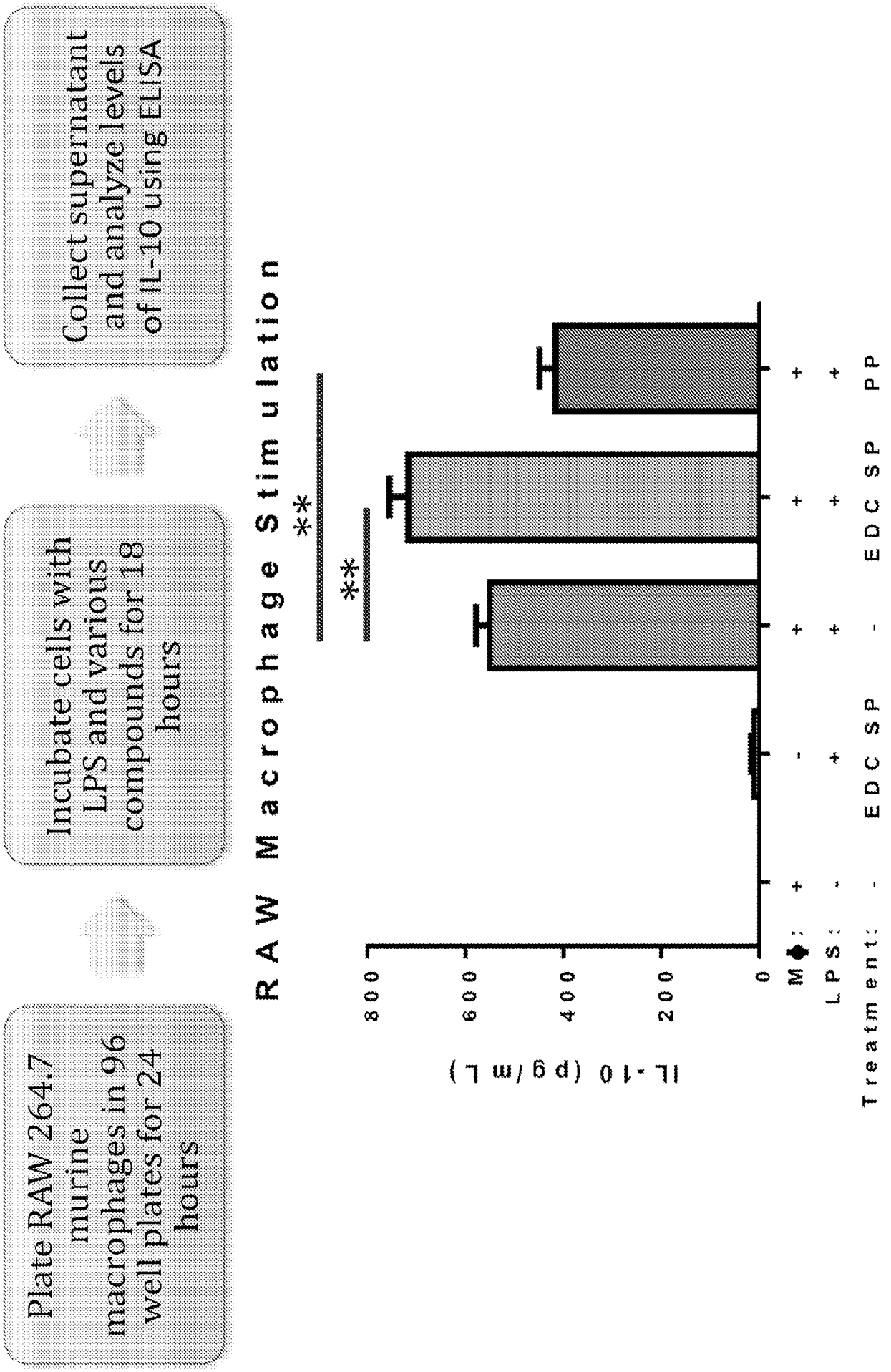
FIGURE 1


Plate RAW 264.7 murine macrophages in 96 well plates for 24 hours
Incubate cells with LPS and various compounds for 18 hours
Collect supernatant and analyze levels of IL-10 using ELISA
RAW Macrophage Stimulation
**
**
IL-10 (pg/mL)
800
600
400
200
0
Treatment: - EDC SP - EDC SP PP
LPS: - + + + + +

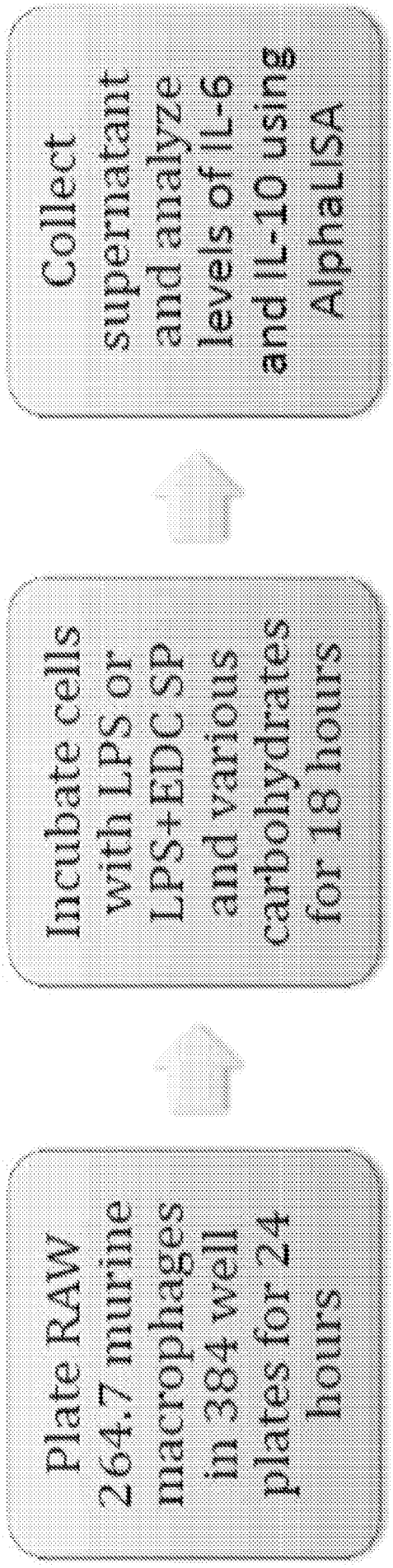

Plate RAW 264.7 murine macrophages in 384 well plates for 24 hours
Incubate cells with LPS or LPS+EDC SP and various carbohydrates for 18 hours
Collect supernatant and analyze levels of IL-6 and IL-10 using AlphaLISA

FIGURE 2

FIGURE 3A

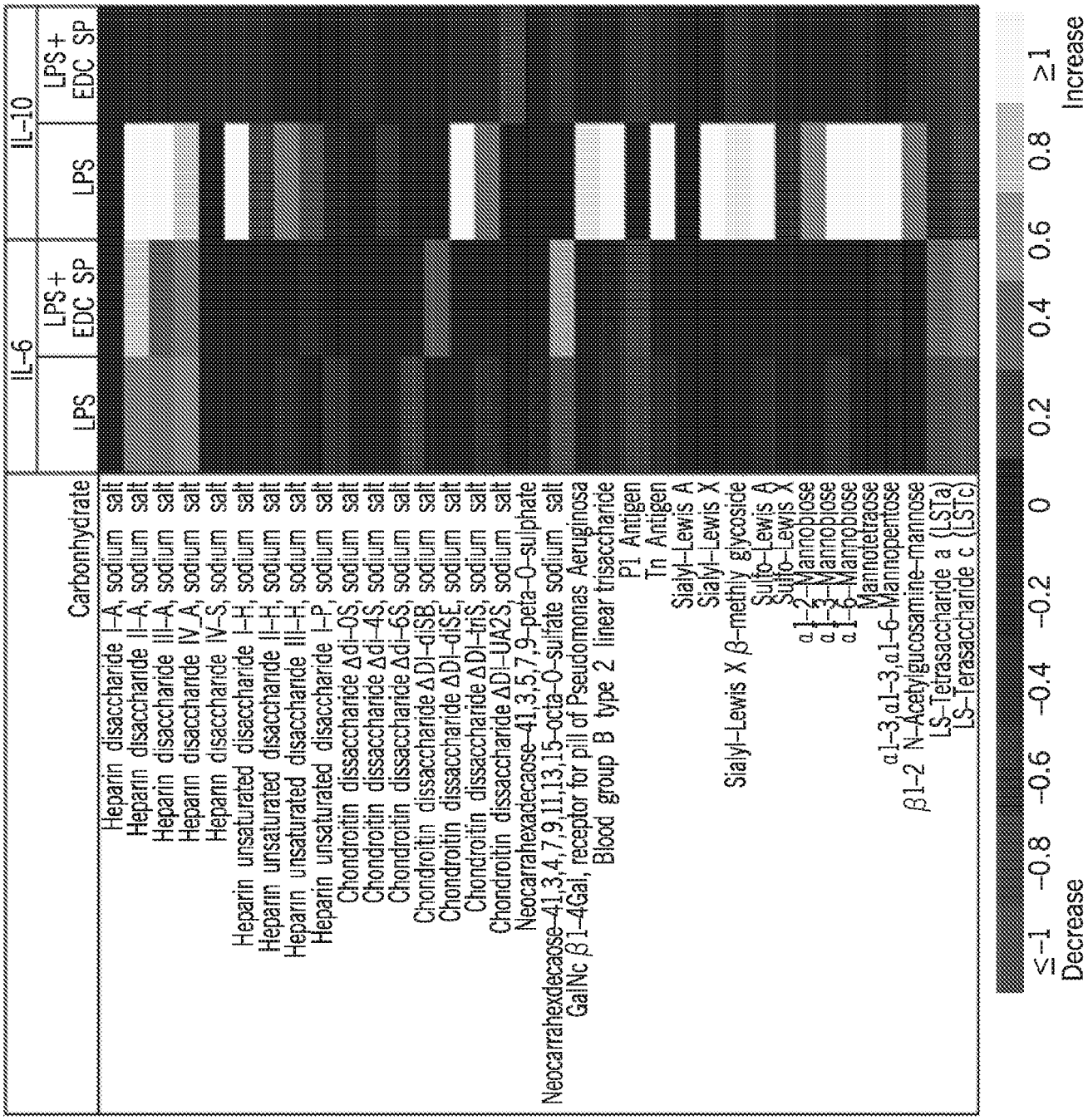

IL-6
IL-10
LPS
LPS + EDC SP
LPS
LPS + EDC SP
Carbonhydrate
Heparin disaccharide I-A, sodium salt
Heparin disaccharide II-A, sodium salt
Heparin disaccharide III-A, sodium salt
Heparin disaccharide IV_A, sodium salt
Heparin disaccharide IV-S, sodium salt
Heparin unsaturated disaccharide I-H, sodium salt
Heparin unsaturated disaccharide II-H, sodium salt
Heparin unsaturated disaccharide III-H, sodium salt
Heparin unsaturated disaccharide I-P, sodium salt
Chondroitin dissaccharide Δdi-0S, sodium salt
Chondroitin dissaccharide Δdi-4S, sodium salt
Chondroitin dissaccharide Δdi-6S, sodium salt
Chondroitin dissaccharide ΔDI-diSB, sodium salt
Chondroitin dissaccharide ΔDI-diSE, sodium salt
Chondroitin dissaccharide ΔDI-triS, sodium salt
Chondroitin dissaccharide ΔDI-UA2S, sodium salt
Neocarrahexadecaose-41,3,5,7,9-peta-O-sulphate
Neocarrahexdecaose-41,3,4,7,9,11,13,15-octa-O-sulfate sodium salt
GalNc β1-4Gal, receptor for pill of Pseudomonas Aeruginosa
Blood group B type 2 linear trisaccharide
P1 Antigen
Tn Antigen
Sialyl-Lewis A
Sialyl-Lewis X
Sialyl-Lewis X β-methly glycoside
Sulfo-Lewis A
Sulfo-Lewis X
α1-2-Mannobiose
α1-3-Mannobiose
α1-6-Mannobiose
Mannotetraose
α1-3,α1-3,α1-6-Mannopentose
β1-2 N-Acetylglucosamine-mannose
LS-Tetrasaccharide a (LSTa)
LS-Terasaccharide c (LSTc)
≤-1 -0.8 -0.6 -0.4 -0.2 0 0.2 0.4 0.6 0.8 ≥1
Decrease
Increase
Fold change of cytokine from baseline LPS or LPS+EDC SP stimulation

FIGURE 3B

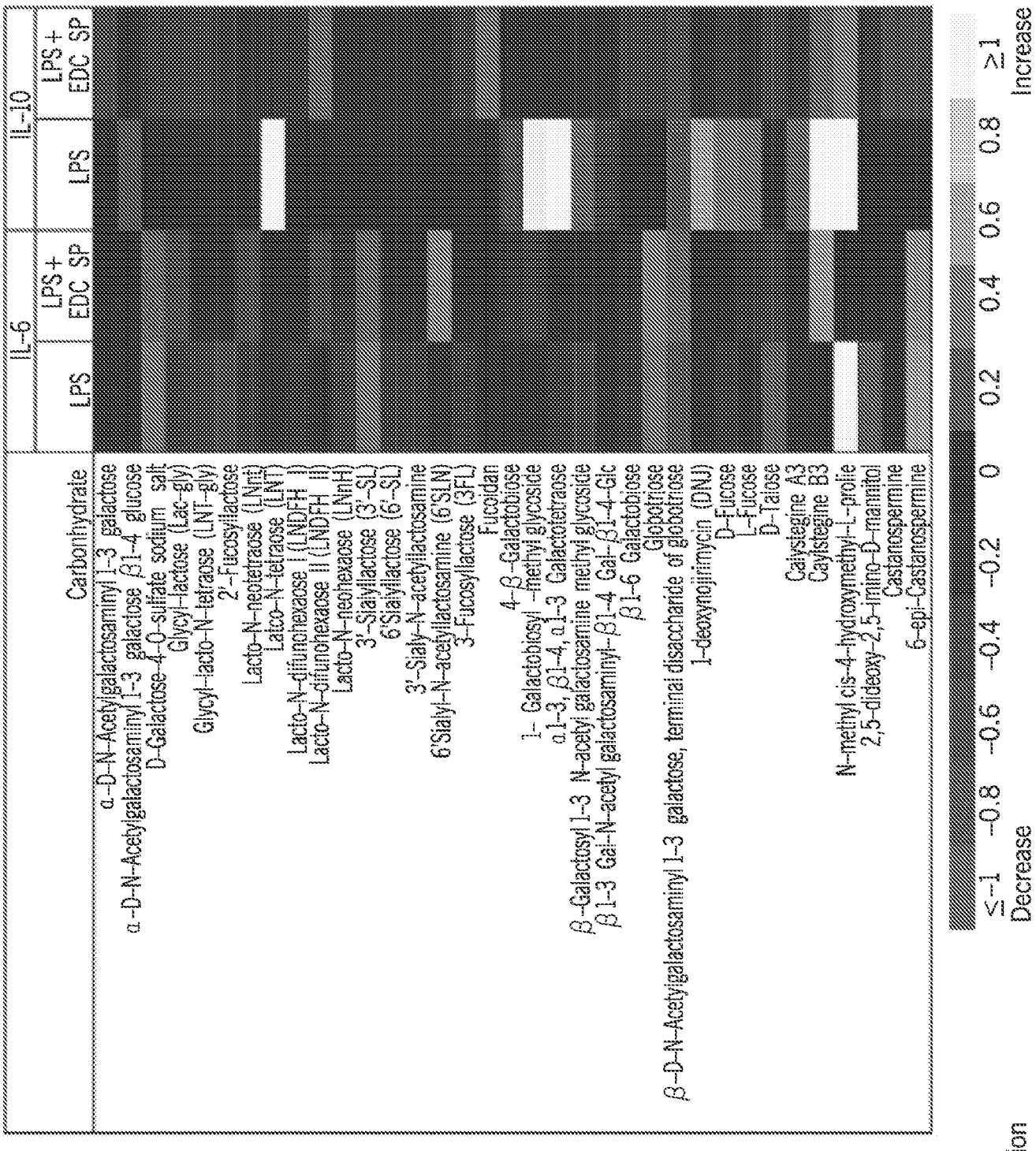
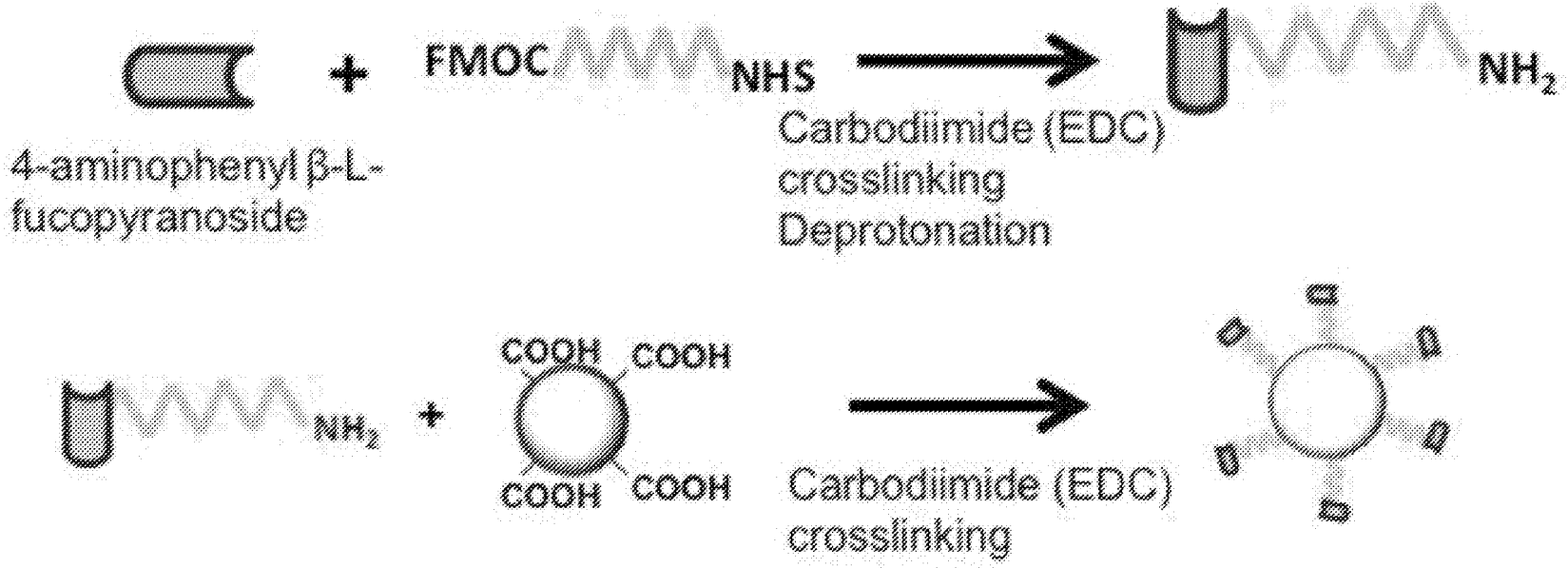
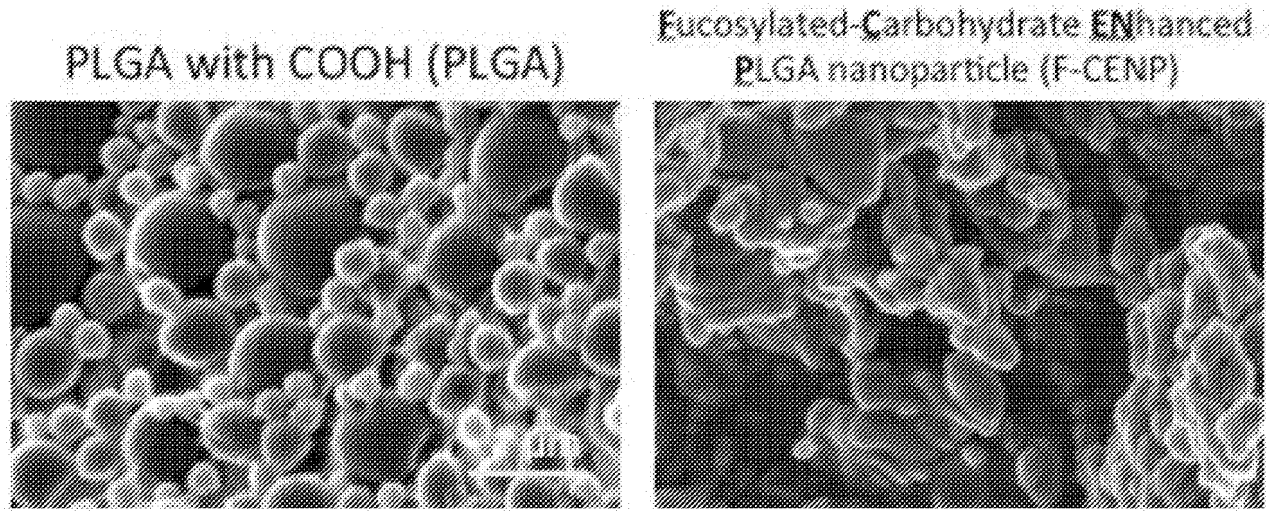

IL-6
LPS
LPS + EDC SP
IL-10
LPS
LPS + EDC SP
Carbohydrate
α-D-N-Acetylgalactosaminyl 1-3 galactose
α-D-N-Acetylgalactosaminyl 1-3 galactose β1-4 glucose
D-Galactose-4-O-sulfate sodium salt
Glycyl-lactose (Lac-gly)
Glycyl-lacto-N-tetraose (LNT-gly)
2'-Fucosyllactose
Lacto-N-neotetraose (LNnt)
Lacto-N-tetraose (LNT)
Lacto-N-difunchexaose I (LNDFH I)
Lacto-N-difunohexaose II (LNDFH II)
Lacto-N-neohexaose (LNnH)
3'-Sialyllactose (3'-SL)
6'Sialyllactose (6'-SL)
3'-Sialy-N-acetyllactosamine
6'Sialyl-N-acetyllactosamine (6'SLN)
3-Fucosyllactose (3FL)
Fucoidan
4-β-Galactobiose
1- Galactobiosyl -methyl glycoside
α1-3, β1-4, α1-3 Galactotetraose
β-Galactosyl 1-3 N-acetyl galactosamine methyl glycoside
β1-3 Gal-N-acetyl galactosaminyl-β1-4 Gal-β1-4-Glc
β1-6 Galactobiose
Globotriose
β-D-N-Acetylgalactosaminyl 1-3 galactose, terminal disaccharide of globotriose
1-deoxynojirimycin (DNJ)
D-Fucose
L-Fucose
D-Talose
Calystegine A3
Calystegine B3
N-methyl cis-4-hydroxymethyl-L-prolie
2,5-dideoxy-2,5-imino-D-mannitol
Castanospermine
6-epi-Castanospermine
≤-1 -0.8 -0.6 -0.4 -0.2 0 0.2 0.4 0.6 0.8 ≥1
Decrease
Increase
Fold change of cytokine from baseline LPS or LPS + EDC SP stimulation

FIGURE 4


FMOC
NHS
NH2
4-aminophenyl β-L-fucopyranoside
Carbodiimide (EDC) crosslinking Deprotonation
COOH
COOH
COOH
COOH
NH2
Carbodiimide (EDC) crosslinking


PLGA with COOH (PLGA)
Fucosylated-Carbohydrate ENhanced PLGA nanoparticle (F-CENP)

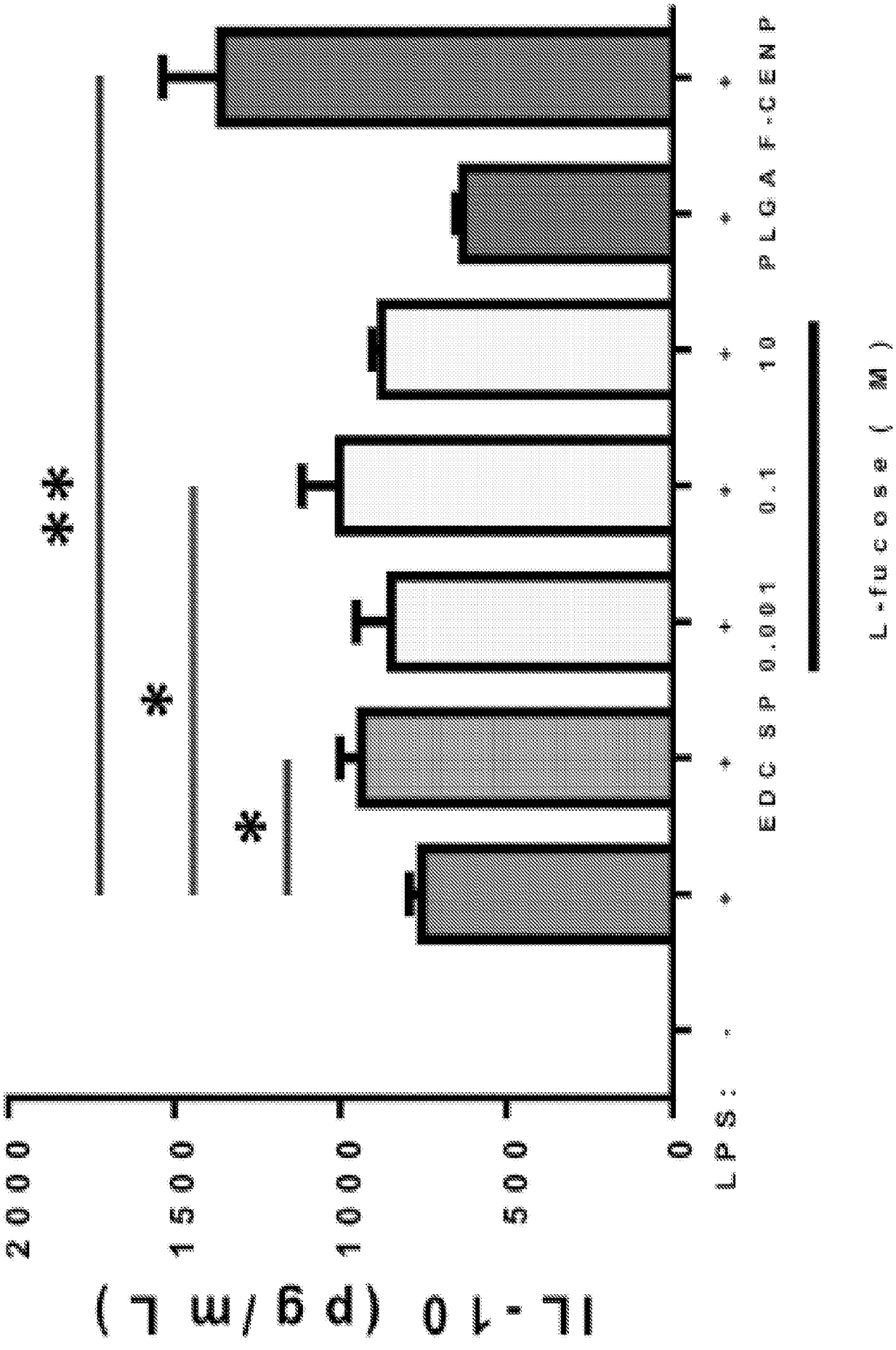

IL-10 (pg/mL)
2000
1500
1000
500
0
LPS: -
+
+
+
+
+
+
EDC SP 0.001 0.1 10
PLGA F-CENP
L-fucose (mM)
*
*
**

FIGURE 5

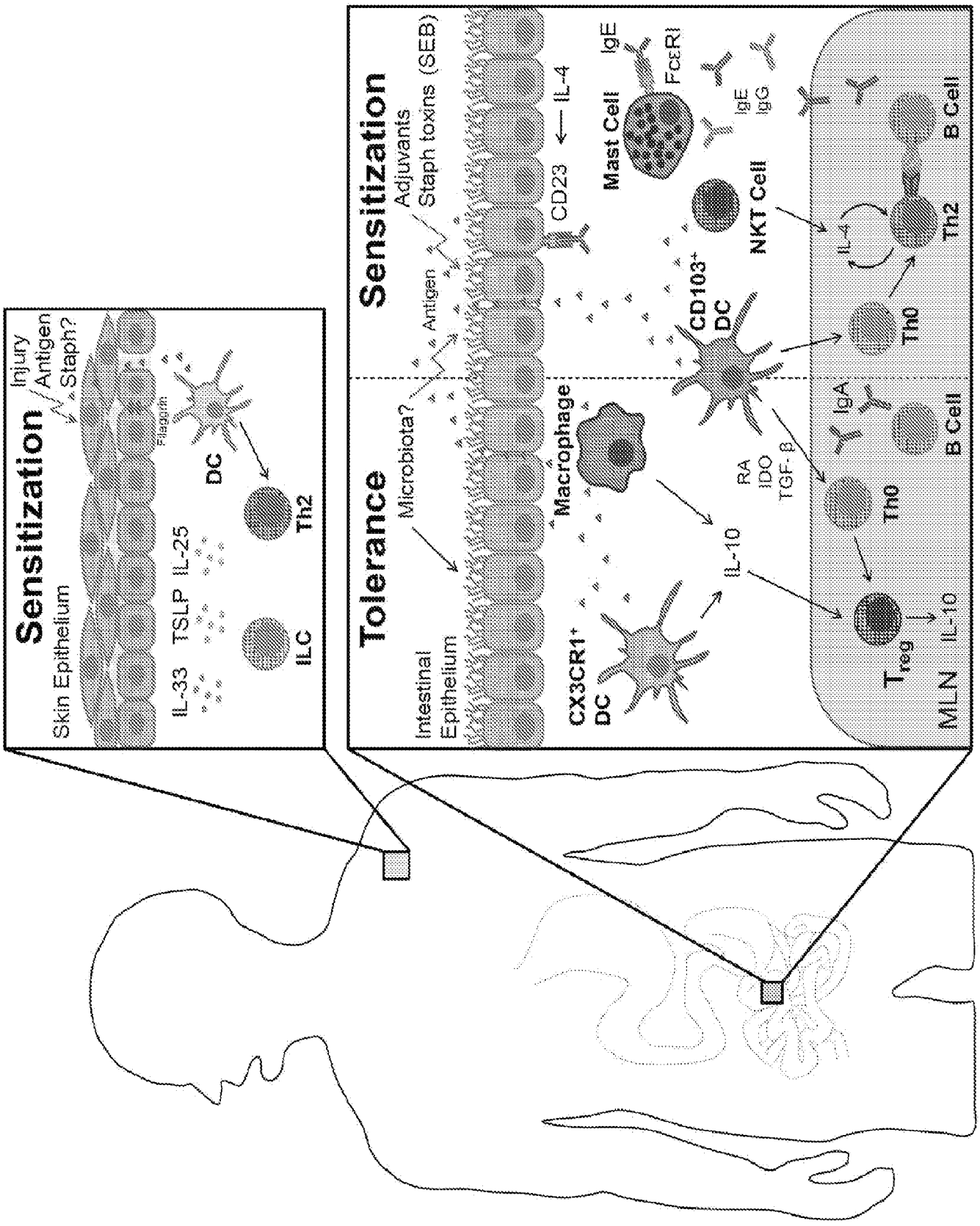
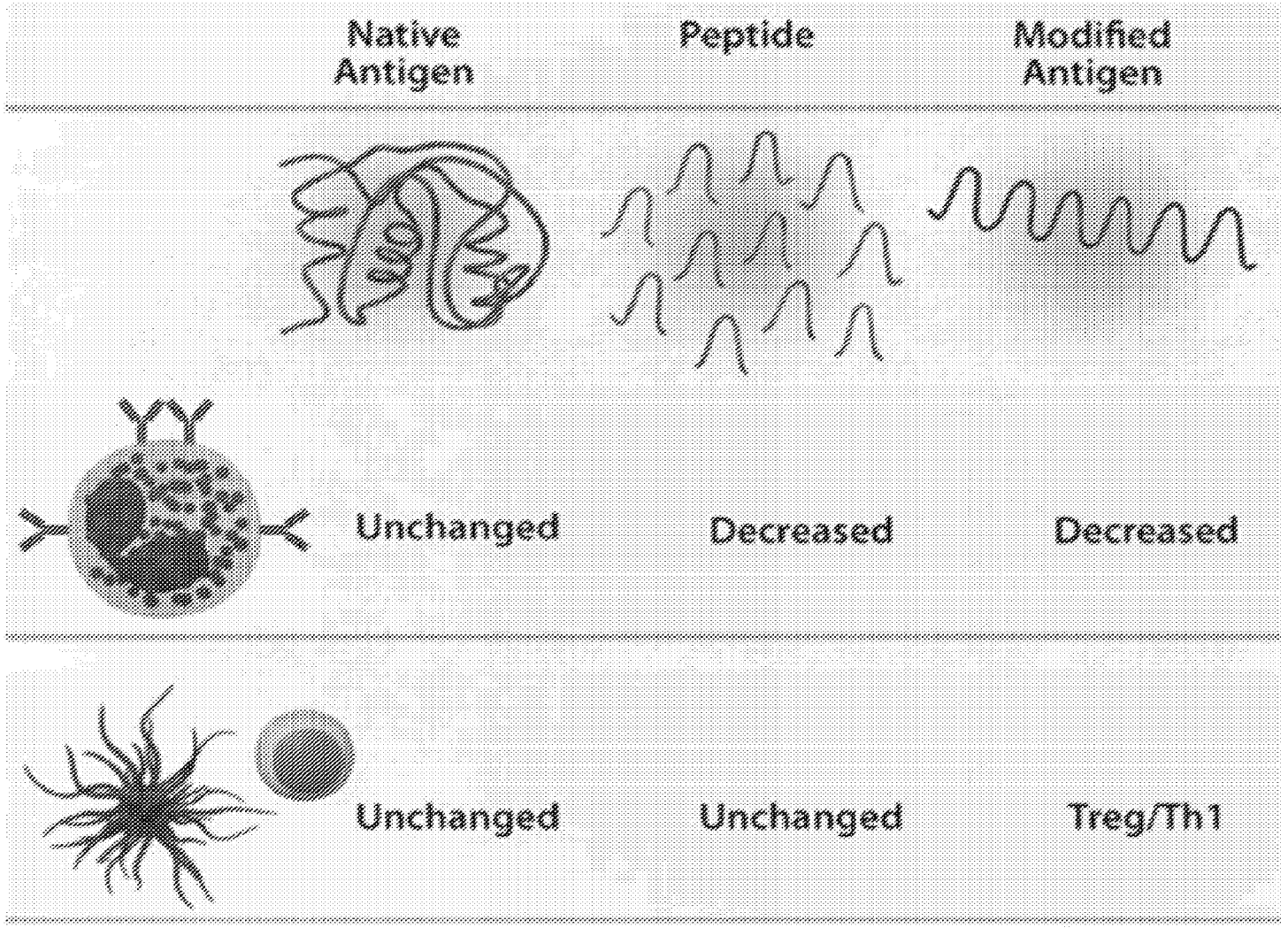

Sensitization
Injury
Antigen
Staph?
Skin Epithelium
Filaggrin
IL-33
TSLP
IL-25
DC
ILC
Th2
Tolerance
Microbiota?
Intestinal Epithelium
Antigen
Macrophage
CX3CR1+ DC
IL-10
RA
IDO
TGF-β
Sensitization
Adjuvants
Staph toxins (SEB)
CD23
IL-4
Mast Cell
IgE
FcεRI
CD103+ DC
NKT Cell
IgE
IgG
IL-4
Th0
Th2
B Cell
IgA
Th0
B Cell
Treg
IL-10
MLN

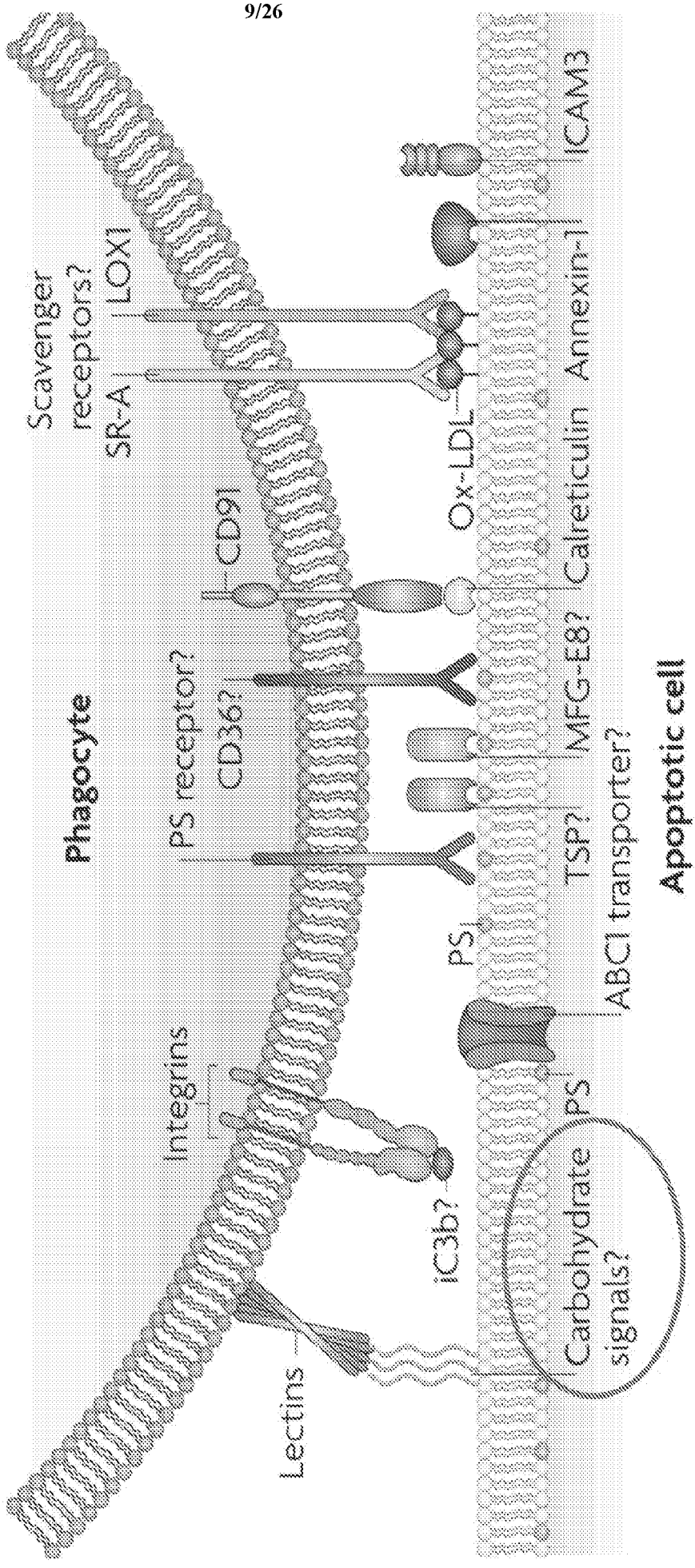
FIGURE 6

FIGURE 7



SUBSTITUTE SHEET (RULE 26)

FIGURE 8


Phagocyte
Scavenger receptors?
SR-A
LOX1
PS receptor?
CD36?
CD91
Integrins
Lectins
iC3b?
Ox-LDL
ICAM3
Annexin-1
Calreticulin
MFG-E8?
TSP?
PS
ABC1 transporter?
PS
Carbohydrate signals?
Apoptotic cell

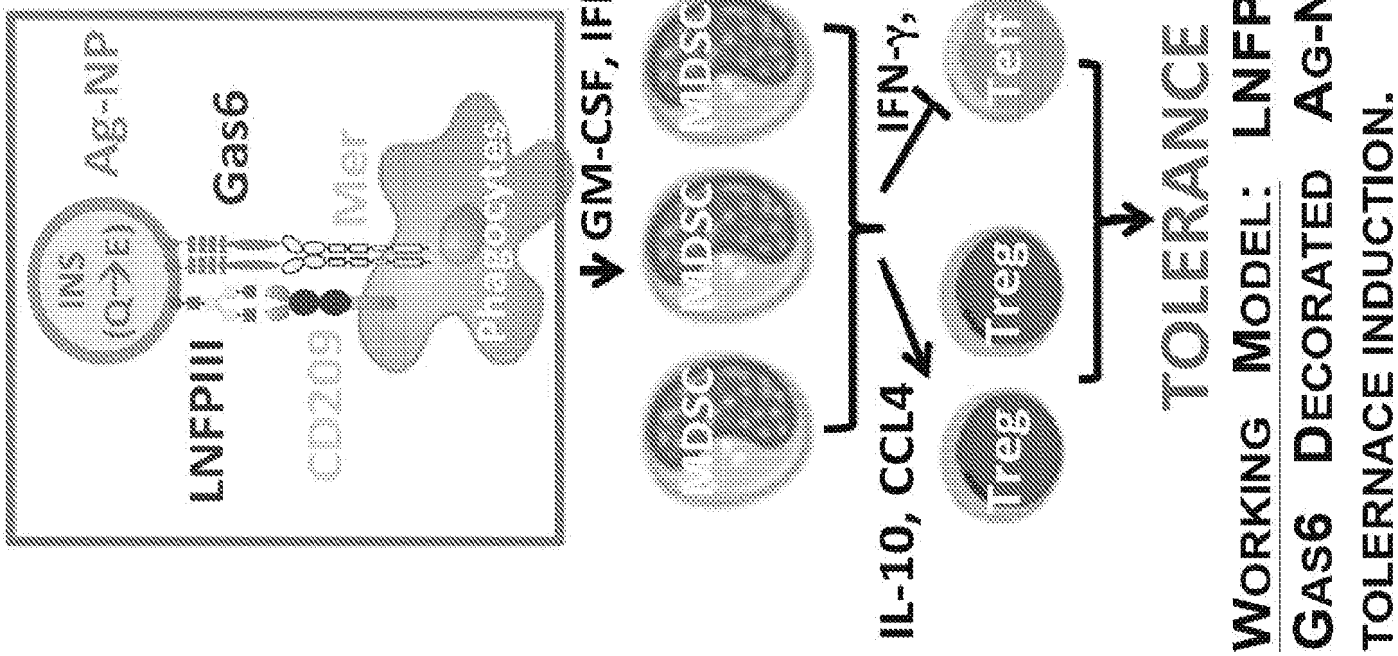


FIGURE 9

WORKING MODEL: LNFPIII AND GAS6 DECORATED AG-NP FOR TOLERNACE INDUCTION.

FIGURE 10A

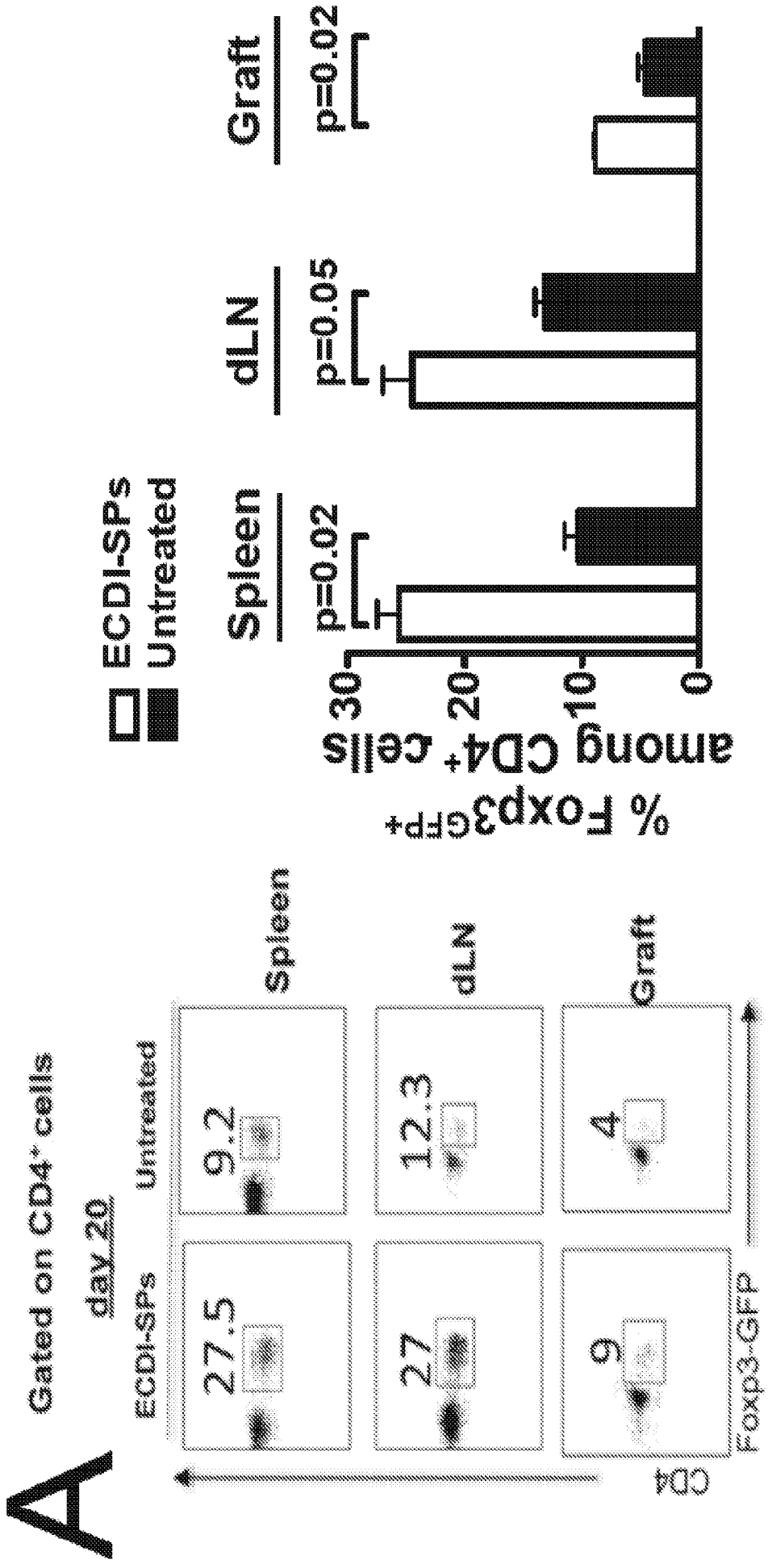
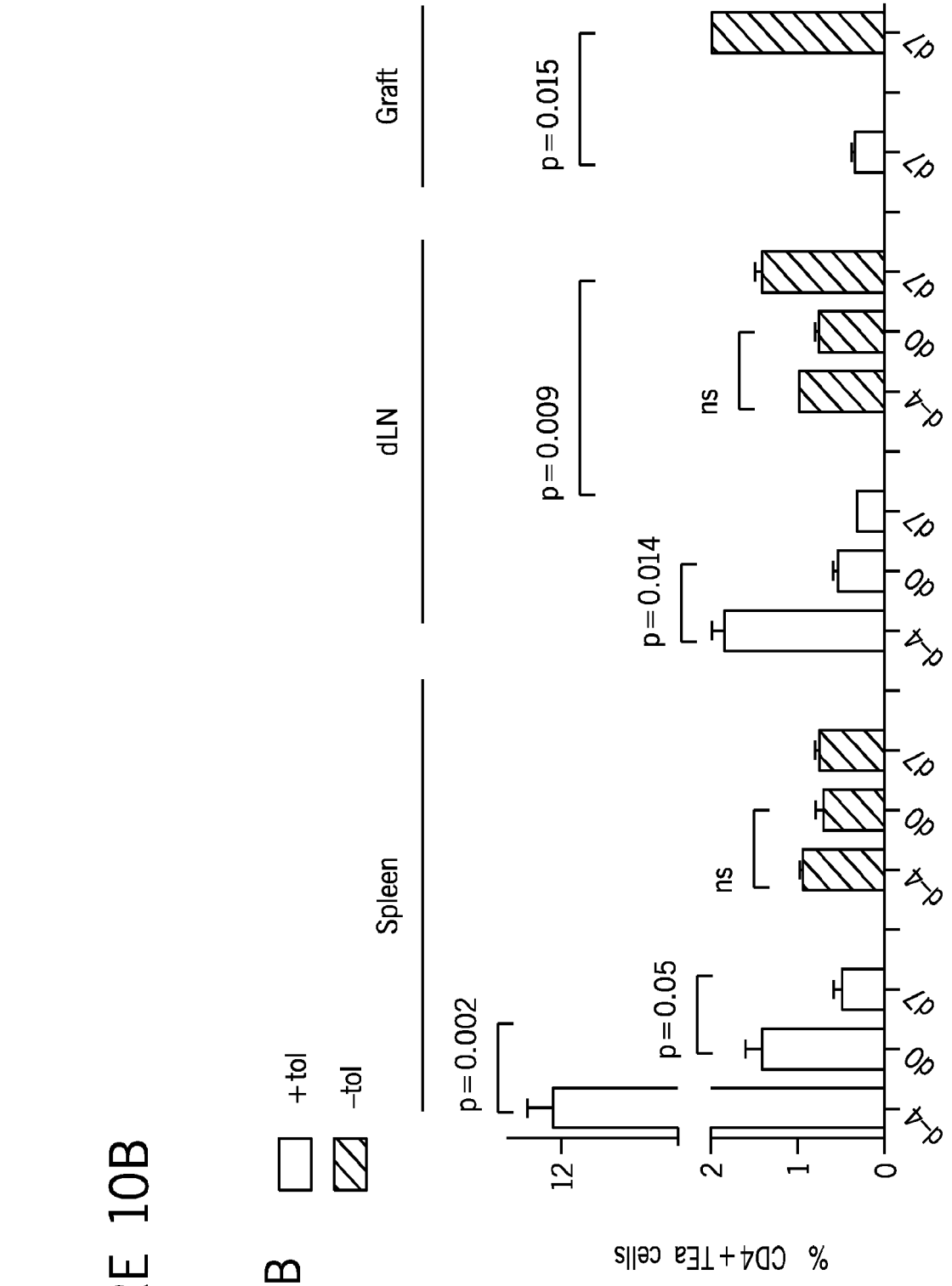

A
Gated on CD4+ cells
day 20
ECDI-SPs
Untreated
Spleen
dLN
Graft
27.5
9.2
27
12.3
9
4
CD4
Foxp3-GFP
% Foxp3GFP+ among CD4+ cells
30
20
10
0
ECDI-SPs
Untreated
Spleen
p=0.02
dLN
p=0.05
Graft
p=0.02

FIGURE 10B


B
+tol
–tol
Spleen
dLN
Graft
p=0.002
p=0.05
ns
p=0.014
p=0.009
ns
p=0.015
% CD4+TEa cells
12
2
1
0
d-4
d0
d7

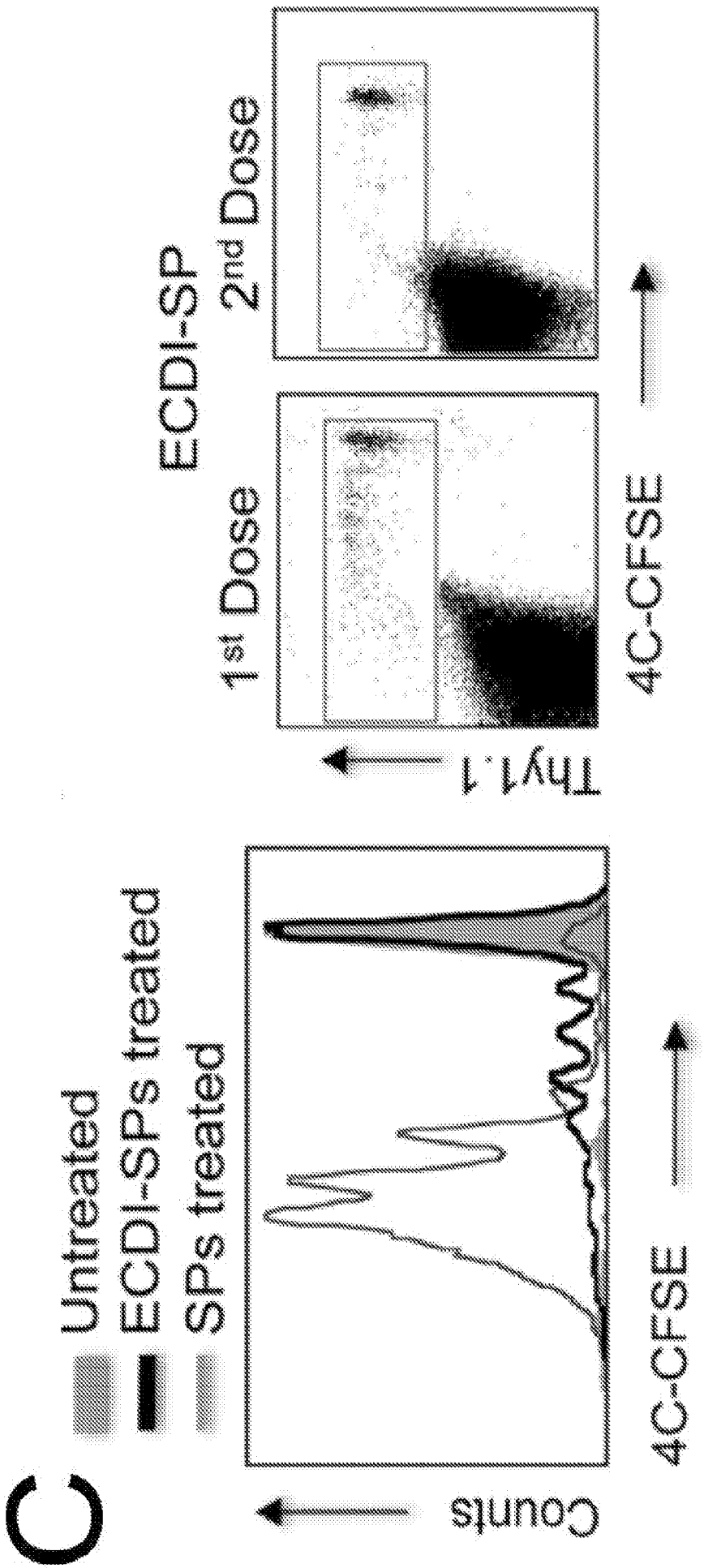

C
Untreated
ECDI-SPs treated
SPs treated
Counts
4C-CFSE
Thy1.1
ECDI-SP
1st Dose
2nd Dose
4C-CFSE

FIGURE 10C

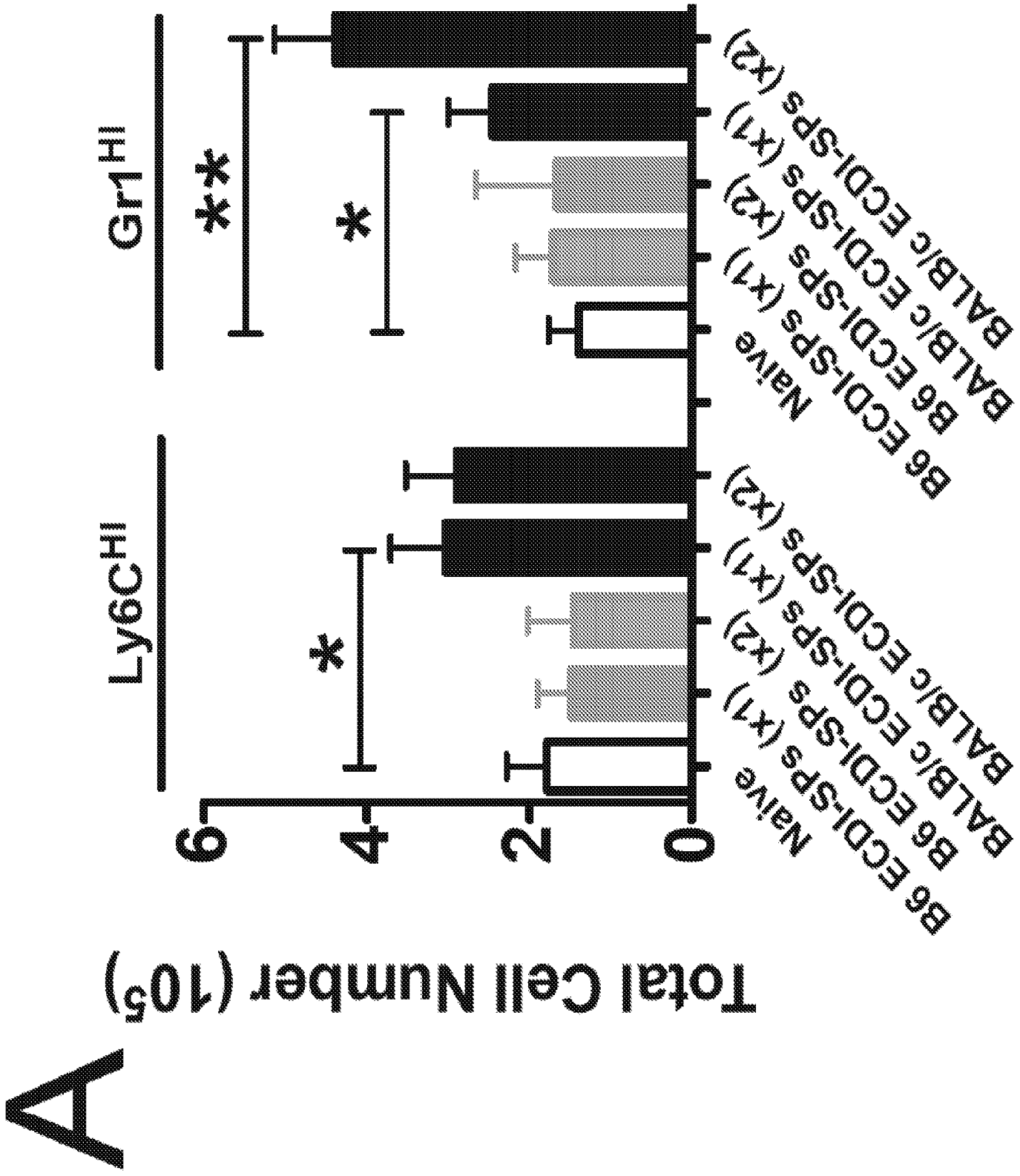

A
Total Cell Number (10^5)
0
2
4
6
Ly6C^{HI}
Gr1^{HI}
*
*
**
Naïve
B6 ECDI-SPs (x1)
B6 ECDI-SPs (x2)
BALB/c ECDI-SPs (x1)
BALB/c ECDI-SPs (x2)

FIGURE 11A

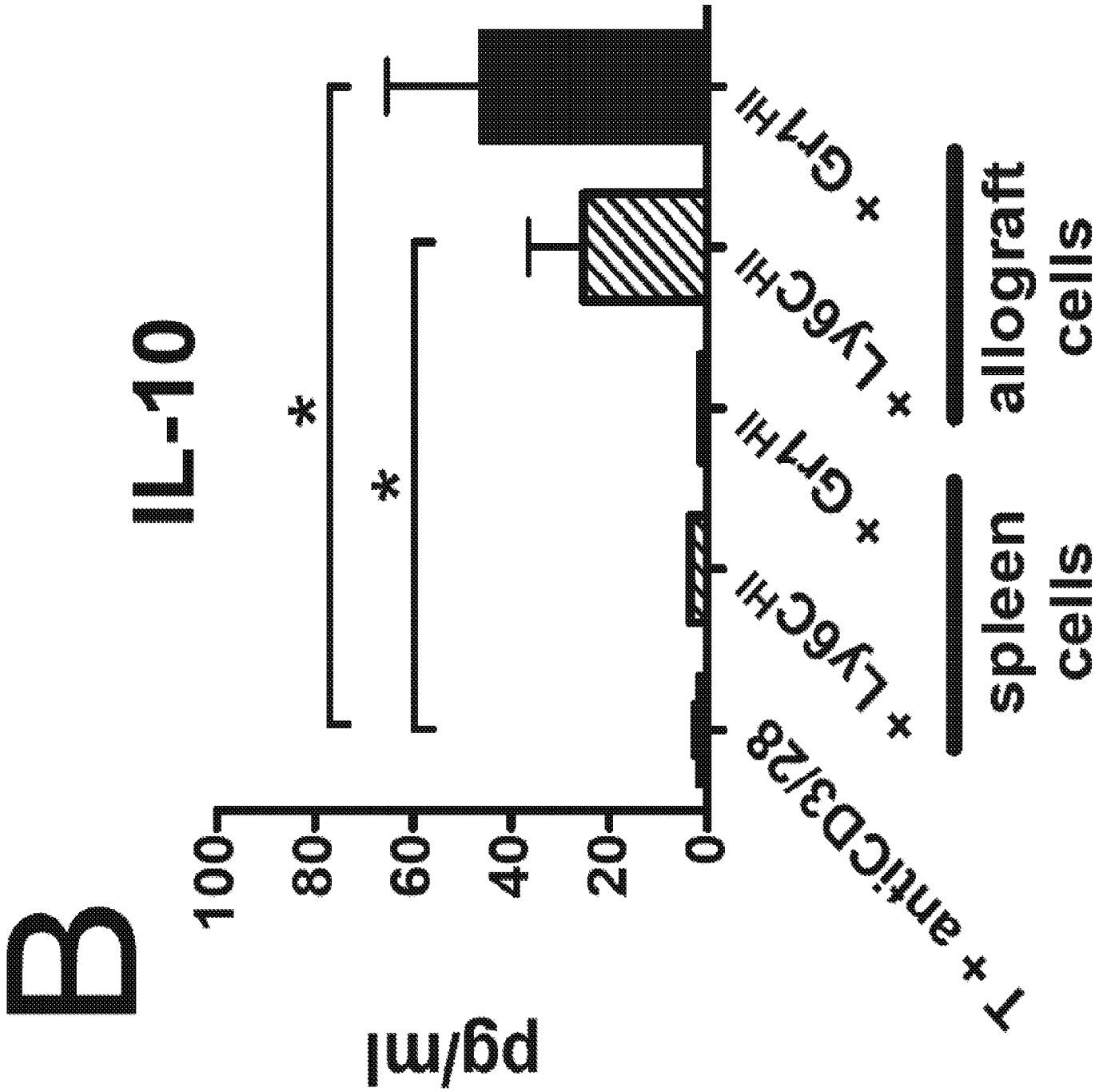

B
IL-10
pg/ml
100
80
60
40
20
0
*
*
T + anticD3/28
+ LyGC^{Hi}
+ GR1^{Hi}
+ LyGC^{Hi}
+ GR1^{Hi}
spleen cells
allograft cells

FIGURE 11B

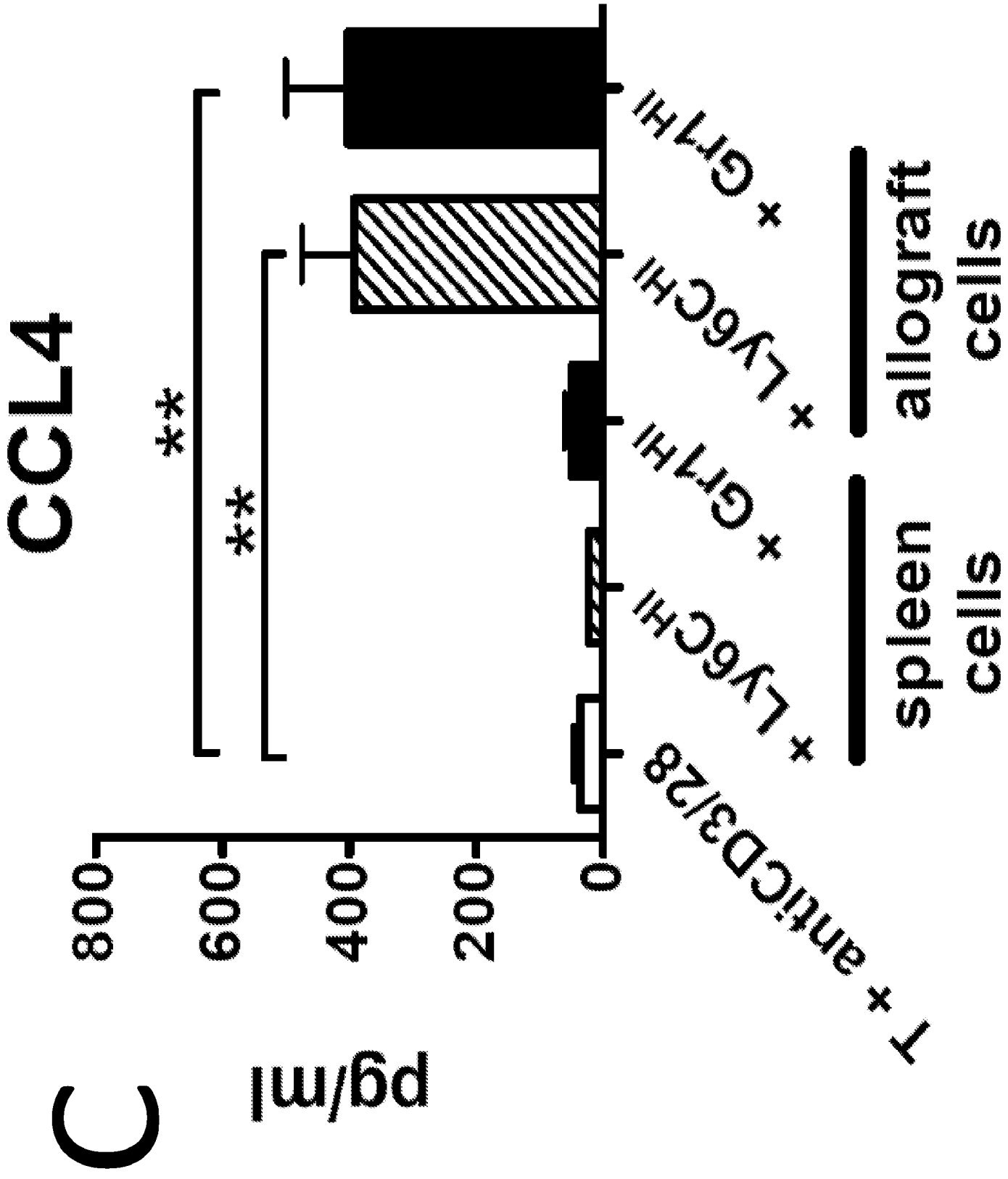

C
CCL4
pg/ml
800
600
400
200
0
**
**
T + antiCD3/28
+ Ly6C^Hi
+ Gr1^Hi
+ Ly6C^Hi
+ Gr1^Hi
spleen cells
allograft cells

FIGURE 11C

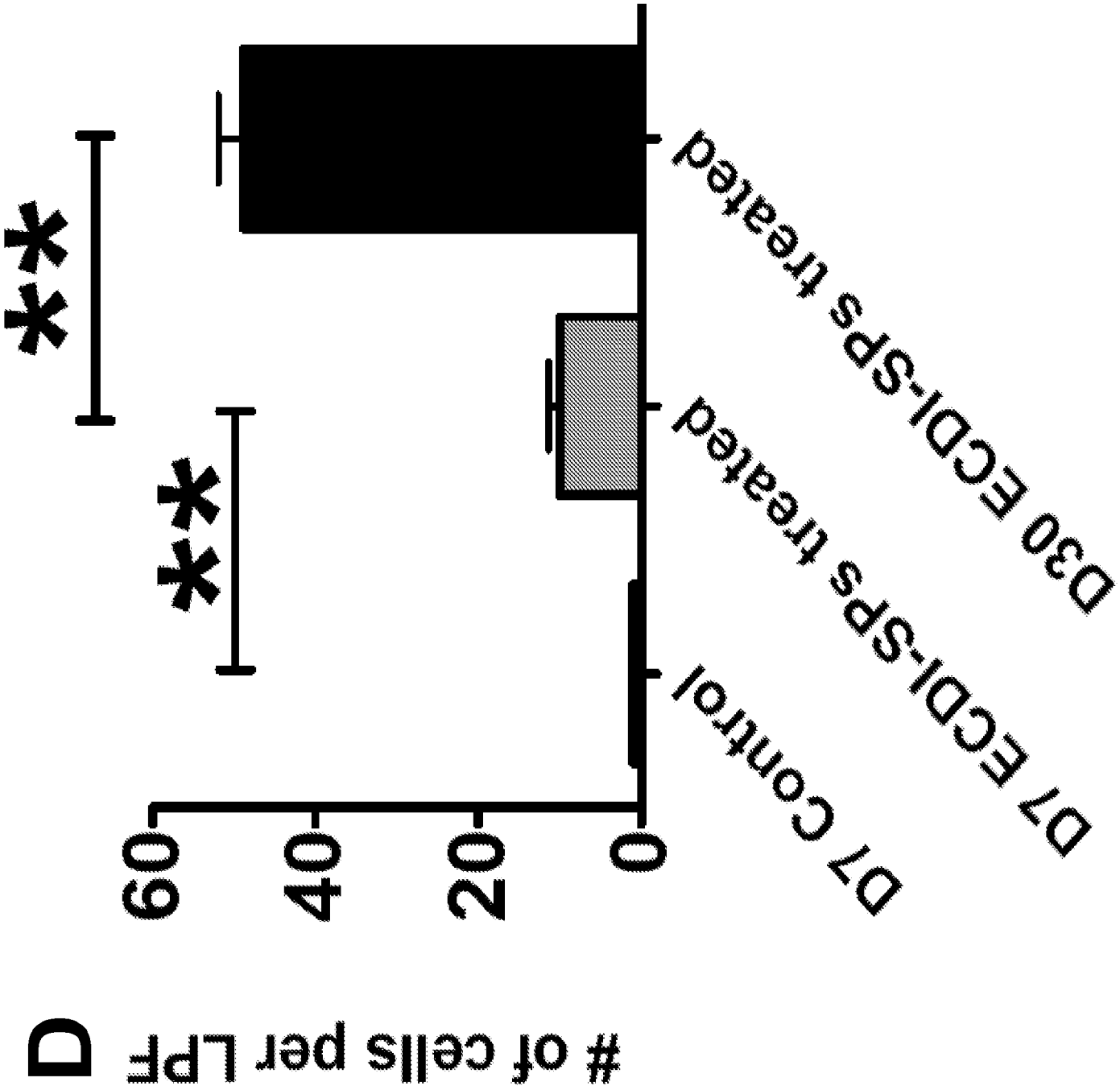

D
# of cells per LPF
60
40
20
0
D7 Control
D7 ECDI-SPS treated
D30 ECDI-SPS treated
**
**

FIGURE 11D

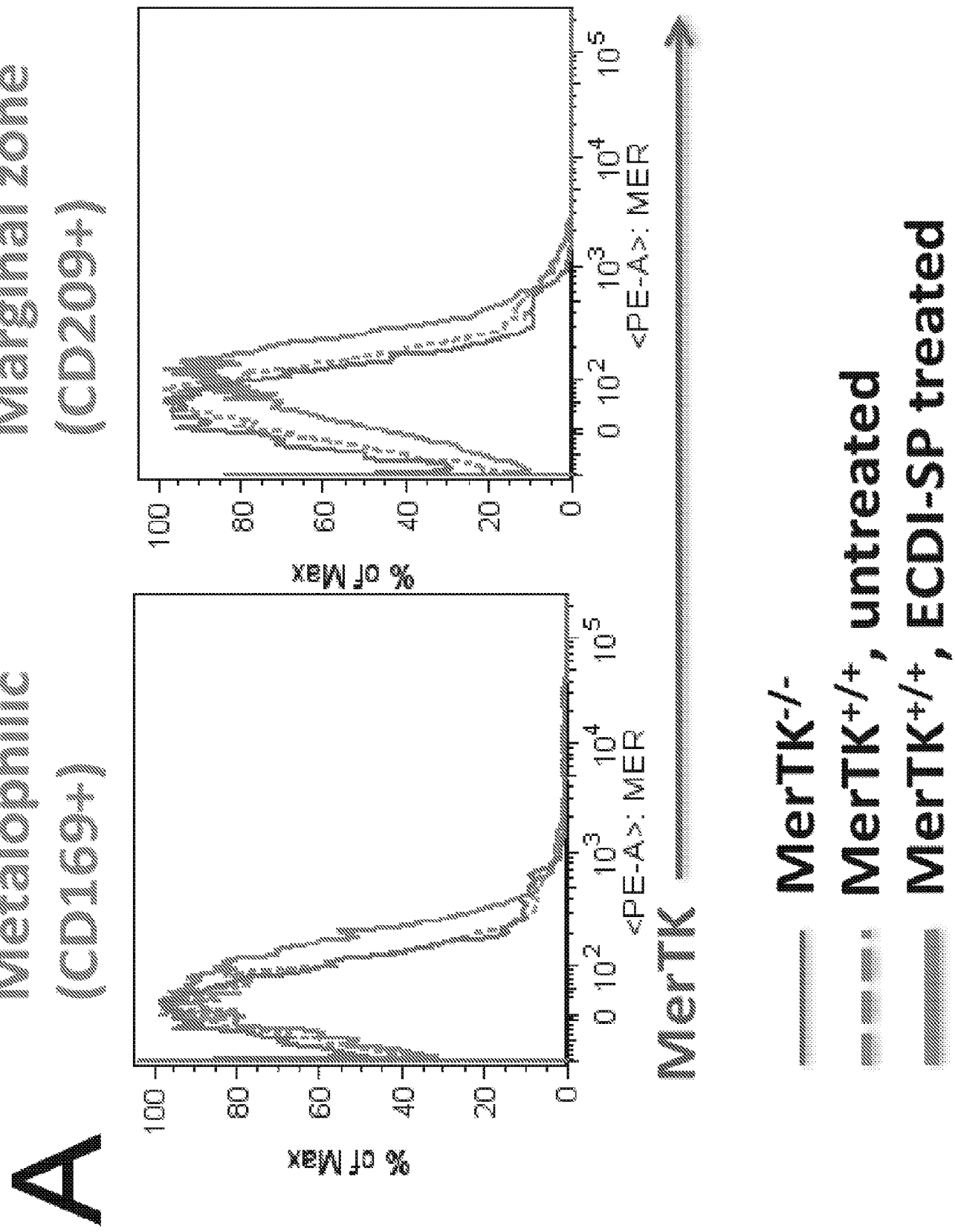

A
Metallophilic
(CD169+)
Marginal zone
(CD209+)
% of Max
<PE-A>: MER
MerTK
MerTK-/-
MerTK+/+, untreated
MerTK+/+, ECDI-SP treated

FIGURE 12A

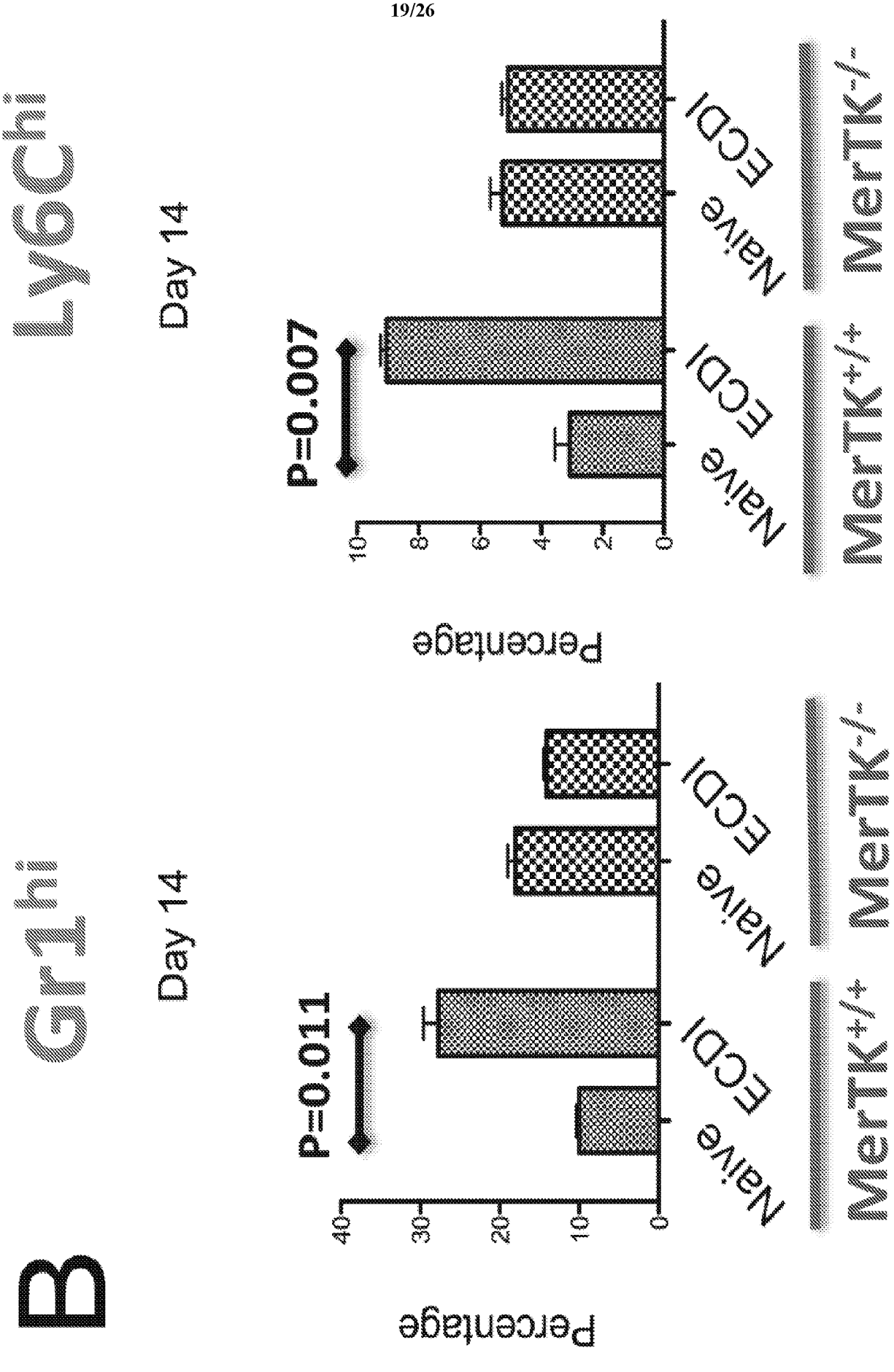

B
Gr1hi
Day 14
Percentage
40
30
20
10
0
P=0.011
Naïve
ECDI
Naïve
ECDI
MerTK+/+
MerTK-/-
Ly6Chi
Day 14
Percentage
10
8
6
4
2
0
P=0.007
Naïve
ECDI
Naïve
ECDI
MerTK+/+
MerTK-/-

FIGURE 12B

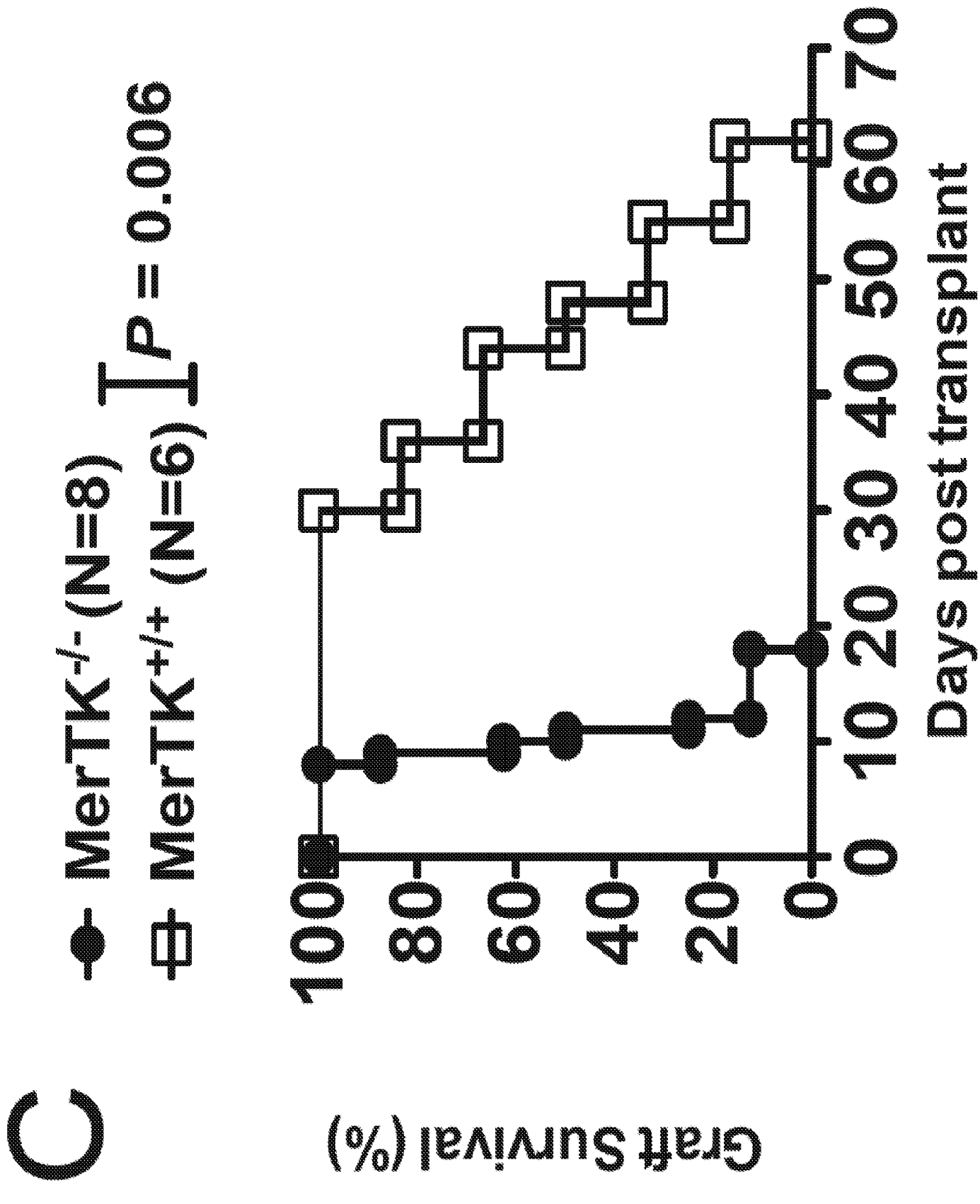

C
MerTK-/- (N=8)
MerTK+/+ (N=6)
P = 0.006
Graft Survival (%)
100
80
60
40
20
0
0 10 20 30 40 50 60 70
Days post transplant

FIGURE 12C

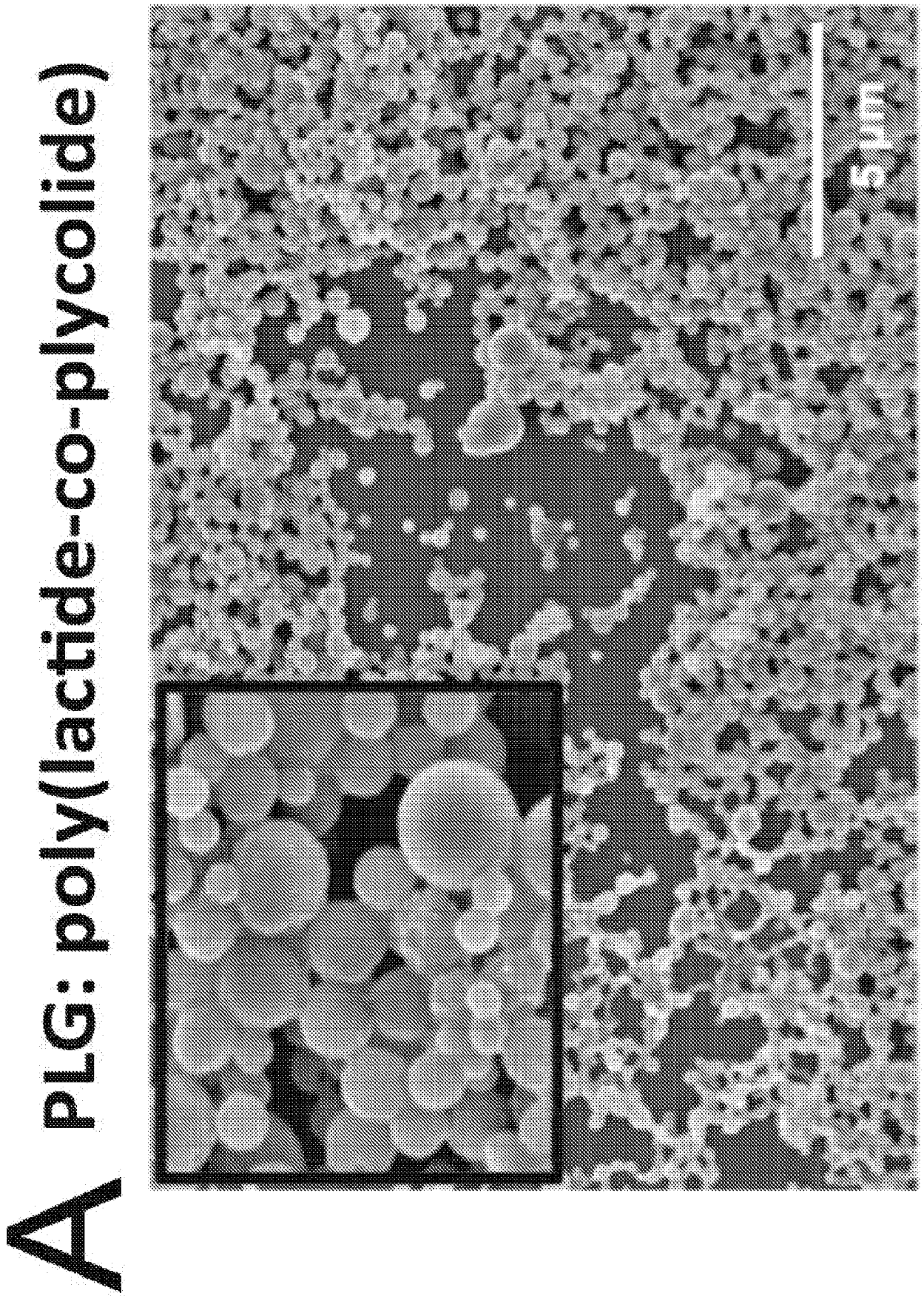
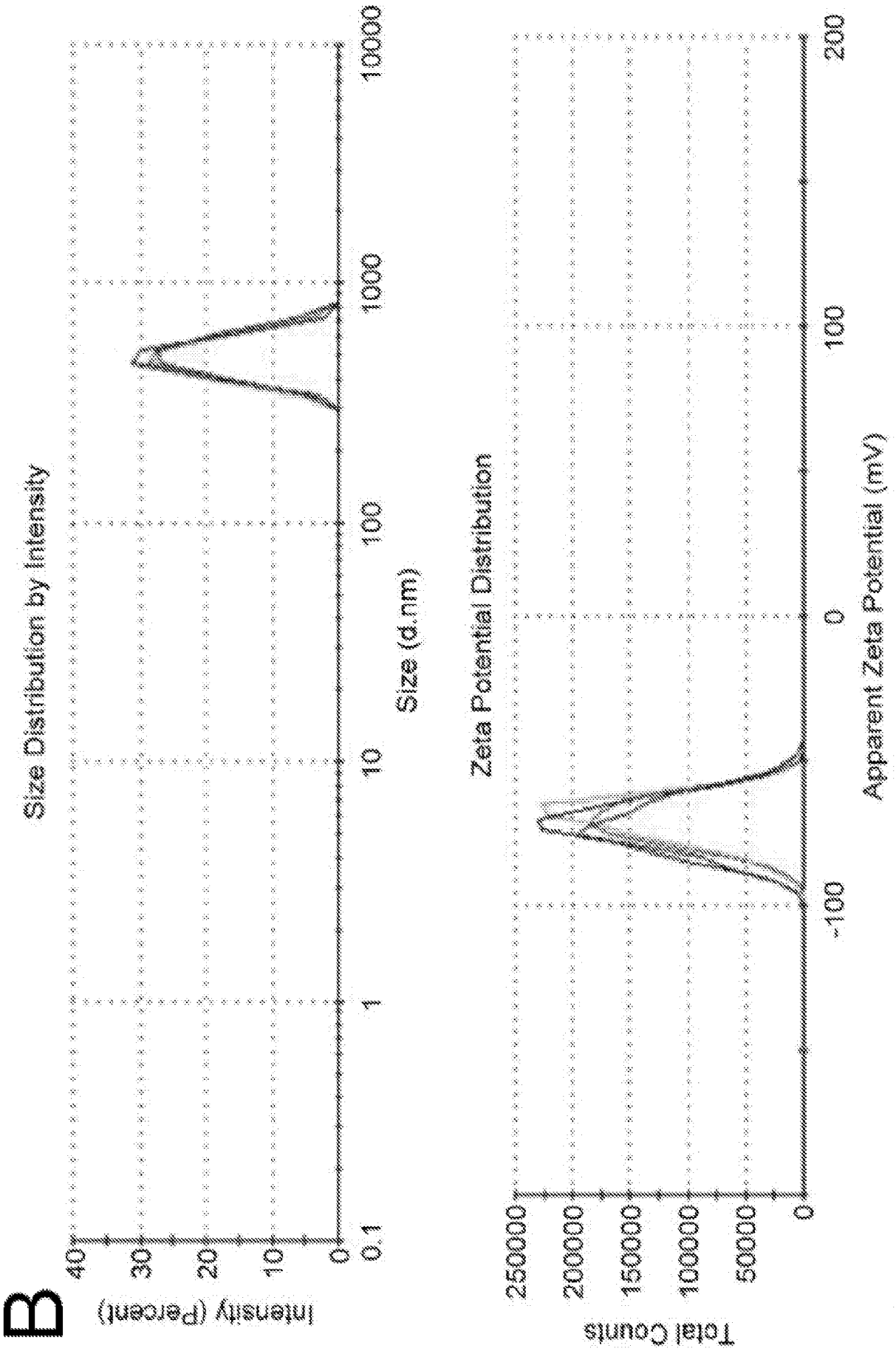


FIGURE 13A

FIGURE 13B


B
Size Distribution by Intensity
Intensity (Percent)
Size (d.nm)
Zeta Potential Distribution
Total Counts
Apparent Zeta Potential (mV)

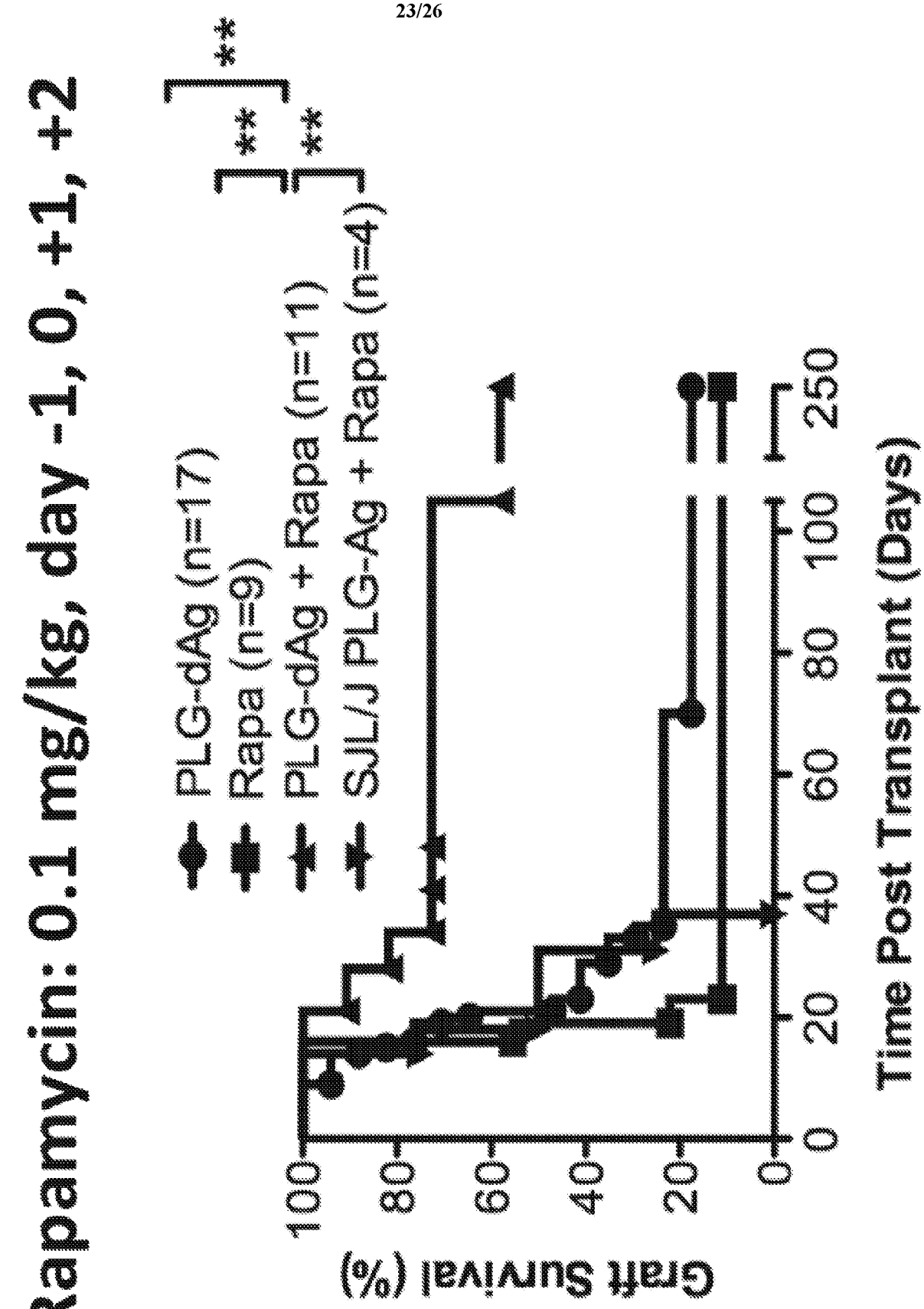

C Rapamycin: 0.1 mg/kg, day -1, 0, +1, +2
PLG-dAg (n=17)
Rapa (n=9)
PLG-dAg + Rapa (n=11)
SJL/J PLG-Ag + Rapa (n=4)
**
**
**
Graft Survival (%)
100
80
60
40
20
0
0
20
40
60
80
100
250
Time Post Transplant (Days)

FIGURE 13C

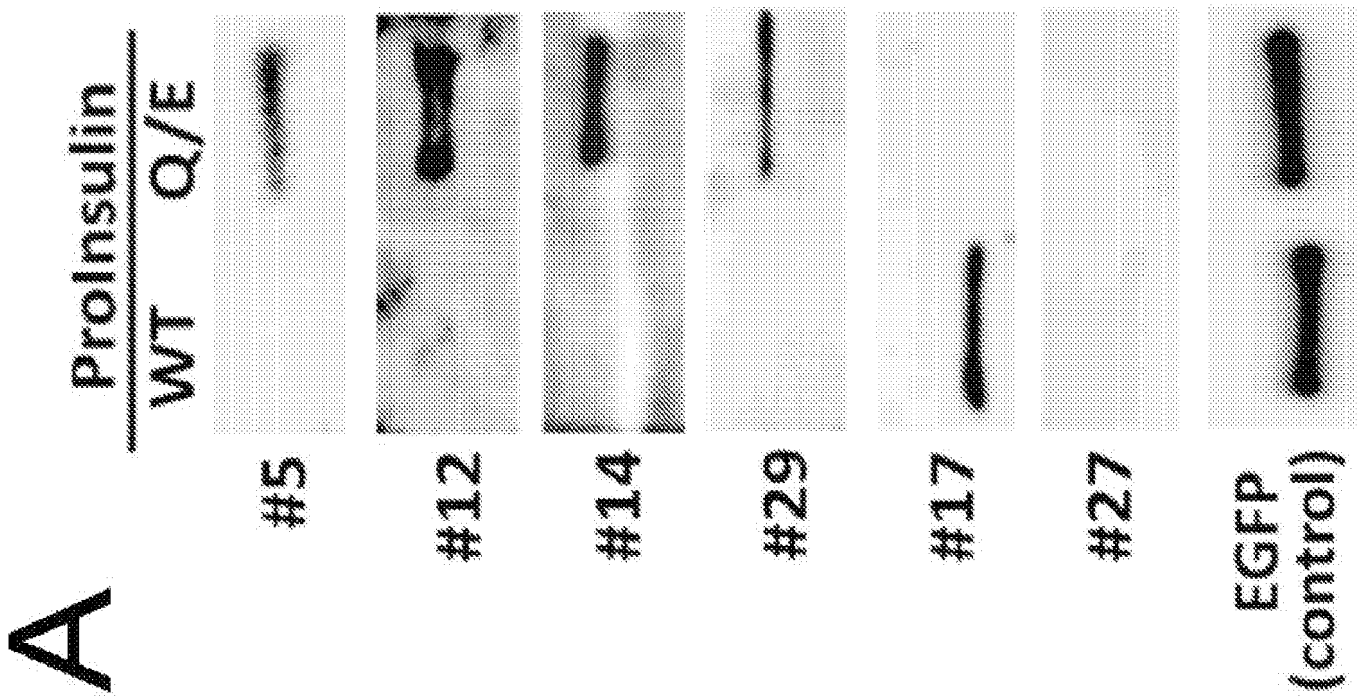


FIGURE 14A

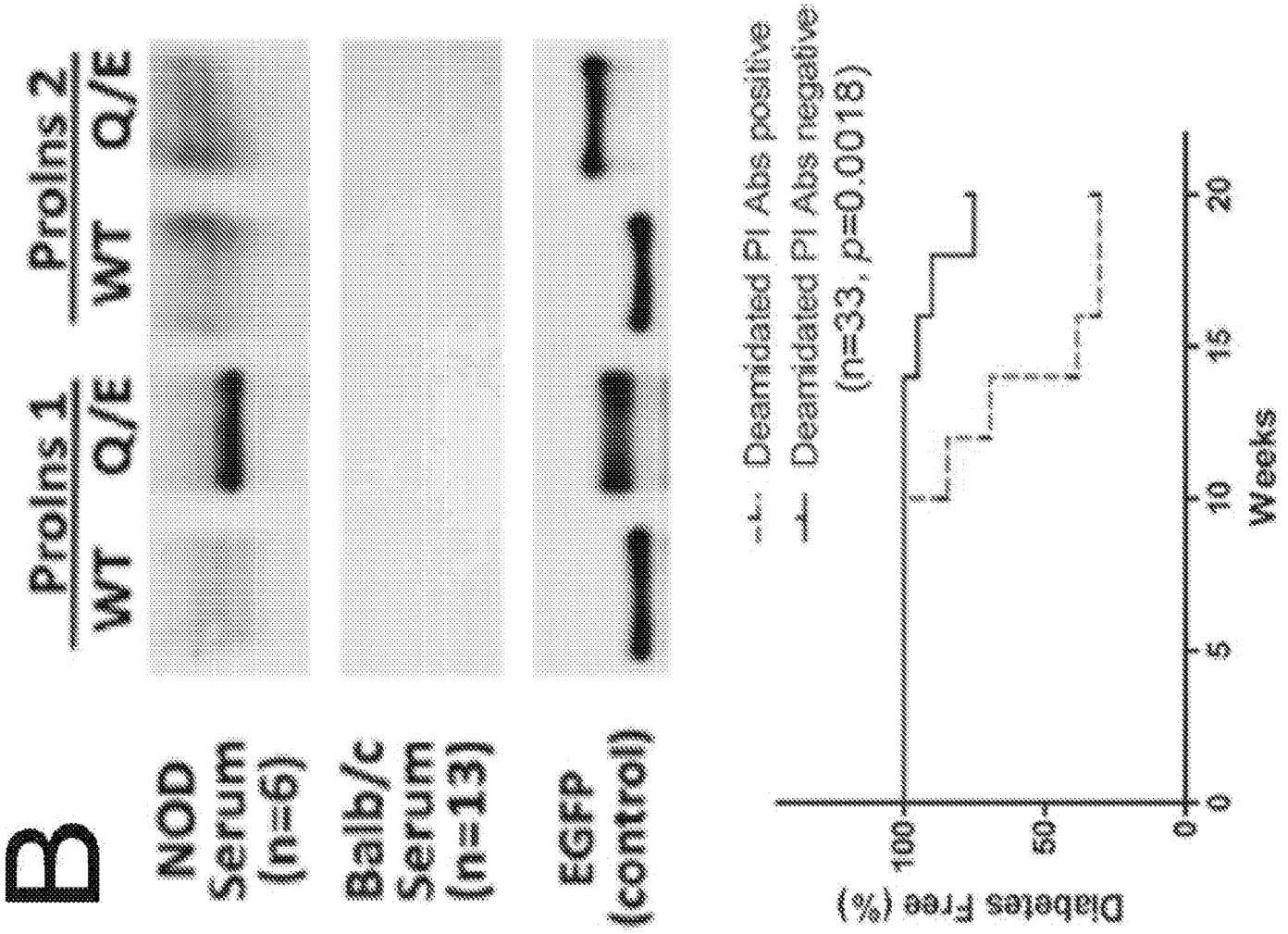
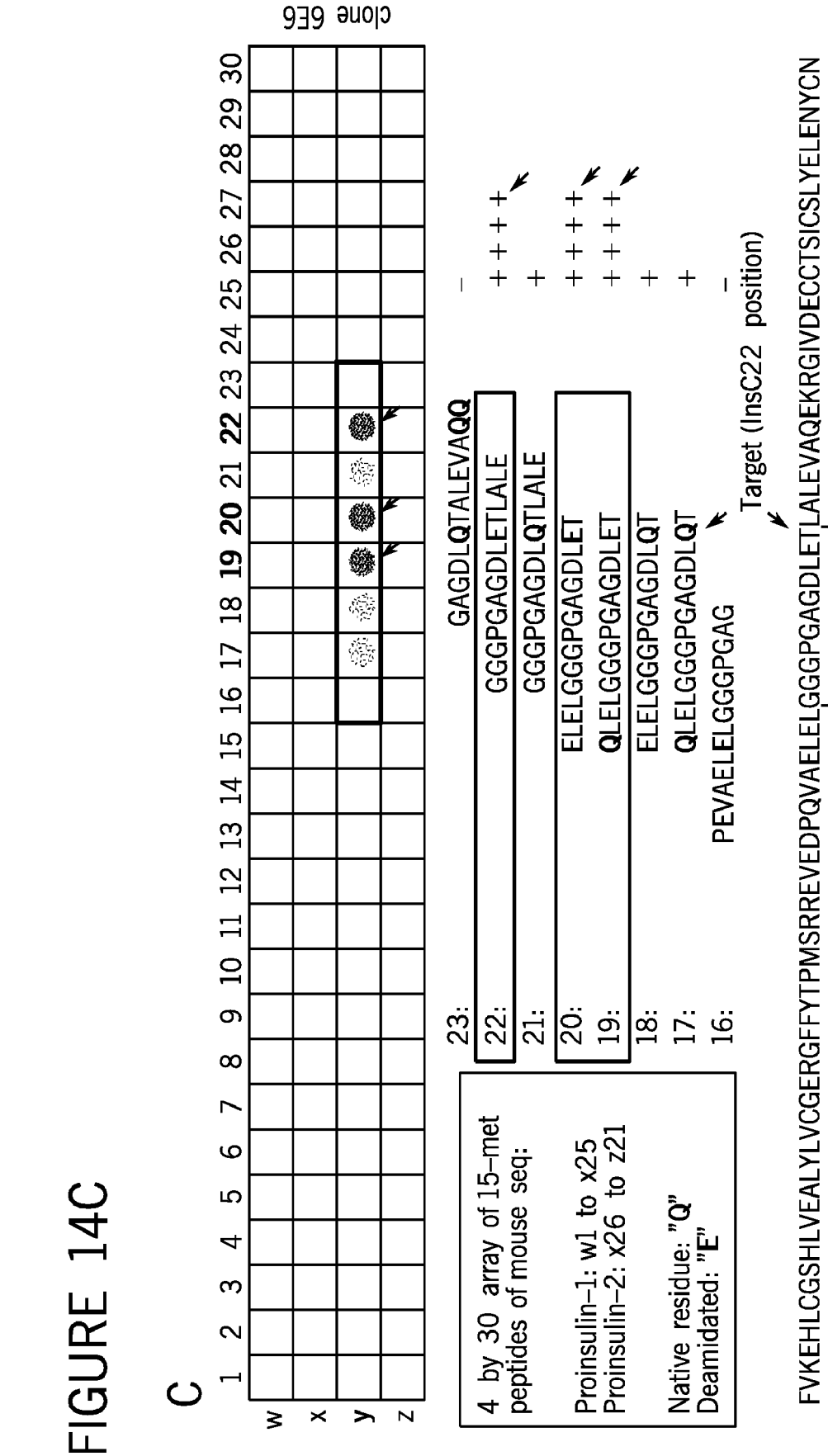

B
Proins 1
WT Q/E
Proins 2
WT Q/E
NOD Serum (n=6)
Balb/c Serum (n=13)
EGFP (control)
Deamidated PI Abs positive
Deamidated PI Abs negative
(n=33, p=0.0018)
Diabetes Free (%)
100
50
0
0
5
10
15
20
Weeks

FIGURE 14B

FIGURE 14C


C
clone 6E6
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30
w
x
y
z
4 by 30 array of 15–met peptides of mouse seq:
Proinsulin–1: w1 to x25
Proinsulin–2: x26 to z21
Native residue: "Q"
Deamidated: "E"
23:
22: GGGPGAGDLETLALE
21: GGGPGAGDLQTLALE
20: ELELGGGPGAGDLET
19: QLELGGGPGAGDLET
18: ELELGGGPGAGDLQT
17: QLELGGGPGAGDLQT
16: PEVAELELGGGPGAG
GAGDLQTALEVAQQ
Target (InsC22 position)
FVKEHLCGSHLVEALYLVCGERGFFYTPMSRREVEDPQVAELELGGGPGAGDLETLALEVAQEKRGIVDECCTSICSLYELENYCN
B–chain
C–peptide
epitope
A–chain