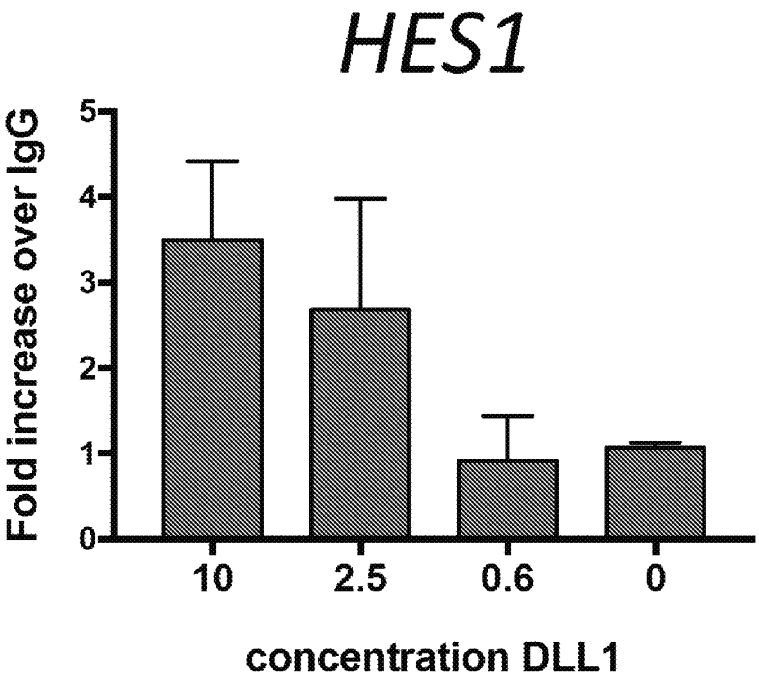

HES1
Fold increase over IgG
5
4
3
2
1
0
10
2.5
0.6
0
concentration DLL1

FIG. 1

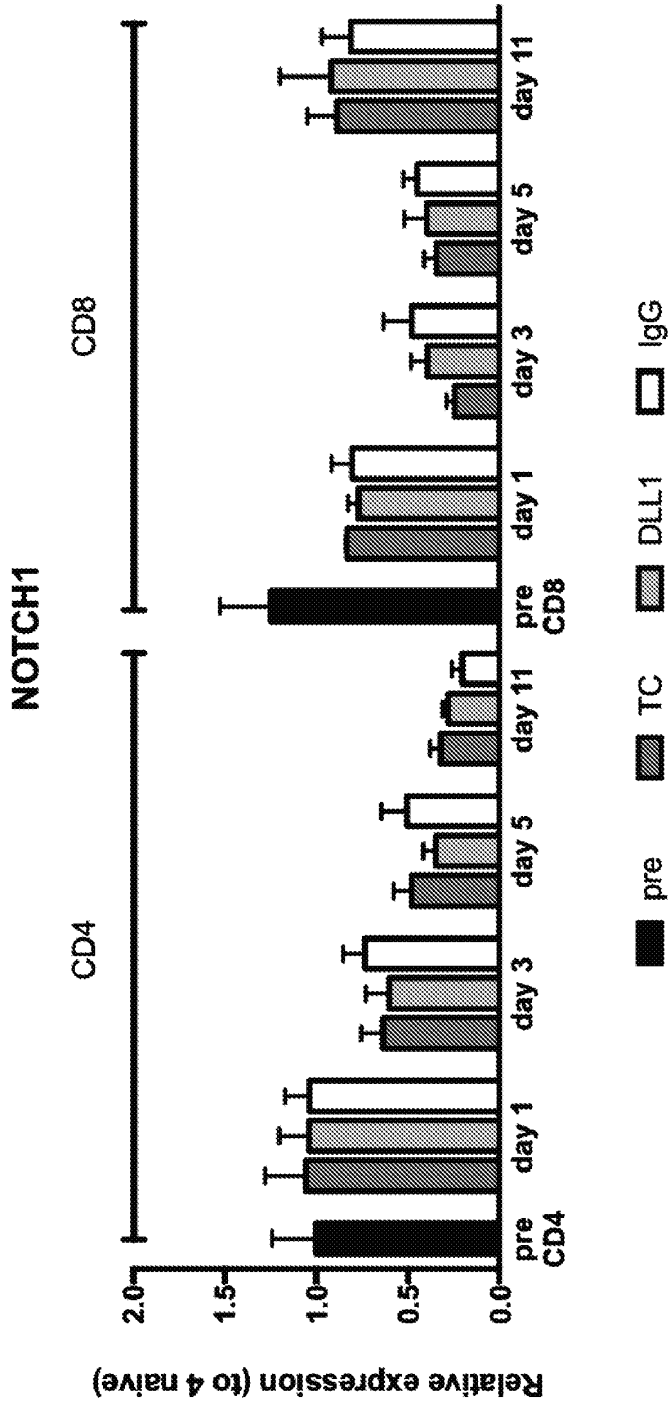

NOTCH1
CD4
CD8
Relative expression (to 4 naive)
2.0
1.5
1.0
0.5
0.0
pre CD4
day 1
day 3
day 5
day 11
pre CD8
day 1
day 3
day 5
day 11
pre
TC
DLL1
IgG

FIG. 2A

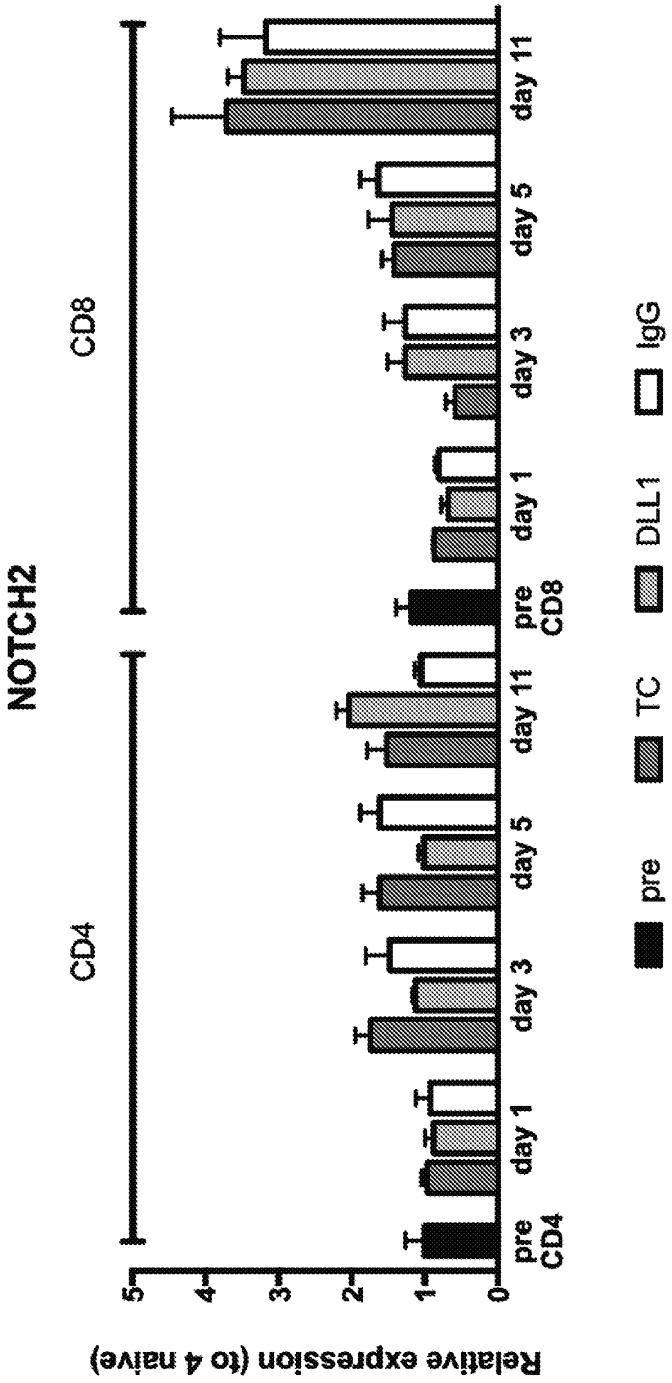

NOTCH2
CD4
CD8
Relative expression (to 4 naive)
0
1
2
3
4
5
pre CD4
day 1
day 3
day 5
day 11
pre CD8
day 1
day 3
day 5
day 11
pre
TC
DLL1
IgG

FIG. 2B

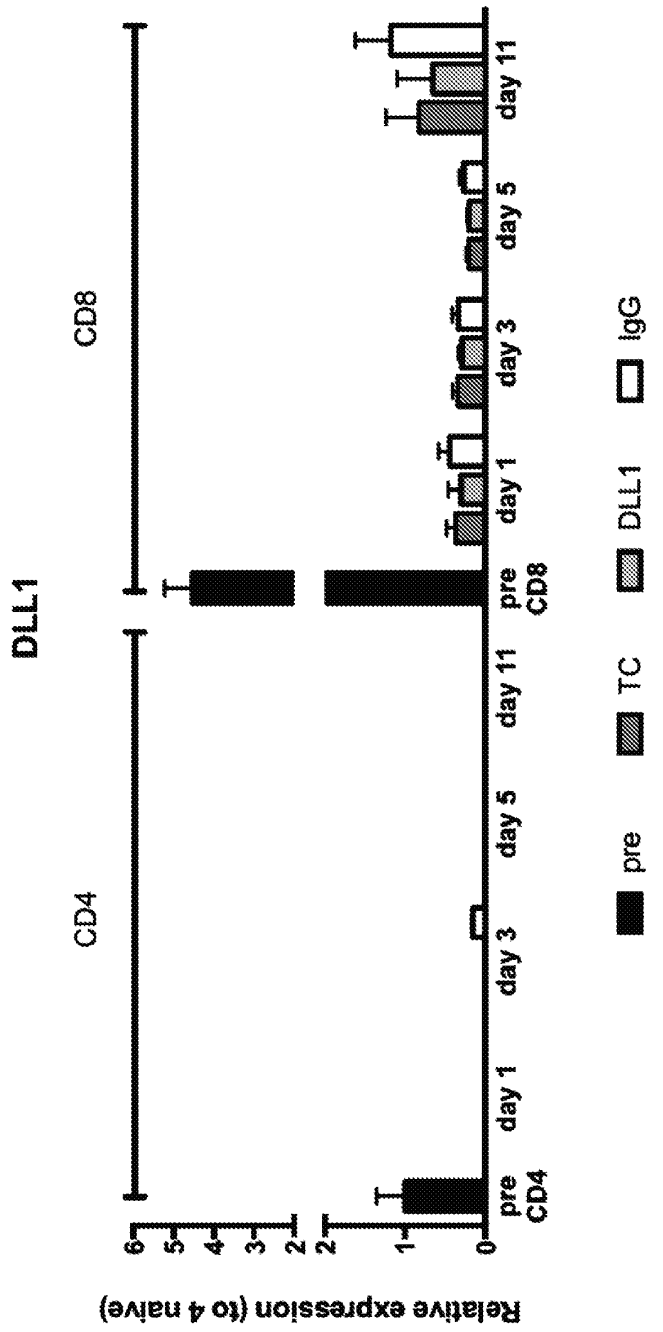

DLL1
CD4
CD8
Relative expression (to 4 naive)
0
1
2
2
3
4
5
6
pre
day 1
day 3
day 5
day 11
pre
day 1
day 3
day 5
day 11
CD4
CD8
pre
TC
DLL1
IgG

FIG. 3A

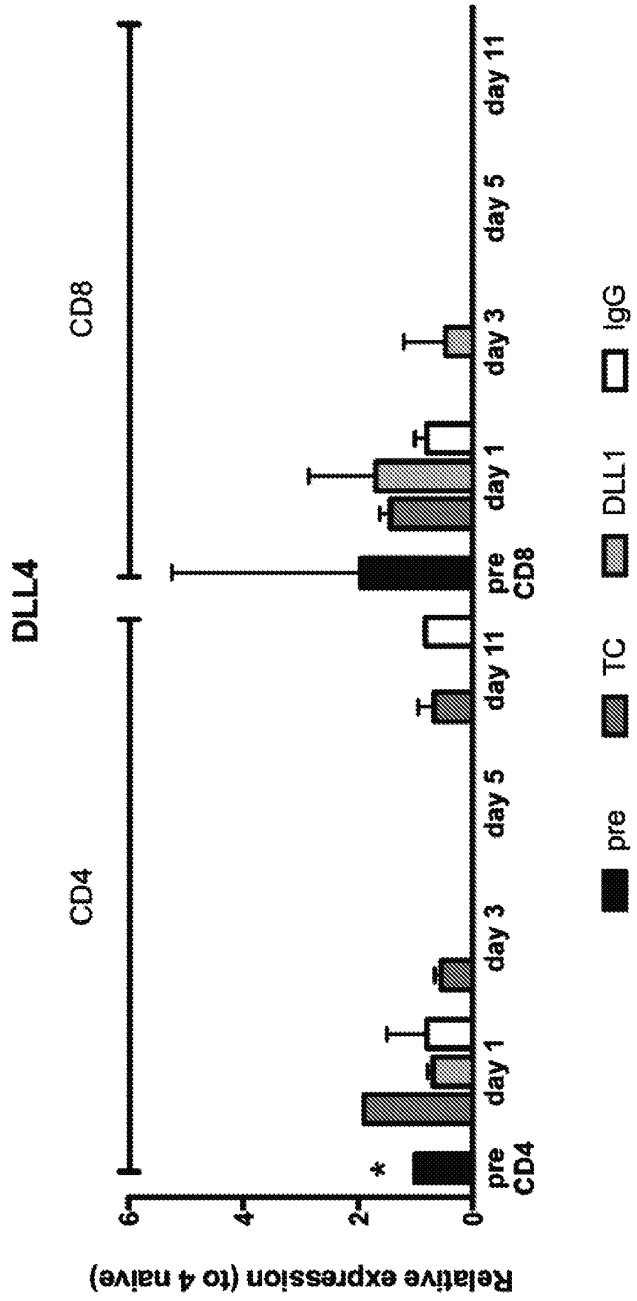

DLL4
CD4
CD8
Relative expression (to 4 naive)
6
4
2
0
*
pre CD4
day 1
day 3
day 5
day 11
pre CD8
day 1
day 3
day 5
day 11
pre
TC
DLL1
IgG

FIG. 3B

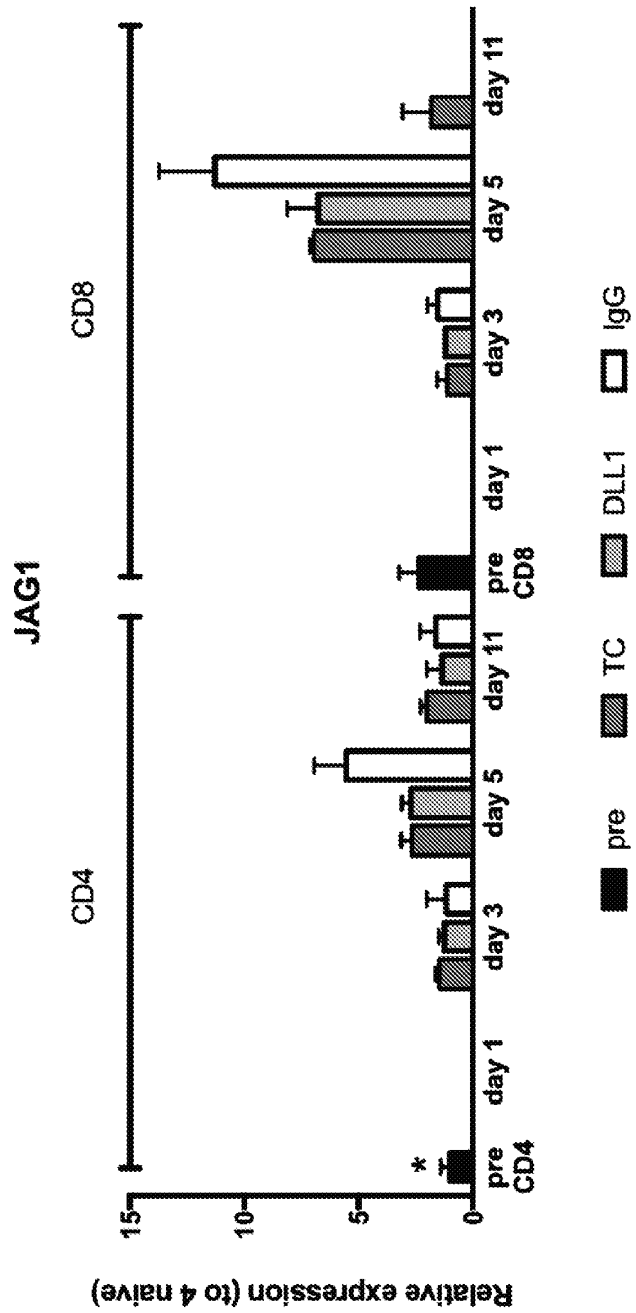

JAG1
CD4
CD8
Relative expression (to 4 naive)
15
10
5
0
*
pre CD4
day 1
day 3
day 5
day 11
pre CD8
day 1
day 3
day 5
day 11
pre
TC
DLL1
IgG

FIG. 3C

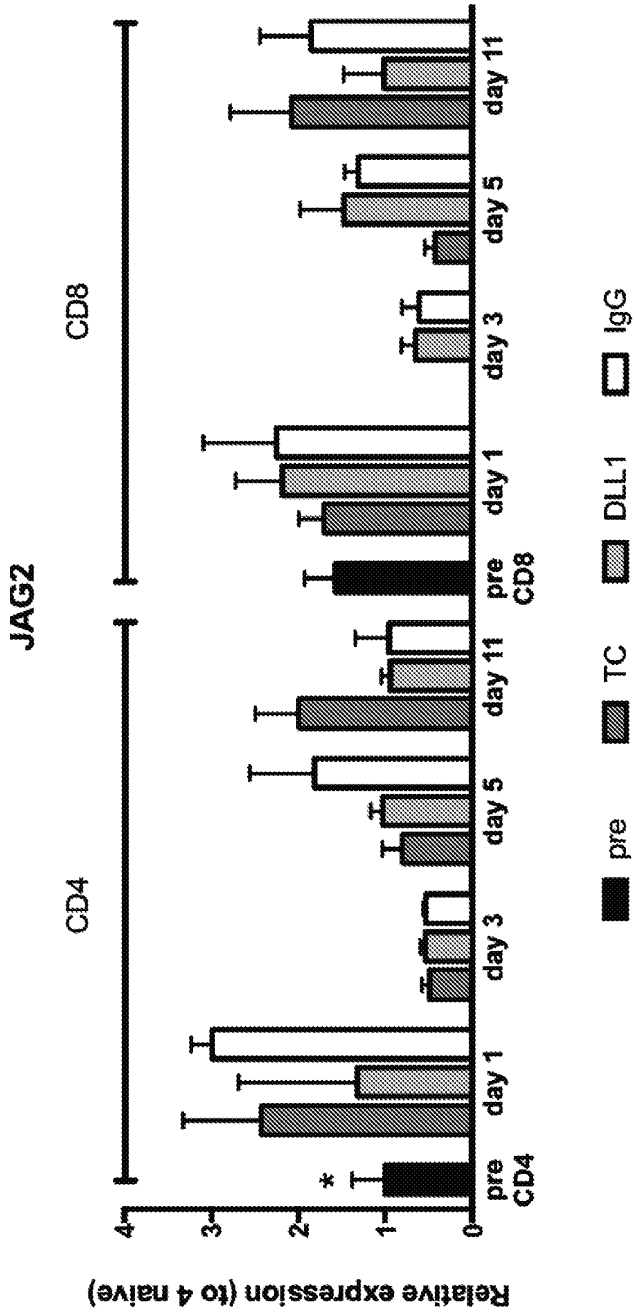

JAG2
CD4
CD8
Relative expression (to 4 naive)
0
1
2
3
4
*
pre
day 1
day 3
day 5
day 11
pre
day 1
day 3
day 5
day 11
CD4
CD8
pre
TC
DLL1
IgG

FIG. 3D

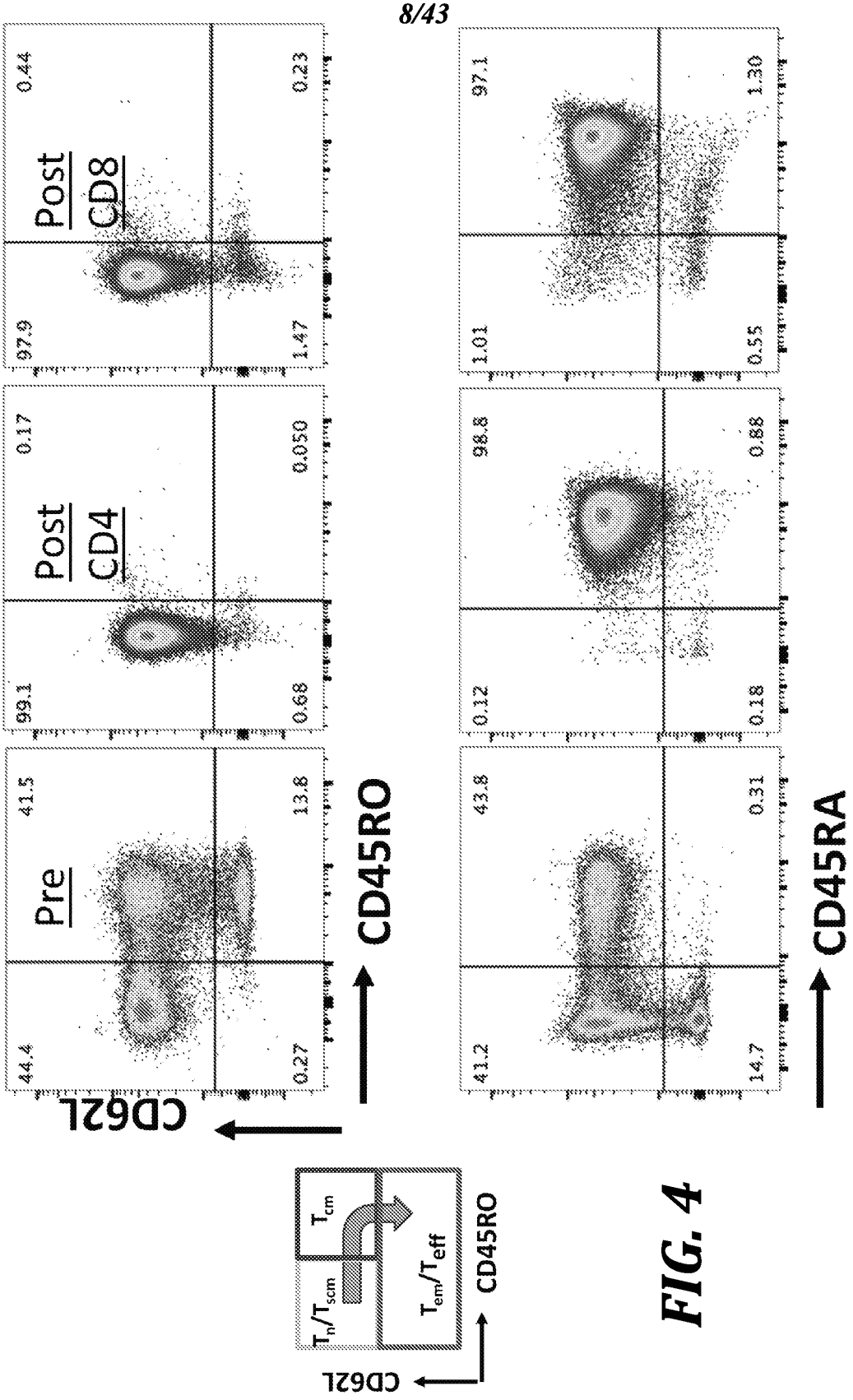

Pre
CD4
CD8
Post
CD62L
CD45RO
CD45RA
44.4
41.5
0.27
13.8
99.1
0.17
0.68
0.050
97.9
0.44
1.47
0.23
41.2
43.8
14.7
0.31
0.12
98.8
0.18
0.88
1.01
97.1
0.55
1.30
T_n/T_{scm}
T_{cm}
T_{em}/T_{eff}

FIG. 4

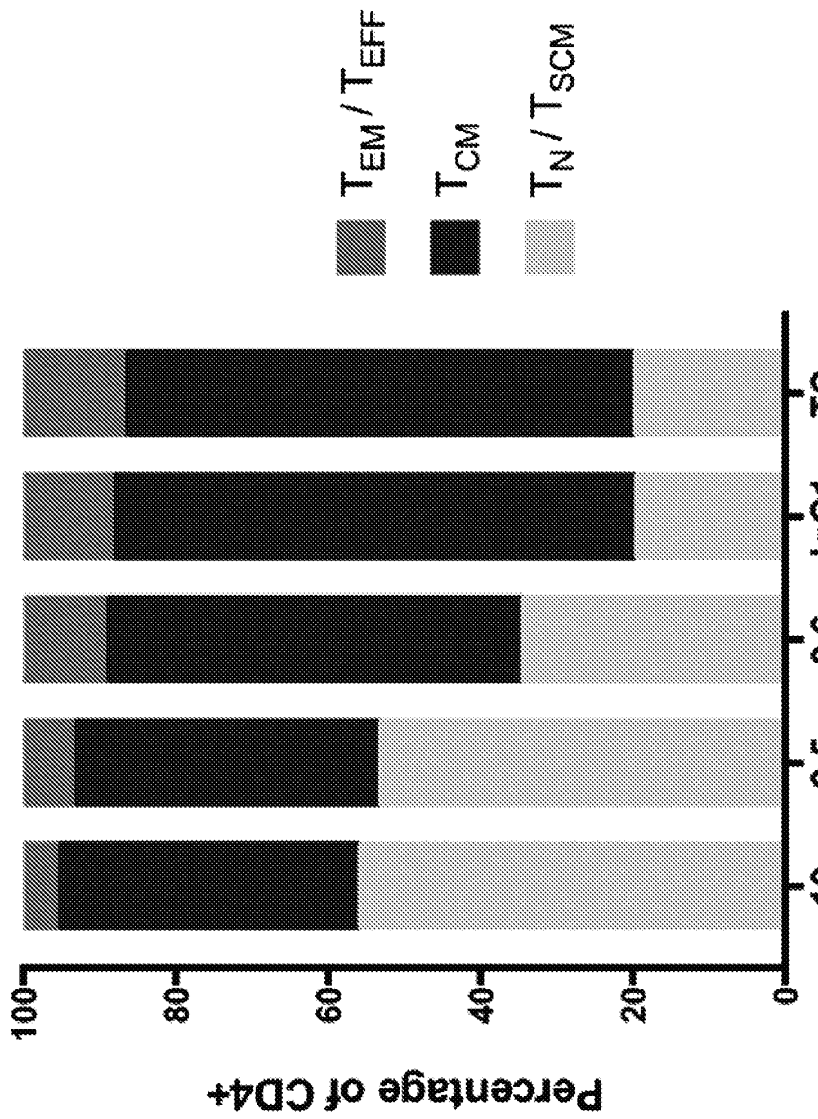

T_{EM} / T_{EFF}
T_{CM}
T_N / T_{SCM}
Percentage of CD4+
100
80
60
40
20
0
10
2.5
0.6
IgG1
TC
DLL1

FIG. 5A

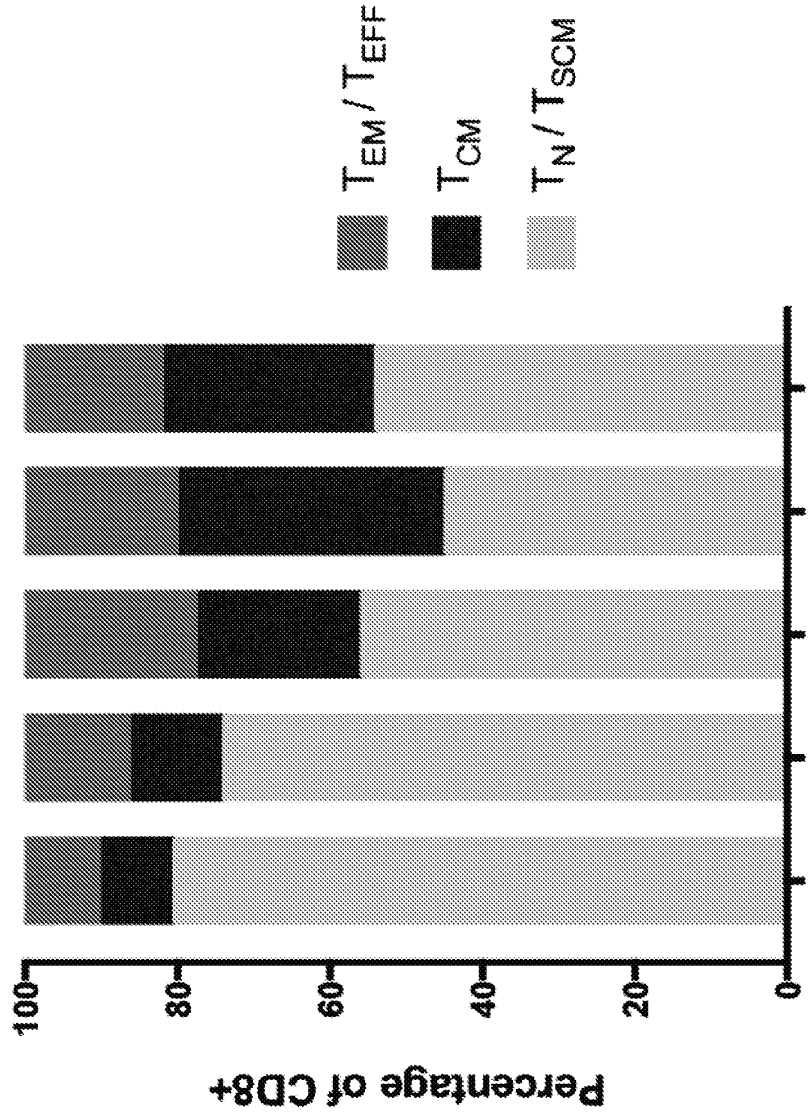

T_{EM} / T_{EFF}
T_{CM}
T_N / T_{SCM}
Percentage of CD8+
100
80
60
40
20
0
10
2.5
0.6
IgG1
TC
DLL1

FIG. 5B

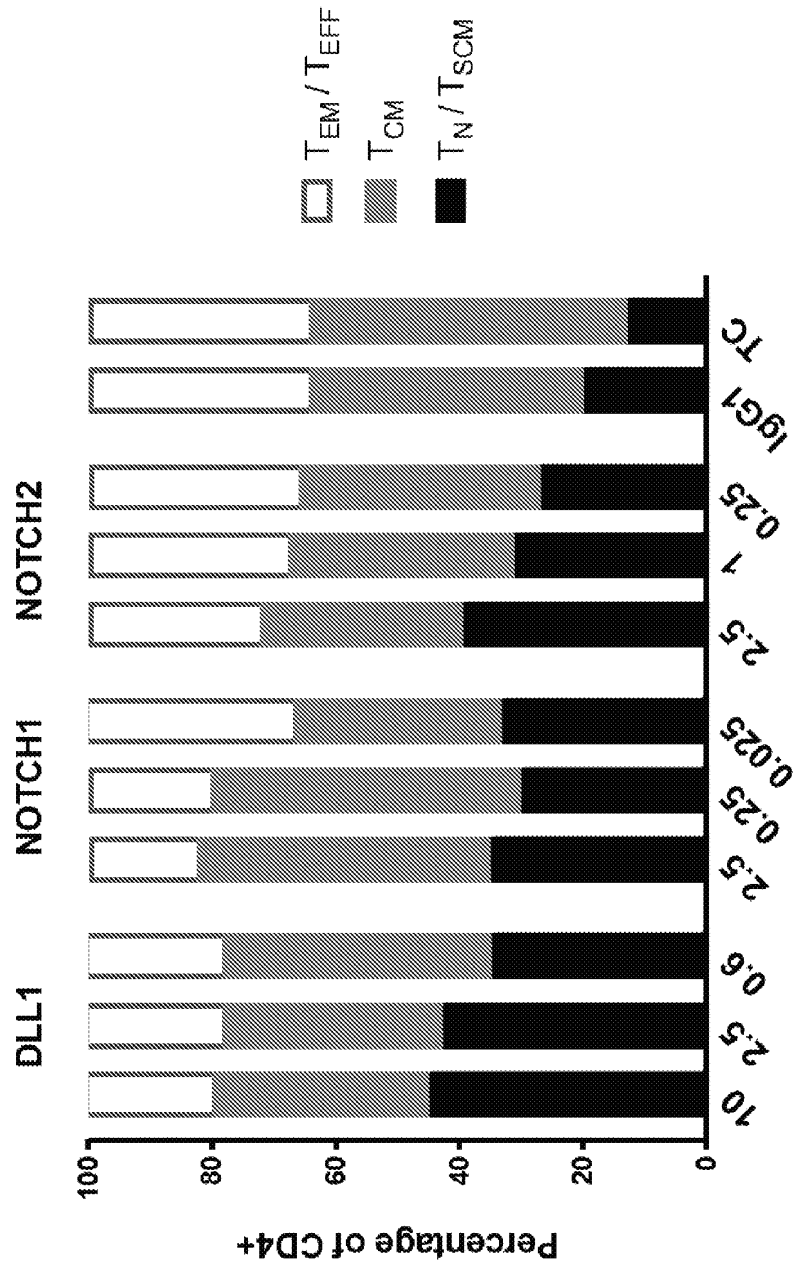

T_{EM} / T_{EFF}
T_{CM}
T_N / T_{SCM}
Percentage of CD4+
100
80
60
40
20
0
DLL1
NOTCH1
NOTCH2
10
2.5
0.6
2.5
0.25
0.025
0.25
2.5
1
0.25
IgG1
TC

FIG. 5C

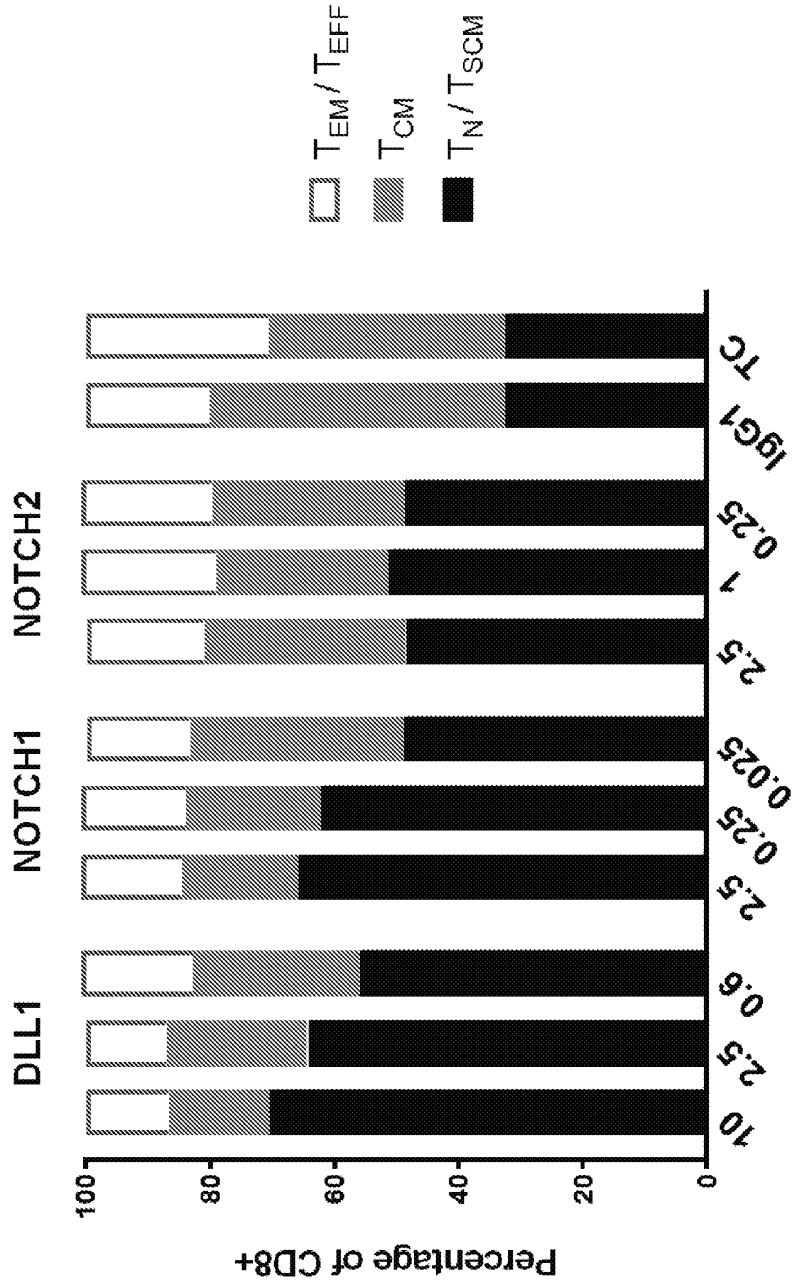

T_{EM} / T_{EFF}
T_{CM}
T_N / T_{SCM}
Percentage of CD8+
100
80
60
40
20
0
DLL1
NOTCH1
NOTCH2
10
2.5
0.6
2.5
0.25
0.025
2.5
1
0.25
IgG1
TC

FIG. 5D

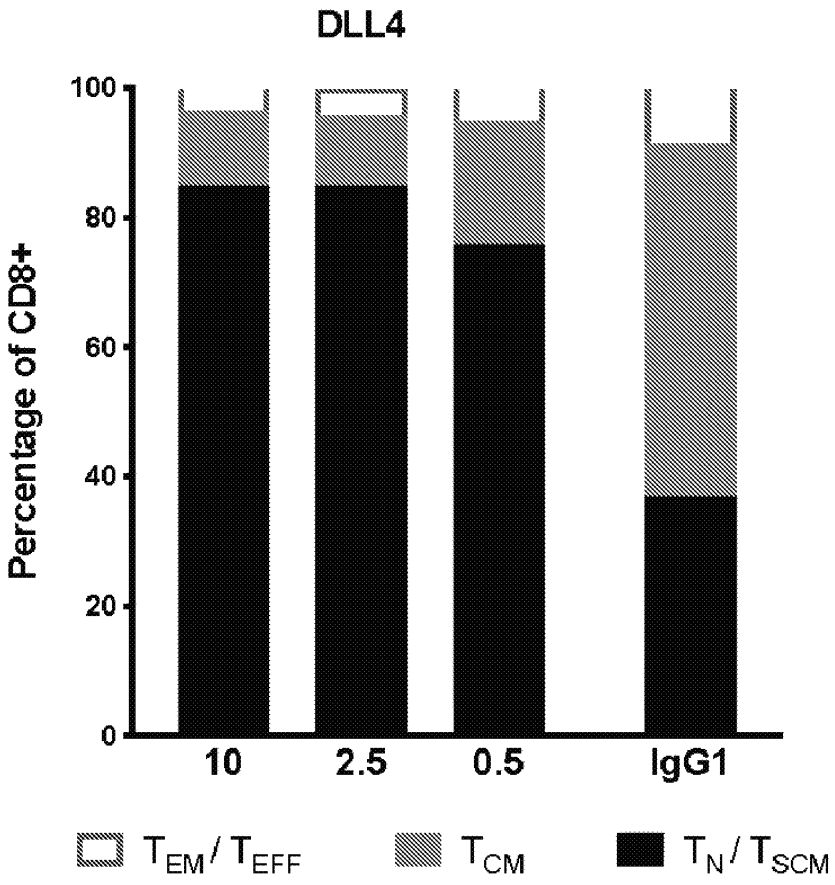

DLL4
Percentage of CD8+
100
80
60
40
20
0
10
2.5
0.5
IgG1
TEM / TEFF
TCM
TN / TSCM

FIG. 5E

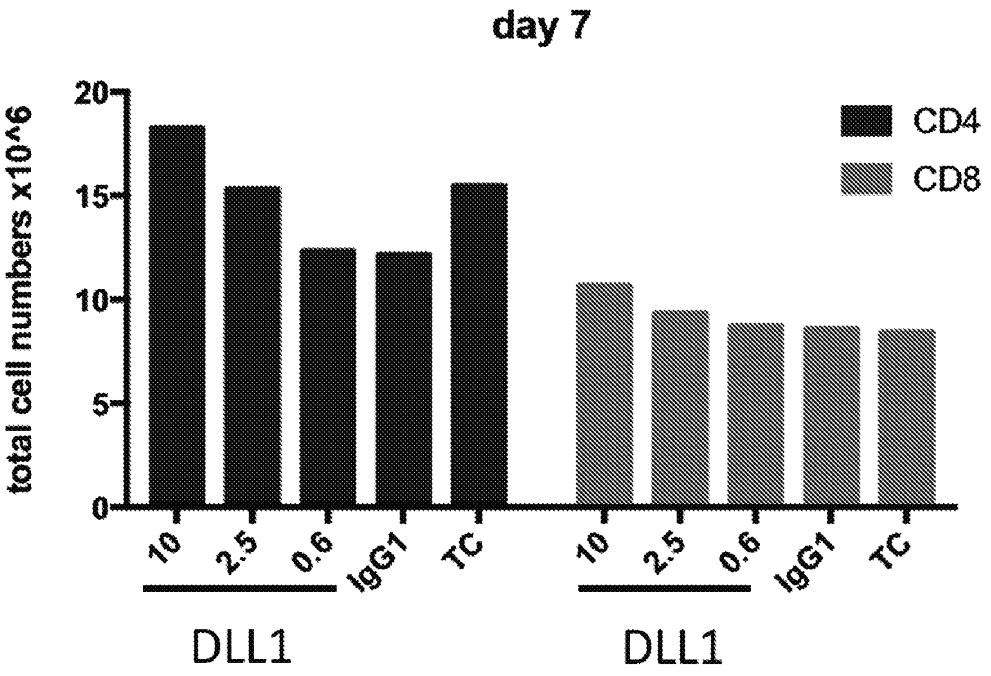

day 7
total cell numbers x10^6
20
15
10
5
0
CD4
CD8
10
2.5
0.6
IgG1
TC
10
2.5
0.6
IgG1
TC
DLL1
DLL1

FIG. 6A

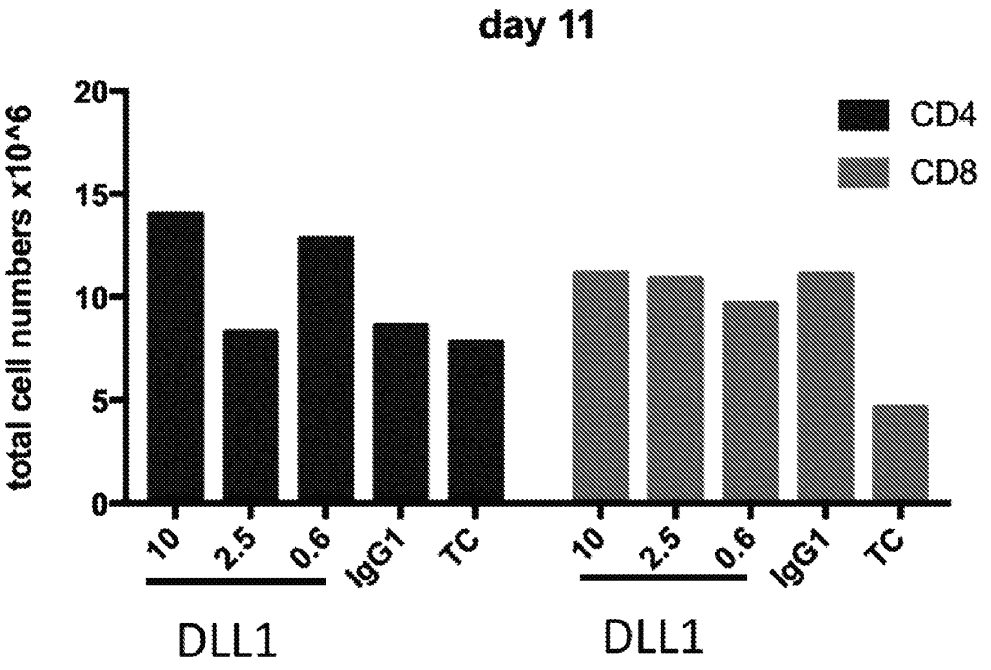

day 11
total cell numbers x10^6
20
15
10
5
0
CD4
CD8
10
2.5
0.6
IgG1
TC
10
2.5
0.6
IgG1
TC
DLL1
DLL1

FIG. 6B

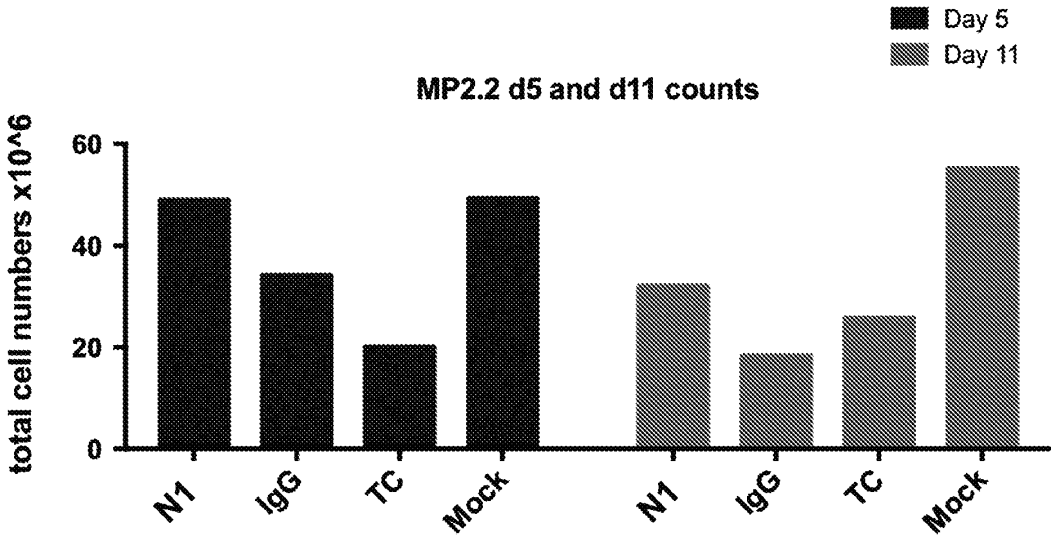

Day 5
Day 11
MP2.2 d5 and d11 counts
total cell numbers x10^6
60
40
20
0
N1
IgG
TC
Mock
N1
IgG
TC
Mock

FIG. 7

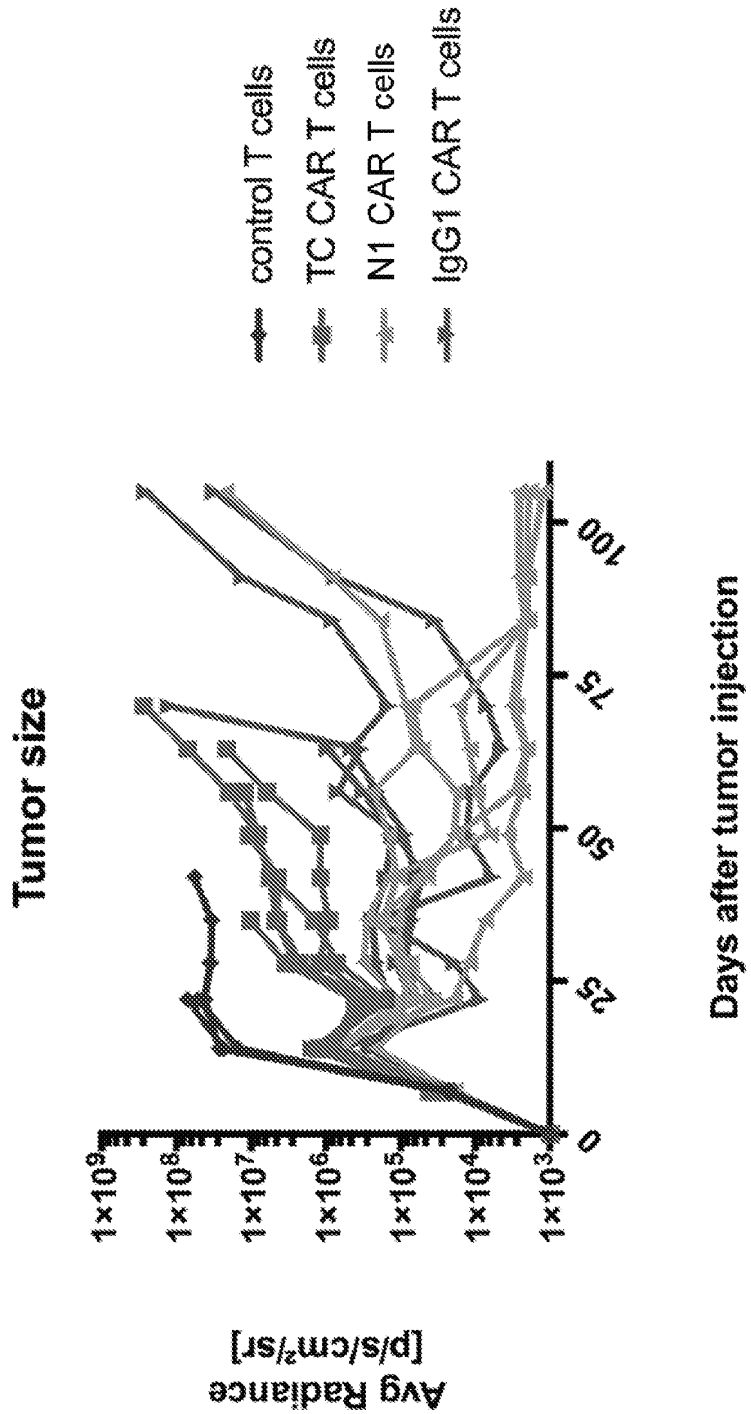

Tumor size
control T cells
TC CAR T cells
N1 CAR T cells
IgG1 CAR T cells
Avg Radiance [p/s/cm²/sr]
1×10⁹
1×10⁸
1×10⁷
1×10⁶
1×10⁵
1×10⁴
1×10³
0
25
50
75
100
Days after tumor injection

FIG. 8

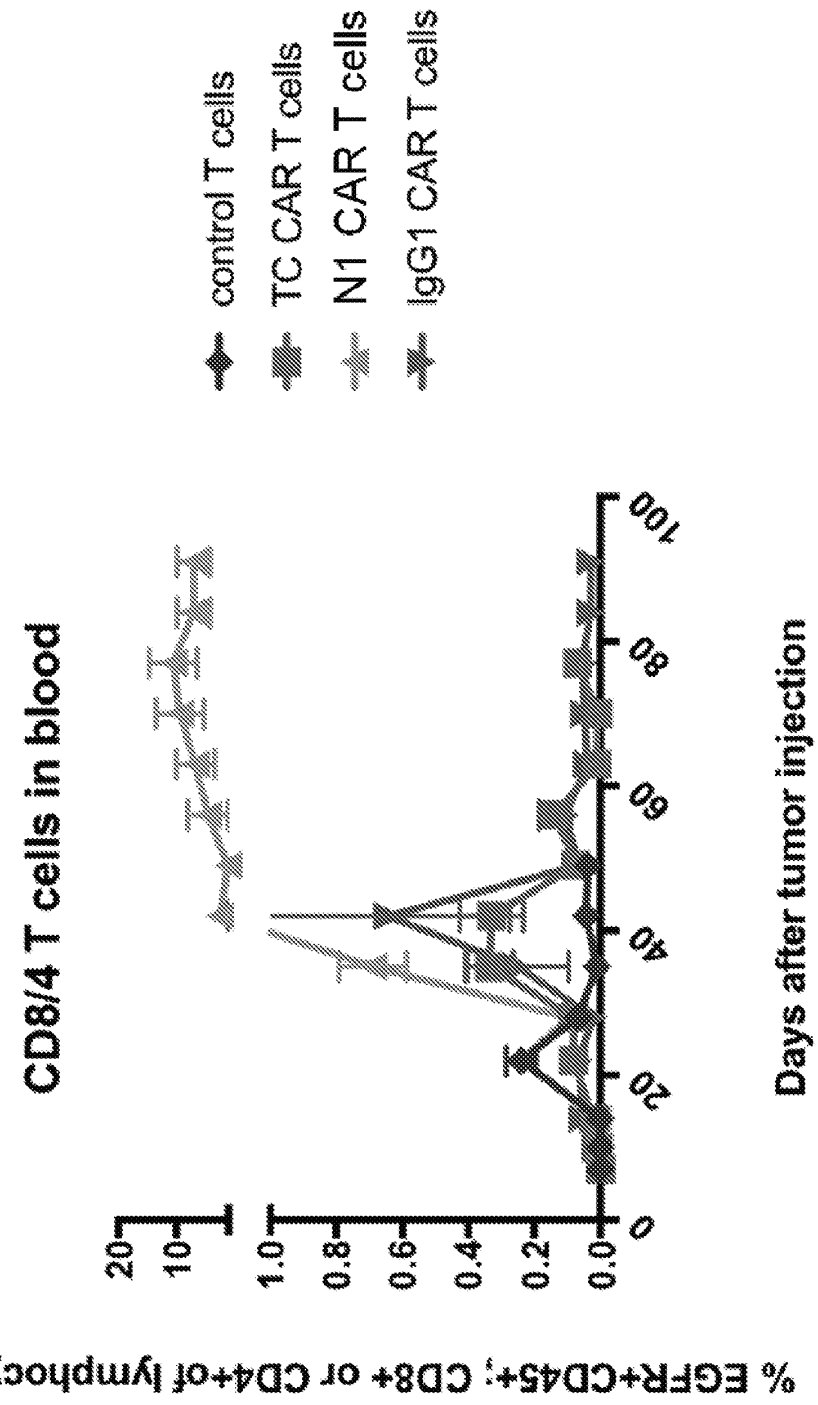

CD8/4 T cells in blood
control T cells
TC CAR T cells
N1 CAR T cells
IgG1 CAR T cells
% EGFR+CD45+; CD8+ or CD4+of lymphocytes
20
10
1.0
0.8
0.6
0.4
0.2
0.0
0
20
40
60
80
100
Days after tumor injection

FIG. 9A

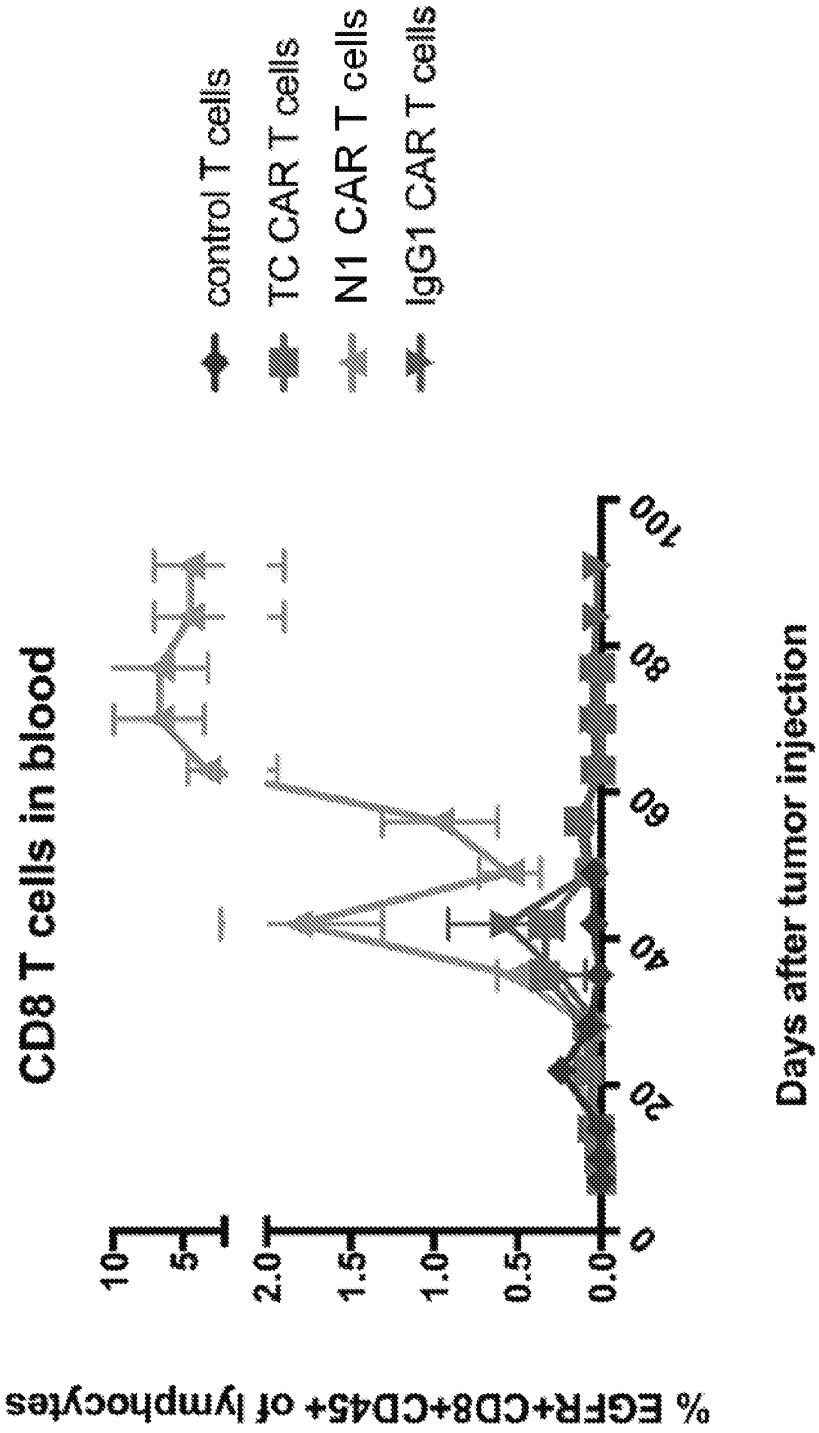

CD8 T cells in blood
% EGFR+CD8+CD45+ of lymphocytes
10
5
2.0
1.5
1.0
0.5
0.0
0
20
40
60
80
100
Days after tumor injection
control T cells
TC CAR T cells
N1 CAR T cells
IgG1 CAR T cells

FIG. 9B

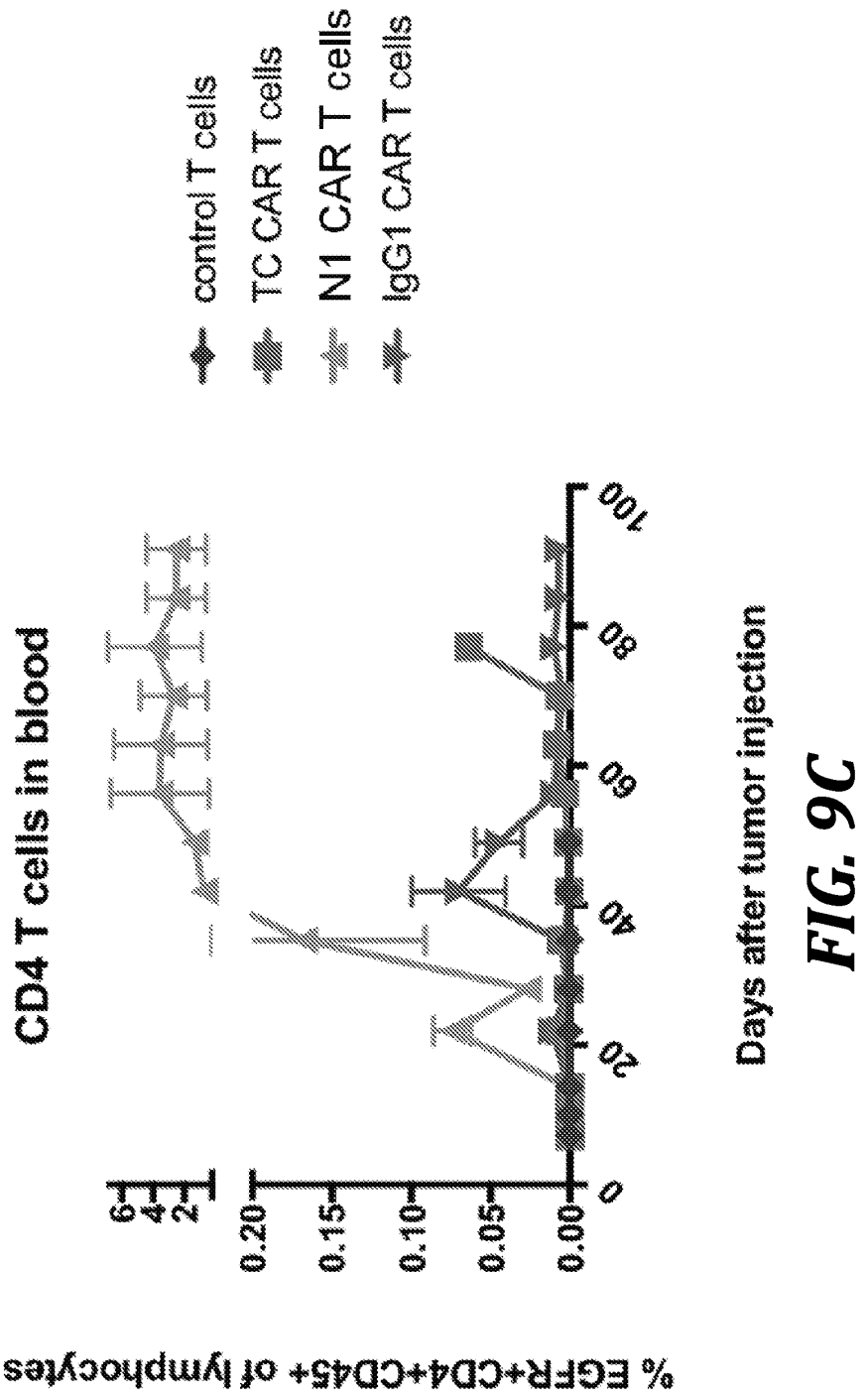

CD4 T cells in blood
control T cells
TC CAR T cells
N1 CAR T cells
IgG1 CAR T cells
% EGFR+CD4+CD45+ of lymphocytes
6
4
2
0.20
0.15
0.10
0.05
0.00
0
20
40
60
80
100
Days after tumor injection

FIG. 9C

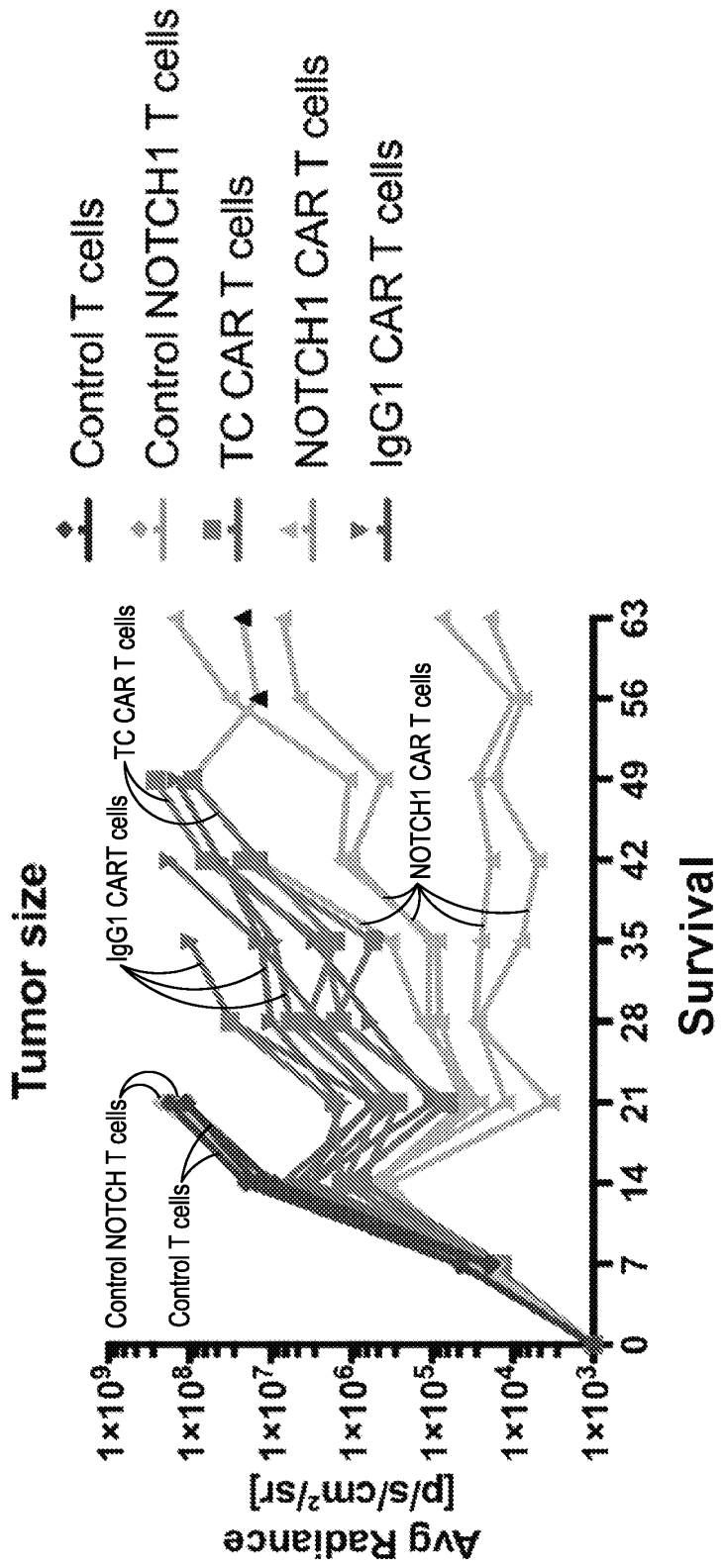

Control T cells
Control NOTCH1 T cells
TC CAR T cells
NOTCH1 CAR T cells
IgG1 CAR T cells
Tumor size
Control NOTCH T cells
Control T cells
IgG1 CART cells
TC CAR T cells
NOTCH1 CAR T cells
Avg Radiance [p/s/cm²/sr]
1×10⁹
1×10⁸
1×10⁷
1×10⁶
1×10⁵
1×10⁴
1×10³
0
7
14
21
28
35
42
49
56
63
Survival

FIG. 10A

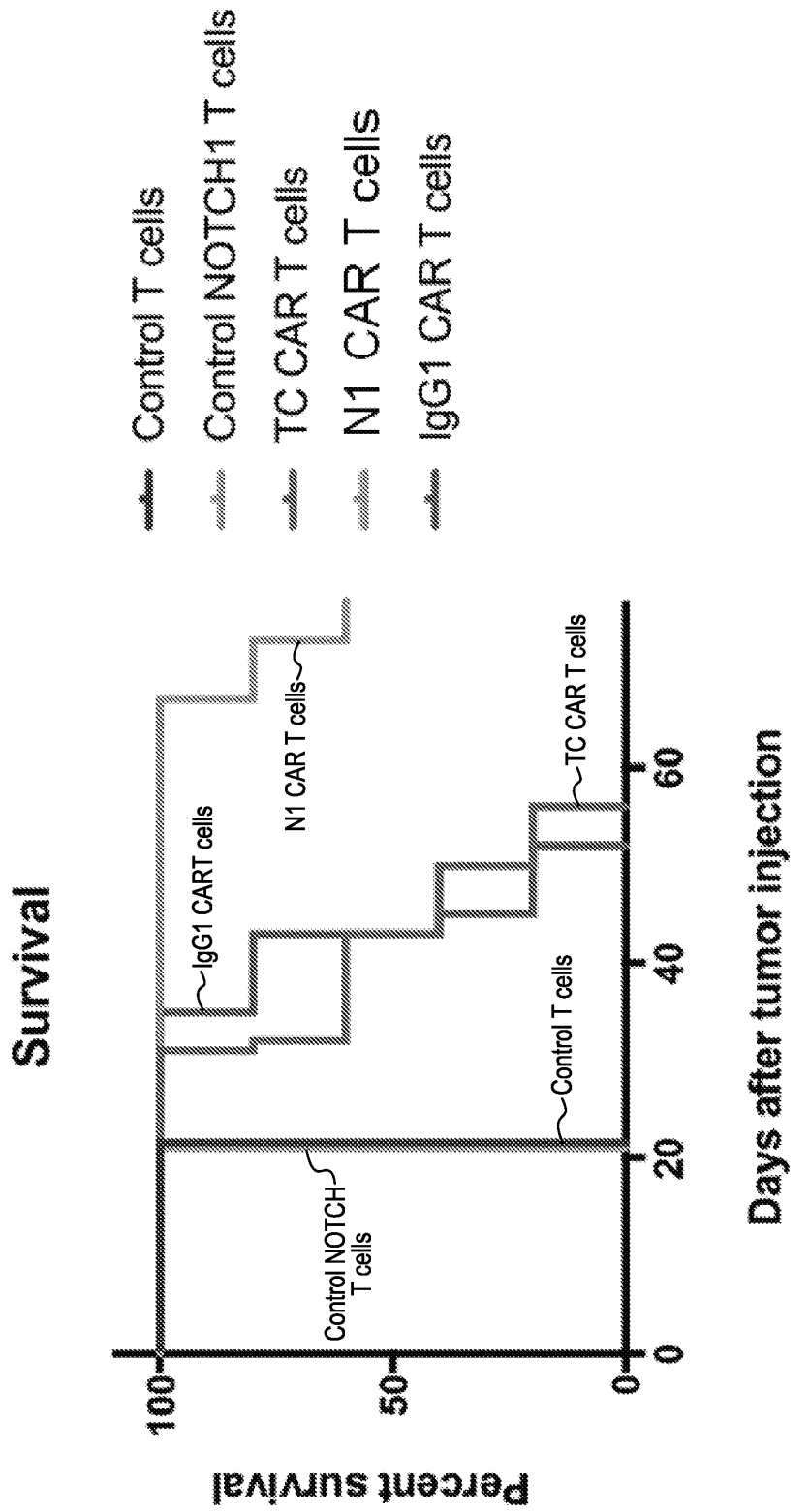

Survival
Control T cells
Control NOTCH1 T cells
TC CAR T cells
N1 CAR T cells
IgG1 CAR T cells
Percent survival
100
50
0
0
20
40
60
Days after tumor injection
IgG1 CART cells
N1 CAR T cells
Control NOTCH T cells
Control T cells
TC CAR T cells

FIG. 10B

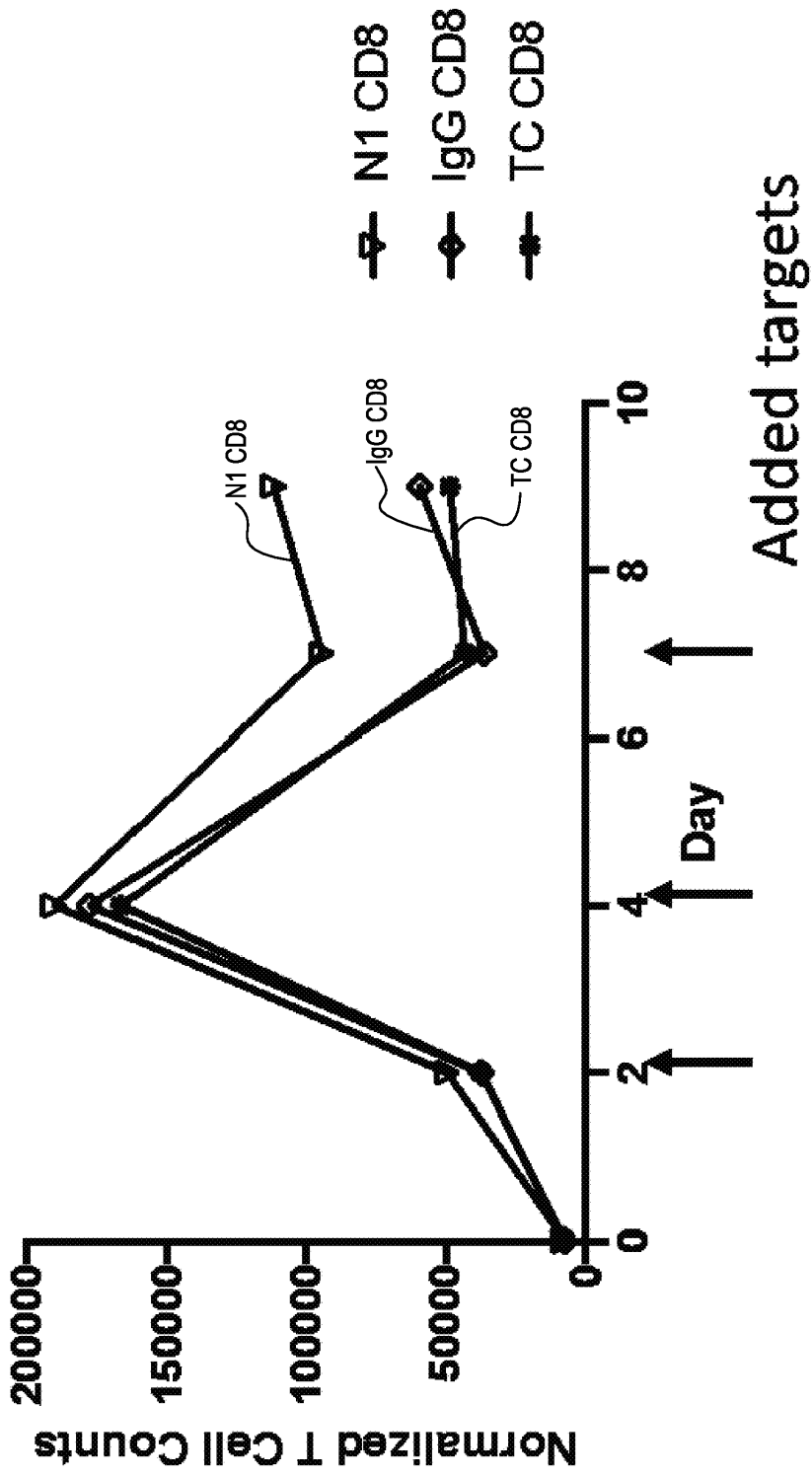

vs K562-CD19 T Cells CD8
Normalized T Cell Counts
200000
150000
100000
50000
0
0
2
4
6
8
10
Day
Added targets
N1 CD8
IgG CD8
TC CD8
N1 CD8
IgG CD8
TC CD8

FIG. 11A

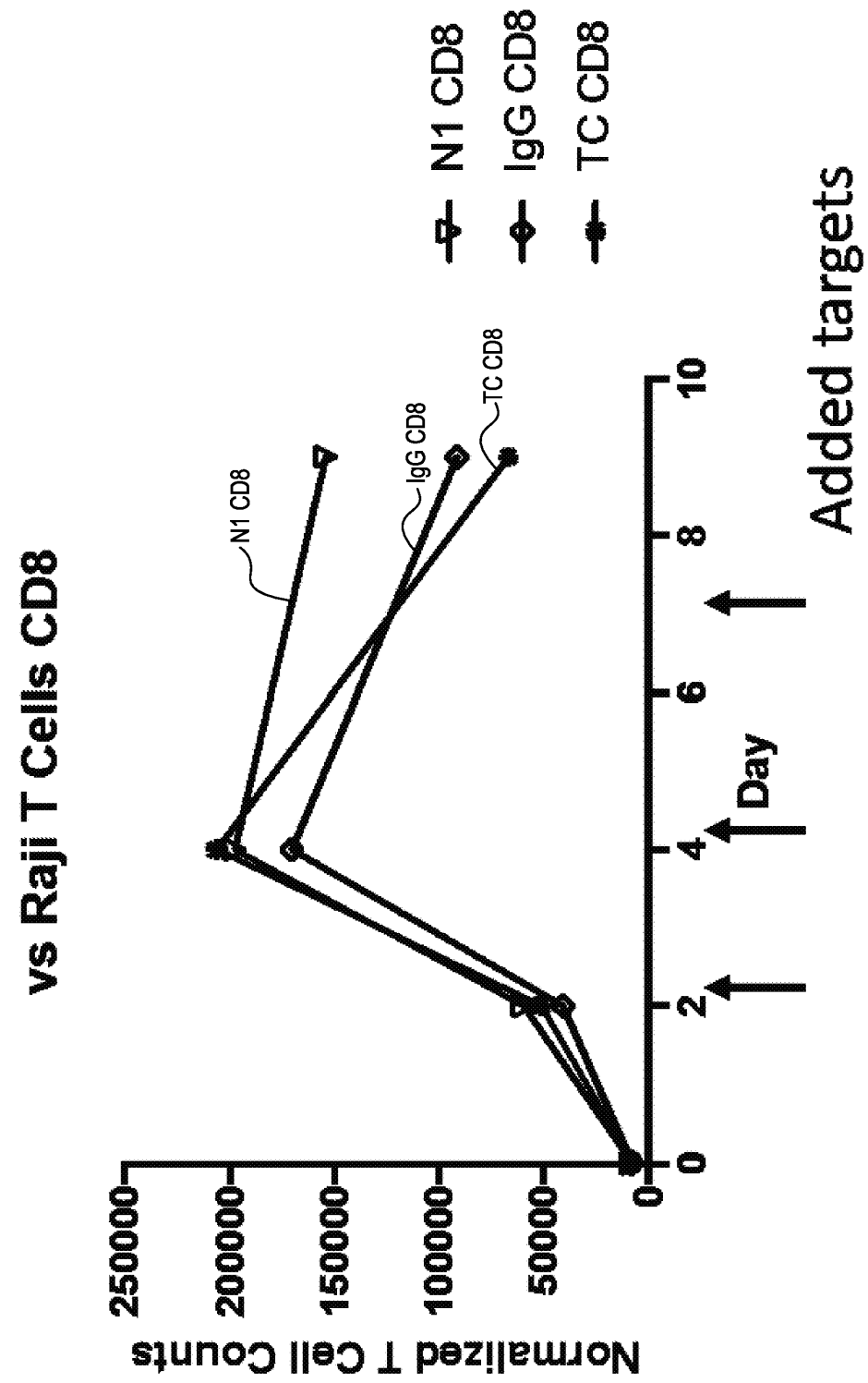

vs Raji T Cells CD8
Normalized T Cell Counts
250000
200000
150000
100000
50000
0
2
4
6
8
10
Day
Added targets
N1 CD8
IgG CD8
TC CD8
N1 CD8
IgG CD8
TC CD8

FIG. 11B

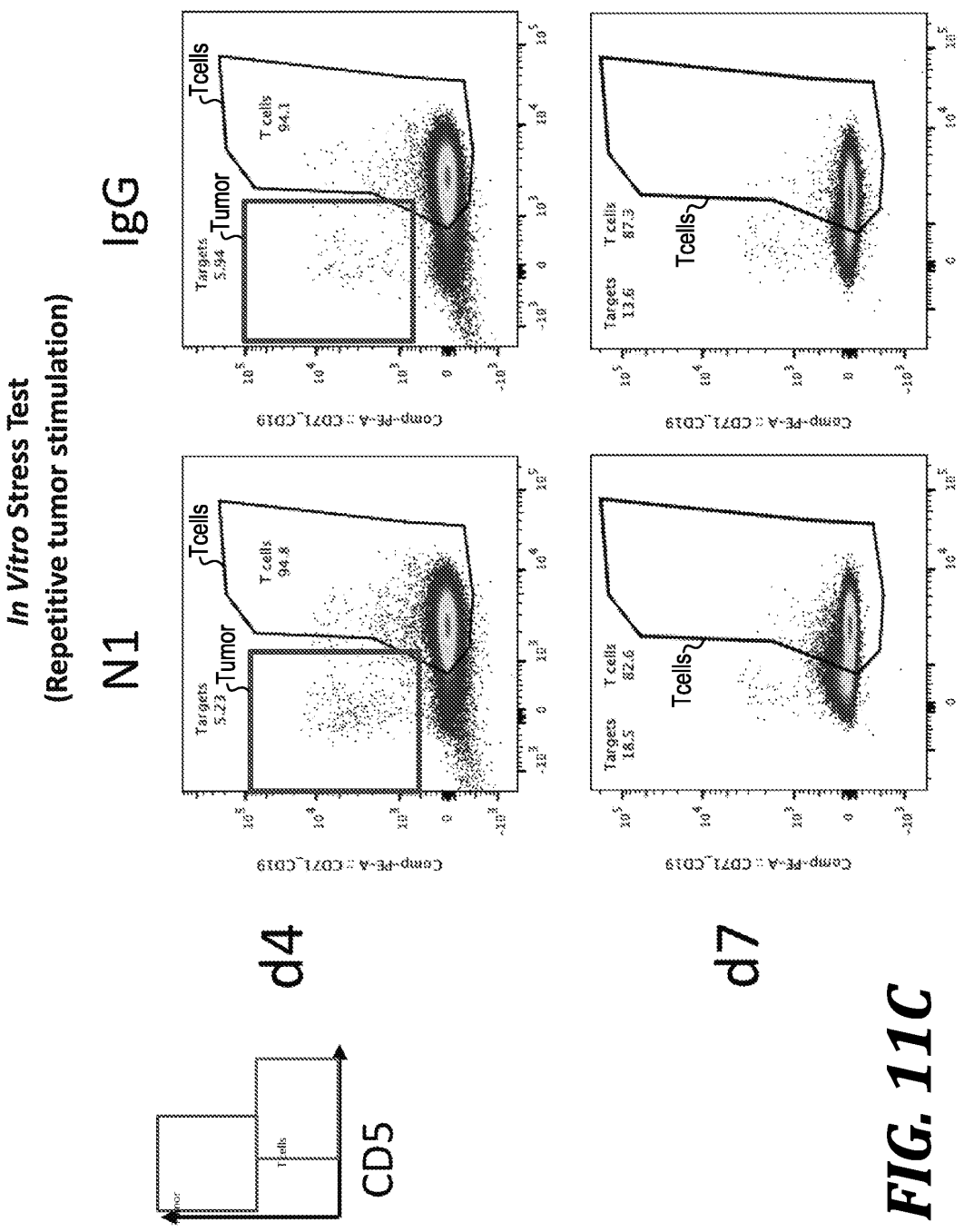

In Vitro Stress Test
(Repetitive tumor stimulation)
N1
IgG
d4
d7
CD19
CD5
Tumor
Tcells
Targets
5.94
T cells
94.1
Targets
5.23
T cells
94.8
Targets
13.6
T cells
87.3
Targets
16.5
T cells
82.6
Comp-PE-A :: CD71_CD19

FIG. 11C

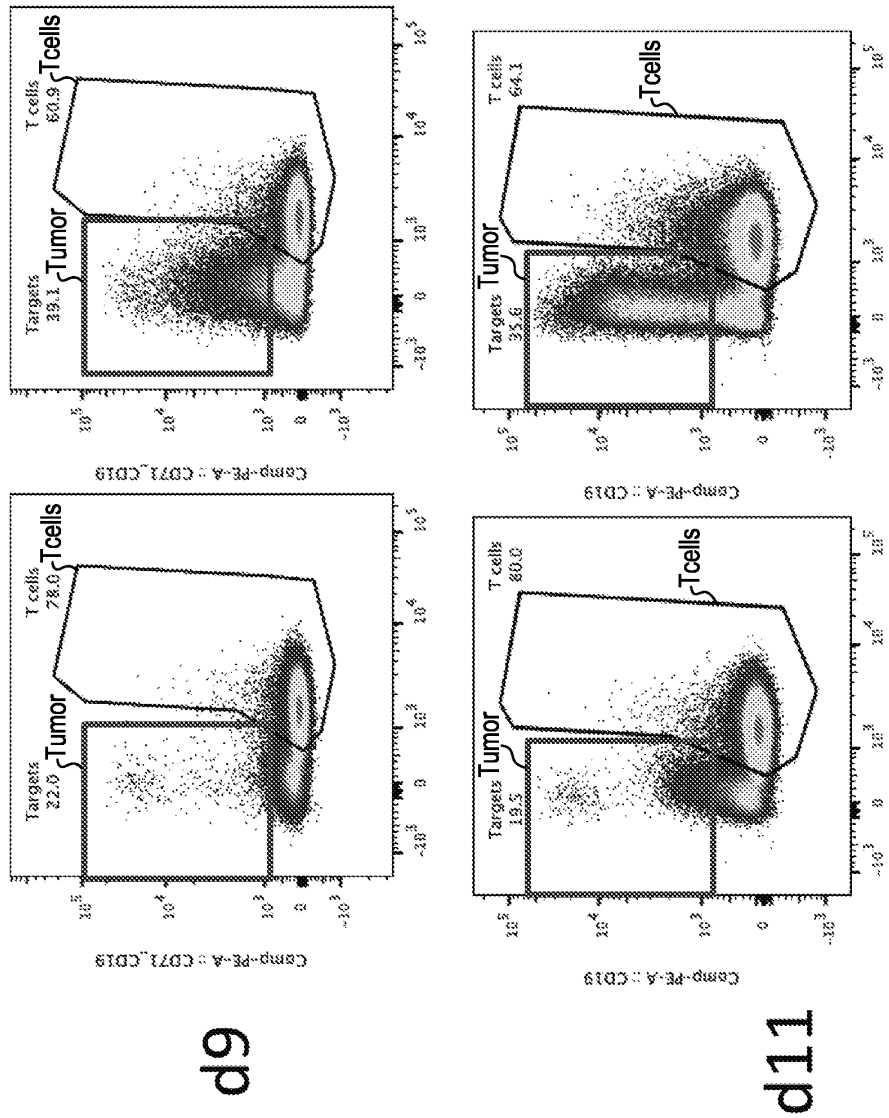

d9
d11
Tumor
Tcells
Targets
T cells
Comp-PE-A :: CD71_CD19
Comp-PE-A :: CD19

FIG. 11C
(CONT.)

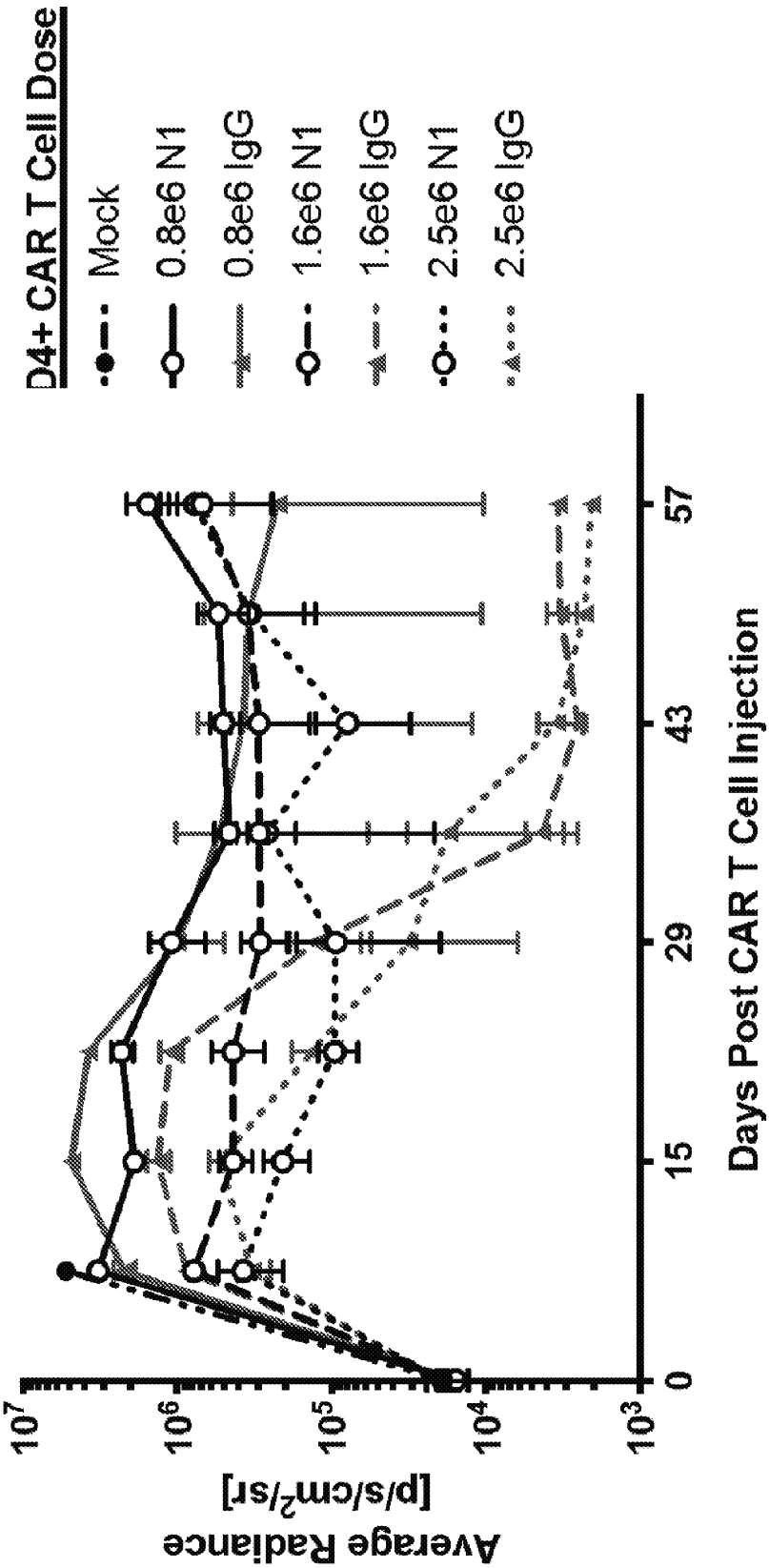

D4+ CAR T Cell Dose
Mock
0.8e6 N1
0.8e6 IgG
1.6e6 N1
1.6e6 IgG
2.5e6 N1
2.5e6 IgG
Average Radiance
[p/s/cm²/sr]
10⁷
10⁶
10⁵
10⁴
10³
0
15
29
43
57
Days Post CAR T Cell Injection

FIG. 12A

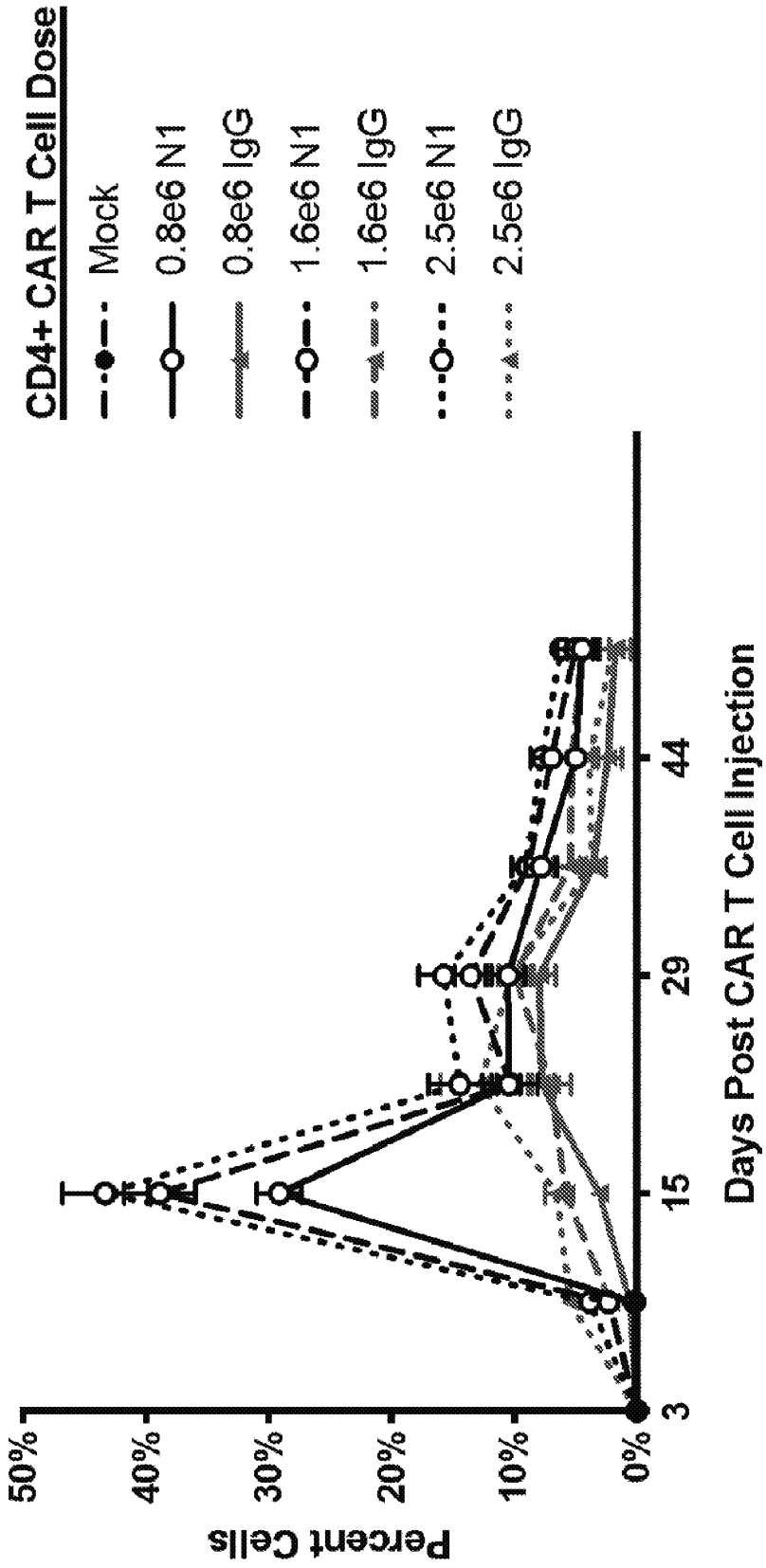

CD4+ CAR T Cell Dose
Mock
0.8e6 N1
0.8e6 IgG
1.6e6 N1
1.6e6 IgG
2.5e6 N1
2.5e6 IgG
Percent Cells
50%
40%
30%
20%
10%
0%
3
15
29
44
Days Post CAR T Cell Injection

FIG. 12B

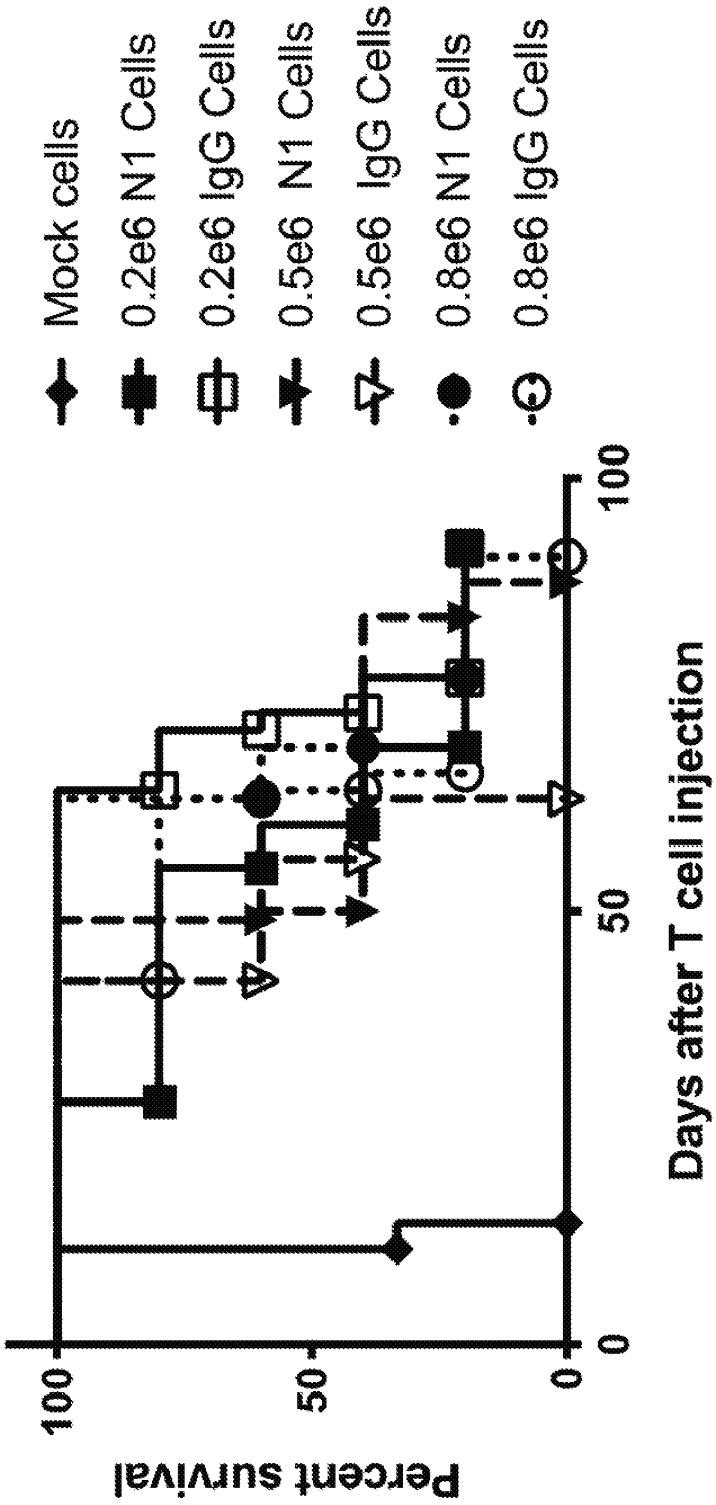

Mock cells
0.2e6 N1 Cells
0.2e6 IgG Cells
0.5e6 N1 Cells
0.5e6 IgG Cells
0.8e6 N1 Cells
0.8e6 IgG Cells
Percent survival
100
50
0
0
50
100
Days after T cell injection

FIG. 13A

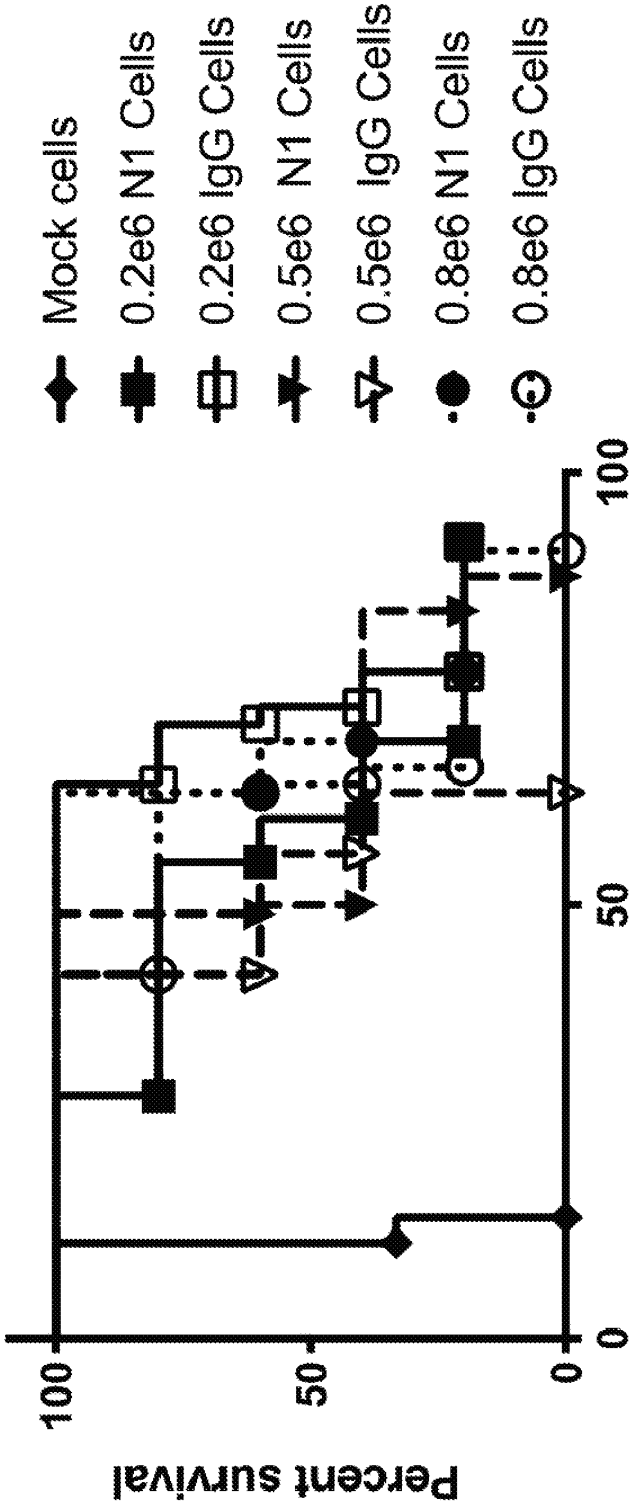

Mock cells
0.2e6 N1 Cells
0.2e6 IgG Cells
0.5e6 N1 Cells
0.5e6 IgG Cells
0.8e6 N1 Cells
0.8e6 IgG Cells
Percent survival
100
50
0
0
50
100
Days after T cell injection

FIG. 13B

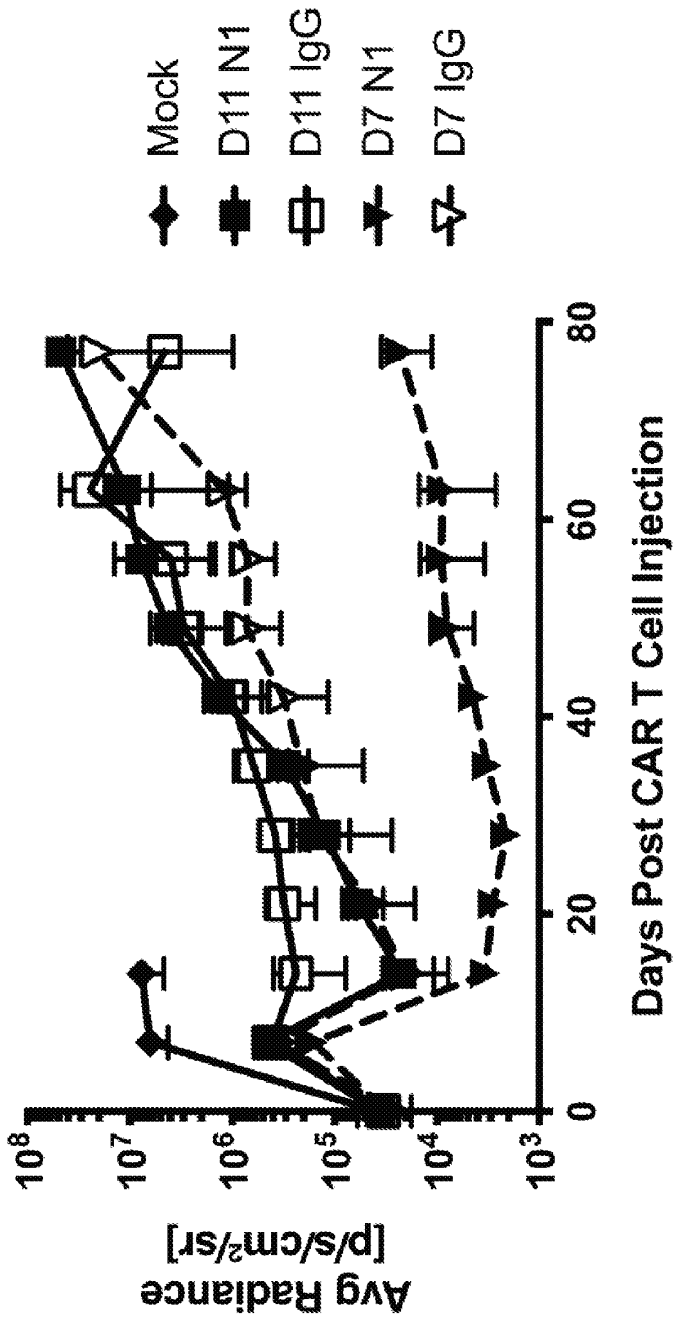

Mock
D11 N1
D11 IgG
D7 N1
D7 IgG
Avg Radiance [p/s/cm²/sr]
10⁸
10⁷
10⁶
10⁵
10⁴
10³
0
20
40
60
80
Days Post CAR T Cell Injection

FIG. 14A

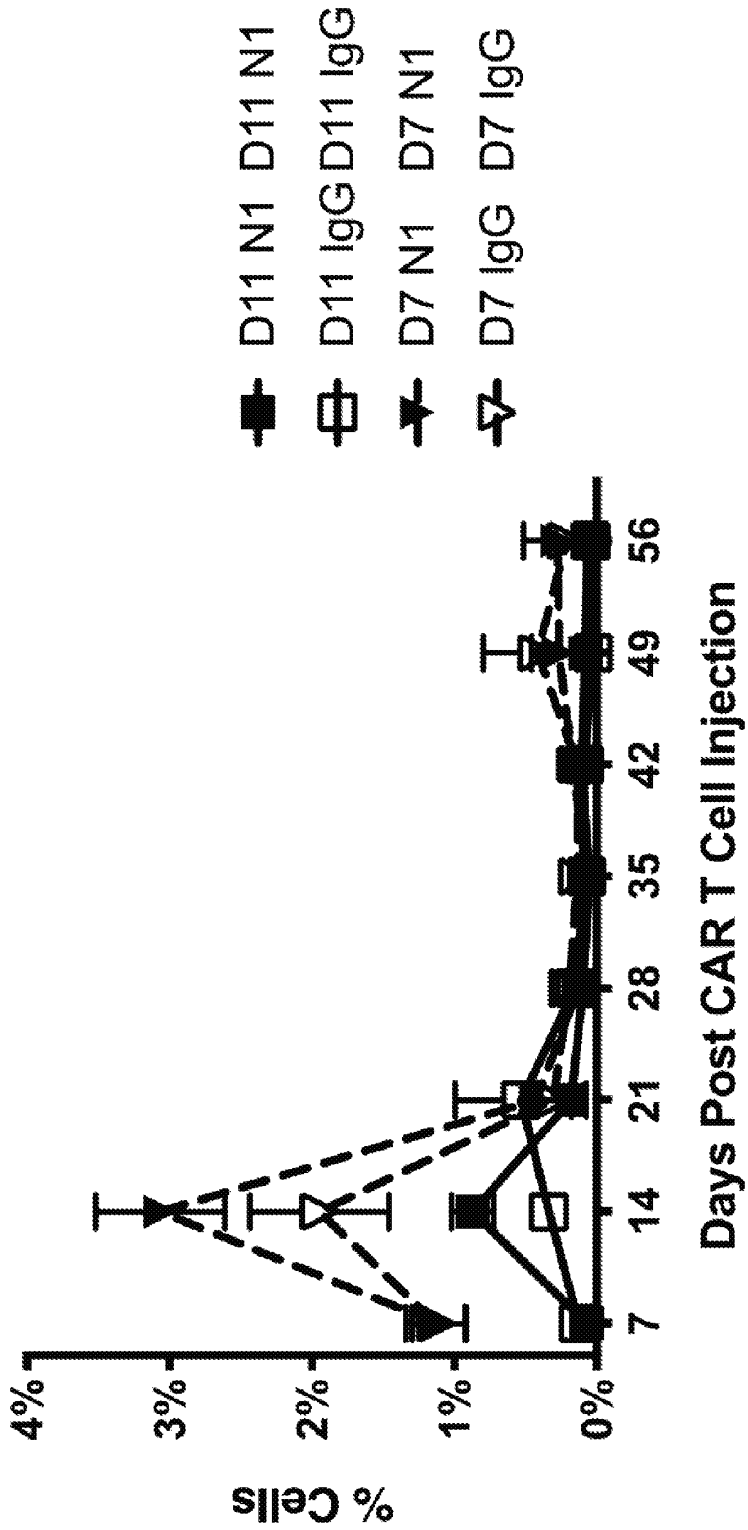

D11 N1 D11 N1
D11 IgG D11 IgG
D7 N1 D7 N1
D7 IgG D7 IgG
% Cells
4%
3%
2%
1%
0%
7
14
21
28
35
42
49
56
Days Post CAR T Cell Injection

FIG. 14B

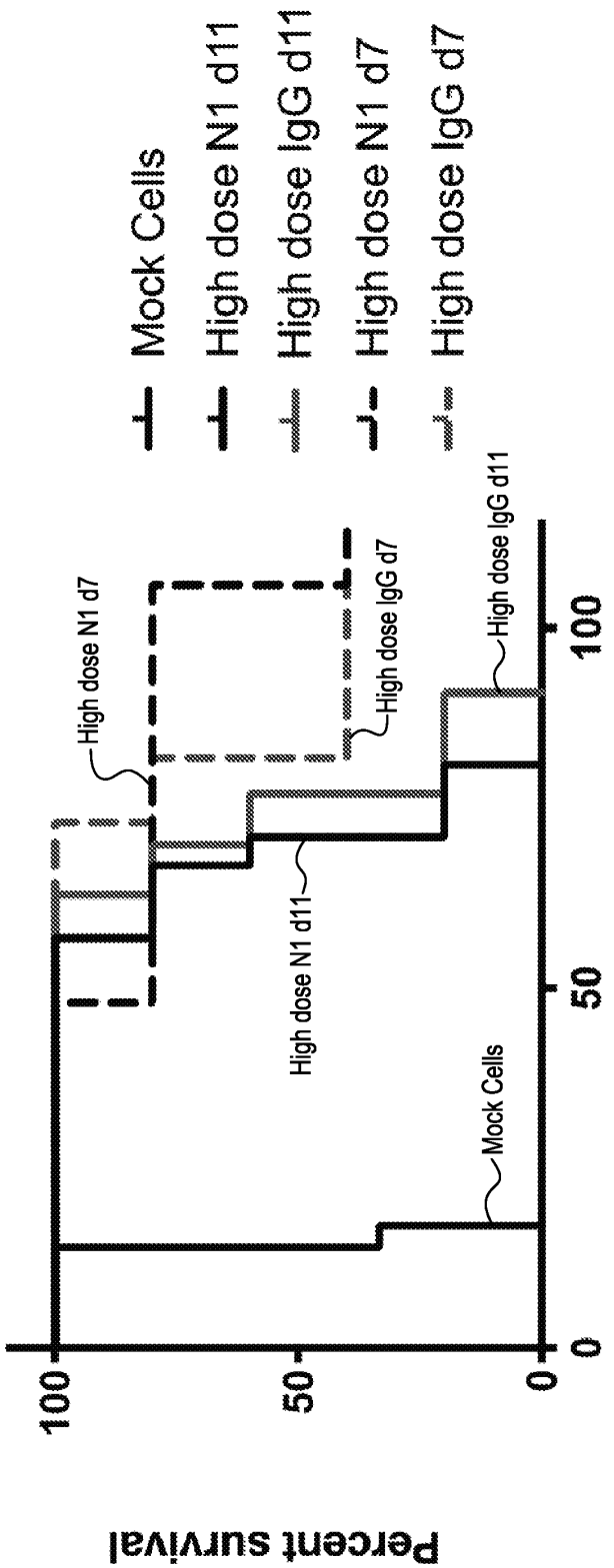

Mock Cells
High dose N1 d11
High dose IgG d11
High dose N1 d7
High dose IgG d7
Percent survival
100
50
0
0
50
100
Days
High dose N1 d7
High dose N1 d11
High dose IgG d7
High dose IgG d11
Mock Cells

FIG. 14C

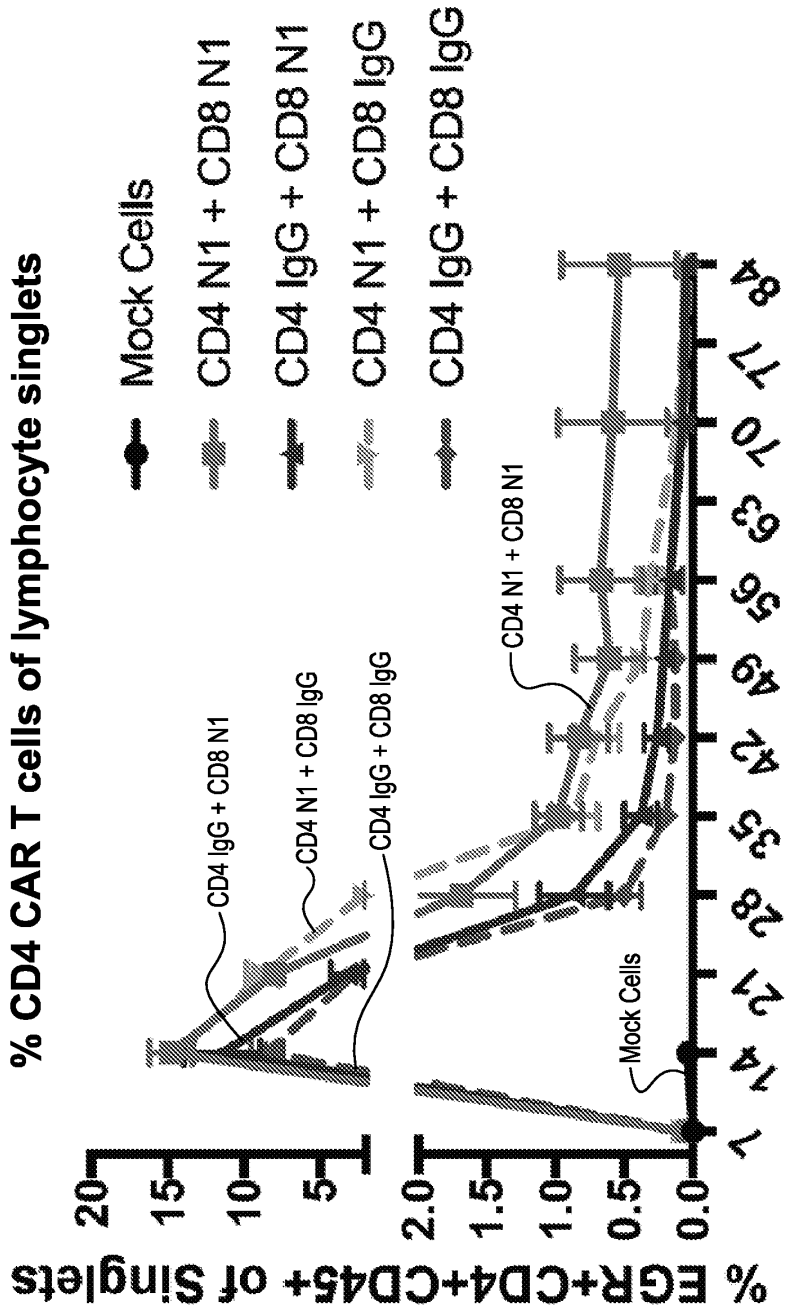

% CD4 CAR T cells of lymphocyte singlets
Mock Cells
CD4 N1 + CD8 N1
CD4 IgG + CD8 N1
CD4 N1 + CD8 IgG
CD4 IgG + CD8 IgG
CD4 IgG + CD8 N1
CD4 N1 + CD8 IgG
CD4 IgG + CD8 IgG
CD4 N1 + CD8 N1
Mock Cells
% EGR+CD4+CD45+ of Singlets
20
15
10
5
2.0
1.5
1.0
0.5
0.0
1
14
21
28
35
42
49
56
63
70
77
84

FIG. 15A

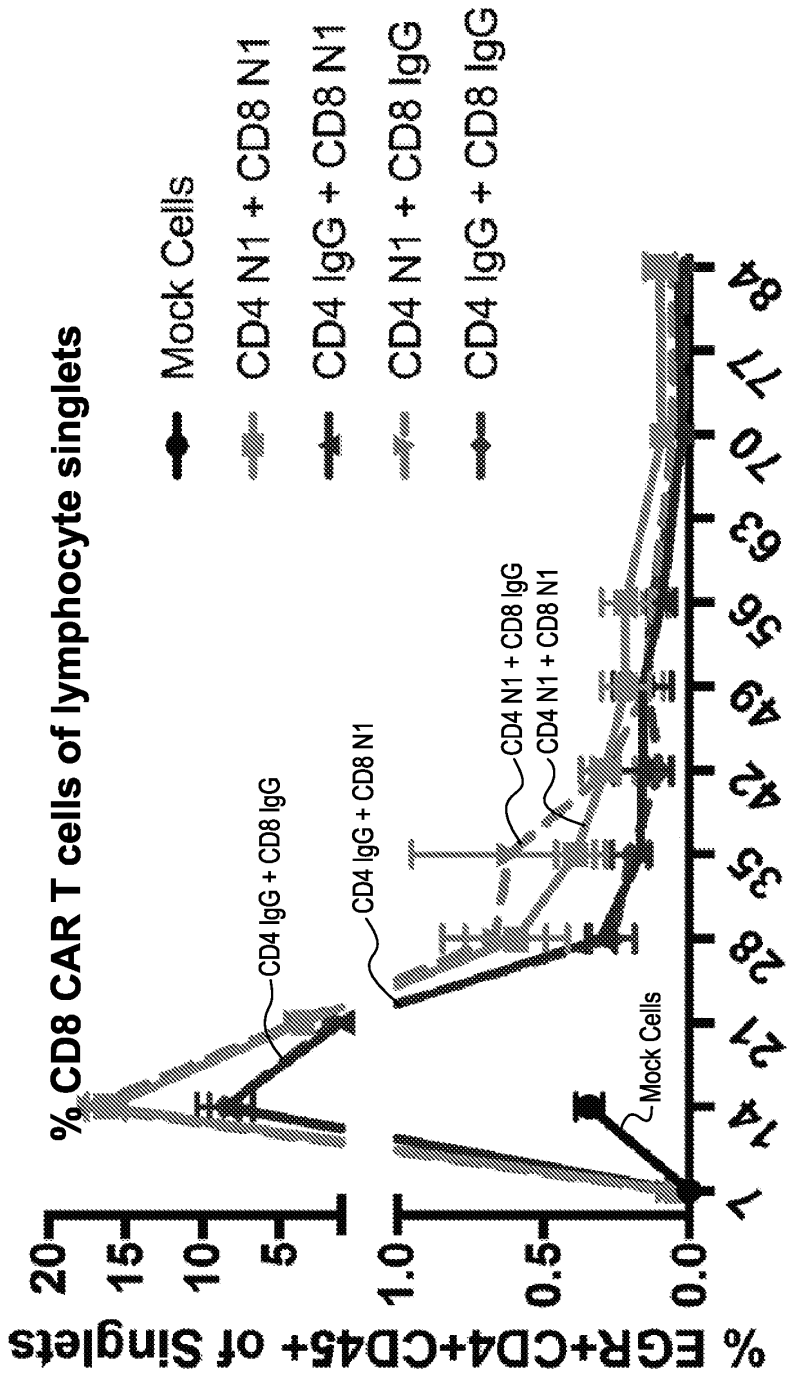

% CD8 CAR T cells of lymphocyte singlets
% EGR+CD4+CD45+ of Singlets
Mock Cells
CD4 N1 + CD8 N1
CD4 IgG + CD8 N1
CD4 N1 + CD8 IgG
CD4 IgG + CD8 IgG
CD4 IgG + CD8 IgG
CD4 IgG + CD8 N1
CD4 N1 + CD8 IgG
CD4 N1 + CD8 N1
Mock Cells
20
15
10
5
1.0
0.5
0.0
7
14
21
28
35
42
49
56
63
70
77
84

FIG. 15B

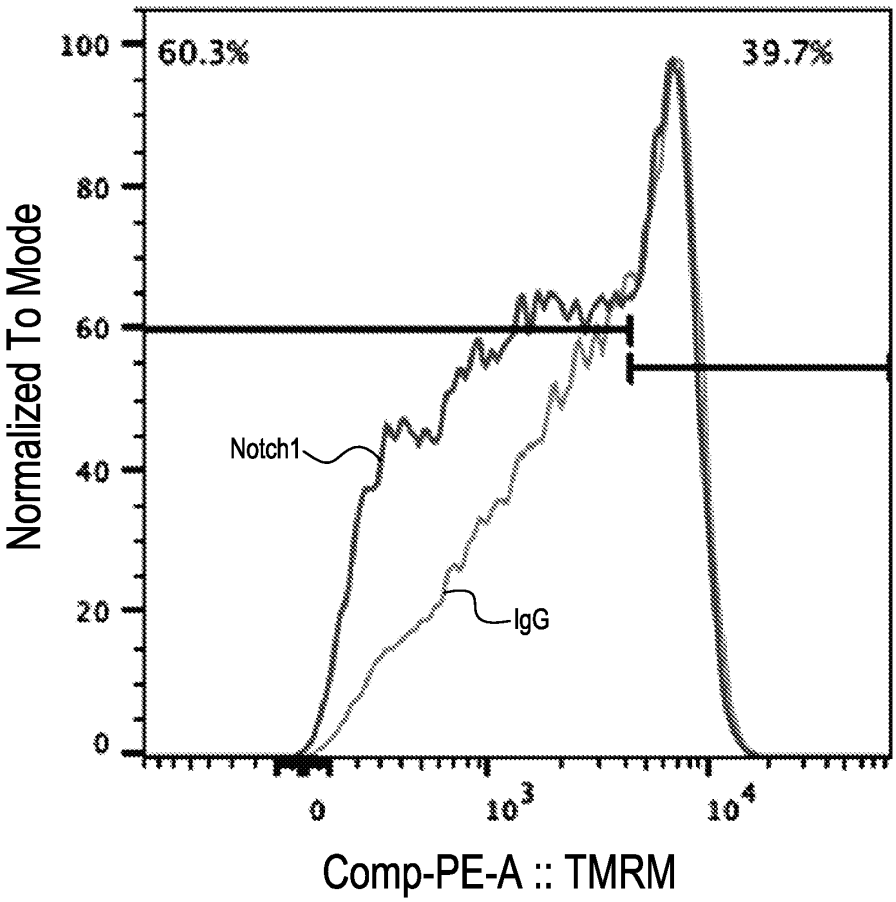

60.3%
39.7%
Normalized To Mode
100
80
60
40
20
0
Notch1
IgG
0
10^3
10^4
Comp-PE-A :: TMRM

FIG. 16A

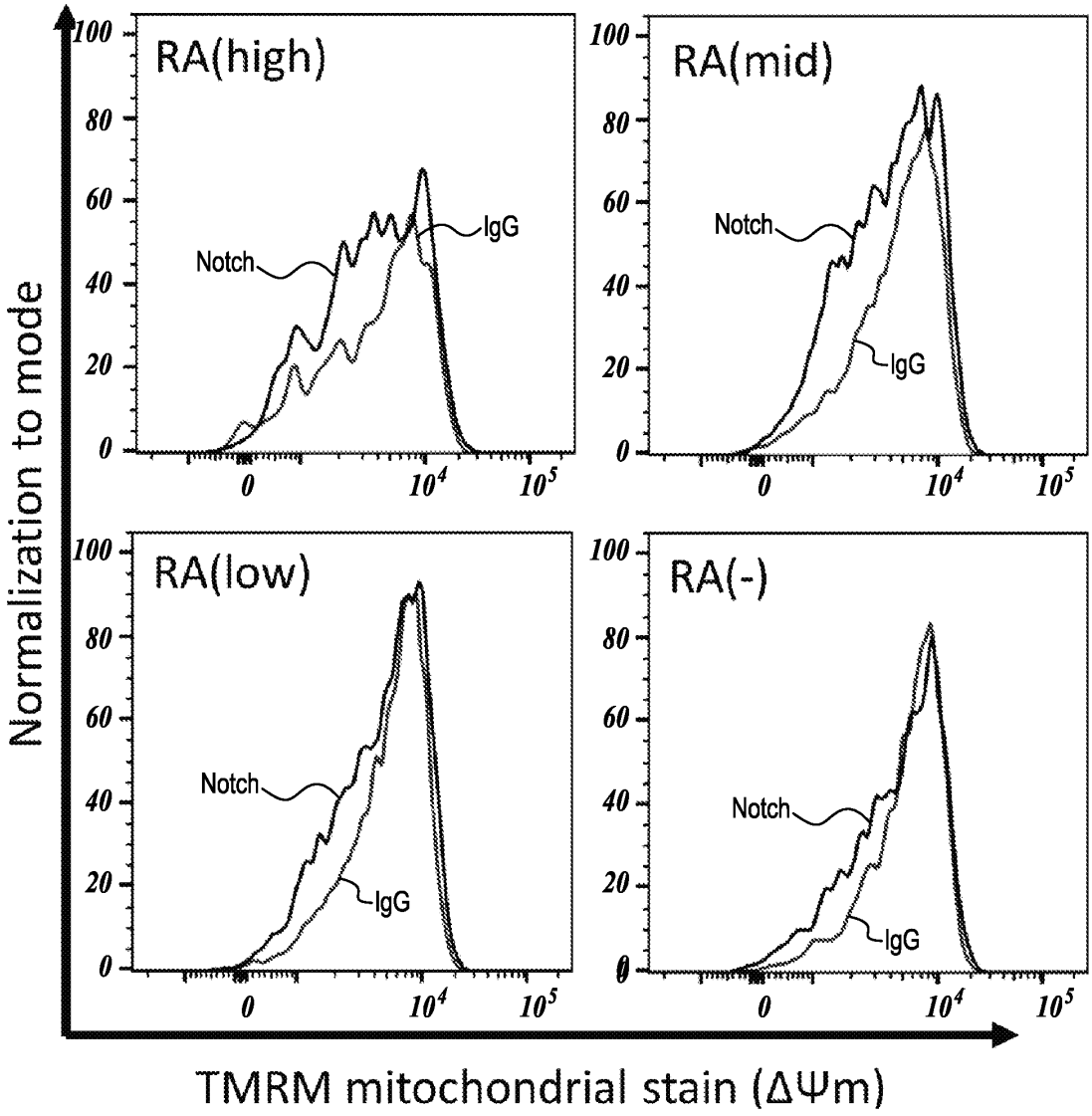

RA(high)
RA(mid)
RA(low)
RA(-)
Notch
IgG
Normalization to mode
TMRM mitochondrial stain (ΔΨm)
0
20
40
60
80
100
0
10^4
10^5

FIG. 16B

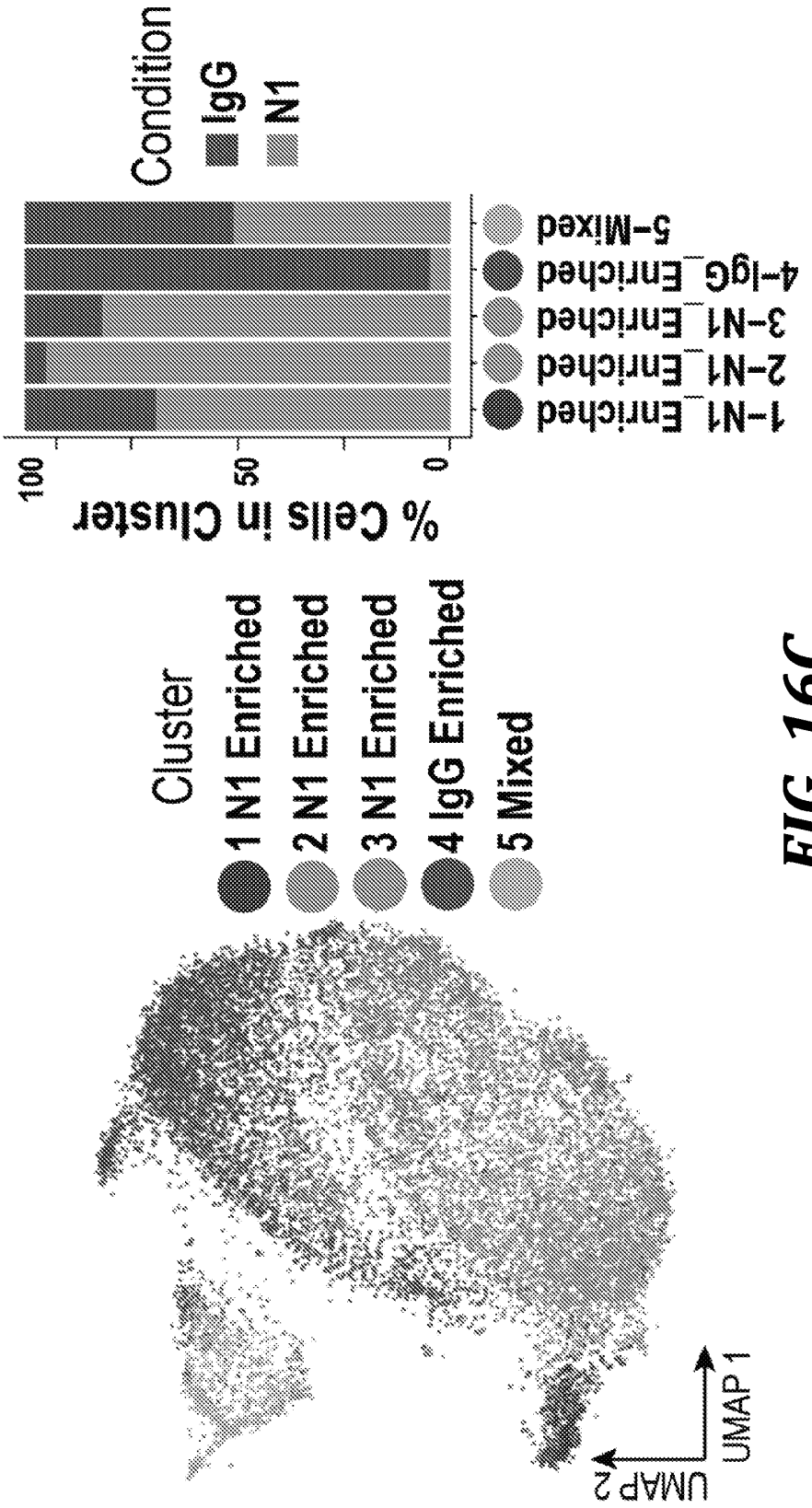

Condition
IgG
N1
% Cells in Cluster
100
50
0
1-N1_Enriched
2-N1_Enriched
3-N1_Enriched
4-IgG_Enriched
5-Mixed
Cluster
1 N1 Enriched
2 N1 Enriched
3 N1 Enriched
4 IgG Enriched
5 Mixed
UMAP 1
UMAP 2

FIG. 16C

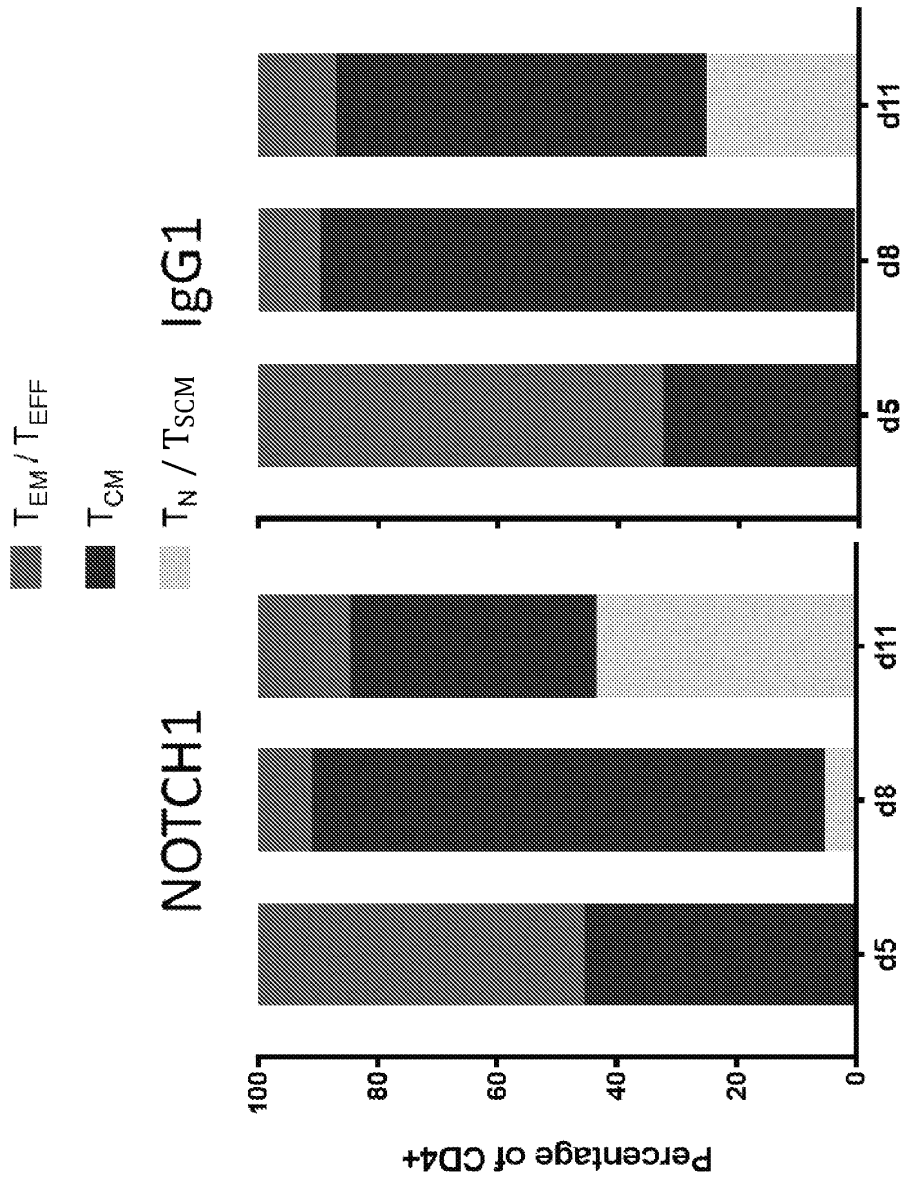

Percentage of CD4+
0
20
40
60
80
100
NOTCH1
IgG1
d5
d8
d11
d5
d8
d11
T_{EM} / T_{EFF}
T_{CM}
T_N / T_{SCM}

FIG. 17

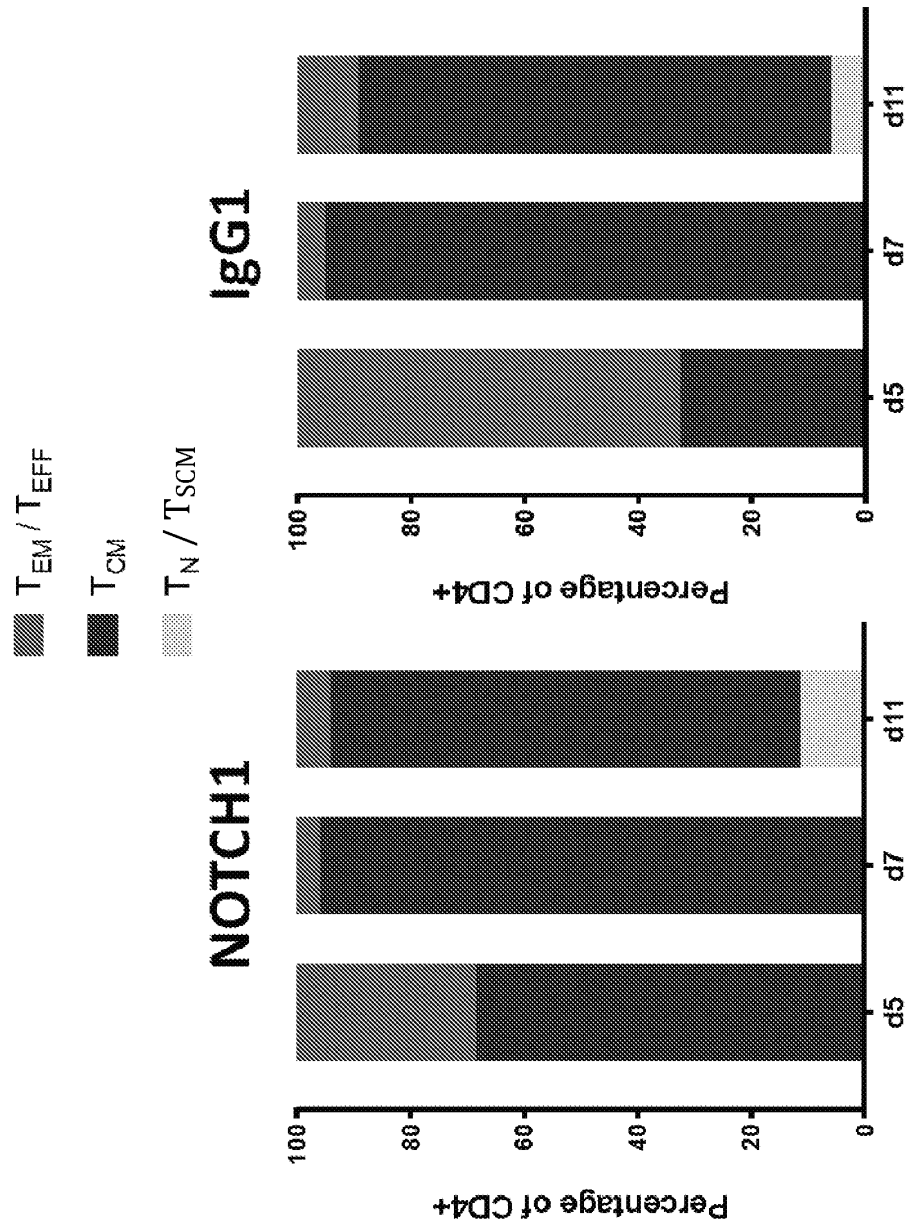

NOTCH1
IgG1
Percentage of CD4+
100
80
60
40
20
0
d5
d7
d11
T_{EM} / T_{EFF}
T_{CM}
T_N / T_{SCM}

FIG. 18A

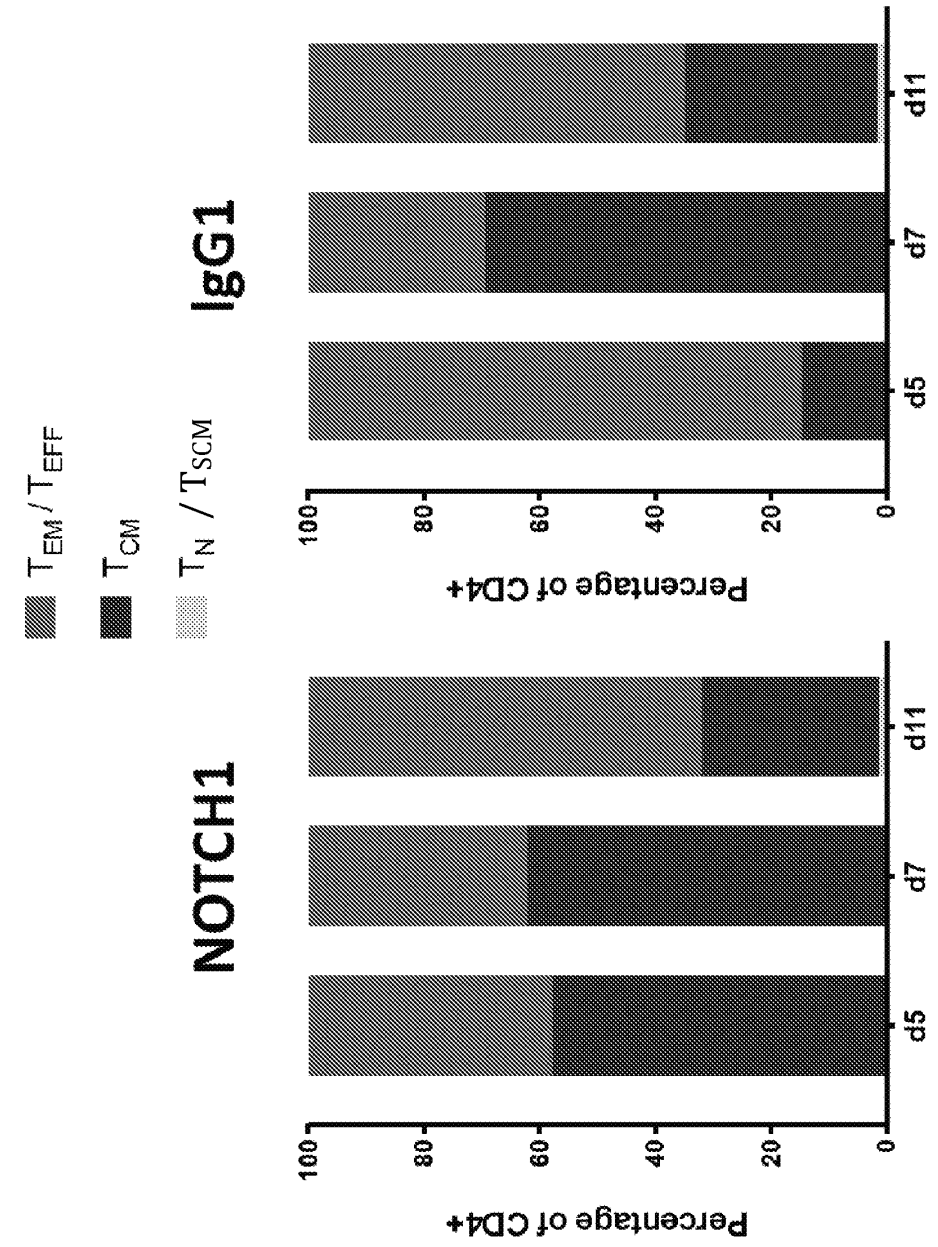

NOTCH1
IgG1
T_{EM} / T_{EFF}
T_{CM}
T_N / T_{SCM}
Percentage of CD4+
0
20
40
60
80
100
d5
d7
d11

FIG. 18B

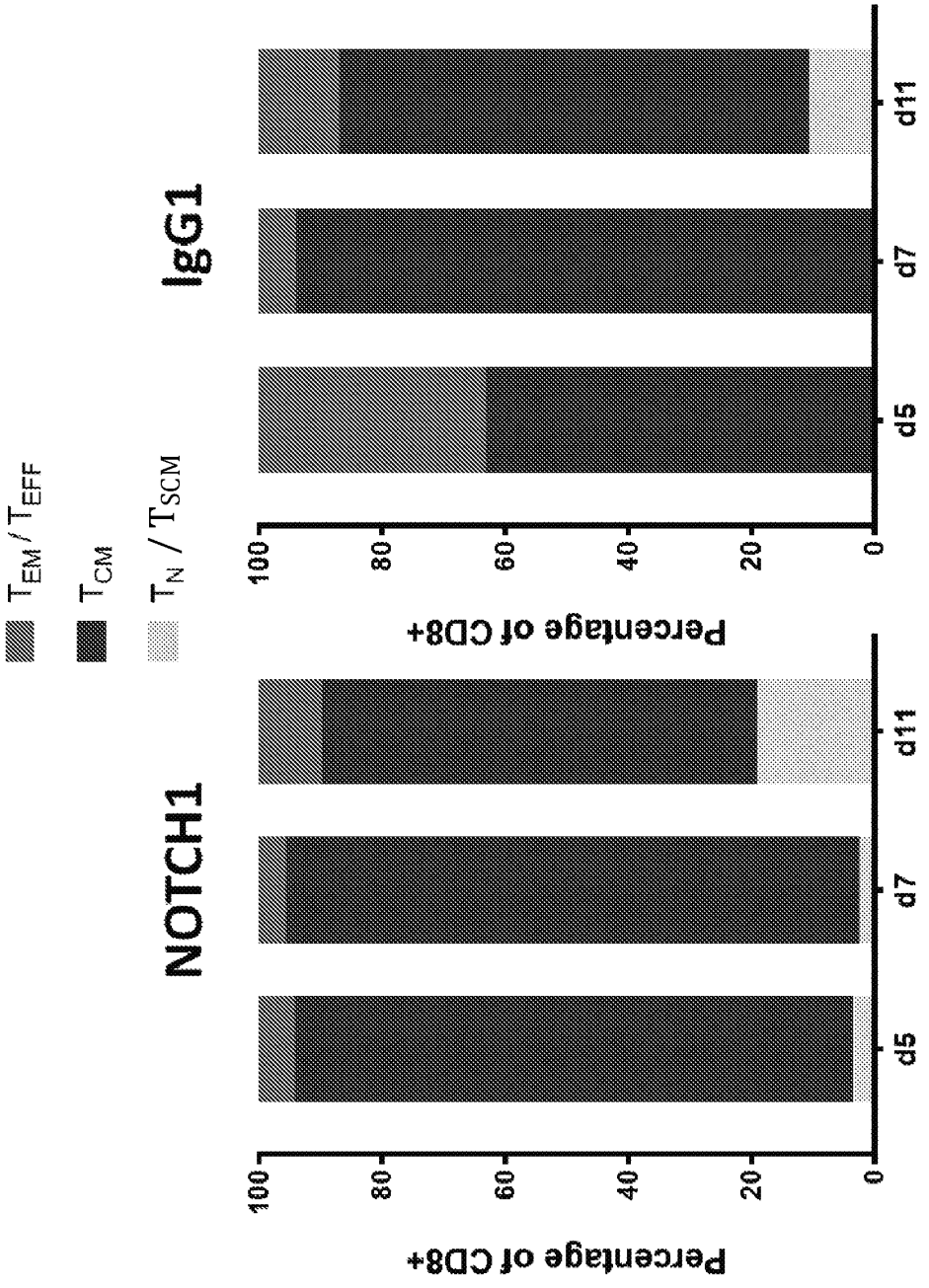

NOTCH1
IgG1
Percentage of CD8+
100
80
60
40
20
0
d5
d7
d11
T_{EM} / T_{EFF}
T_{CM}
T_N / T_{SCM}

FIG. 19A

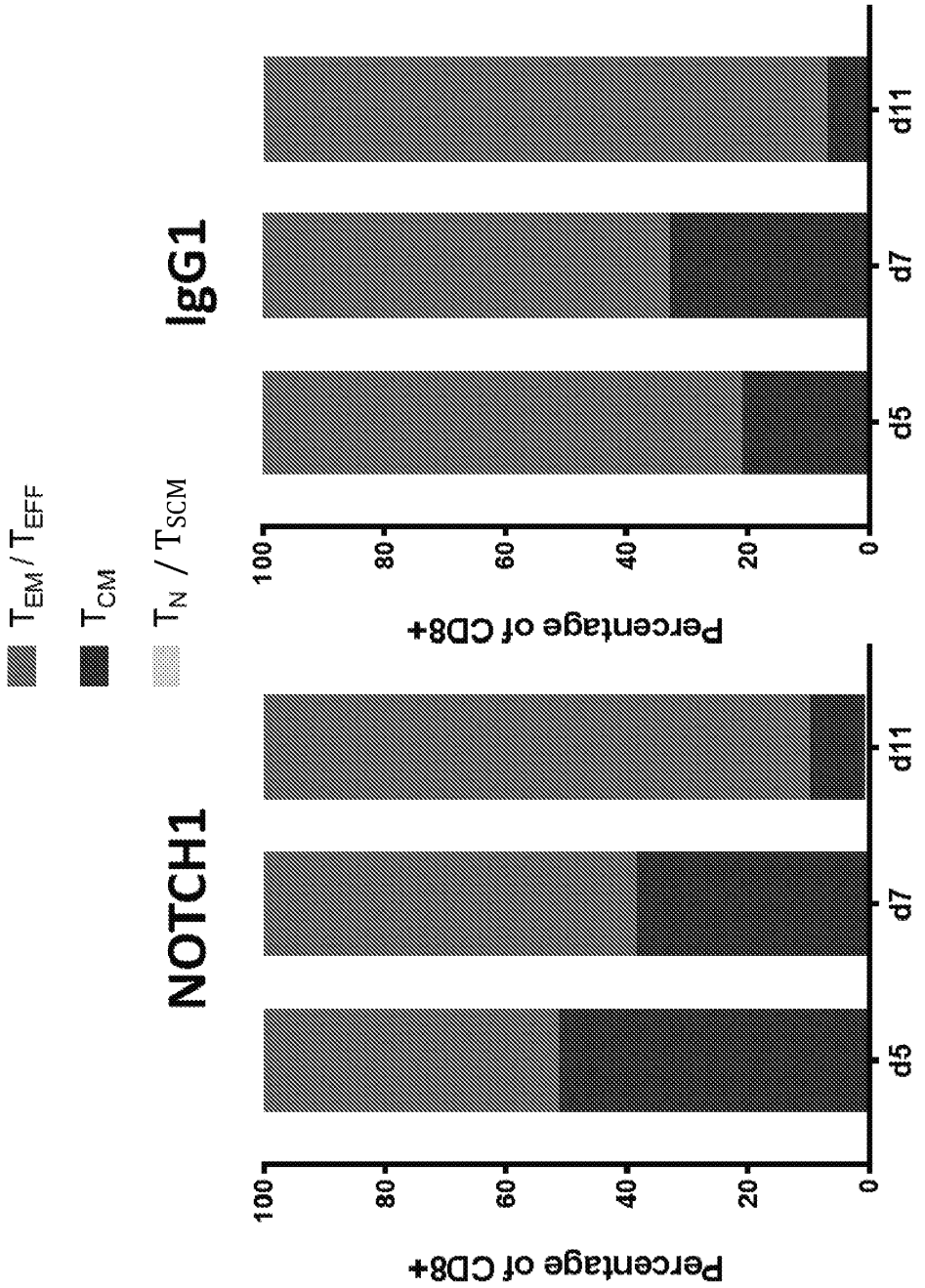

NOTCH1
IgG1
T_{EM} / T_{EFF}
T_{CM}
T_N / T_{SCM}
Percentage of CD8+
0
20
40
60
80
100
d5
d7
d11

FIG. 19B

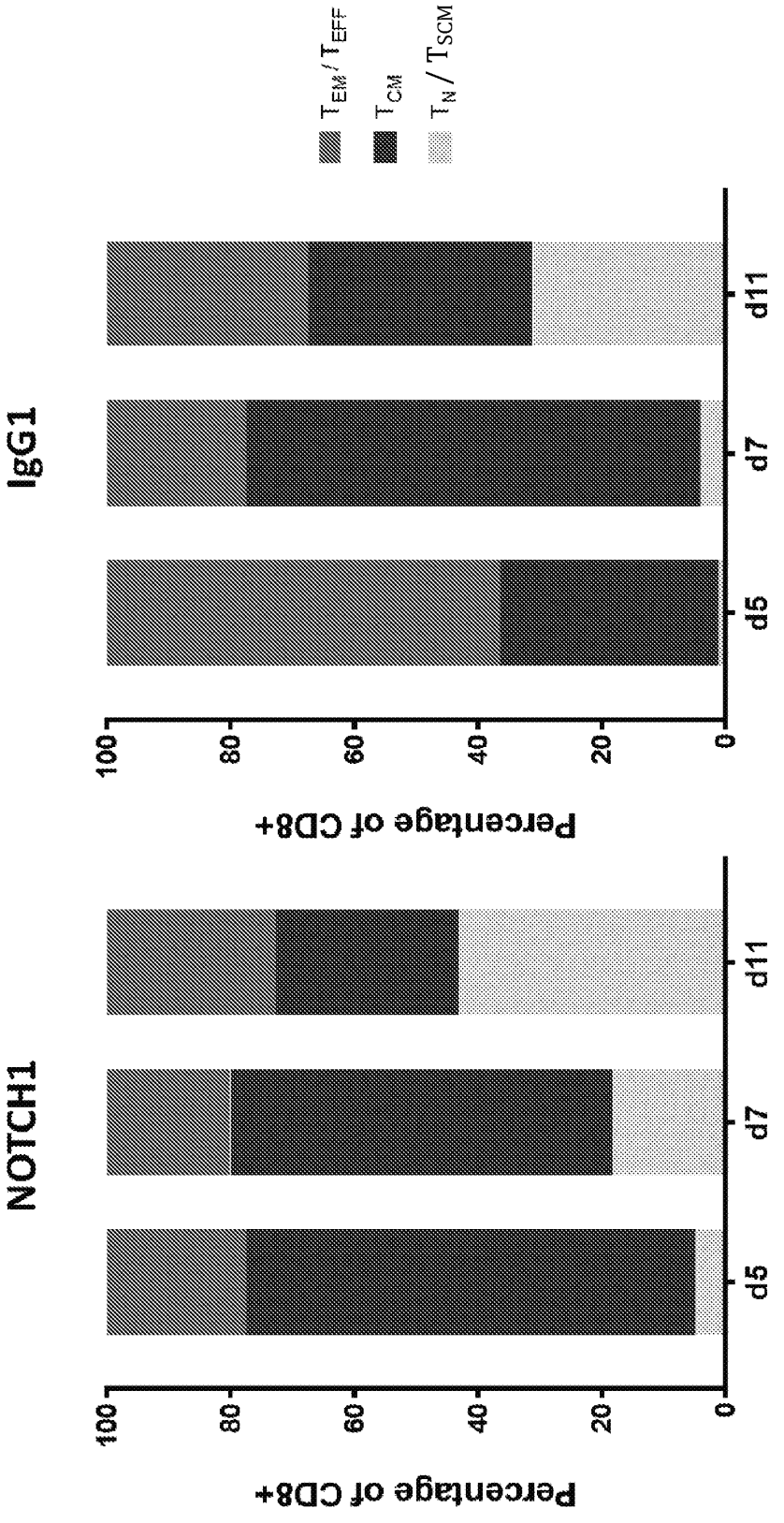

NOTCH1
IgG1
Percentage of CD8+
100
80
60
40
20
0
d5
d7
d11
T_{EM} / T_{EFF}
T_{CM}
T_N / T_{SCM}

FIG. 20