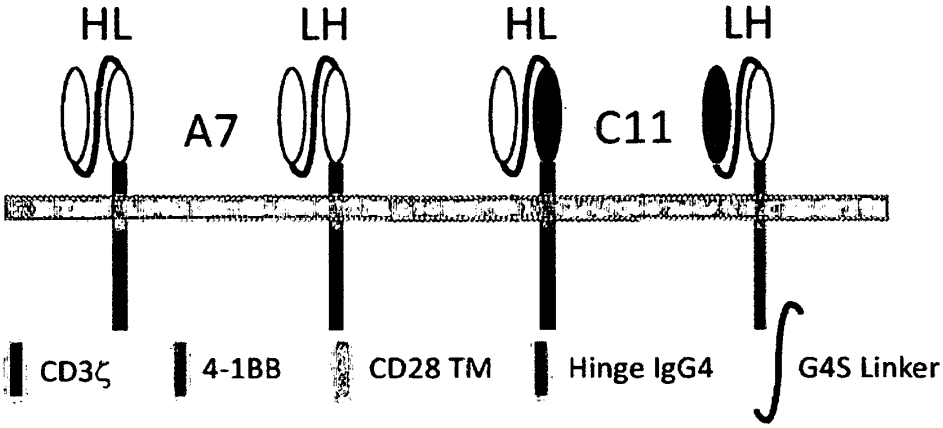

HL
LH
HL
LH
A7
C11
CD3ζ
4-1BB
CD28 TM
Hinge IgG4
G4S Linker

*FIG. 1A*

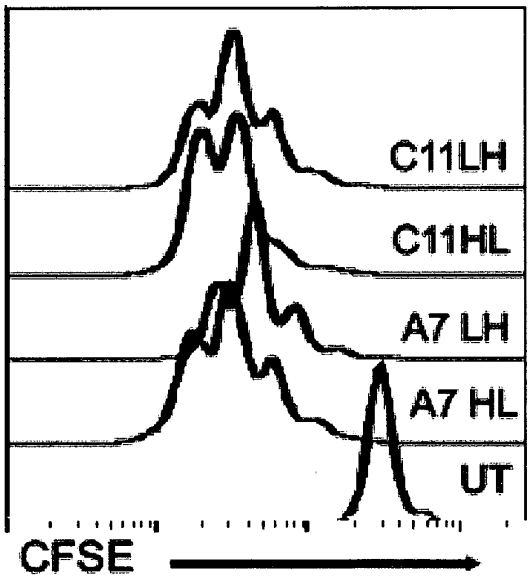

C11LH
C11HL
A7 LH
A7 HL
UT
CFSE

*FIG. 1B*

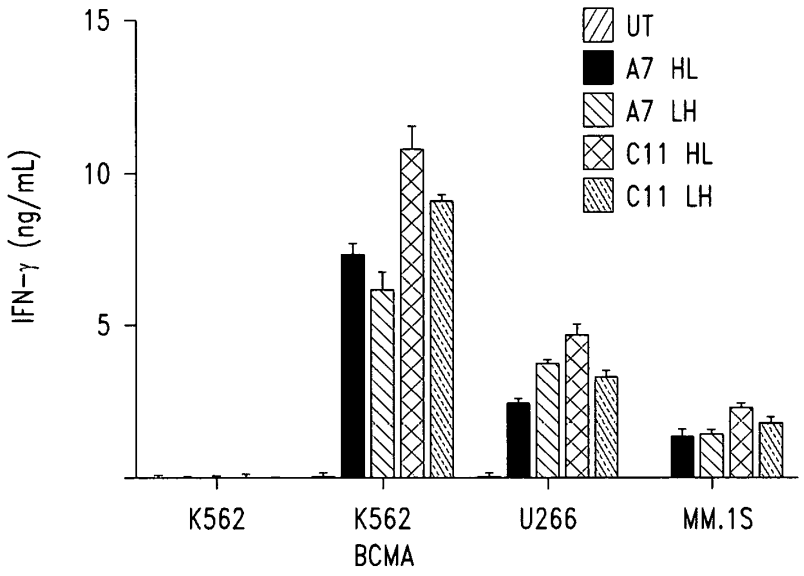

15
10
5
IFN-γ (ng/mL)
UT
A7 HL
A7 LH
C11 HL
C11 LH
K562
K562 BCMA
U266
MM.1S

*FIG. 1C*

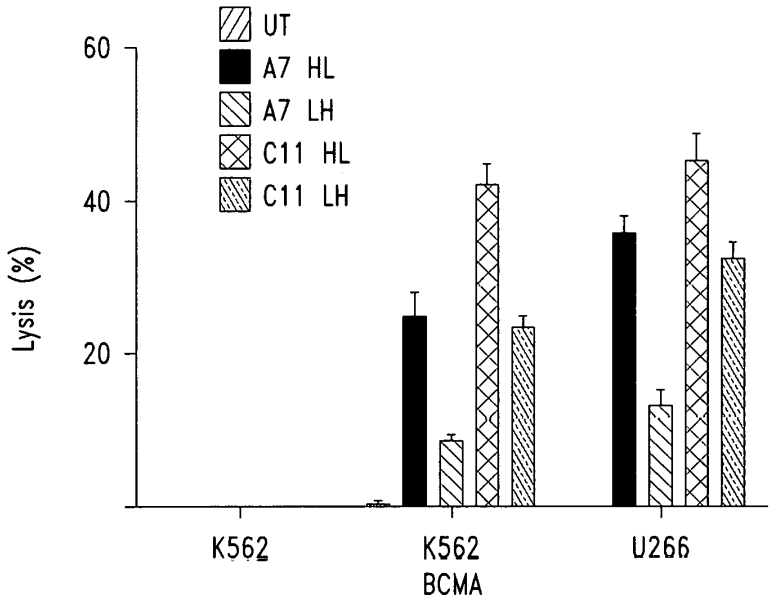

60
40
20
Lysis (%)
UT
A7 HL
A7 LH
C11 HL
C11 LH
K562
K562 BCMA
U266

*FIG. 1D*

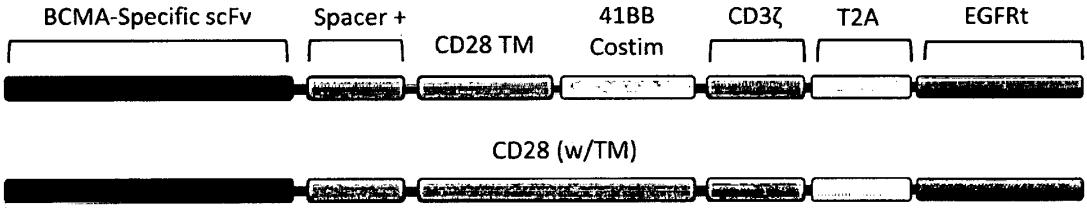

BCMA-Specific scFv
Spacer +
CD28 TM
41BB
Costim
CD3ζ
T2A
EGFRt
CD28 (w/TM)

*FIG. 1E*

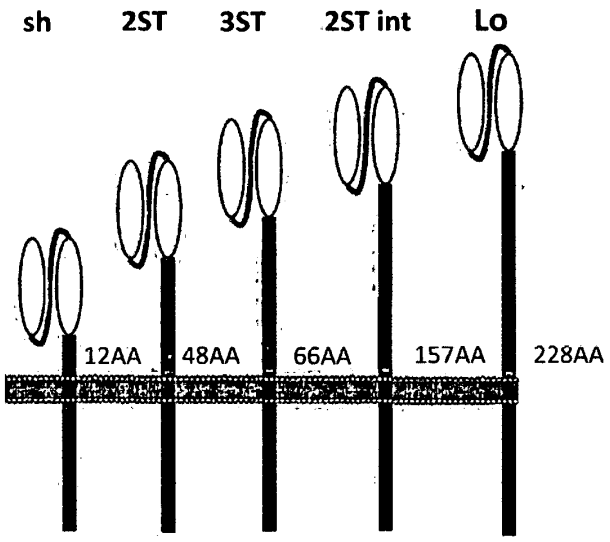

sh
2ST
3ST
2ST int
Lo
12AA
48AA
66AA
157AA
228AA

*FIG. 1F*

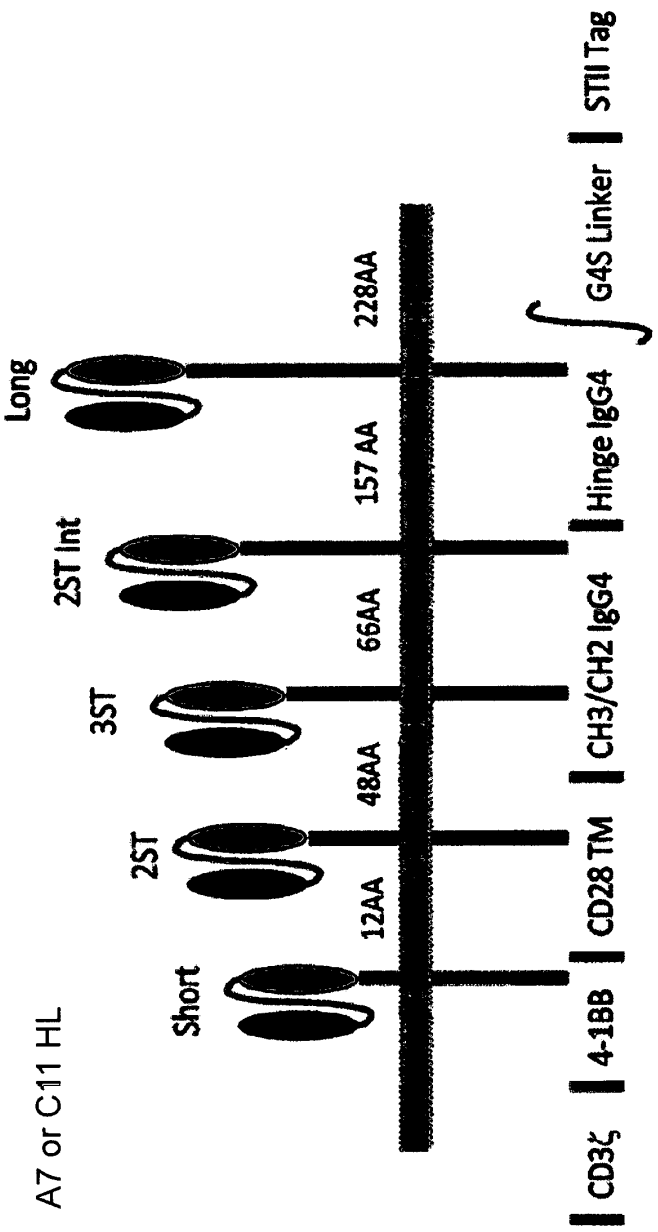

A7 or C11 HL
Short
2ST
3ST
2ST Int
Long
12AA
48AA
66AA
157 AA
228AA
CD3ζ
4-1BB
CD28 TM
CH3/CH2 IgG4
Hinge IgG4
G4S Linker
STII Tag

*FIG. 1G*

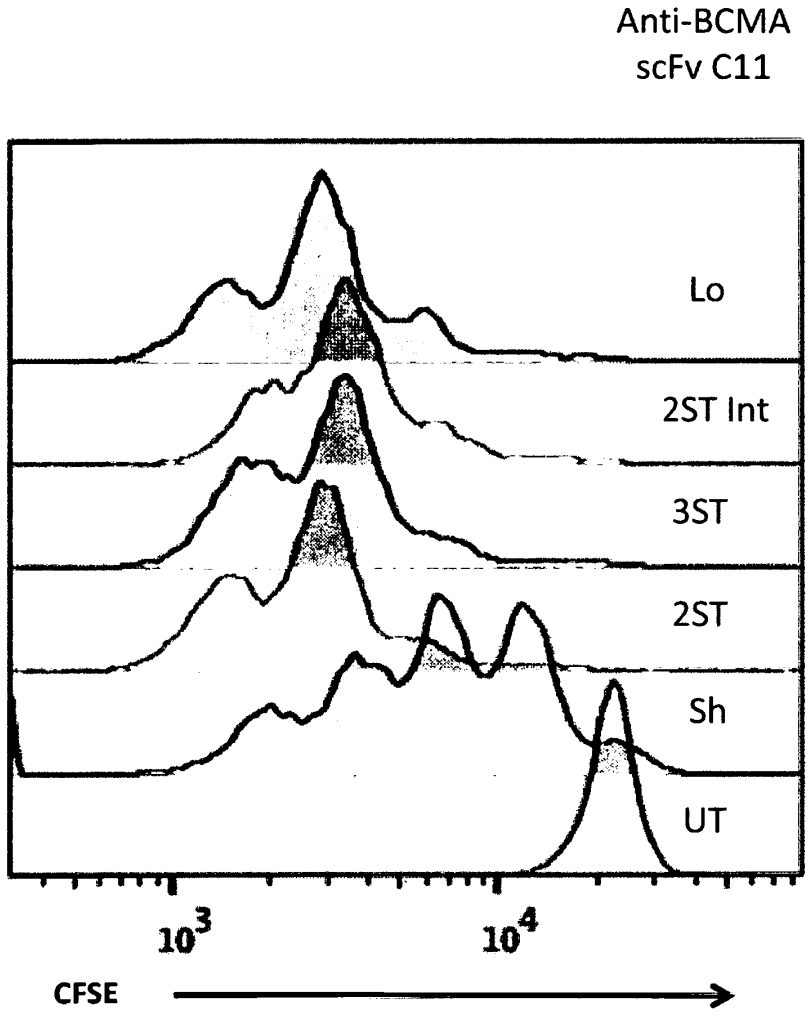



*FIG. 1H*

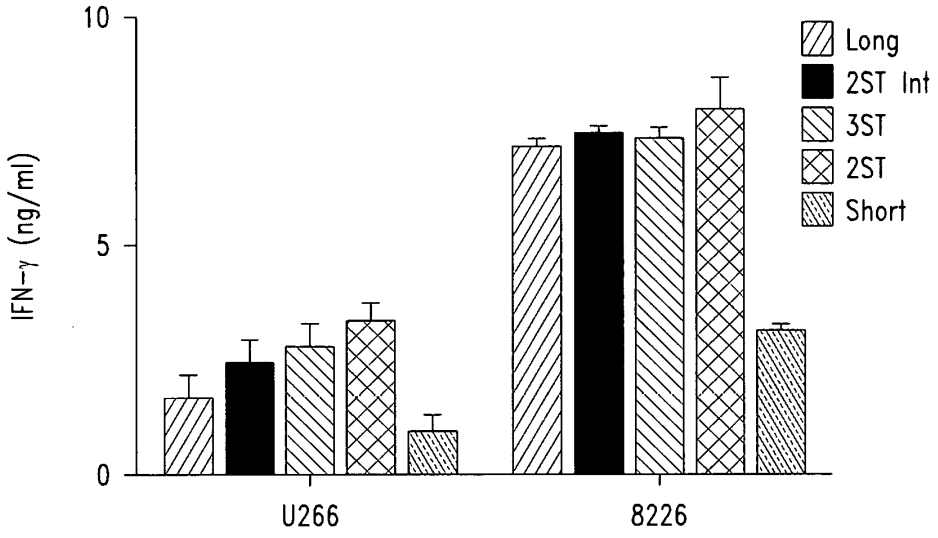

10
5
0
IFN-γ (ng/ml)
U266
8226
Long
2ST Int
3ST
2ST
Short

*FIG. 1I*

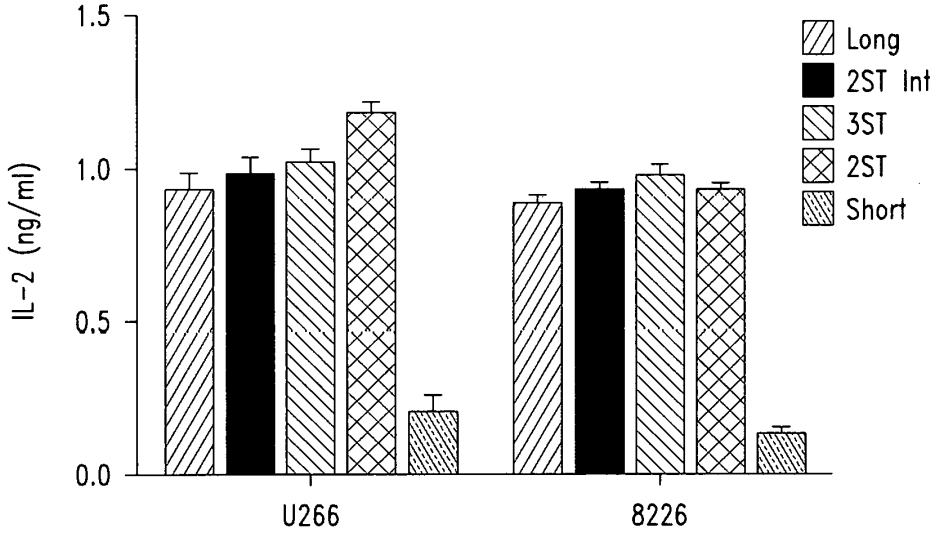

1.5
1.0
0.5
0.0
IL-2 (ng/ml)
U266
8226
Long
2ST Int
3ST
2ST
Short

*FIG. 1J*

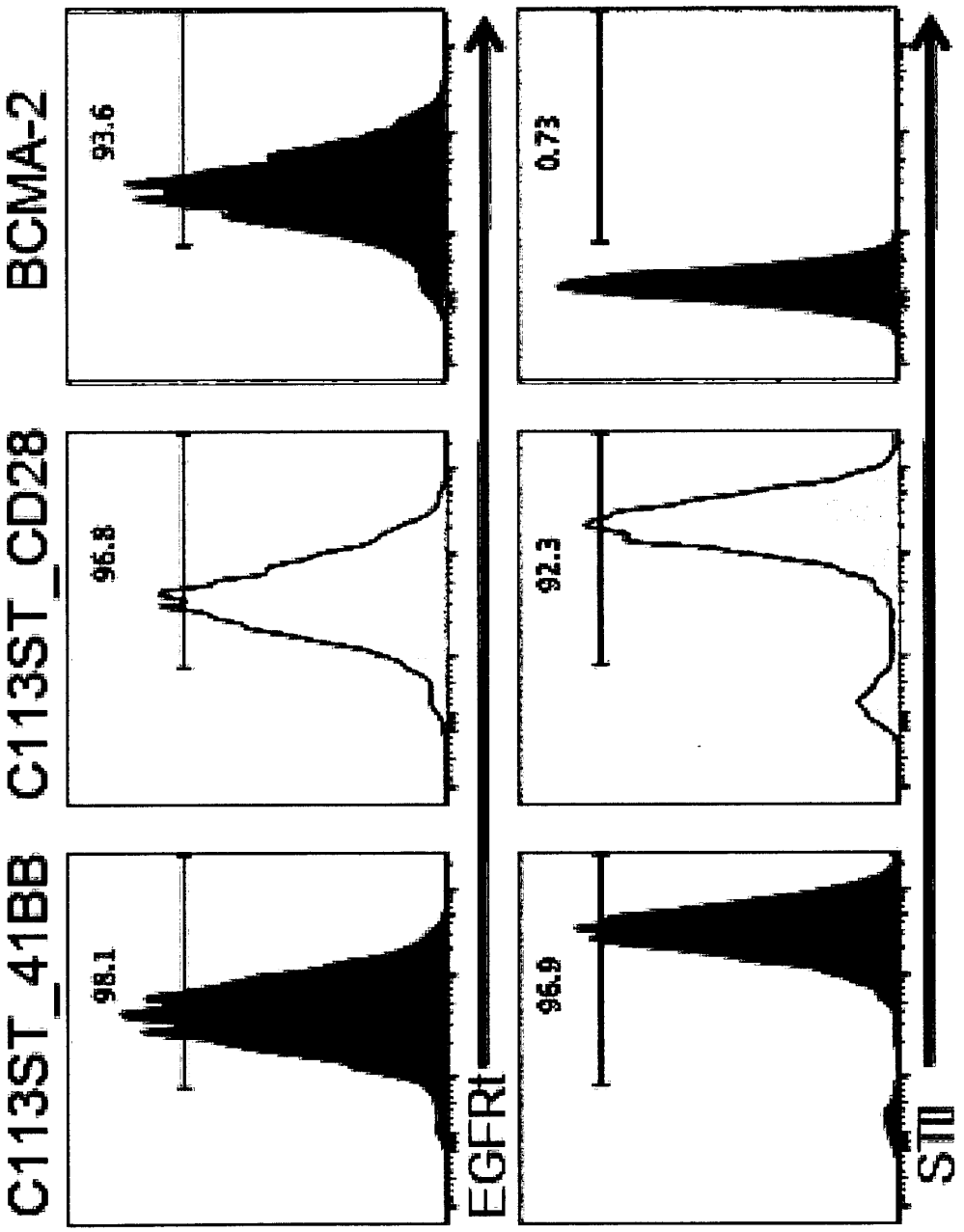

C113ST_41BB
C113ST_CD28
BCMA-2
98.1
96.8
93.6
96.9
92.3
0.73
EGFRt
STII

*FIG. 1K*

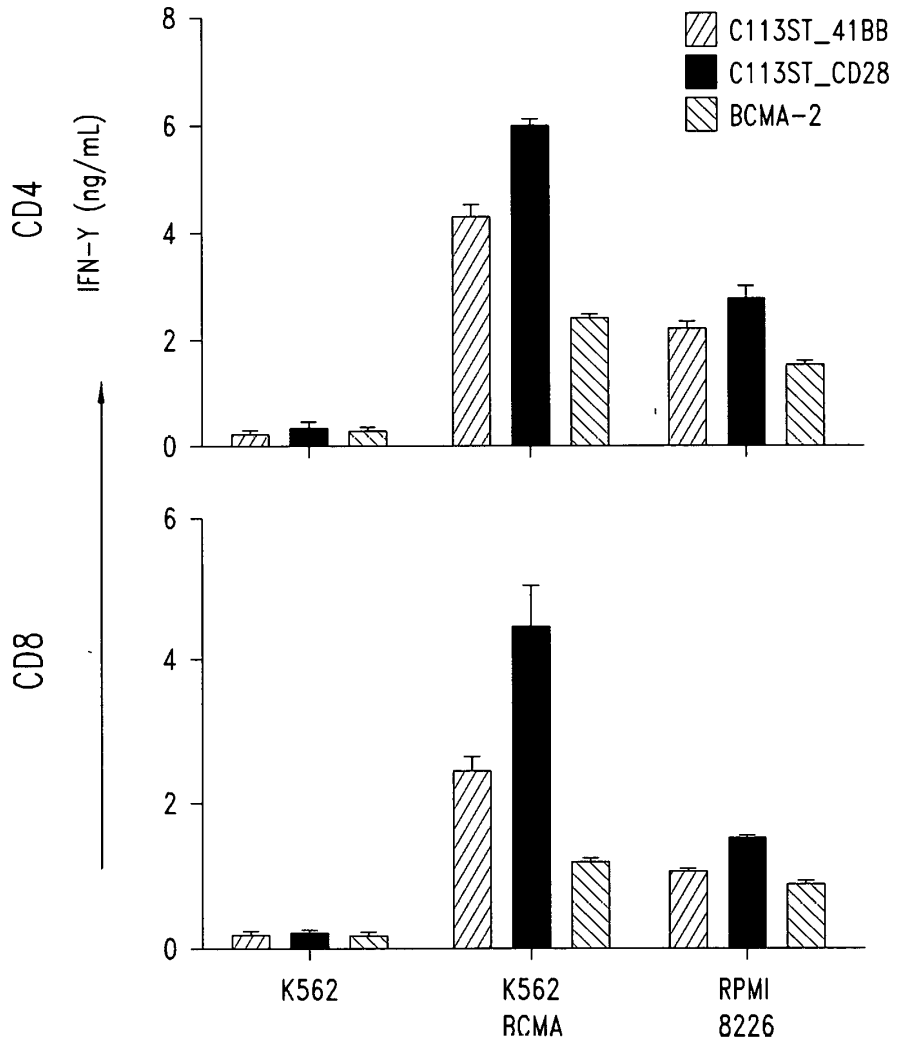

C113ST_41BB
C113ST_CD28
BCMA-2
CD4
IFN-Y (ng/mL)
CD8
8
6
4
2
0
6
4
2
0
K562
K562 RCMA
RPMI 8226

*FIG. 1L*

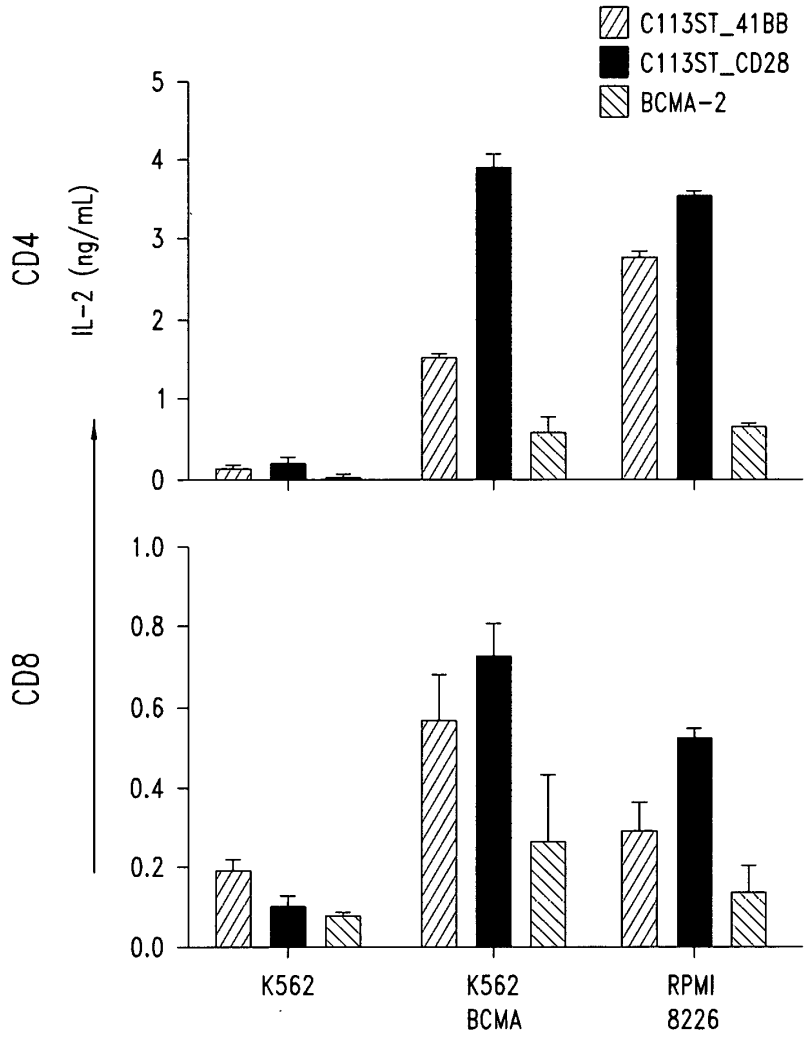

C113ST_41BB
C113ST_CD28
BCMA-2
CD4
IL-2 (ng/mL)
5
4
3
2
1
0
CD8
1.0
0.8
0.6
0.4
0.2
0.0
K562
K562 BCMA
RPMI 8226

*FIG. 1M*

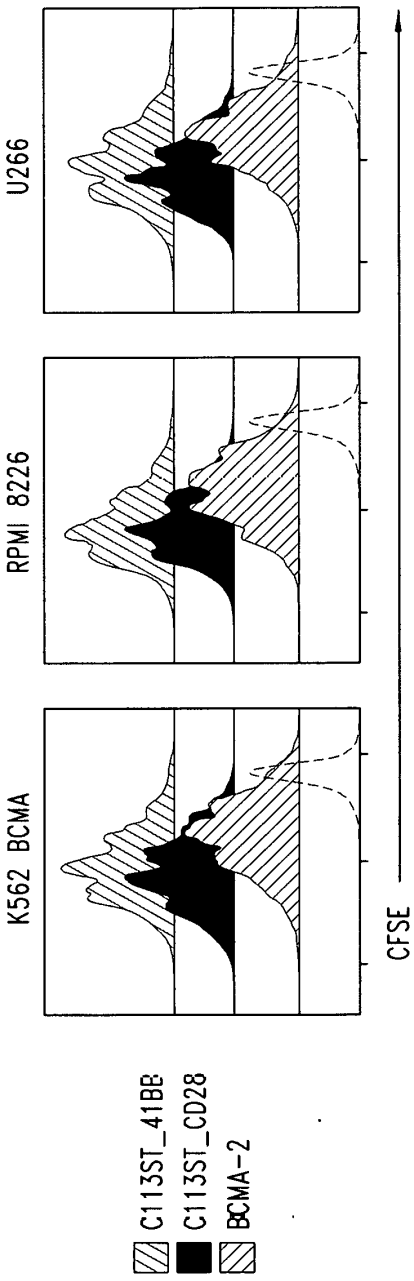



*FIG. 1N*

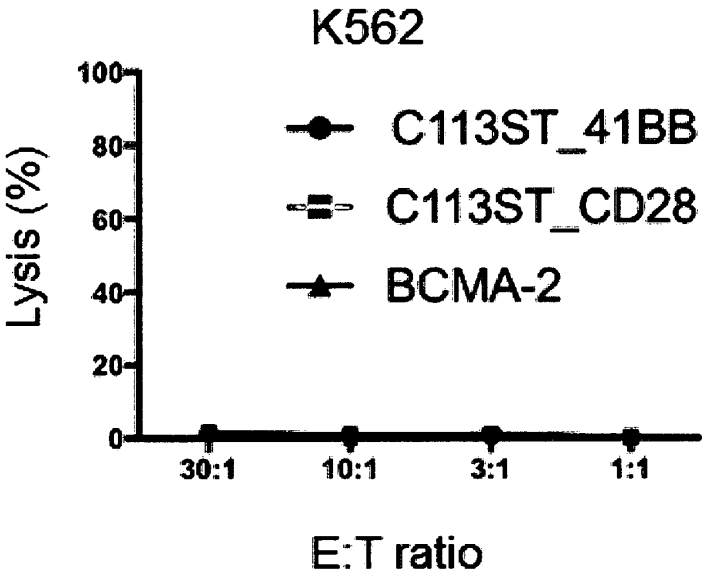

K562
Lysis (%)
100
80
60
40
20
0
C113ST_41BB
C113ST_CD28
BCMA-2
30:1
10:1
3:1
1:1
E:T ratio

*FIG. 1O*

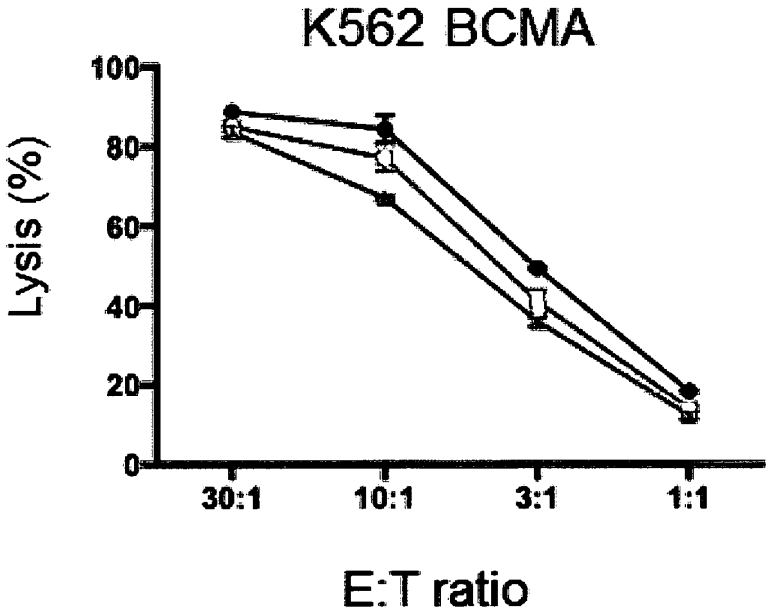

K562 BCMA
Lysis (%)
100
80
60
40
20
0
30:1
10:1
3:1
1:1
E:T ratio

*FIG. 1P*

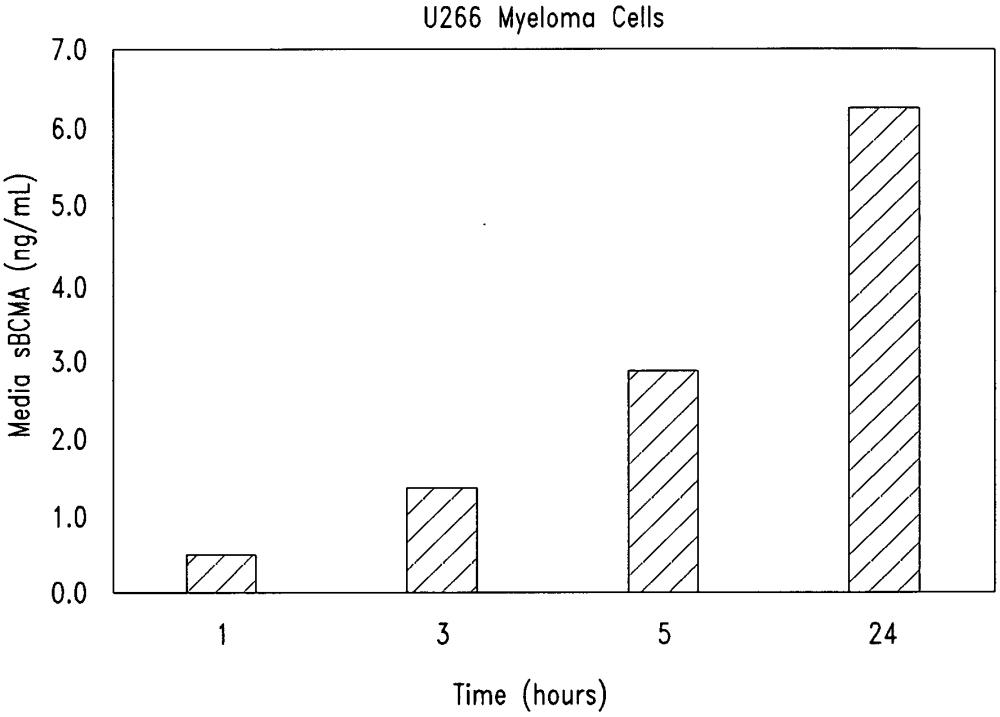

U266 Myeloma Cells
Media sBCMA (ng/mL)
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
1
3
5
24
Time (hours)

*FIG. 2A*

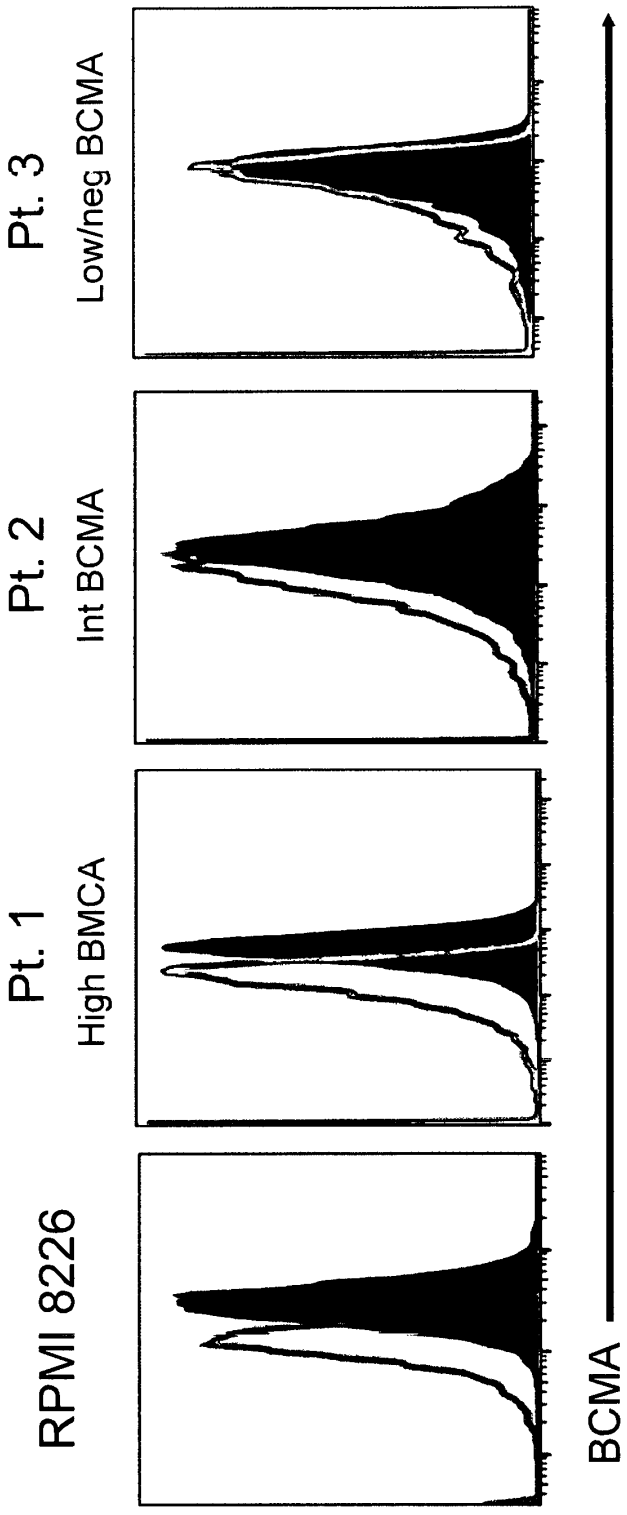



*FIG. 2B*

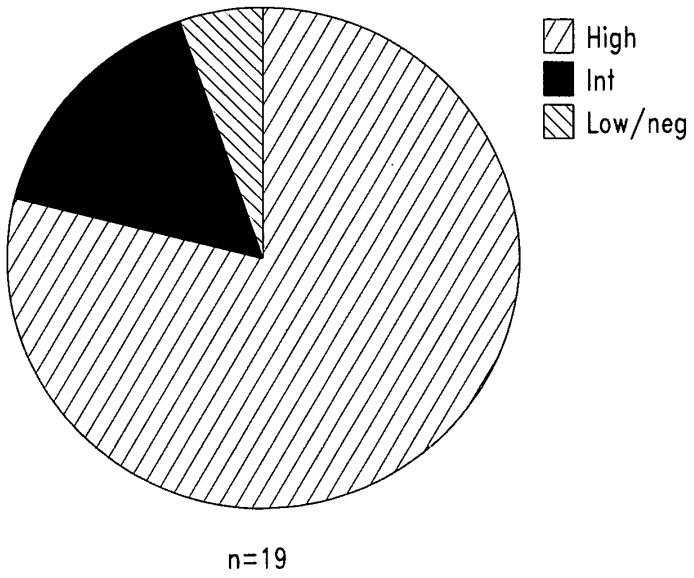

High
Int
Low/neg
n=19

*FIG. 2C*

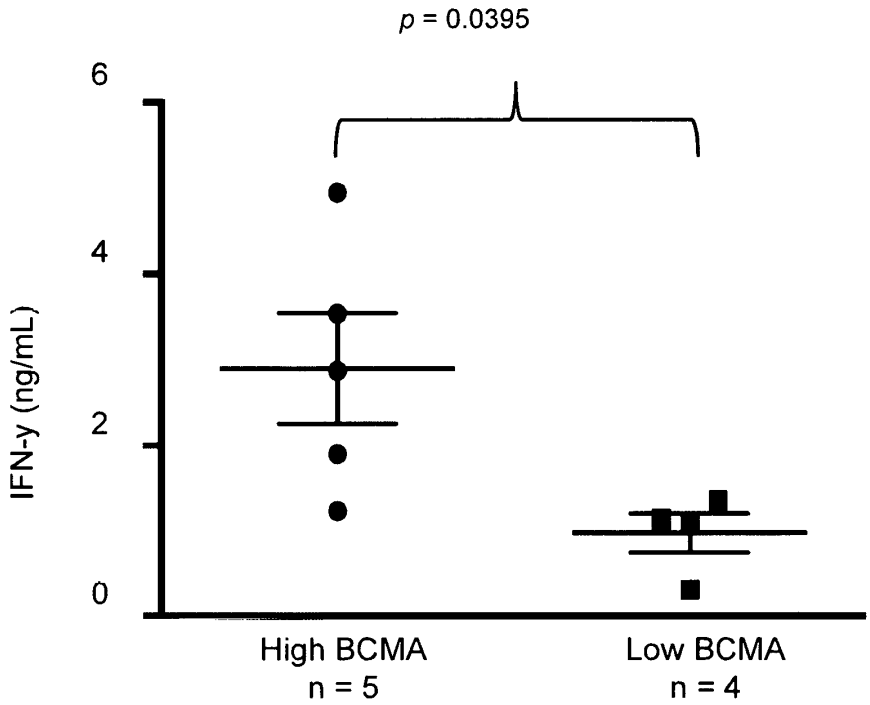

p = 0.0395
6
4
2
0
IFN-γ (ng/mL)
High BCMA
n = 5
Low BCMA
n = 4

*FIG. 2D*

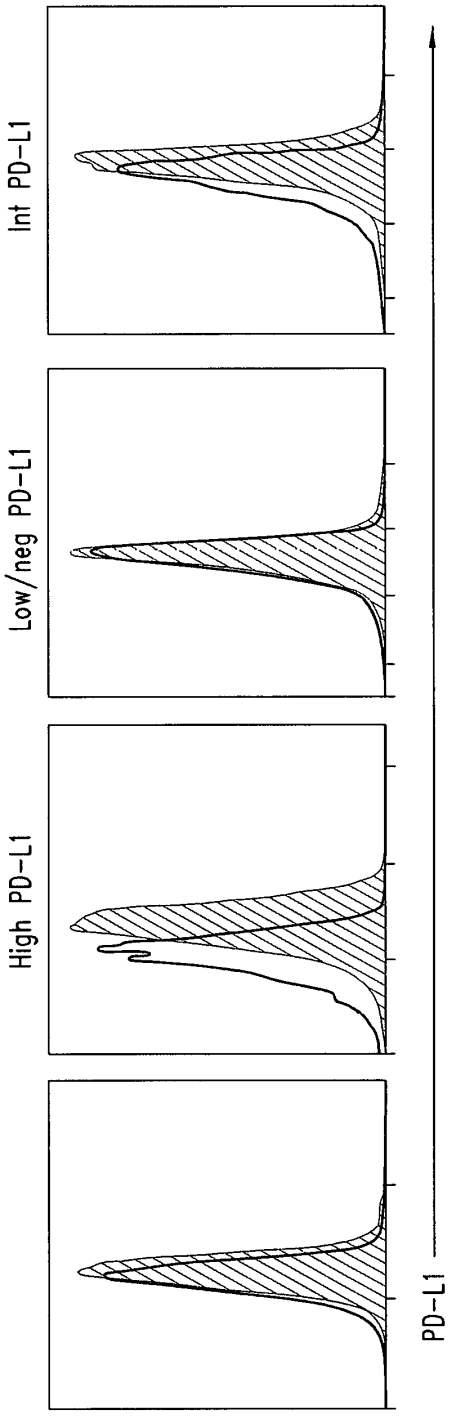



*FIG. 2E*

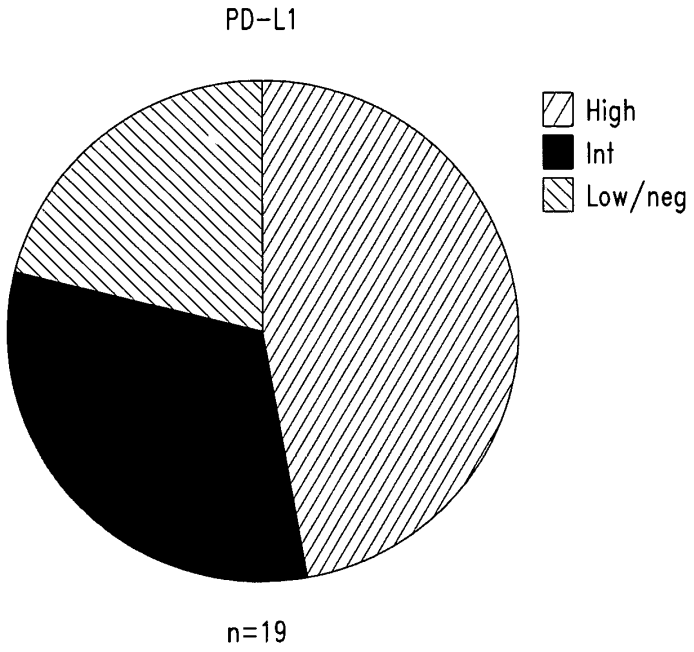

PD-L1
High
Int
Low/neg
n=19

*FIG. 2F*

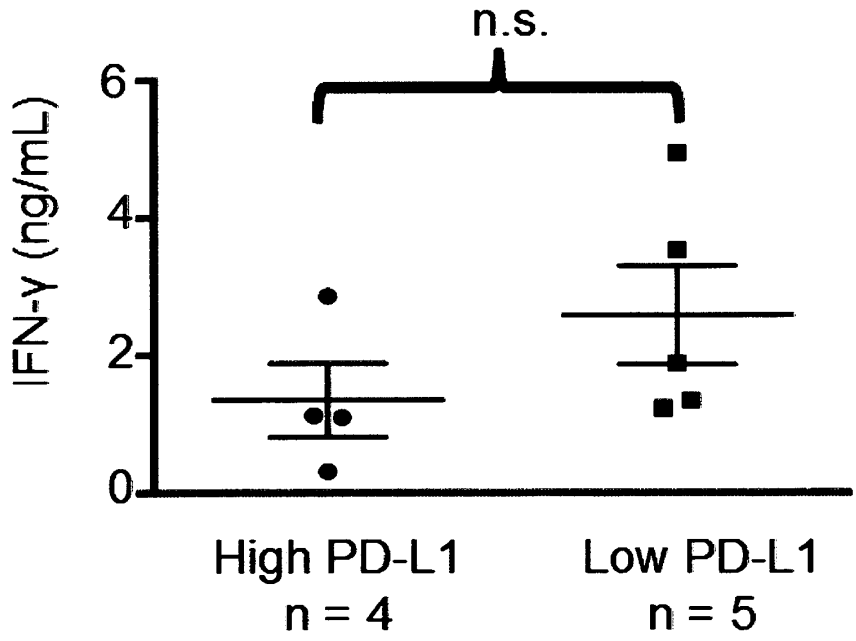

n.s.
IFN-γ (ng/mL)
6
4
2
0
High PD-L1
n = 4
Low PD-L1
n = 5

*FIG. 2G*

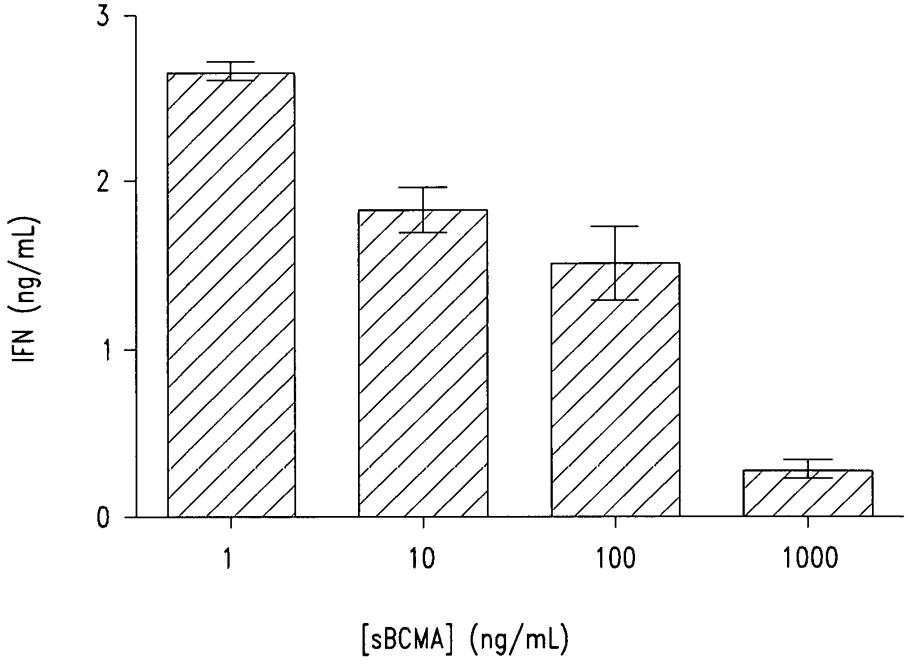

3
2
1
0
IFN (ng/mL)
1
10
100
1000
[sBCMA] (ng/mL)

*FIG. 2H*

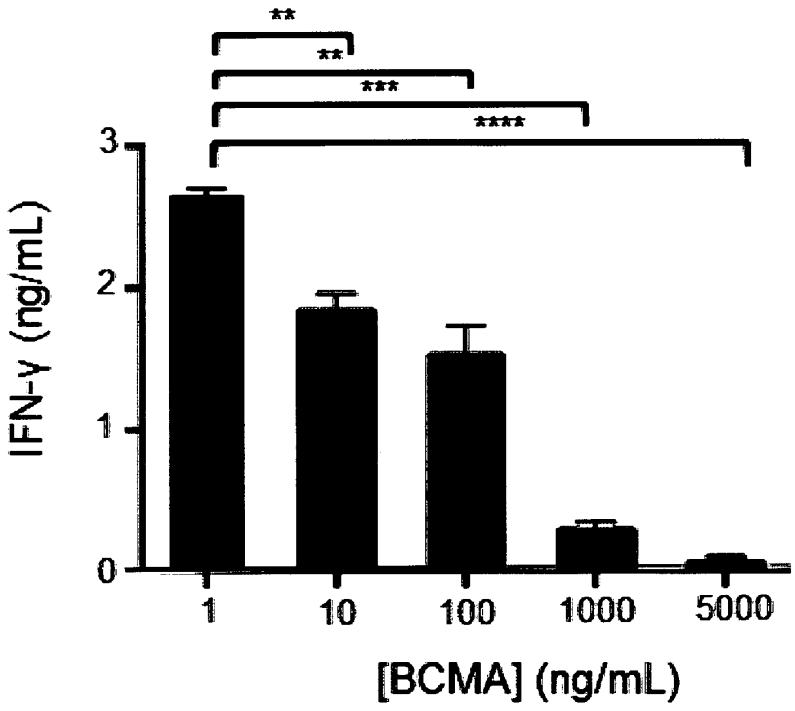

**
**
***
****
3
2
1
0
IFN-γ (ng/mL)
1
10
100
1000
5000
[BCMA] (ng/mL)

*FIG. 2I*

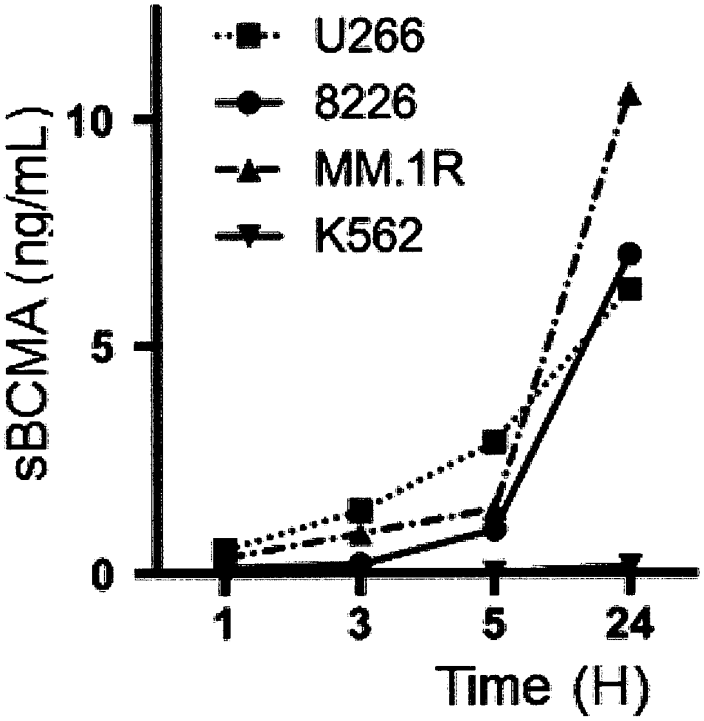

U266
8226
MM.1R
K562
sBCMA (ng/mL)
10
5
0
1
3
5
24
Time (H)

*FIG. 2J*

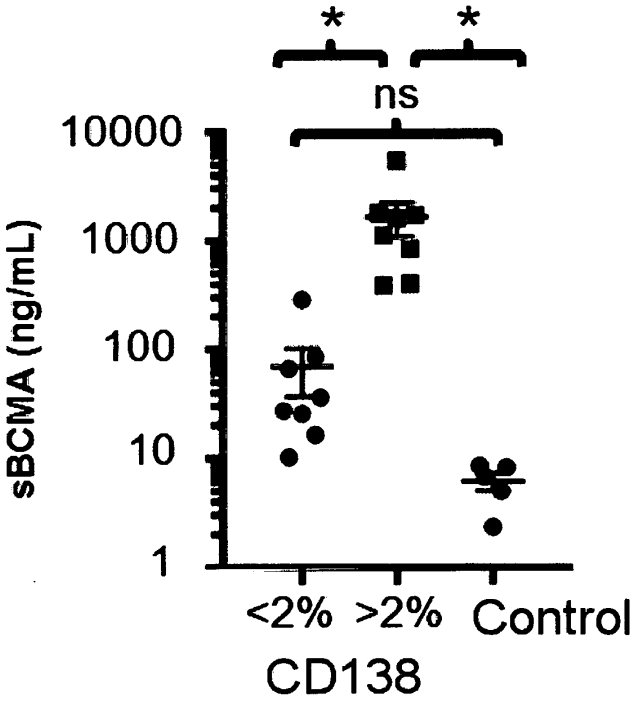

*
*
ns
10000
1000
100
10
1
sBCMA (ng/mL)
<2%
>2%
Control
CD138

*FIG. 2K*

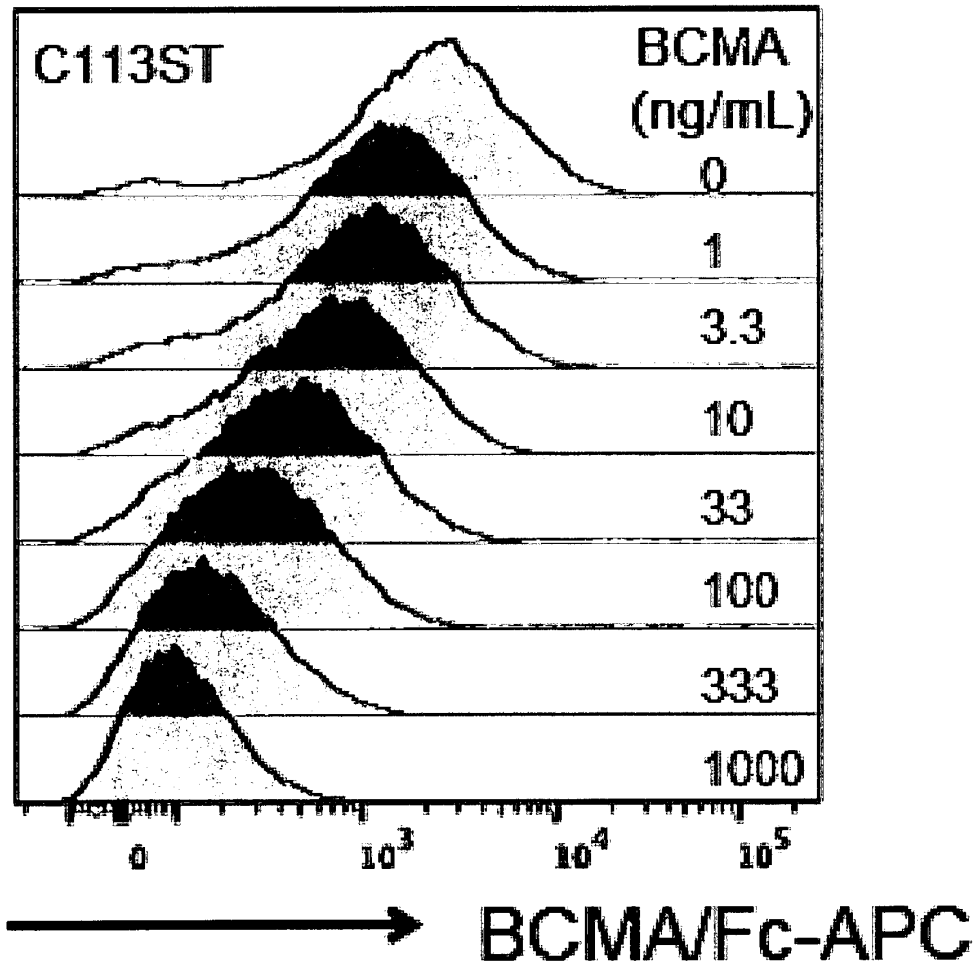

C113ST
BCMA (ng/mL)
0
1
3.3
10
33
100
333
1000
0
$10^3$
$10^4$
$10^5$
BCMA/Fc-APC

*FIG. 2L*

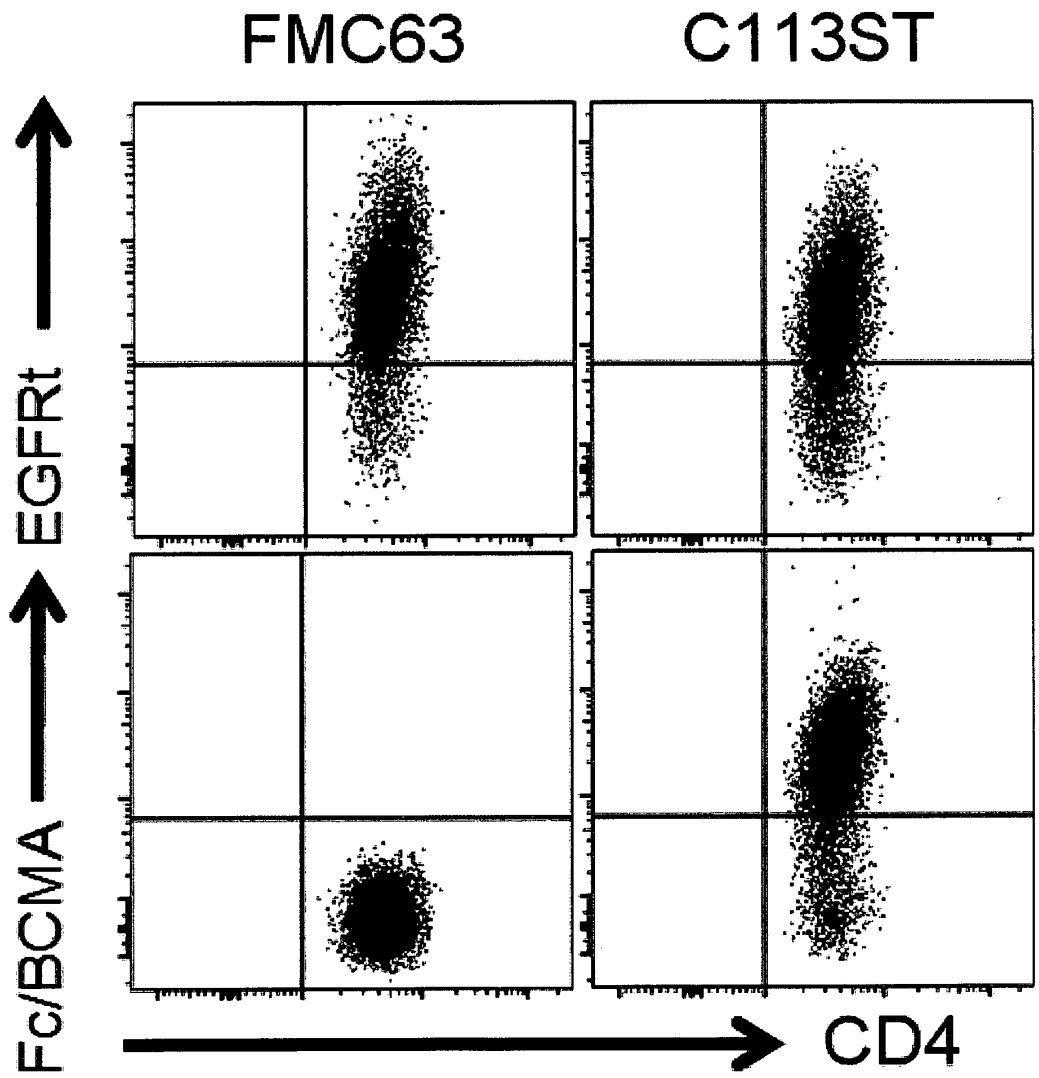



*FIG. 2M*

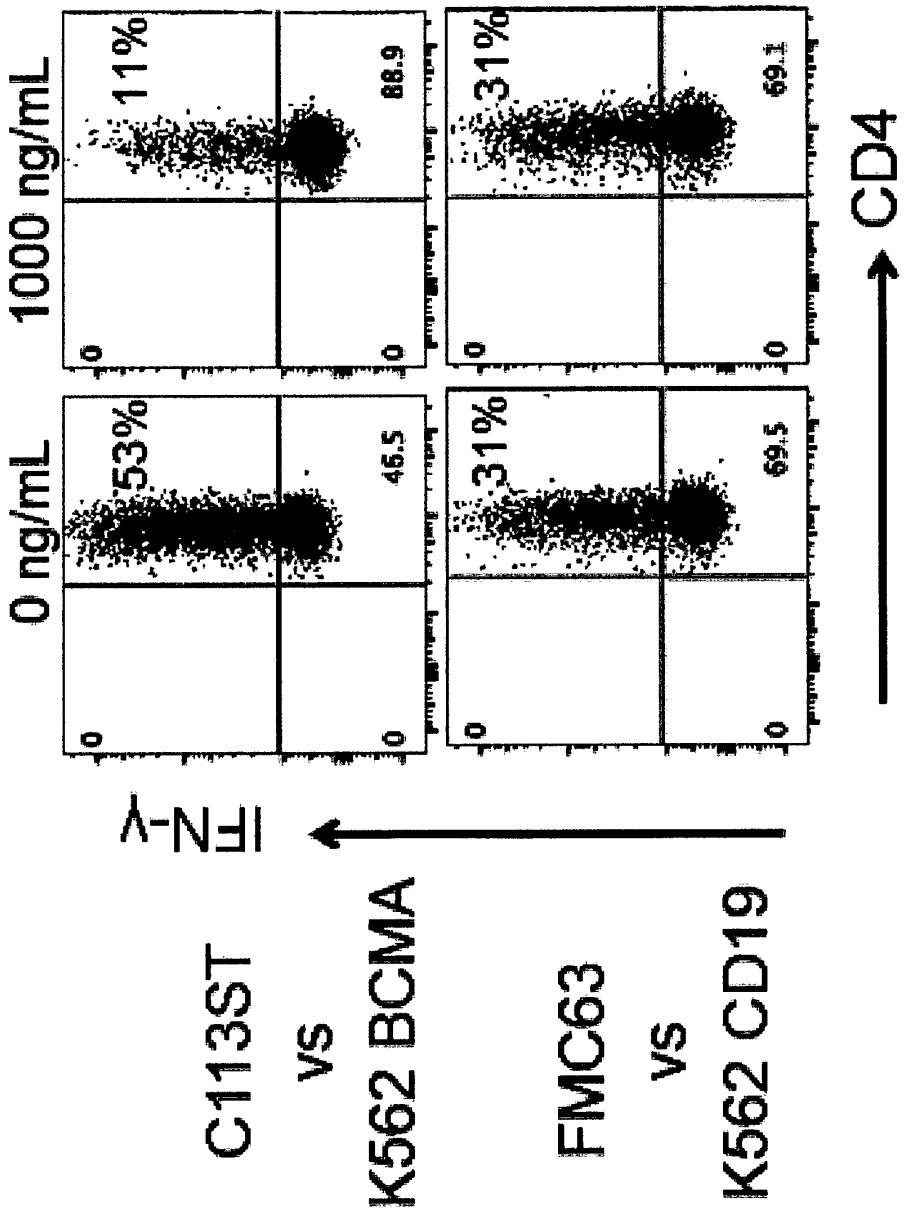

C113ST
vs
K562 BCMA
FMC63
vs
K562 CD19
IFN-γ
CD4
0 ng/mL
1000 ng/mL
53%
46.5
11%
88.9
31%
69.5
31%
69.1
0
0

*FIG. 2N*

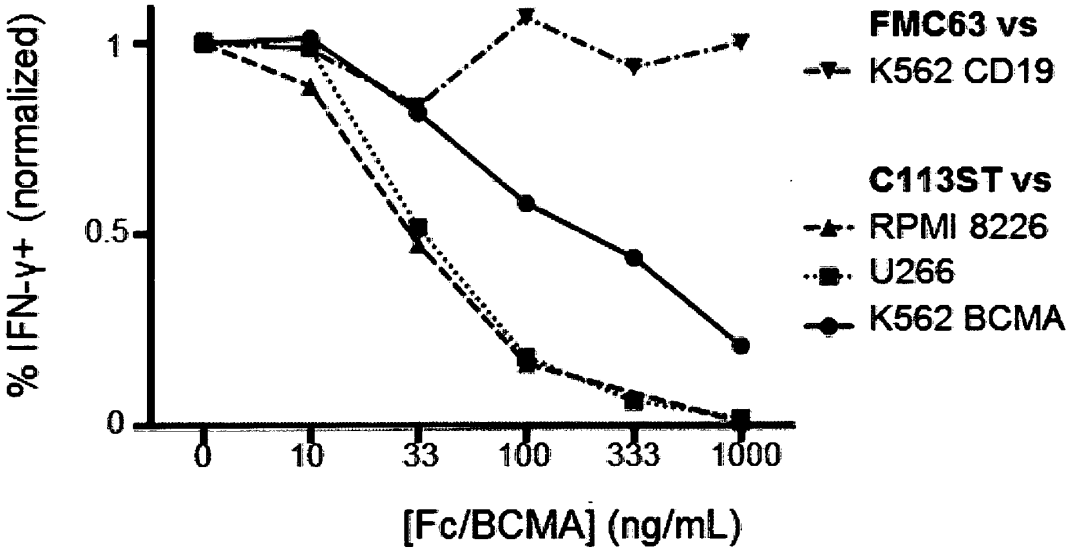

% IFN-γ+ (normalized)
1
0.5
0
0
10
33
100
333
1000
[Fc/BCMA] (ng/mL)
FMC63 vs
K562 CD19
C113ST vs
RPMI 8226
U266
K562 BCMA

*FIG. 2O*

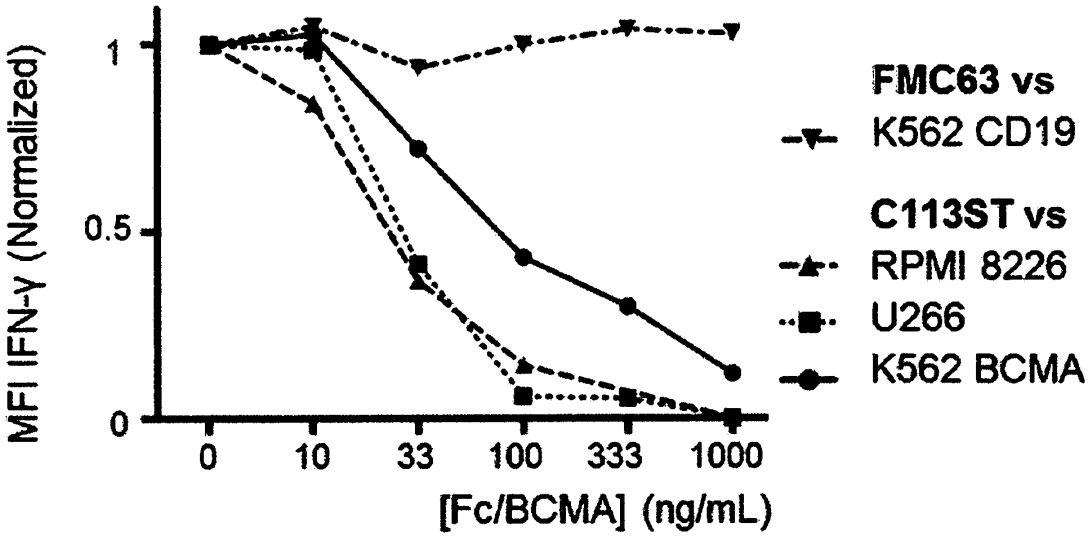

MFI IFN-γ (Normalized)
1
0.5
0
0
10
33
100
333
1000
[Fc/BCMA] (ng/mL)
FMC63 vs
K562 CD19
C113ST vs
RPMI 8226
U266
K562 BCMA

*FIG. 2P*

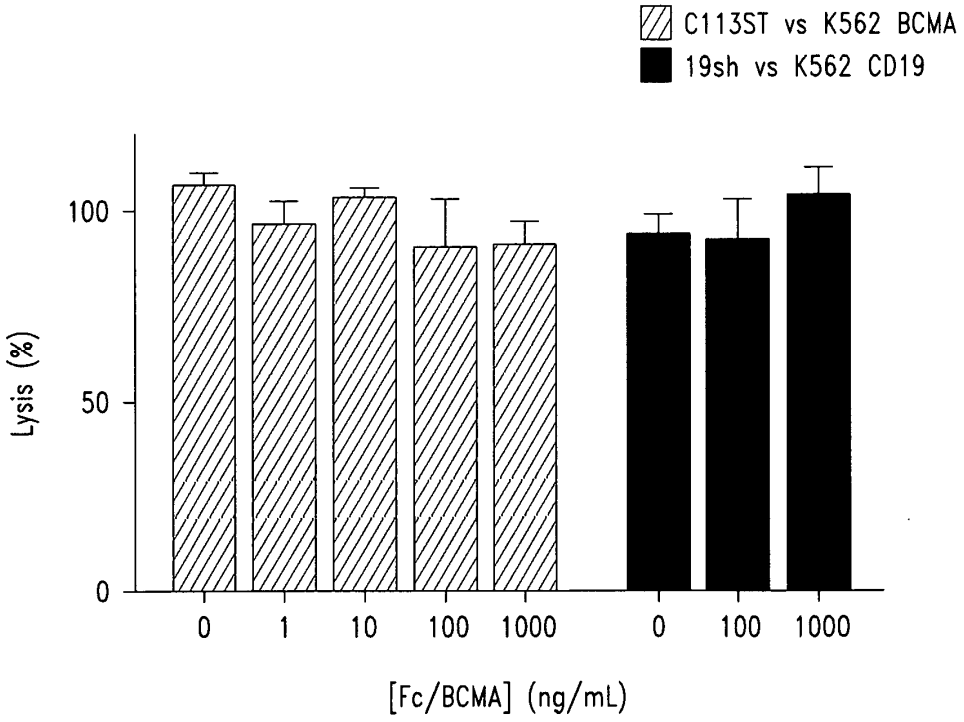

C113ST vs K562 BCMA
19sh vs K562 CD19
Lysis (%)
100
50
0
0
1
10
100
1000
0
100
1000
[Fc/BCMA] (ng/mL)

*FIG. 2Q*

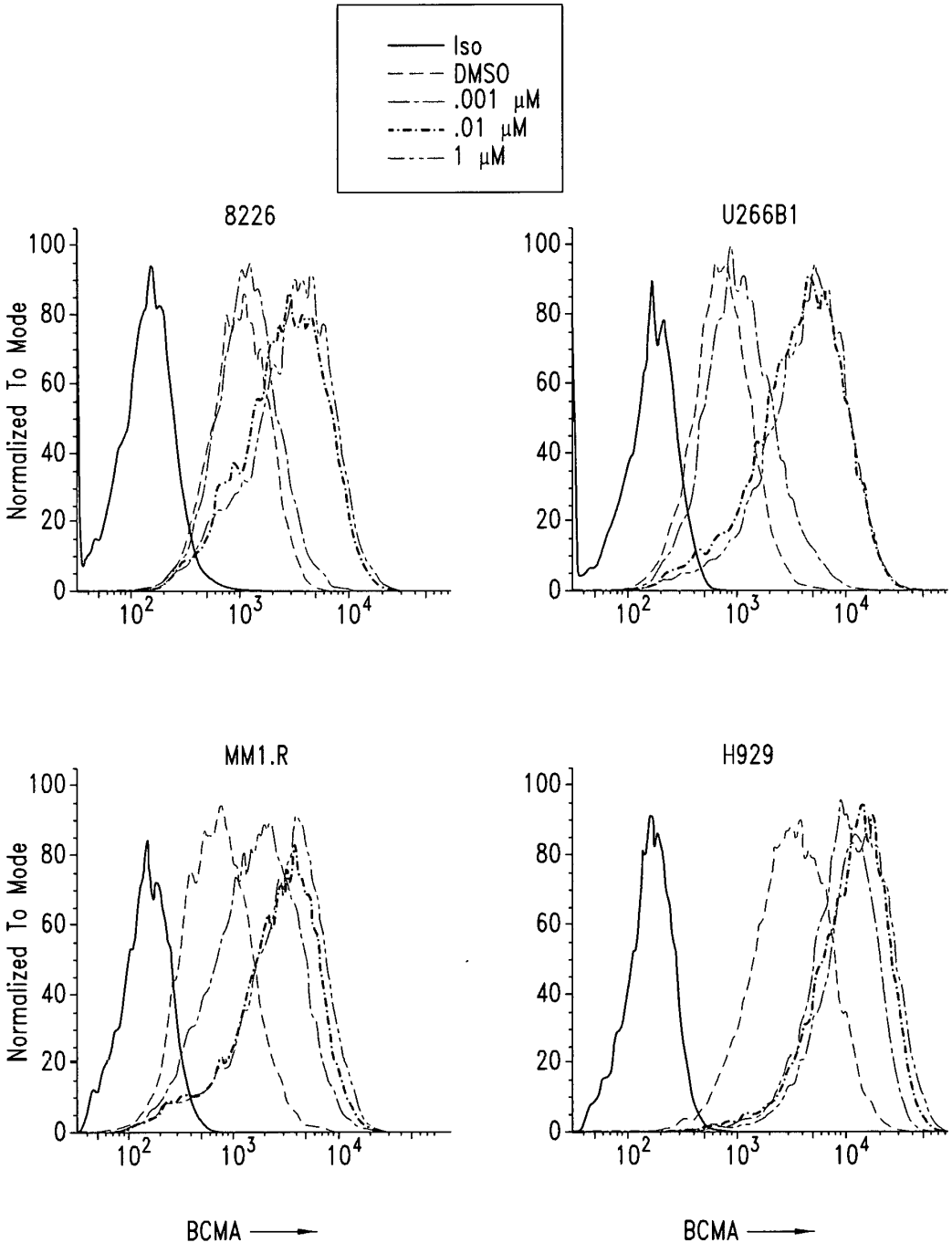

Iso
DMSO
.001 μM
.01 μM
1 μM
8226
U266B1
MM1.R
H929
Normalized To Mode
100
80
60
40
20
0
$10^2$
$10^3$
$10^4$
BCMA

*FIG. 3A*

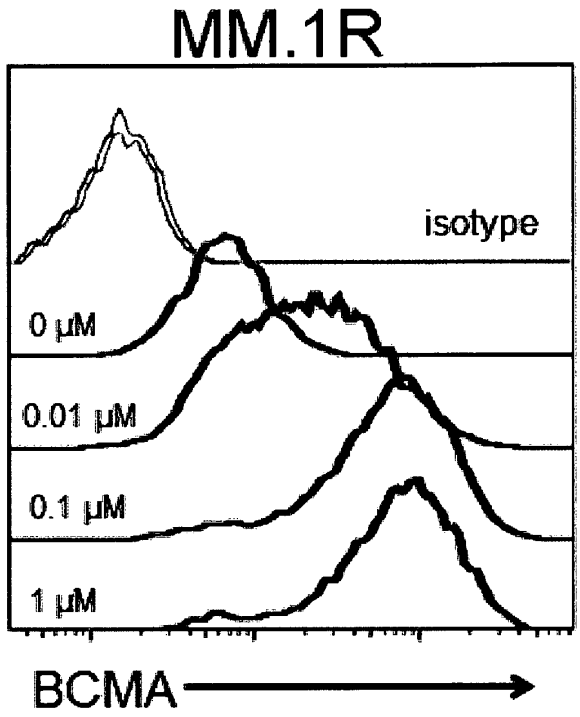

MM.1R
isotype
0 μM
0.01 μM
0.1 μM
1 μM
BCMA

*FIG. 3B*

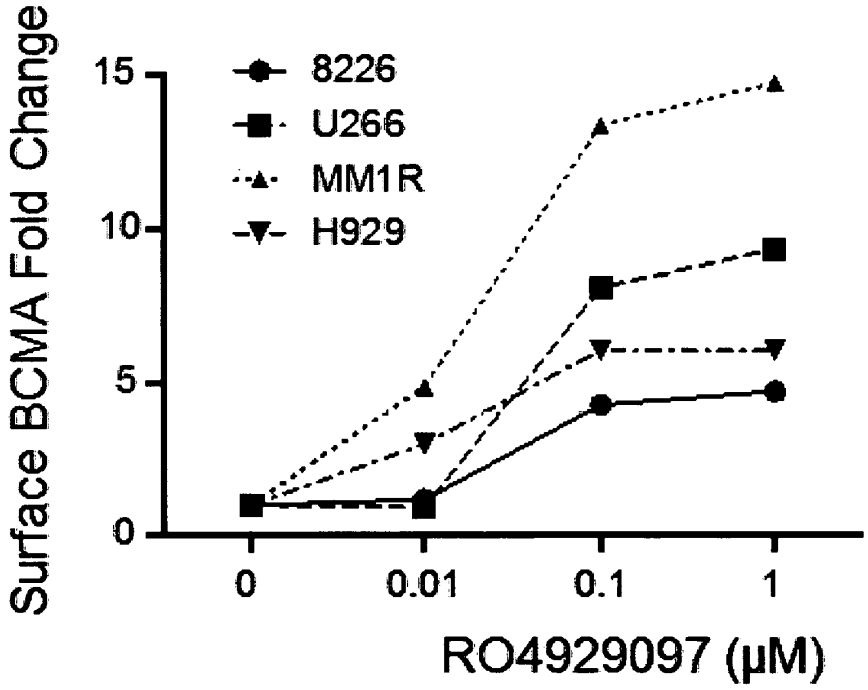

Surface BCMA Fold Change
15
10
5
0
8226
U266
MM1R
H929
0
0.01
0.1
1
RO4929097 (μM)

*FIG. 3C*

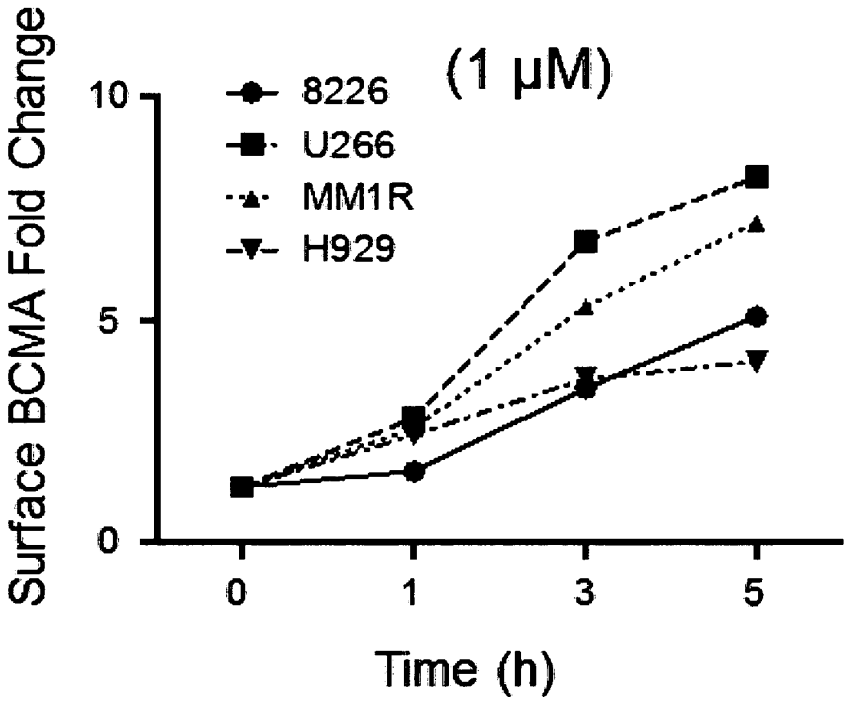

Surface BCMA Fold Change
(1 μM)
8226
U266
MM1R
H929
10
5
0
0
1
3
5
Time (h)

*FIG. 3D*

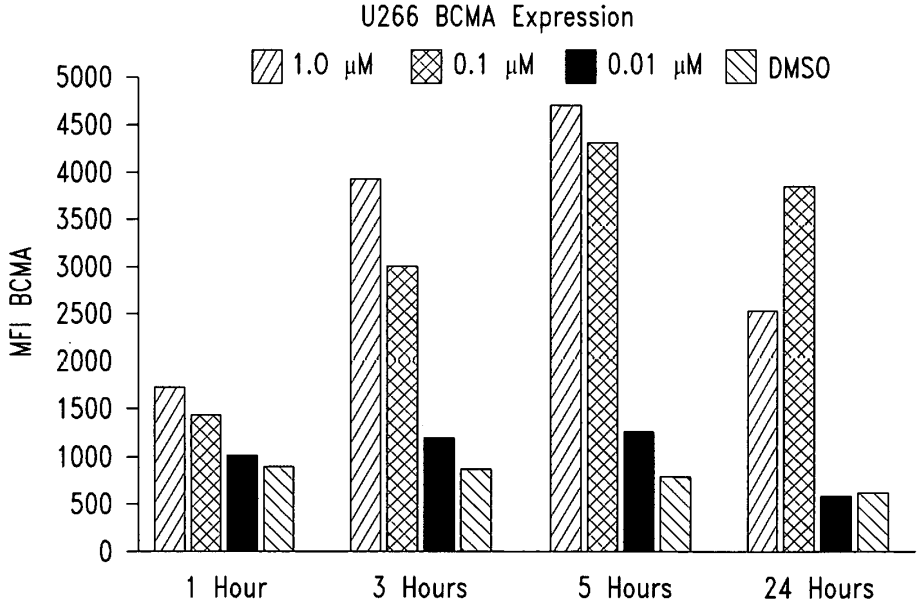

U266 BCMA Expression
1.0 μM
0.1 μM
0.01 μM
DMSO
MFI BCMA
5000
4500
4000
3500
3000
2500
2000
1500
1000
500
0
1 Hour
3 Hours
5 Hours
24 Hours

*FIG. 3E*

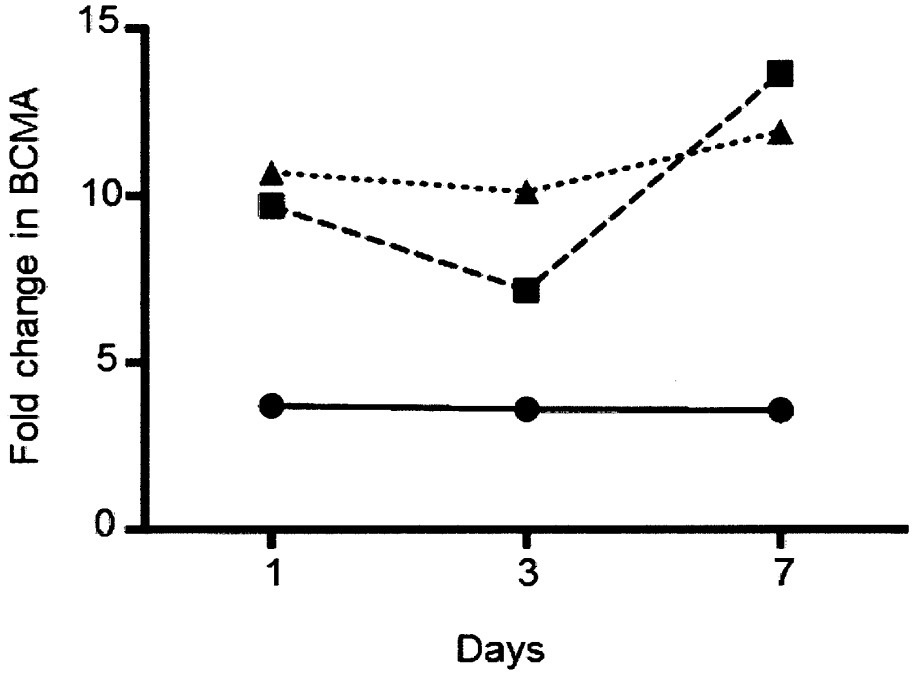

15
10
5
0
Fold change in BCMA
1
3
7
Days

*FIG. 3F*

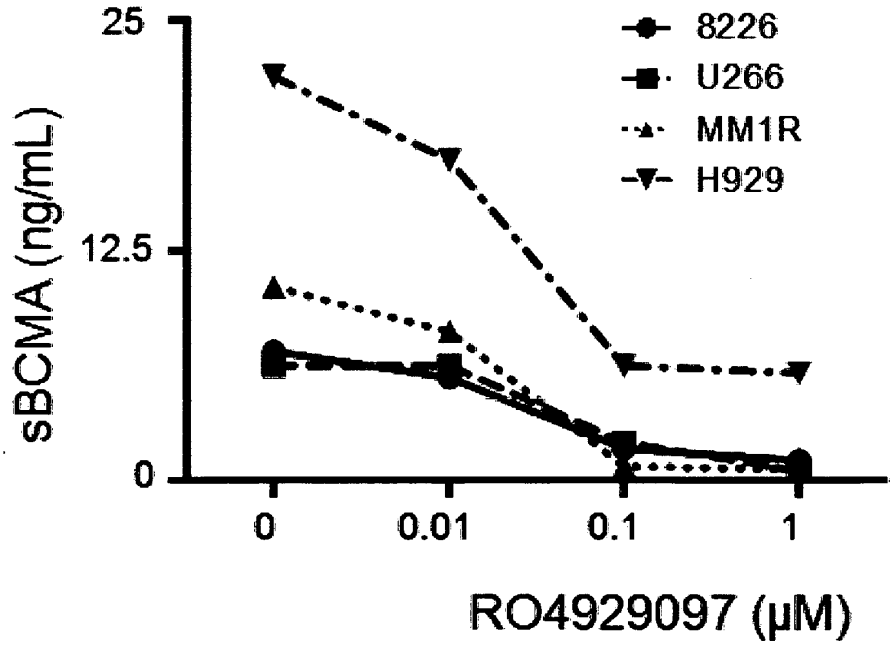

25
12.5
0
sBCMA (ng/mL)
0
0.01
0.1
1
RO4929097 (μM)
8226
U266
MM1R
H929

*FIG. 3G*

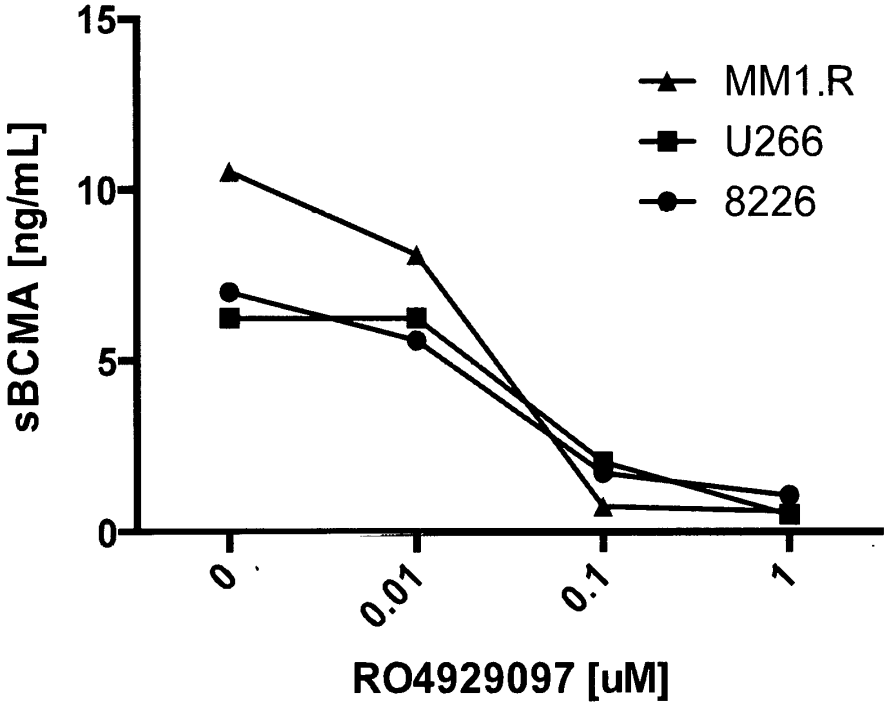



*FIG. 3H*

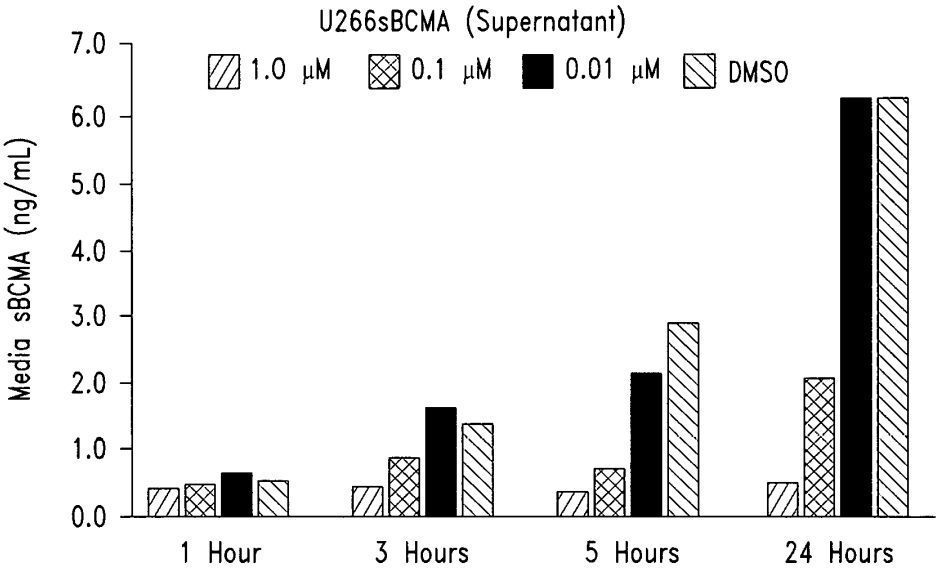

U266sBCMA (Supernatant)
1.0 µM
0.1 µM
0.01 µM
DMSO
Media sBCMA (ng/mL)
7.0
6.0
5.0
4.0
3.0
2.0
1.0
0.0
1 Hour
3 Hours
5 Hours
24 Hours

*FIG. 3I*

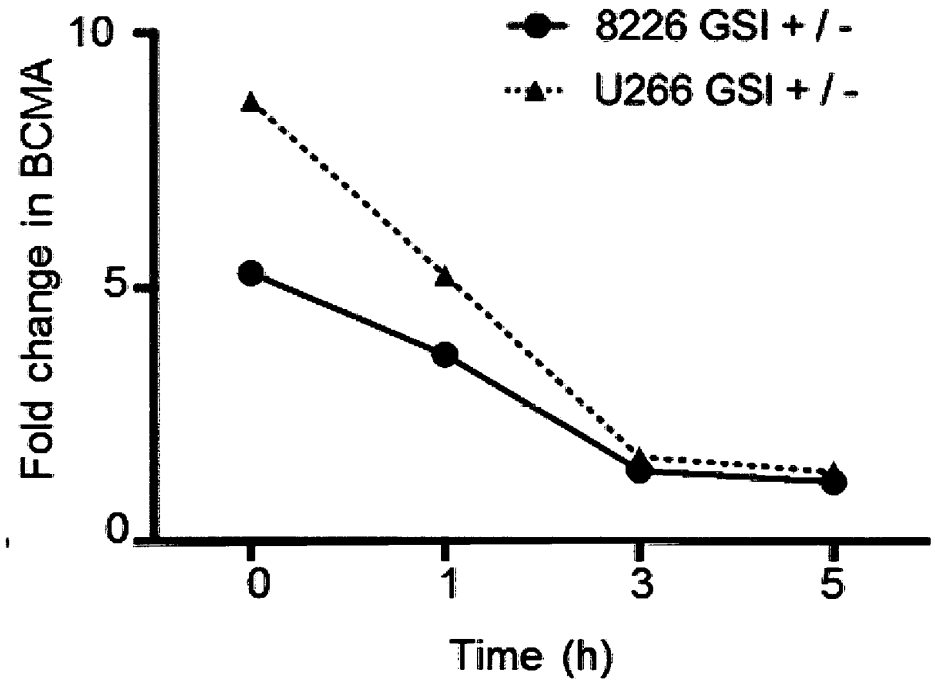

8226 GSI + / -
U266 GSI + / -
Fold change in BCMA
10
5
0
0
1
3
5
Time (h)

*FIG. 3J*

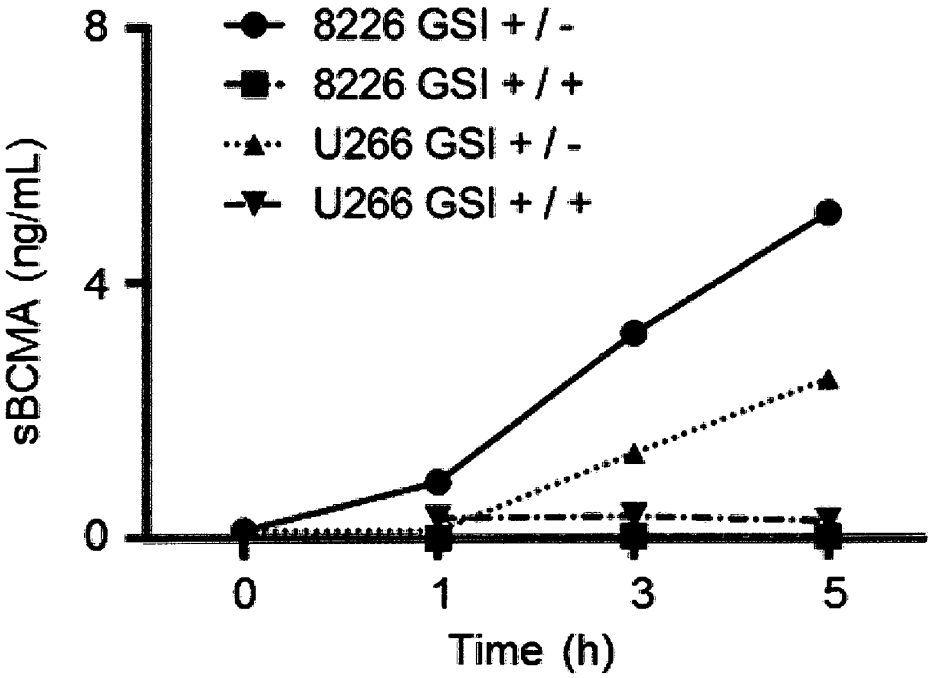



*FIG. 3K*

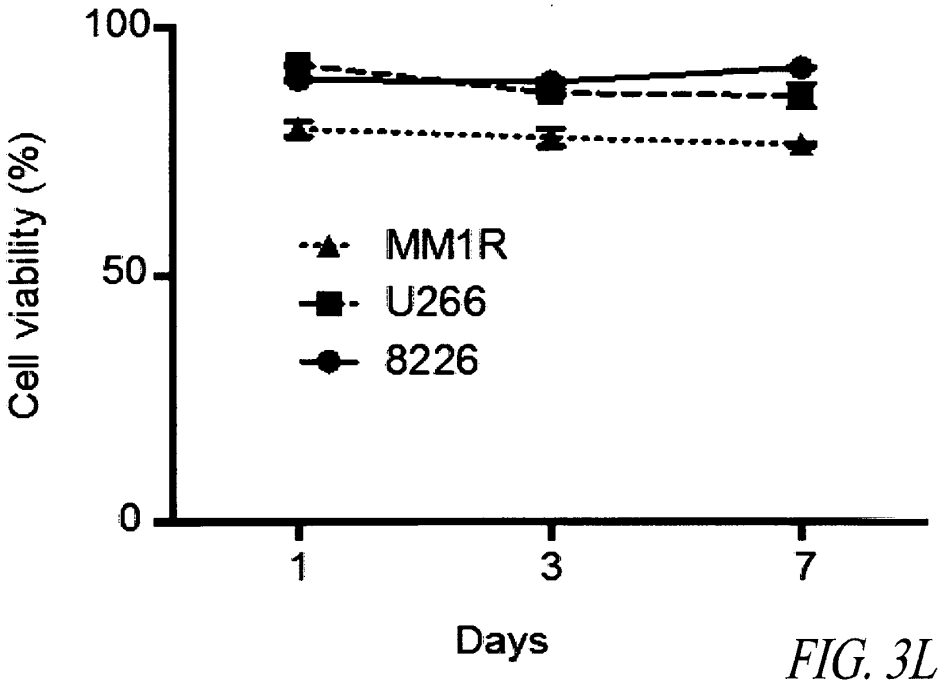

100
50
0
Cell viability (%)
MM1R
U266
8226
1
3
7
Days

*FIG. 3L*

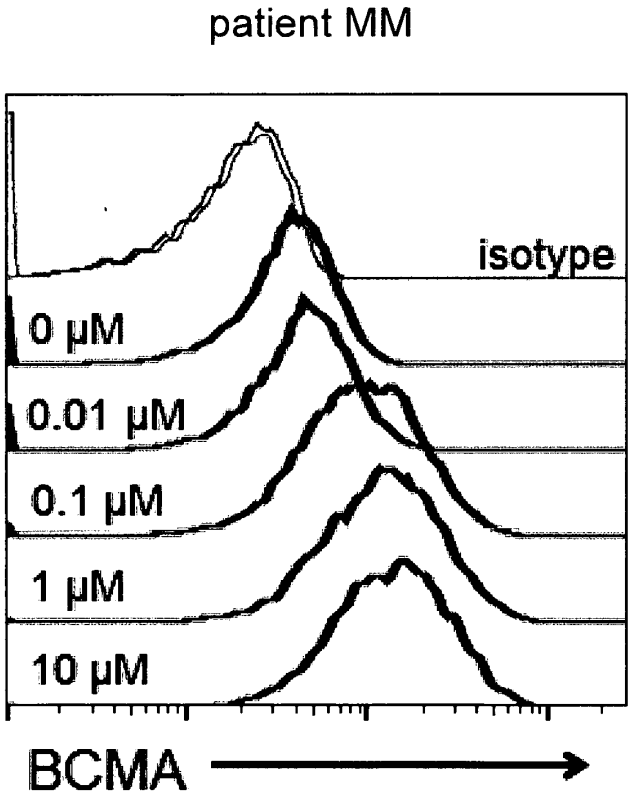

patient MM
isotype
0 μM
0.01 μM
0.1 μM
1 μM
10 μM
BCMA

*FIG. 3M*

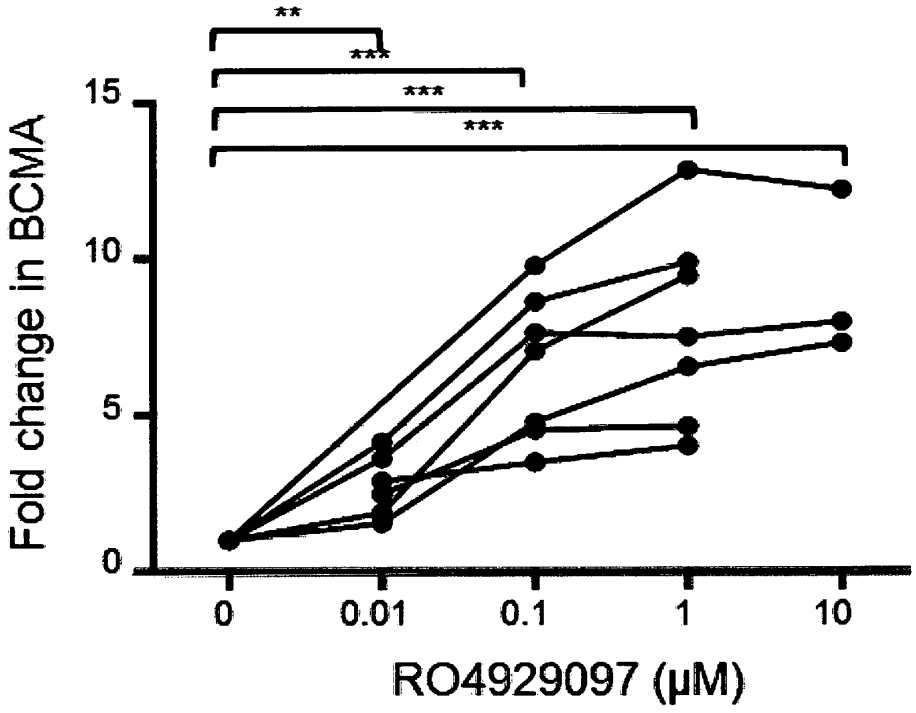



*FIG. 3N*

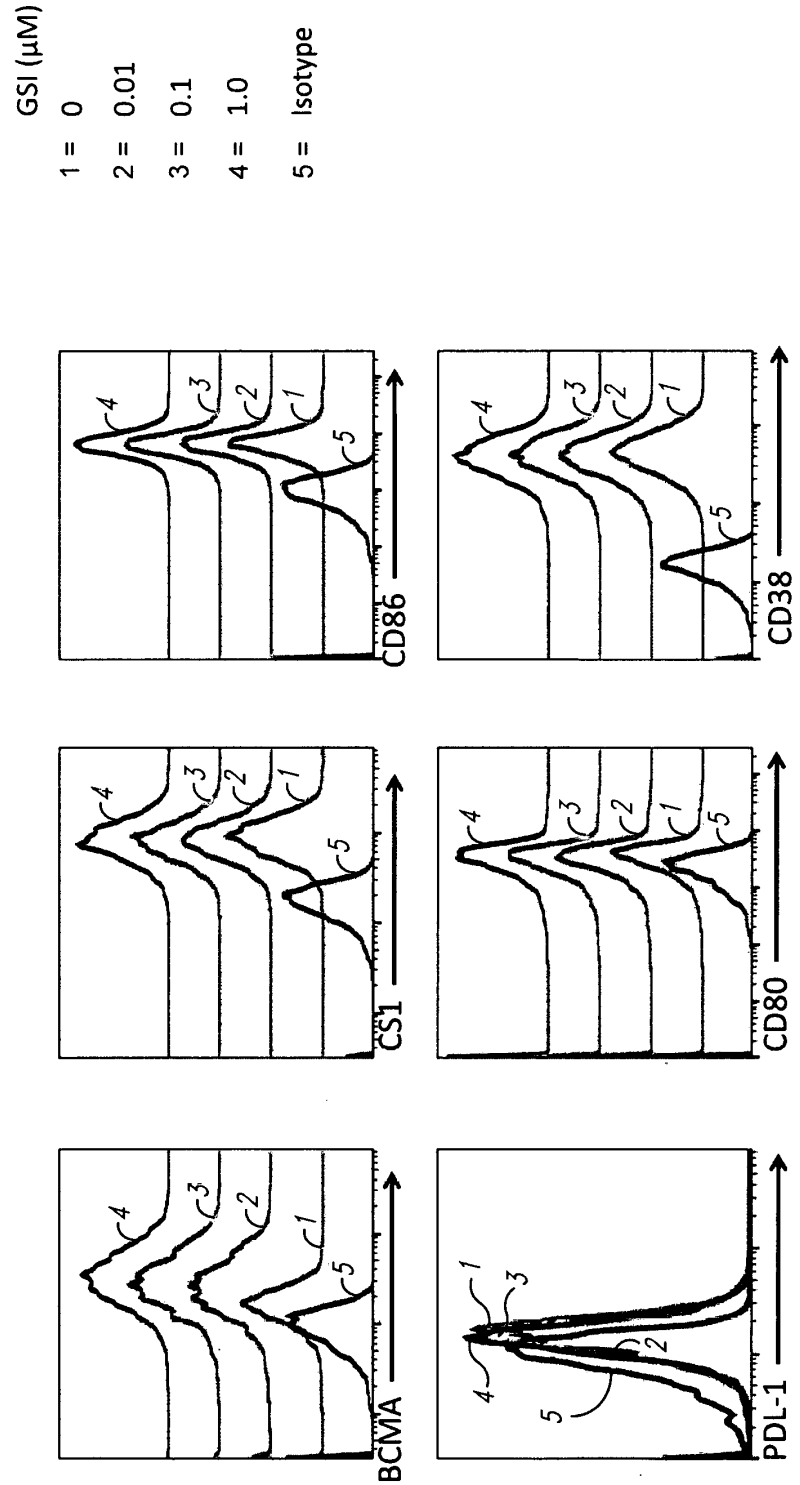



FIG. 3O

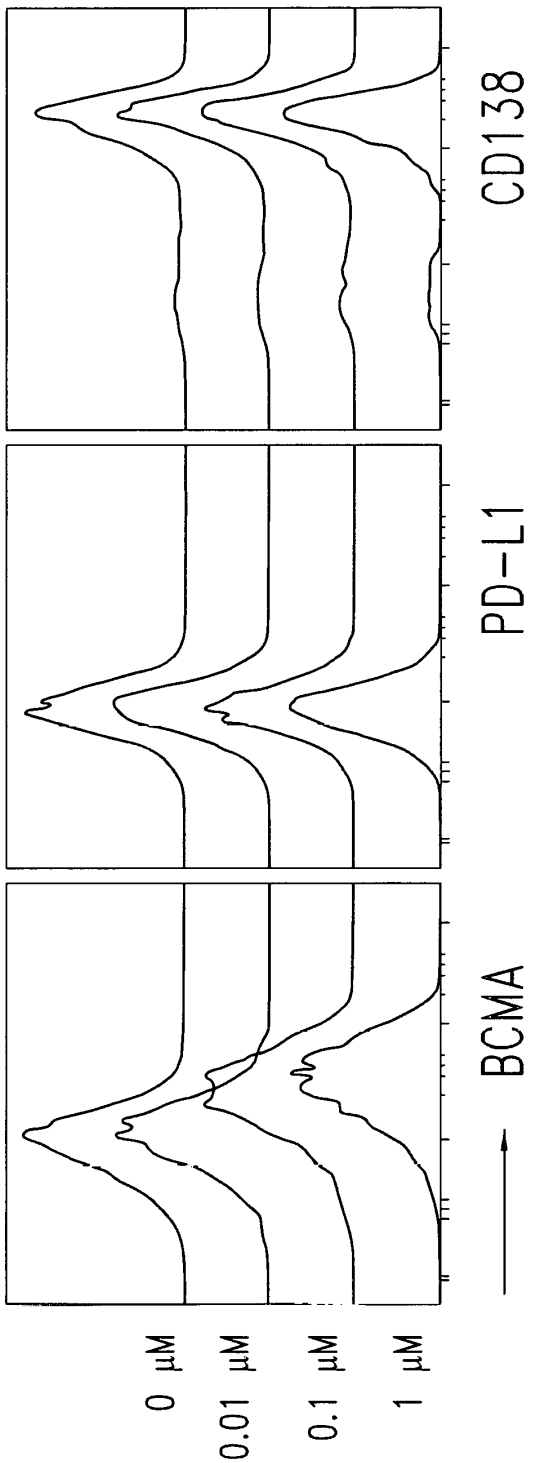



*FIG. 3P*

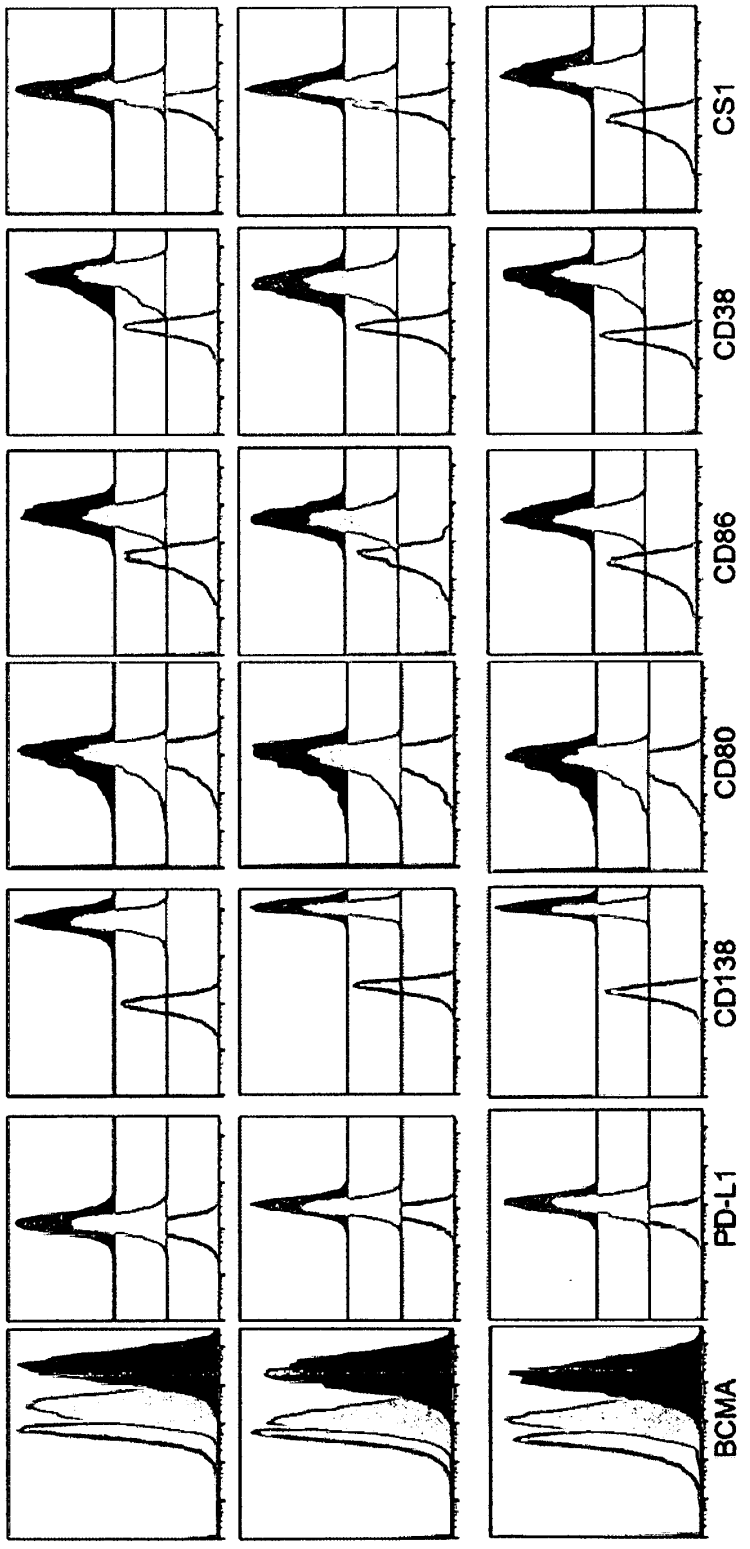



*FIG. 3Q*

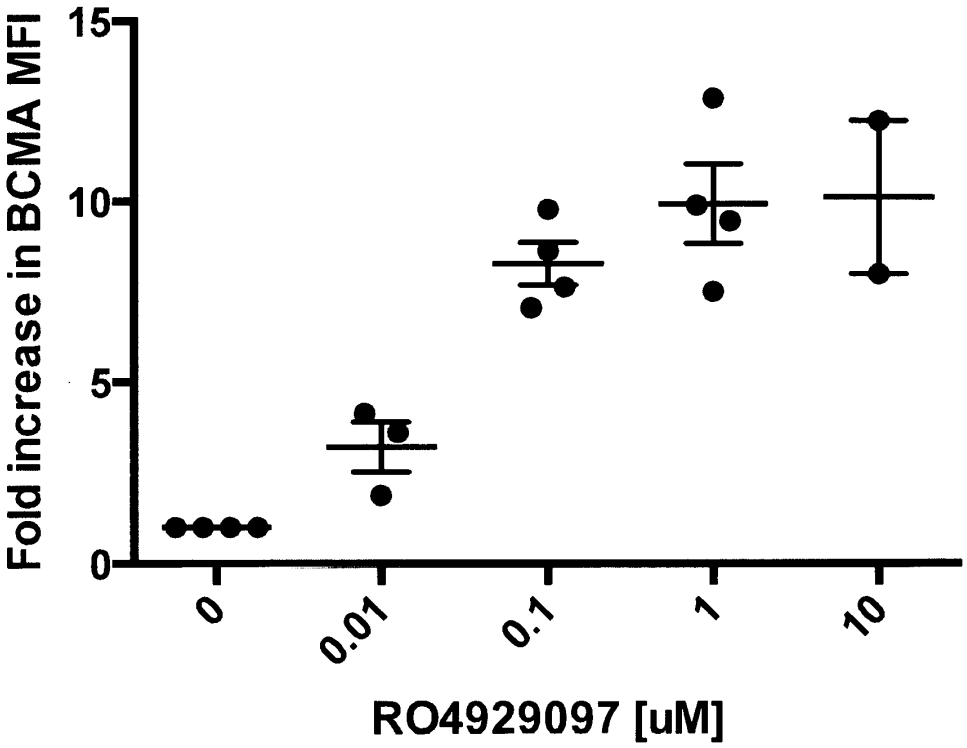



*FIG. 3R*

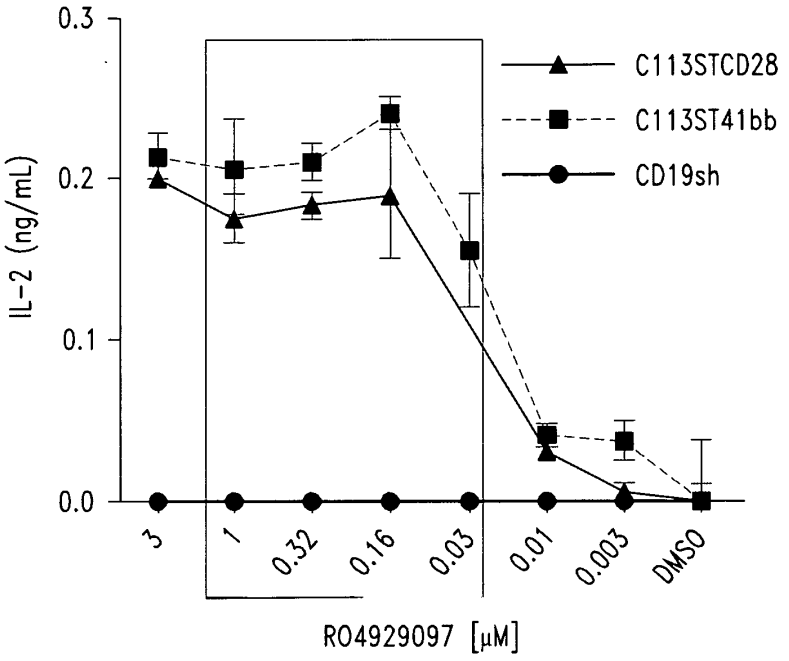

0.3
0.2
0.1
0.0
IL-2 (ng/mL)
C113STCD28
C113ST41bb
CD19sh
3
1
0.32
0.16
0.03
0.01
0.003
DMSO
RO4929097 [μM]

*FIG. 4A*

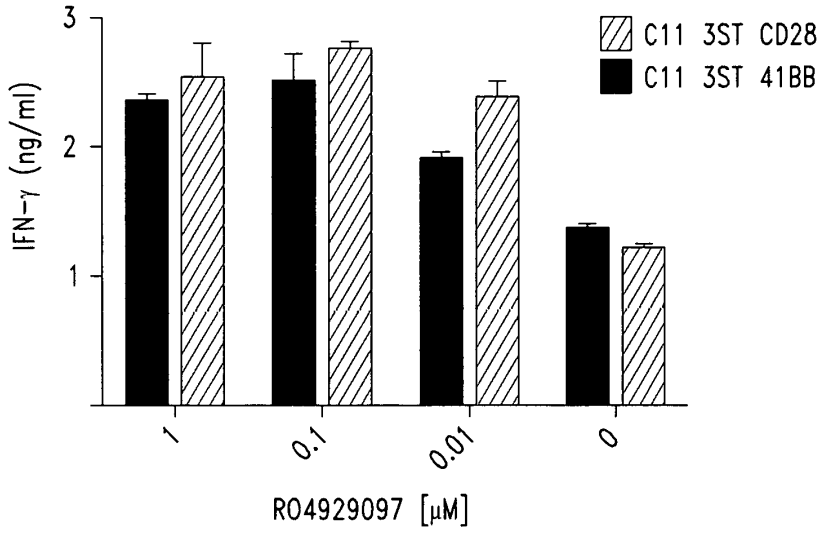

3
2
1
IFN-γ (ng/ml)
C11 3ST CD28
C11 3ST 41BB
1
0.1
0.01
0
RO4929097 [μM]

*FIG. 4B*

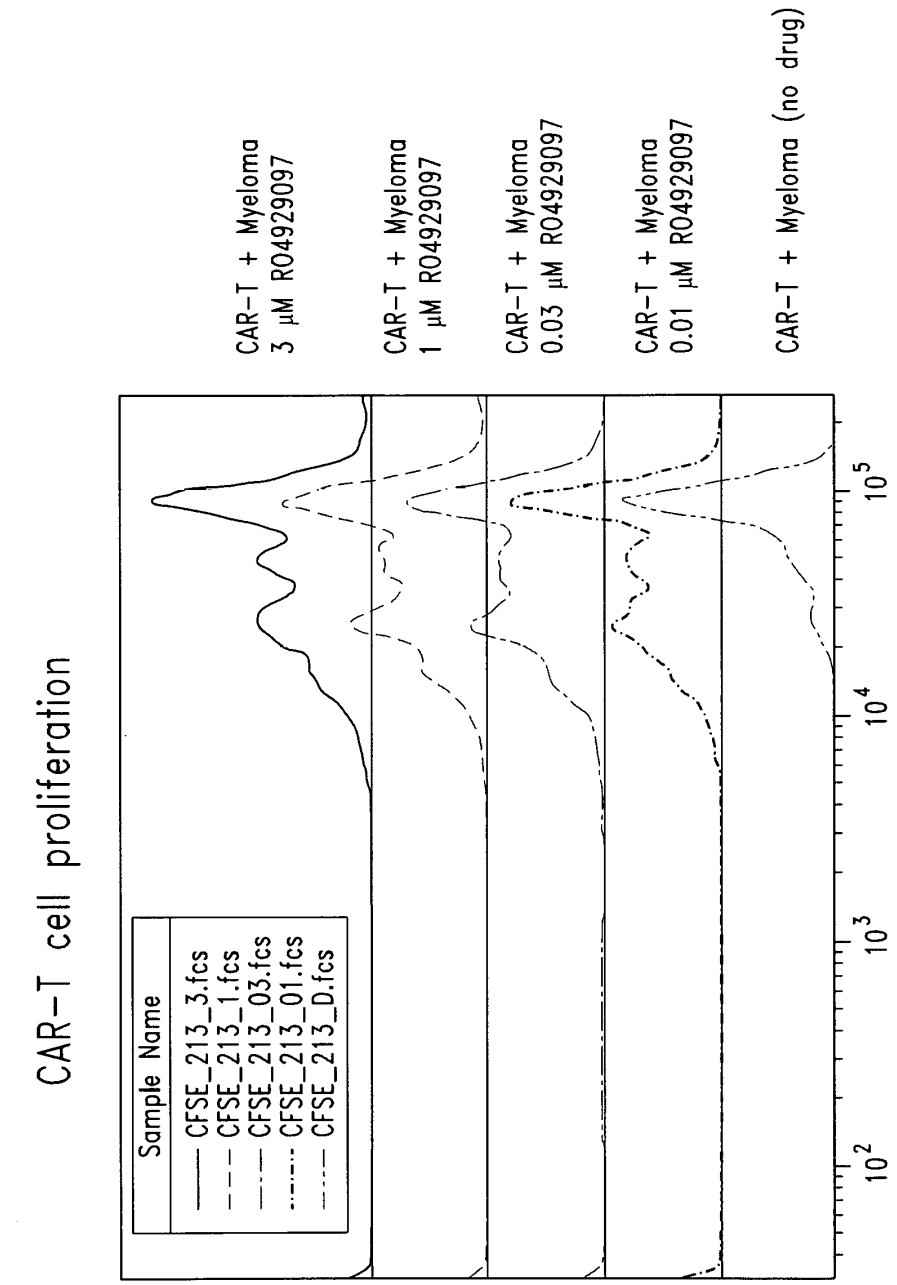

CAR-T cell proliferation
Sample Name
CFSE_213_3.fcs
CFSE_213_1.fcs
CFSE_213_03.fcs
CFSE_213_01.fcs
CFSE_213_D.fcs
CAR-T + Myeloma 3 µM RO4929097
CAR-T + Myeloma 1 µM RO4929097
CAR-T + Myeloma 0.03 µM RO4929097
CAR-T + Myeloma 0.01 µM RO4929097
CAR-T + Myeloma (no drug)
$10^2$
$10^3$
$10^4$
$10^5$
FITC-A :: CFSE

*FIG. 4C*

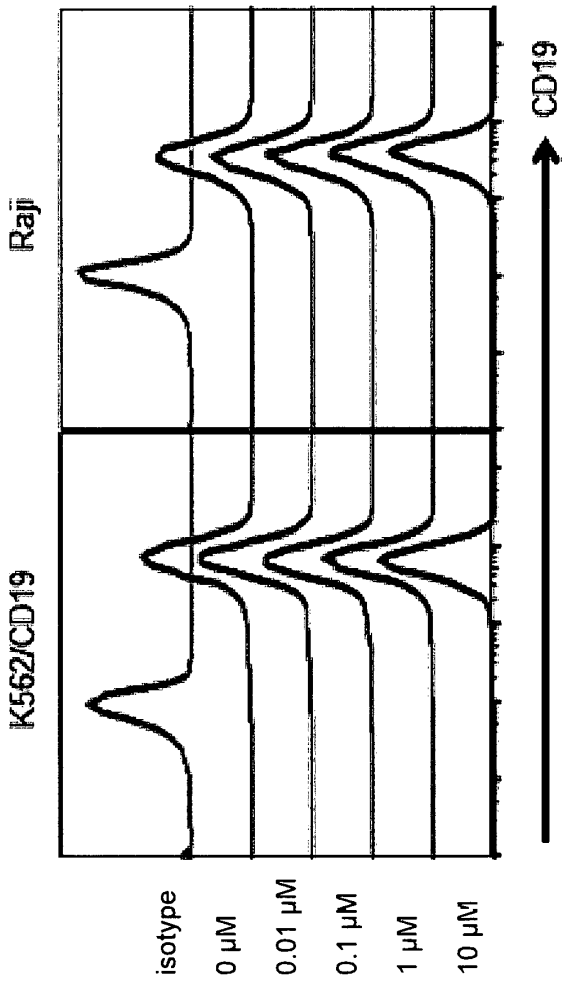



*FIG. 5A*

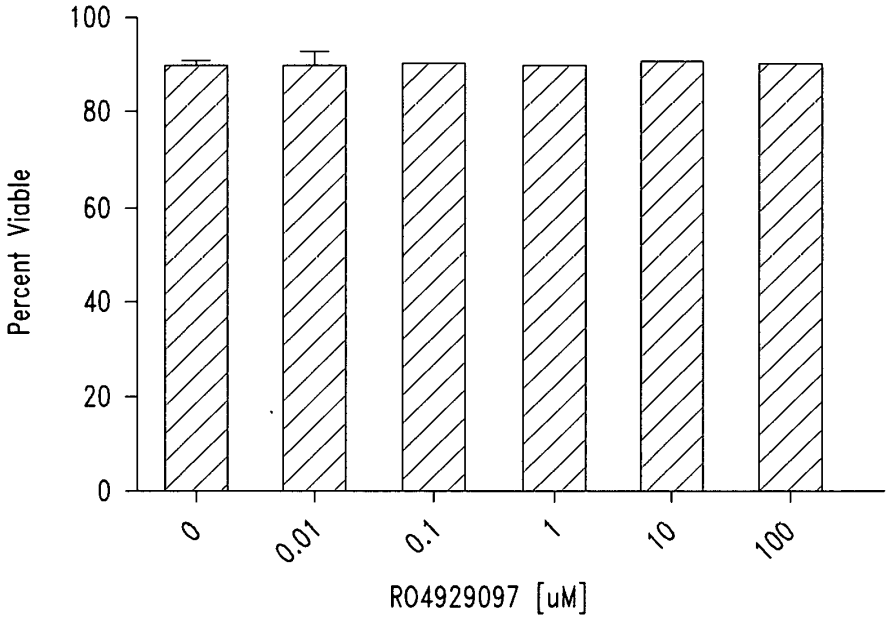



*FIG. 5B*

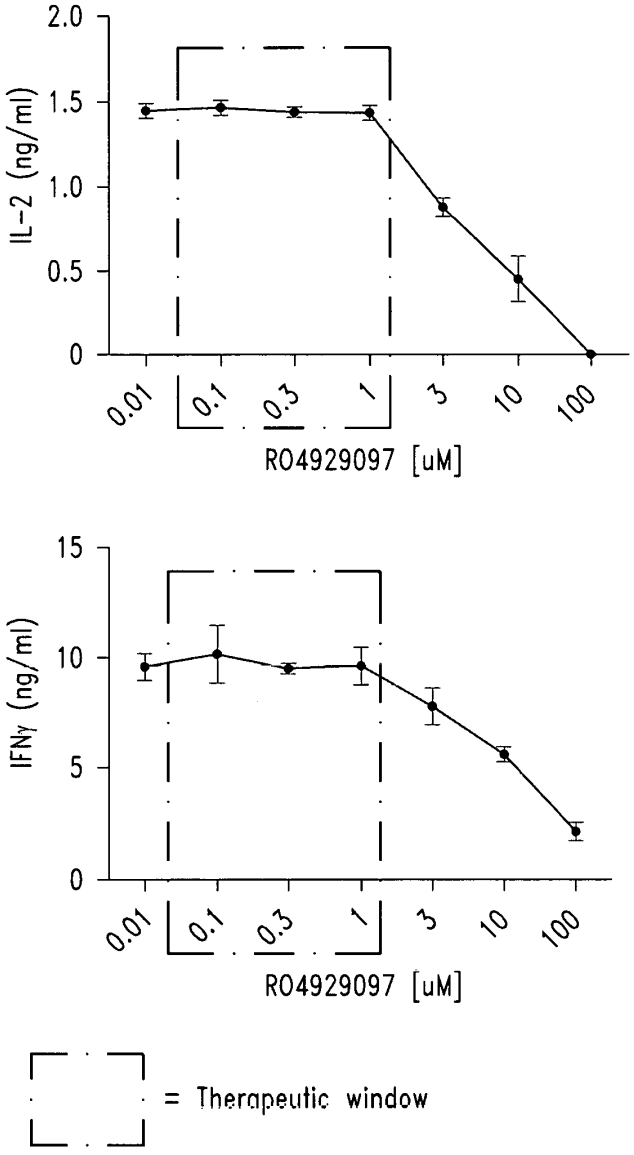

2.0
1.5
1.0
0.5
0
IL-2 (ng/ml)
0.01
0.1
0.3
1
3
10
100
RO4929097 [uM]
15
10
5
0
IFNγ (ng/ml)
0.01
0.1
0.3
1
3
10
100
RO4929097 [uM]
= Therapeutic window

*FIG. 5C*

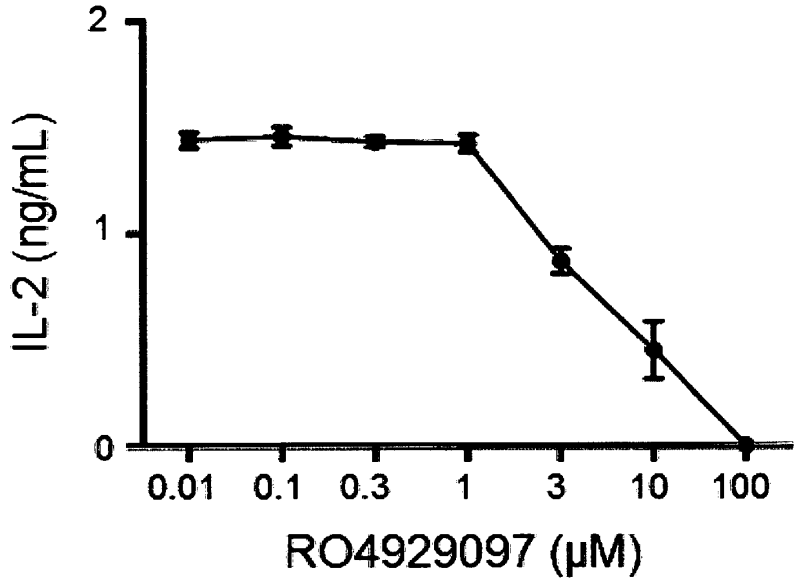

2
1
0
IL-2 (ng/mL)
0.01
0.1
0.3
1
3
10
100
RO4929097 (μM)

*FIG. 5D*

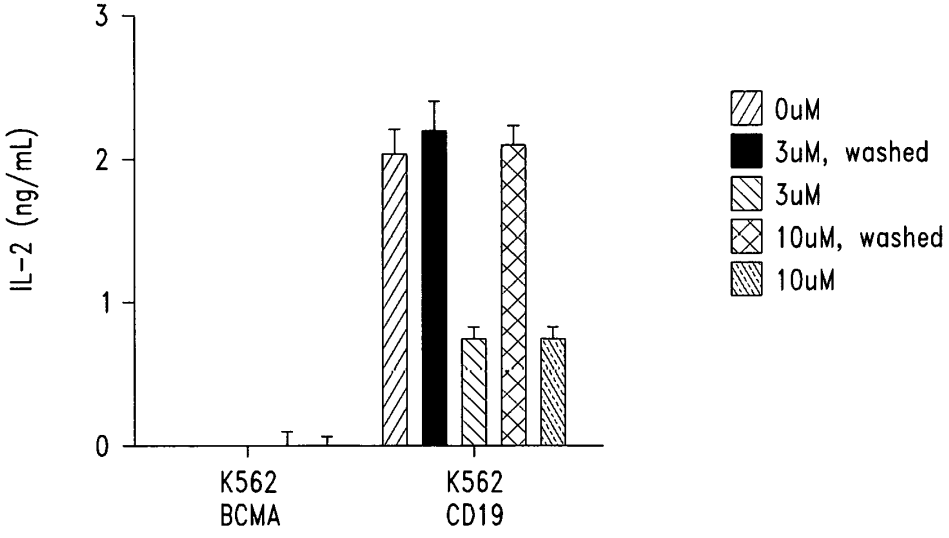

3
2
1
0
IL-2 (ng/mL)
K562
BCMA
K562
CD19
0uM
3uM, washed
3uM
10uM, washed
10uM

*FIG. 5E*

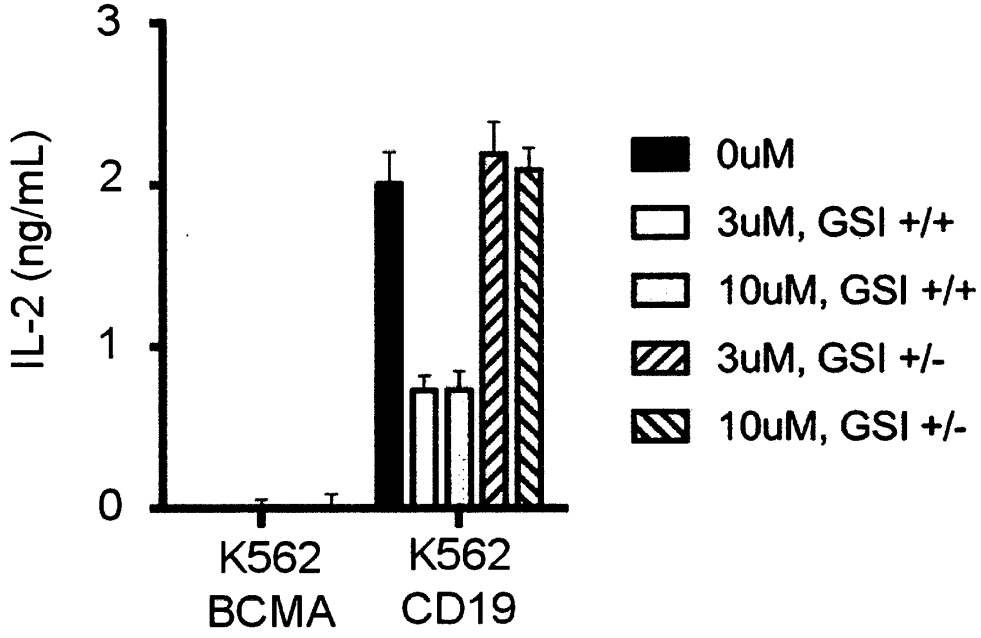

3
2
1
0
IL-2 (ng/mL)
K562 BCMA
K562 CD19
0uM
3uM, GSI +/+
10uM, GSI +/+
3uM, GSI +/-
10uM, GSI +/-

*FIG. 5F*

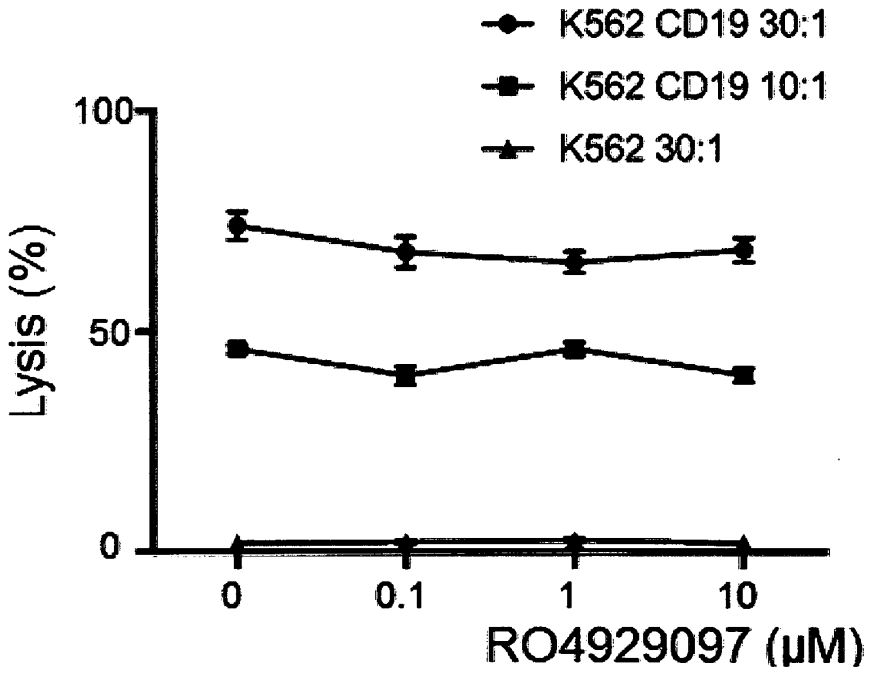

K562 CD19 30:1
K562 CD19 10:1
K562 30:1
100
50
0
Lysis (%)
0
0.1
1
10
RO4929097 (μM)

*FIG. 5G*

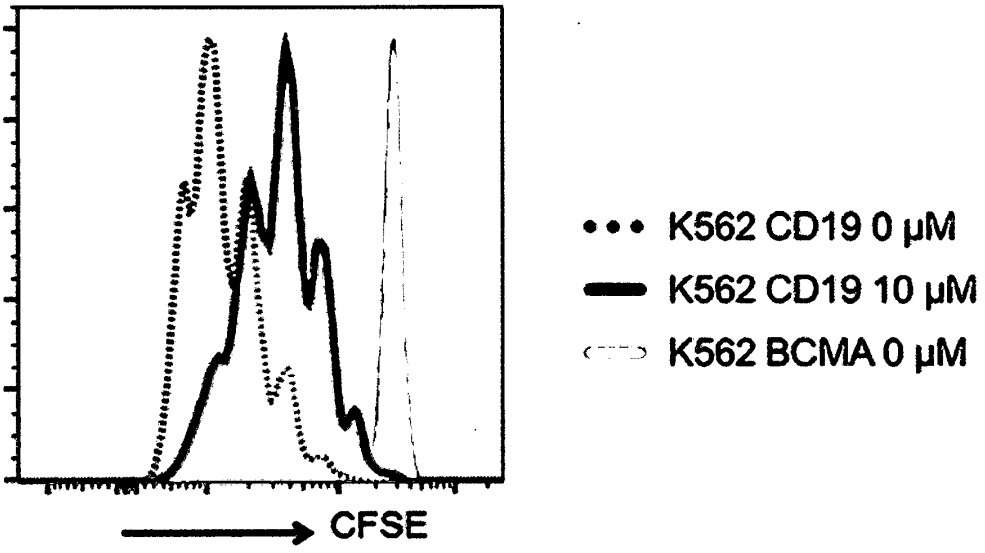

K562 CD19 0 μM
K562 CD19 10 μM
K562 BCMA 0 μM
CFSE

*FIG. 5H*

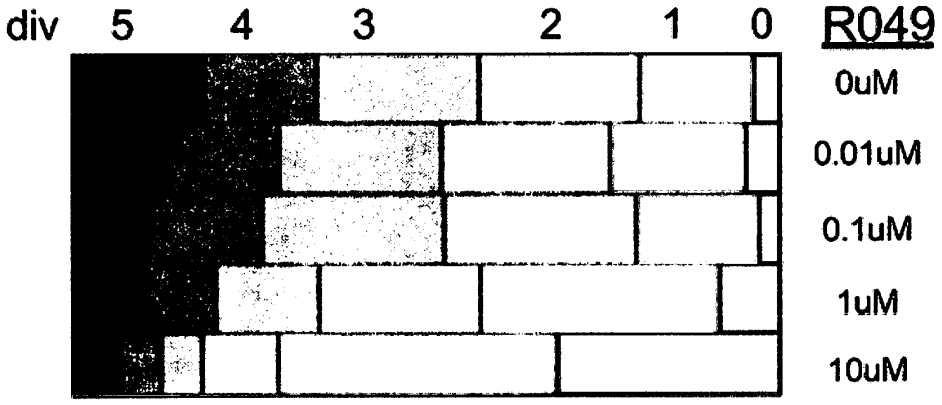

div 5 4 3 2 1 0
R049
0uM
0.01uM
0.1uM
1uM
10uM

*FIG. 5I*

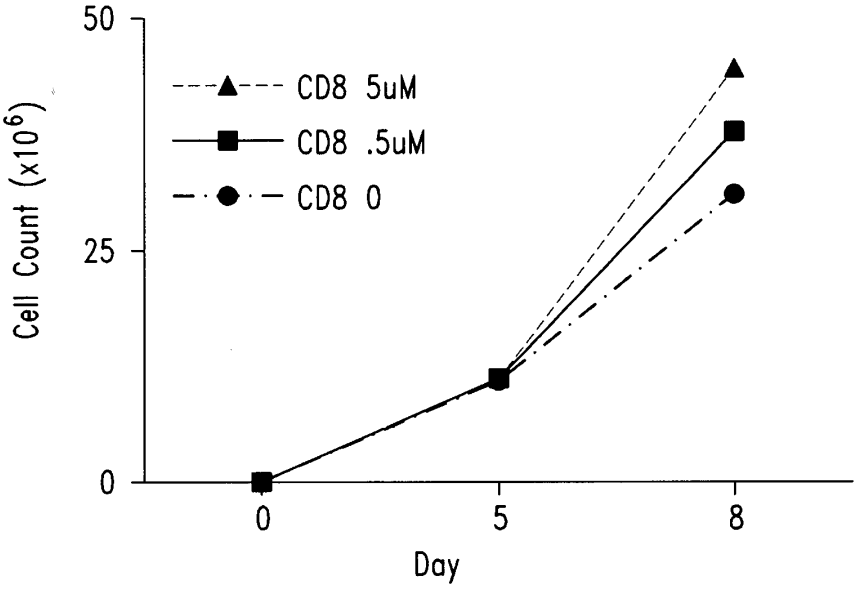

50
25
0
Cell Count (x10^6)
CD8 5uM
CD8 .5uM
CD8 0
0
5
8
Day

*FIG. 5J*

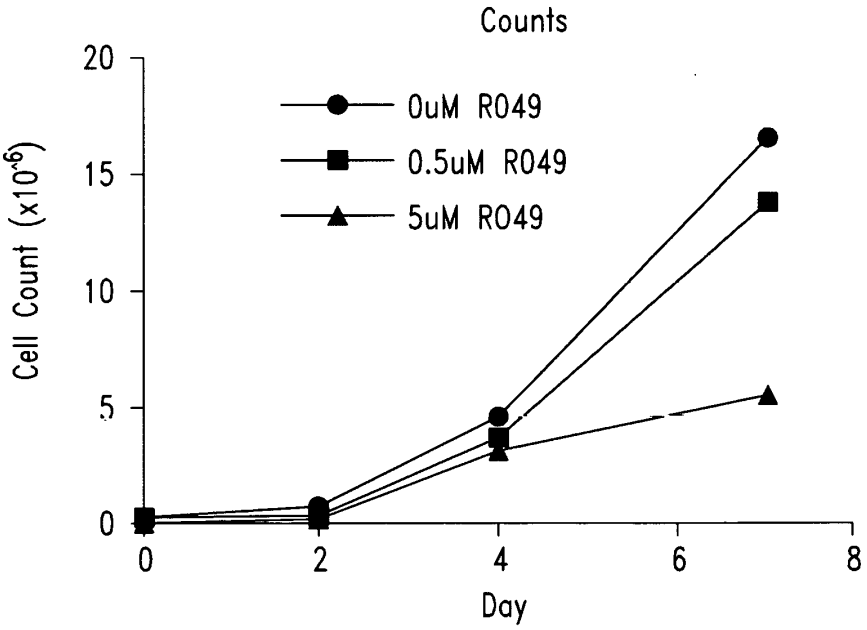

Counts
20
15
10
5
0
Cell Count (x10^6)
0uM R049
0.5uM R049
5uM R049
0
2
4
6
8
Day

*FIG. 5K*

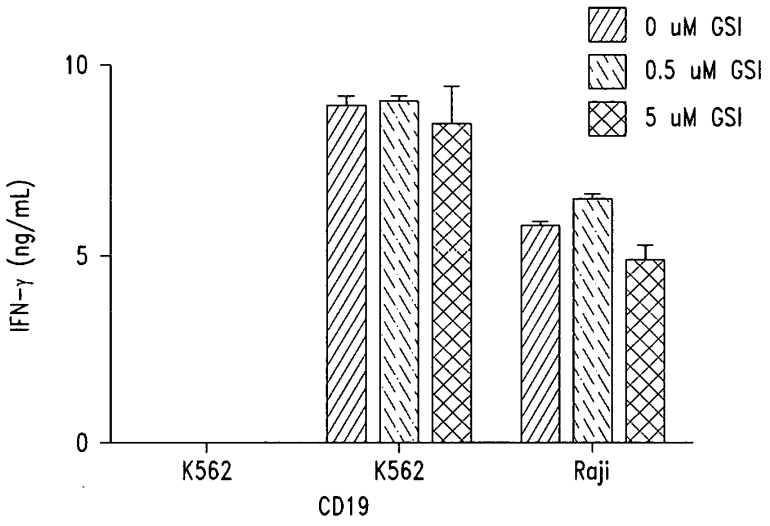



*FIG. 5L*

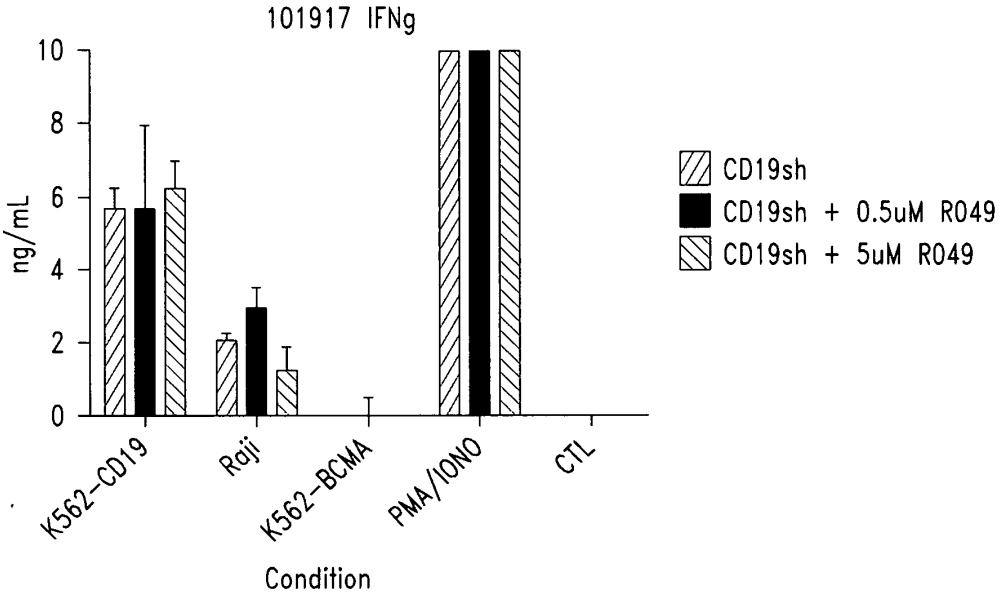

101917 IFNg
10
8
6
4
2
0
ng/mL
K562-CD19
Raji
K562-BCMA
PMA/IONO
CTL
Condition
CD19sh
CD19sh + 0.5uM R049
CD19sh + 5uM R049

*FIG. 5M*

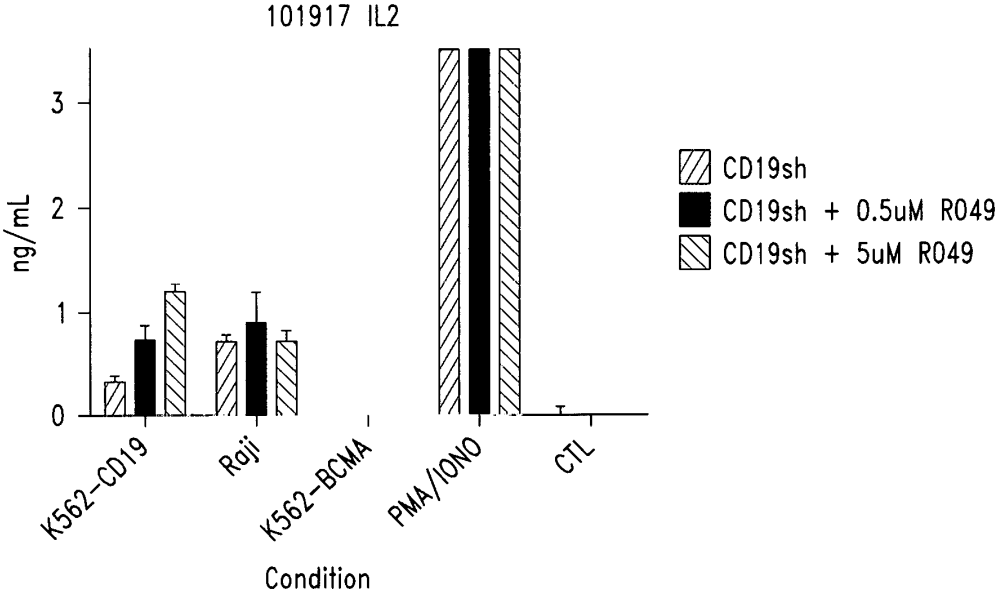

101917 IL2
3
2
1
0
ng/mL
K562-CD19
Raji
K562-BCMA
PMA/IONO
CTL
Condition
CD19sh
CD19sh + 0.5uM R049
CD19sh + 5uM R049

*FIG. 5N*

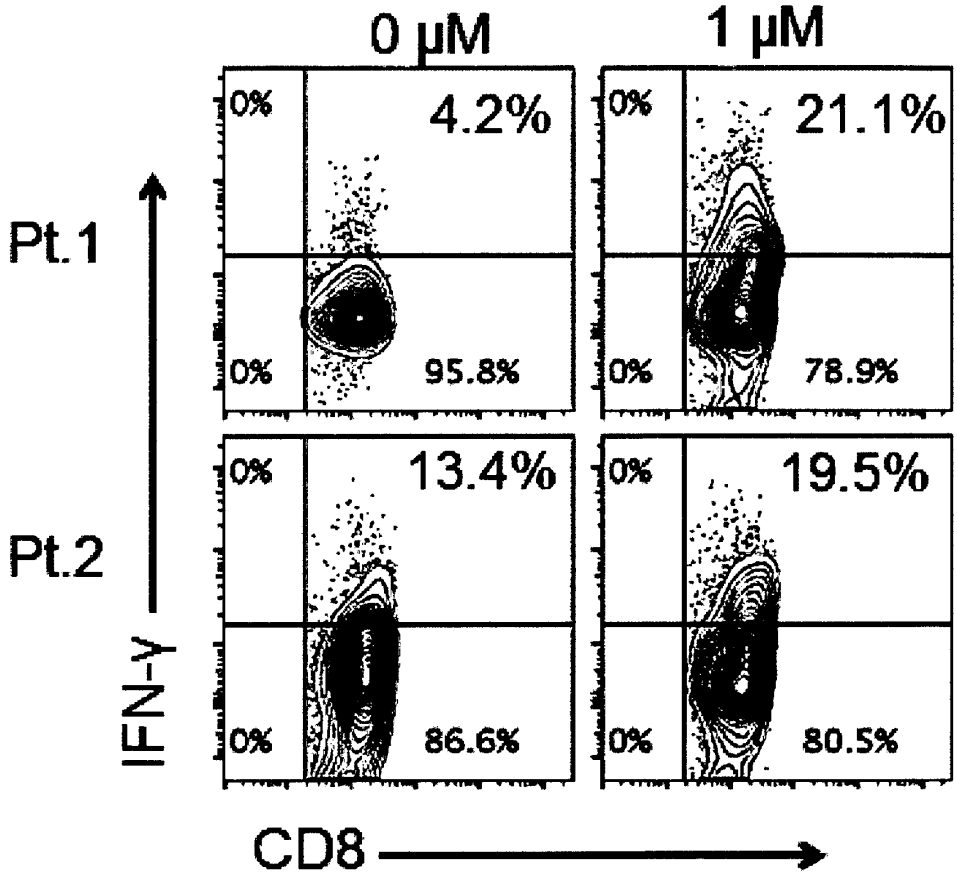



*FIG. 5O*

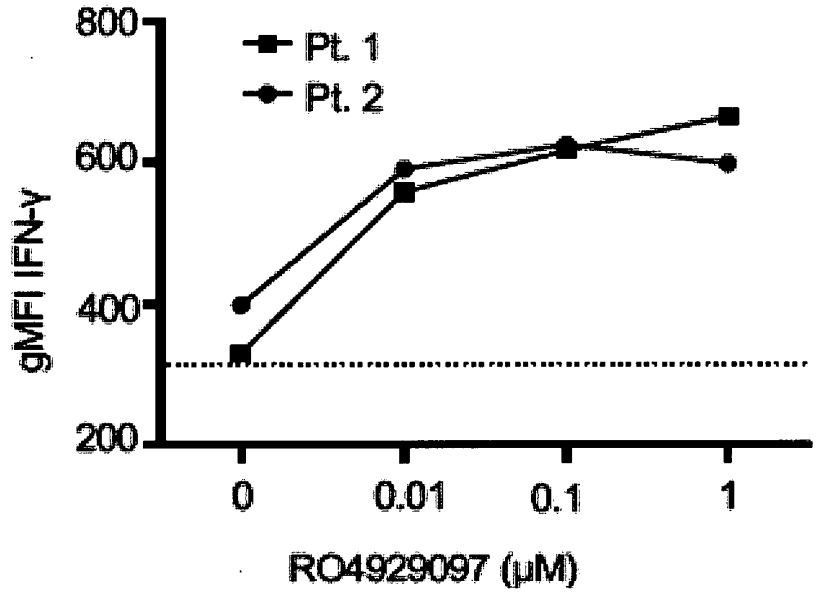

800
600
400
200
gMFI IFN-γ
Pt. 1
Pt. 2
0
0.01
0.1
1
RO4929097 (μM)

*FIG. 5P*

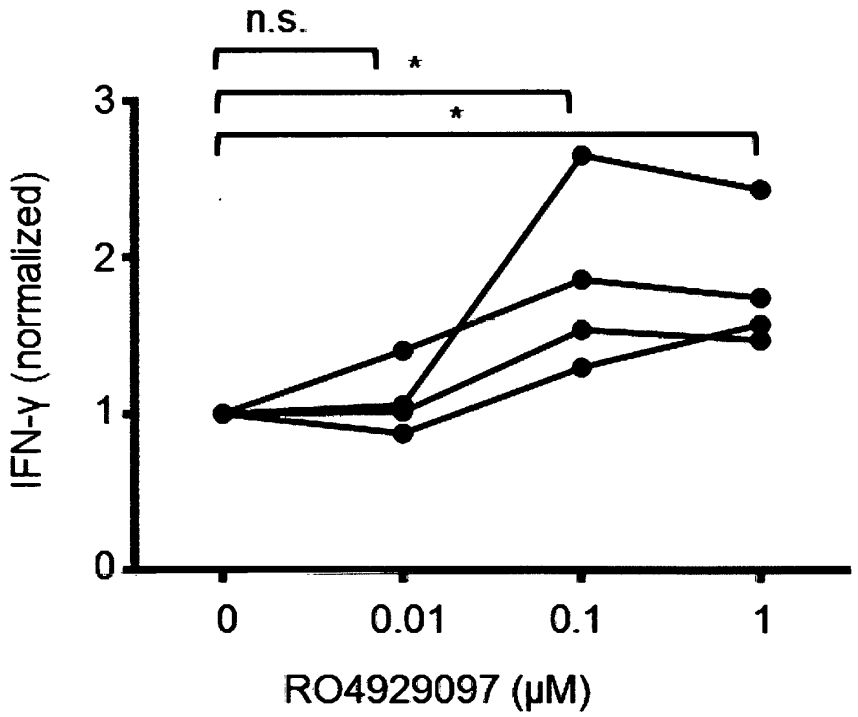

n.s.
*
*
3
2
1
0
IFN-γ (normalized)
0
0.01
0.1
1
RO4929097 (μM)

*FIG. 5Q*

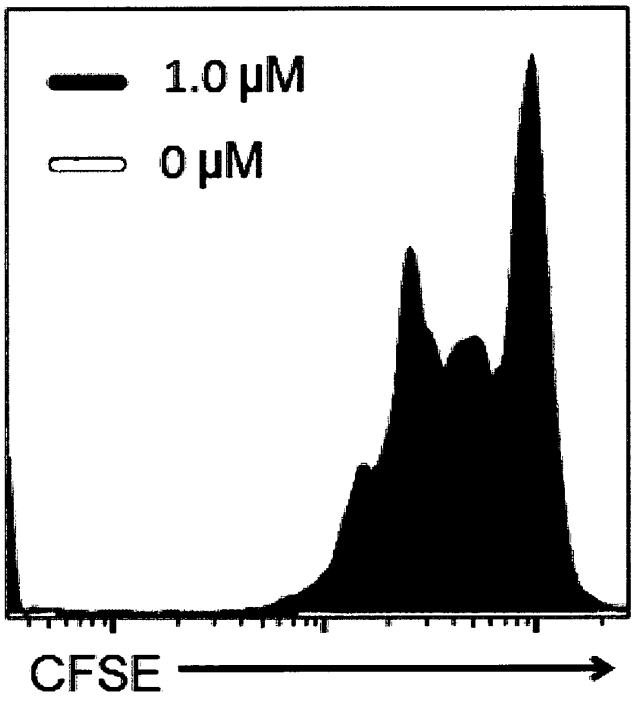



*FIG. 5R*

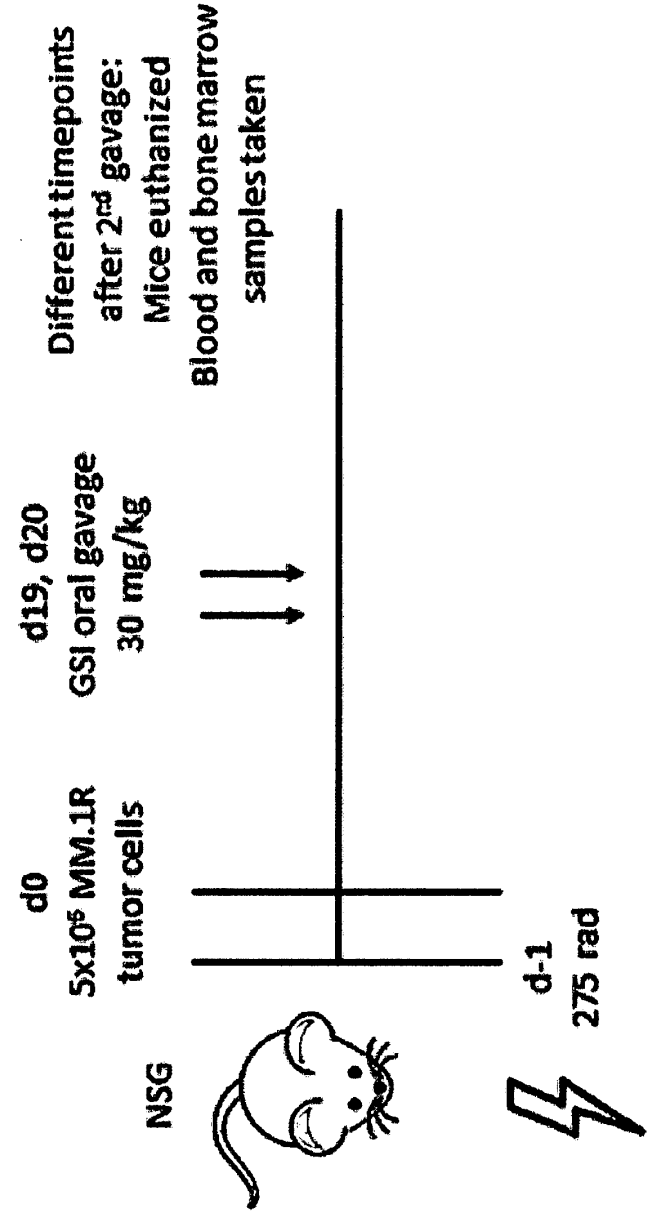

Different timepoints after 2nd gavage: Mice euthanized Blood and bone marrow samples taken
d19, d20 GSI oral gavage 30 mg/kg
d0 5x10⁵ MM.1R tumor cells
NSG
d-1 275 rad

*FIG. 6A*

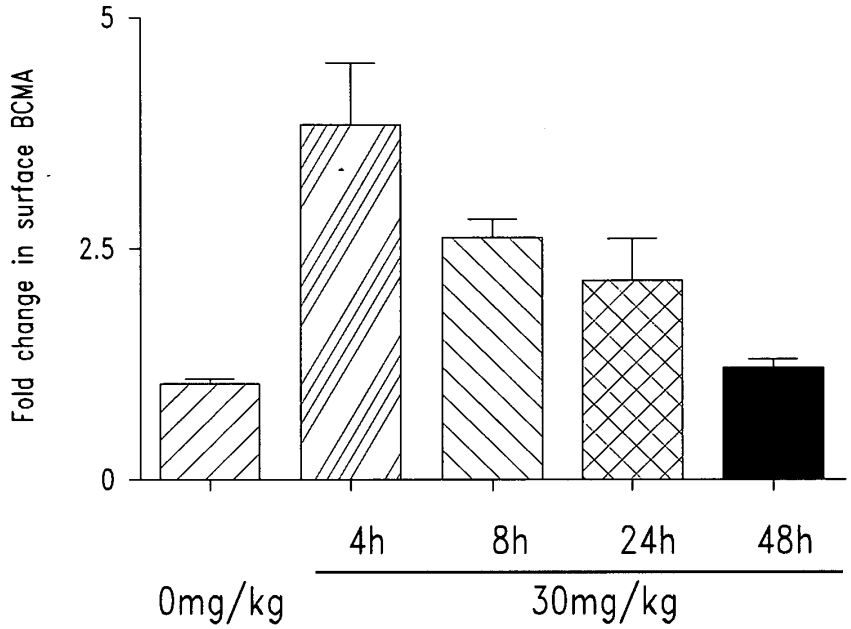

Fold change in surface BCMA
5
2.5
0
4h
8h
24h
48h
0mg/kg
30mg/kg

*FIG. 6B*

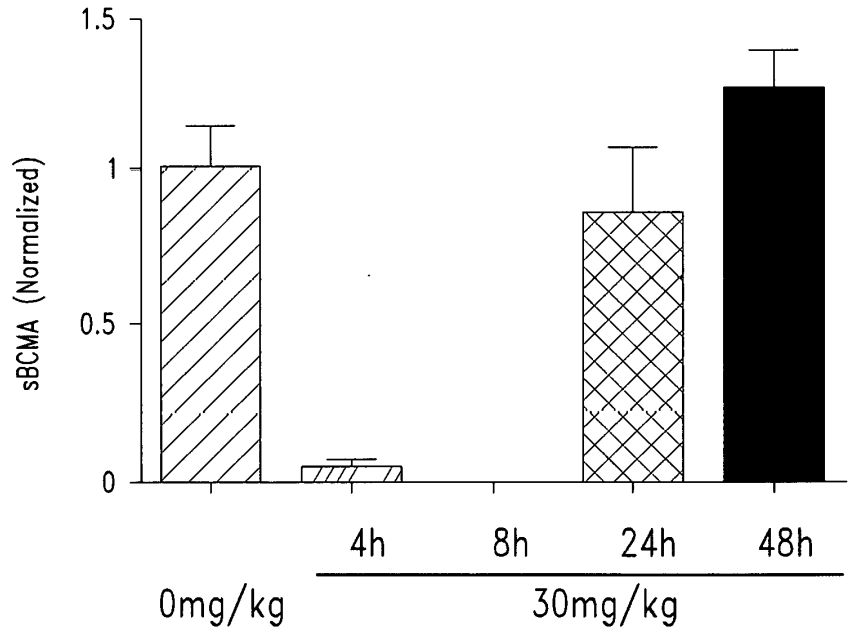

sBCMA (Normalized)
1.5
1
0.5
0
4h
8h
24h
48h
0mg/kg
30mg/kg

*FIG. 6C*

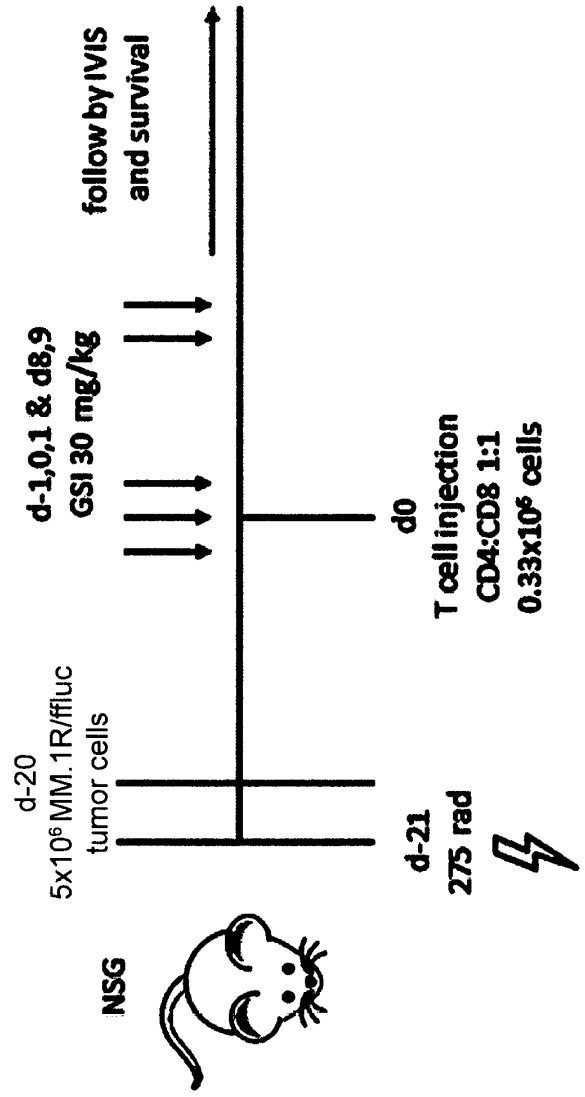

follow by IVIS
and survival
d-1,0,1 & d8,9
GSI 30 mg/kg
d0
T cell injection
CD4:CD8 1:1
0.33x10⁶ cells
d-20
5x10⁶ MM.1R/ffluc
tumor cells
d-21
275 rad
NSG

*FIG. 7A*

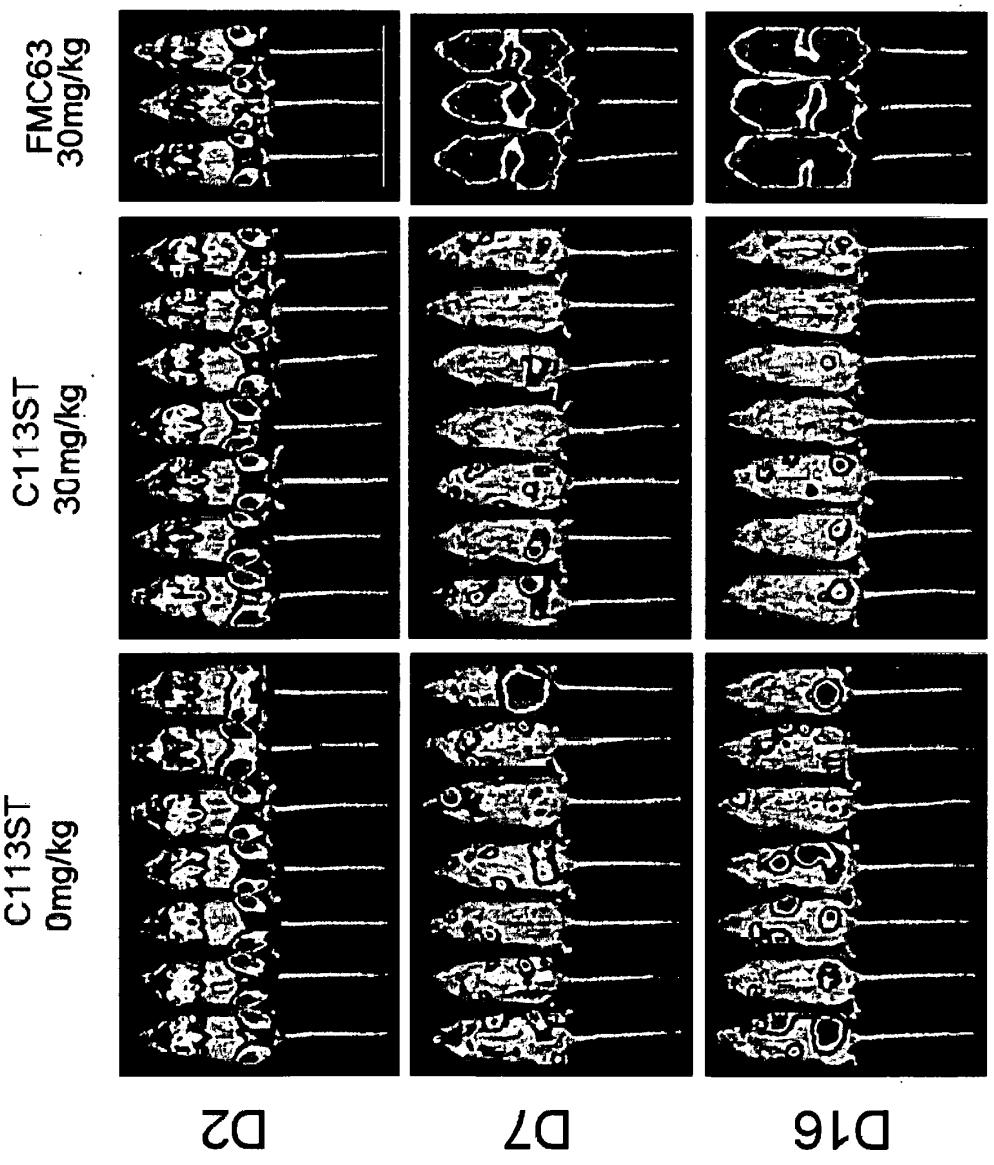



*FIG. 7B*

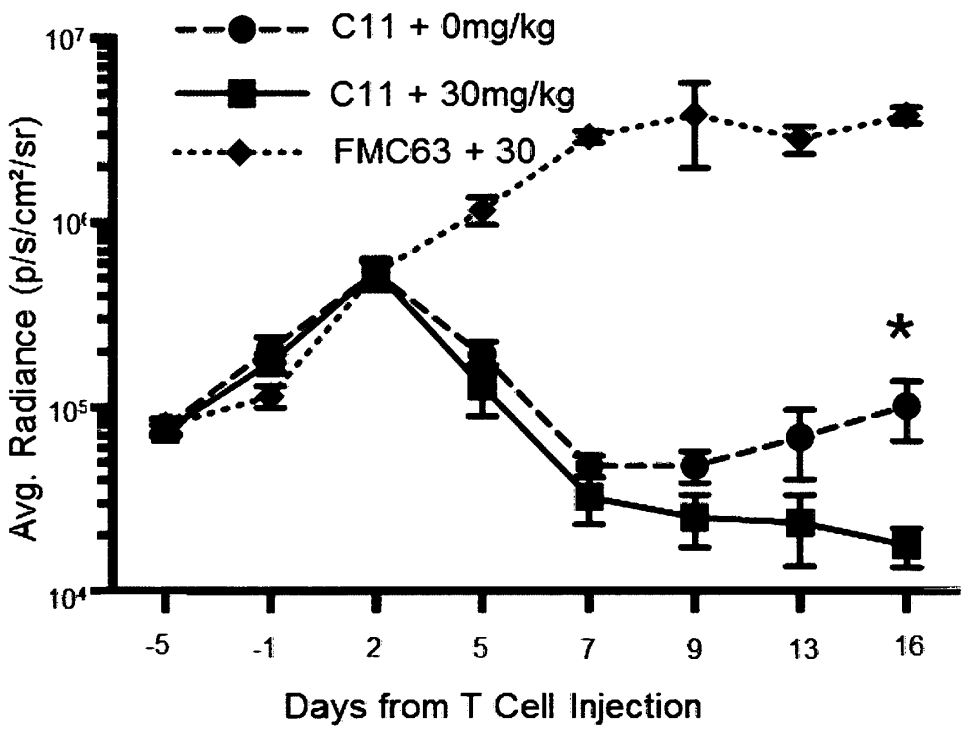

C11 + 0mg/kg
C11 + 30mg/kg
FMC63 + 30
Avg. Radiance (p/s/cm²/sr)
10⁷
10⁶
10⁵
10⁴
*
-5
-1
2
5
7
9
13
16
Days from T Cell Injection

*FIG. 7C*

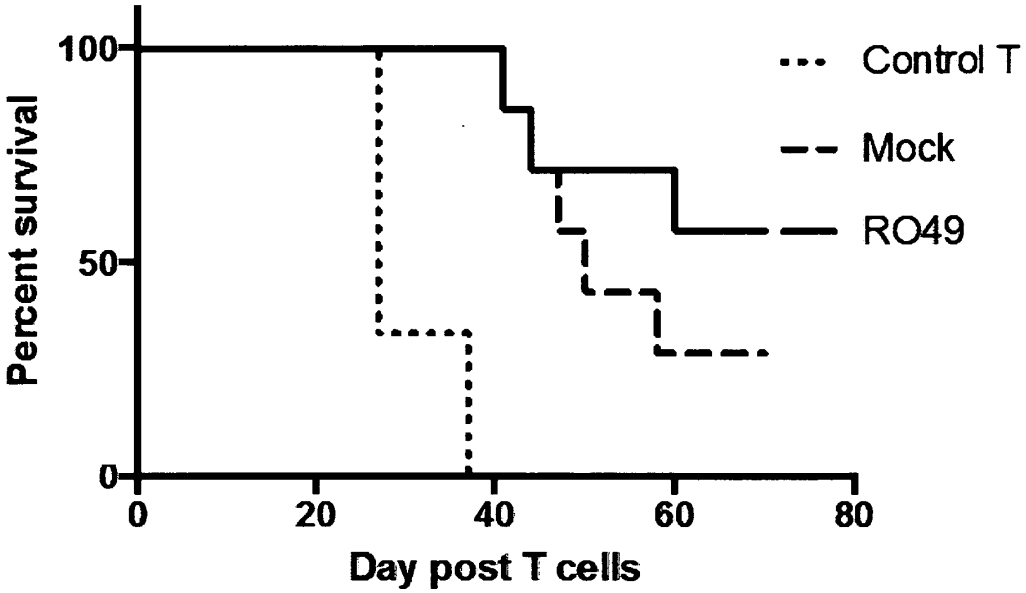

Control T
Mock
RO49
Percent survival
100
50
0
0
20
40
60
80
Day post T cells

*FIG. 7D*

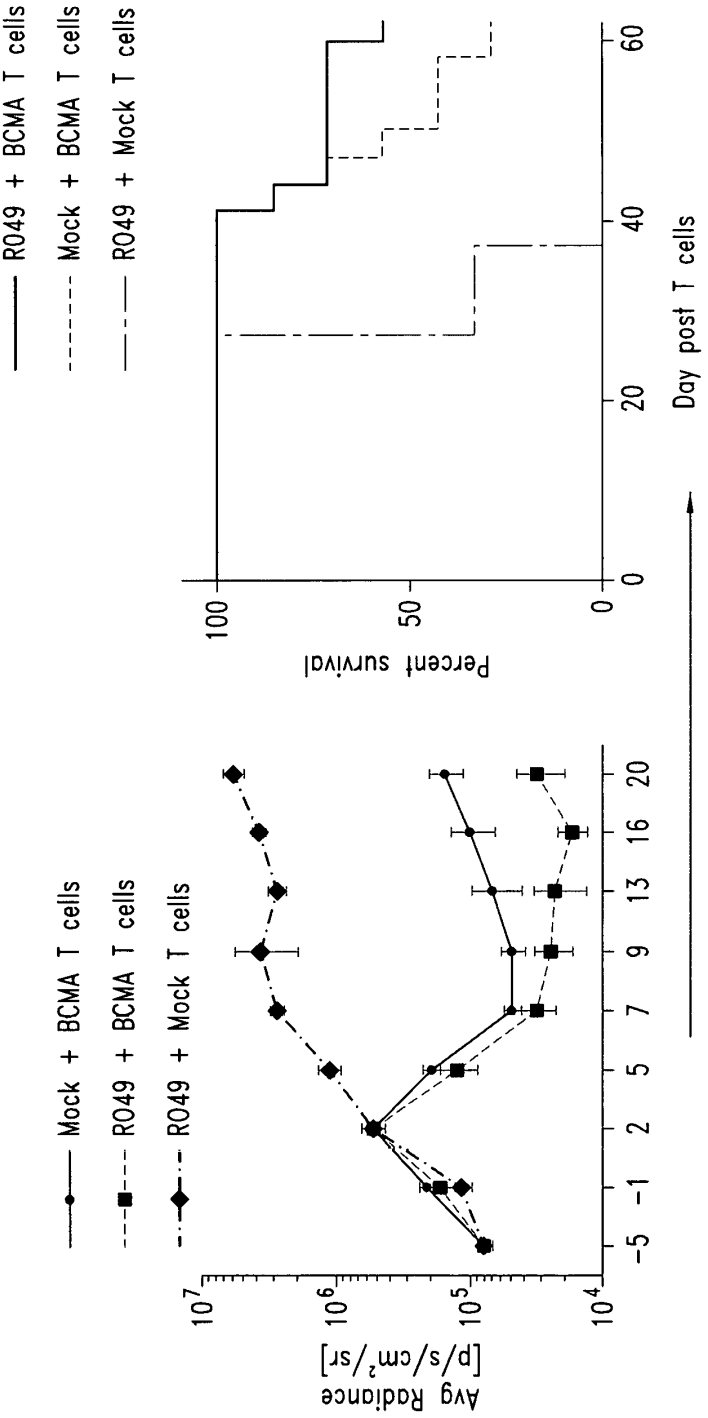

R049 + BCMA T cells
Mock + BCMA T cells
R049 + Mock T cells
Percent survival
100
50
0
0
20
40
60
Day post T cells
Mock + BCMA T cells
R049 + BCMA T cells
R049 + Mock T cells
Avg Radiance [p/s/cm²/sr]
10⁷
10⁶
10⁵
10⁴
-5
-1
2
5
7
9
13
16
20

*FIG. 7E*

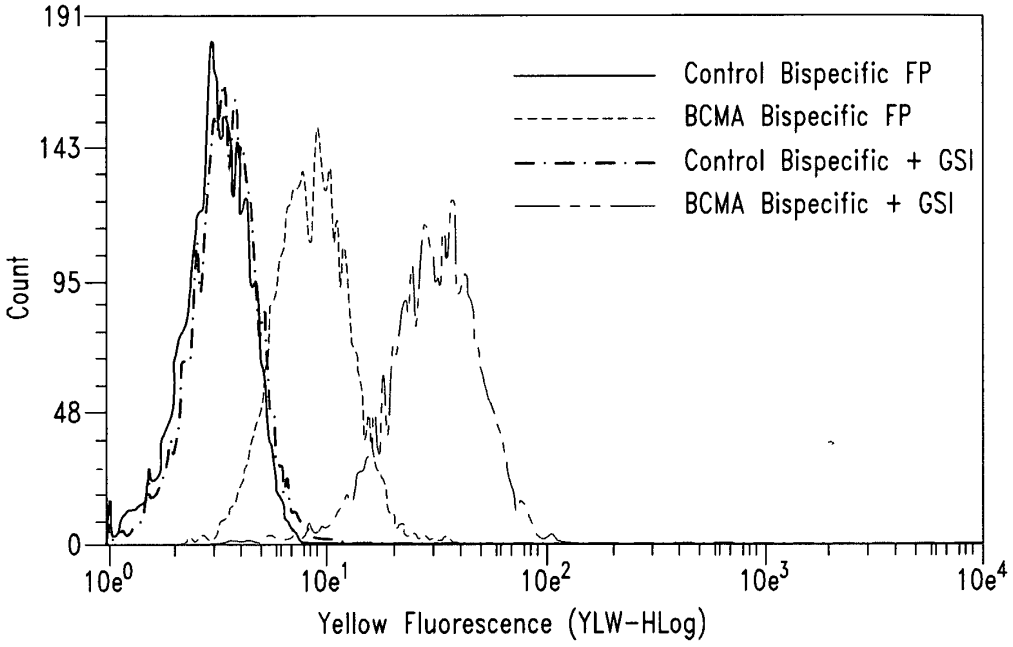

191
143
95
48
0
Count
10e0
10e1
10e2
10e3
10e4
Yellow Fluorescence (YLW-HLog)
Control Bispecific FP
BCMA Bispecific FP
Control Bispecific + GSI
BCMA Bispecific + GSI

*FIG. 8*