Figure 1

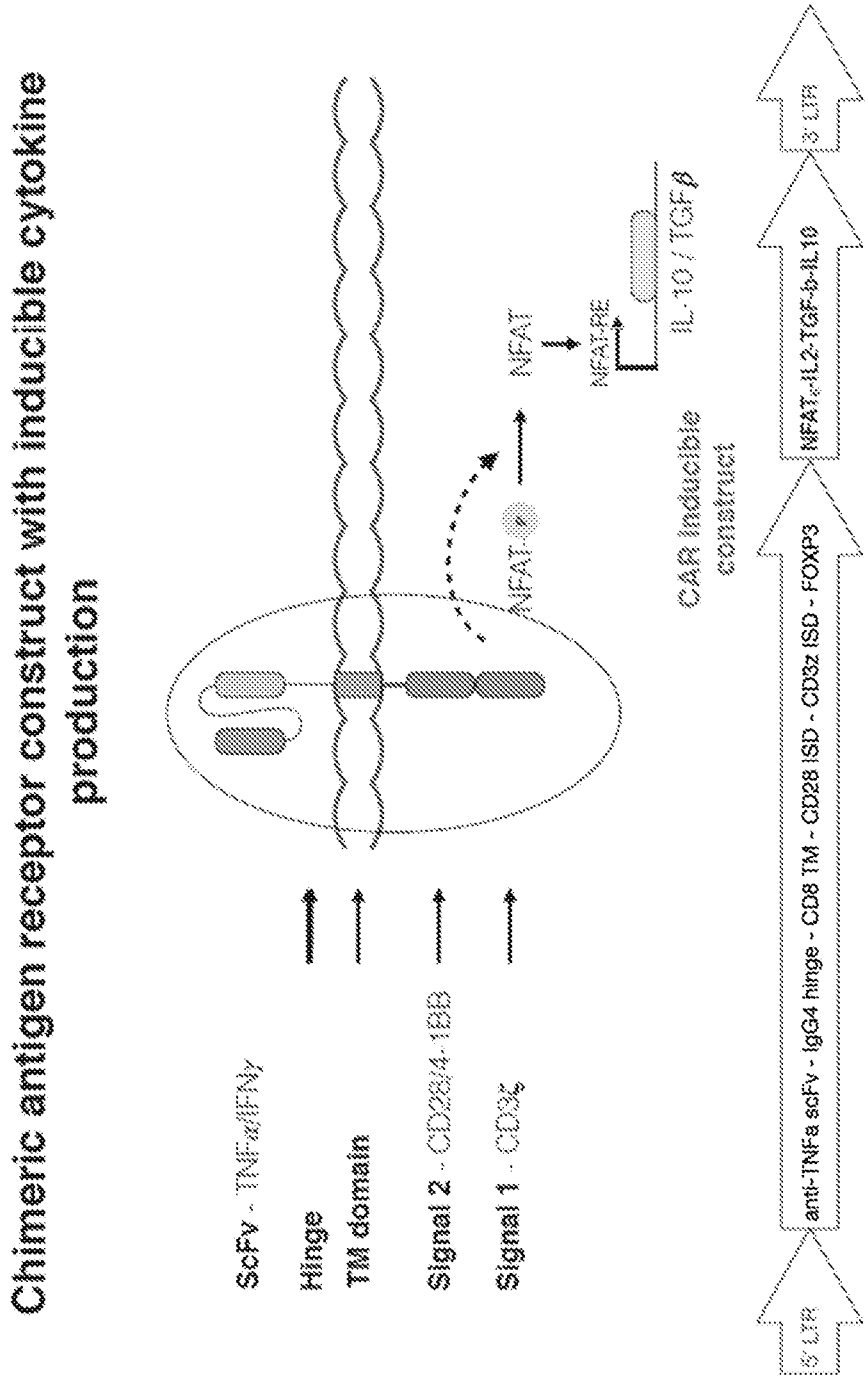

Chimeric antigen receptor construct with inducible cytokine production
ScFv - TNFα/IFNγ
Hinge
TM domain
Signal 2 - CD28/4-1BB
Signal 1 - CD3ζ
NFAT
NFAT-RE
IL-10 / TGFβ
CAR inducible construct
5' LTR
anti-TNFa scFv - IgG4 hinge - CD8 TM - CD28 ISD - CD3z ISD - FOXP3
NFAT-IL2-TGF-b-IL10
3' LTR
Inducible chimeric antigen receptor - Recognition of TNFα/IFNγ by scFv leads to signalling via CD3z and CD28, activating NFAT, which in turn induces expression of cytokines (IL10/TGFb).

Figure 2A

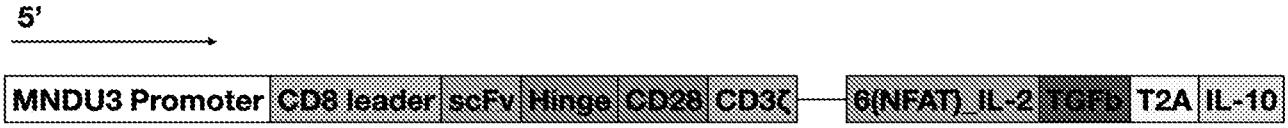


Figure 2B

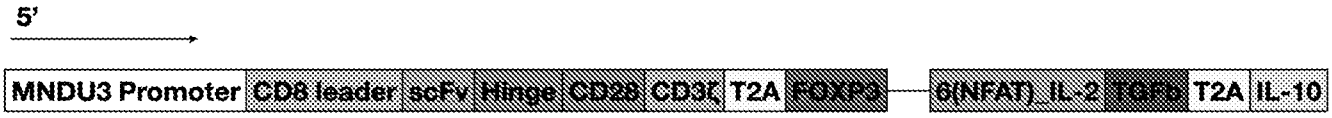


Figure 3A

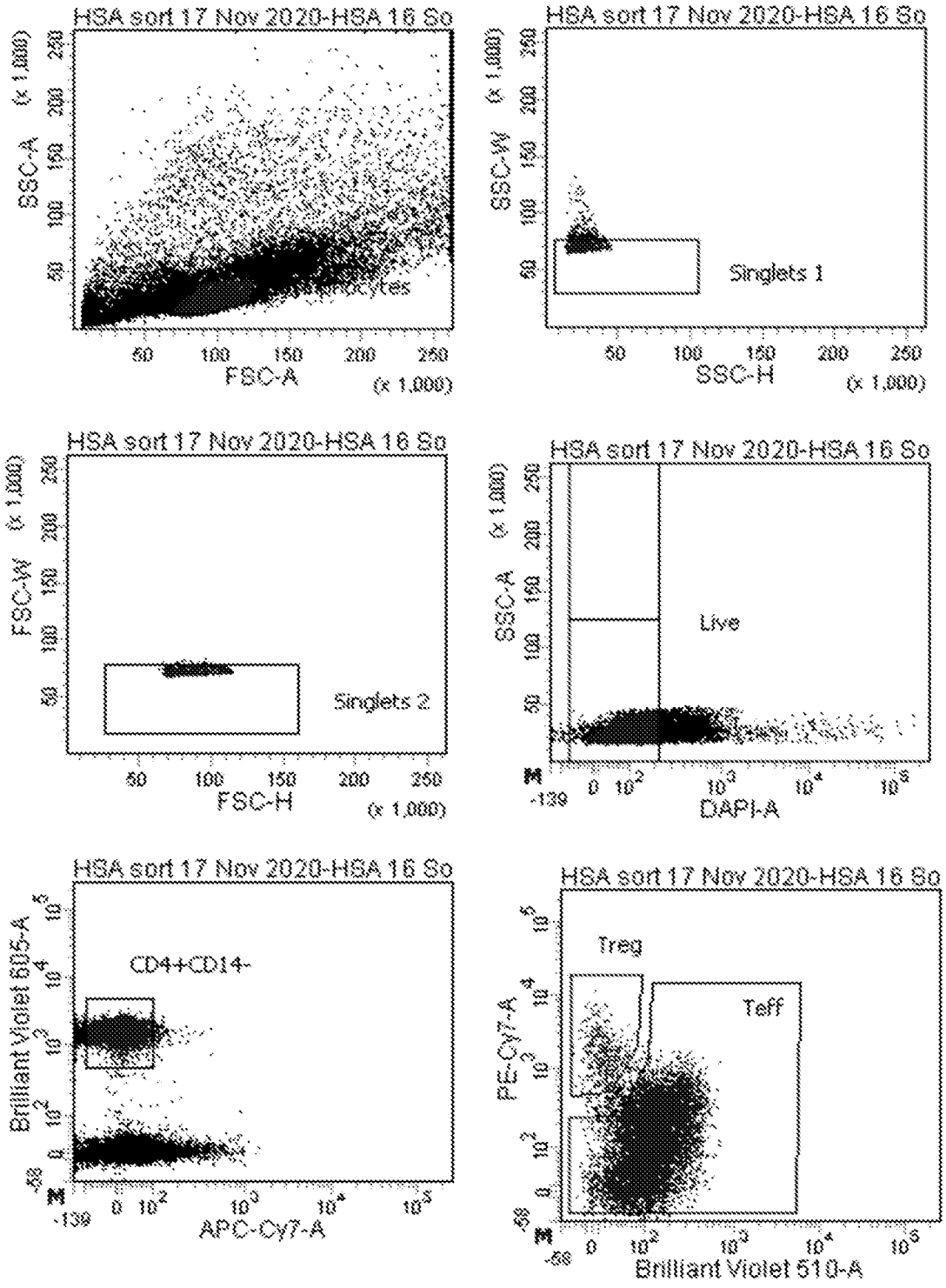

HSA sort 17 Nov 2020-HSA 16 So
SSC-A
(x 1,000)
FSC-A
(x 1,000)
Lymphocytes
SSC-W
SSC-H
Singlets 1
FSC-W
FSC-H
Singlets 2
Live
DAPI-A
CD4+CD14-
Brilliant Violet 605-A
APC-Cy7-A
Treg
Teff
PE-Cy7-A
Brilliant Violet 510-A

Figure 3B

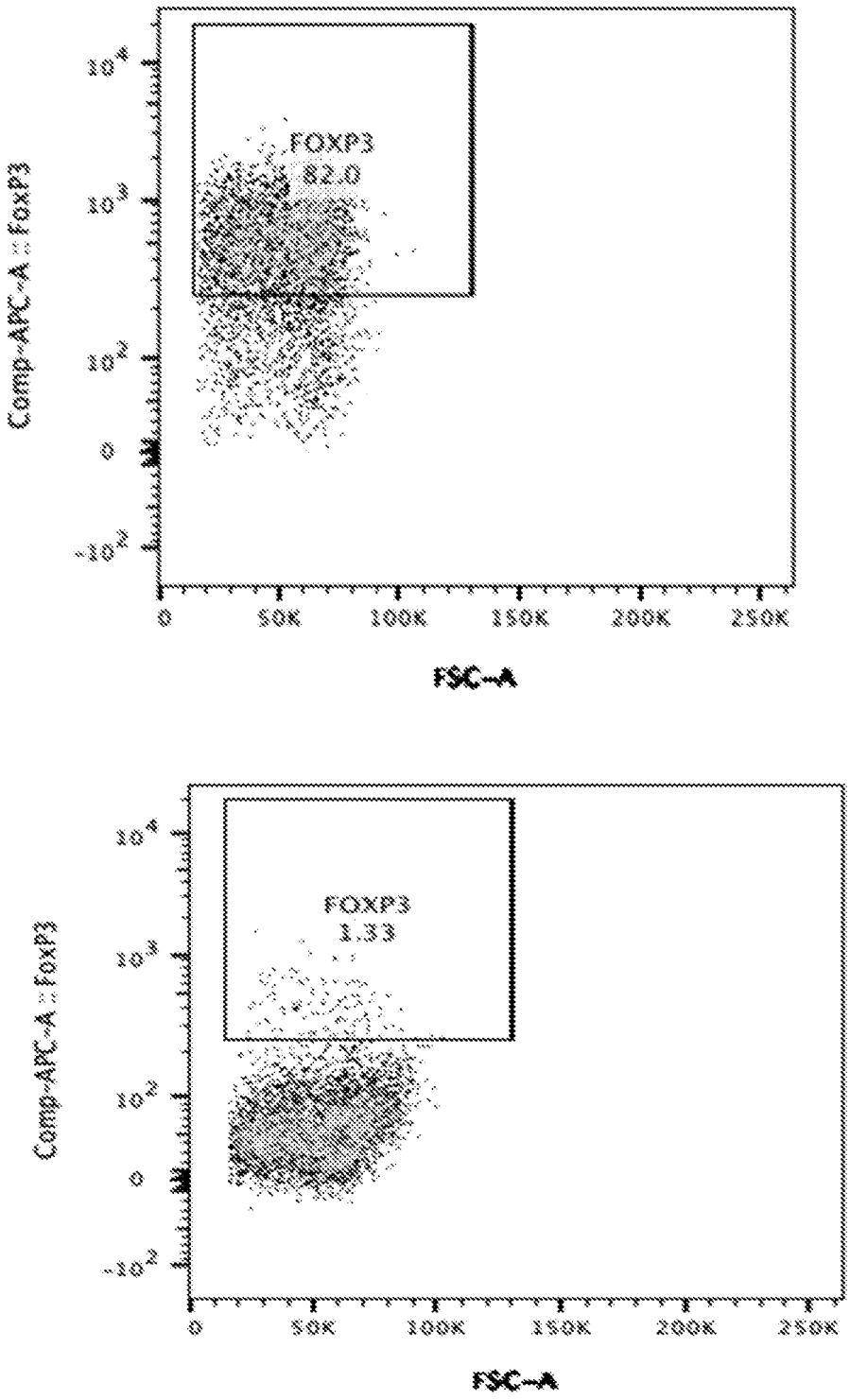

FOXP3
82.0
Comp-APC-A :: FoxP3
FSC-A
FOXP3
1.33
Comp-APC-A :: FoxP3
FSC-A

Figure 4A

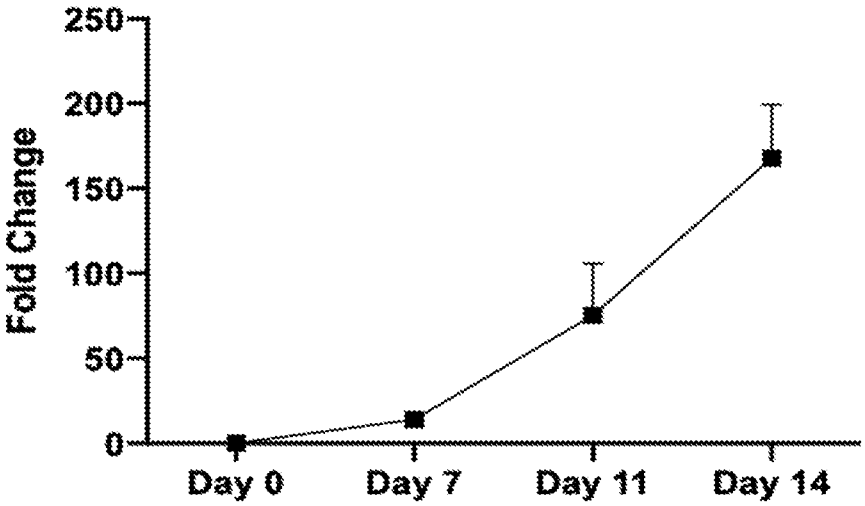
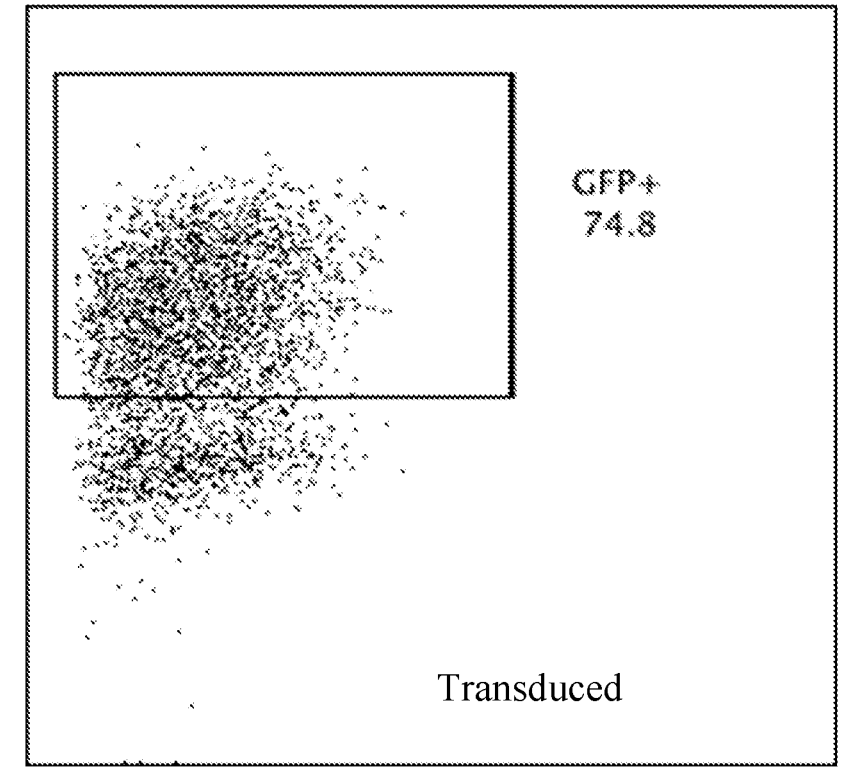


Figure 4B



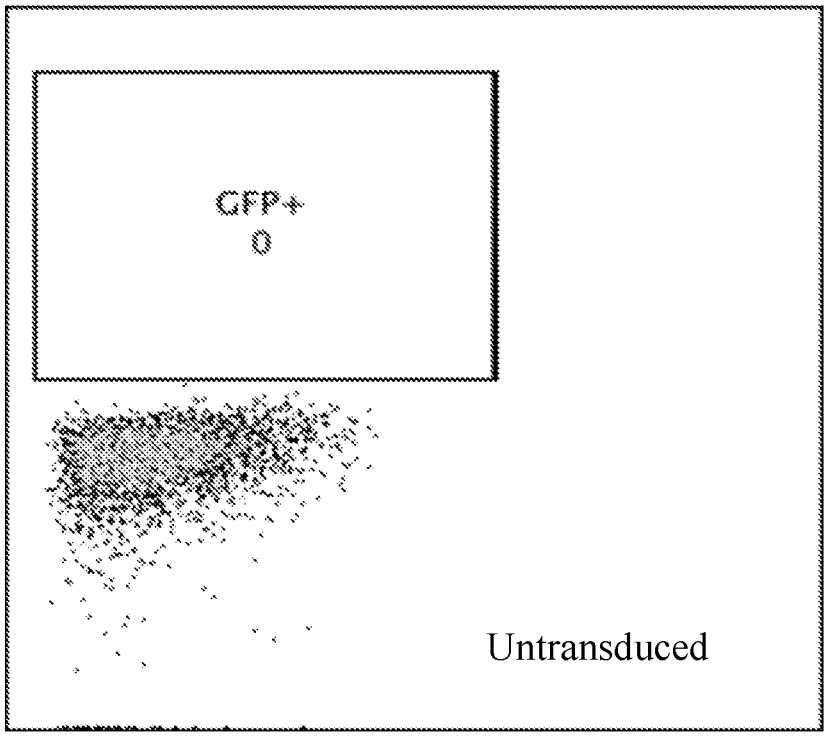


Figure 5

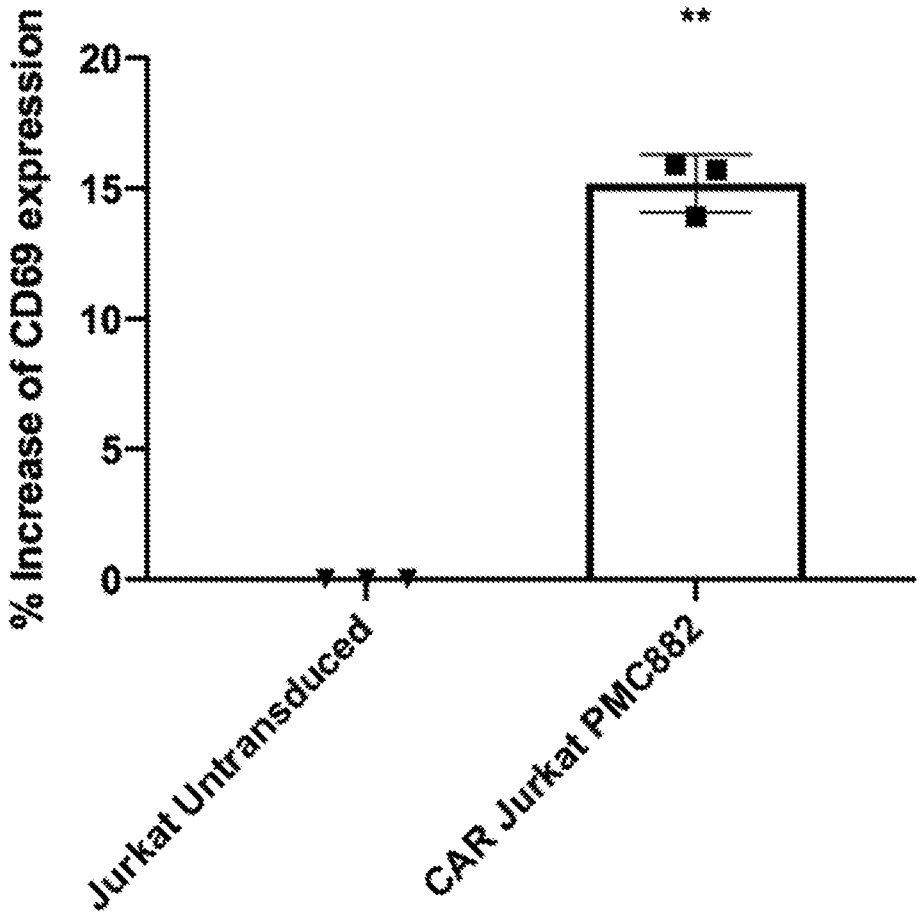

**
20
15
10
5
0
% Increase of CD69 expression
Jurkat Untransduced
CAR Jurkat PMC882

Figure 6A

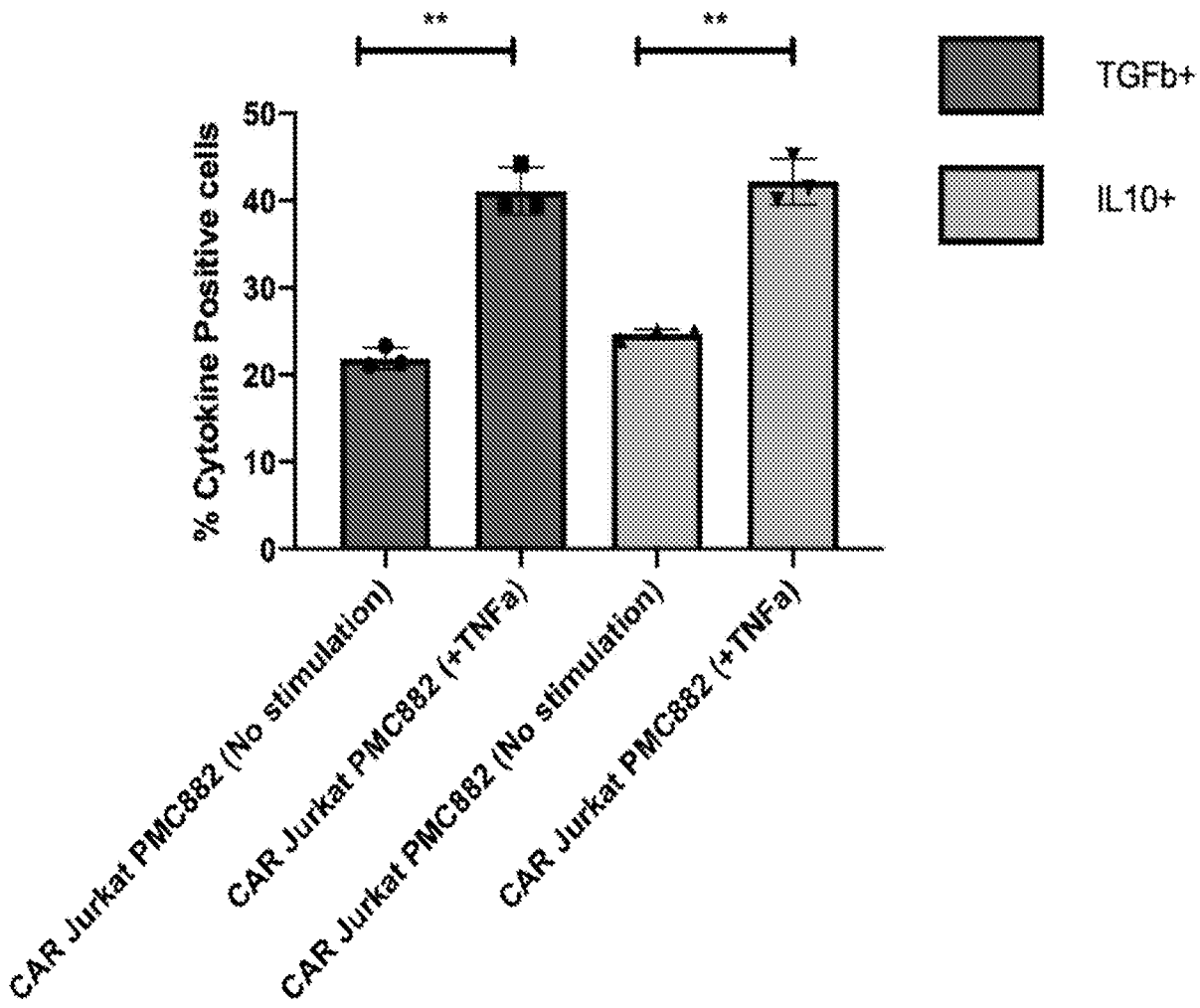

**
**
TGFb+
IL10+
% Cytokine Positive cells
50
40
30
20
10
0
CAR Jurkat PMC882 (No stimulation)
CAR Jurkat PMC882 (+TNFa)
CAR Jurkat PMC882 (No stimulation)
CAR Jurkat PMC882 (+TNFa)

Figure 6B

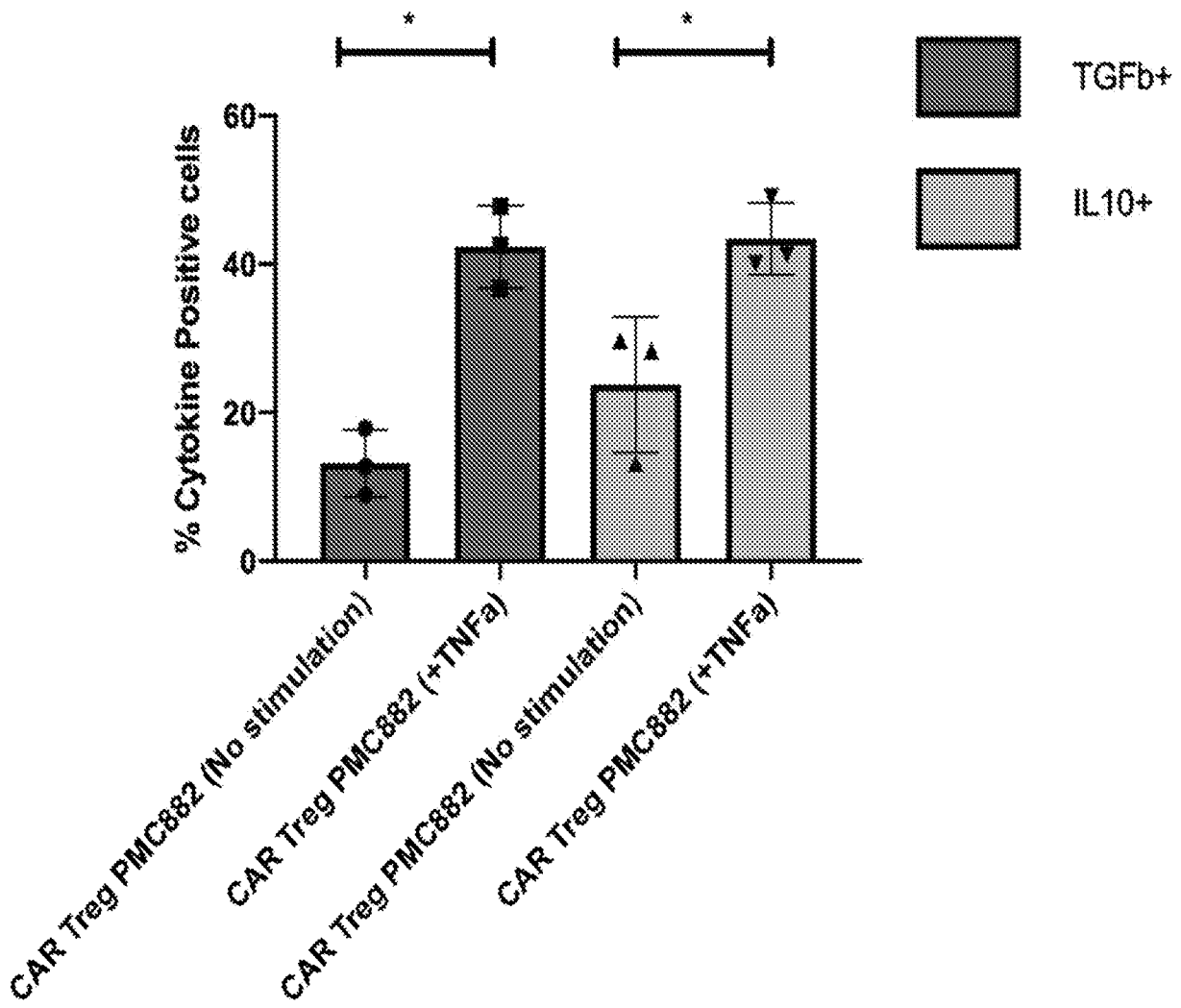

*
*
TGFb+
IL10+
% Cytokine Positive cells
60
40
20
0
CAR Treg PMC882 (No stimulation)
CAR Treg PMC882 (+TNFa)
CAR Treg PMC882 (No stimulation)
CAR Treg PMC882 (+TNFa)

Figure 6C

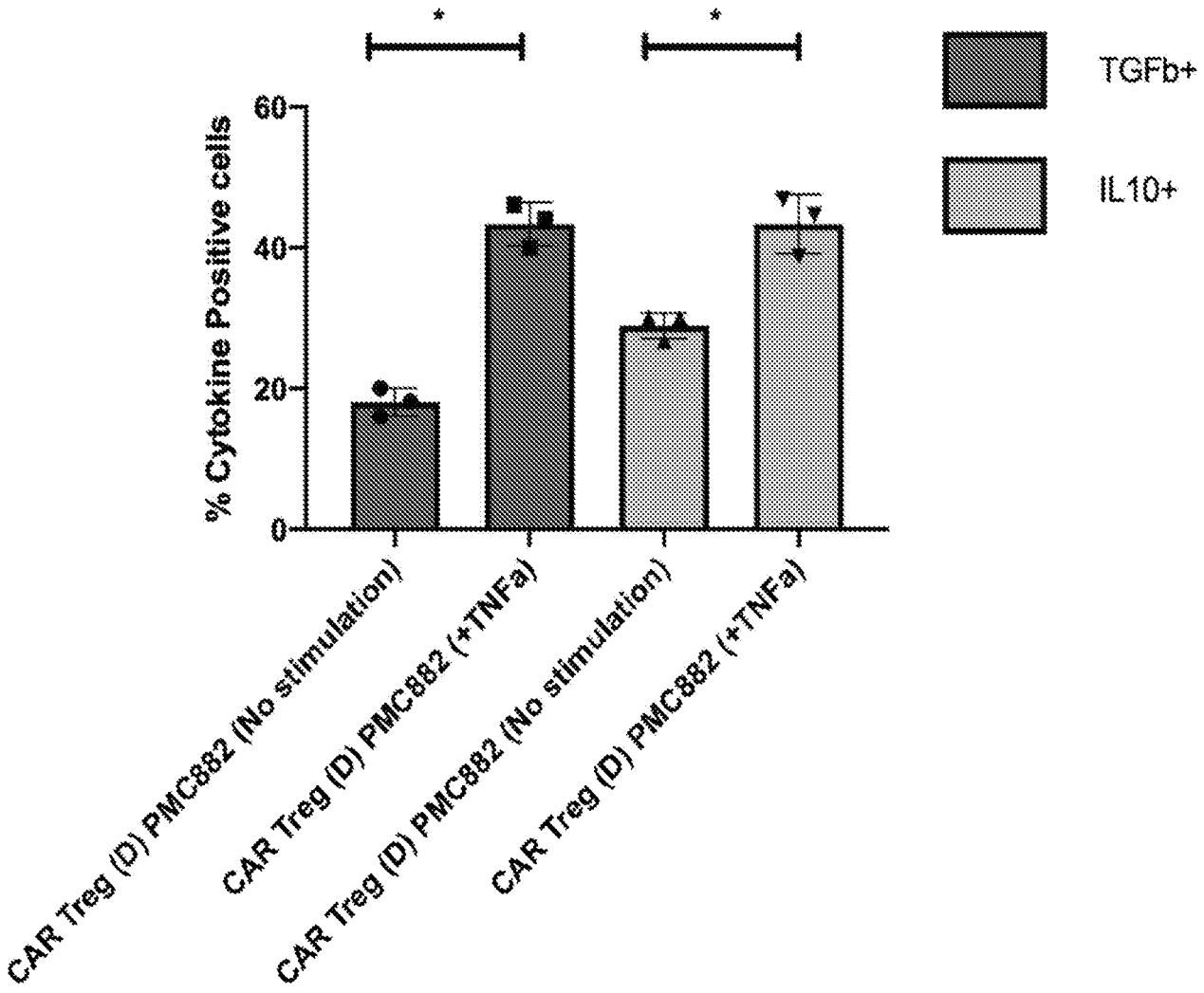

*
*
TGFb+
IL10+
% Cytokine Positive cells
60
40
20
0
CAR Treg (D) PMC882 (No stimulation)
CAR Treg (D) PMC882 (+TNFa)
CAR Treg (D) PMC882 (No stimulation)
CAR Treg (D) PMC882 (+TNFa)

Figure 7A

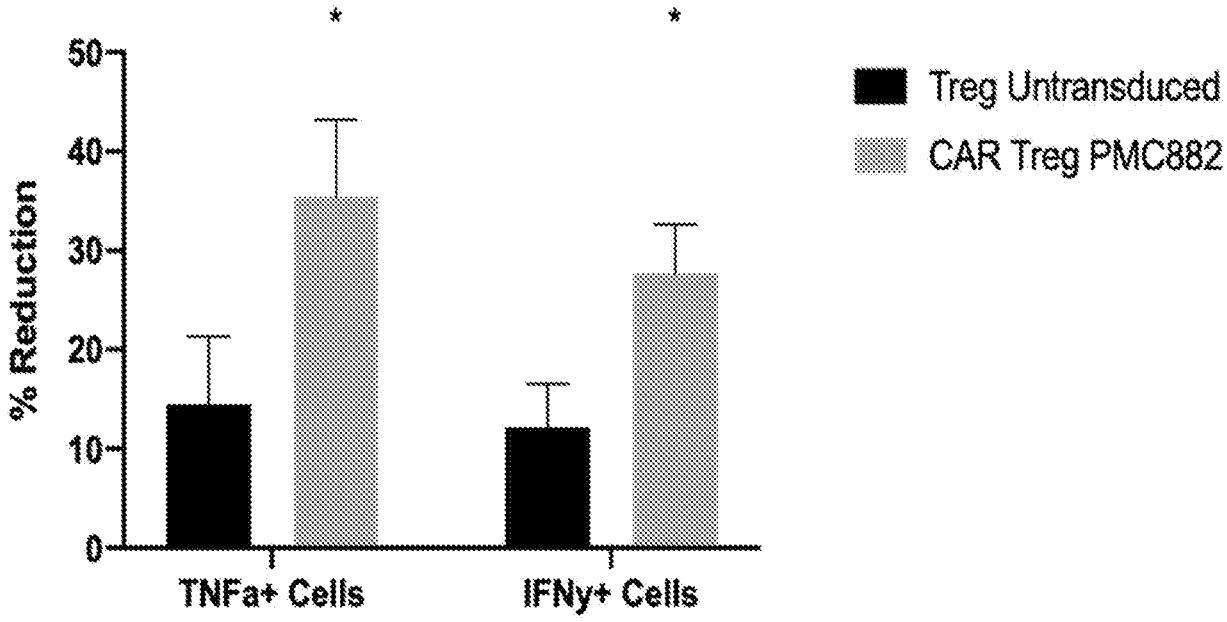


Figure 7B

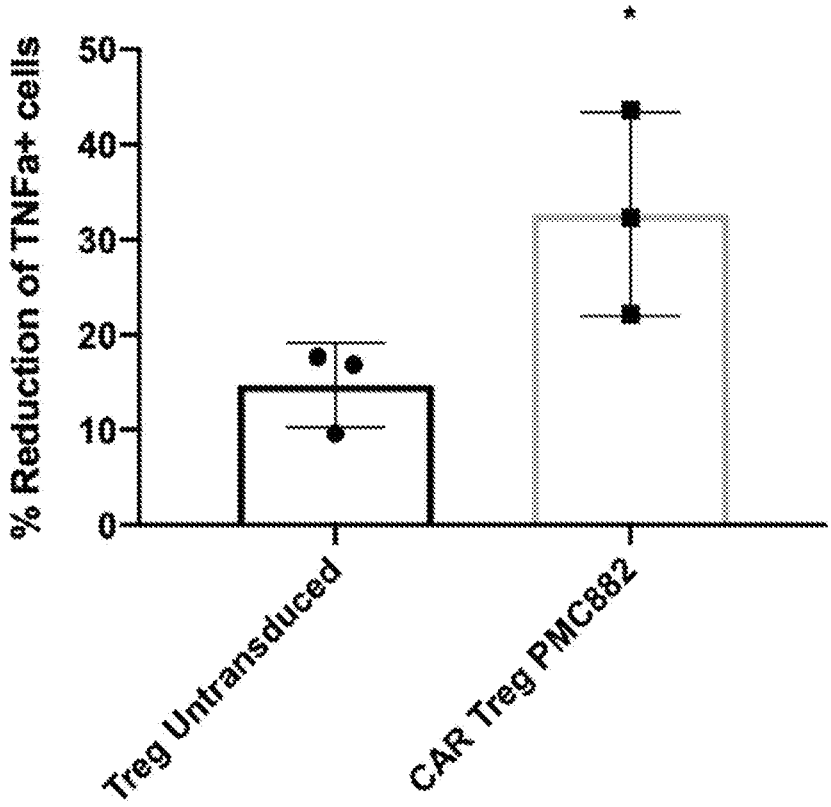


Figure 8A

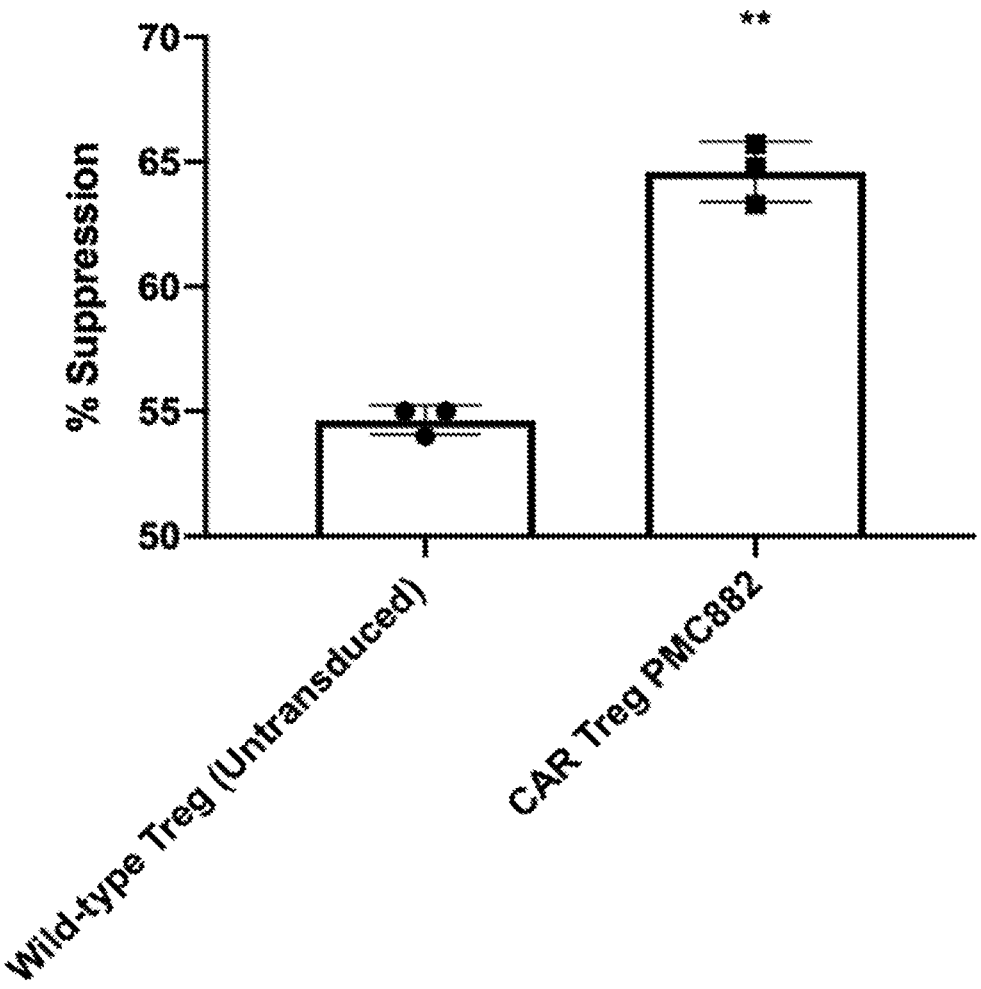

**
70
65
60
55
50
% Suppression
Wild-type Treg (Untransduced)
CAR Treg PMC882

Figure 8B

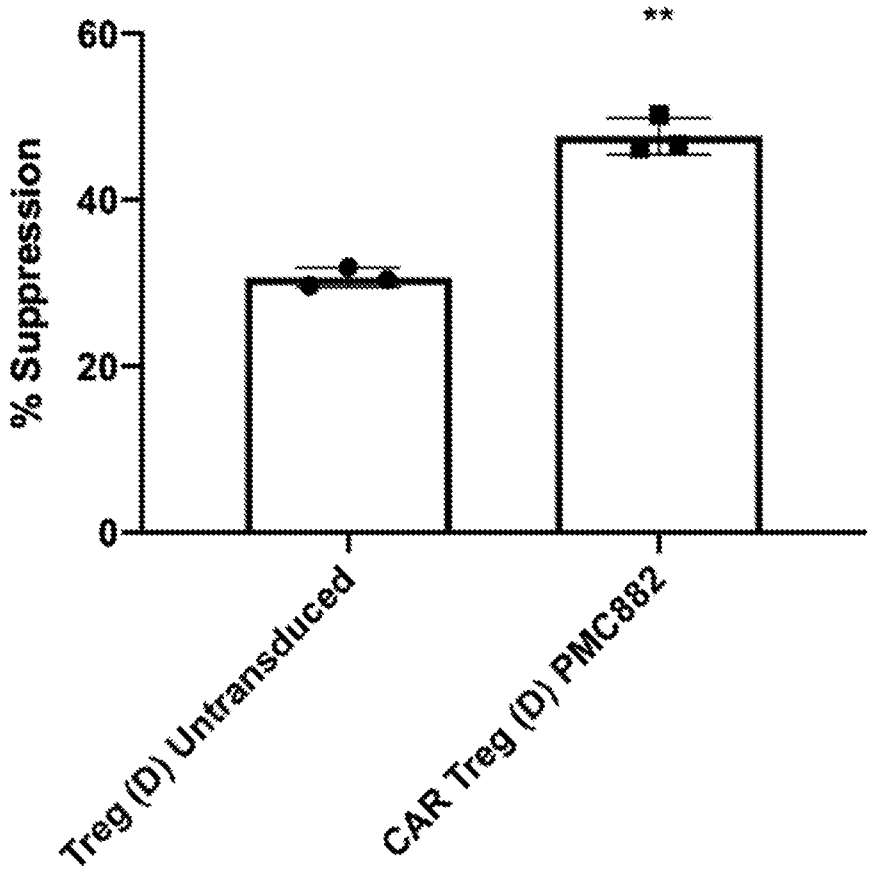


Figure 9A

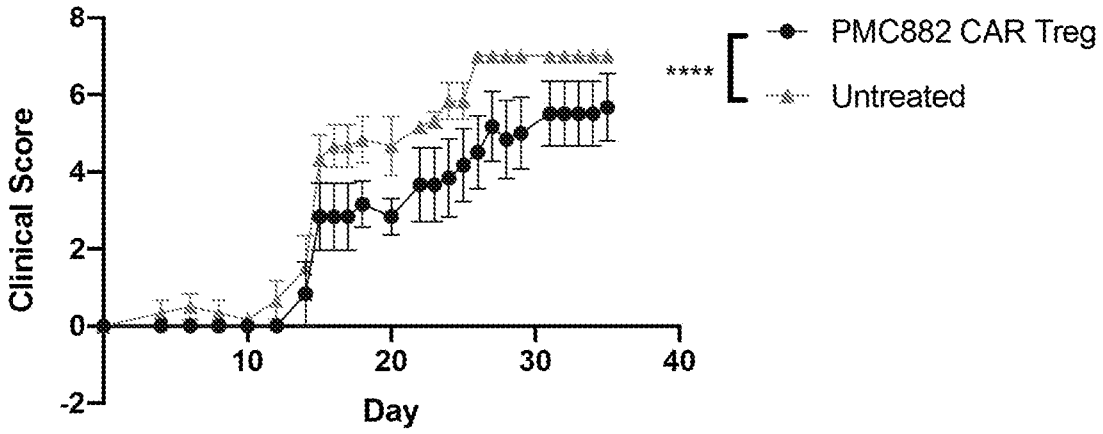


Figure 9B

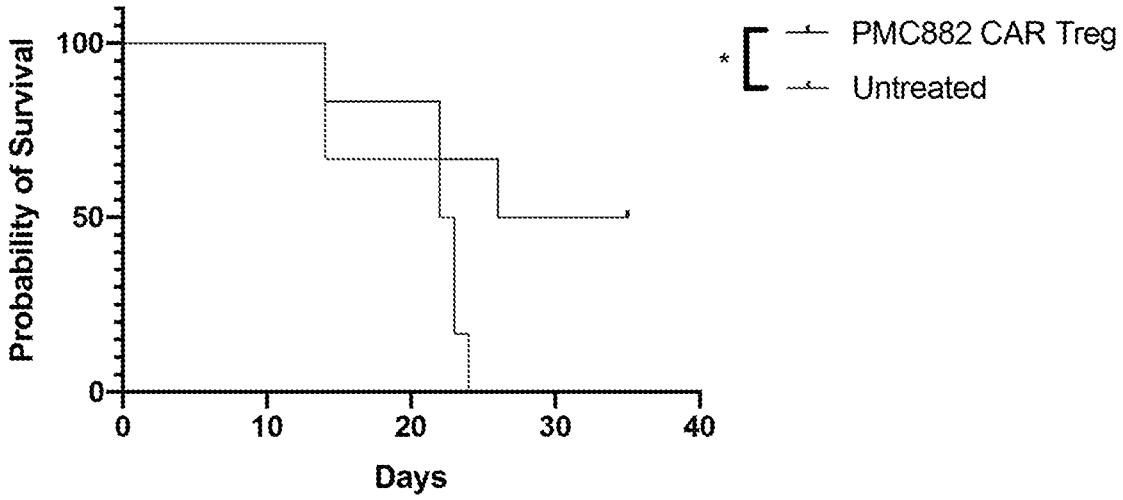


Figure 9C

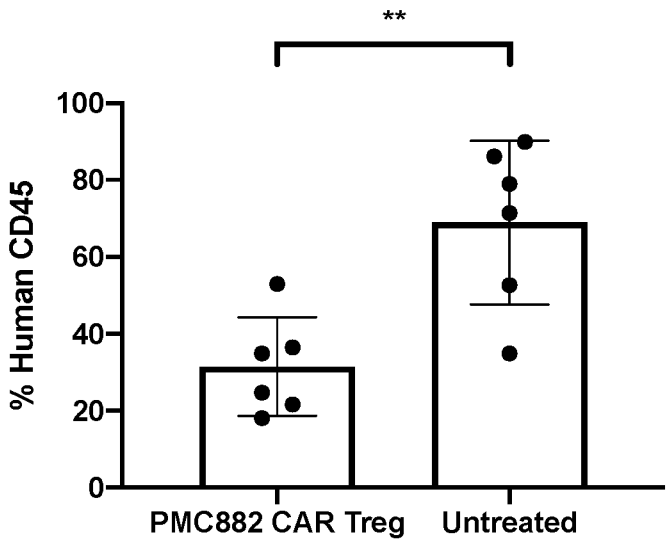


Figure 9D

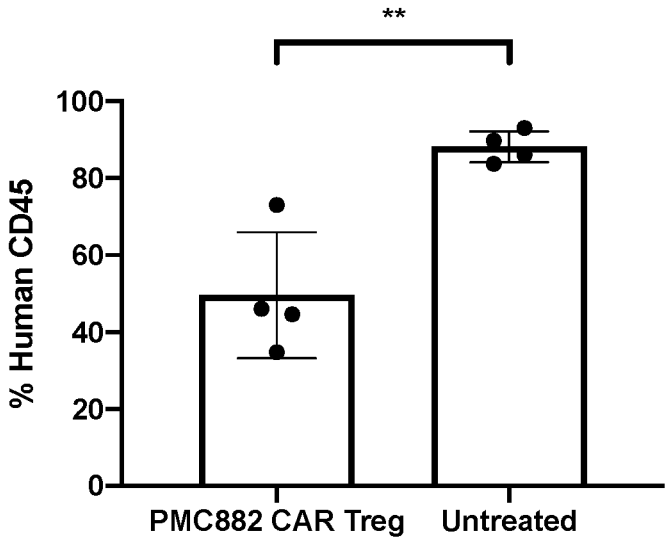


Figure 9E

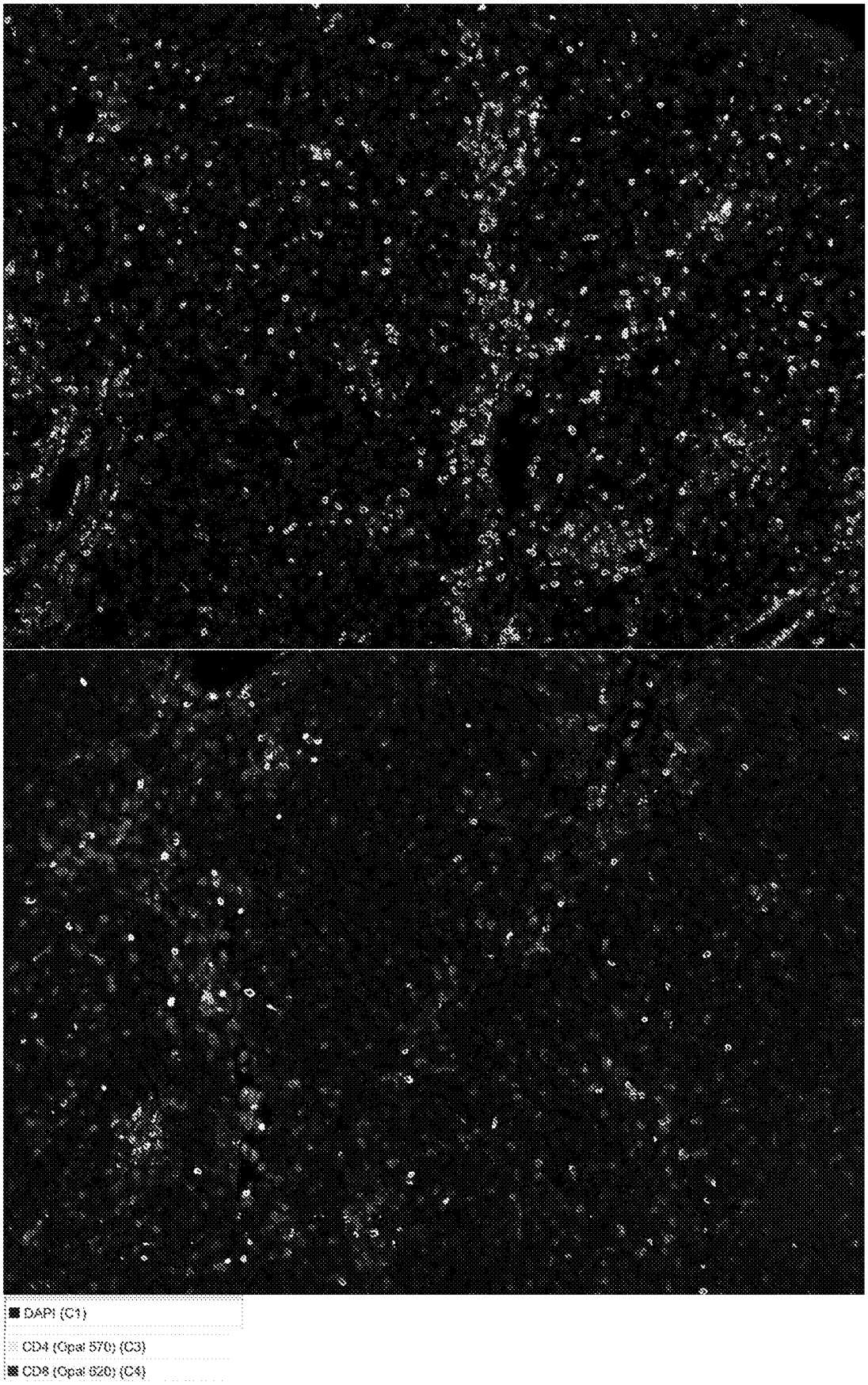
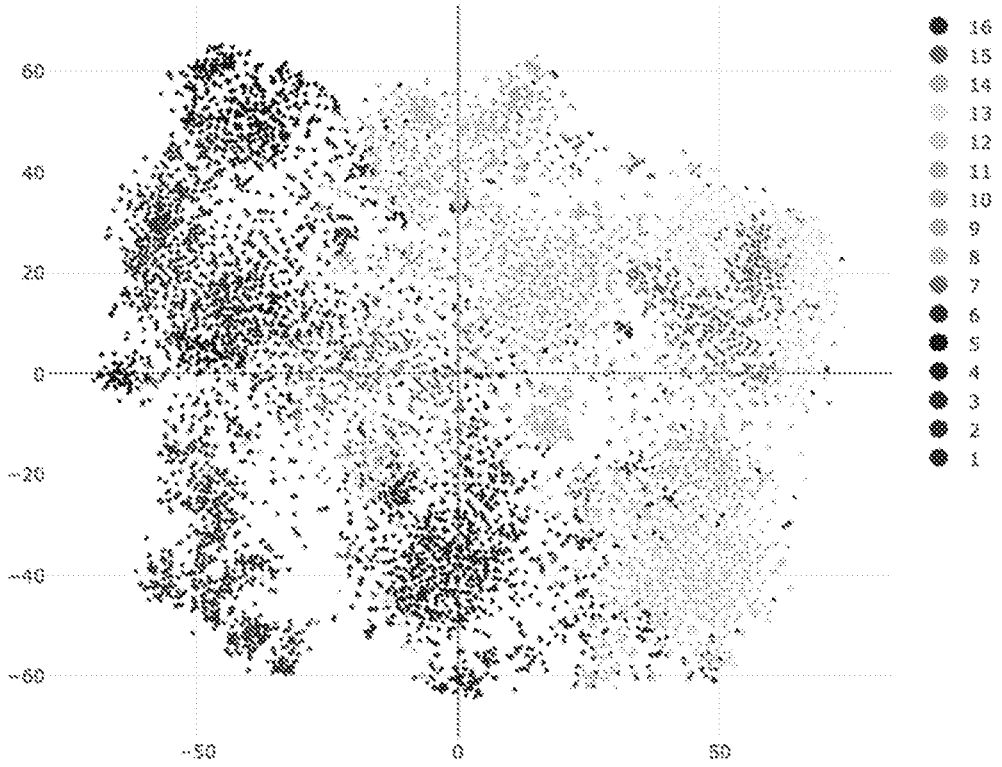


Figure 9F


60
40
20
0
-20
-40
-60
-50
0
50
16
15
14
13
12
11
10
9
8
7
6
5
4
3
2
1

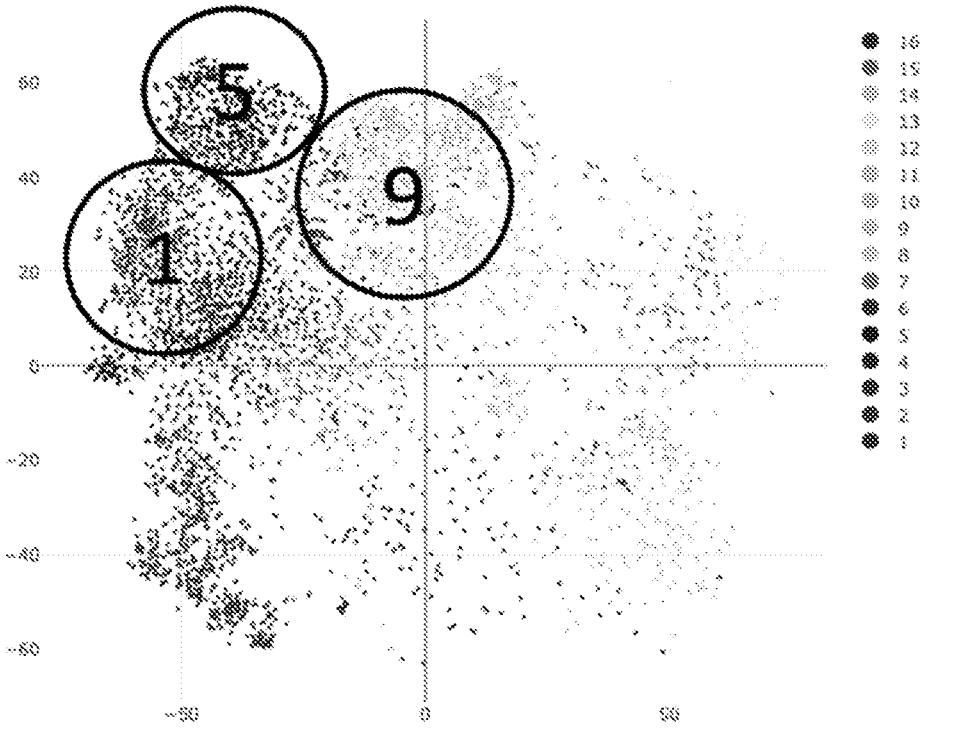

5
9
1
60
40
20
0
-20
-40
-60
-50
0
50
16
15
14
13
12
11
10
9
8
7
6
5
4
3
2
1

Figure 9F continued

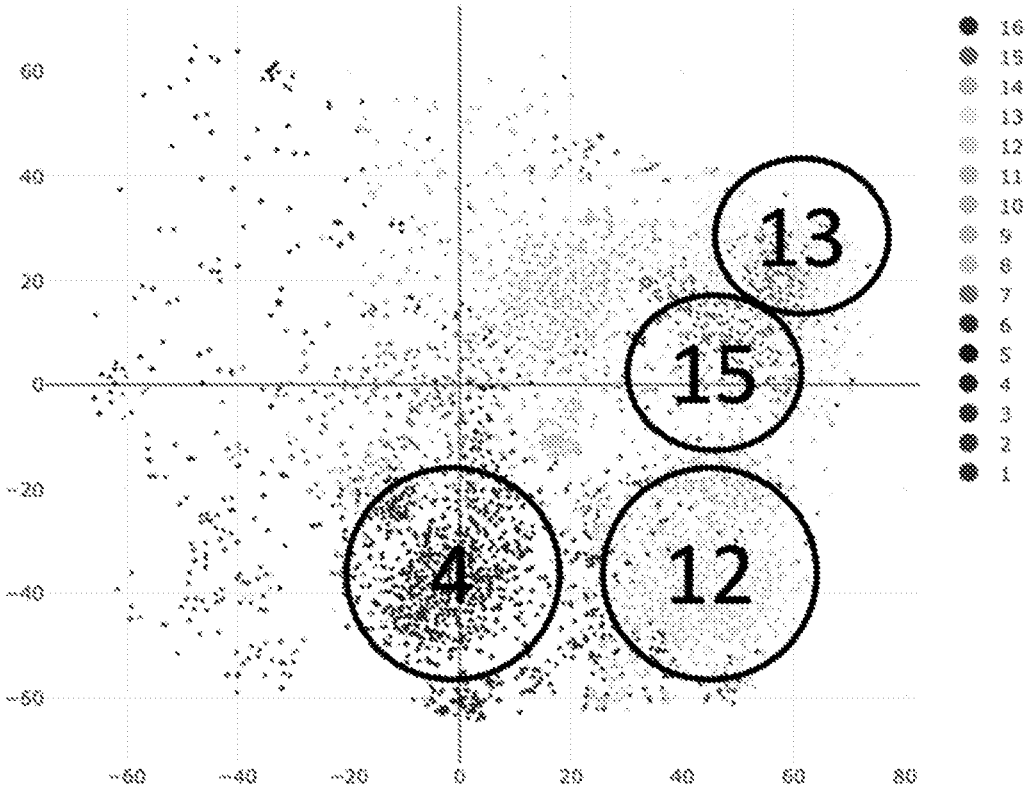


Figure 9G


1.00
0.75
0.50
0.25
0.00
Expression (linear median)
1.00
0.75
0.50
0.25
0.00
Expression (linear median)

Figure 10A

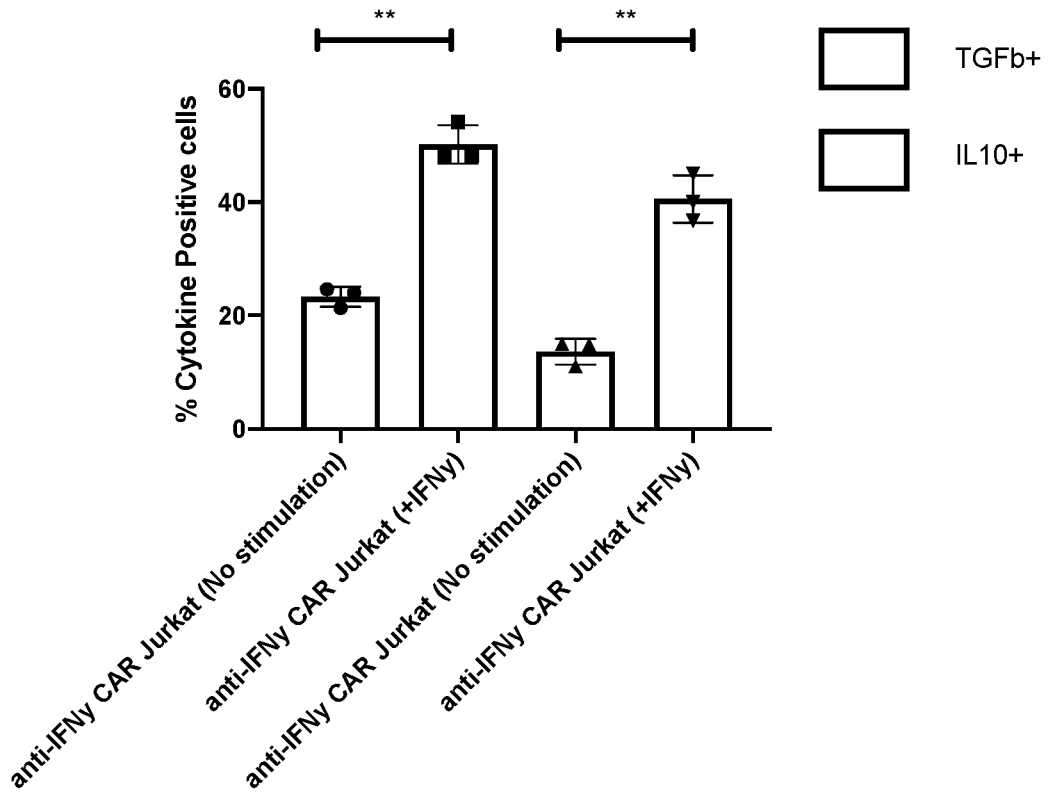


Figure 10B

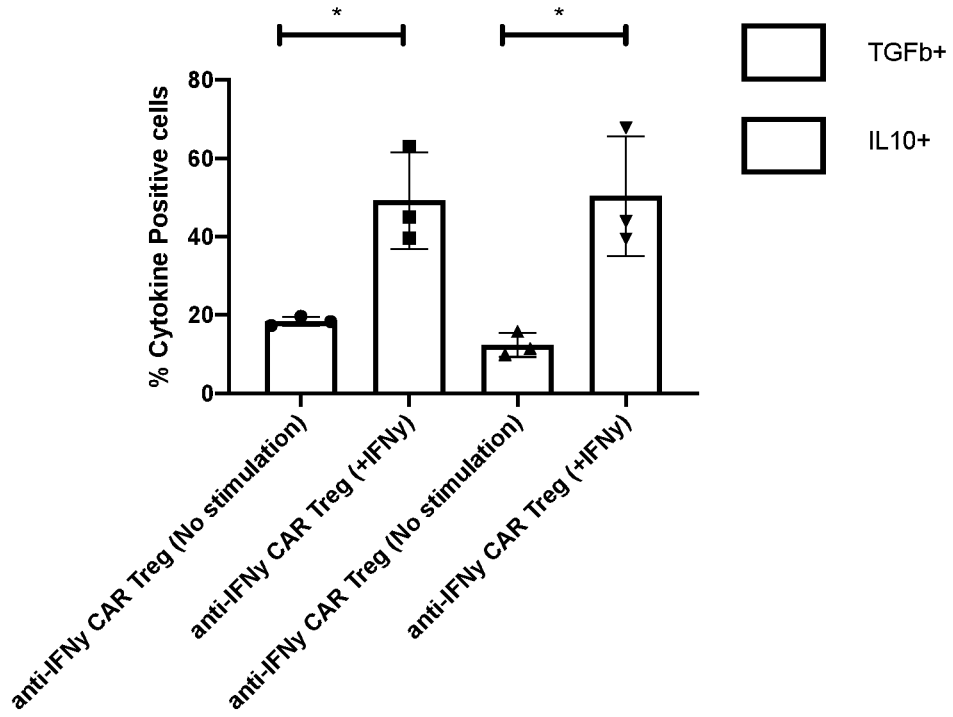


Figure 11

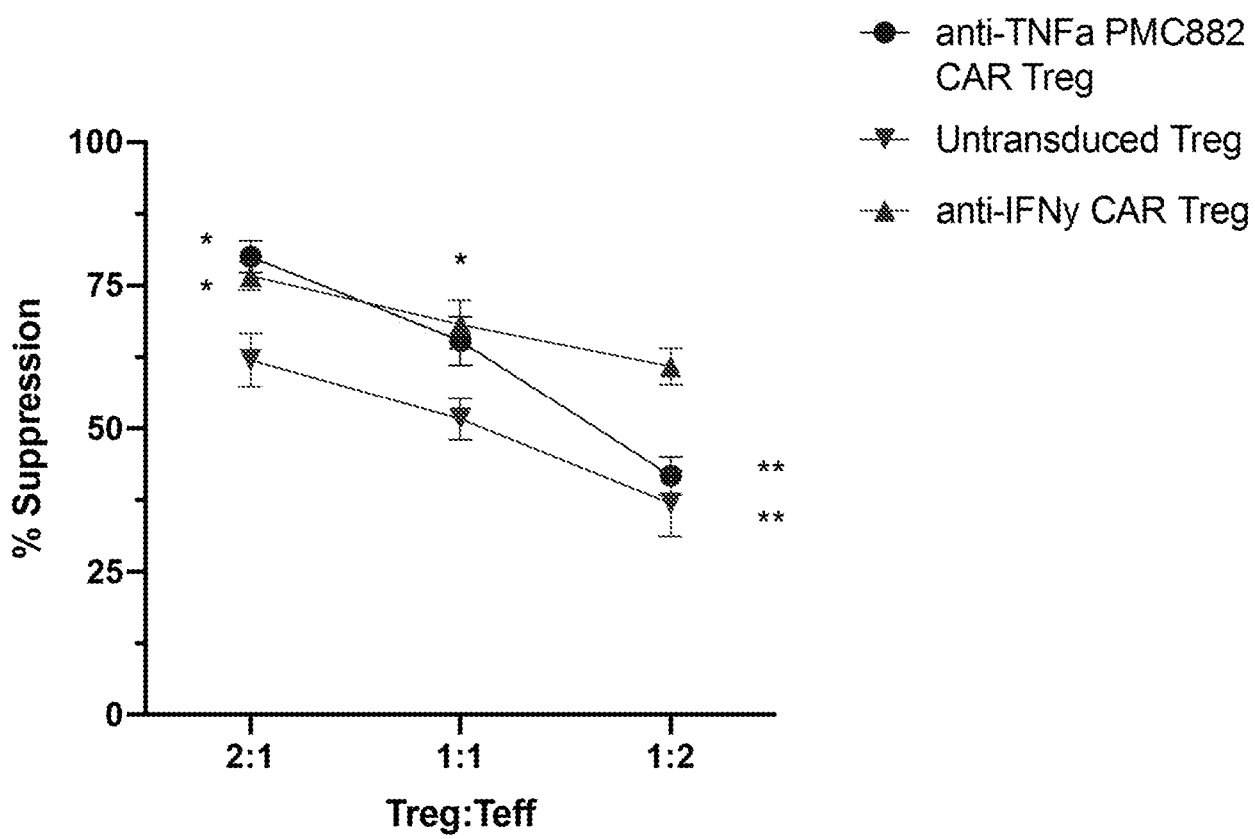

anti-TNFa PMC882 CAR Treg
Untransduced Treg
anti-IFNy CAR Treg
% Suppression
100
75
50
25
0
2:1
1:1
1:2
Treg:Teff
*
*
*
**
**

Figure 12A

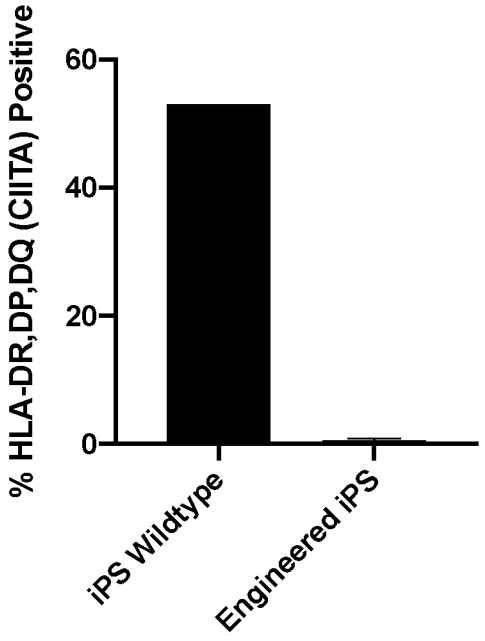


Figure 12B

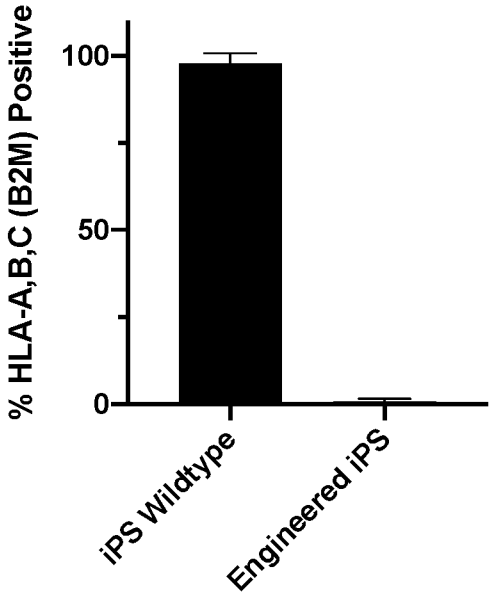


Figure 12C

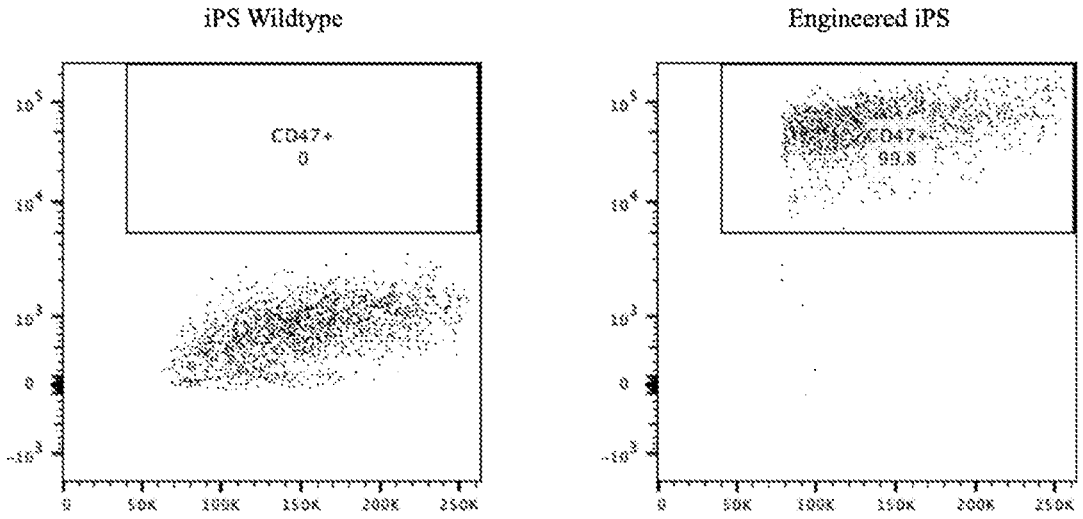


Figure 12D

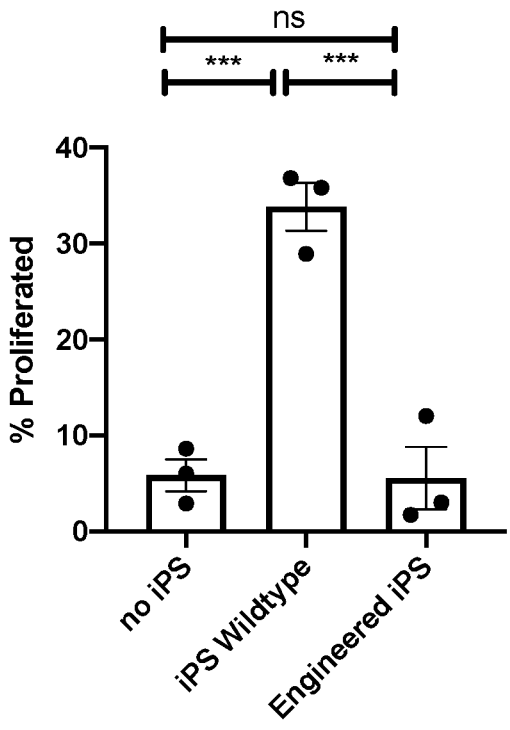


Figure 13A

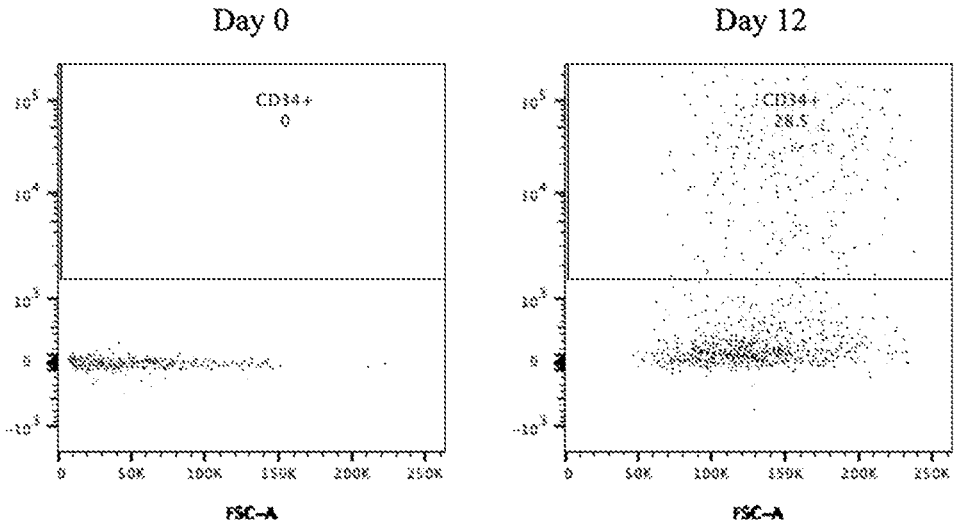


Figure 13B

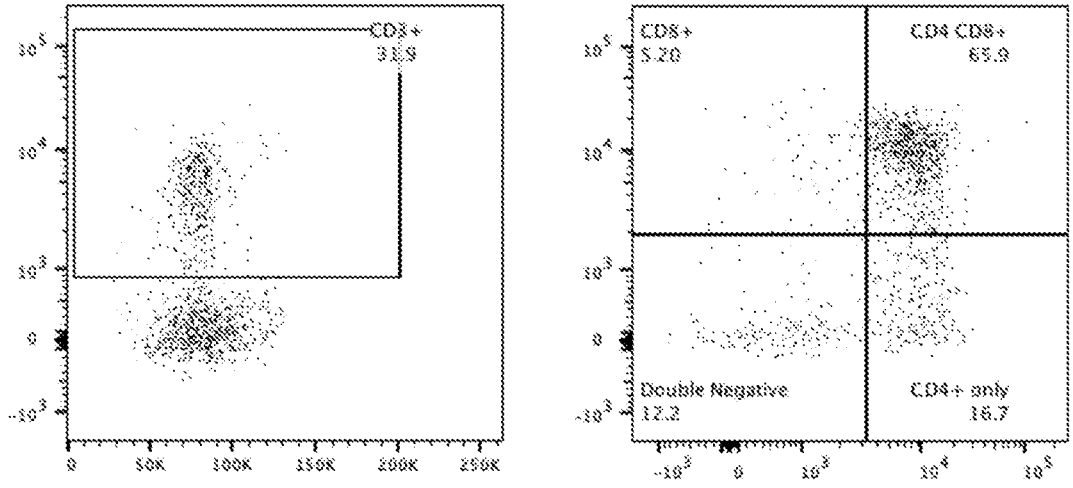


Figure 13C

