

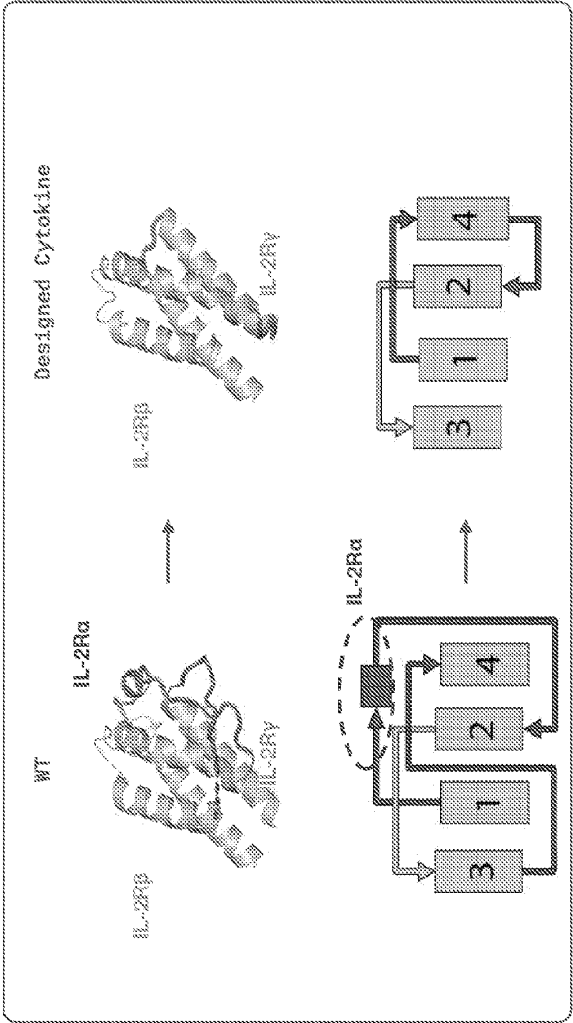


FIG. 1A

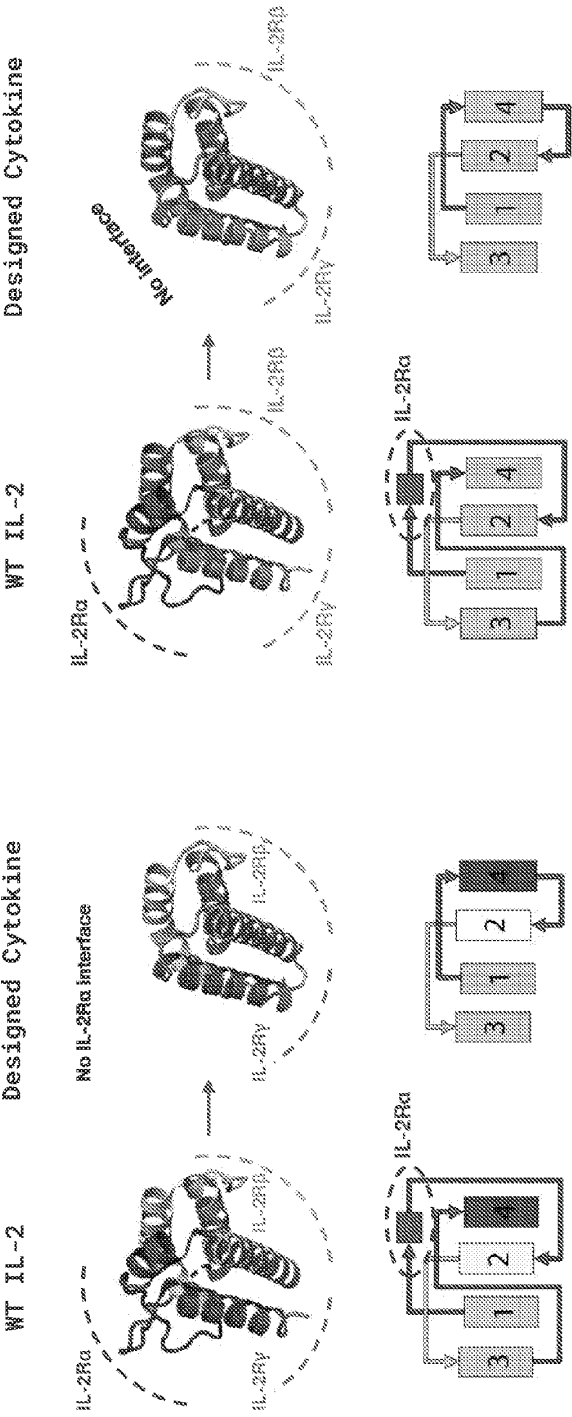


FIG. 1B

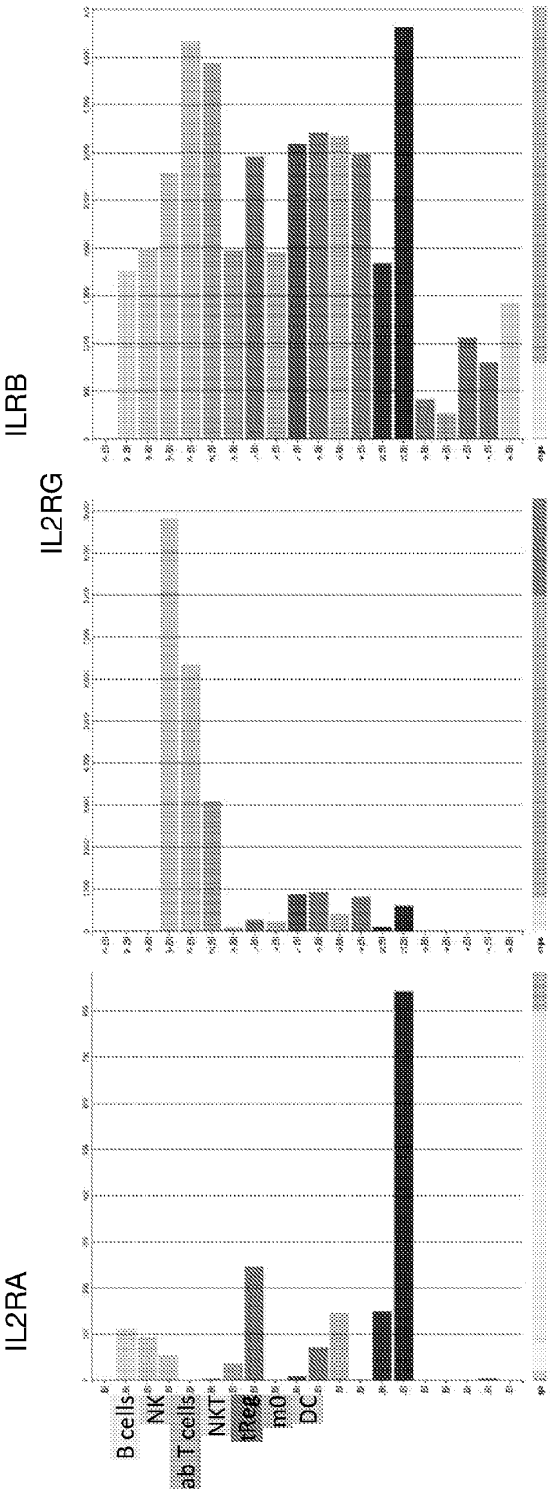


FIG. 2

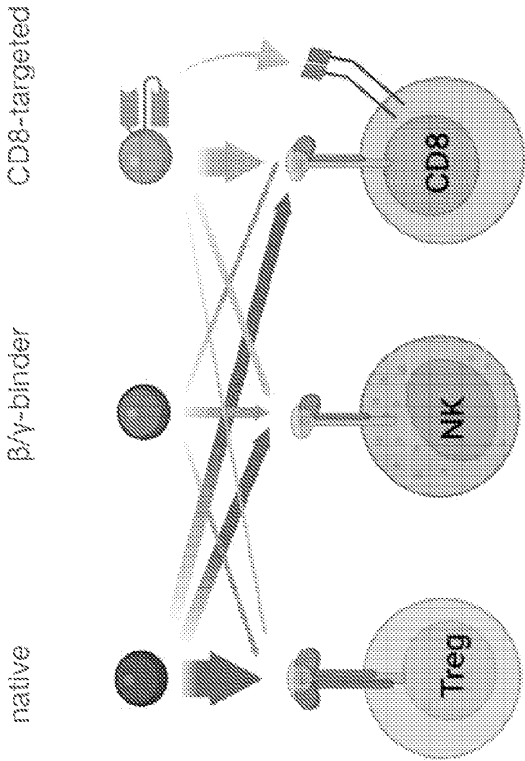


FIG. 3A

Use of Designed Cytokine in a variety of cell types and for different applications

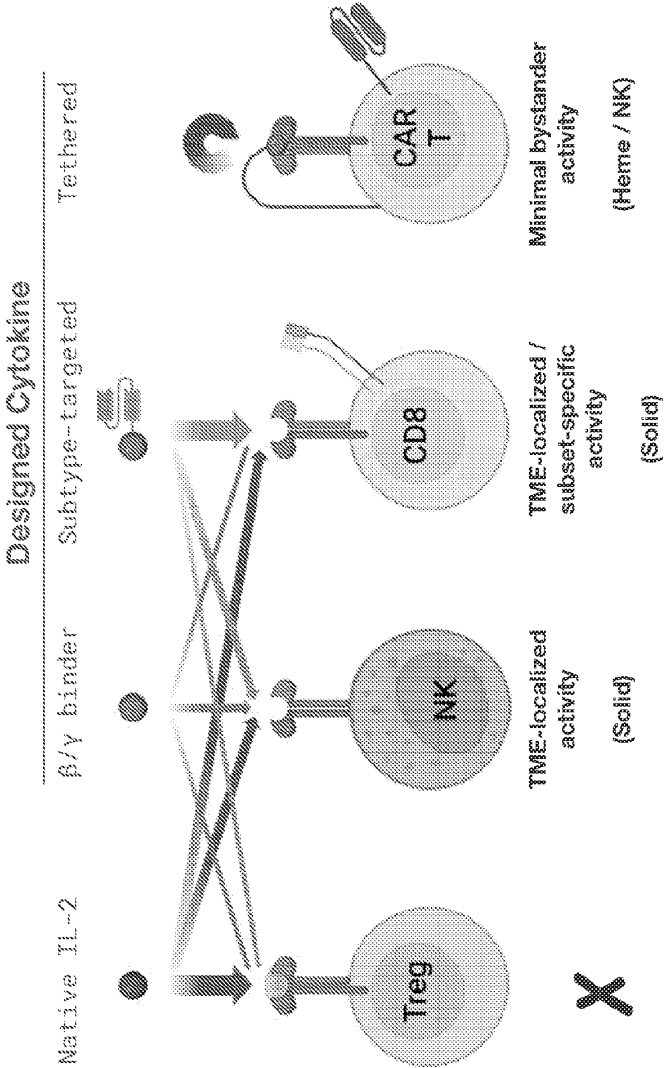


FIG. 3B

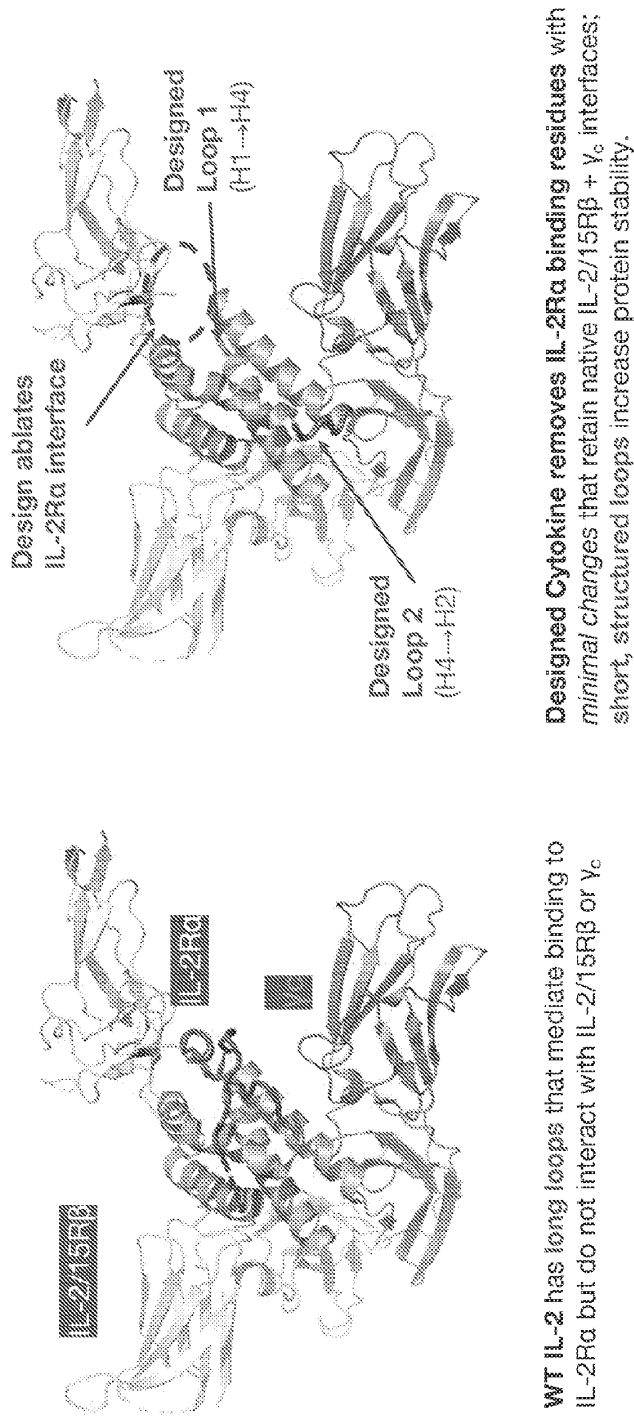


FIG. 4

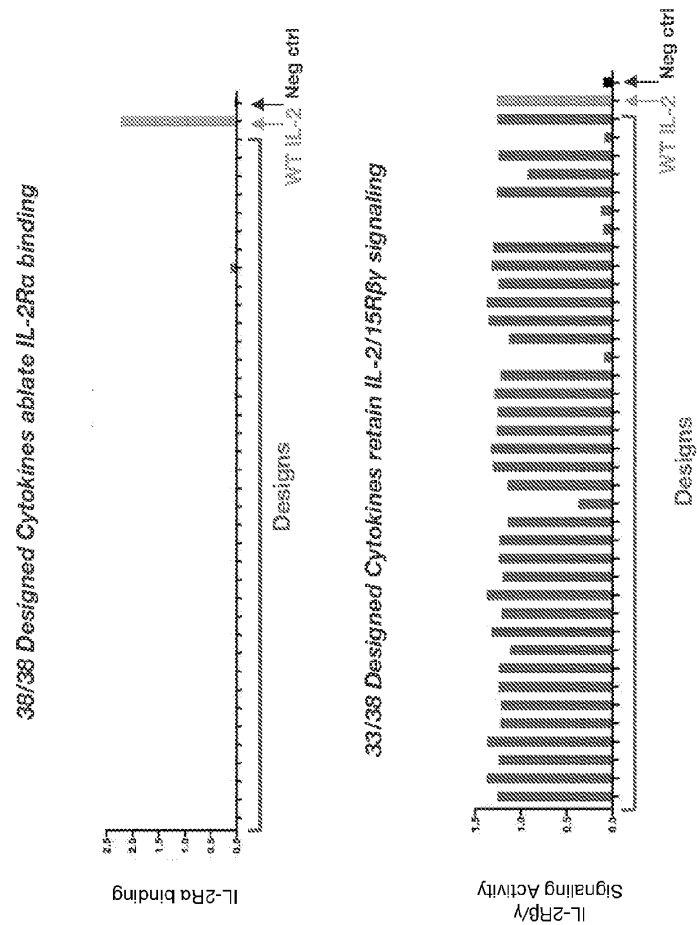
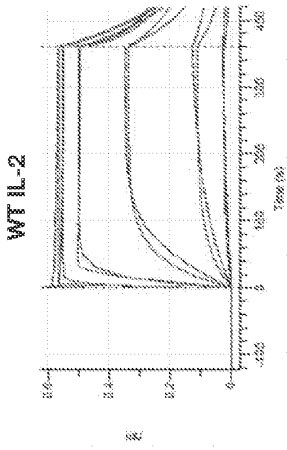
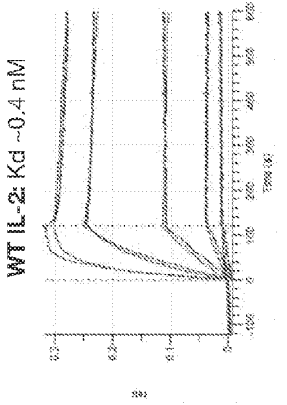


FIG. 5A

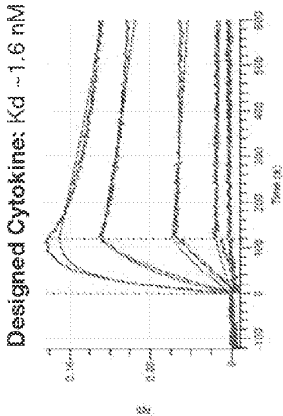
Designed Cytokine shows no detectable binding to IL-2Ra



Designed Cytokine binds to IL-2/15Rβγ with very low nanomolar affinity



Designed Cytokine



0.9 nM to 3000 nM cytokine tested

FIG. 5B

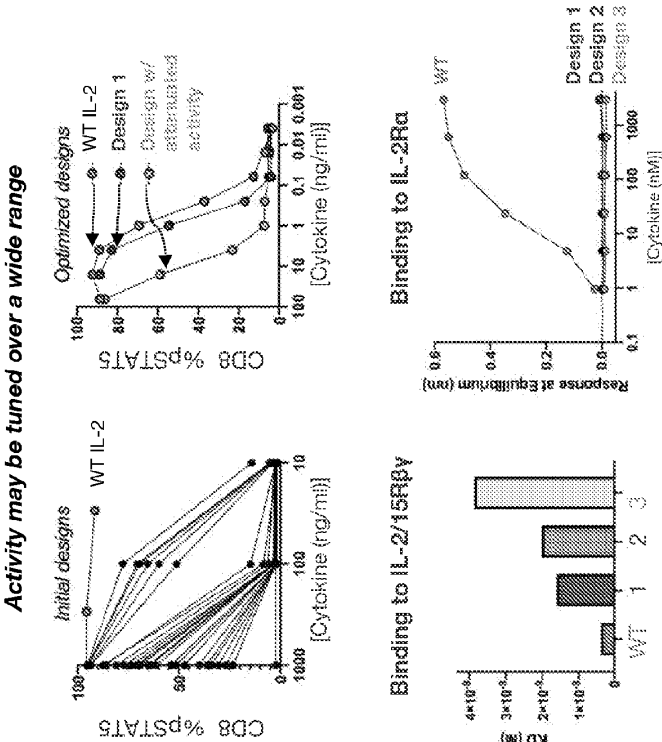
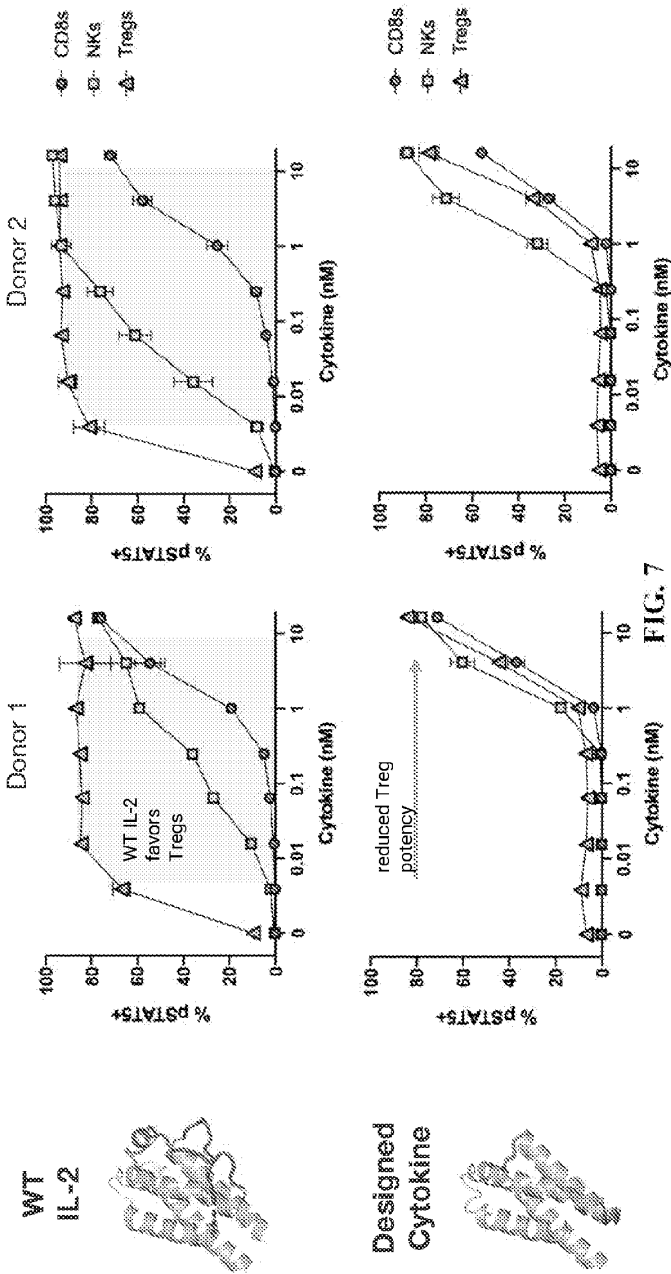


FIG. 6

Designed Cytokine eliminates Treg preference, preserving NK and CD8 activity



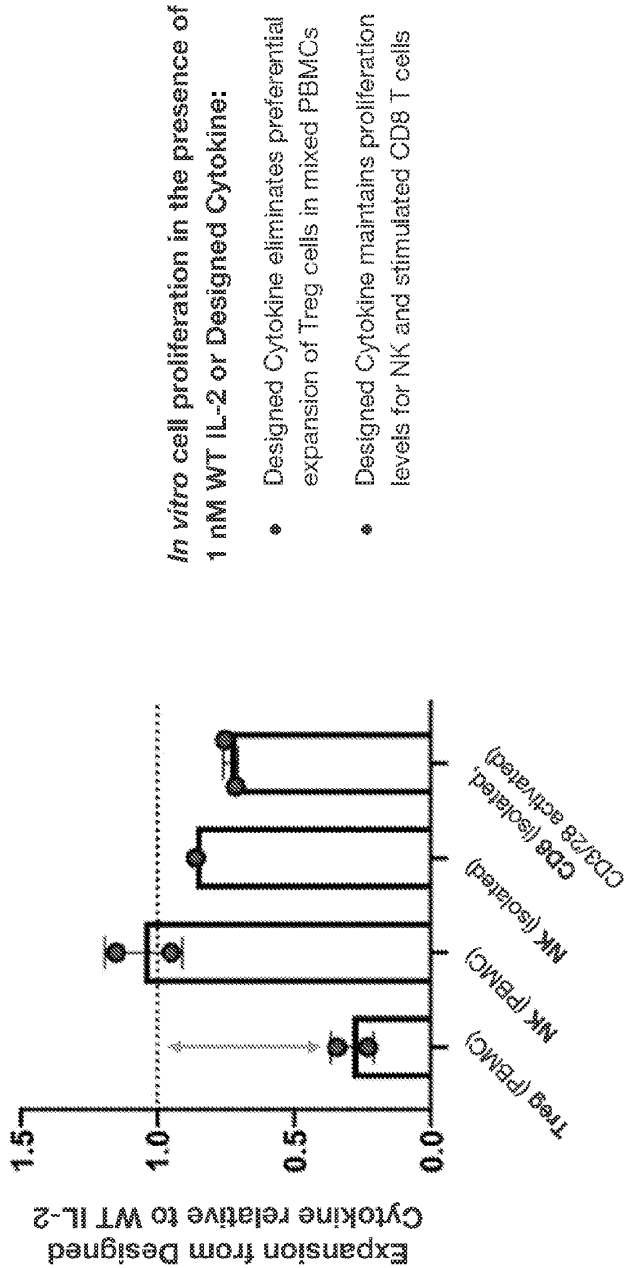


FIG. 8

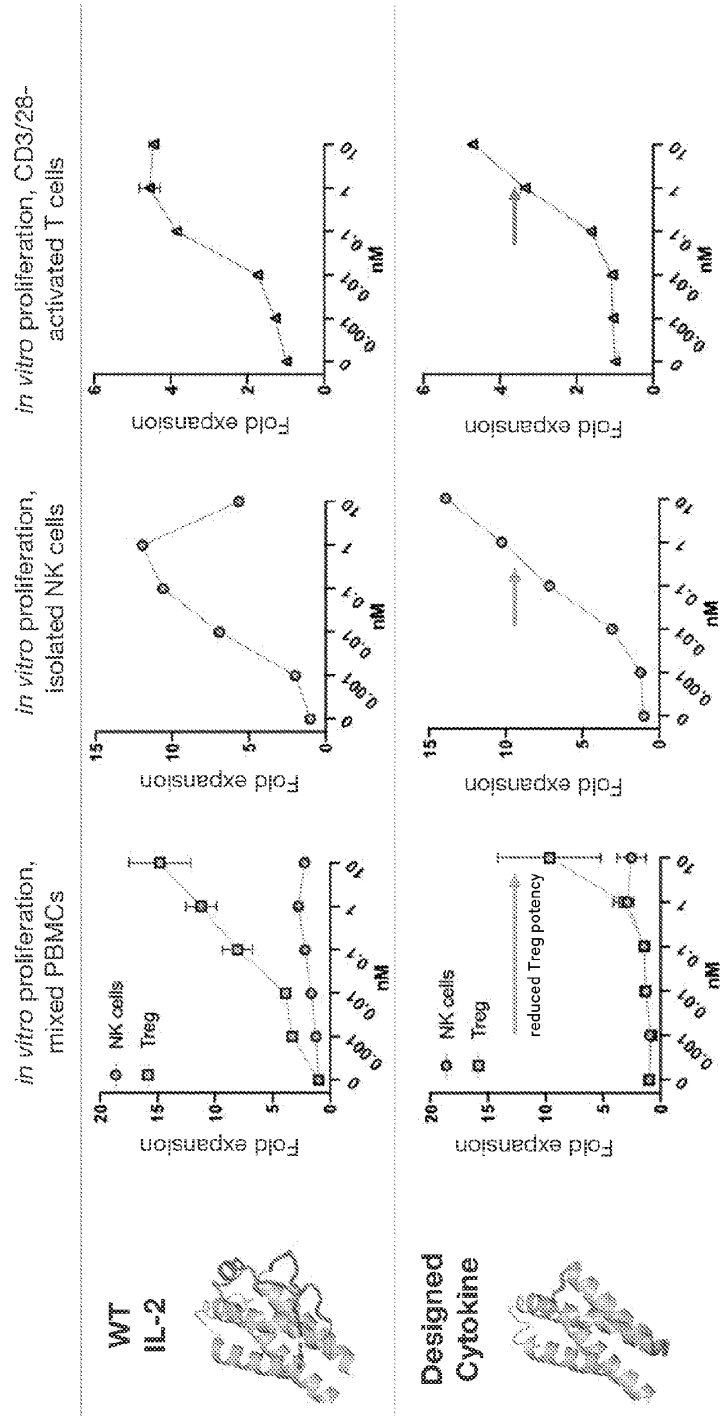


FIG. 9

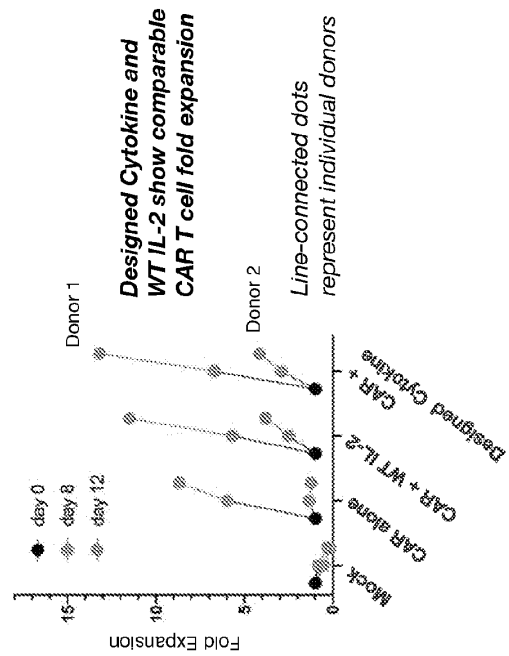
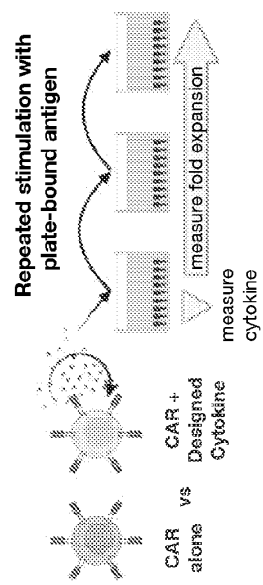
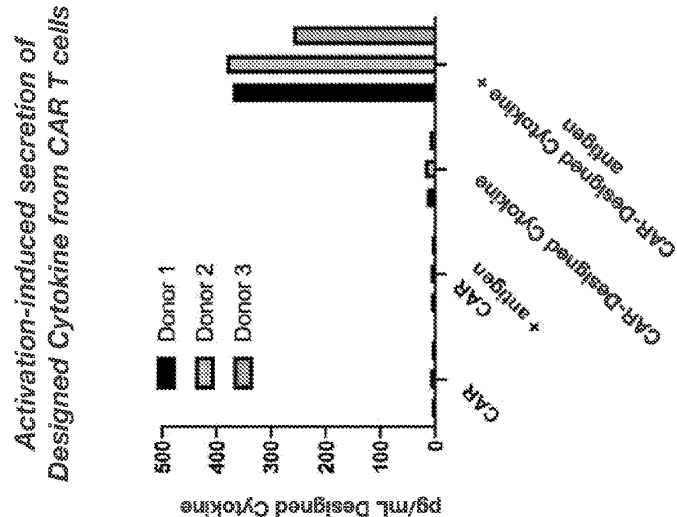


FIG. 10





Engineering desired promoters for Designed Cytokine:

- “Off” state shows no T cell expansion in cytokine free media
- “On” state levels increase expansion relative to no cytokine condition
- Maintain circulating levels below thresholds known to be safe in blood
- Evaluate antigen-induced production in response to stimulation using plate-bound antigen & target cells
- Measure levels of secreted Designed Cytokine
- Measure levels of mRNA and copy number using ddPCR

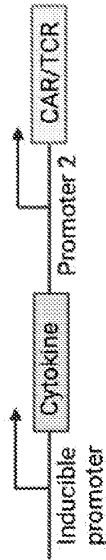


FIG. 11

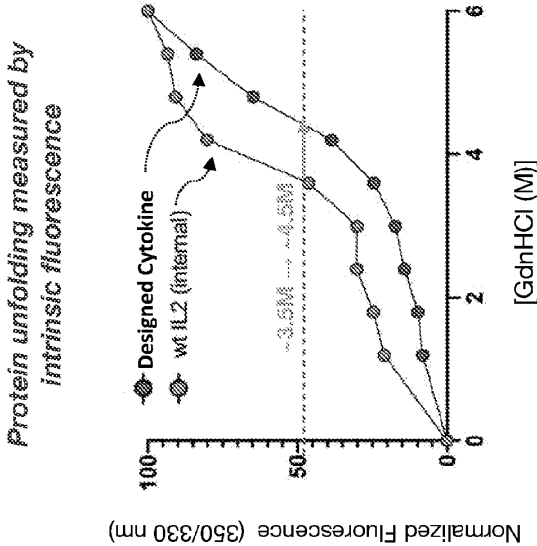
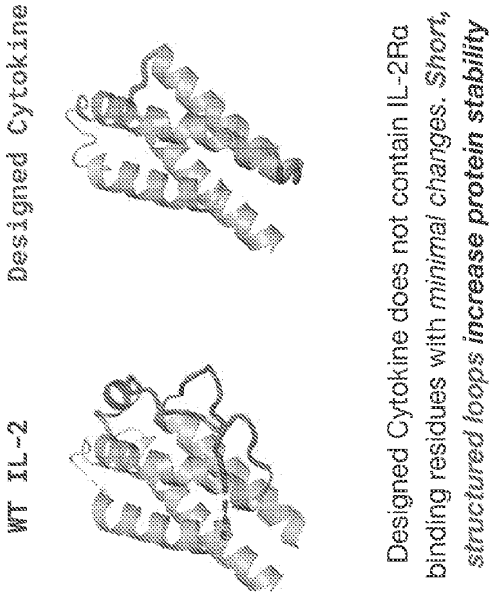


FIG. 12



Designed Cytokine demonstrates IL15-like effects on NK cells in vitro

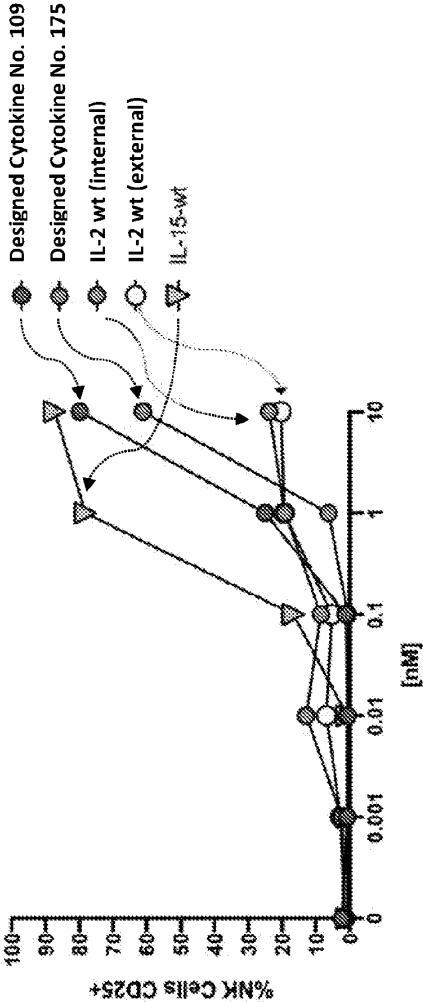


FIG. 13

TME-targeting of Designed Cytokines having Targeting Moieties

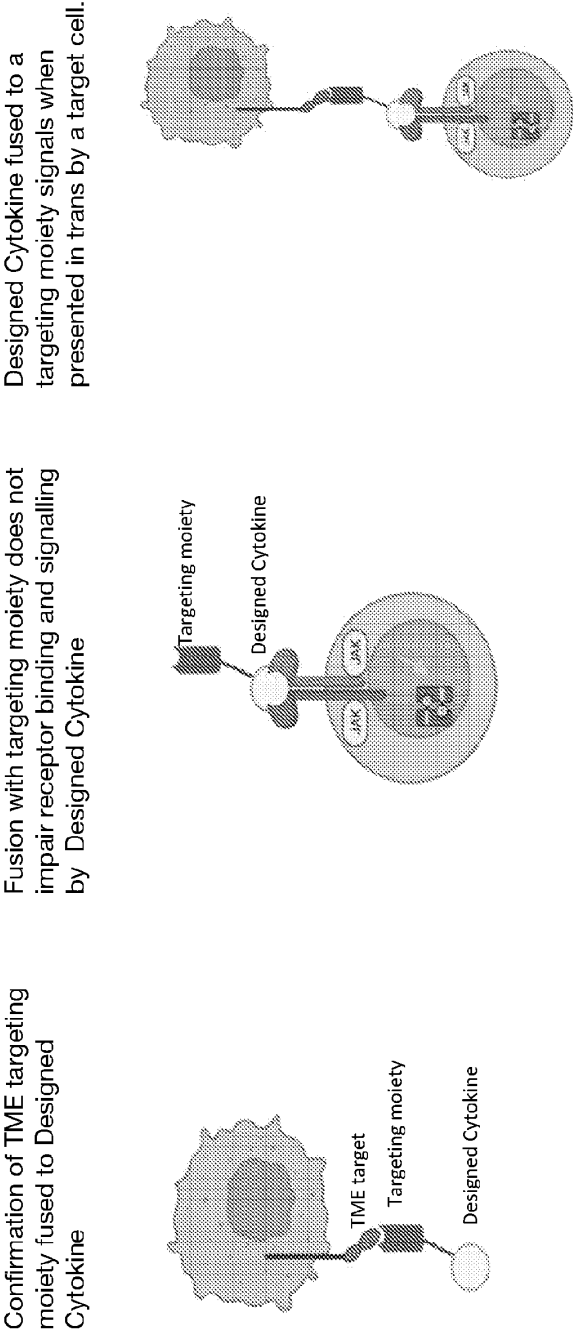


FIG. 14A

TME-targeting of Designed Cytokines

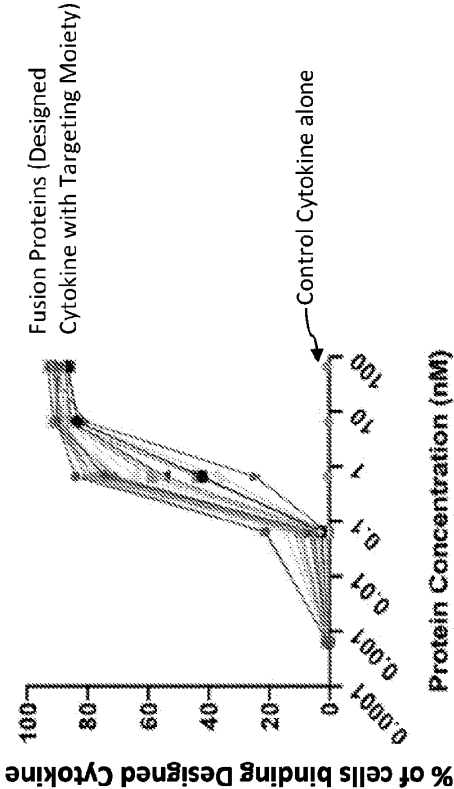


FIG. 14B

TME-targeting of Designed Cytokines

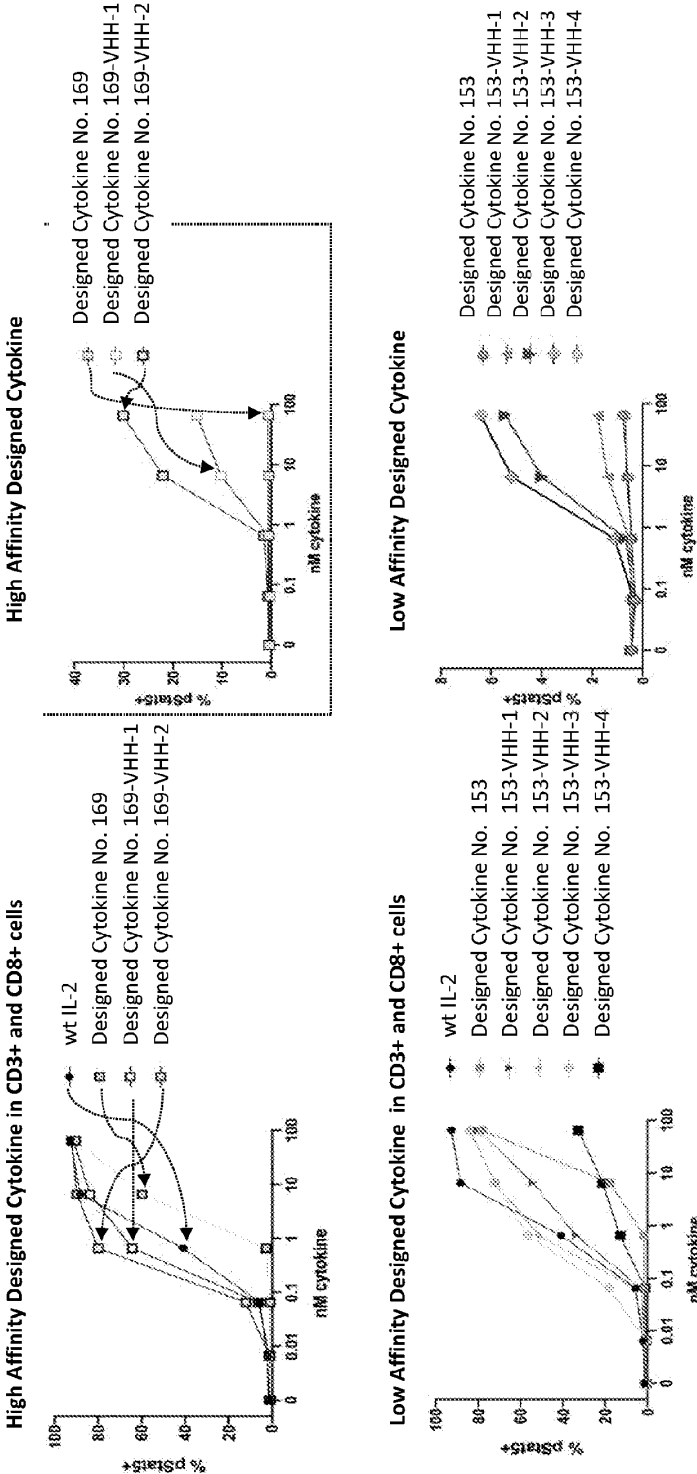


FIG. 14C

Designed Cytokine No. 201 shows no detectable binding to IL2Ra
WT IL-2 binds with nanomolar affinity

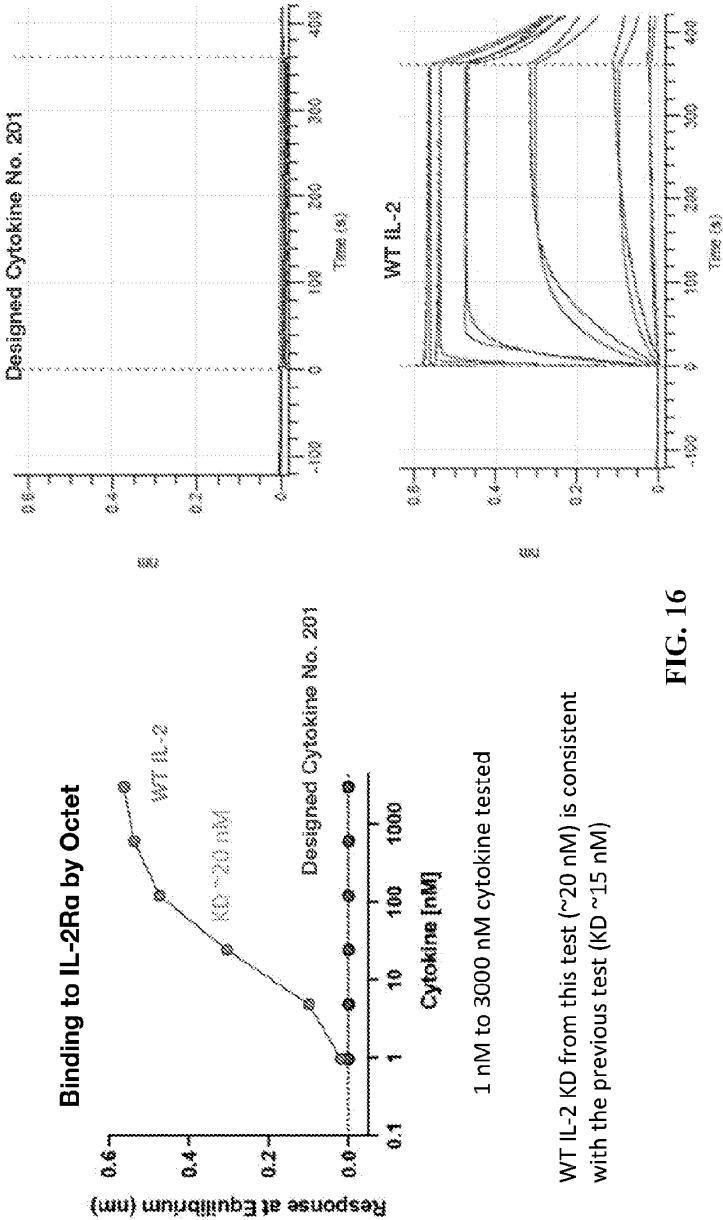


FIG. 16

1 nM to 3000 nM cytokine tested

WT IL-2 KD from this test (~20 nM) is consistent with the previous test (KD ~15 nM)

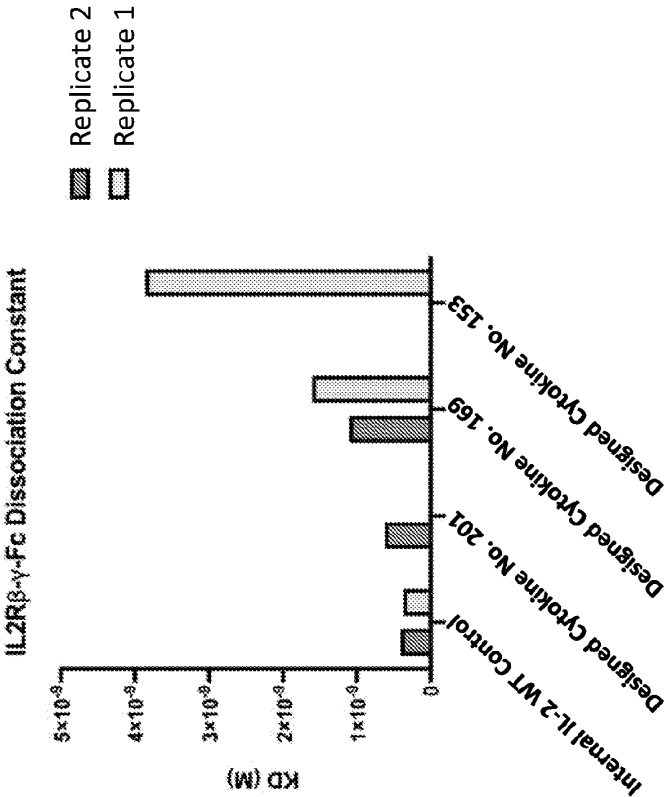
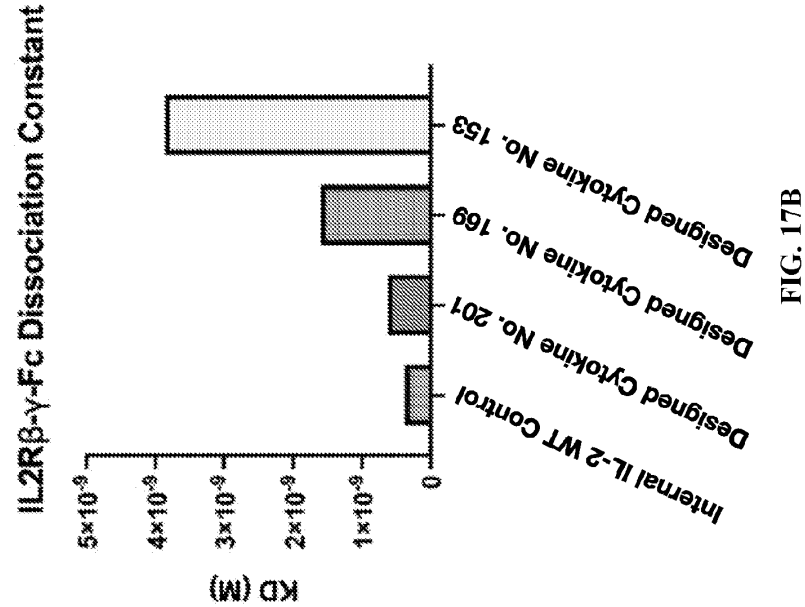
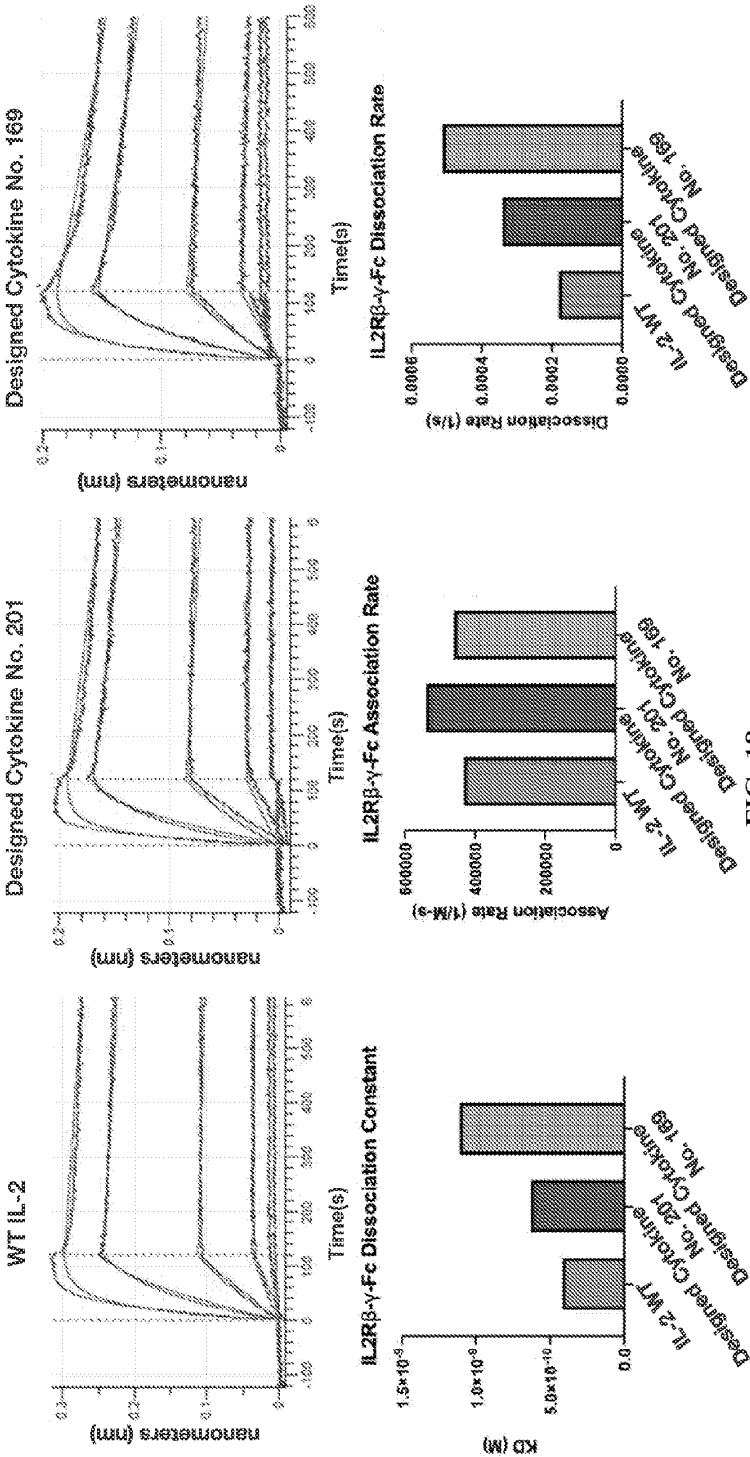


FIG. 17A



Binding affinity (by Octet) to IL-2Rβγ



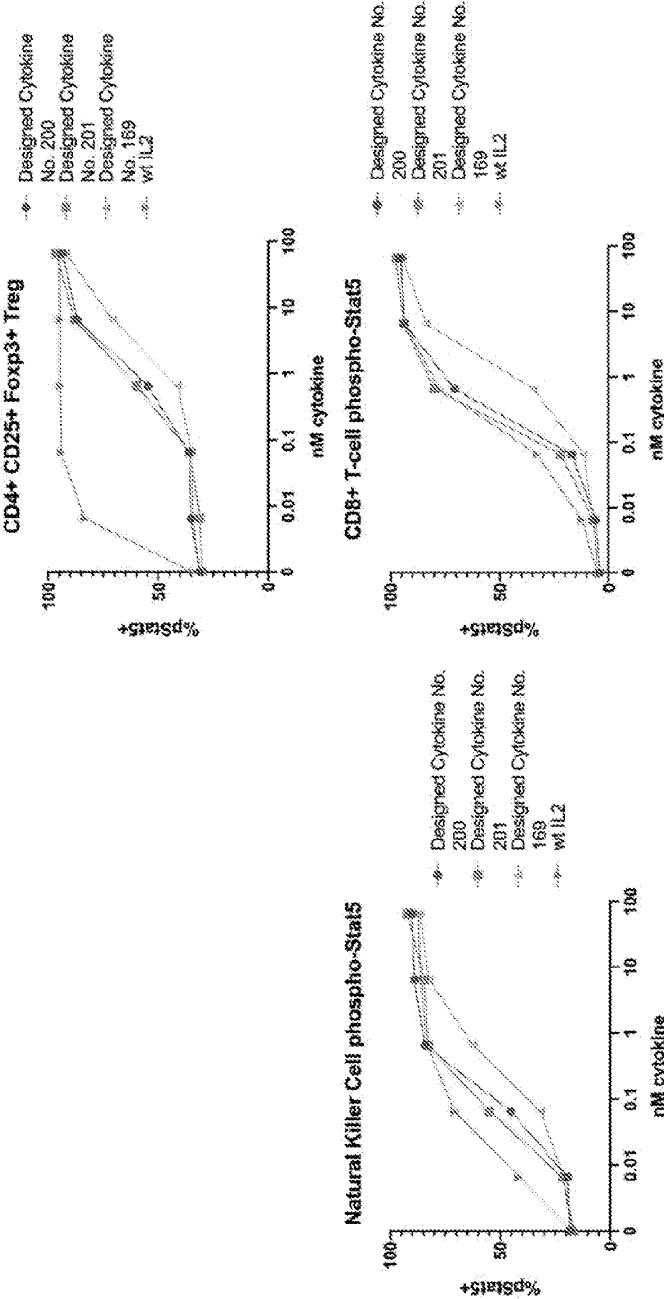


FIG. 19

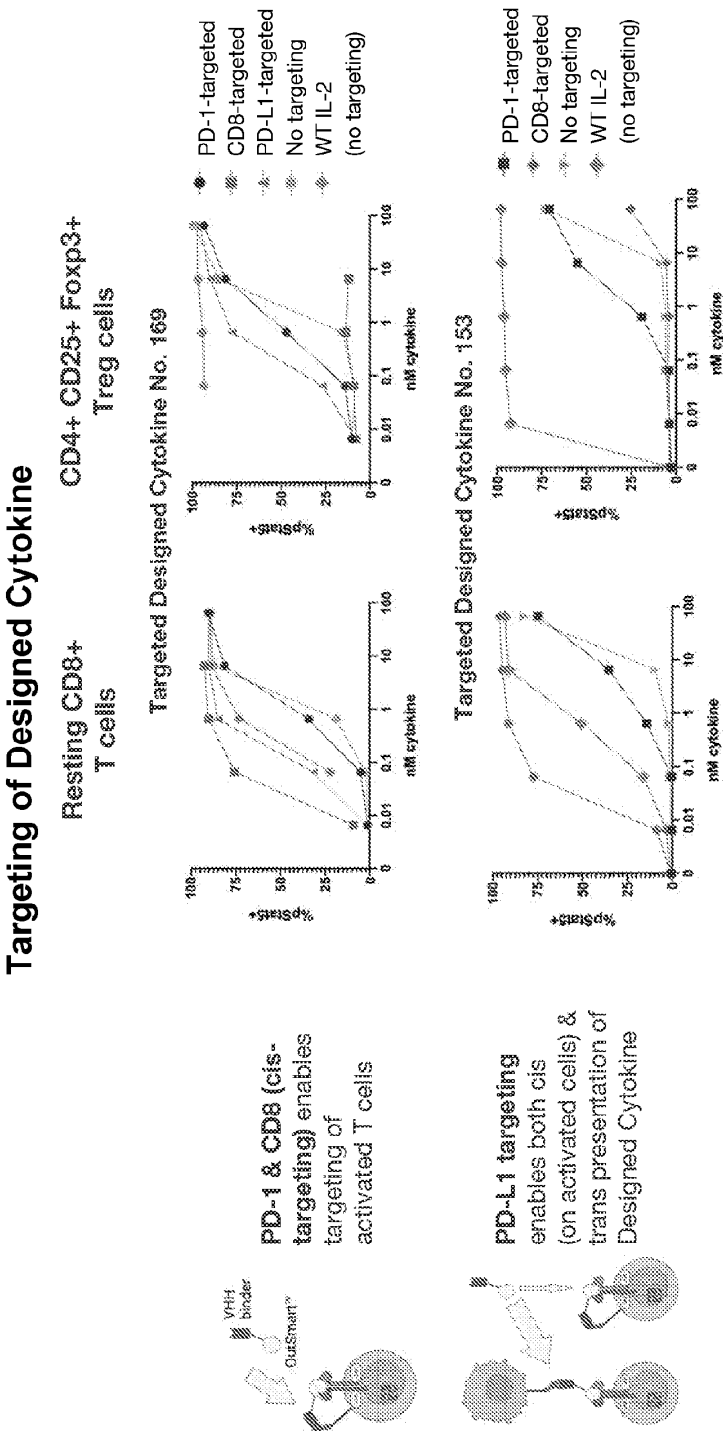


FIG. 20

Targeting of Designed Cytokine

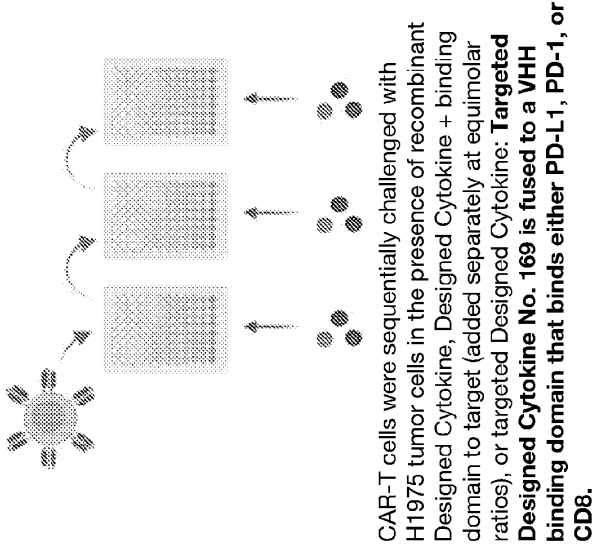


FIG. 21A

Killing activity
(AUC, lower is more killing)

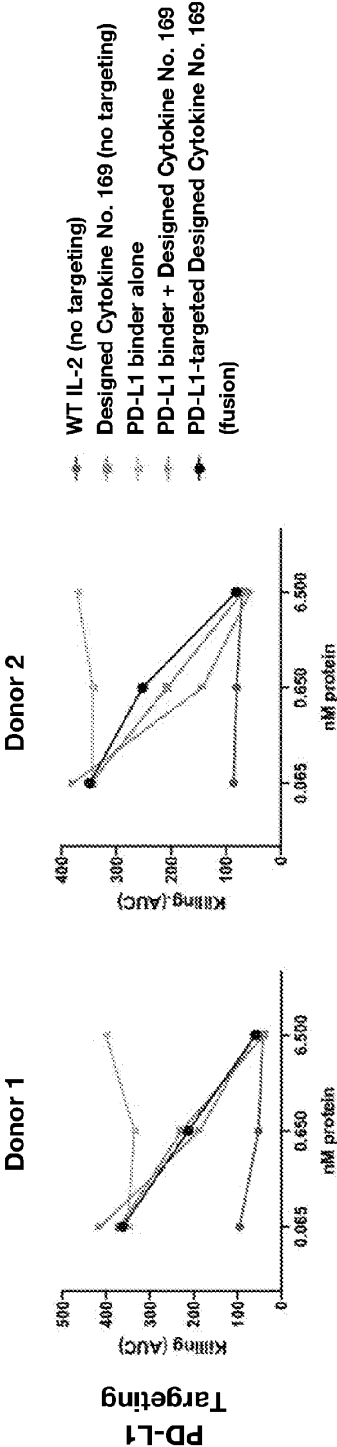


FIG. 21B

Killing activity
(AUC, lower is more killing)

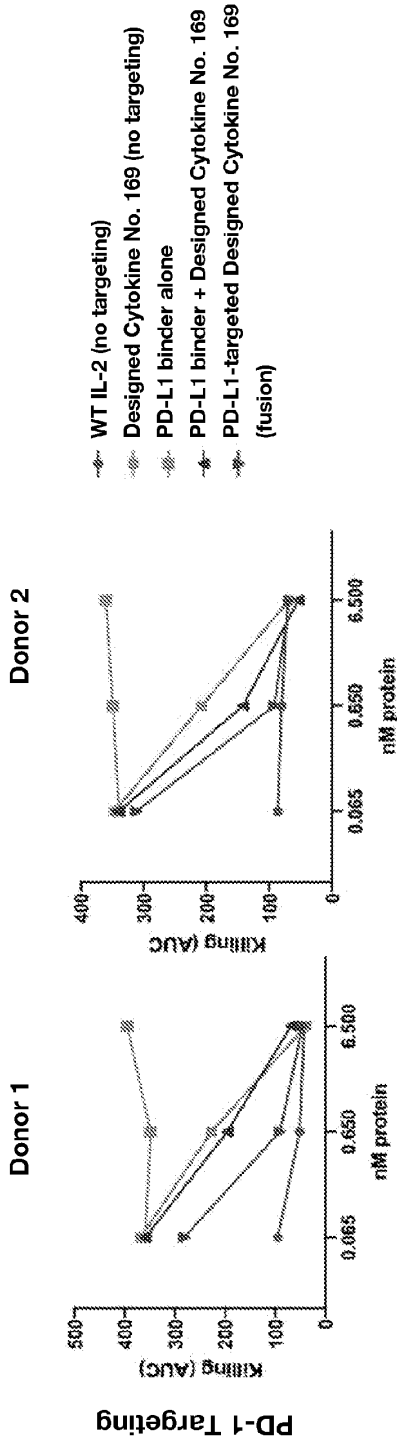


FIG. 21C

Killing activity
(AUC, lower is more killing)

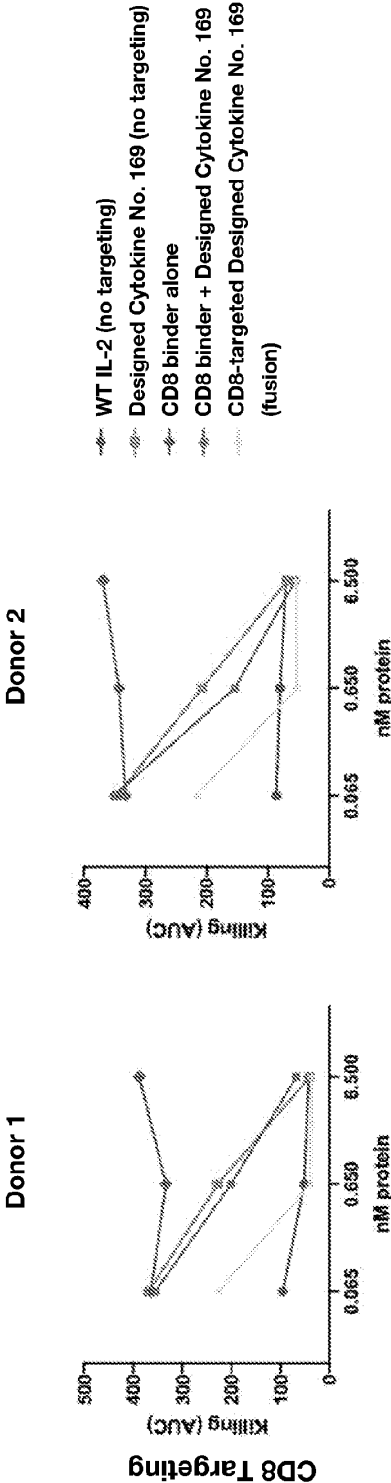


FIG. 21D

Phospho-STAT5 Activity of Designed Cytokines Derived from
Designed Cytokine No. 153

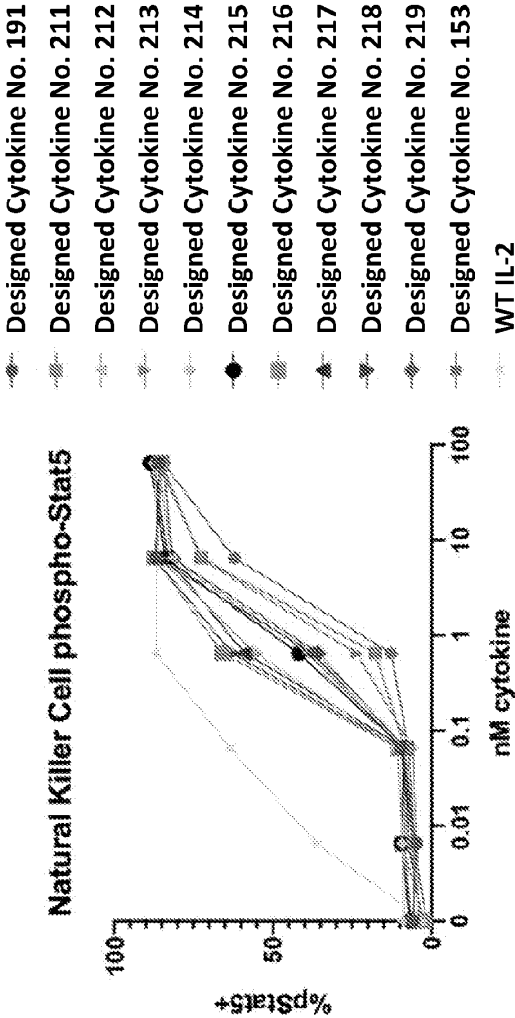


FIG. 22A

Phospho-STAT5 Activity of Designed Cytokines Derived from Designed Cytokine No. 153

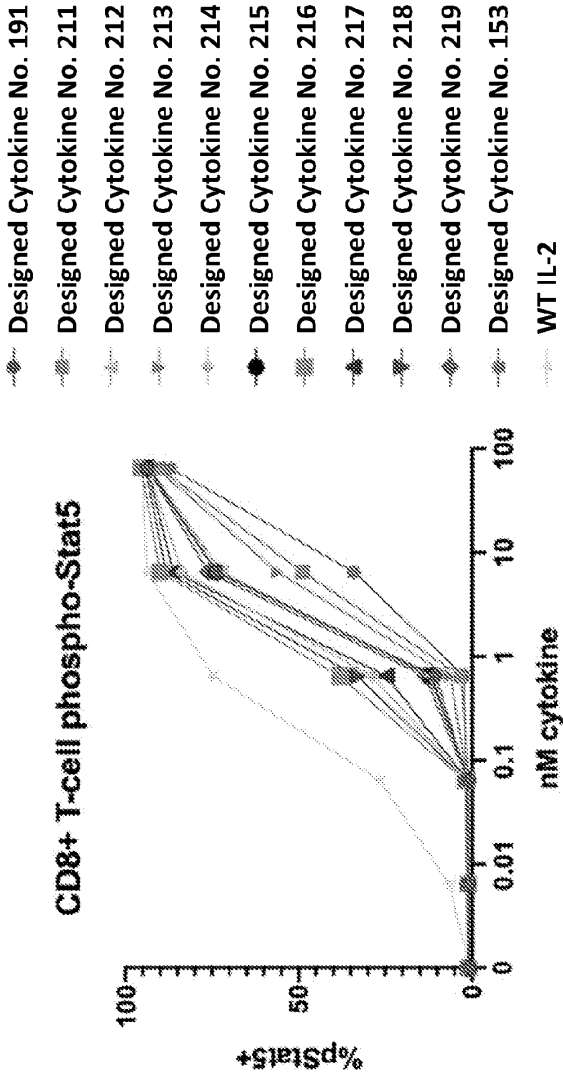
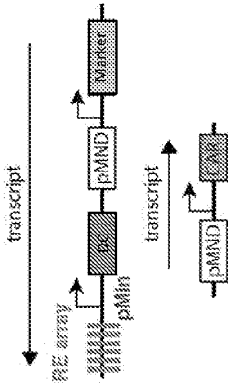


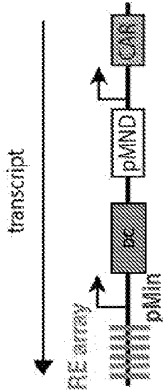
FIG. 22B

Inducible Designed Cytokines (DCs), Vectors and Circuits

2V-A: 2 Vector All in one



1V-A: 1 Vector All in one



AUTOREGULATION (AUT)

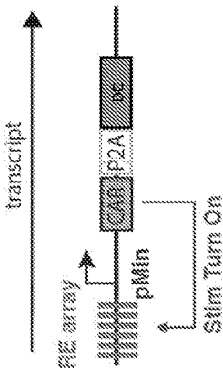


FIG. 23A

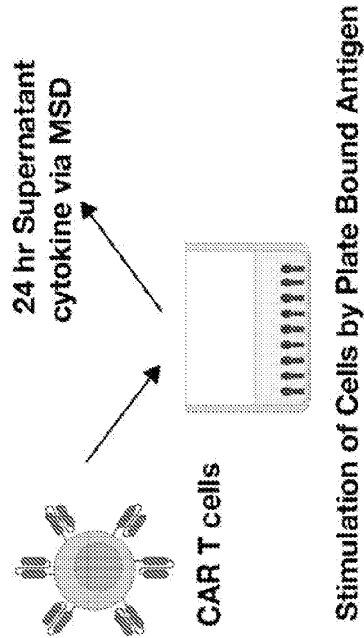


FIG. 23B

Inducible Designed Cytokines (DCs) show increased expression relative to Control

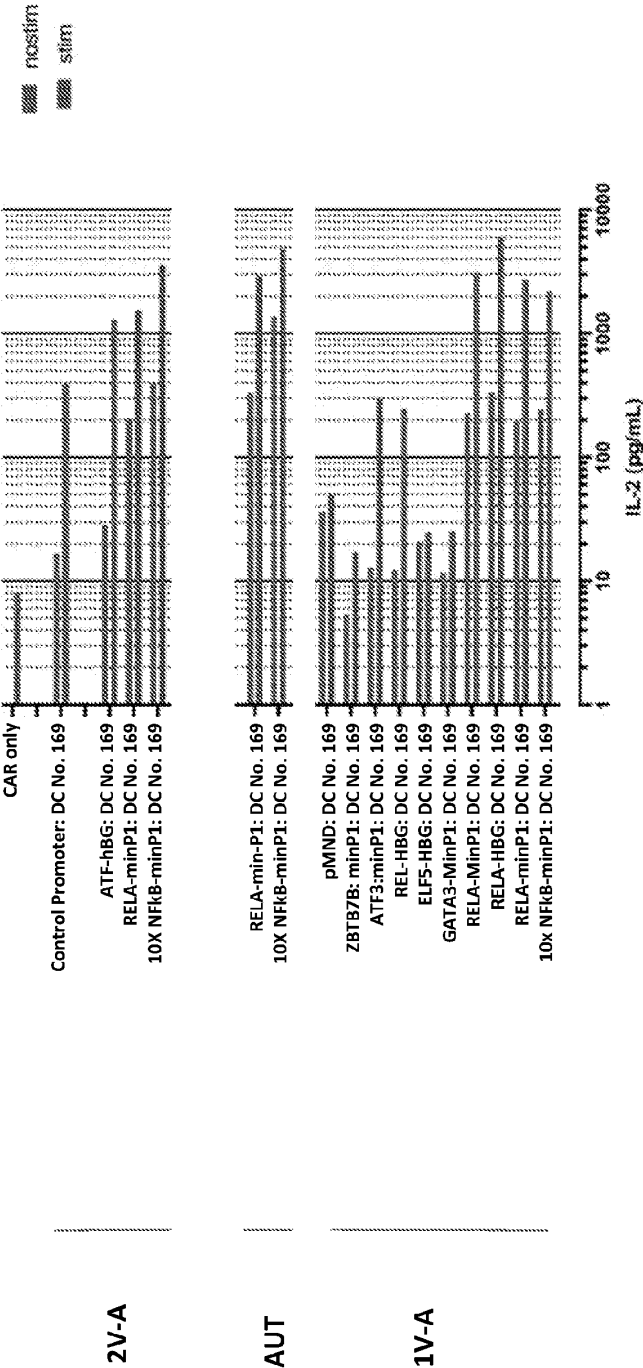
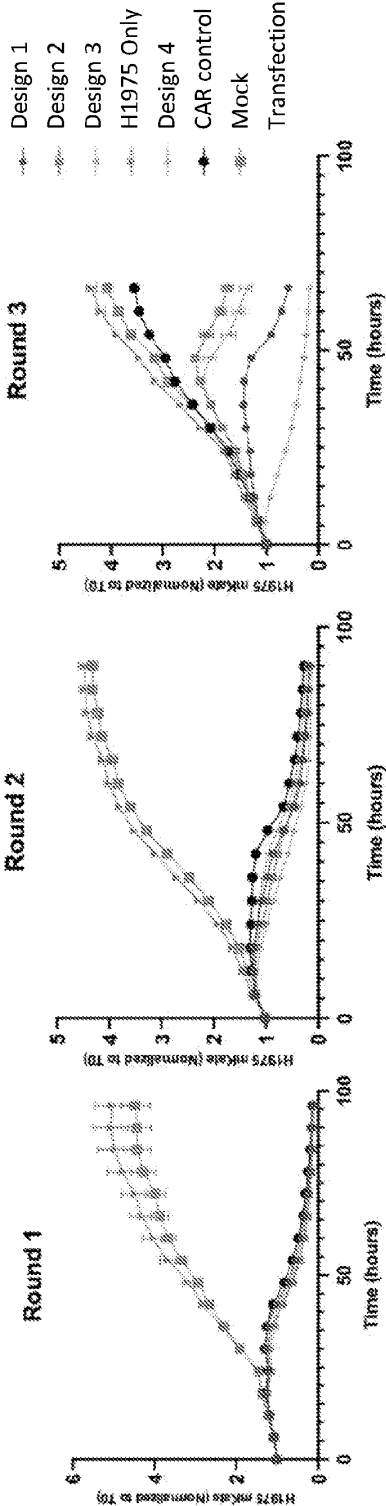


FIG. 23C

Sequential Killing with Inducible Designed Cytokines



- These constructs continued to kill at the end of round 3
 - Design 1: V1-a, RELA-HBG: DC No. 169
 - Design 2: V1-a, RELA-minP1: DC No. 169
 - Design 3: V1-a, GATA3-MinP1: DC No. 169
 - Design 4: Autoregulated, RELA-minP1 : DC No. 169
 - CAR only control

FIG. 24

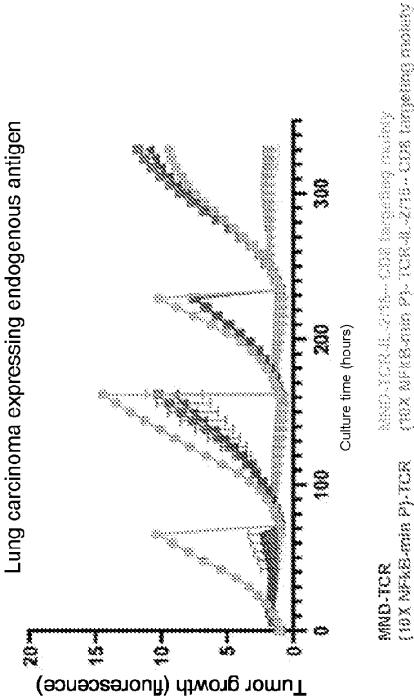


FIG. 25

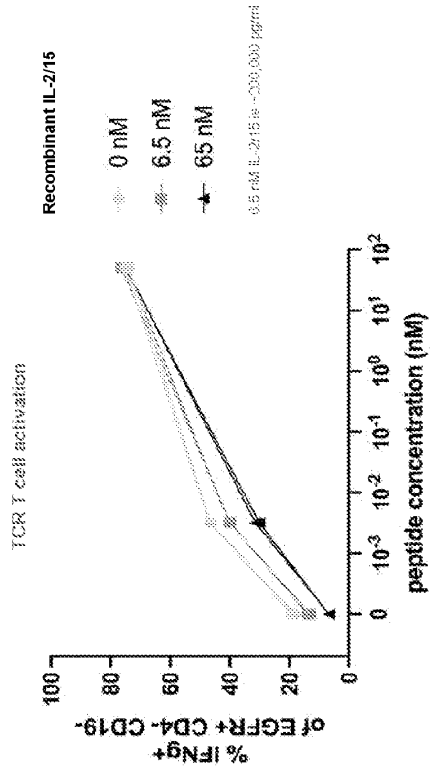


FIG. 26

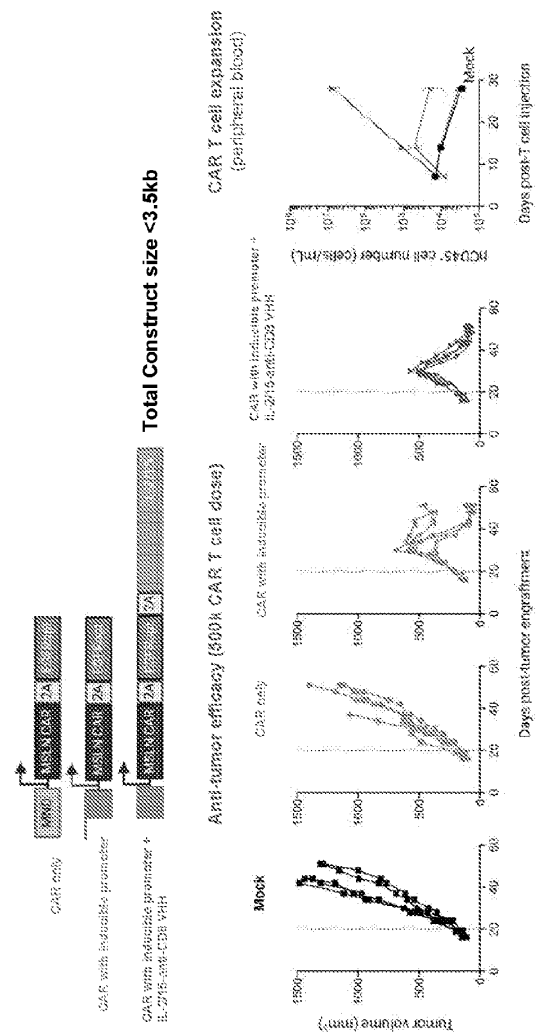


FIG. 27

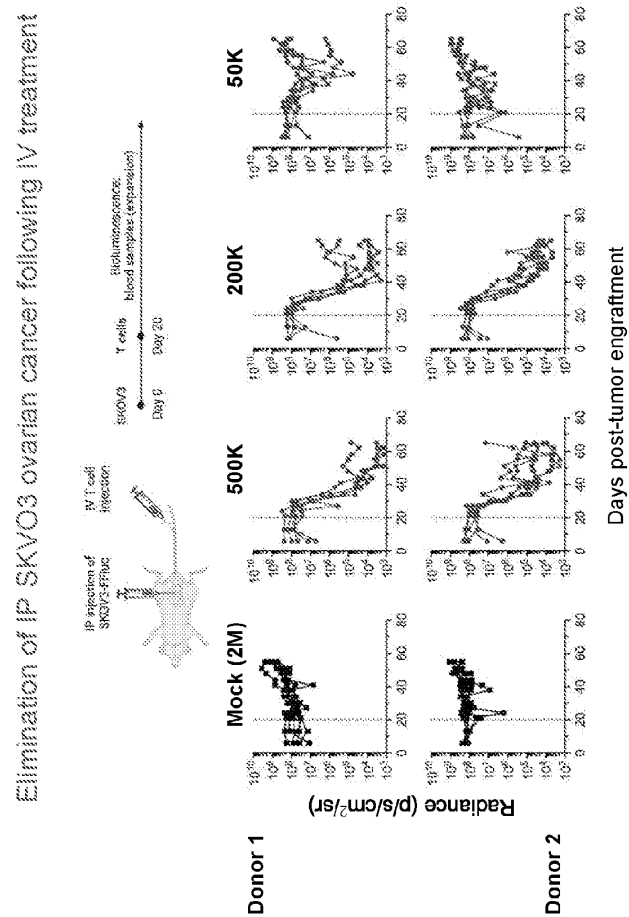


FIG. 28

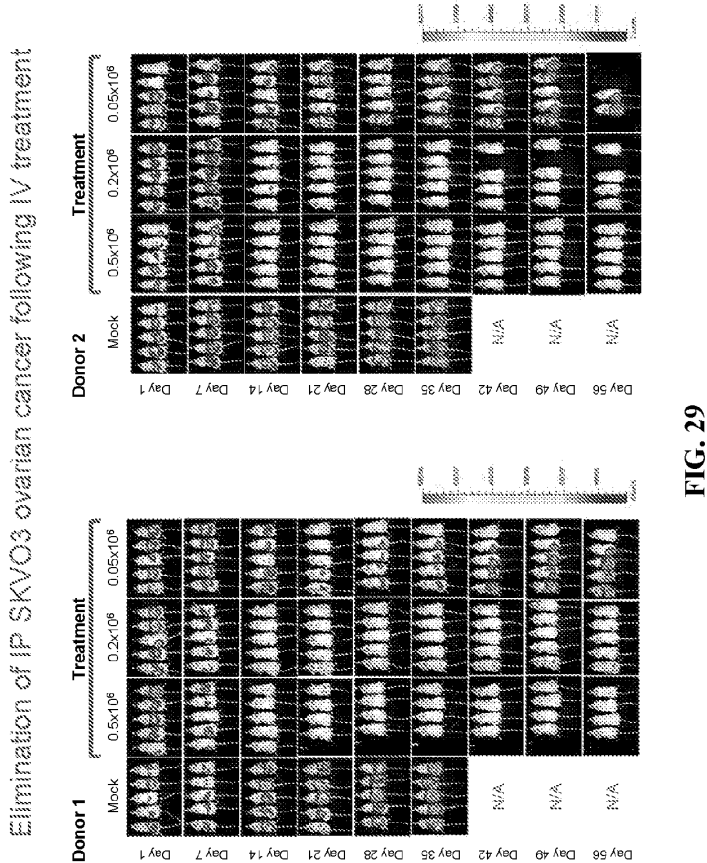


FIG. 29

Elimination of IP SKOV3 ovarian cancer following IV Treatment

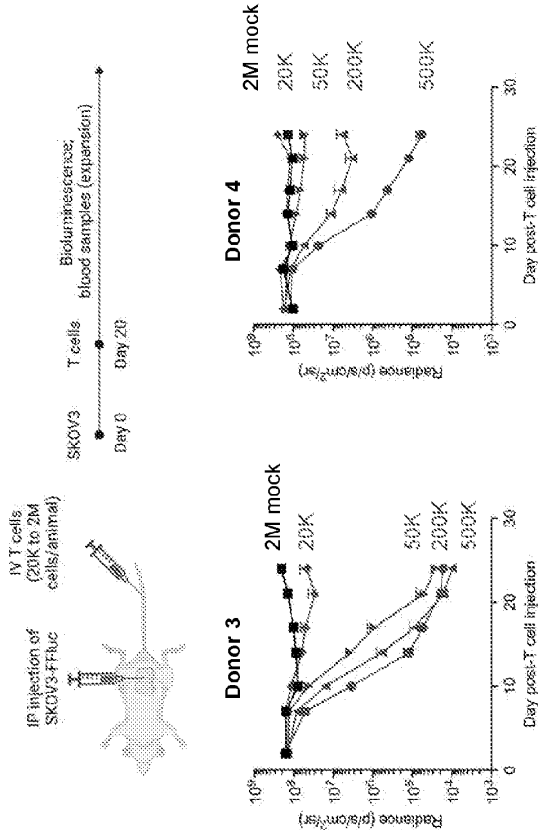


FIG. 30