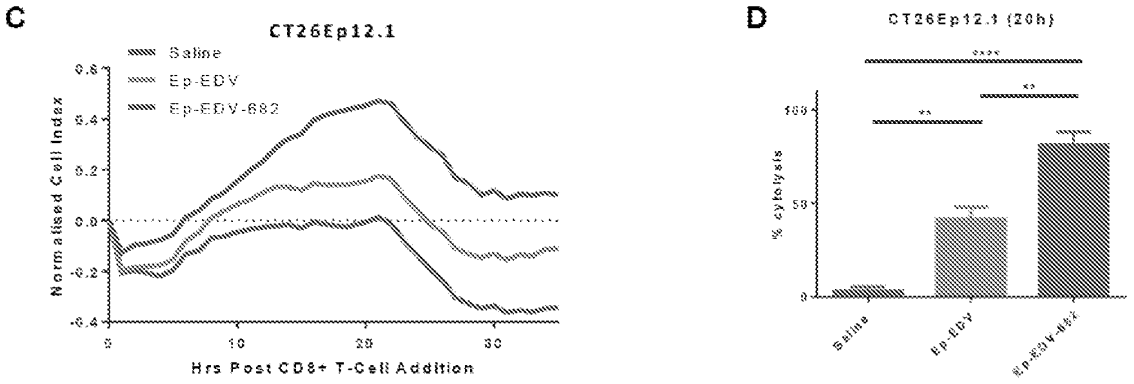


FIG. 31
(Continued)

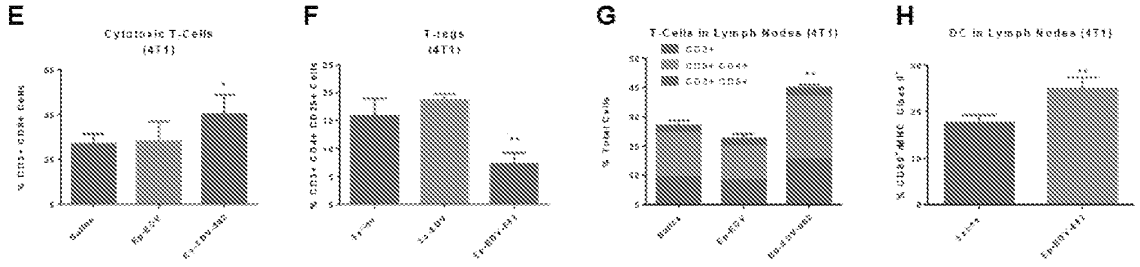


FIG. 31
(Continued)

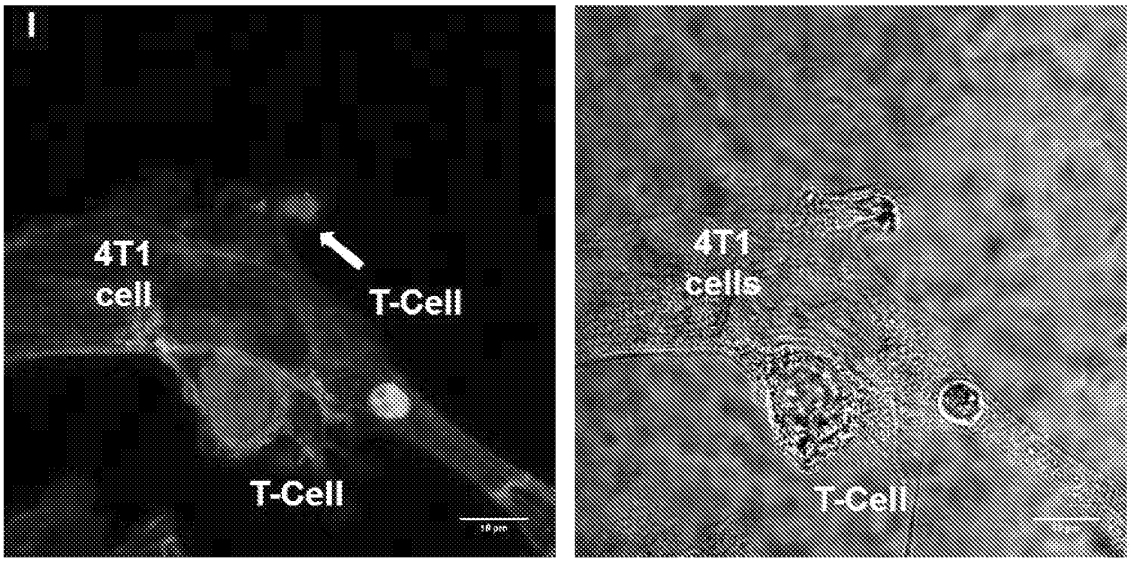


FIG. 32

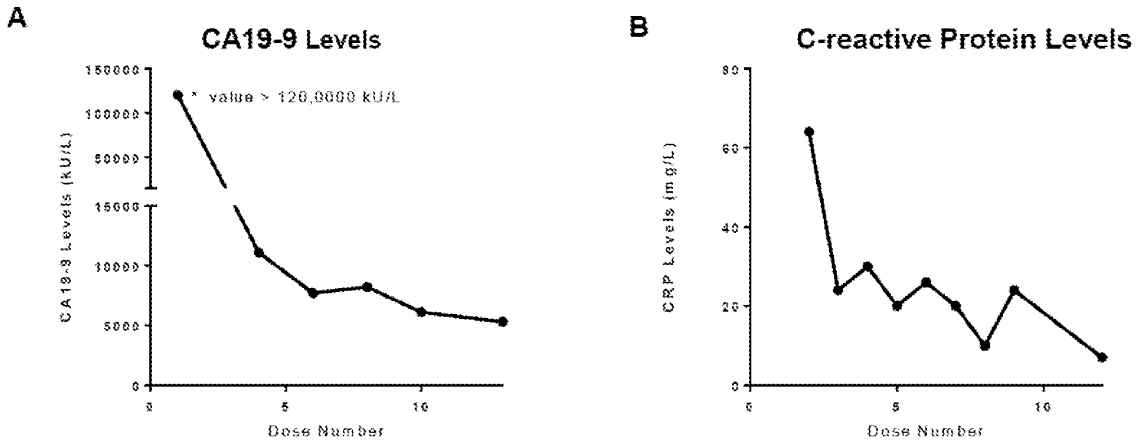

A
CA19-9 Levels
* value > 120,0000 kU/L
CA19-9 Levels (kU/L)
150000
100000
50000
15000
10000
5000
0
0
5
10
Dose Number
B
C-reactive Protein Levels
CRP Levels (mg/L)
80
60
40
20
0
0
5
10
Dose Number

FIG. 32
(Continued)

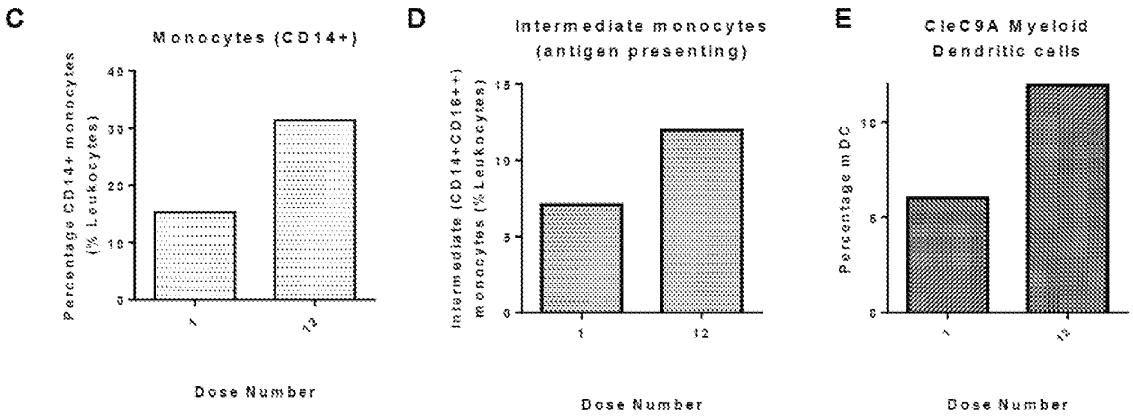


FIG. 32
(Continued)

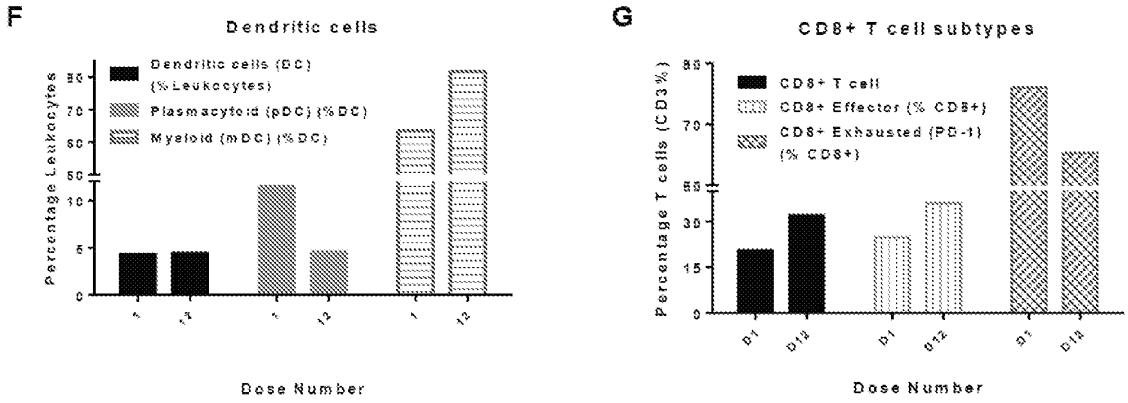


FIG. 33

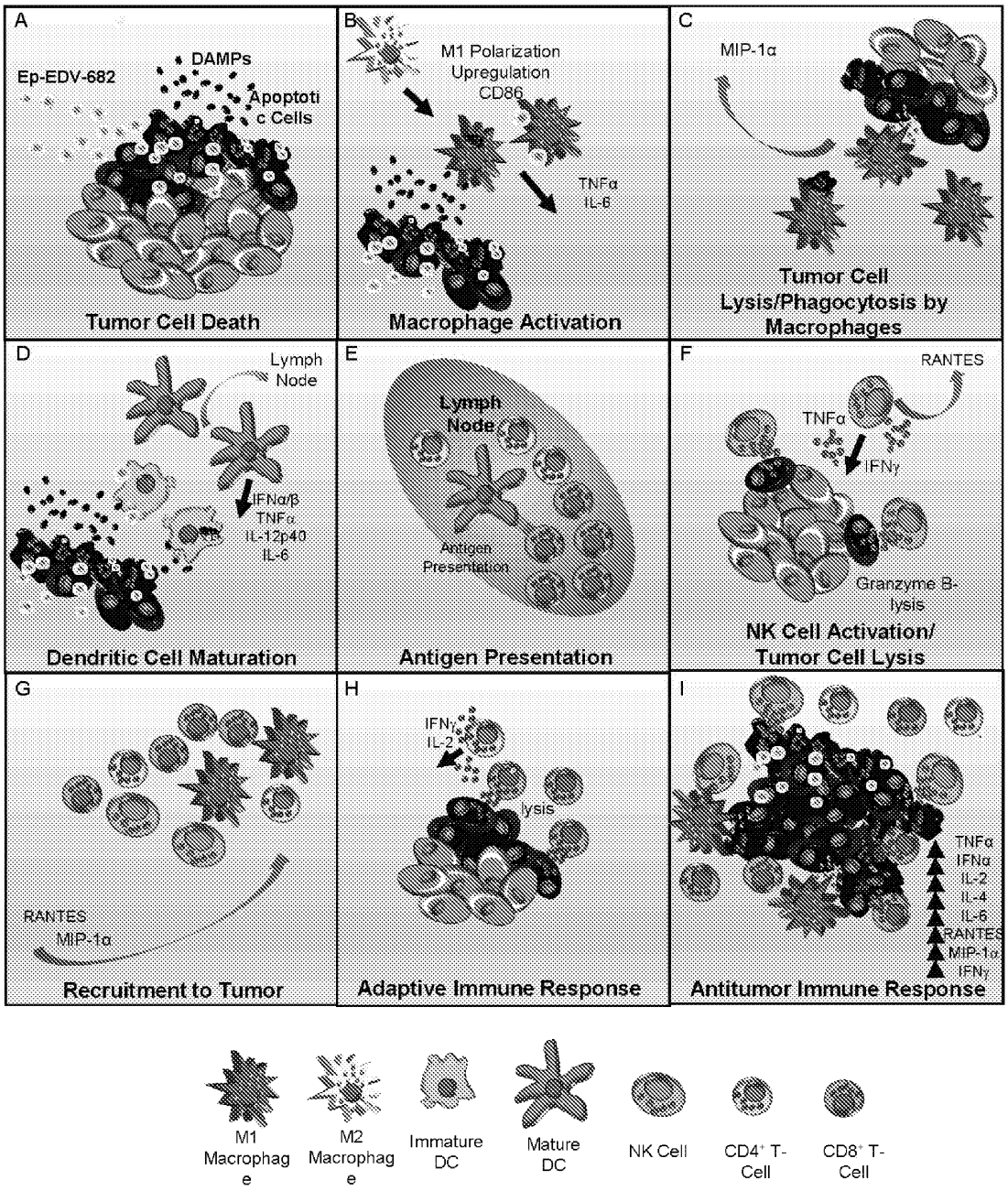

A
Ep-EDV-682
DAMPs
Apoptotic Cells
Tumor Cell Death
B
M1 Polarization
Upregulation
CD86
TNFα
IL-6
Macrophage Activation
C
MIP-1α
Tumor Cell
Lysis/Phagocytosis by
Macrophages
D
Lymph
Node
IFNα/β
TNFα
IL-12p40
IL-6
Dendritic Cell Maturation
E
Lymph
Node
Antigen
Presentation
Antigen Presentation
F
RANTES
TNFα
IFNγ
Granzyme B-
lysis
NK Cell Activation/
Tumor Cell Lysis
G
RANTES
MIP-1α
Recruitment to Tumor
H
IFNγ
IL-2
lysis
Adaptive Immune Response
I
TNFα
IFNα
IL-2
IL-4
IL-6
RANTES
MIP-1α
IFNγ
Antitumor Immune Response
M1
Macrophage
M2
Macrophage
Immature
DC
Mature
DC
NK Cell
CD4+ T-
Cell
CD8+ T-
Cell

FIG. 34

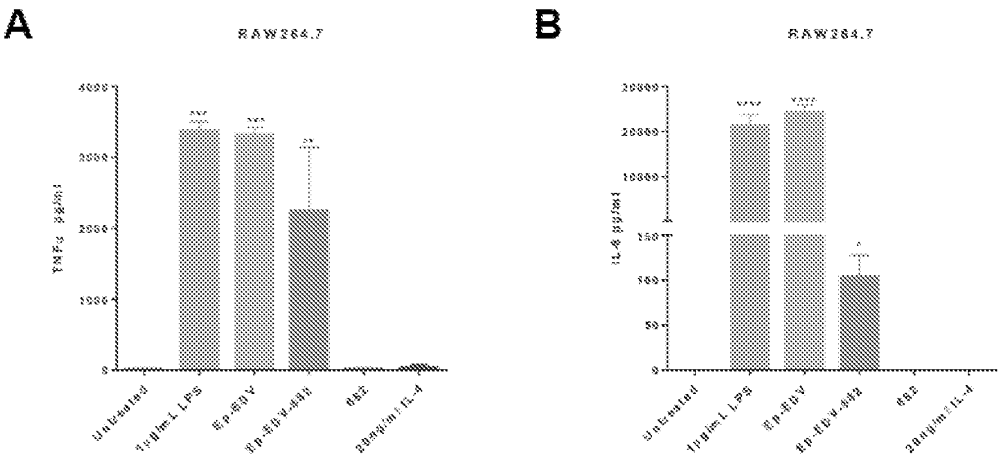

A
RAW264.7
TNFα pg/ml
Unstimulated
1μg/mL LPS
Ep-EpV
Ep-EpV-648
648
20ng/ml IL-4
B
RAW264.7
IL-6 pg/ml
Unstimulated
1μg/mL LPS
Ep-EpV
Ep-EpV-648
648
20ng/ml IL-4

FIG. 34

(Continued)

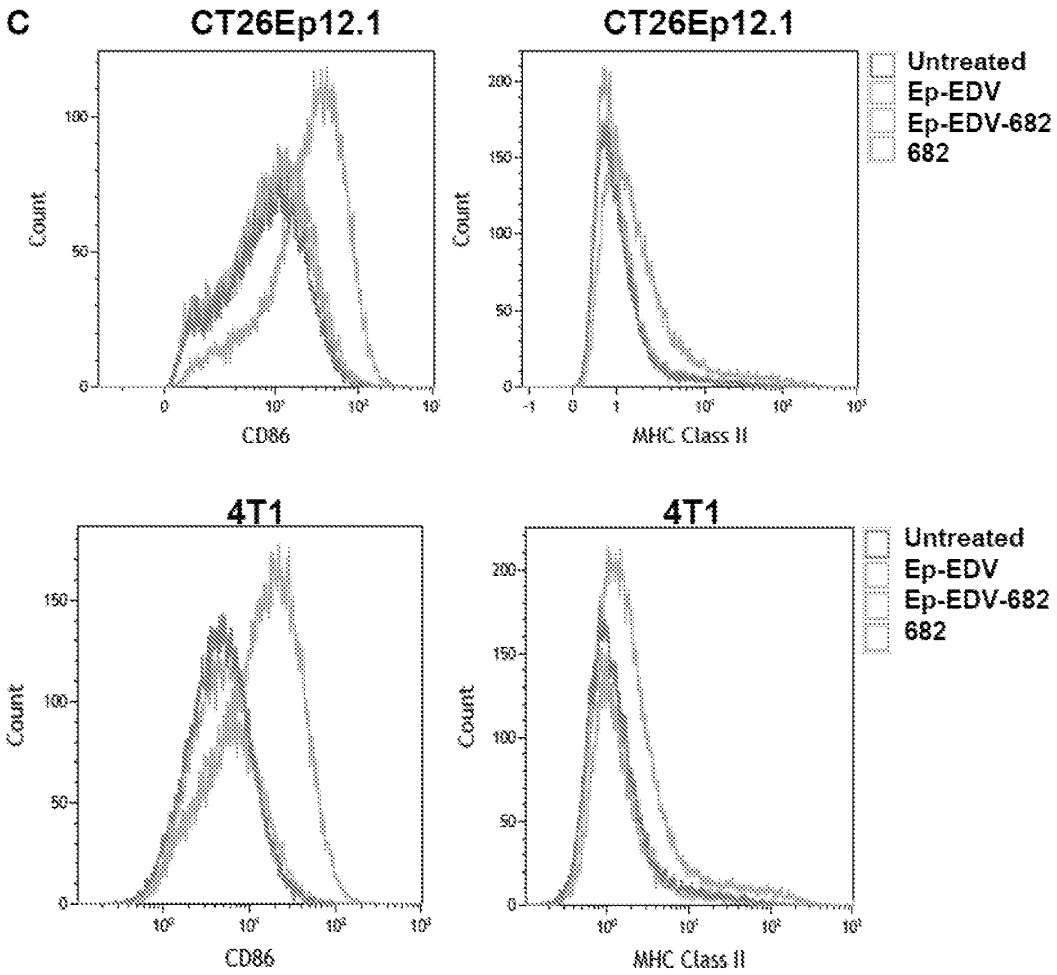


FIG. 34
(Continued)

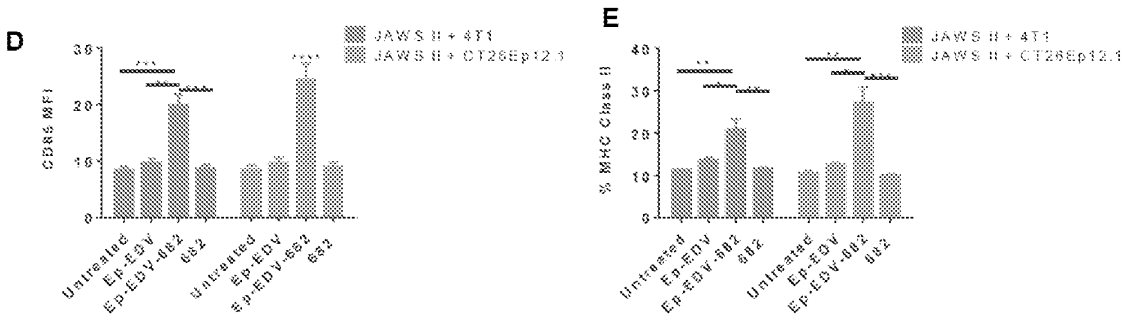


FIG. 35

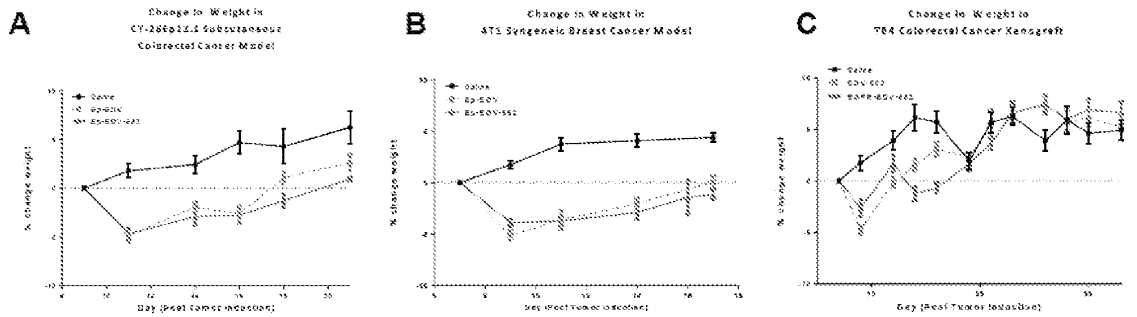


FIG. 35
(Continued)

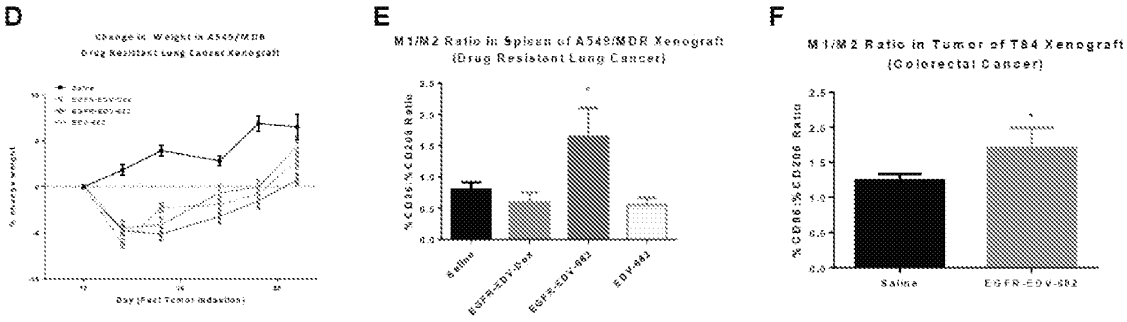


FIG. 35
(Continued)

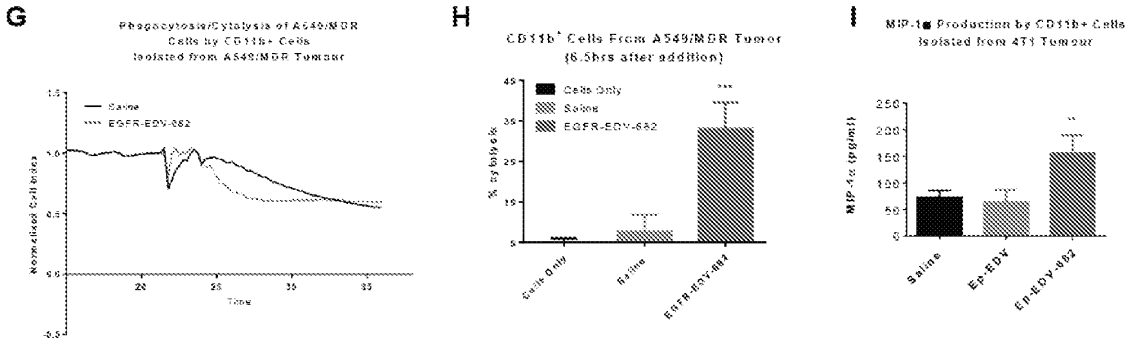


FIG. 36

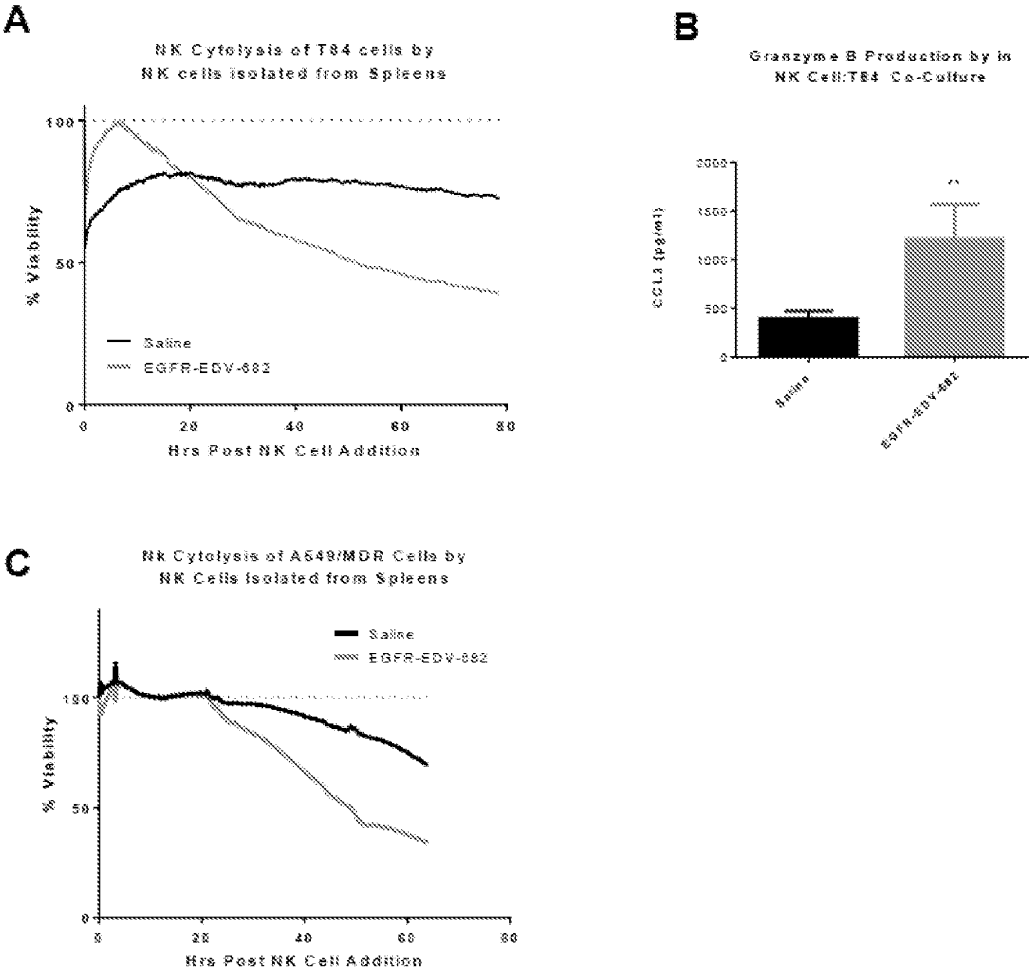
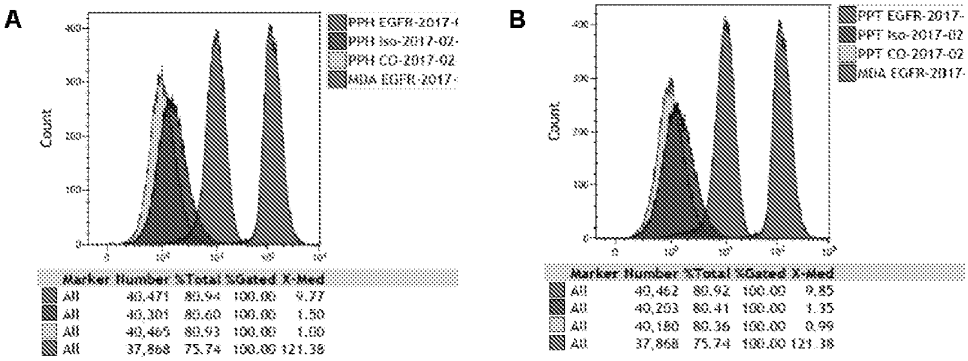
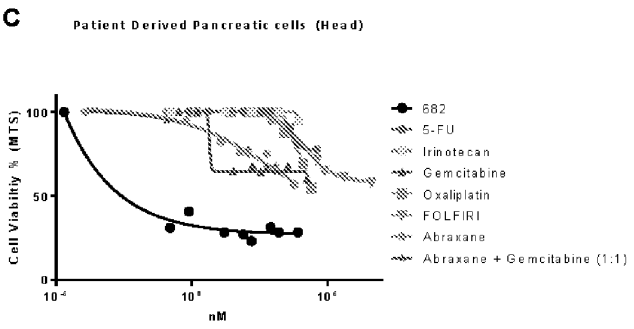

A
NK Cytolysis of T84 cells by
NK cells isolated from Spleens
% Viability
100
50
0
0
20
40
60
80
Hrs Post NK Cell Addition
Saline
EGFR-EDV-682
B
Granzyme B Production by in
NK Cell:T84 Co-Culture
CCL3 (pg/ml)
3000
2500
2000
1500
1000
500
0
Saline
EGFR-EDV-682
C
Nk Cytolysis of A549/MDR Cells by
NK Cells isolated from Spleens
Saline
EGFR-EDV-682
% Viability
100
50
0
0
20
40
60
80
Hrs Post NK Cell Addition

FIG. 37



Head - 287,152 receptors/cell

Tail - 296,840 receptors/cell



	IC-50 (nM)
682	< 0.1
5-FU	> 38,000
Irinotecan	> 10,000
Gemcitabine	> 16,000
Oxaliplatin	> 25,000
FOLFIRI	> 38.000
Abraxane	> 5,000
Abraxane +Gemcitabine	> 5,000

FIG. 38

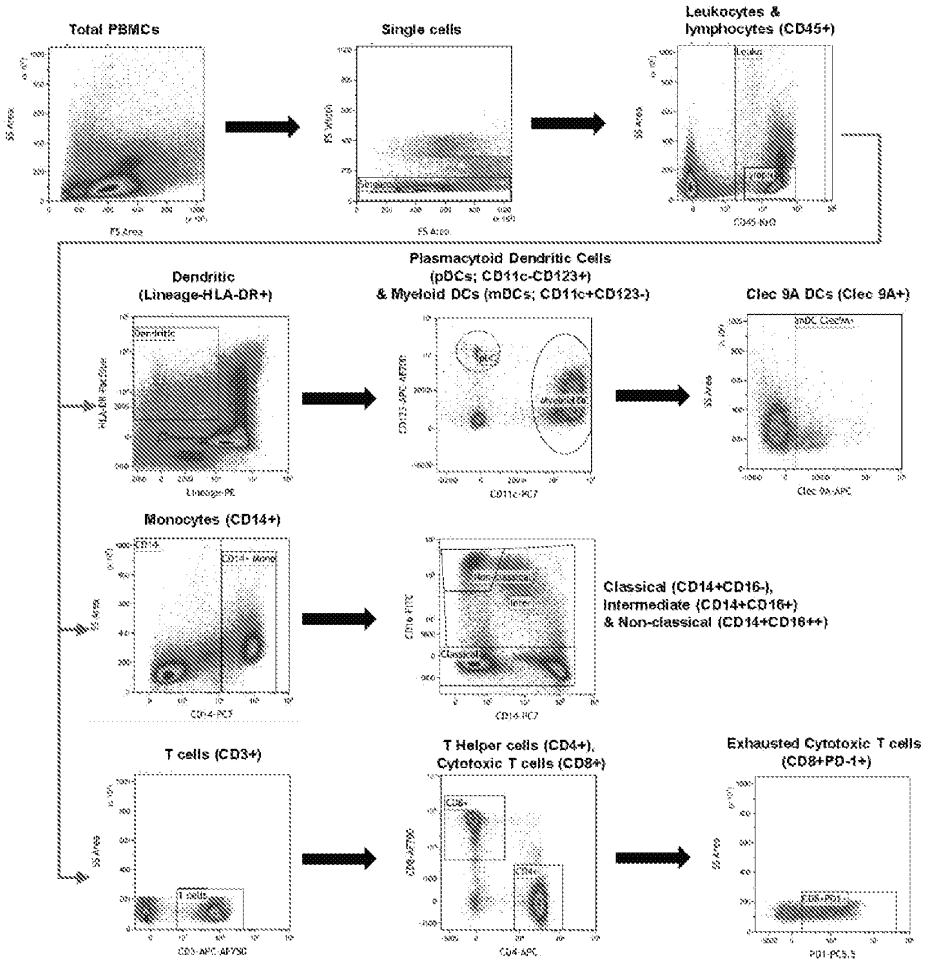

Total PBMCs
Single cells
Leukocytes & lymphocytes (CD45+)
Dendritic (Lineage-HLA-DR+)
Plasmacytoid Dendritic Cells (pDCs; CD11c-CD123+) & Myeloid DCs (mDCs; CD11c+CD123-)
Clec 9A DCs (Clec 9A+)
Monocytes (CD14+)
Classical (CD14+CD16-), Intermediate (CD14+CD16+) & Non-classical (CD14+CD16++)
T cells (CD3+)
T Helper cells (CD4+), Cytotoxic T cells (CD8+)
Exhausted Cytotoxic T cells (CD8+PD-1+)

FIG. 39

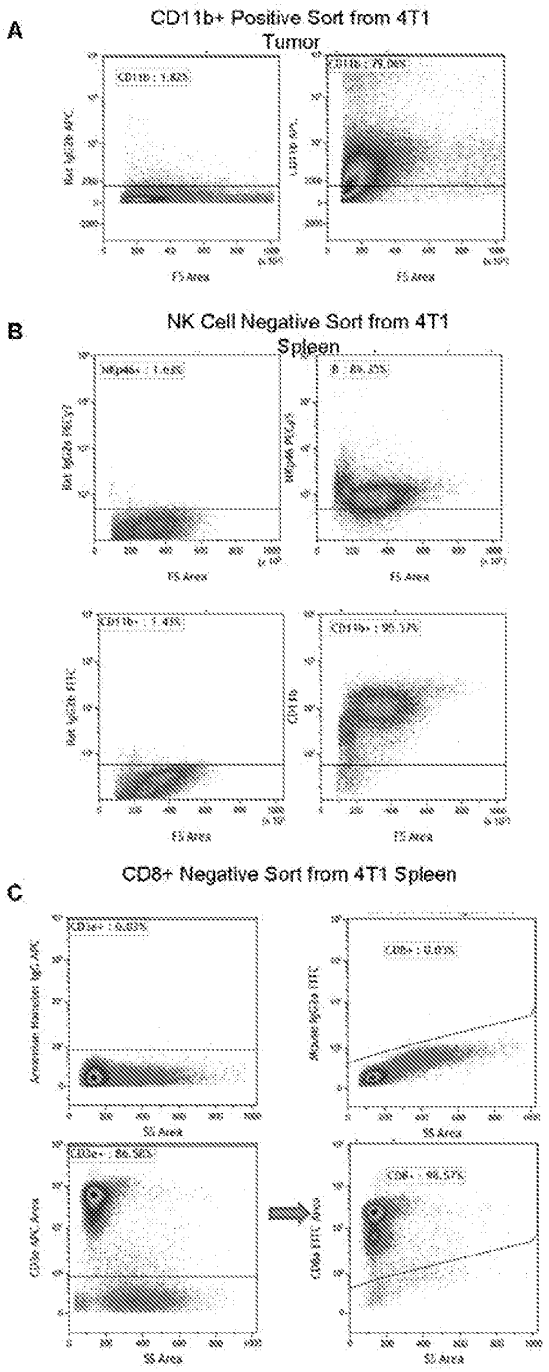
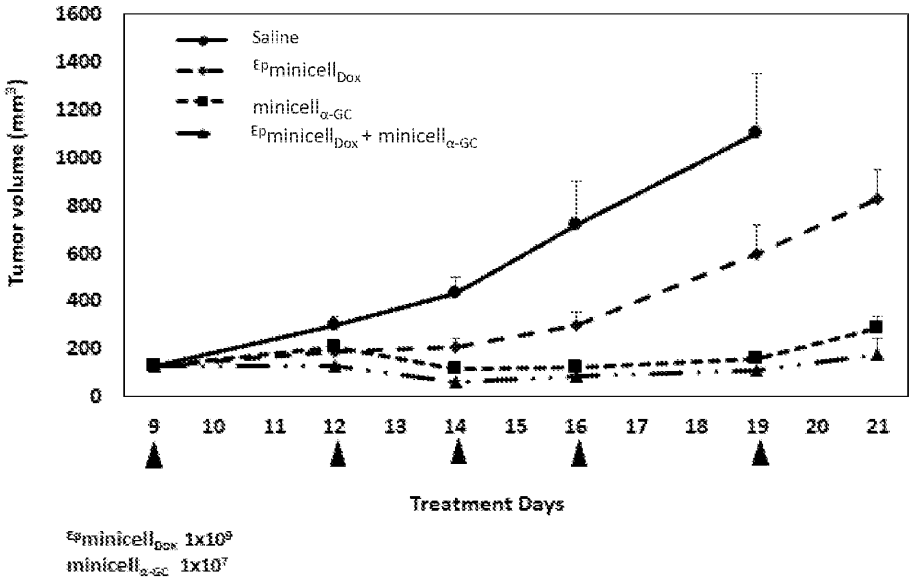

A
CD11b+ Positive Sort from 4T1 Tumor
B
NK Cell Negative Sort from 4T1 Spleen
C
CD8+ Negative Sort from 4T1 Spleen

FIG. 40



EPminicell$_{Dox}$ 1x10^9
minicell$_{\alpha\text{-GC}}$ 1x10^7

FIG. 41

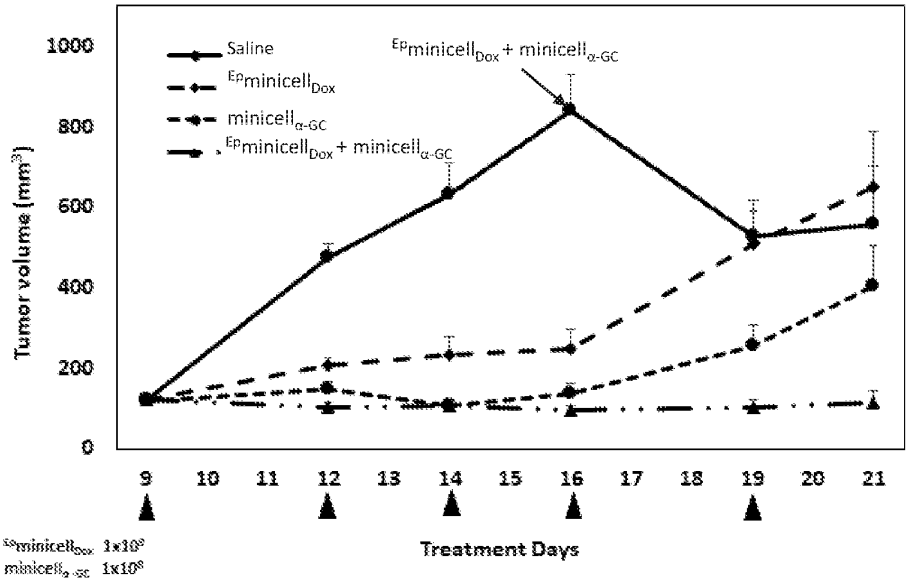

Saline
EpminicellDox
minicellα-GC
EpminicellDox + minicellα-GC
EpminicellDox + minicellα-GC
Tumor volume (mm3)
1000
800
600
400
200
0
9
10
11
12
13
14
15
16
17
18
19
20
21
Treatment Days
EpminicellDox 1x10^9
minicellα-GC 1x10^9

FIG. 42

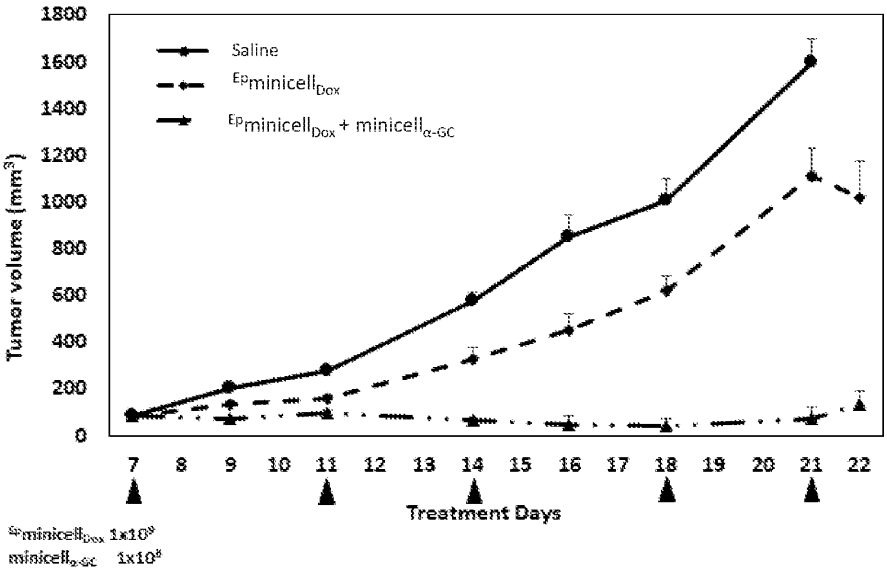

1800
1600
1400
1200
1000
800
600
400
200
0
Tumor volume (mm3)
Saline
EpminicellDox
EpminicellDox + minicellα-GC
7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22
Treatment Days
EpminicellDox 1x10^9
minicellα-GC 1x10^8

FIG. 43

CT26 (Epclone12.1) isograft; n=3; Tumor volume: 200-250mm^3; Dose: 1; Sacrifice Time: 24 hr

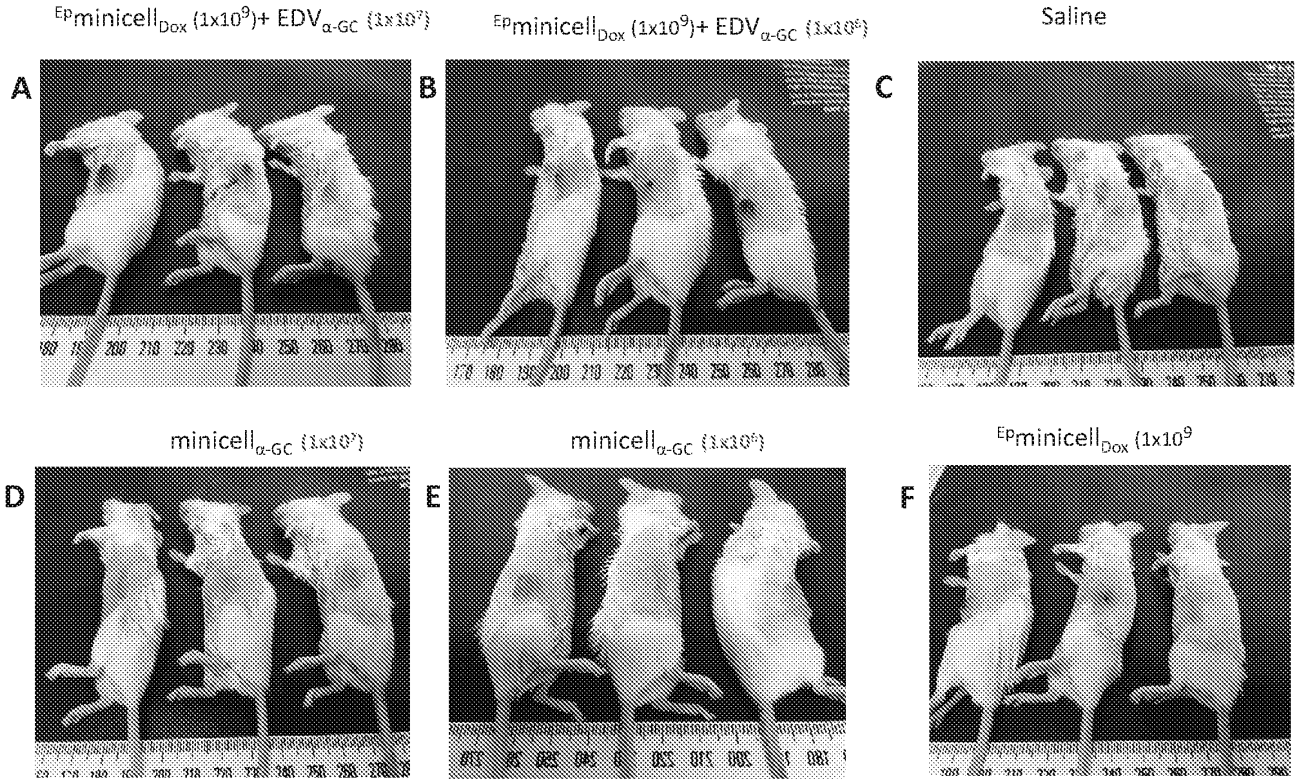
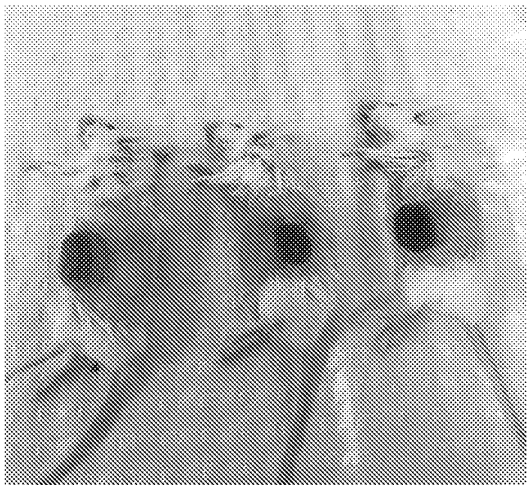


FIG. 44

CT26 (Epclone12.1) isograft; n=3; Tumor volume: 600-800mm^3; Dose: 1; Sacrifice Time: 24 hr

A

^{Ep}minicell$_{Dox}$ (1x10^9)+ minicell$_{\alpha\text{-}GC}$ (1x10^7)
16hr



B

^{Ep}minicell$_{Dox}$ (1x10^9)+ minicell$_{\alpha\text{-}GC}$ (1x10^7)
24hr

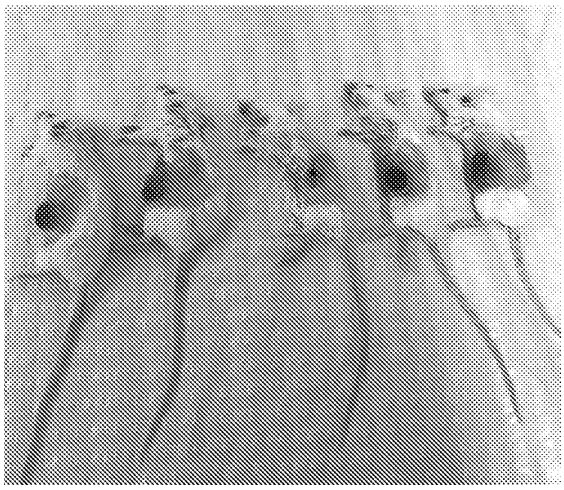
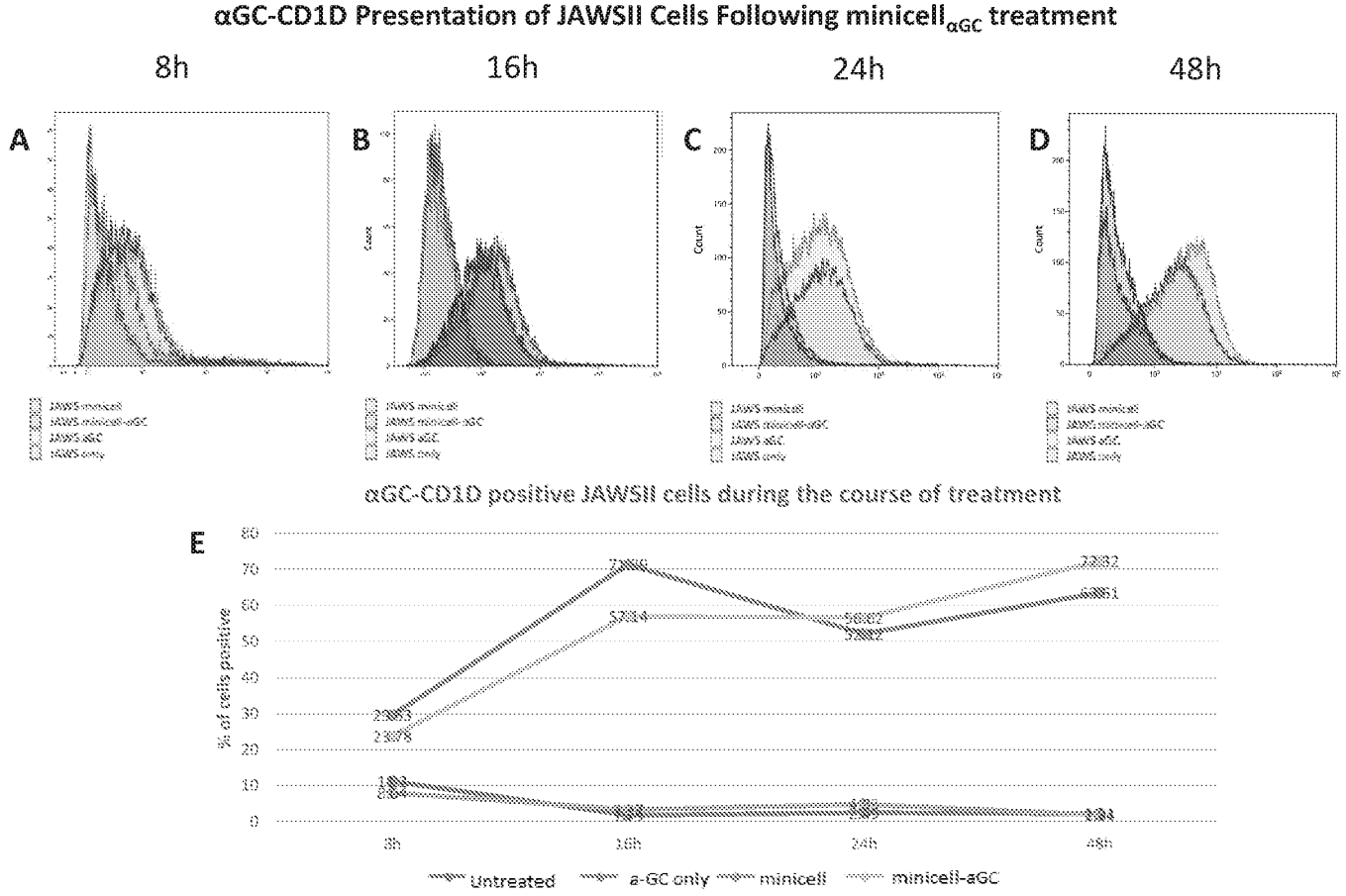


FIG. 45


αGC-CD1D Presentation of JAWSII Cells Following minicell$_{αGC}$ treatment
8h
16h
24h
48h
A
B
C
D
Count
αGC-CD1D positive JAWSII cells during the course of treatment
E
% of cells positive
0
10
20
30
40
50
60
70
80
8h
16h
24h
48h
Untreated
a-GC only
minicell
minicell-aGC