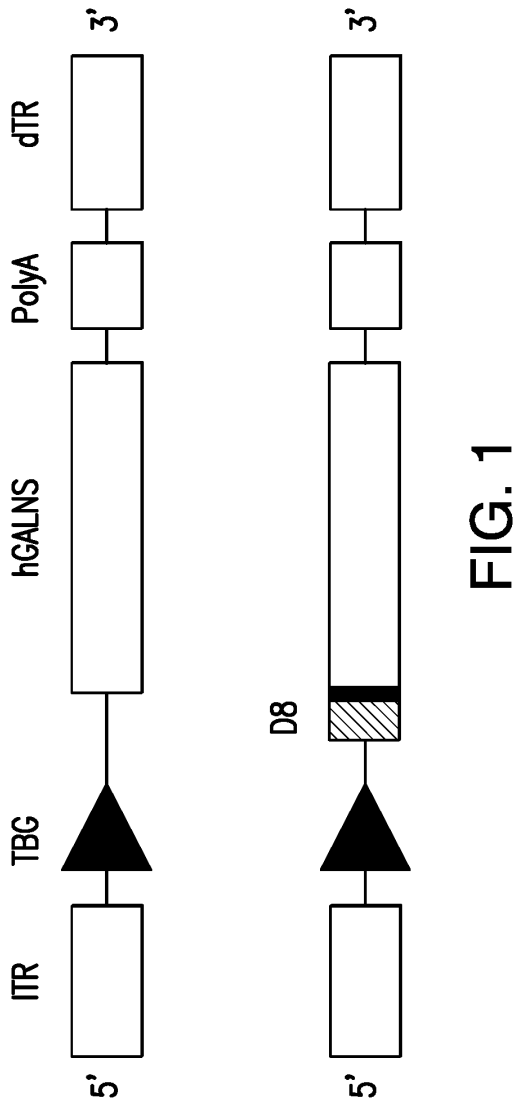


FIG. 1

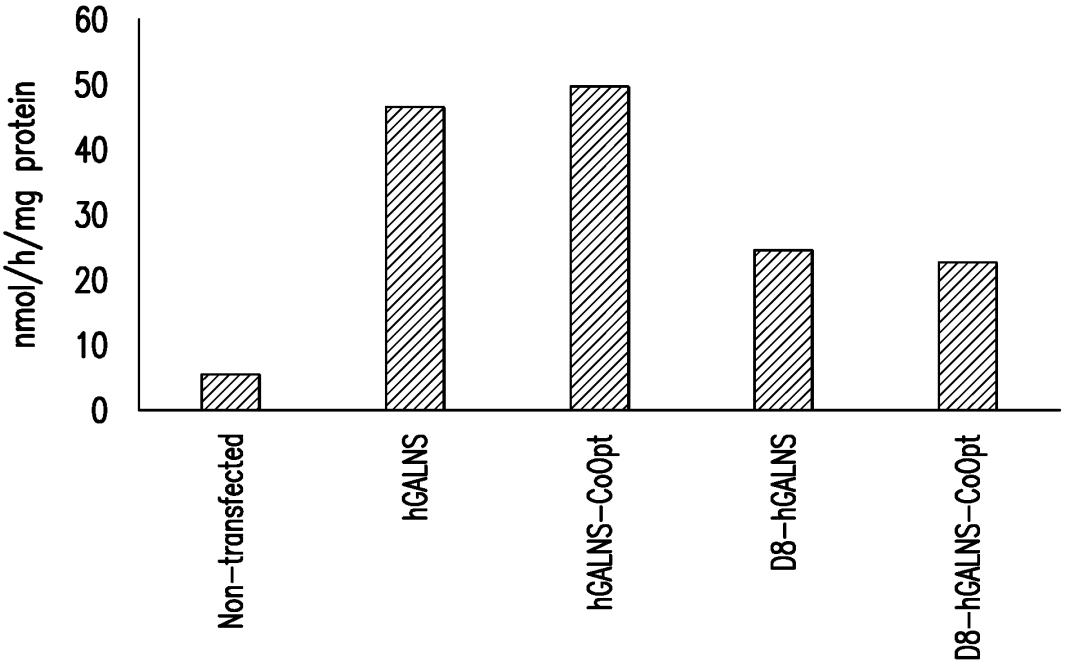

nmol/h/mg protein
60
50
40
30
20
10
0
Non-transfected
hGALNS
hGALNS-CoOpt
D8-hGALNS
D8-hGALNS-CoOpt

FIG. 2A

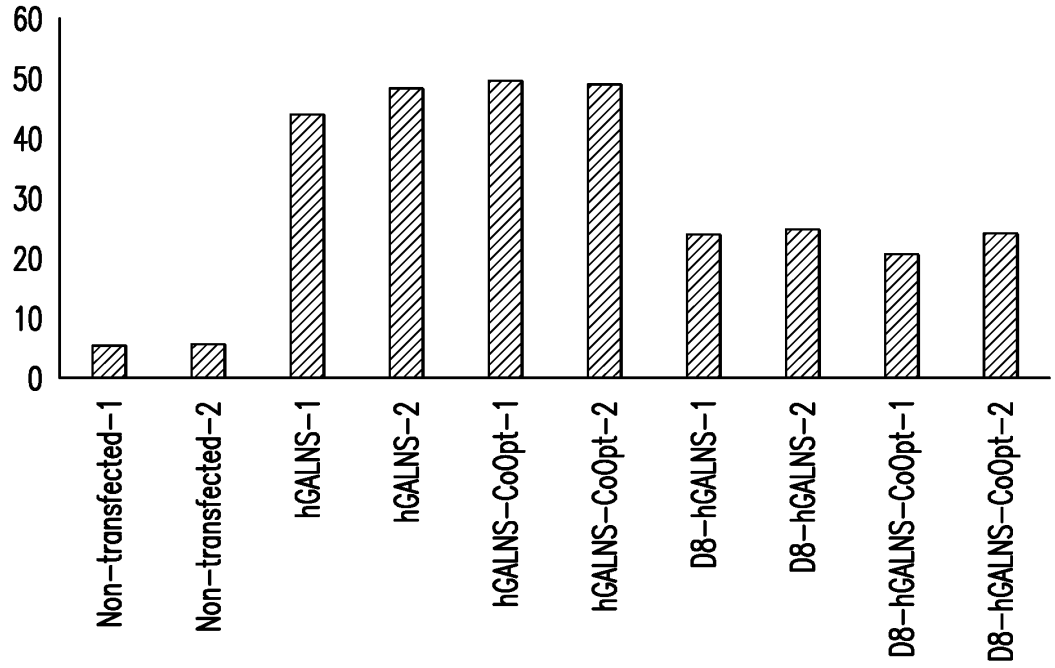

60
50
40
30
20
10
0
Non−transfected−1
Non−transfected−2
hGALNS−1
hGALNS−2
hGALNS−CoOpt−1
hGALNS−CoOpt−2
D8−hGALNS−1
D8−hGALNS−2
D8−hGALNS−CoOpt−1
D8−hGALNS−CoOpt−2

FIG. 2B

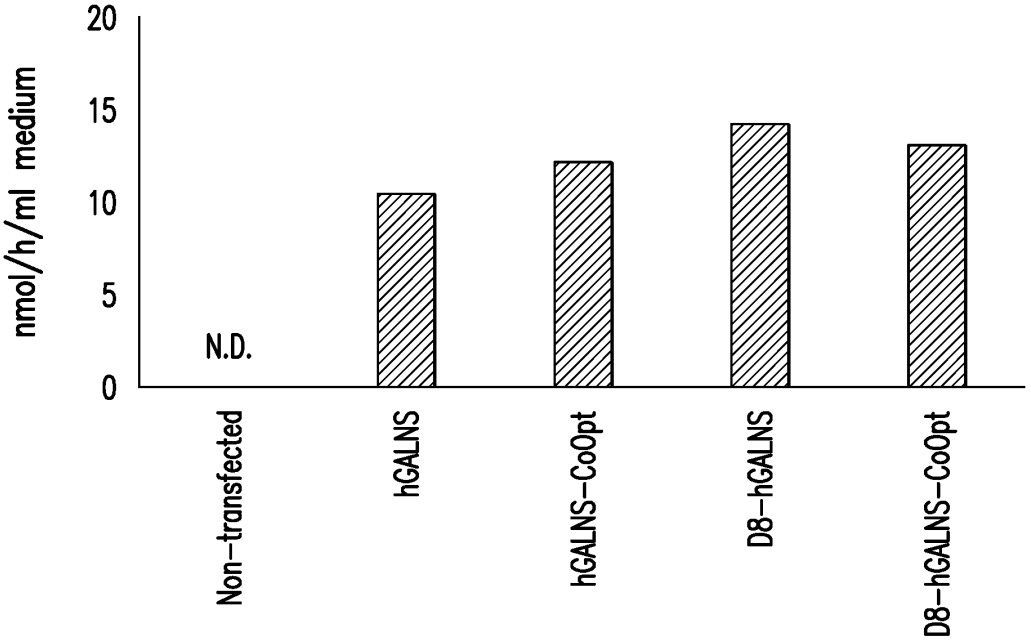

20
15
10
5
0
nmol/h/ml medium
N.D.
Non-transfected
hGALNS
hGALNS-CoOpt
D8-hGALNS
D8-hGALNS-CoOpt

FIG. 2C

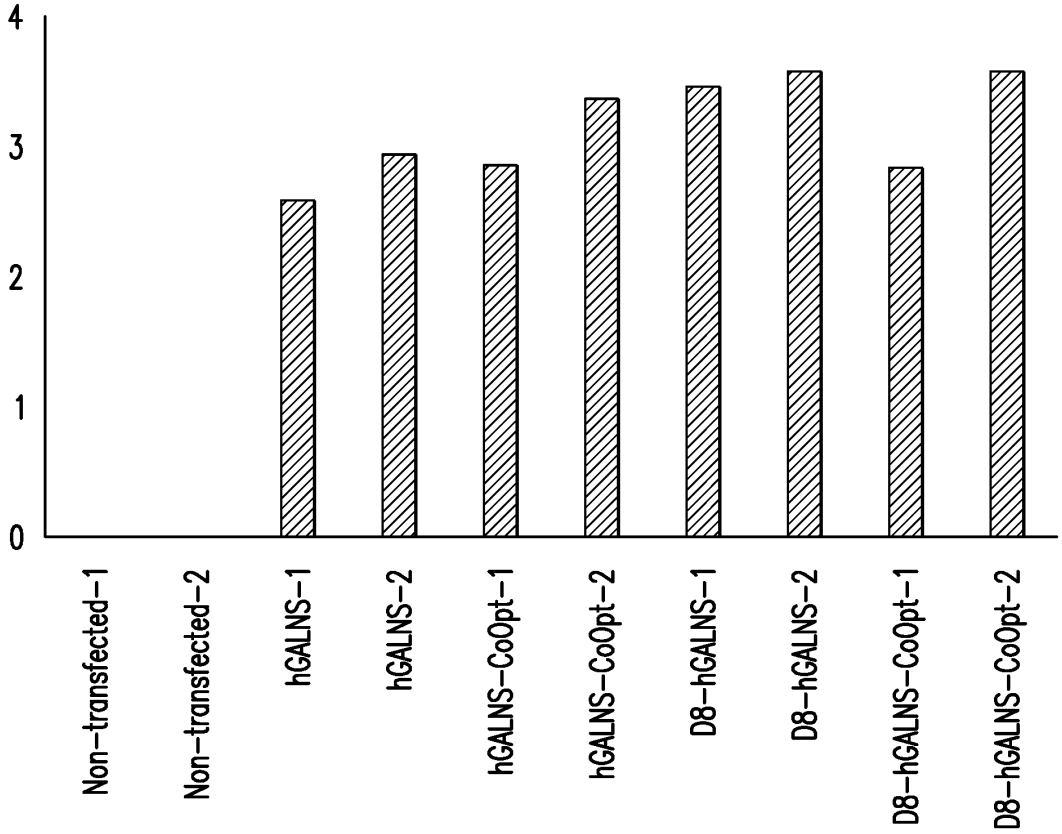

4
3
2
1
0
Non−transfected−1
Non−transfected−2
hGALNS−1
hGALNS−2
hGALNS−CoOpt−1
hGALNS−CoOpt−2
D8−hGALNS−1
D8−hGALNS−2
D8−hGALNS−CoOpt−1
D8−hGALNS−CoOpt−2

FIG. 2D

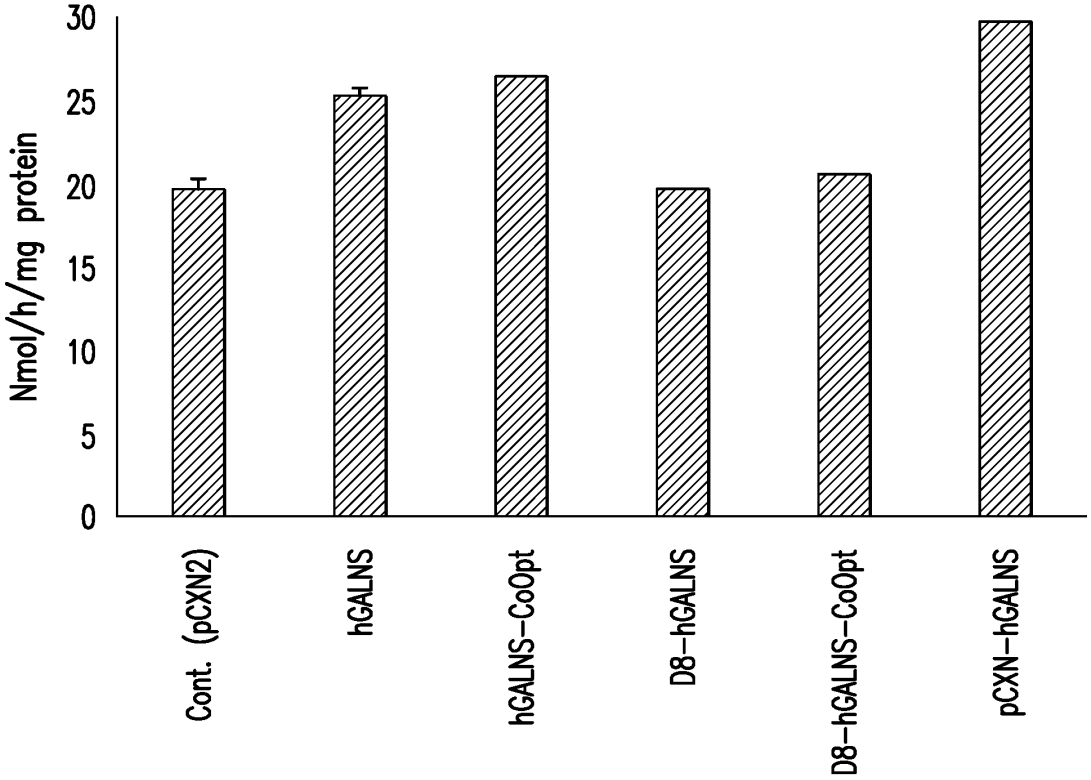

Nmol/h/mg protein
30
25
20
15
10
5
0
Cont. (pCXN2)
hGALNS
hGALNS-CoOpt
D8-hGALNS
D8-hGALNS-CoOpt
pCXN-hGALNS

FIG. 3

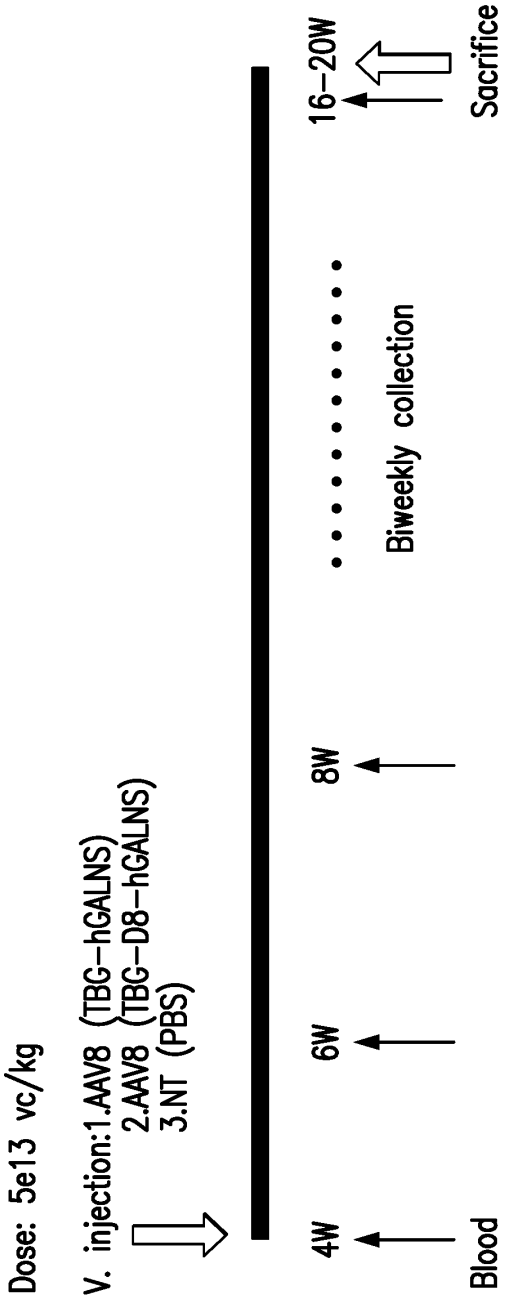

Dose: 5e13 vc/kg
V. injection:1.AAV8 (TBG-hGALNS)
2.AAV8 (TBG-D8-hGALNS)
3.NT (PBS)
4W
6W
8W
16-20W
Blood
Biweekly collection
Sacrifice
Biomarker; Enzyme activity and KS levels in Blood - Biweekly

FIG. 4

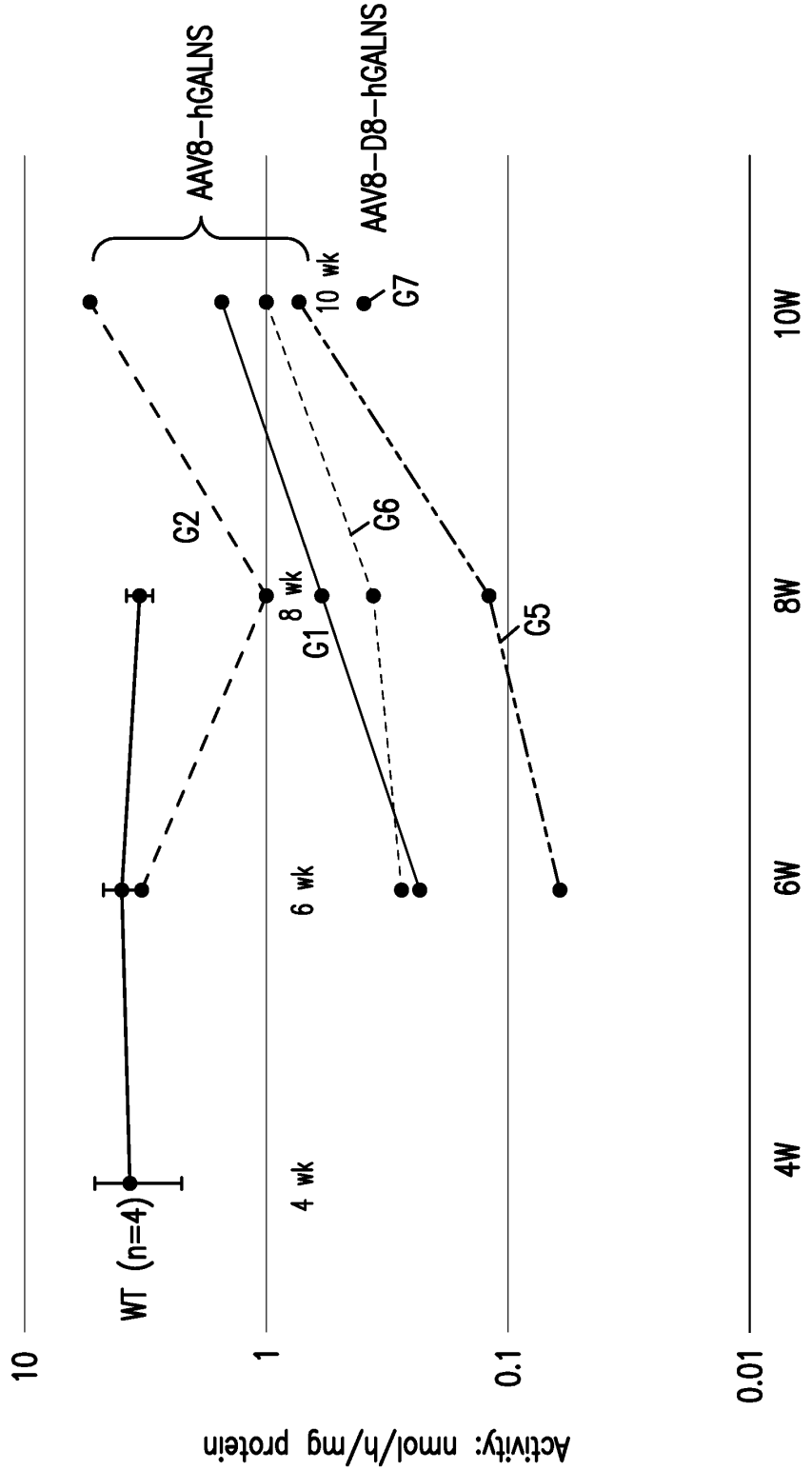

AAV8–hGALNS
AAV8–D8–hGALNS
10 wk
G7
G2
G6
8 wk
G1
G5
6 wk
4 wk
WT (n=4)
10
1
0.1
0.01
4W
6W
8W
10W
Activity: nmol/h/mg protein

FIG. 5A

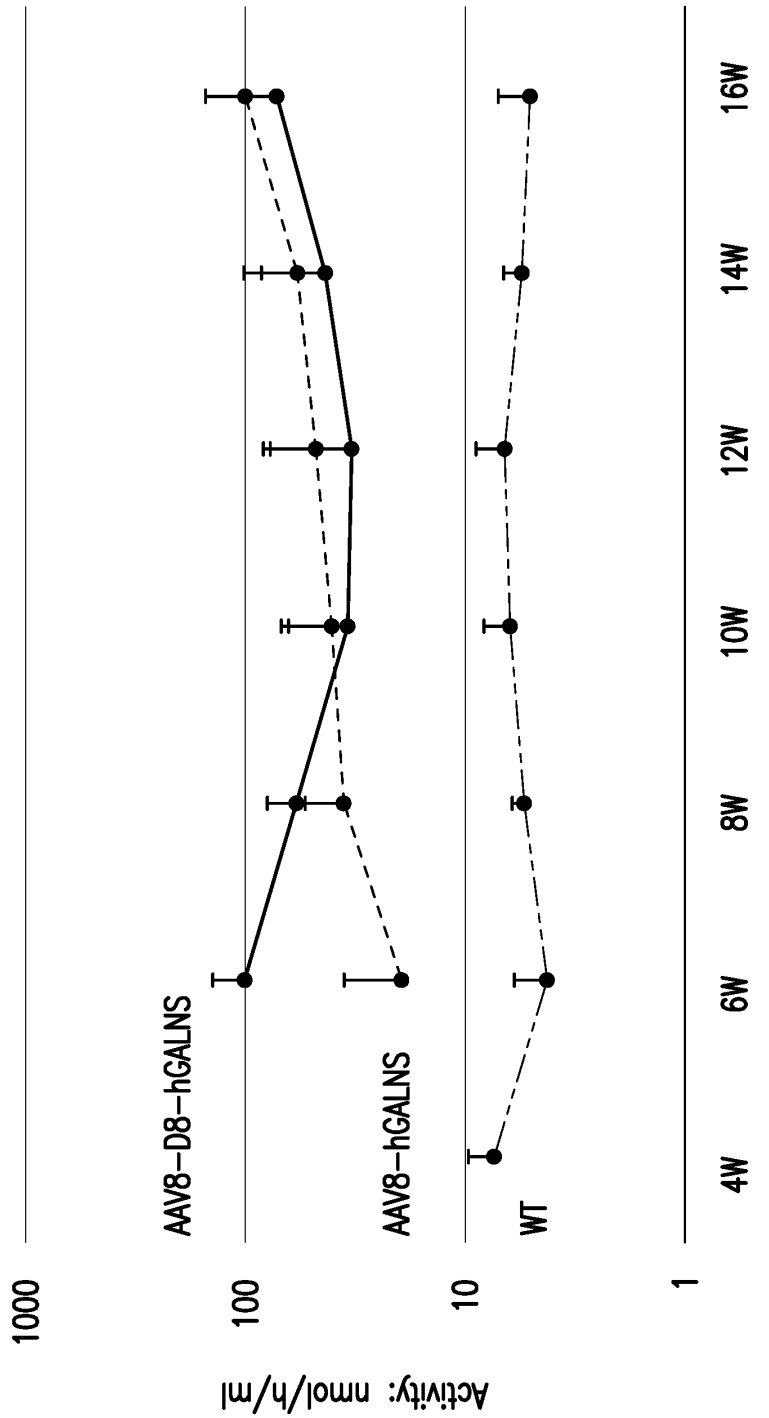

Activity: nmol/h/ml
1000
100
10
1
AAV8-D8-hGALNS
AAV8-hGALNS
WT
4W
6W
8W
10W
12W
14W
16W

FIG. 5B

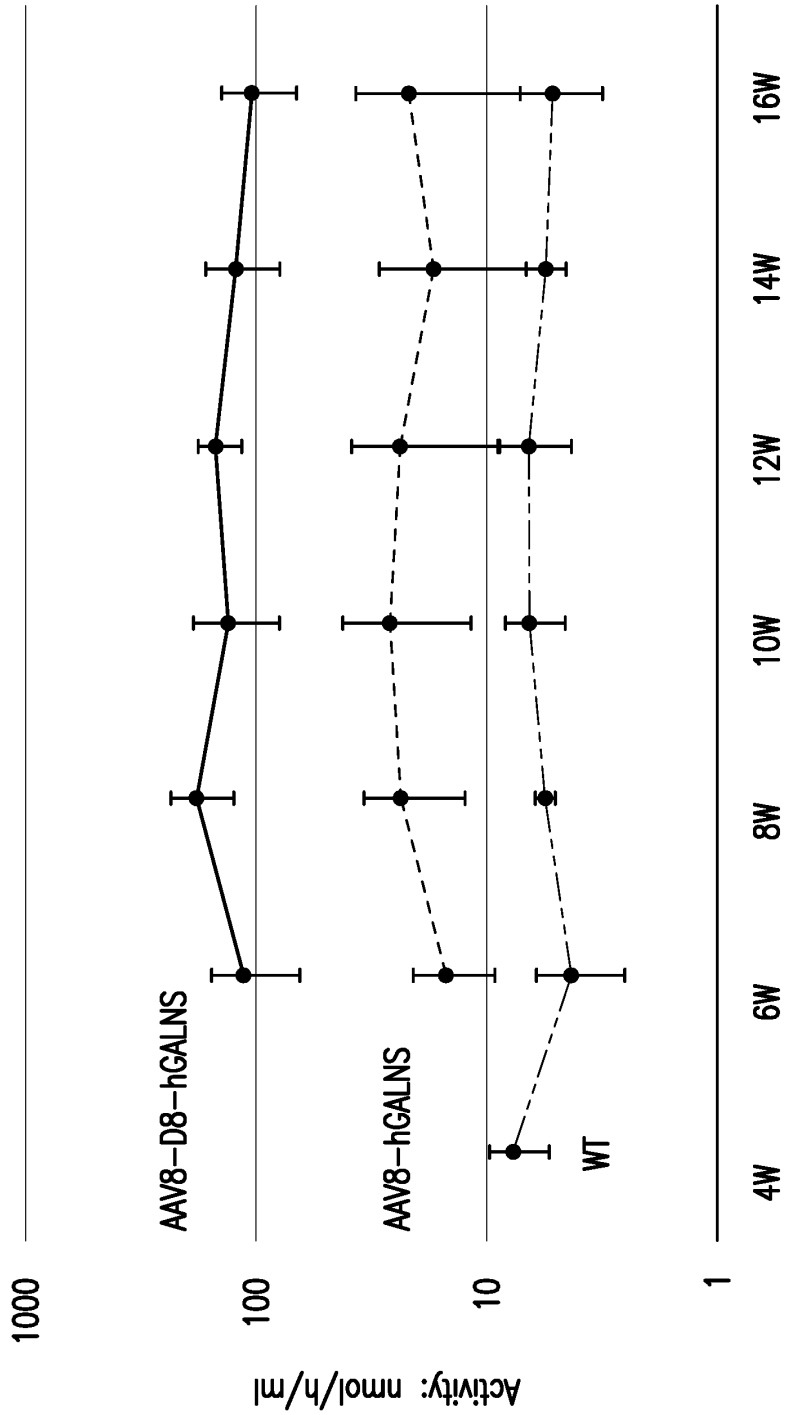

Activity: nmol/h/ml
1000
100
10
1
4W
6W
8W
10W
12W
14W
16W
AAV8-D8-hGALNS
AAV8-hGALNS
WT

FIG. 6

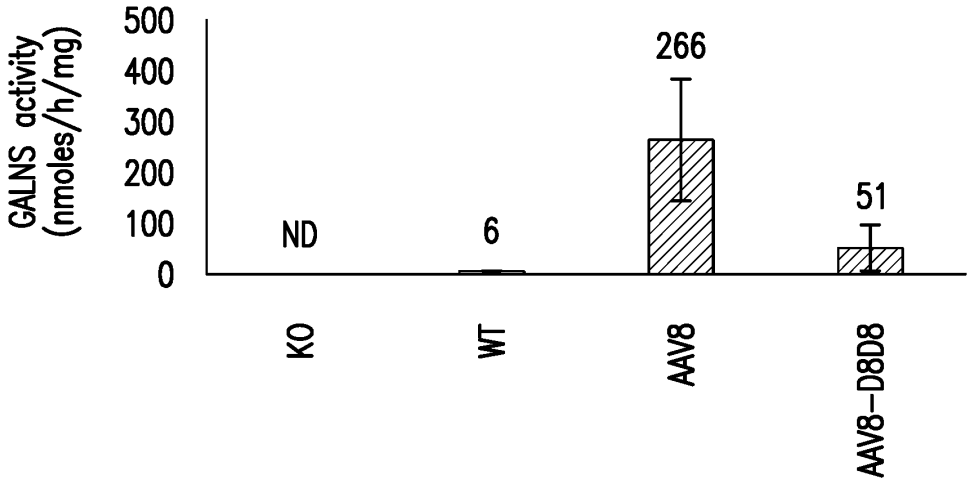

GALNS activity (nmoles/h/mg)
500
400
300
200
100
0
ND
6
266
51
KO
WT
AAV8
AAV8–D8D8

FIG. 7A

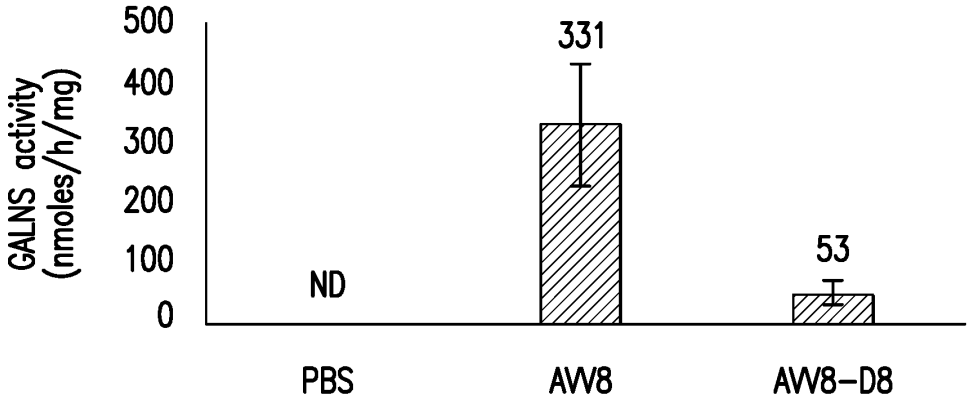

GALNS activity
(nmoles/h/mg)
500
400
300
200
100
0
ND
331
53
PBS
AW8
AW8-D8

FIG. 7B

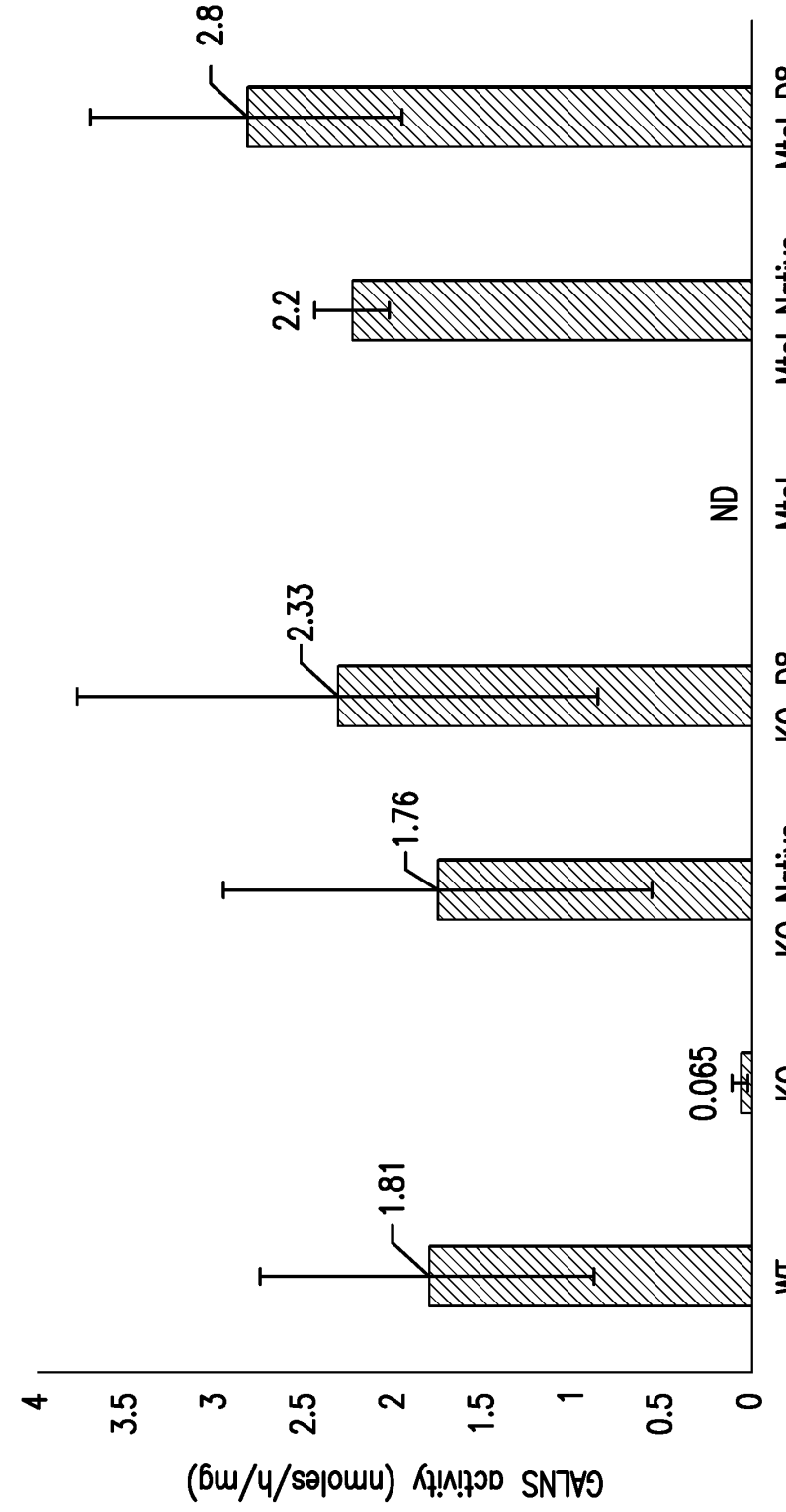

GALNS activity (nmoles/h/mg)
0
0.5
1
1.5
2
2.5
3
3.5
4
1.81
0.065
1.76
2.33
ND
2.2
2.8
WT
KO
KO Native GALNS
KO–D8
Mtol
Mtol Native GALNS
Mtol D8

FIG. 7C

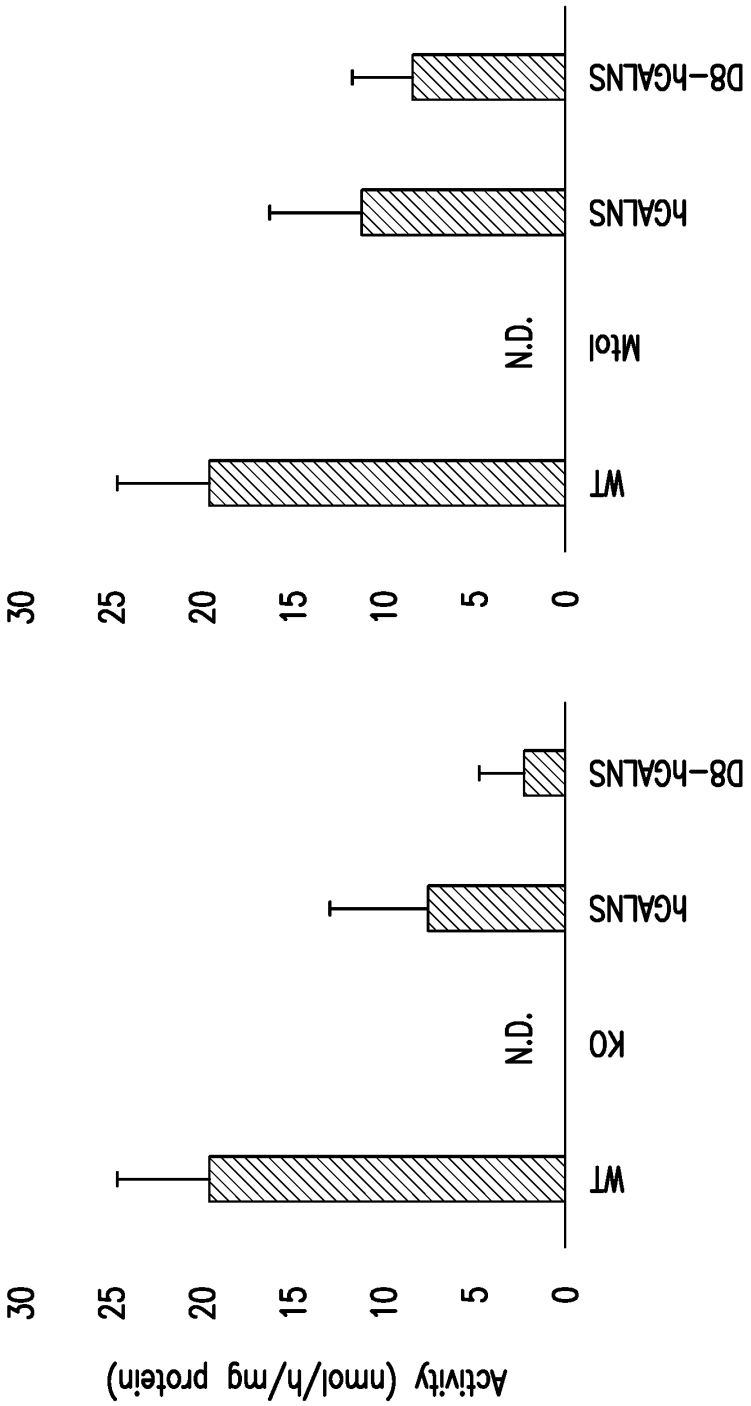

Activity (nmol/h/mg protein)
0
5
10
15
20
25
30
WT
N.D.
KO
hGALNS
Db-hGALNS
WT
N.D.
Mtol
hGALNS
Db-hGALNS

FIG. 7D

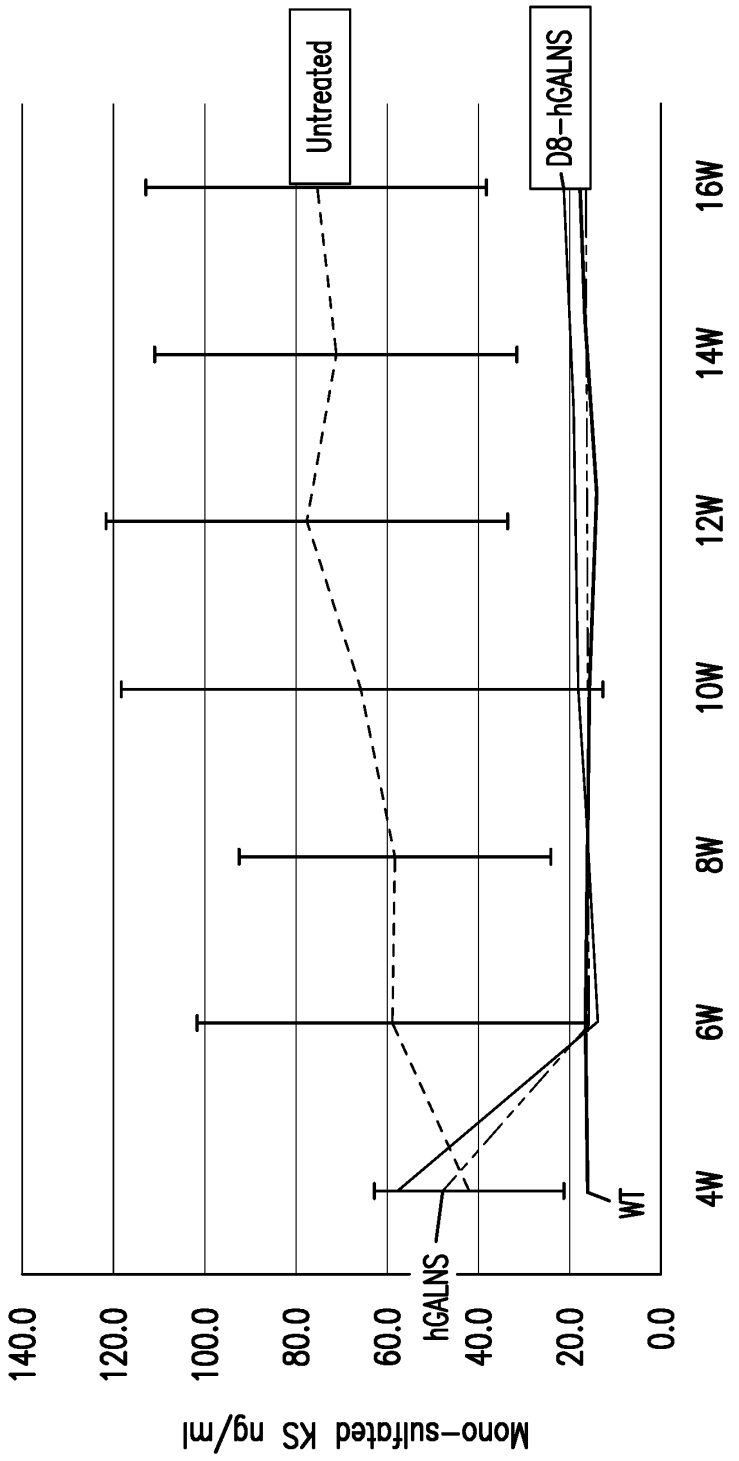

Untreated
D8-hGALNS
WT
hGALNS
4W
6W
8W
10W
12W
14W
16W
Weeks
140.0
120.0
100.0
80.0
60.0
40.0
20.0
0.0
Mono-sulfated KS ng/ml

FIG. 8

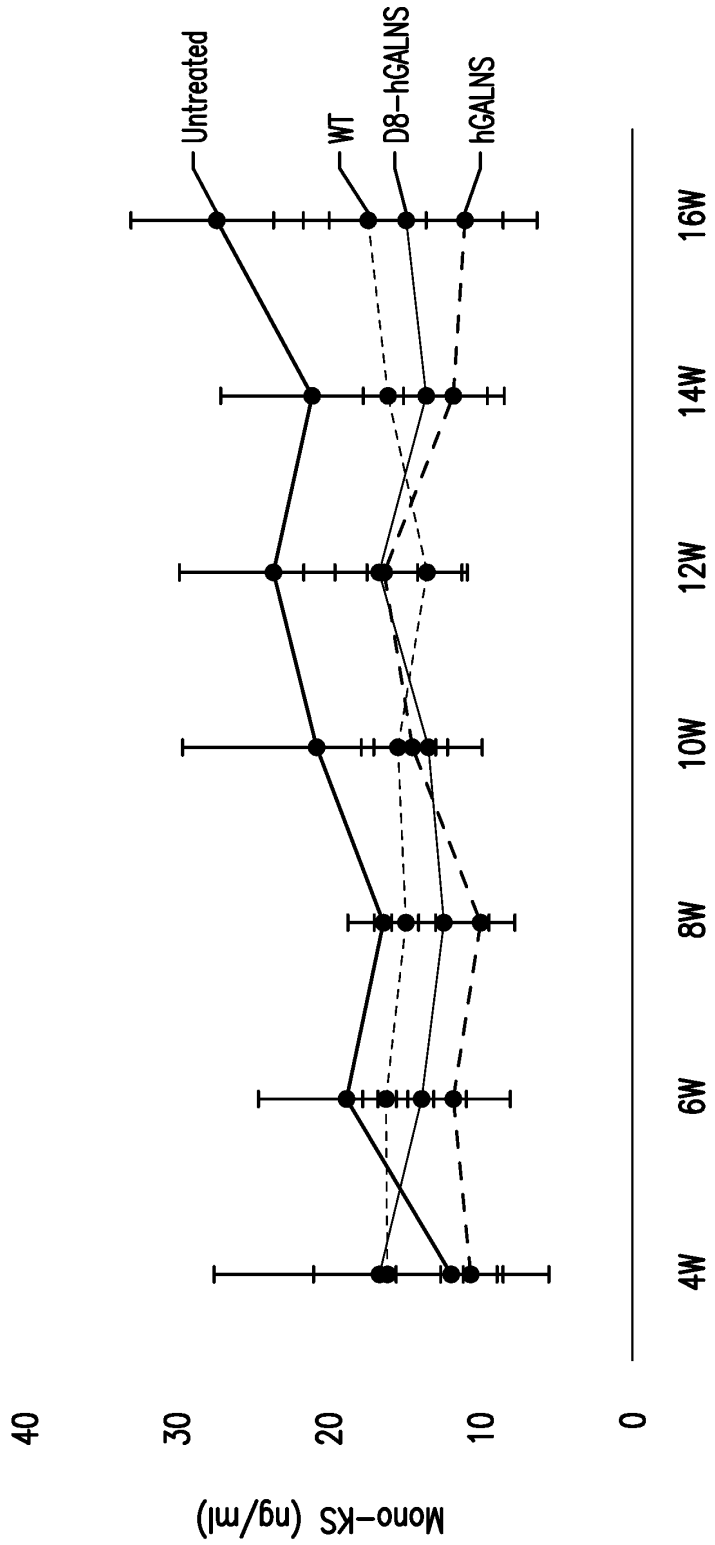

Untreated
WT
D8-hGALNS
hGALNS
4W
6W
8W
10W
12W
14W
16W
0
10
20
30
40
Mono-KS (ng/ml)

FIG. 9A

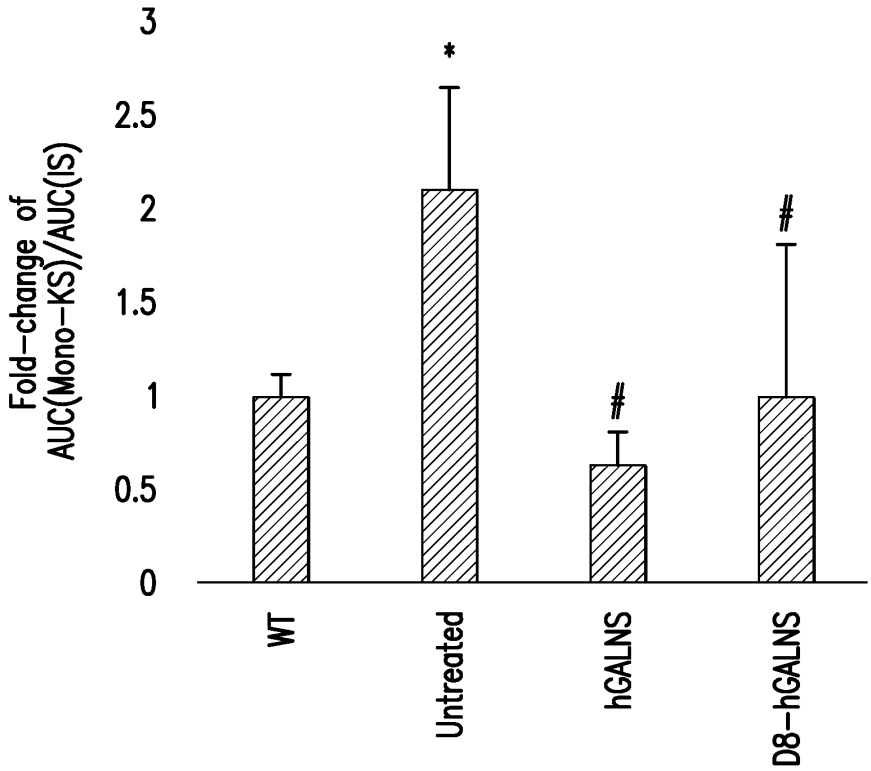

Fold-change of AUC(Mono-KS)/AUC(IS)
3
2.5
2
1.5
1
0.5
0
*
#
#
WT
Untreated
hGALNS
D8-hGALNS

FIG. 9B

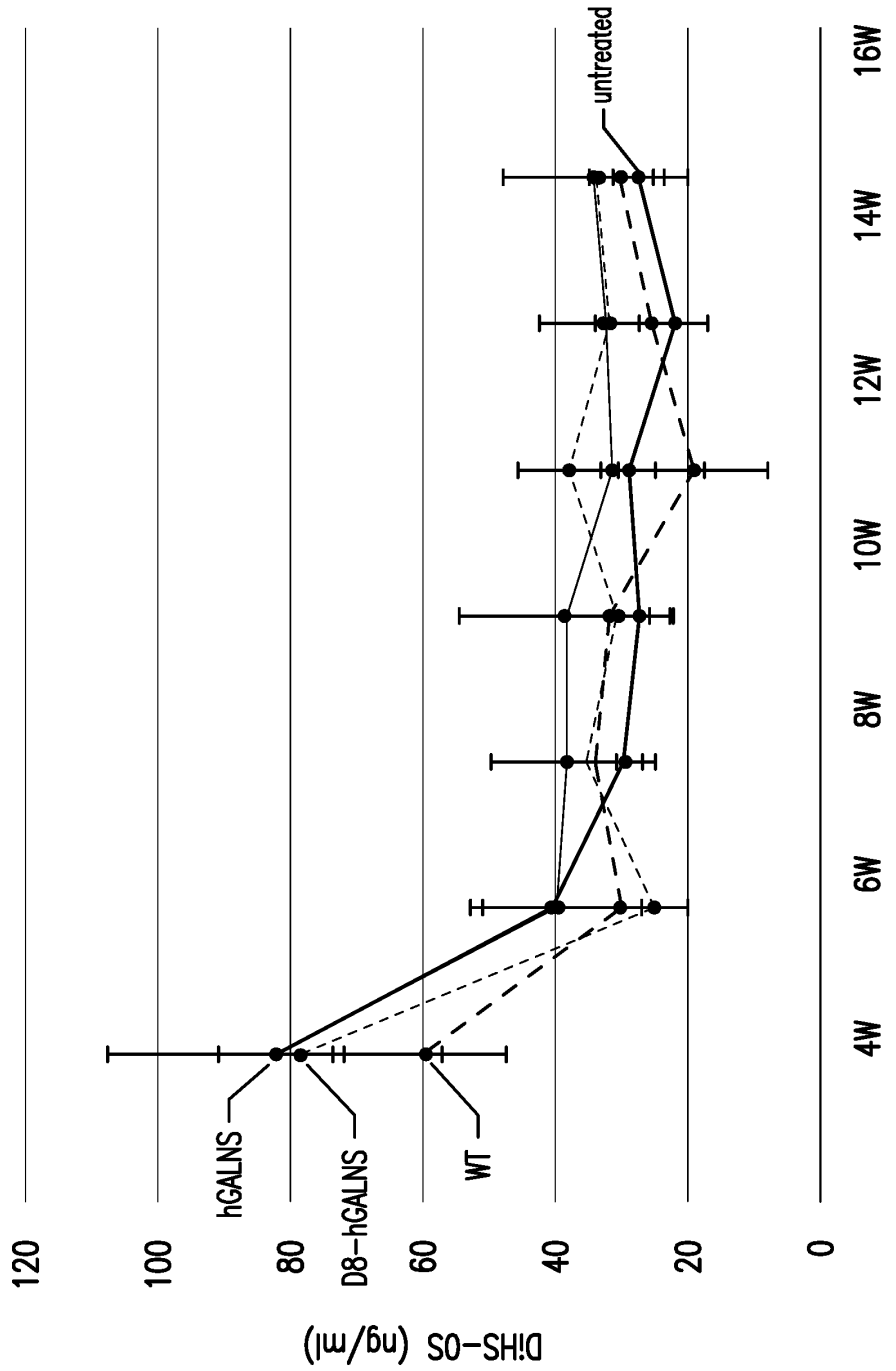

DIHS-OS (ng/ml)
120
100
80
60
40
20
0
4W
6W
8W
10W
12W
14W
16W
hGALNS
D8-hGALNS
WT
untreated

FIG. 10

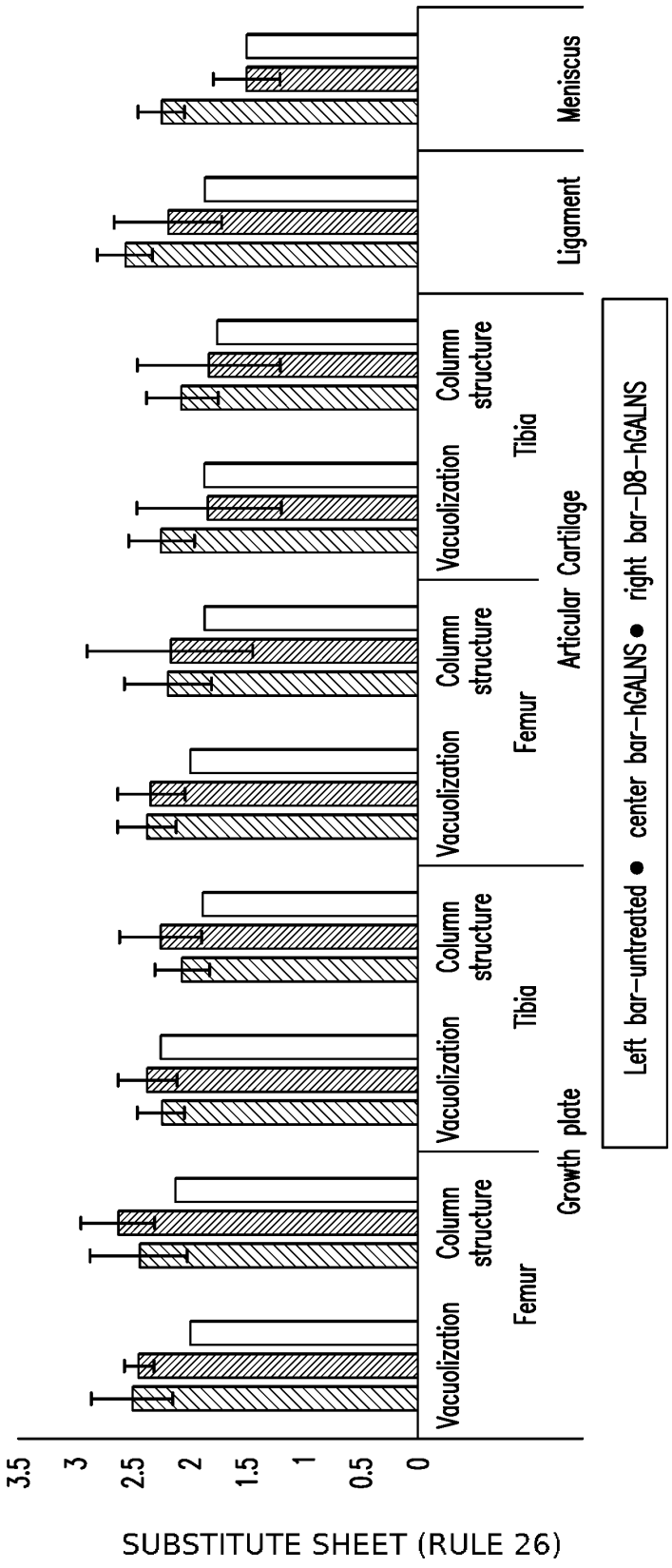

3.5
3
2.5
2
1.5
1
0.5
0
Vacuolization
Column structure
Femur
Growth plate
Vacuolization
Column structure
Tibia
Vacuolization
Column structure
Femur
Articular Cartilage
Vacuolization
Column structure
Tibia
Ligament
Meniscus
Left bar–untreated ● center bar–hGALNS ● right bar–D8–hGALNS

FIG. 11A

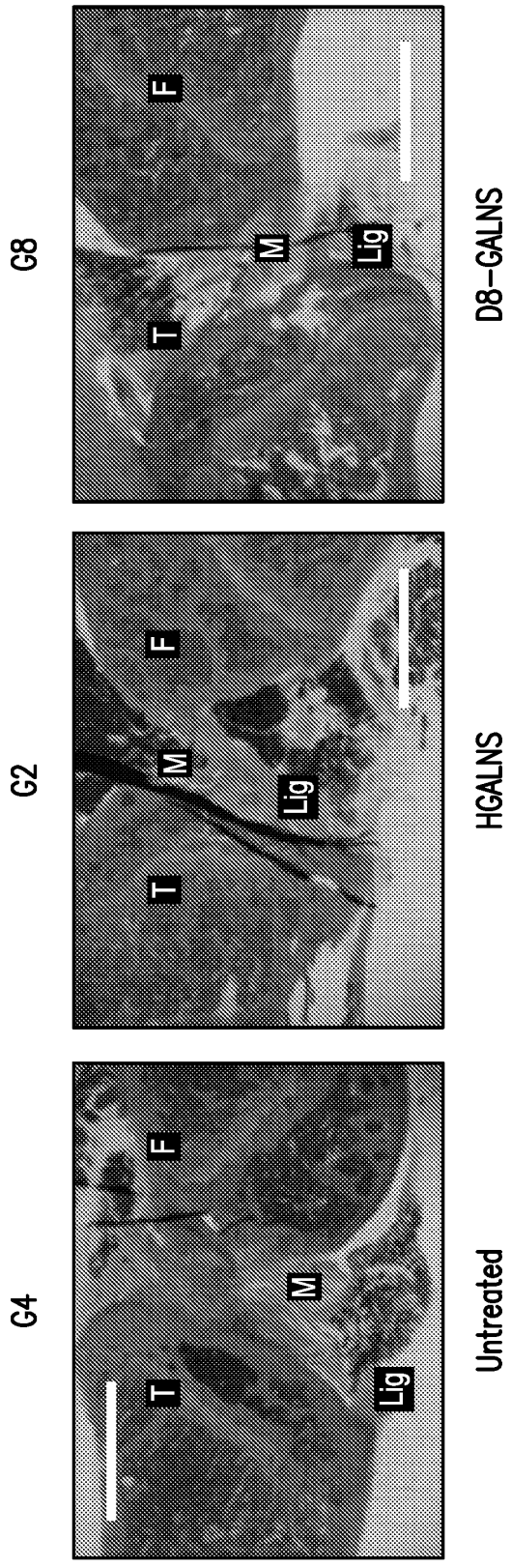


FIG. 11B

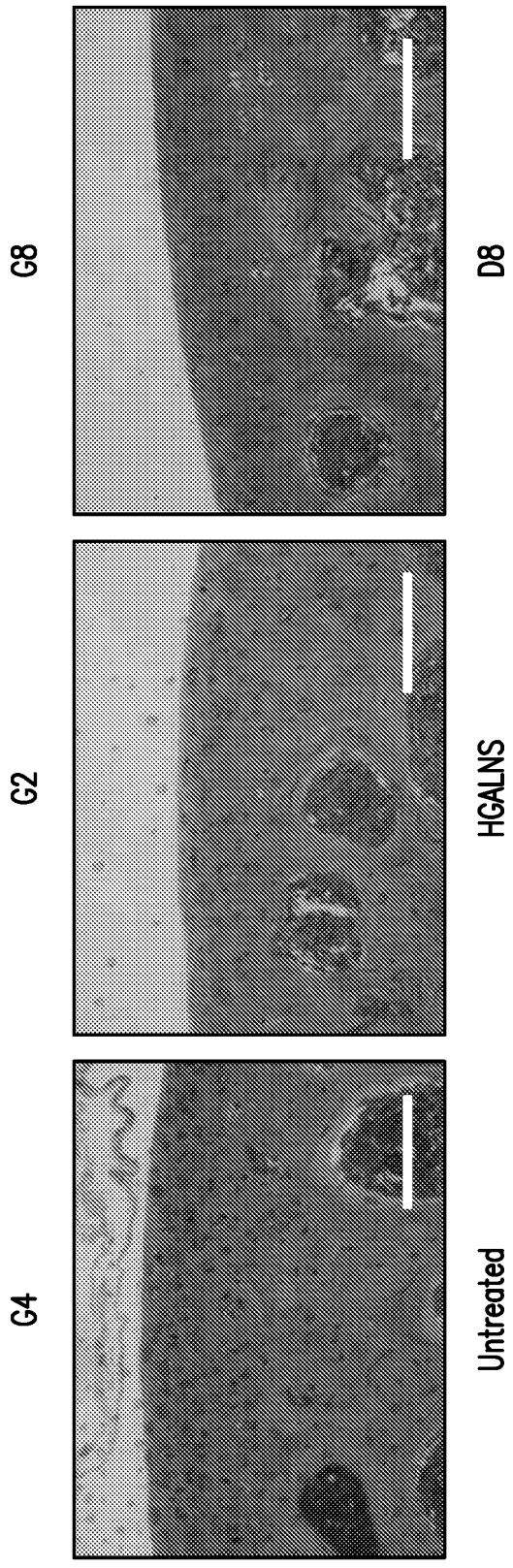


FIG. 11C

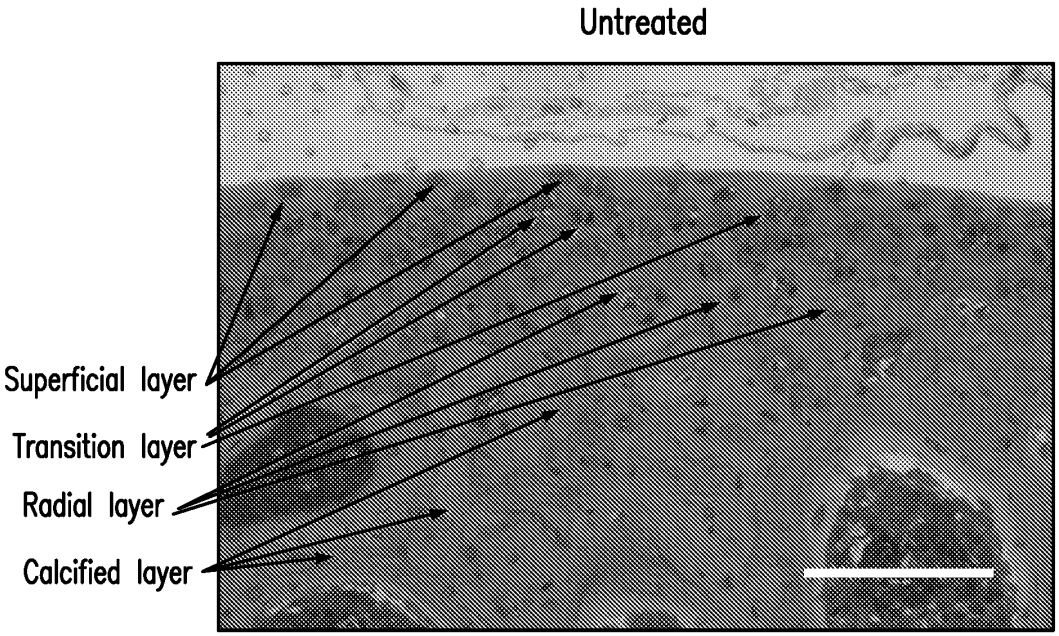

Untreated
Superficial layer
Transition layer
Radial layer
Calcified layer

FIG. 11D

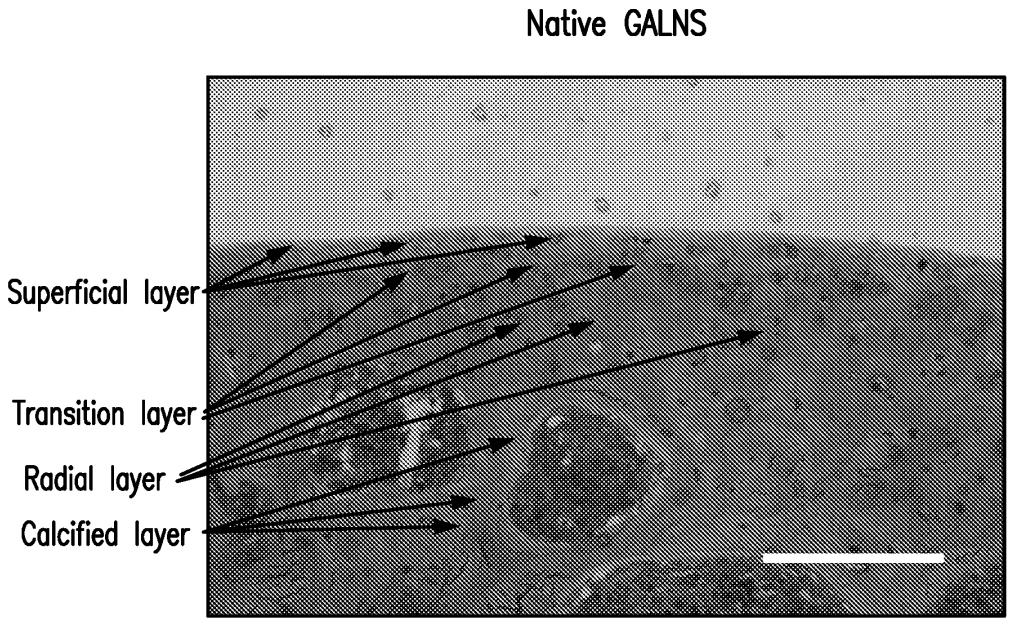

Native GALNS
Superficial layer
Transition layer
Radial layer
Calcified layer

FIG. 11E

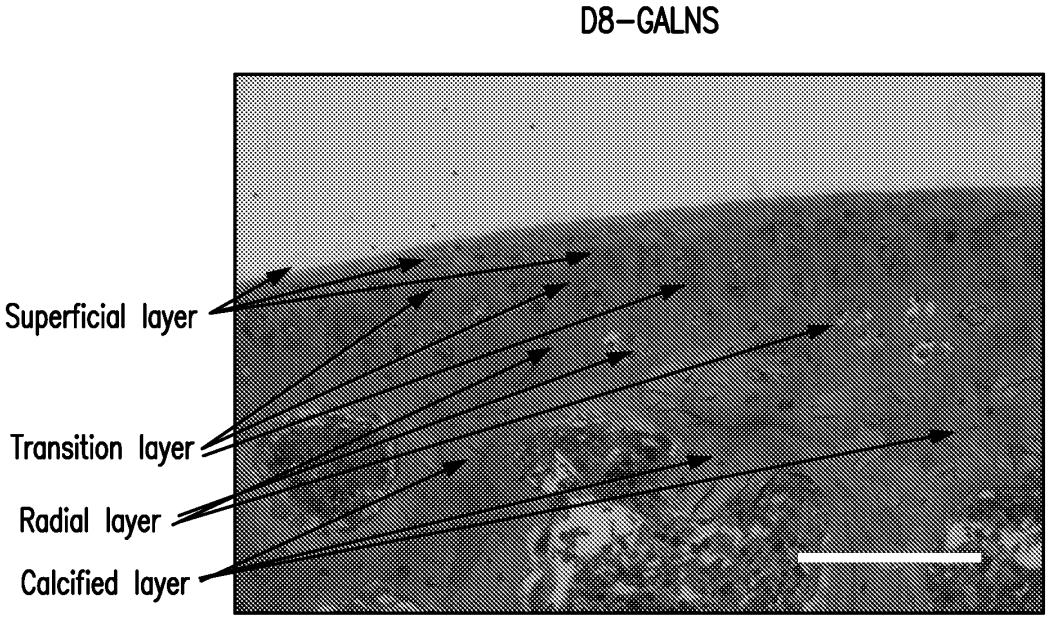

D8-GALNS
Superficial layer
Transition layer
Radial layer
Calcified layer

FIG. 11F

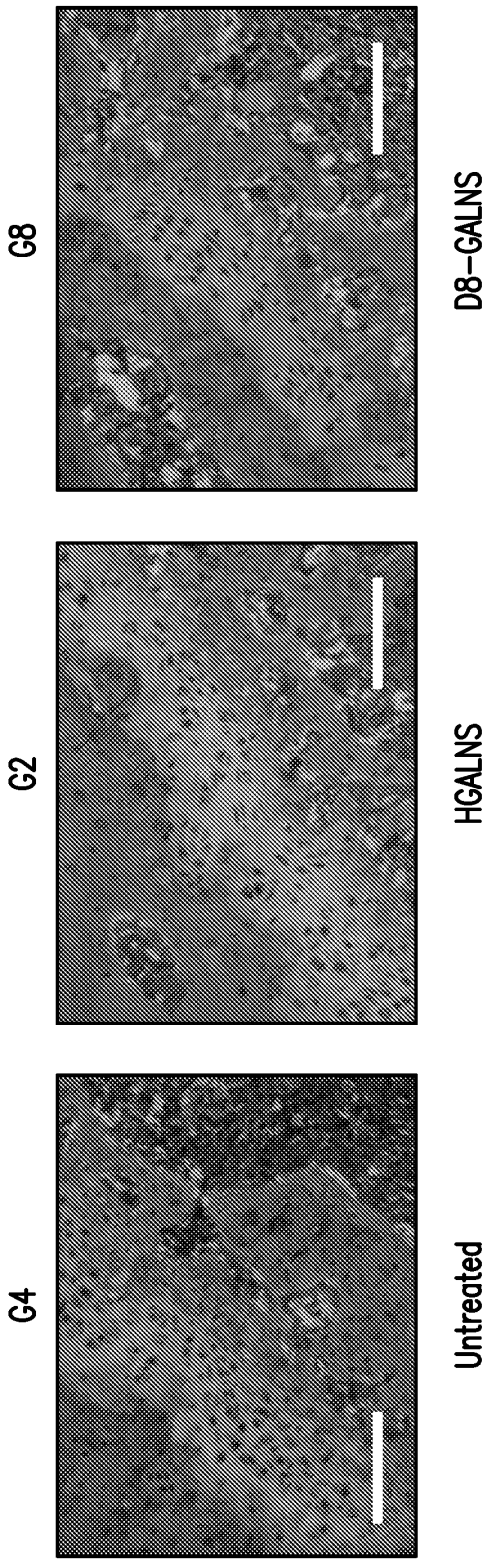


FIG. 11G

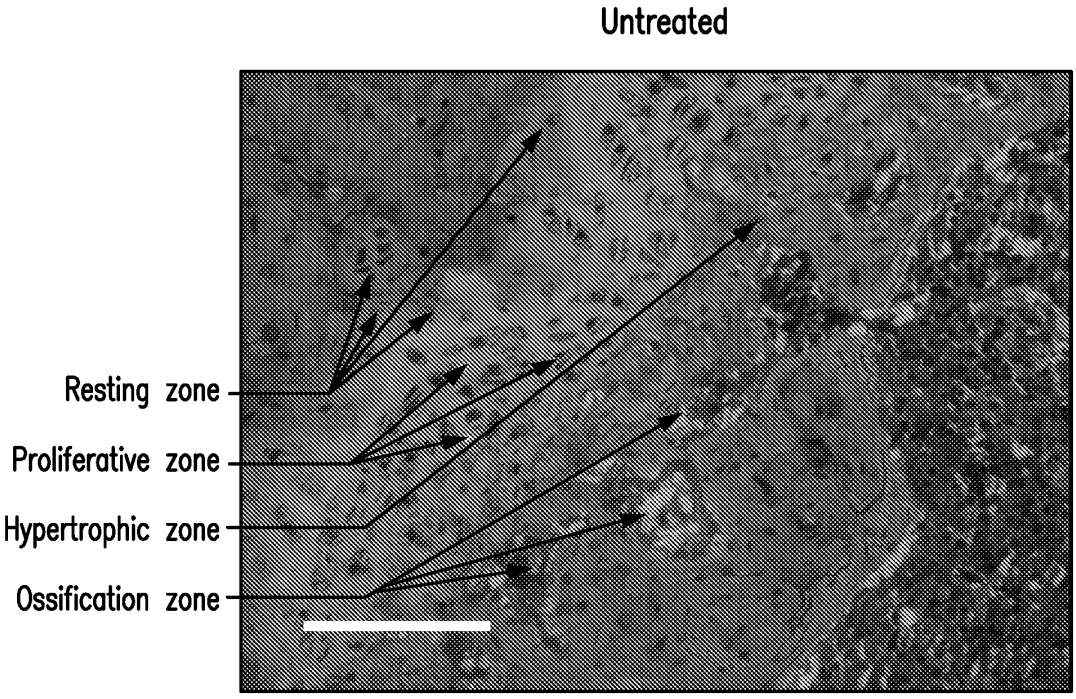

Untreated
Resting zone
Proliferative zone
Hypertrophic zone
Ossification zone

FIG. 11H

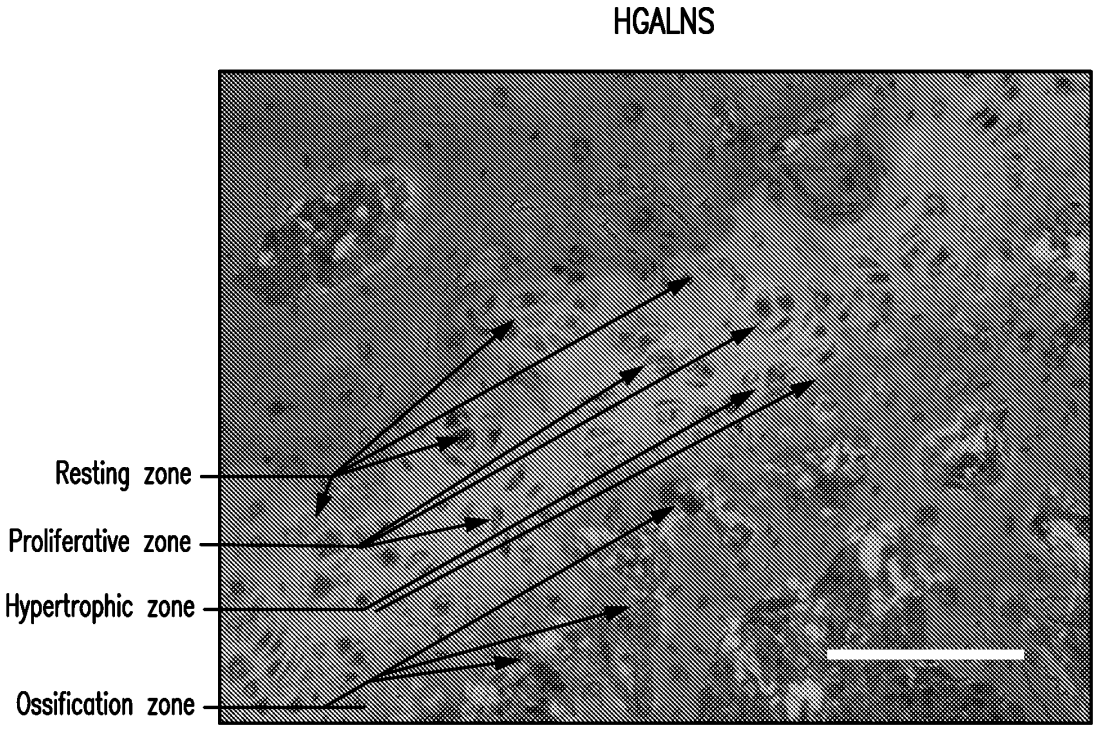

HGALNS
Resting zone
Proliferative zone
Hypertrophic zone
Ossification zone

FIG. 11I

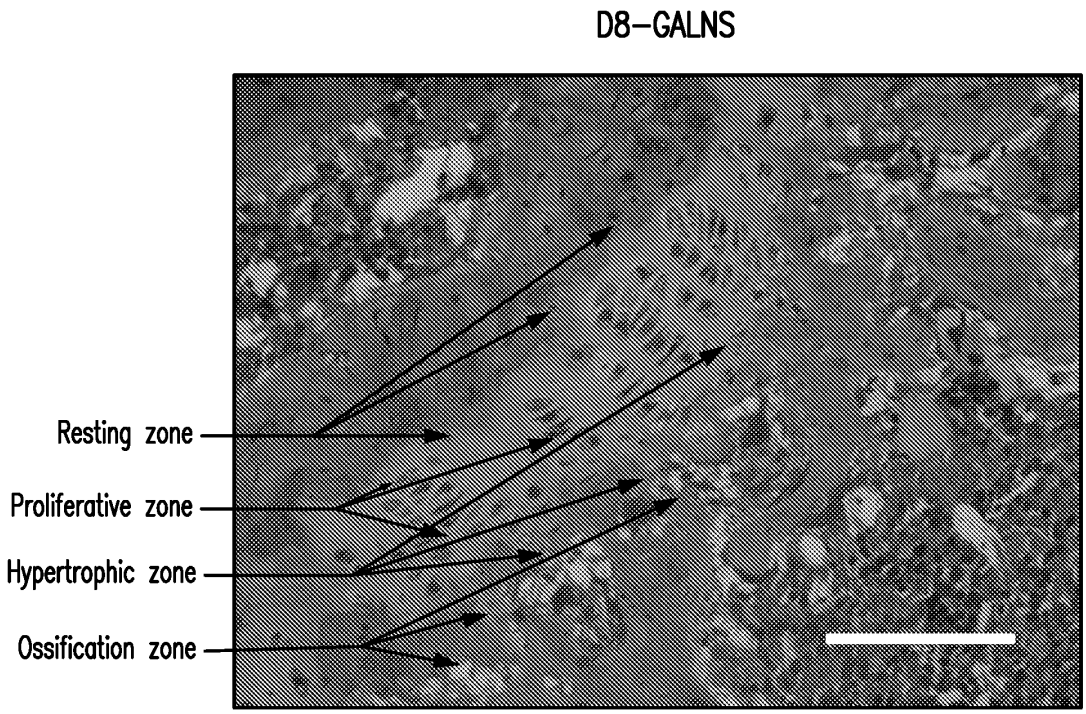

D8-GALNS
Resting zone
Proliferative zone
Hypertrophic zone
Ossification zone

FIG. 11J

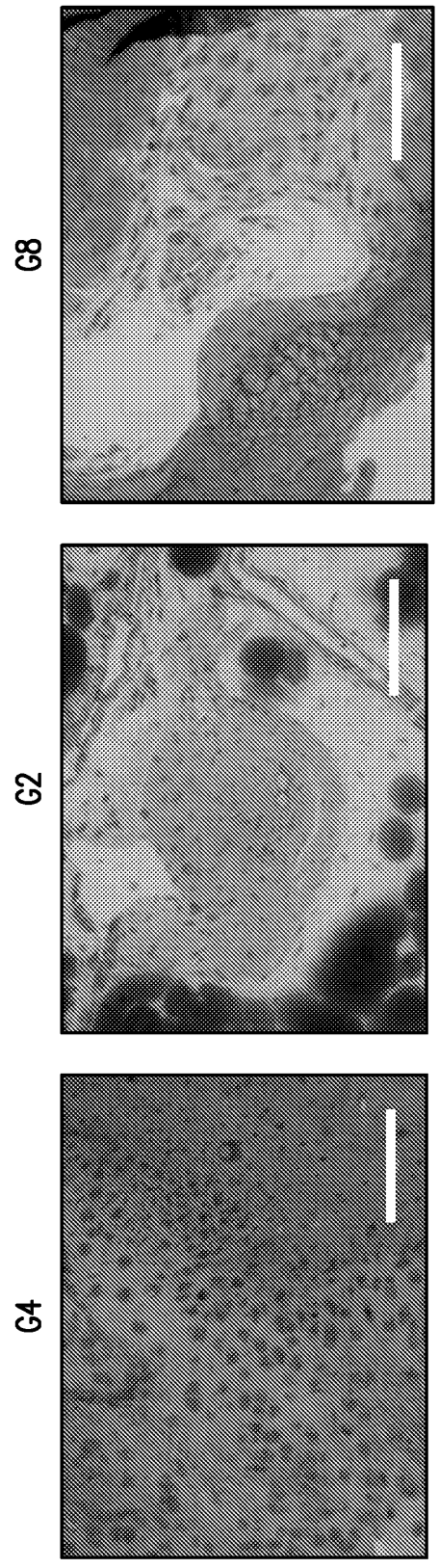


FIG. 11K

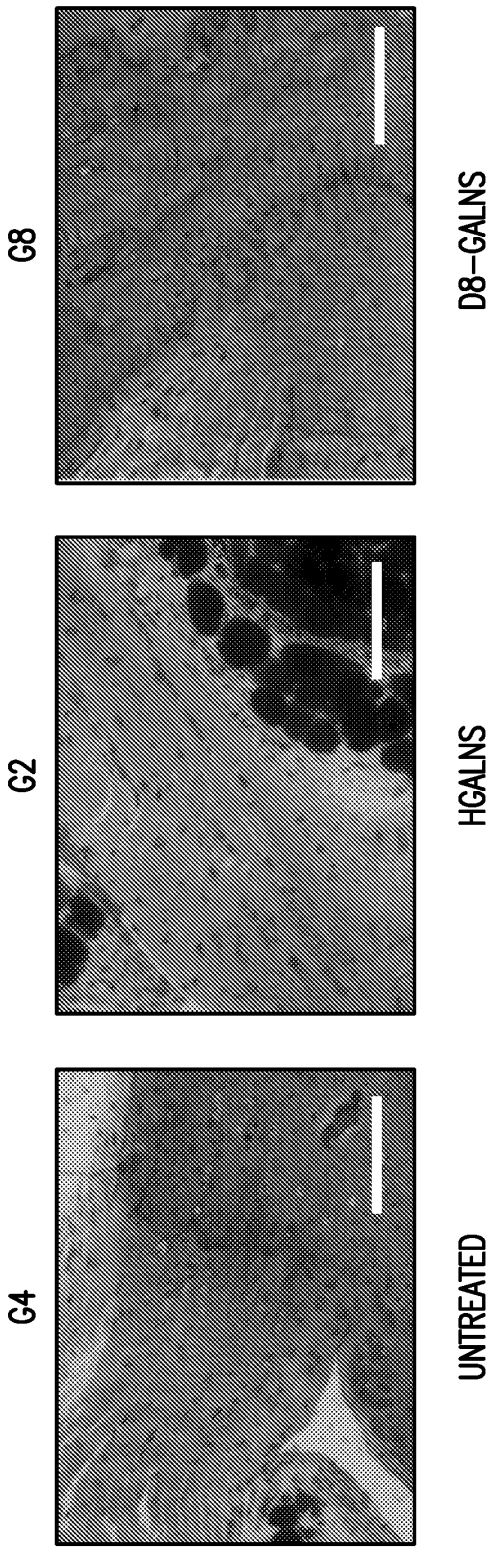


FIG. 11L

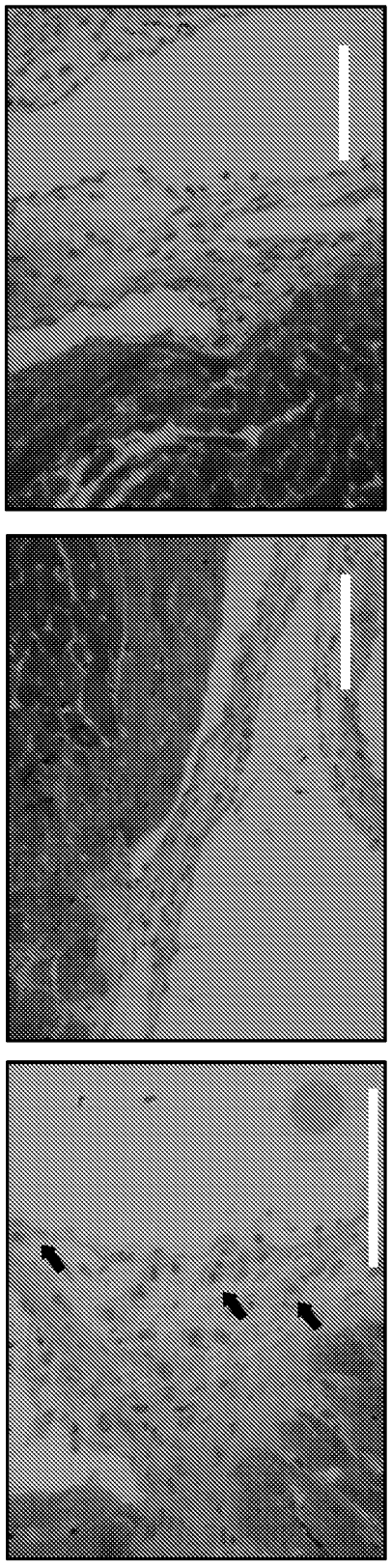


FIG. 11M

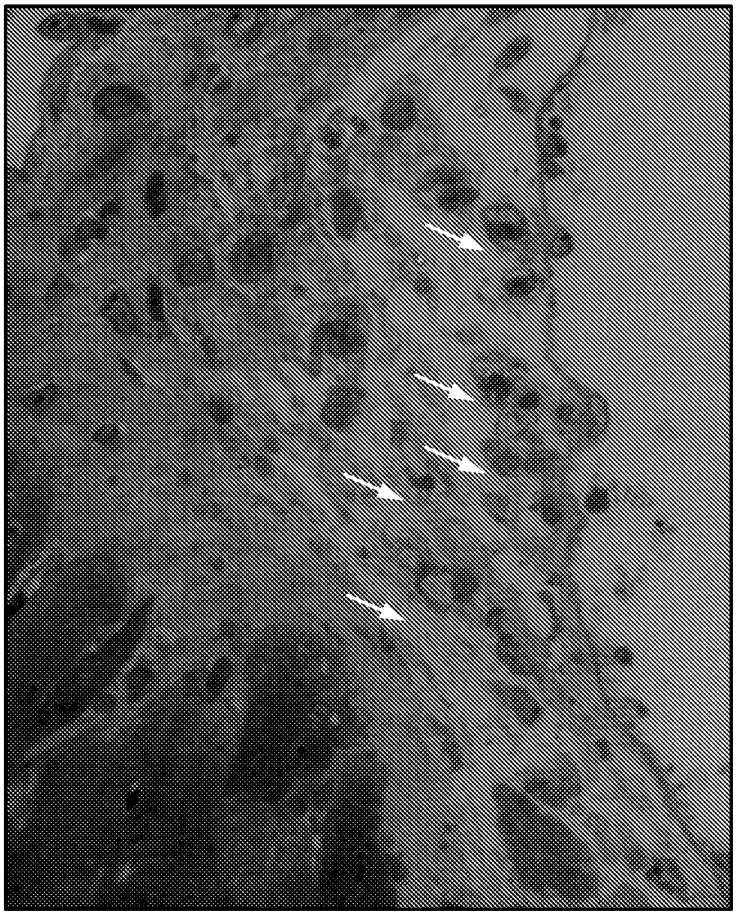


FIG. 11N

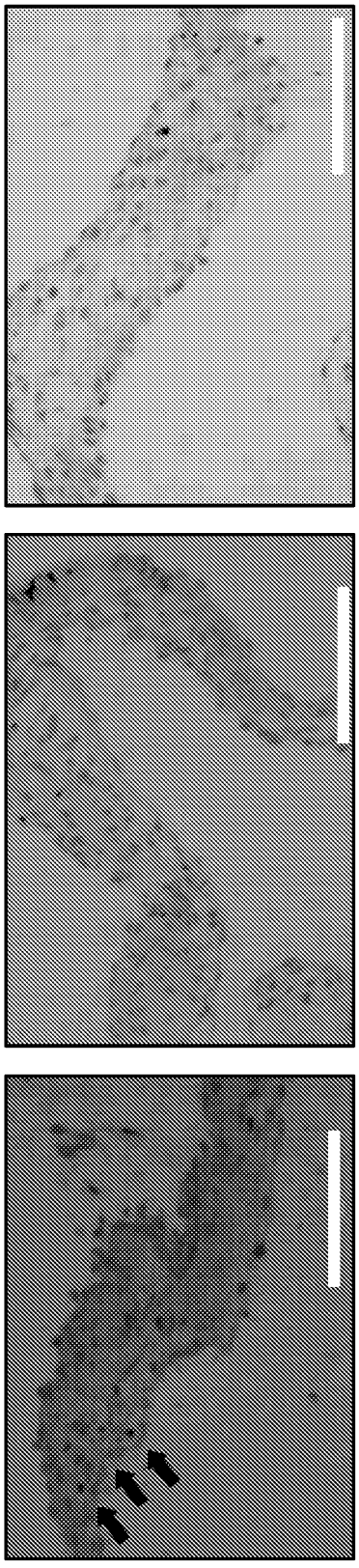


FIG. 11O

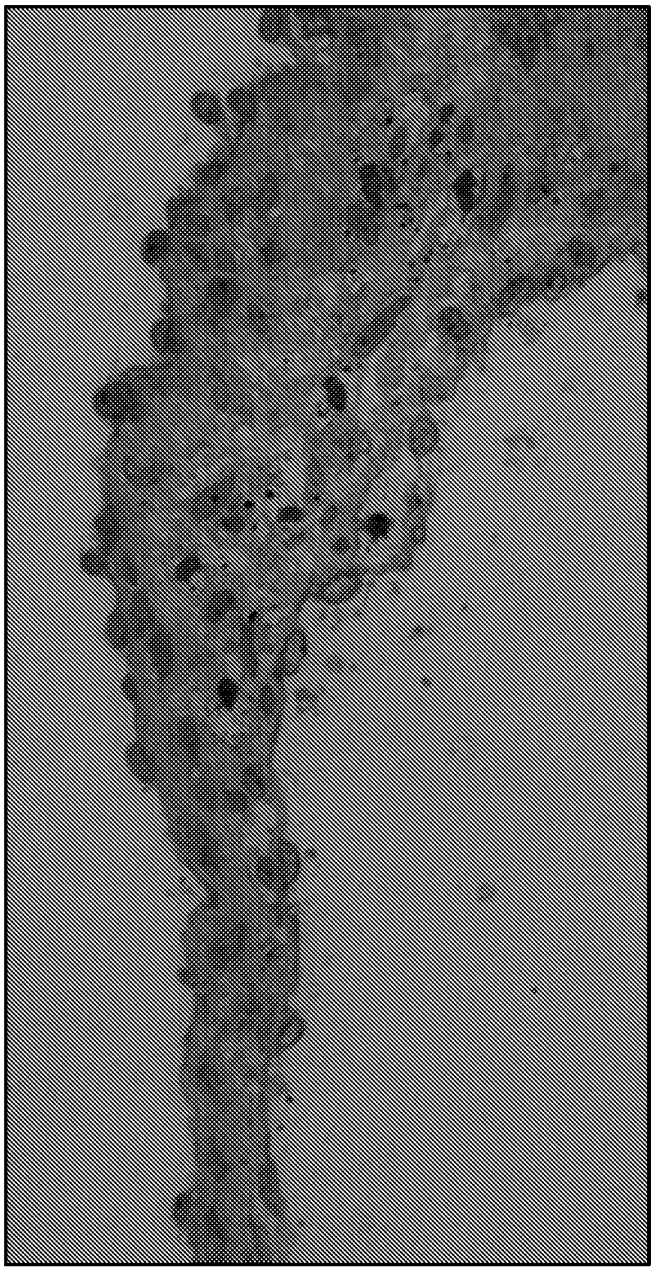


FIG. 11P

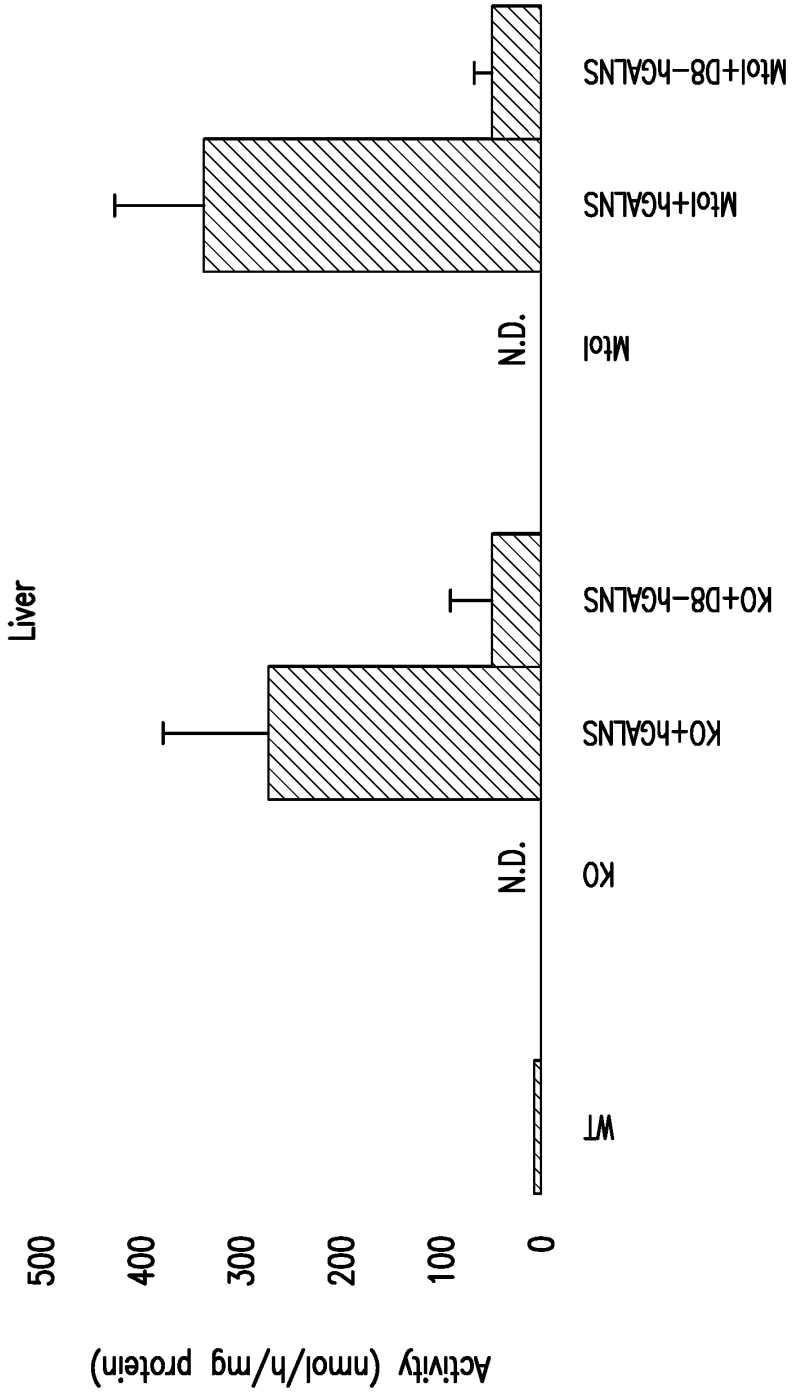

Liver
Activity (nmol/h/mg protein)
0
100
200
300
400
500
WT
KO
KO+hGALNS
KO+DB-hGALNS
Mtol
Mtol+hGALNS
Mtol+DB-hGALNS
N.D.
N.D.

FIG. 12A

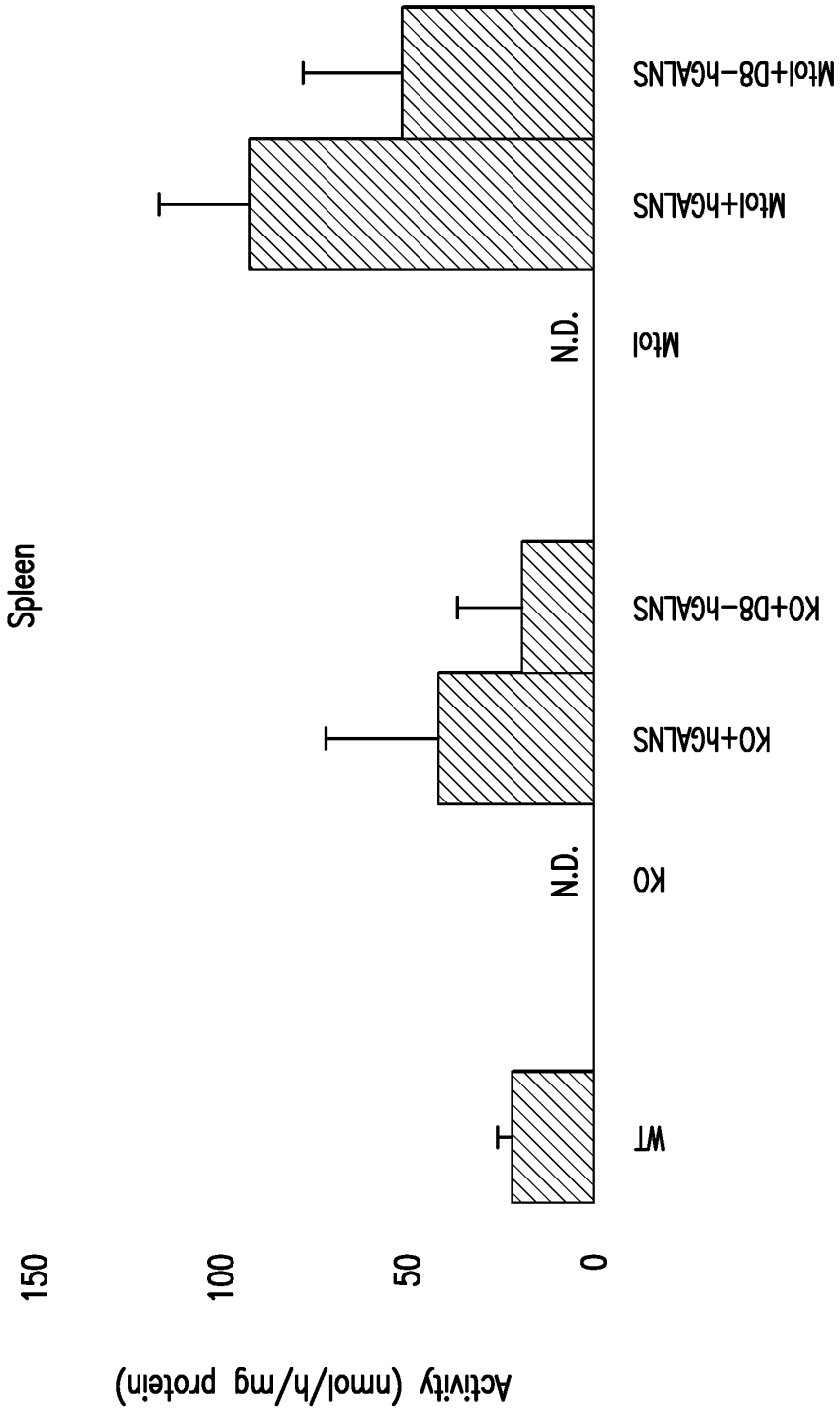

Spleen
Activity (nmol/h/mg protein)
150
100
50
0
WT
N.D.
KO
KO+hGALNS
KO+D8-hGALNS
N.D.
Mtol
Mtol+hGALNS
Mtol+D8-hGALNS

FIG. 12B

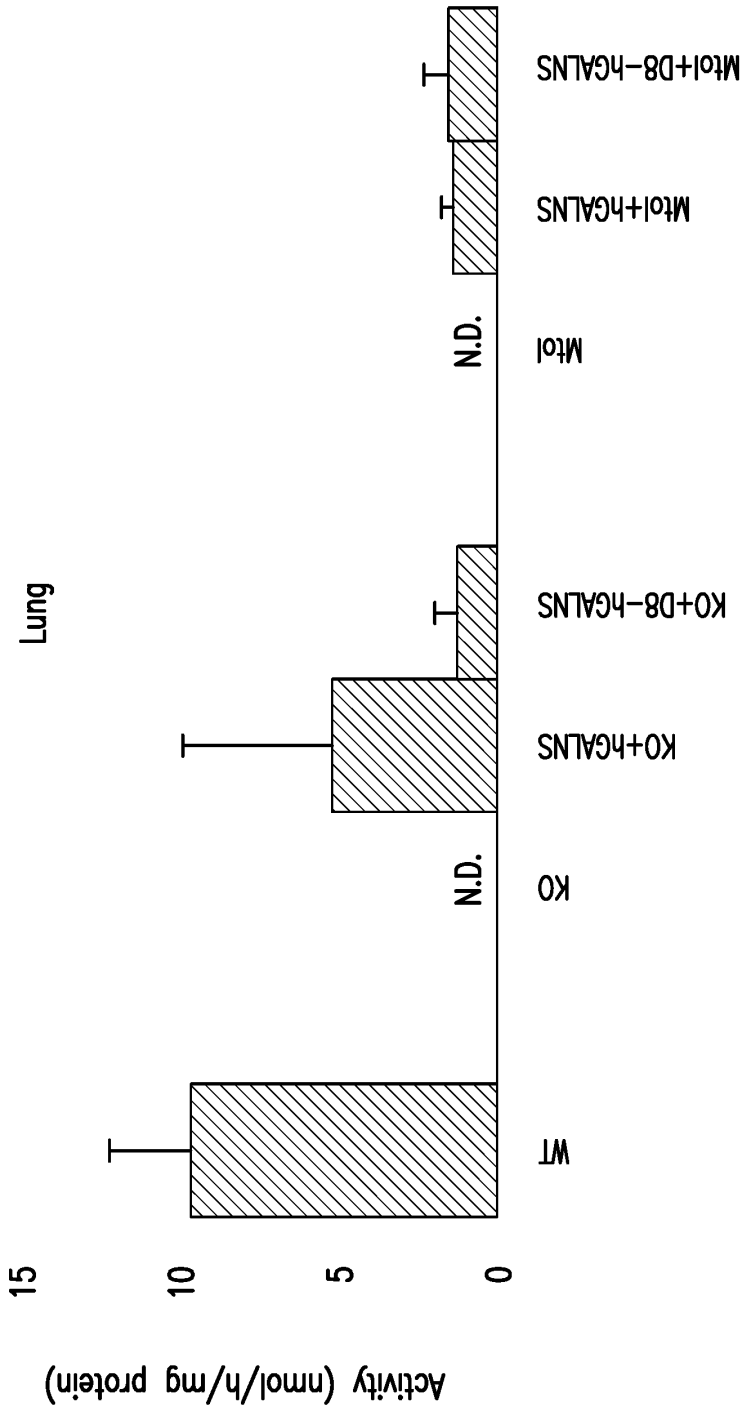

Lung
Activity (nmol/h/mg protein)
15
10
5
0
WT
N.D.
KO
KO+hGALNS
KO+DB-hGALNS
Mtoi
N.D.
Mtoi+hGALNS
Mtoi+DB-hGALNS

FIG. 12C

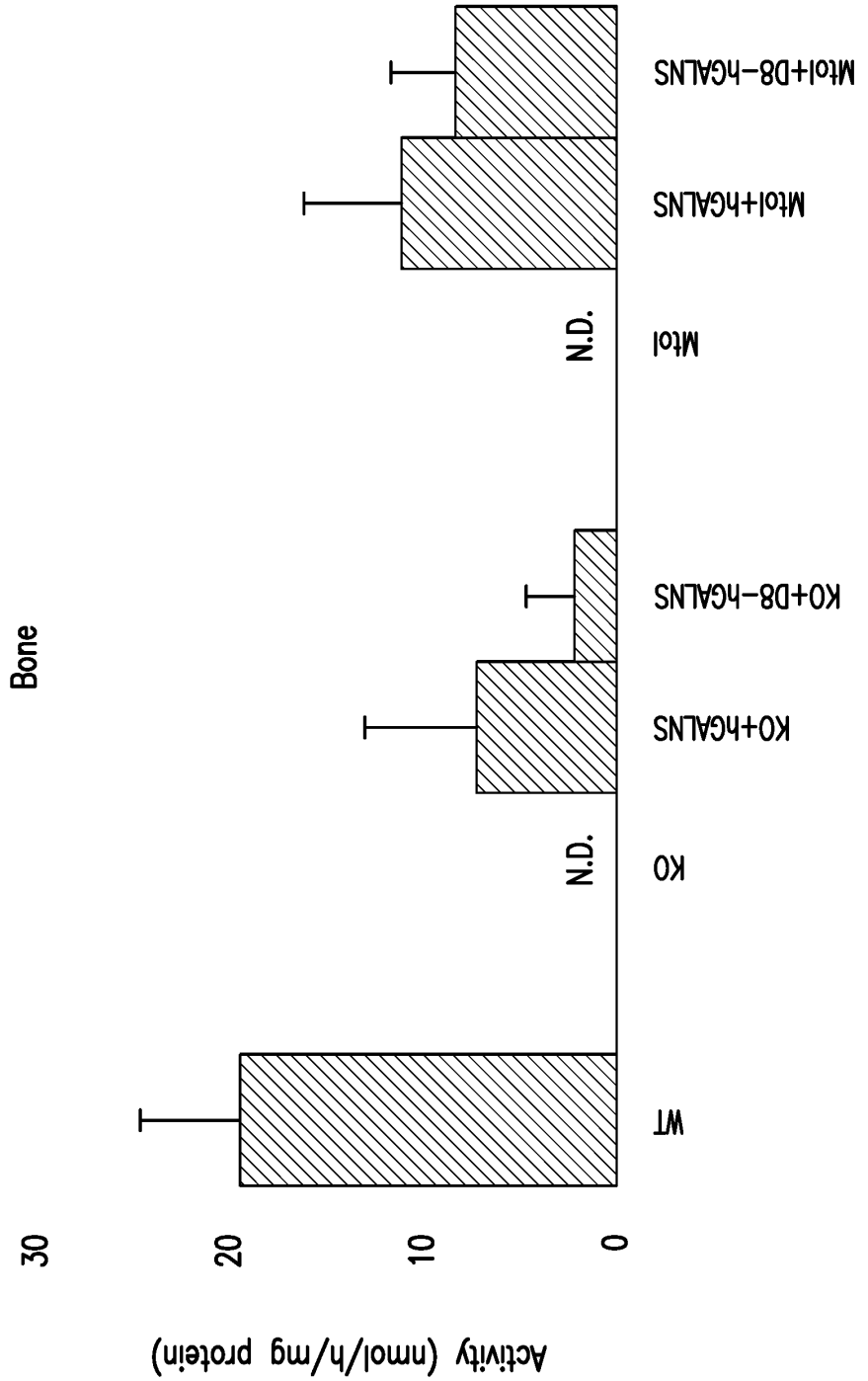

Bone
Activity (nmol/h/mg protein)
0
10
20
30
WT
KO
N.D.
KO+hGALNS
KO+D8-hGALNS
Mtol
N.D.
Mtol+hGALNS
Mtol+D8-hGALNS

FIG. 13A

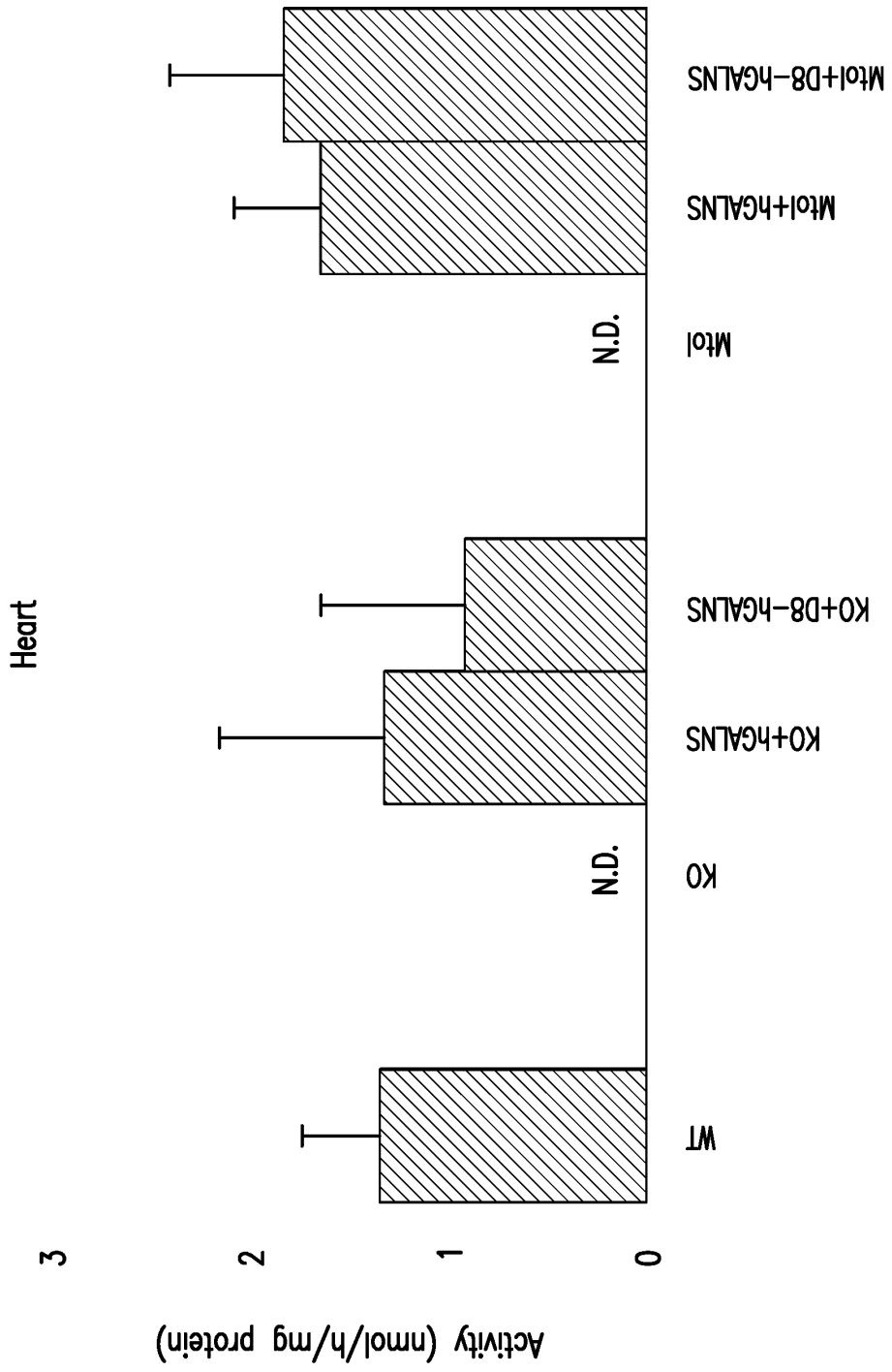

Heart
Activity (nmol/h/mg protein)
0
1
2
3
WT
KO
N.D.
KO+hGALNS
KO+DB-hGALNS
Mtol
N.D.
Mtol+hGALNS
Mtol+DB-hGALNS

FIG. 13B

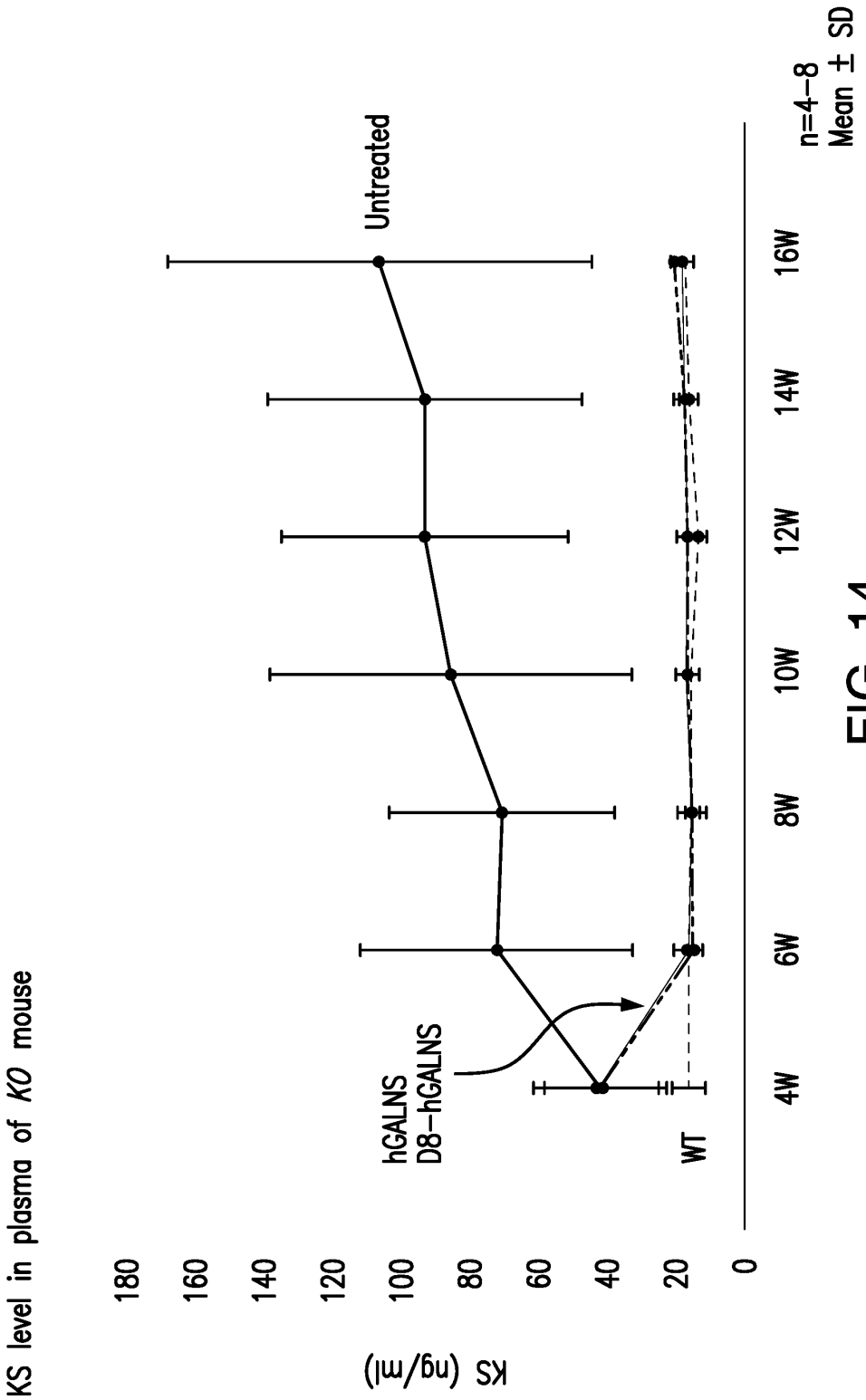

KS level in plasma of KO mouse
KS (ng/ml)
180
160
140
120
100
80
60
40
20
0
Untreated
hGALNS
D8–hGALNS
WT
4W
6W
8W
10W
12W
14W
16W
n=4–8
Mean ± SD

FIG. 14

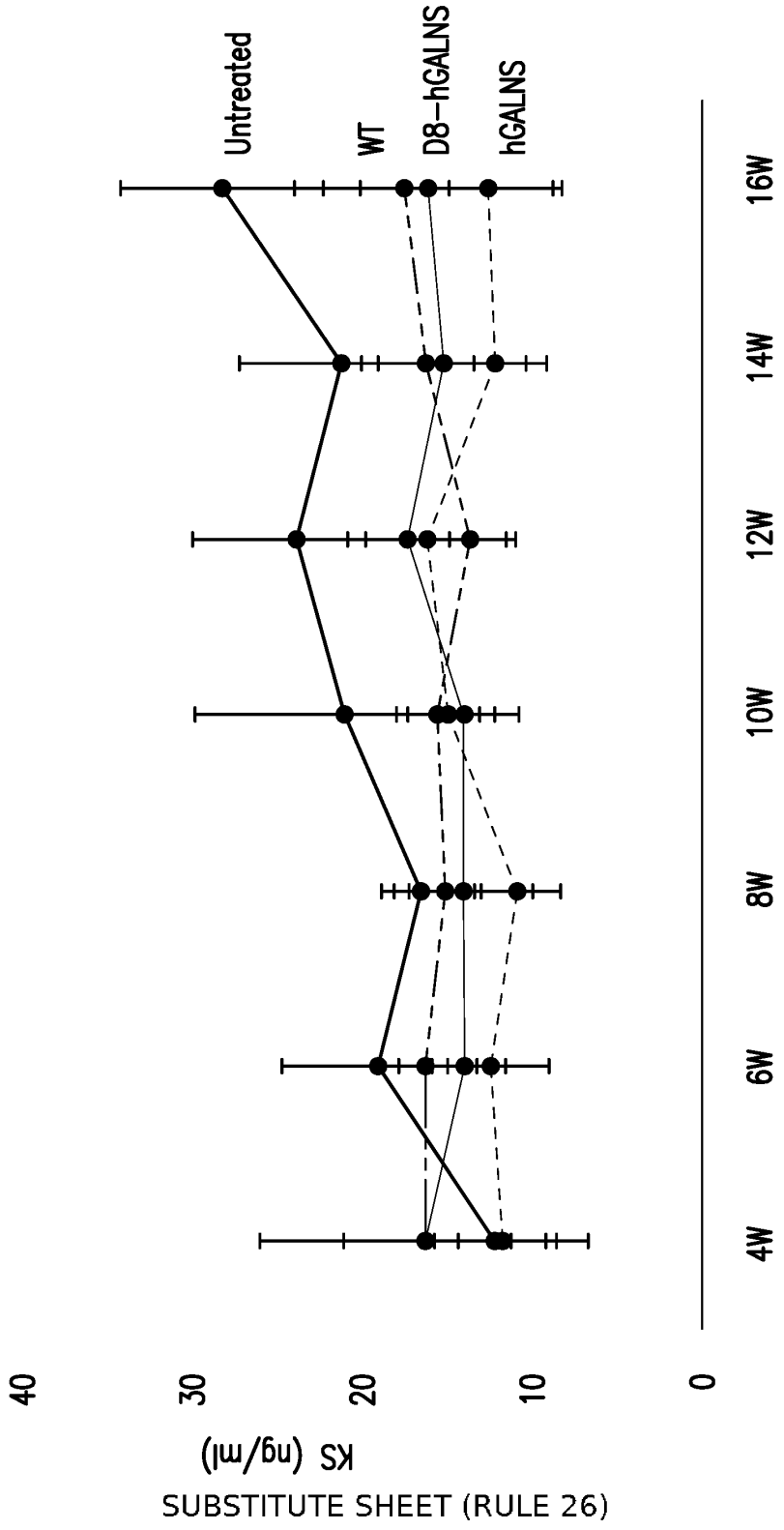

Untreated
WT
D8−hGALNS
hGALNS
4W
6W
8W
10W
12W
14W
16W
0
10
20
30
40
KS (ng/ml)

FIG. 15A

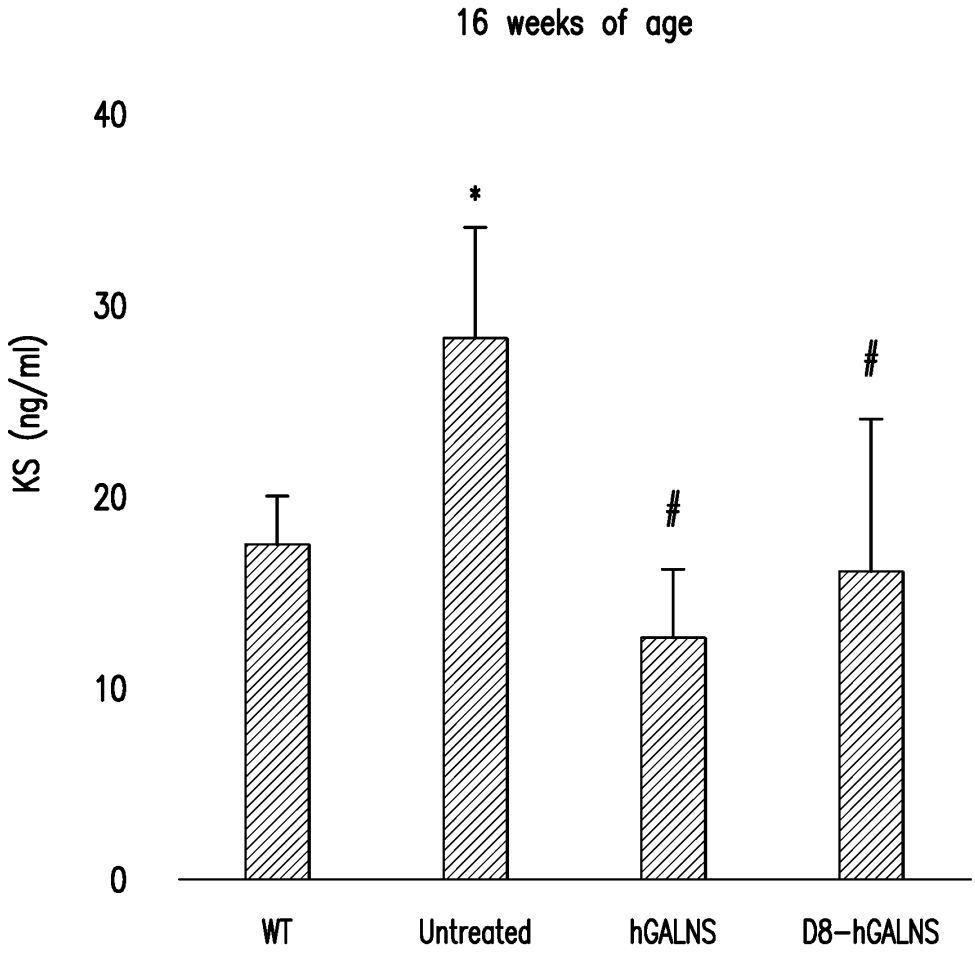

16 weeks of age
KS (ng/ml)
40
30
20
10
0
*
#
#
WT
Untreated
hGALNS
D8-hGALNS

FIG. 15B

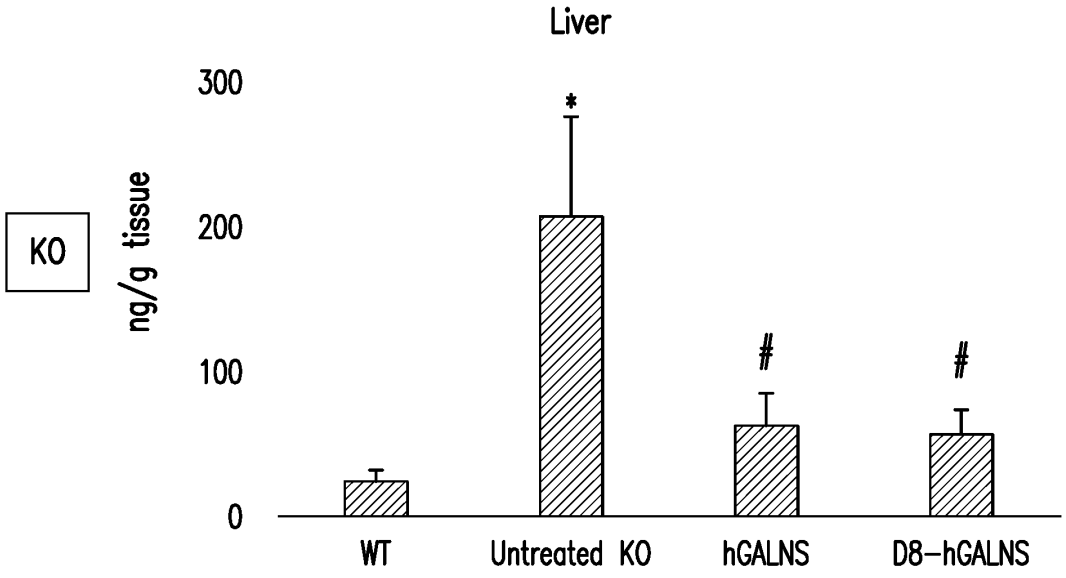

Liver
KO
ng/g tissue
300
200
100
0
*
#
#
WT
Untreated KO
hGALNS
D8-hGALNS

FIG. 16A

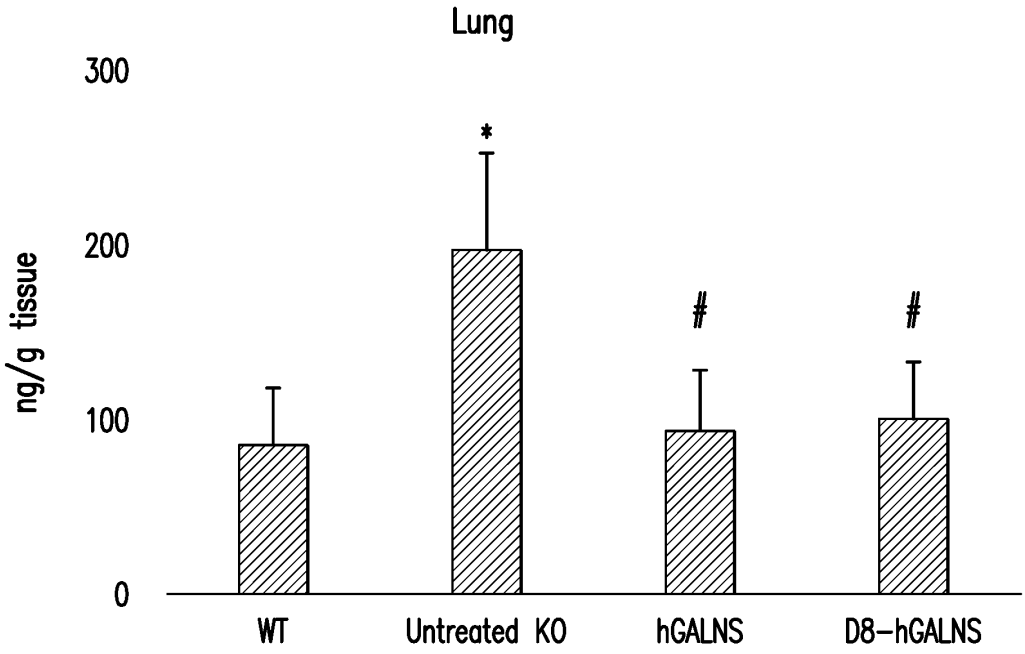

Lung
300
200
100
0
ng/g tissue
*
#
#
WT
Untreated KO
hGALNS
D8-hGALNS

FIG. 16B

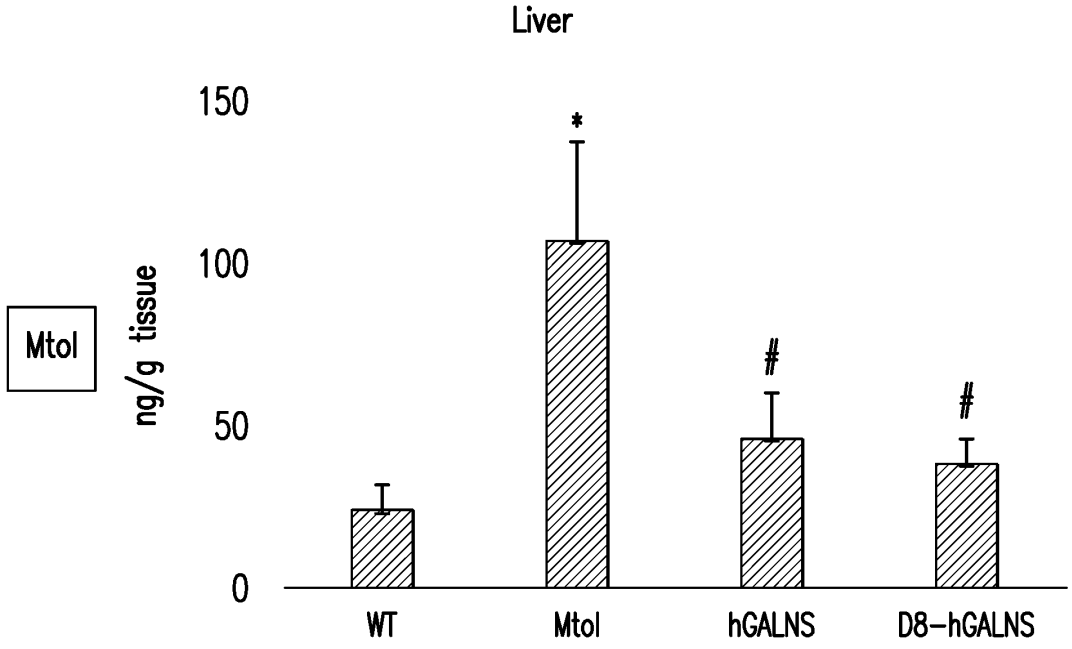
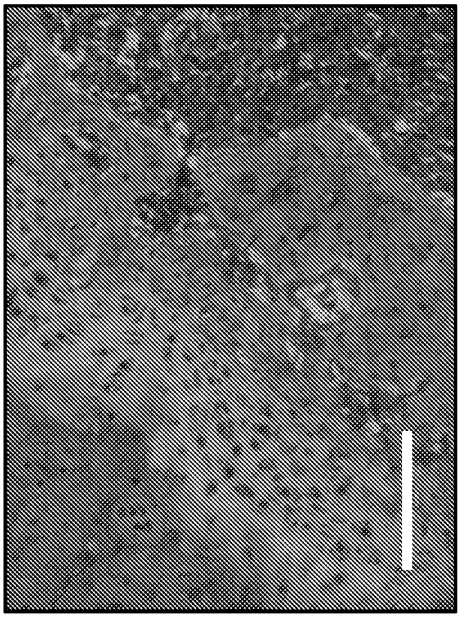
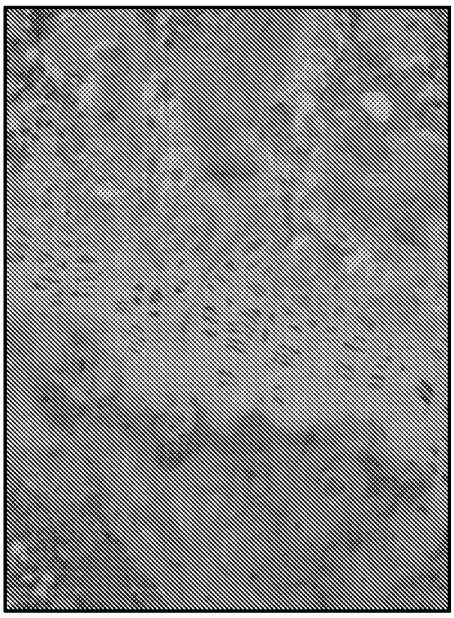


FIG. 16C



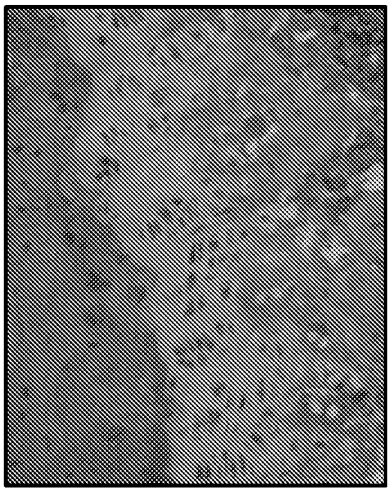
Untreated

FIG. 17B



WT

FIG. 17A



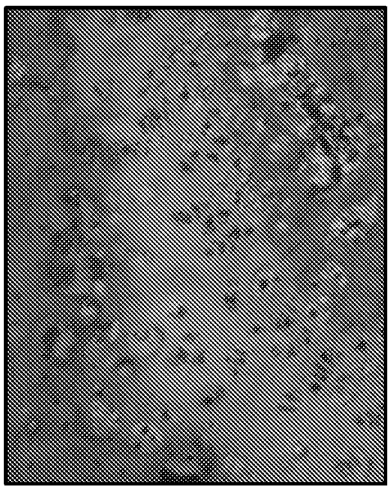
AAV8-D8-GALNS

FIG. 17E



AAV8-hGALNS

FIG. 17D



Untreated

FIG. 17C

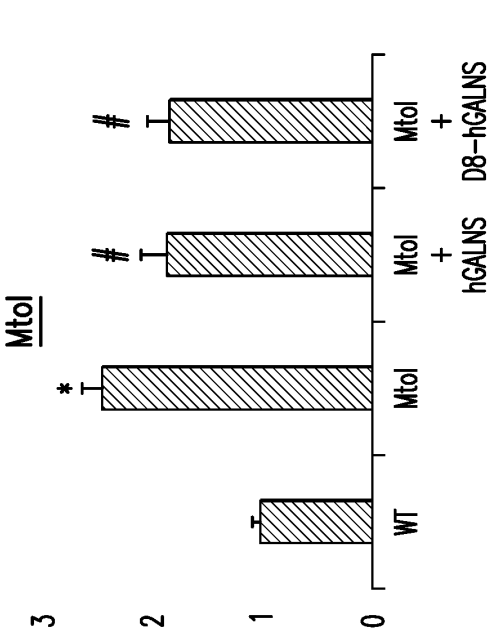


FIG. 18A

FIG. 18B

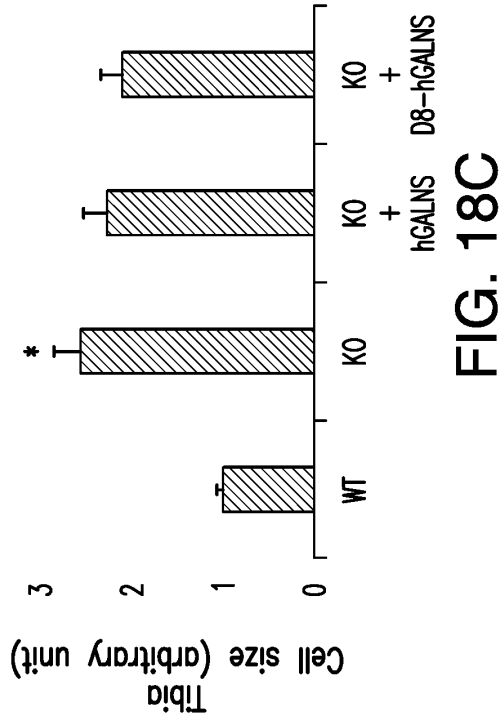


FIG. 18C

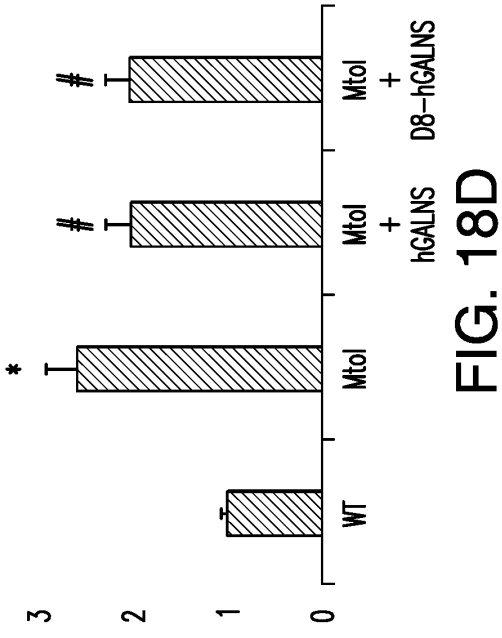


FIG. 18D

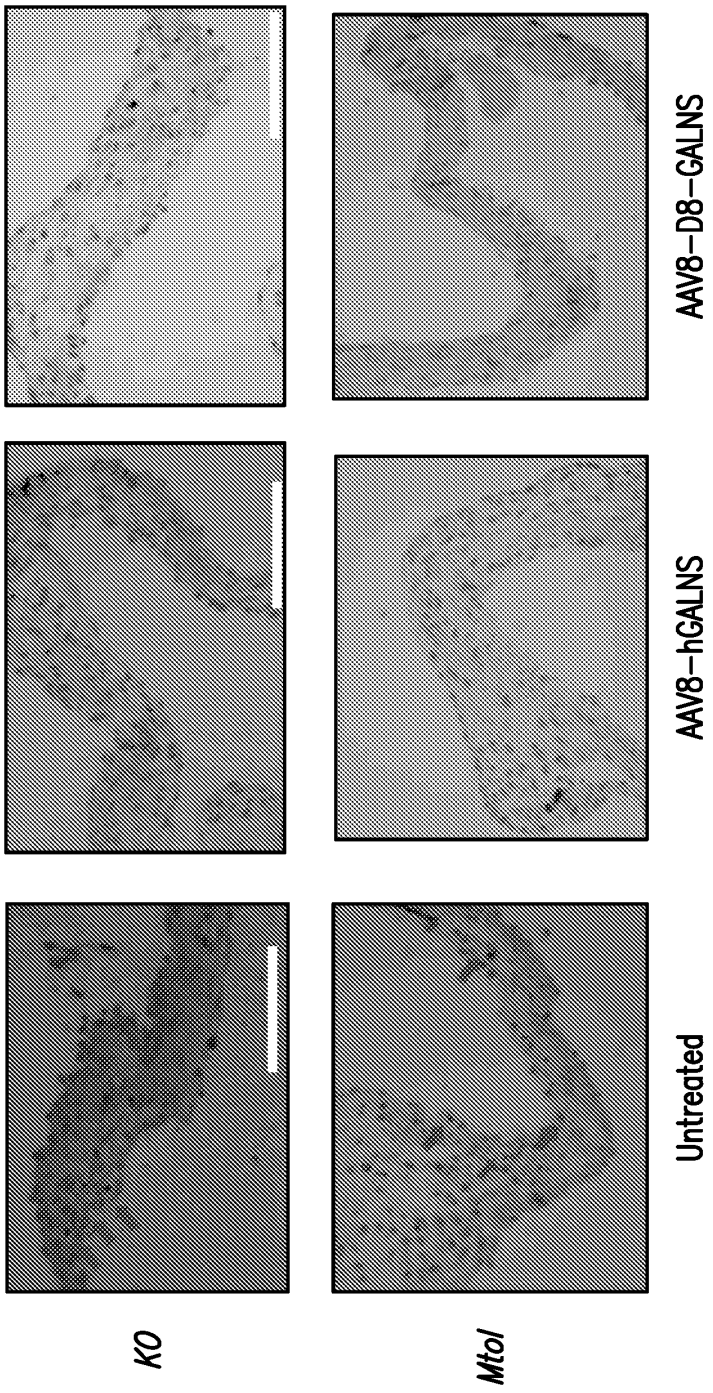


FIG. 19

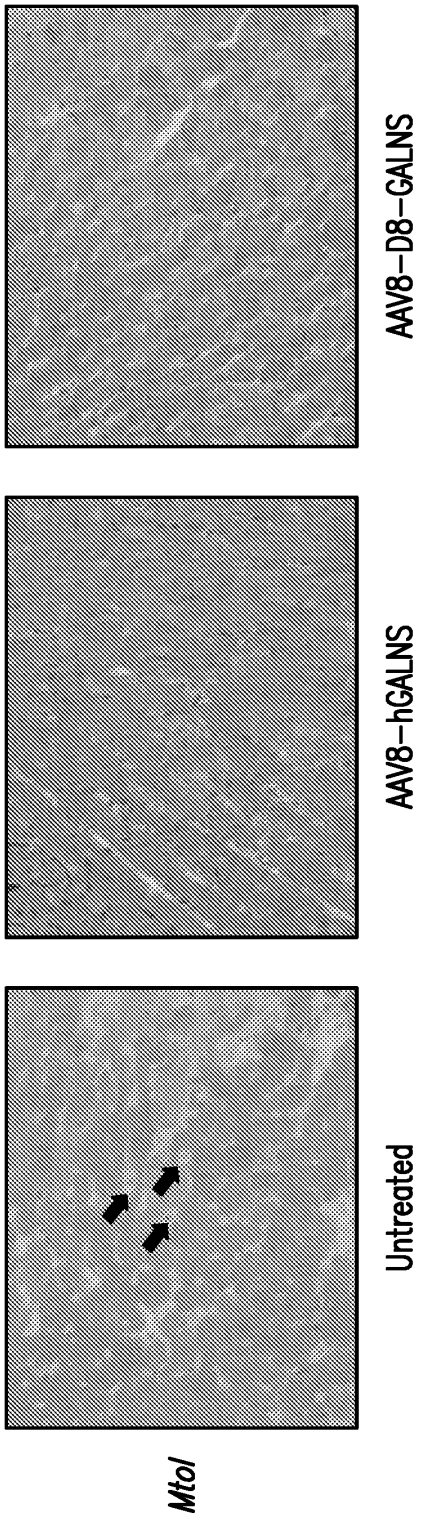


FIG. 20

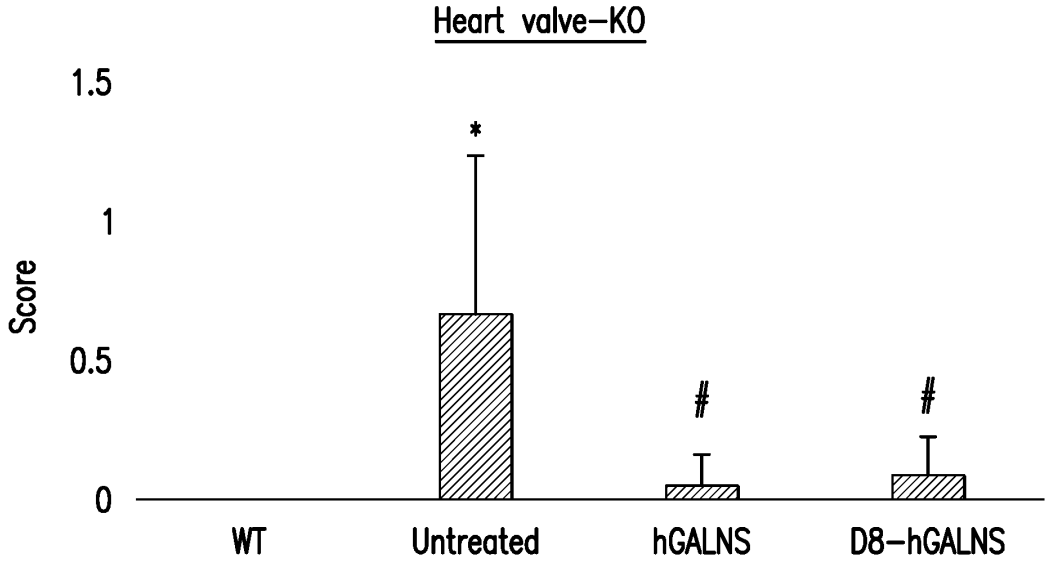


FIG. 21A

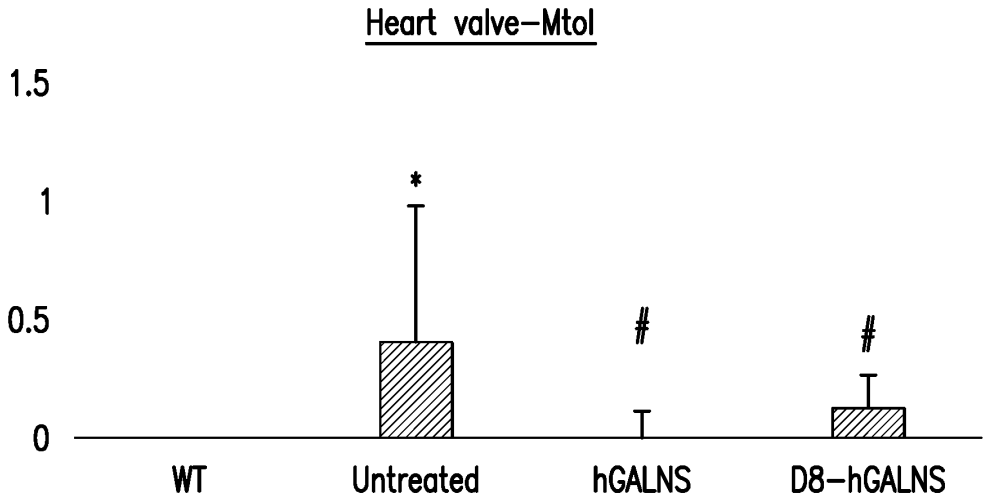


FIG. 21B

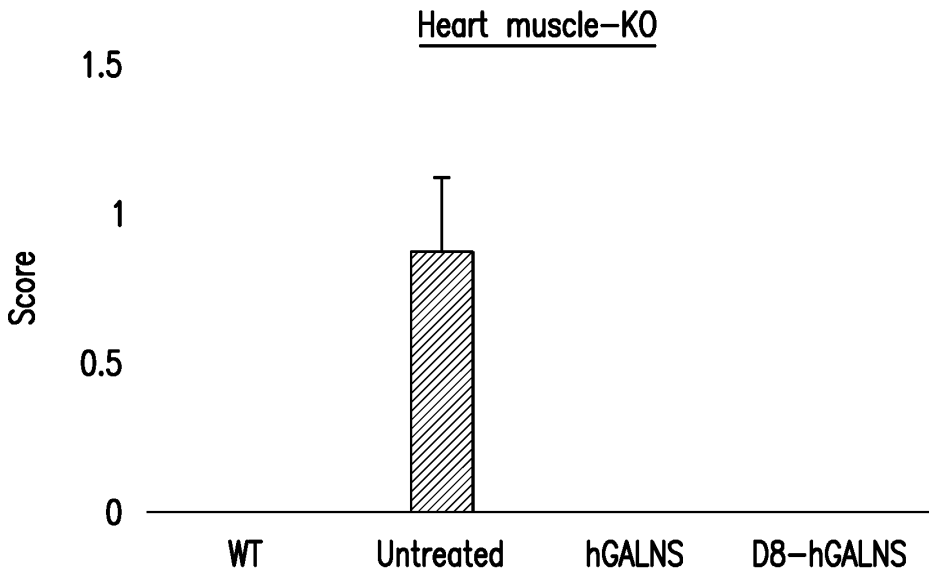


FIG. 21C

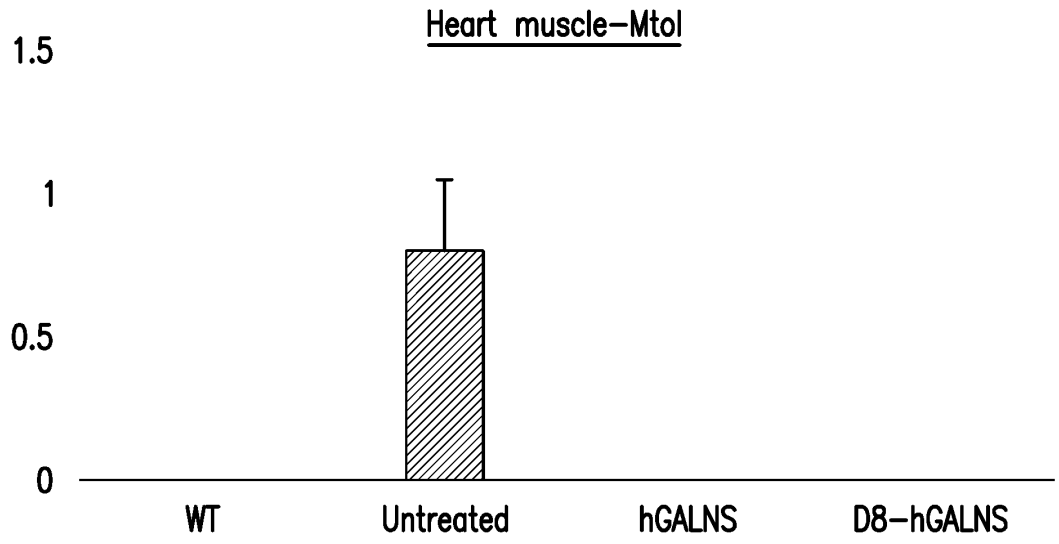


FIG. 21D

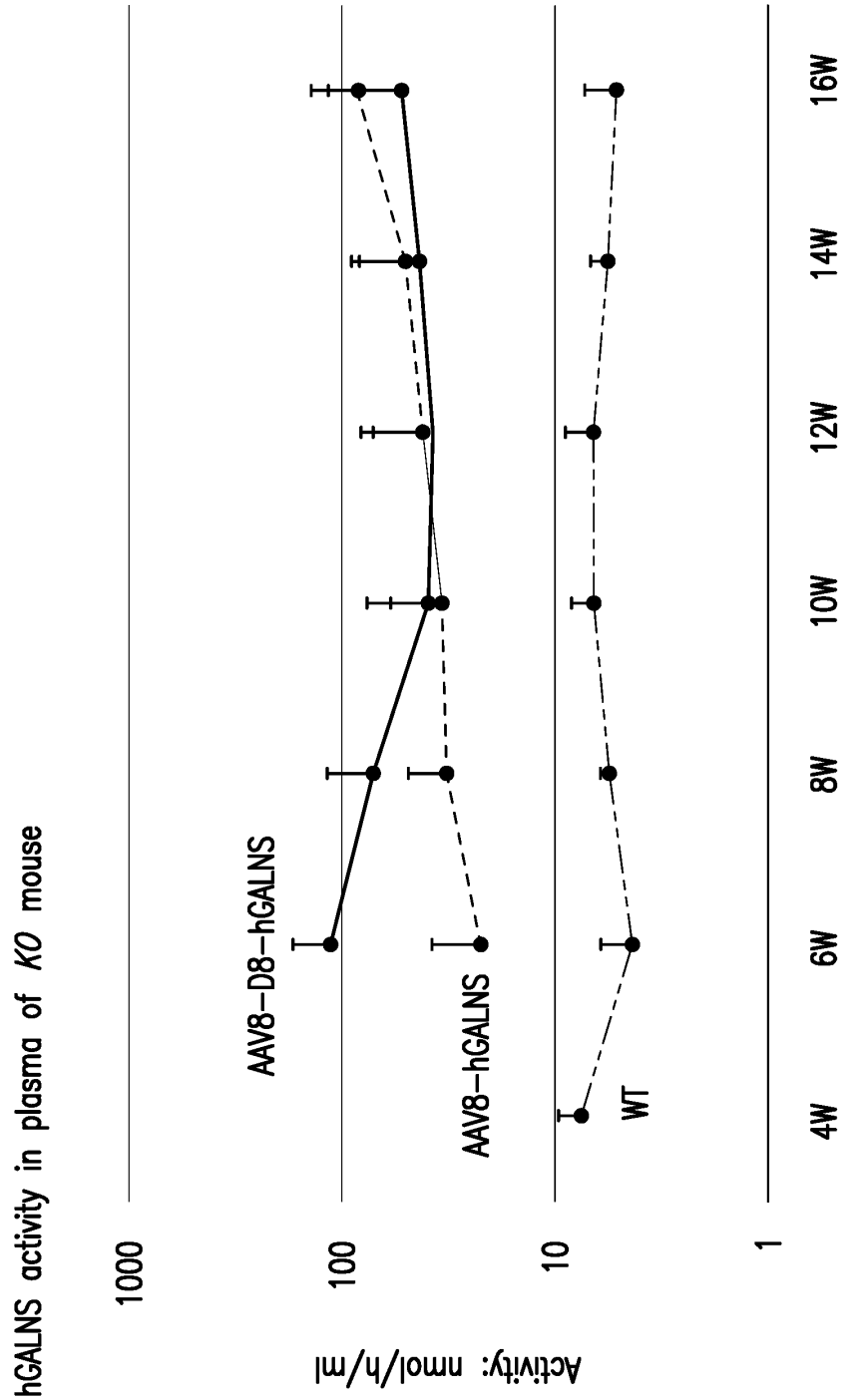

hGALNS activity in plasma of KO mouse
Activity: nmol/h/ml
1000
100
10
1
AAV8-D8-hGALNS
AAV8-hGALNS
WT
4W
6W
8W
10W
12W
14W
16W

FIG. 22

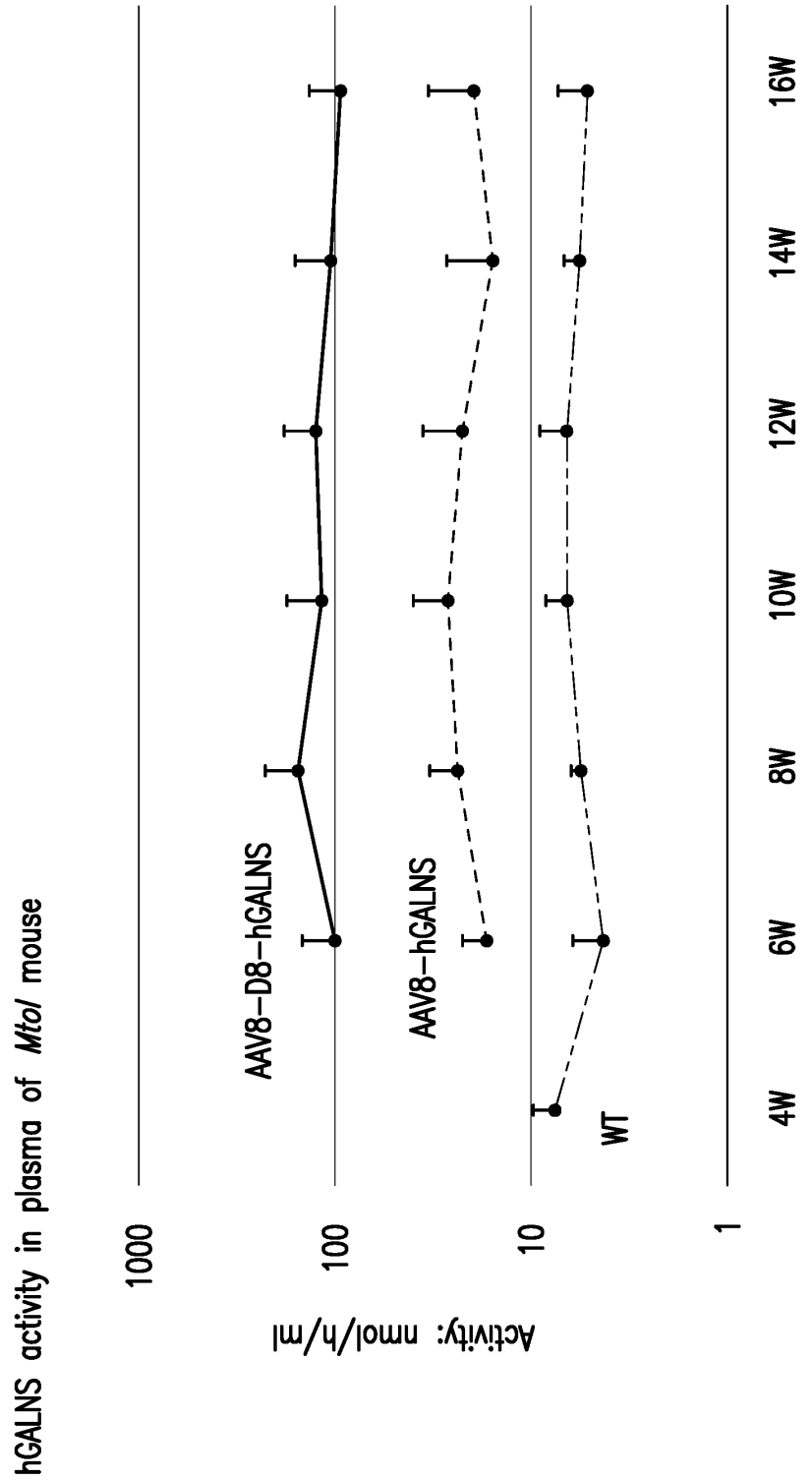

hGALNS activity in plasma of Mtol mouse
Activity: nmol/h/ml
1000
100
10
1
AAV8-D8-hGALNS
AAV8-hGALNS
WT
4W
6W
8W
10W
12W
14W
16W

FIG. 23

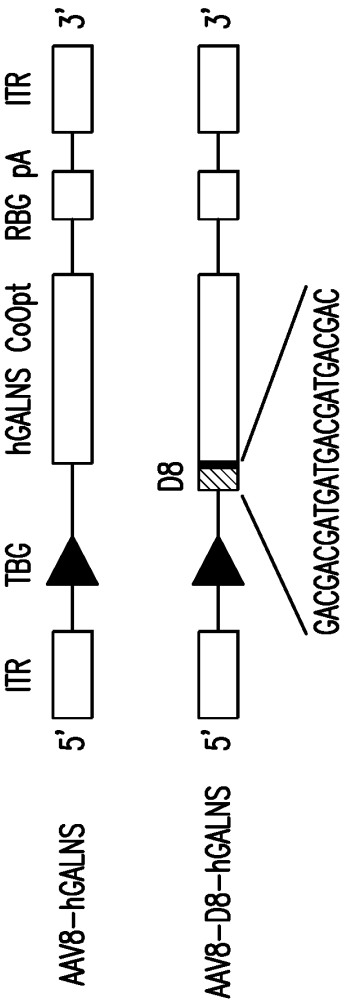


FIG. 24A

FIG. 24B FIG. 24C

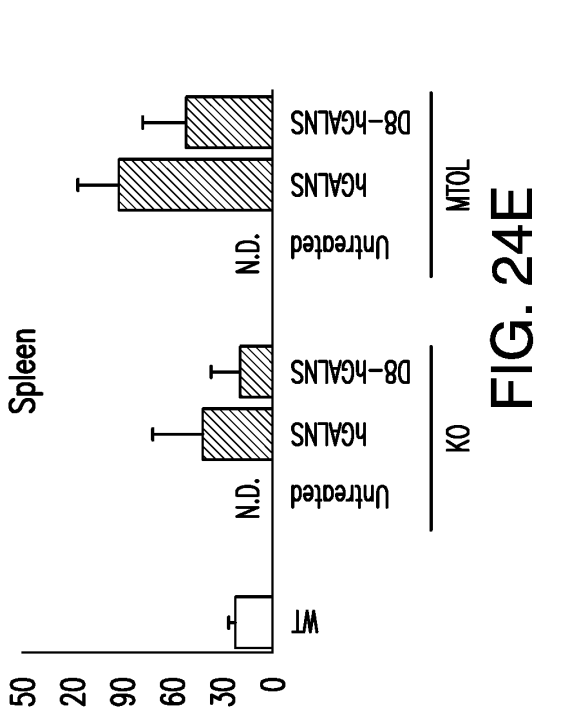


FIG. 24E

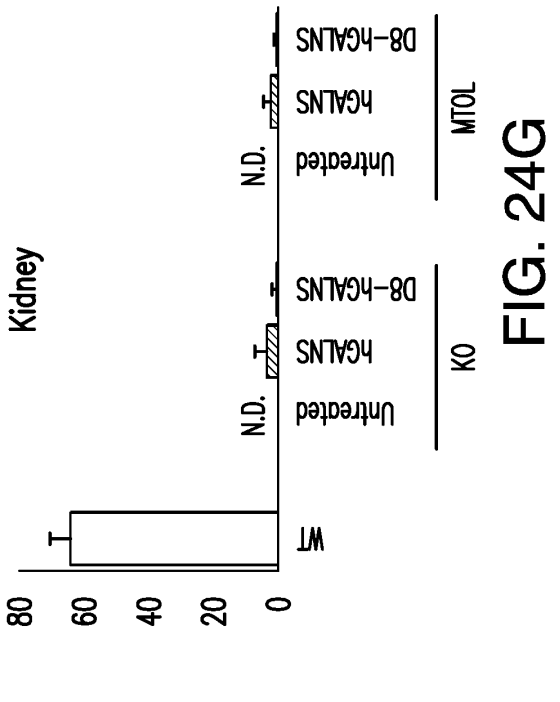


FIG. 24G

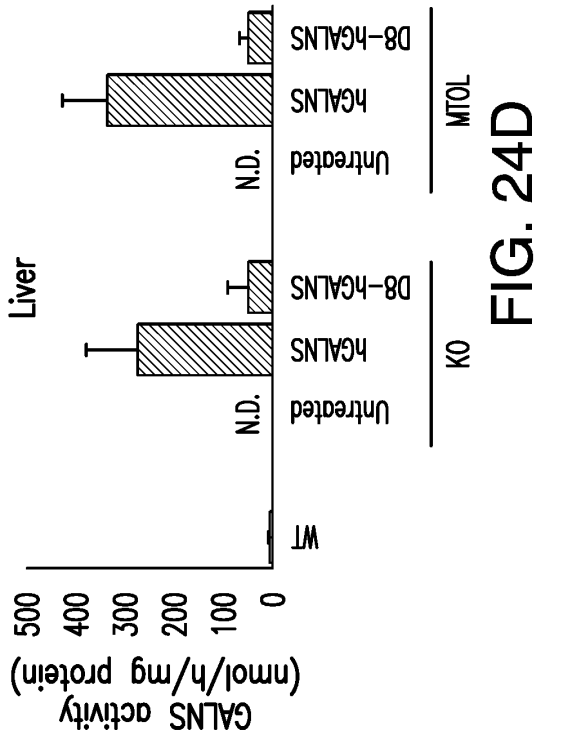


FIG. 24D

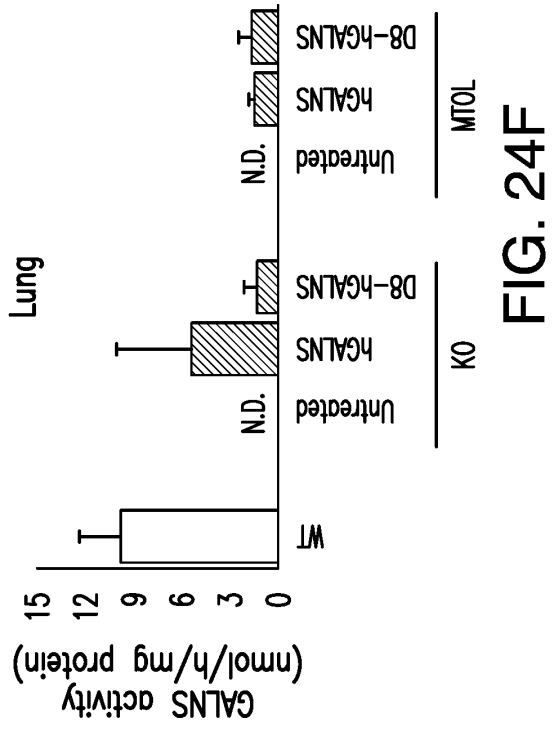


FIG. 24F

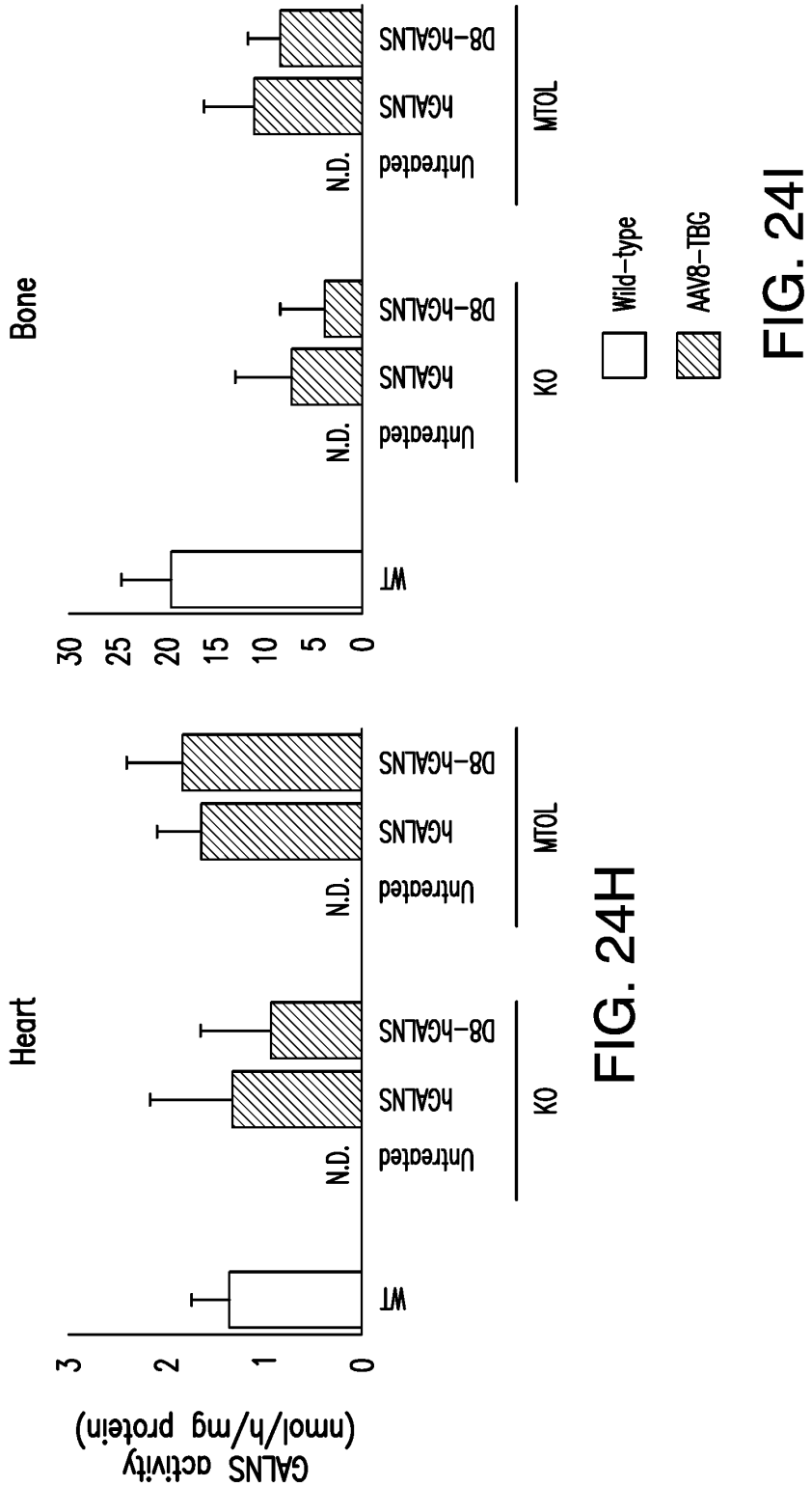


FIG. 24H

FIG. 24I

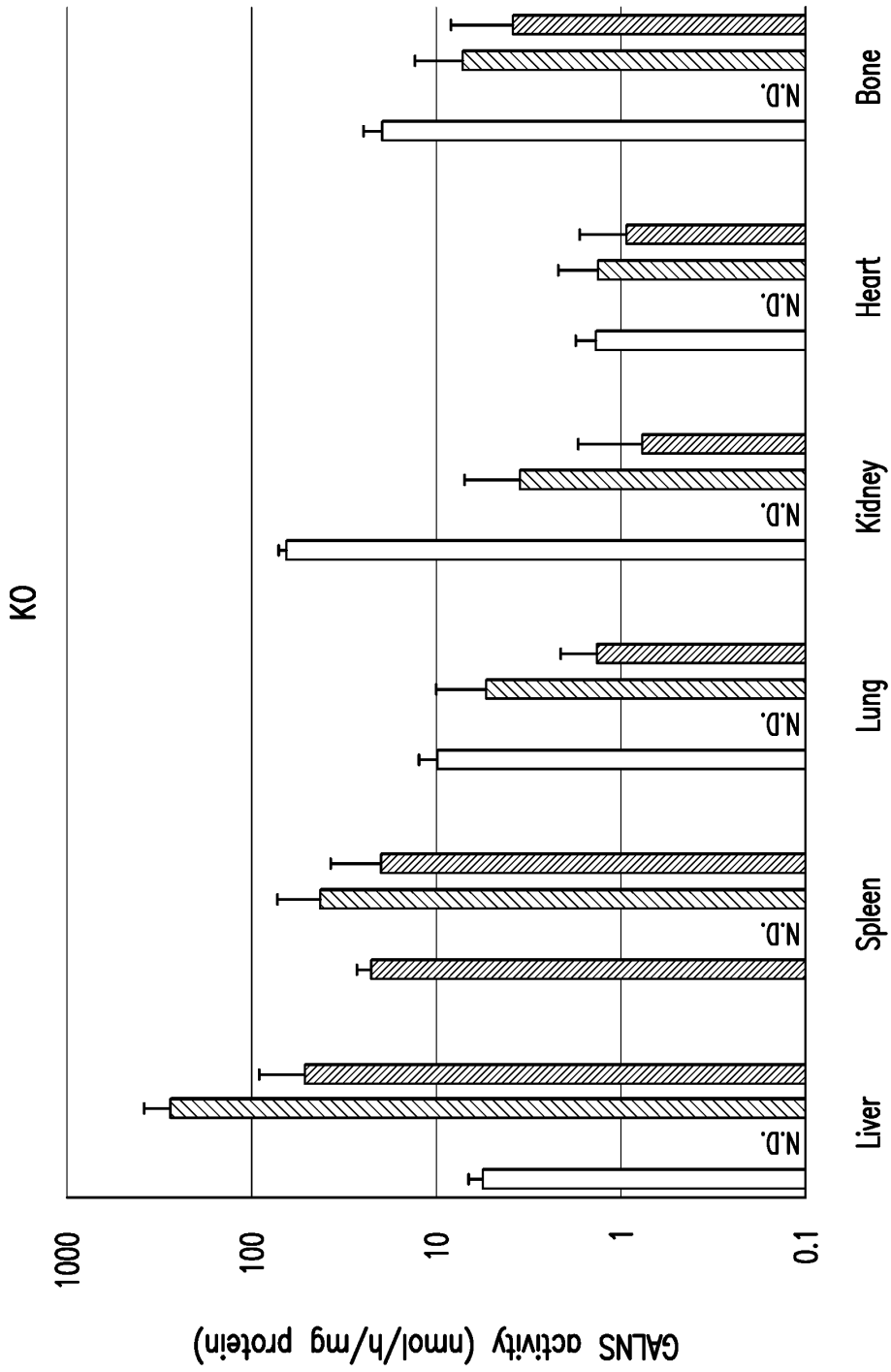

KO
GALNS activity (nmol/h/mg protein)
1000
100
10
1
0.1
Liver
Spleen
Lung
Kidney
Heart
Bone
N.D.
N.D.
N.D.
N.D.
N.D.
N.D.

FIG. 24J

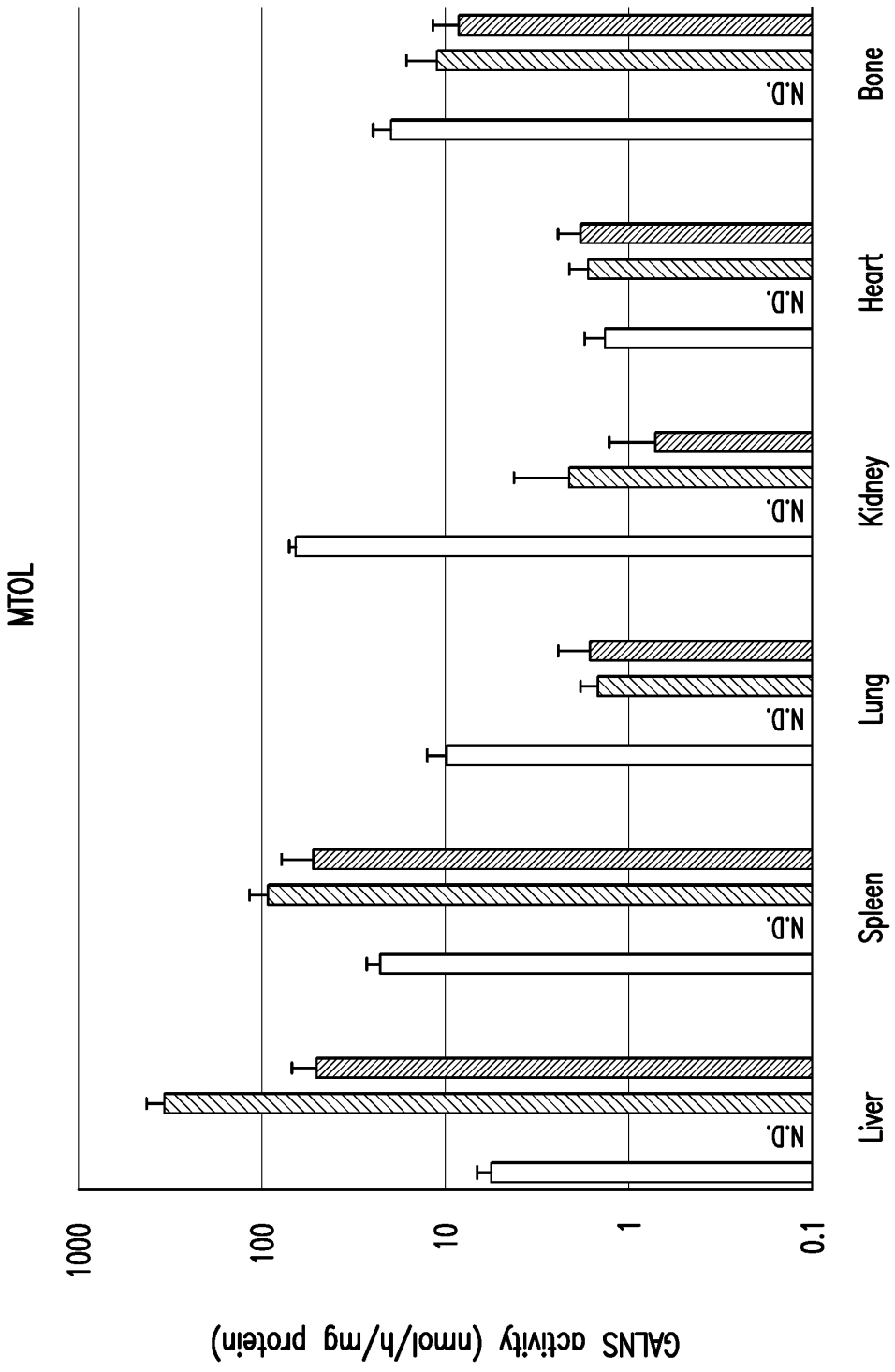

MTOL
GALNS activity (nmol/h/mg protein)
1000
100
10
1
0.1
Liver
Spleen
Lung
Kidney
Heart
Bone
N.D.
N.D.
N.D.
N.D.
N.D.

FIG. 24K

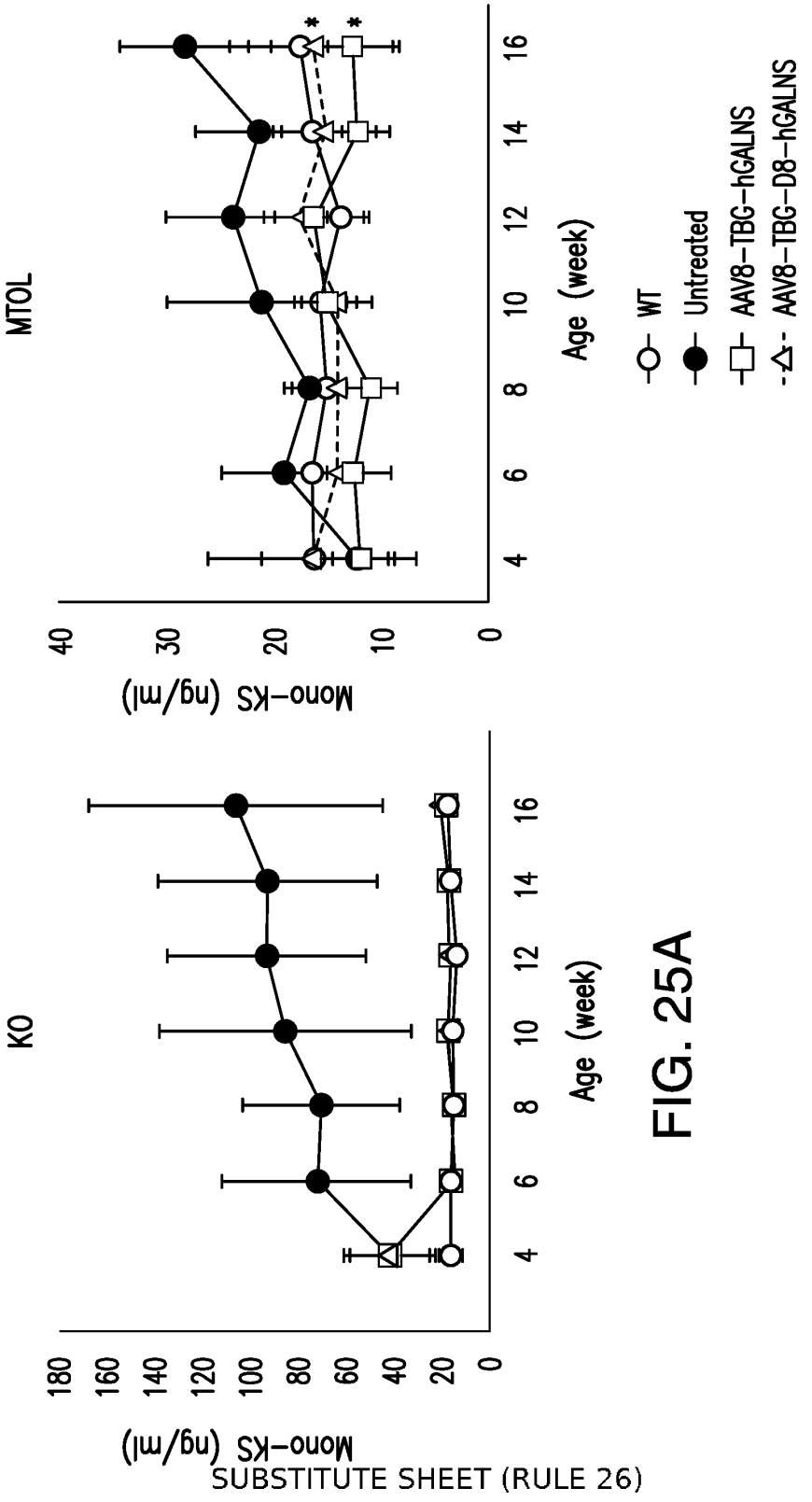


FIG. 25A

FIG. 25B

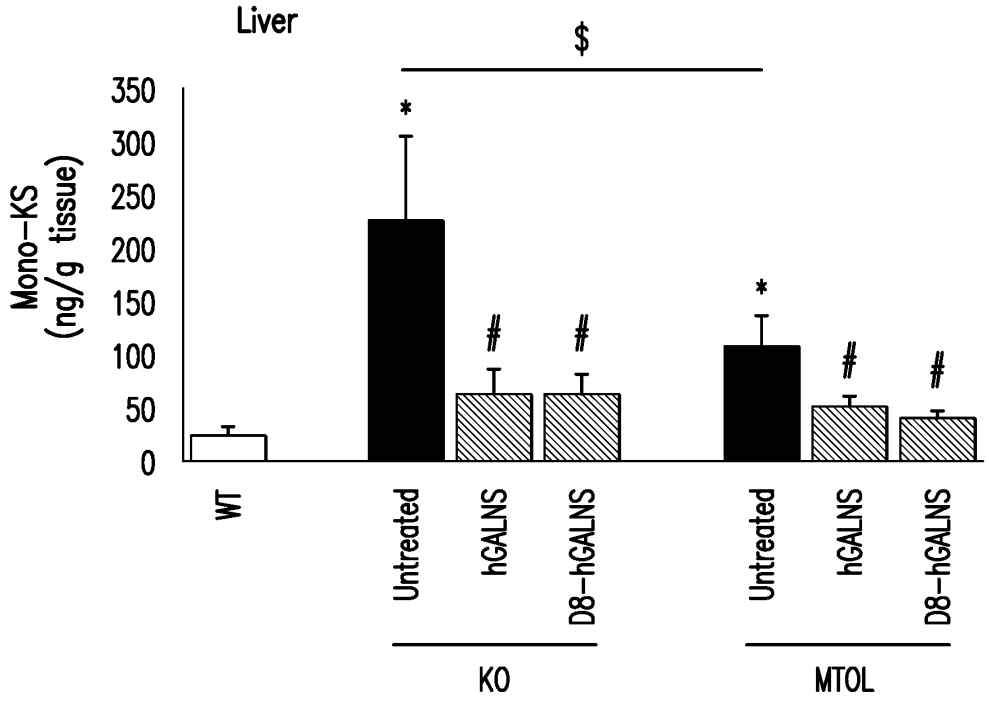


FIG. 25C

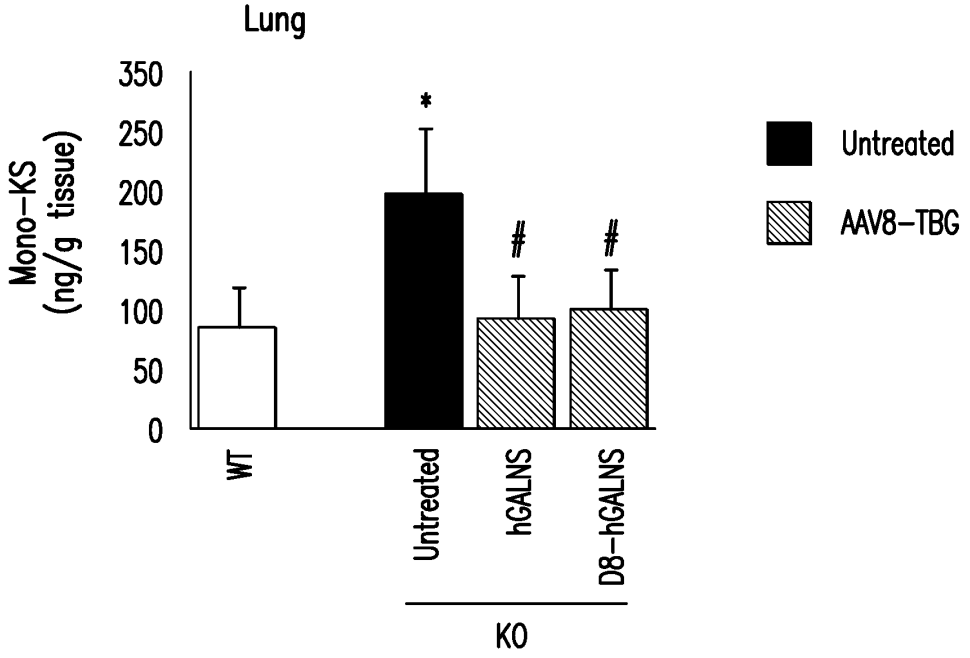


FIG. 25D

FIG. 26A

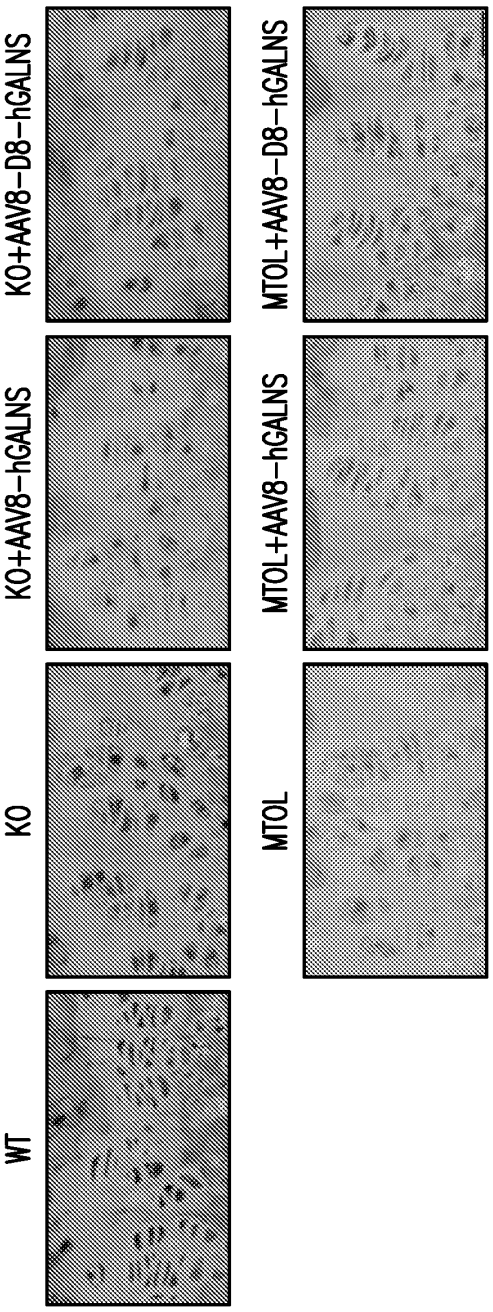
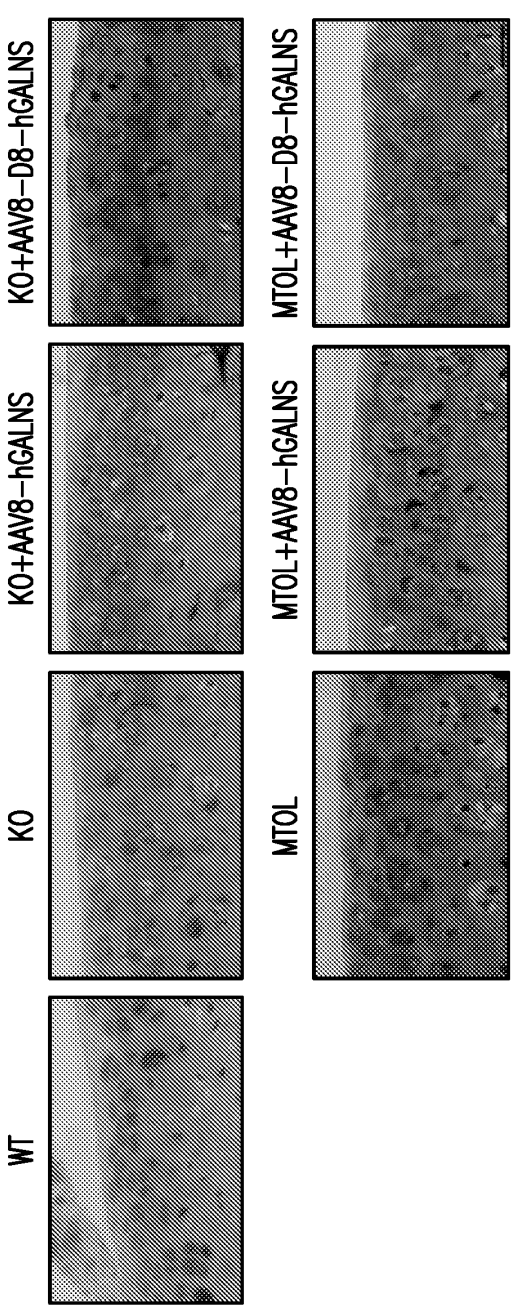


FIG. 26B



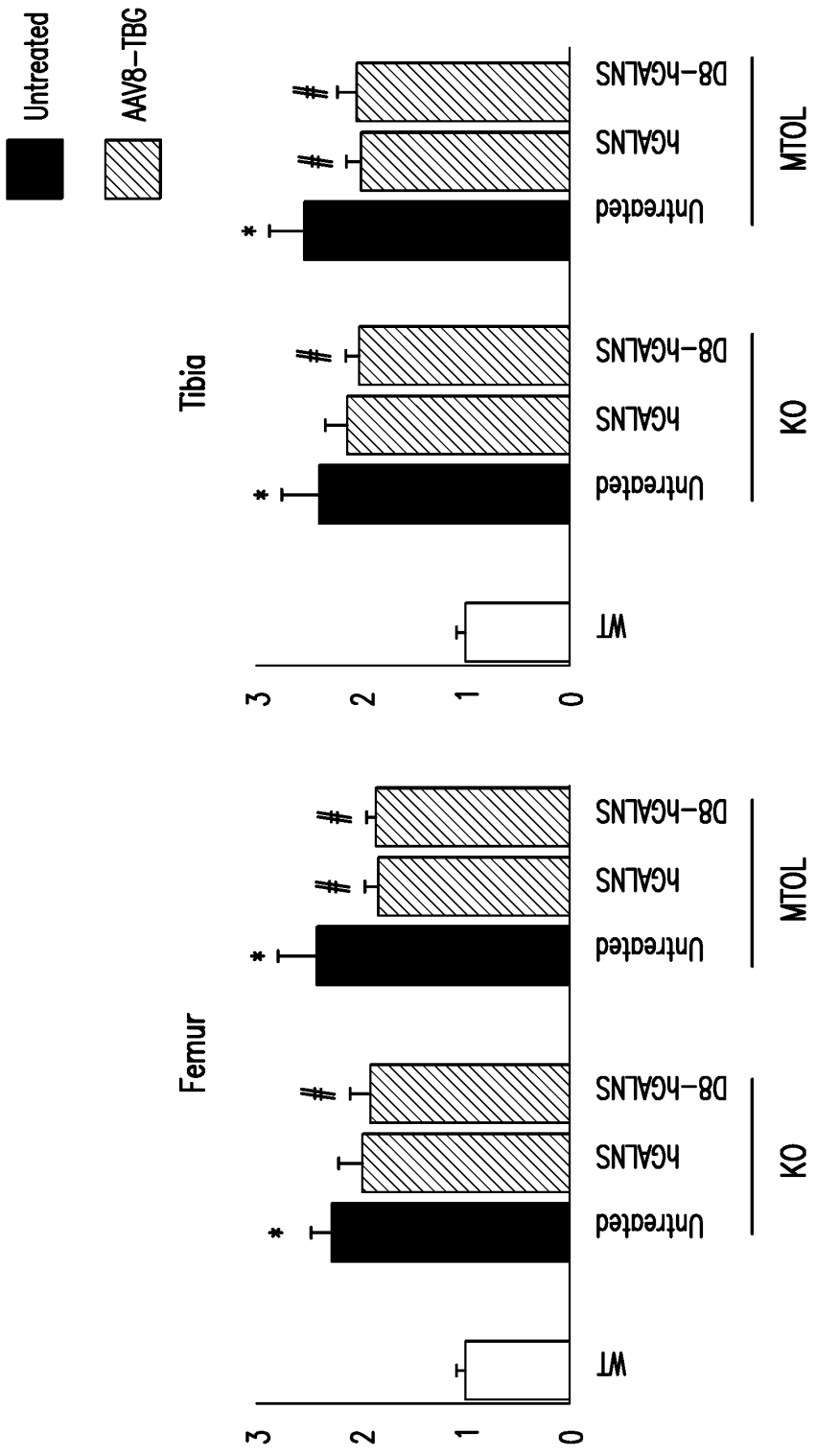

Untreated
AAV8-TBG
Tibia
Femur
Chondrocyte size in growth plate
(arbitrary unit)
0
1
2
3
WT
Untreated
hGALNS
D8-hGALNS
KO
MTOL
*
#

FIG. 26C

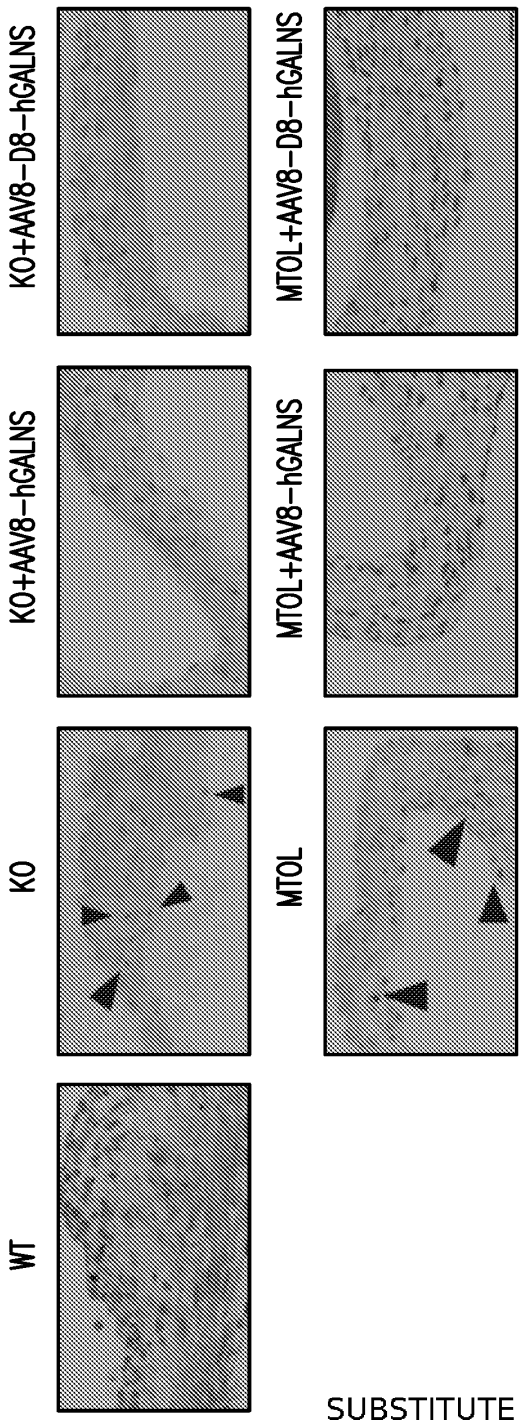


FIG. 27A

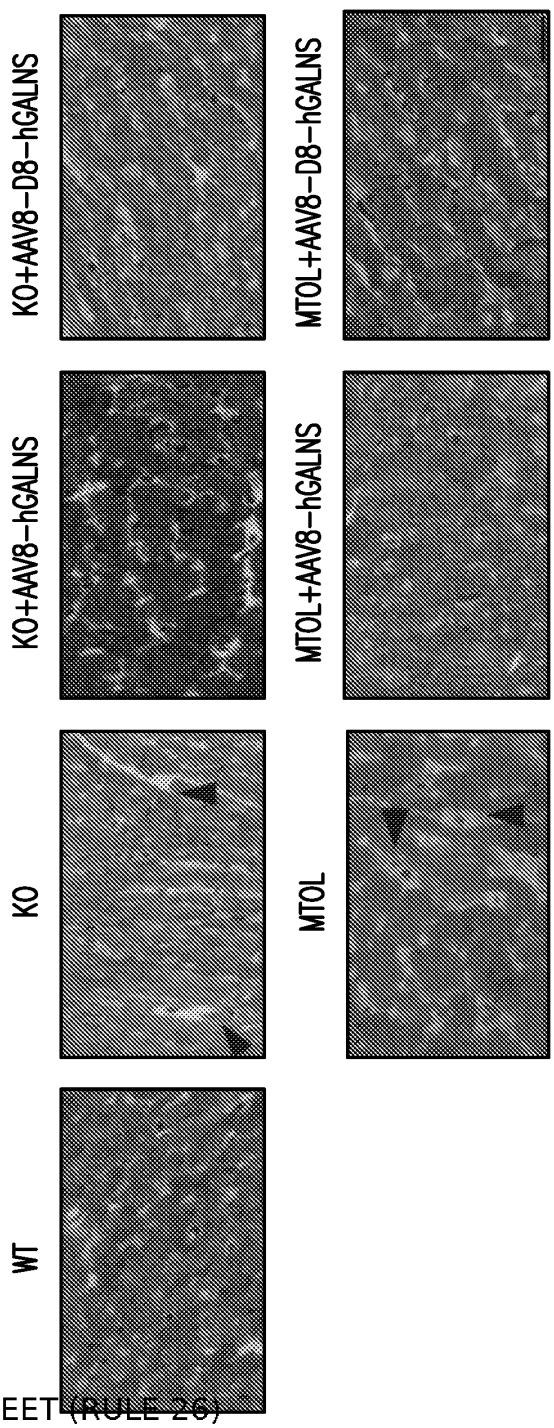


FIG. 27B

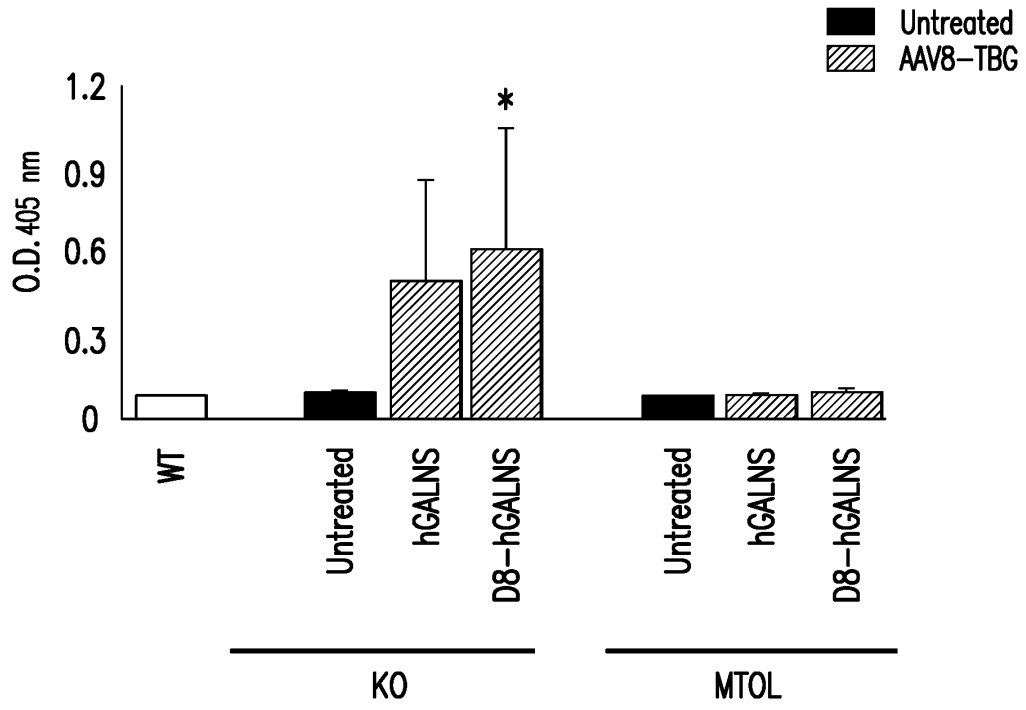

Untreated
AAV8-TBG
O.D. 405 nm
1.2
0.9
0.6
0.3
0
*
WT
Untreated
hGALNS
D8-hGALNS
Untreated
hGALNS
D8-hGALNS
KO
MTOL

FIG. 28

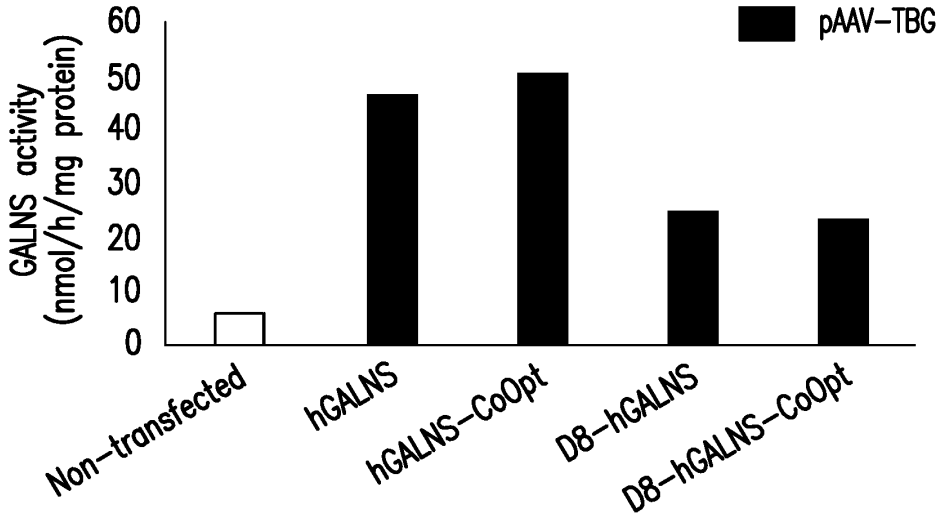


FIG. 29A

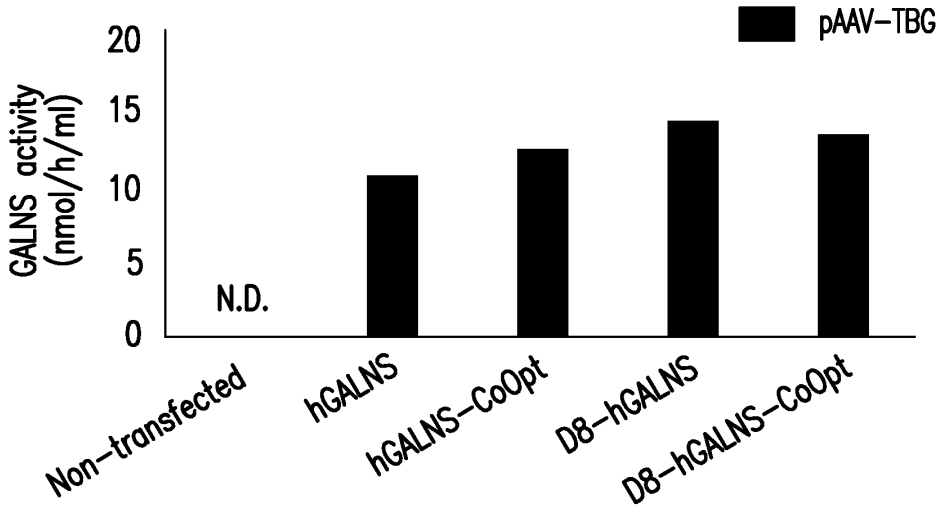


FIG. 29B

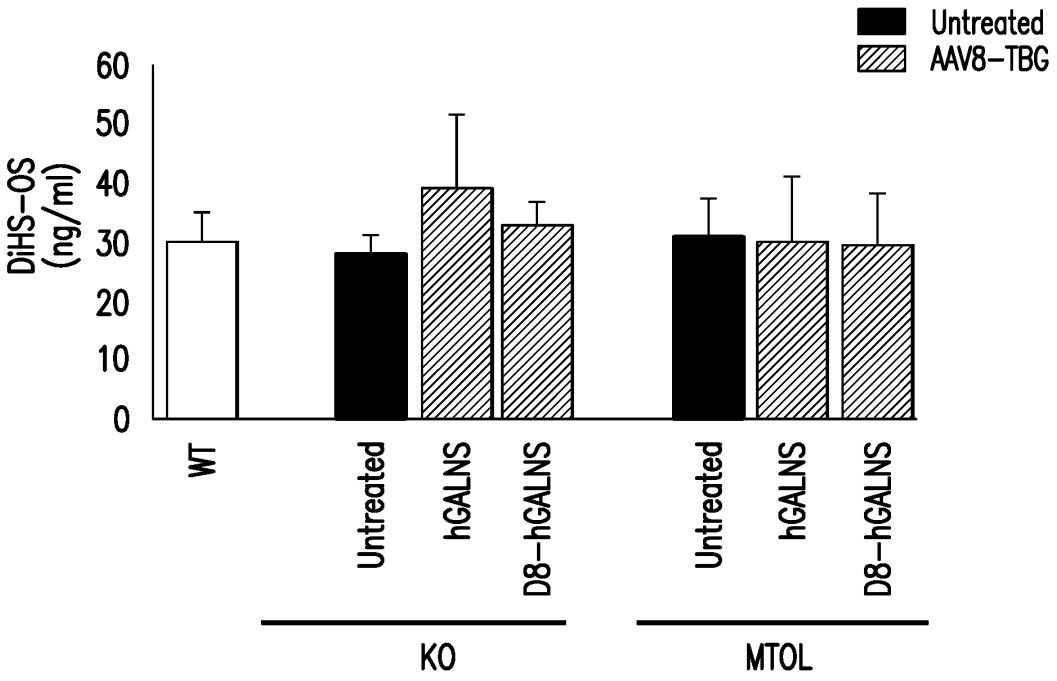

Untreated
AAV8-TBG
DiHS-OS (ng/ml)
60
50
40
30
20
10
0
WT
Untreated
hGALNS
D8-hGALNS
Untreated
hGALNS
D8-hGALNS
KO
MTOL

FIG. 30

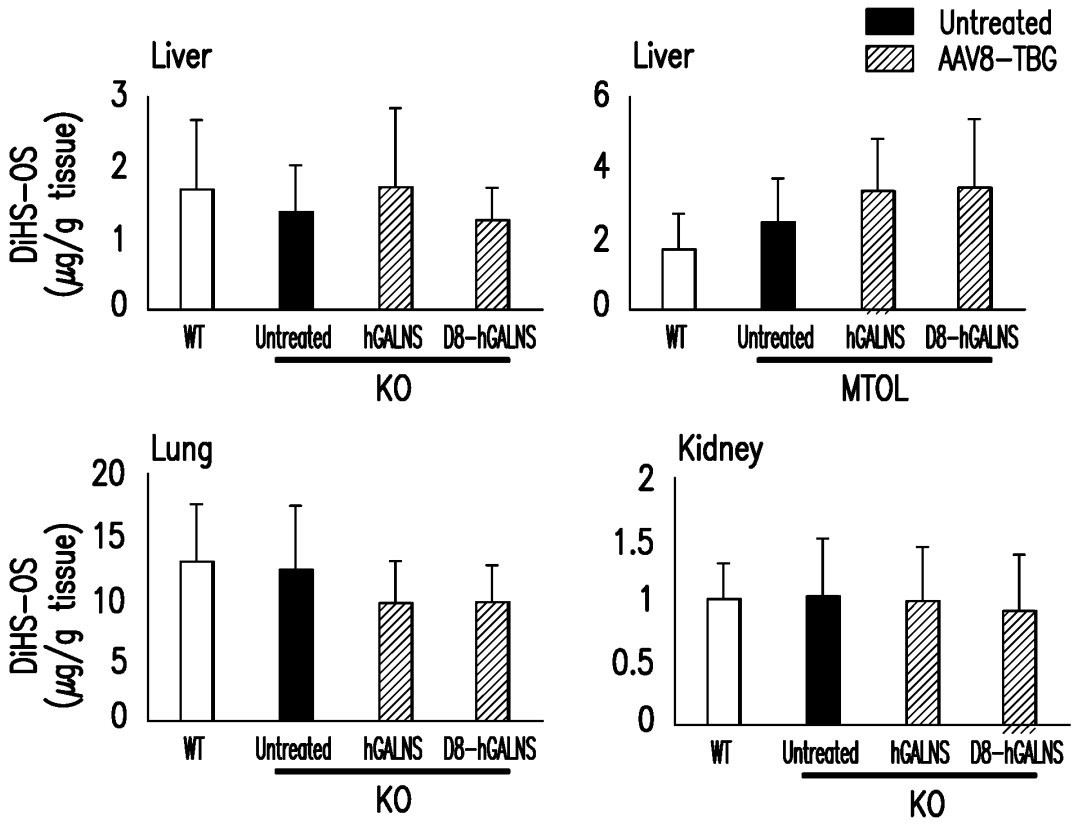

Liver
DiHS-OS (μg/g tissue)
3
2
1
0
WT
Untreated
hGALNS
D8-hGALNS
KO
Liver
Untreated
AAV8-TBG
6
4
2
0
WT
Untreated
hGALNS
D8-hGALNS
MTOL
Lung
DiHS-OS (μg/g tissue)
20
15
10
5
0
WT
Untreated
hGALNS
D8-hGALNS
KO
Kidney
2
1.5
1
0.5
0
WT
Untreated
hGALNS
D8-hGALNS
KO

FIG. 31

FIG. 32A

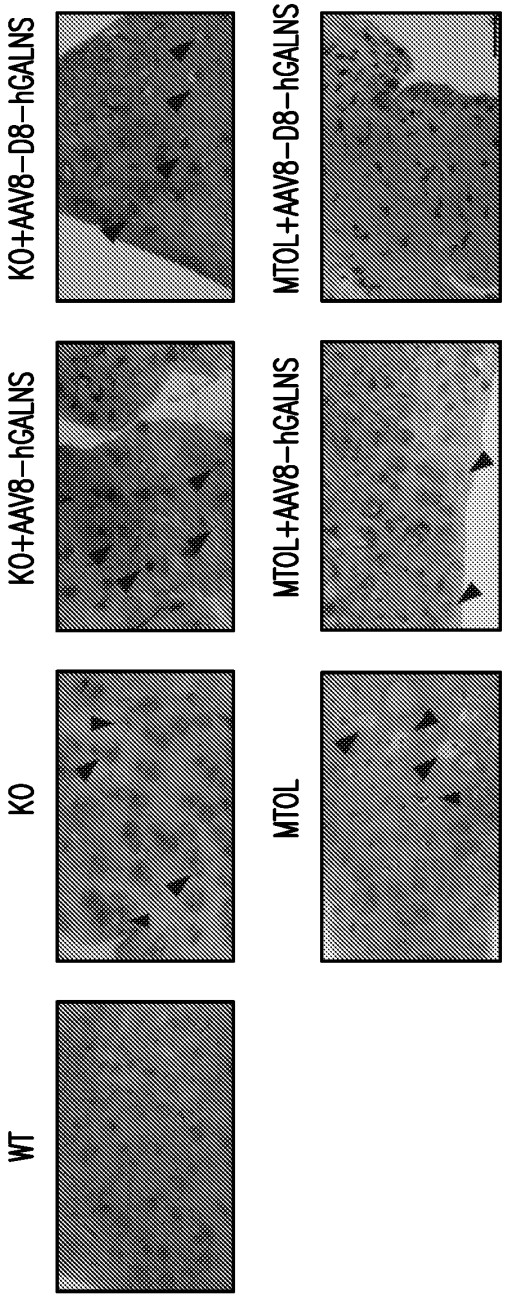
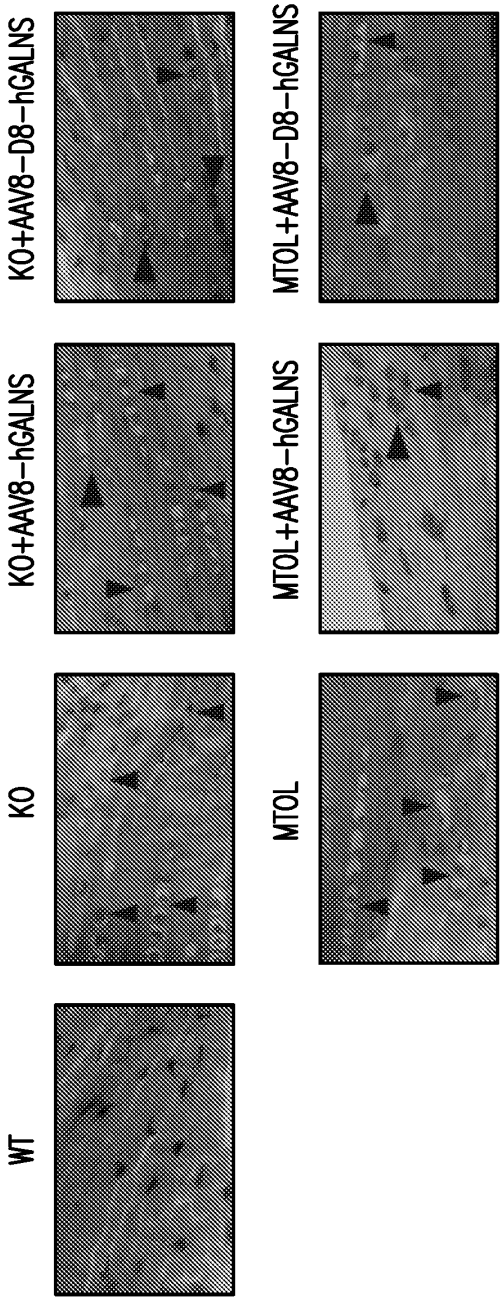


FIG. 32B



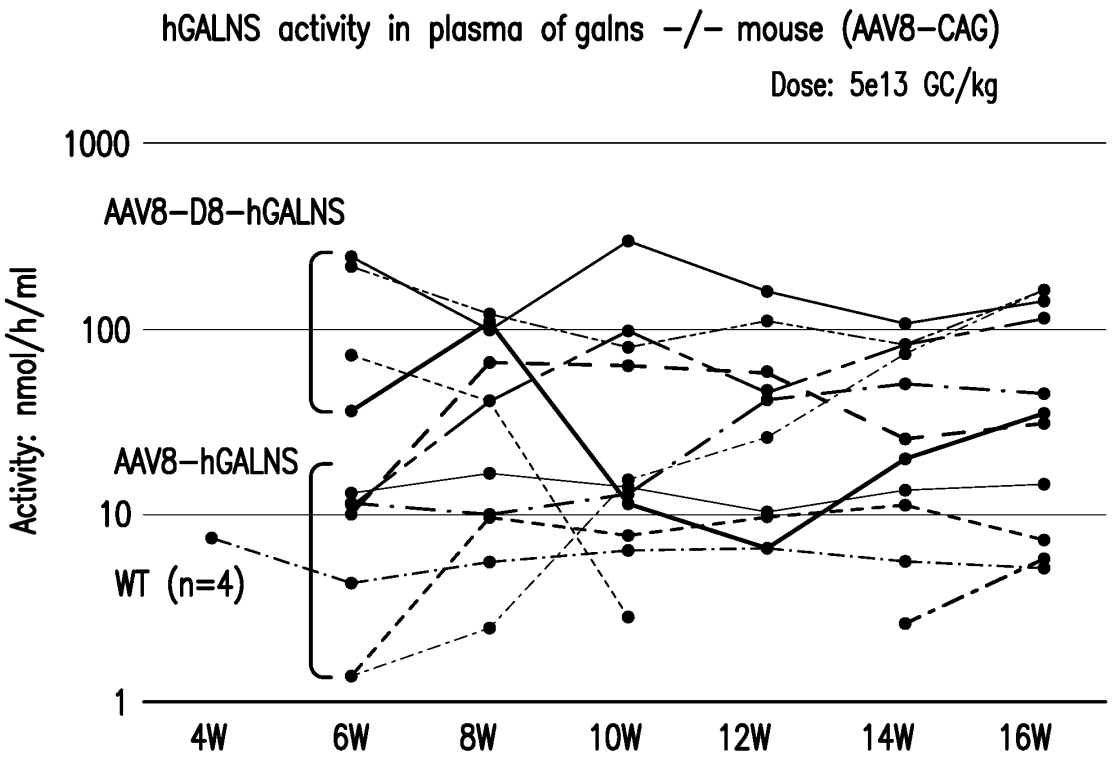

hGALNS activity in plasma of galns −/− mouse (AAV8−CAG)
Dose: 5e13 GC/kg
1000
100
10
1
AAV8−D8−hGALNS
AAV8−hGALNS
WT (n=4)
Activity: nmol/h/ml
4W
6W
8W
10W
12W
14W
16W

FIG. 33

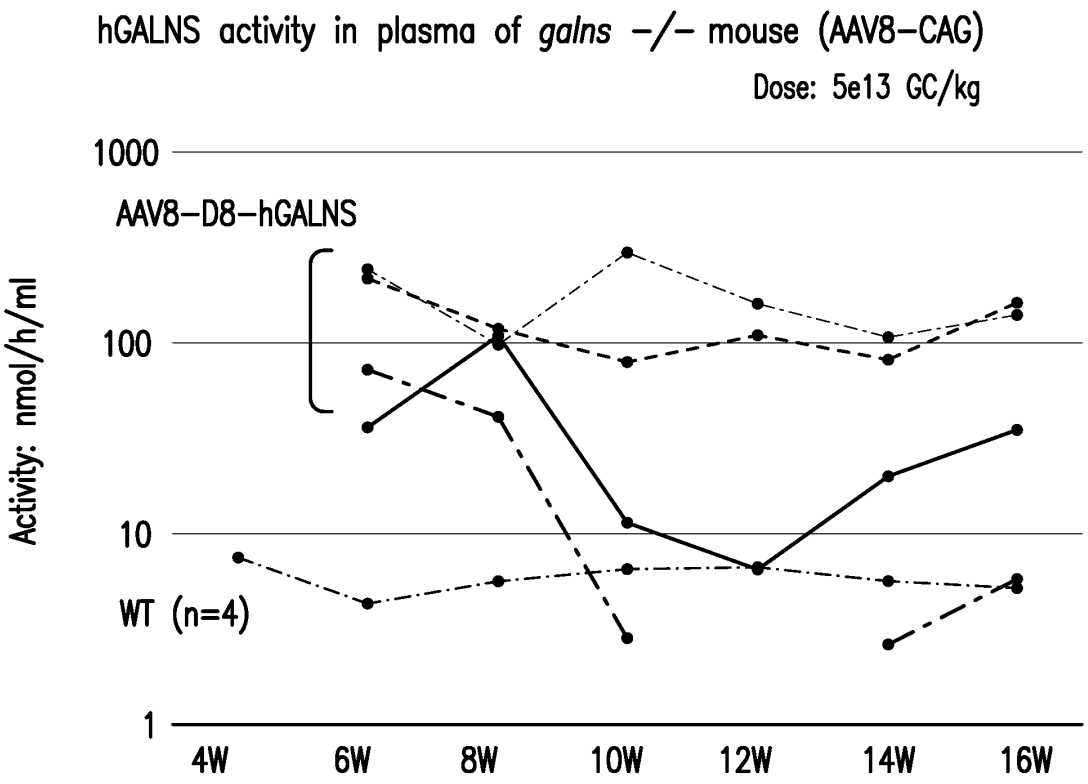

hGALNS activity in plasma of galns −/− mouse (AAV8−CAG)
Dose: 5e13 GC/kg
1000
AAV8−D8−hGALNS
100
10
WT (n=4)
1
Activity: nmol/h/ml
4W
6W
8W
10W
12W
14W
16W

FIG. 34

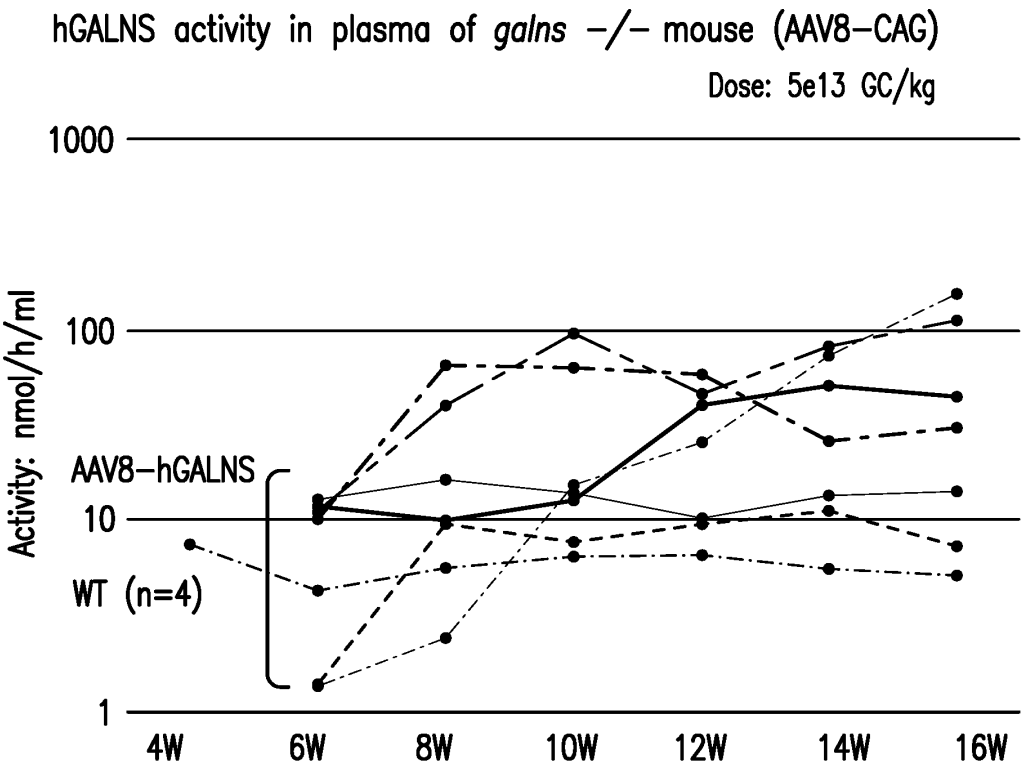

hGALNS activity in plasma of galns −/− mouse (AAV8−CAG)
Dose: 5e13 GC/kg
1000
100
10
1
Activity: nmol/h/ml
AAV8−hGALNS
WT (n=4)
4W
6W
8W
10W
12W
14W
16W

FIG. 35

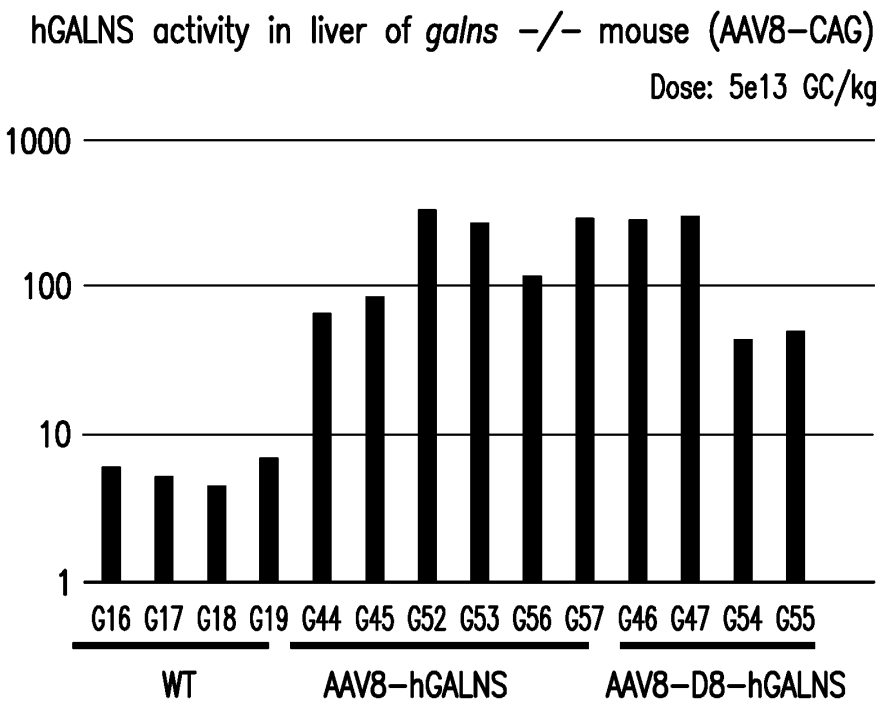

hGALNS activity in liver of galns −/− mouse (AAV8−CAG)
Dose: 5e13 GC/kg
1000
100
10
1
G16 G17 G18 G19 G44 G45 G52 G53 G56 G57 G46 G47 G54 G55
WT
AAV8−hGALNS
AAV8−D8−hGALNS

FIG. 36

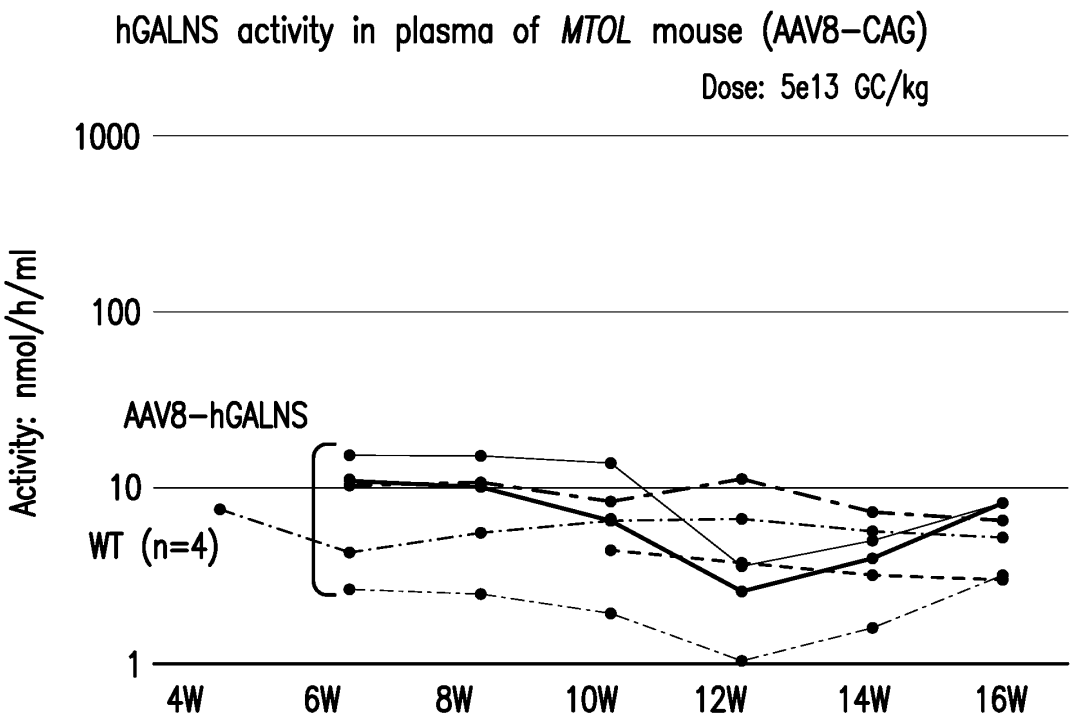

hGALNS activity in plasma of MTOL mouse (AAV8-CAG)
Dose: 5e13 GC/kg
1000
100
10
1
Activity: nmol/h/ml
AAV8-hGALNS
WT (n=4)
4W
6W
8W
10W
12W
14W
16W

FIG. 37

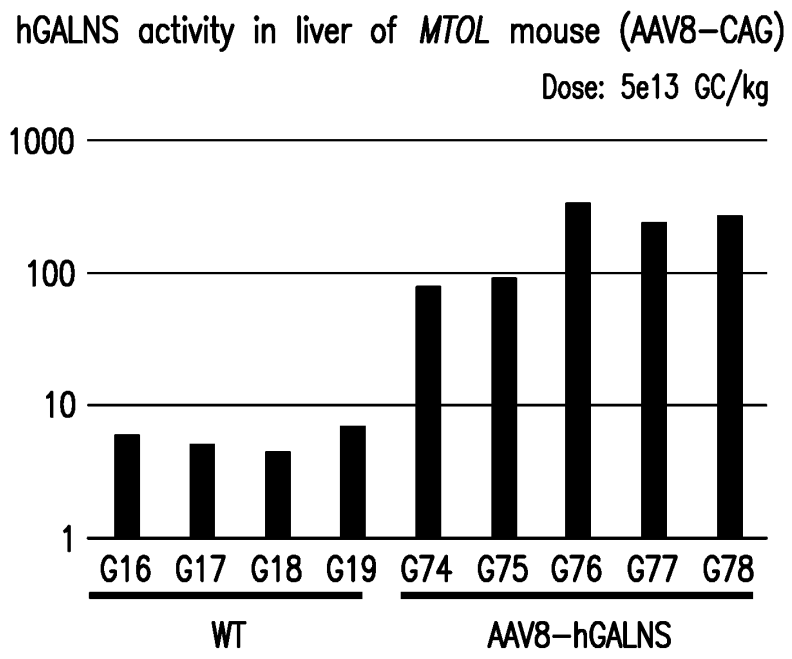

hGALNS activity in liver of *MTOL* mouse (AAV8–CAG)
Dose: 5e13 GC/kg
1000
100
10
1
G16 G17 G18 G19 G74 G75 G76 G77 G78
WT
AAV8–hGALNS

FIG. 38

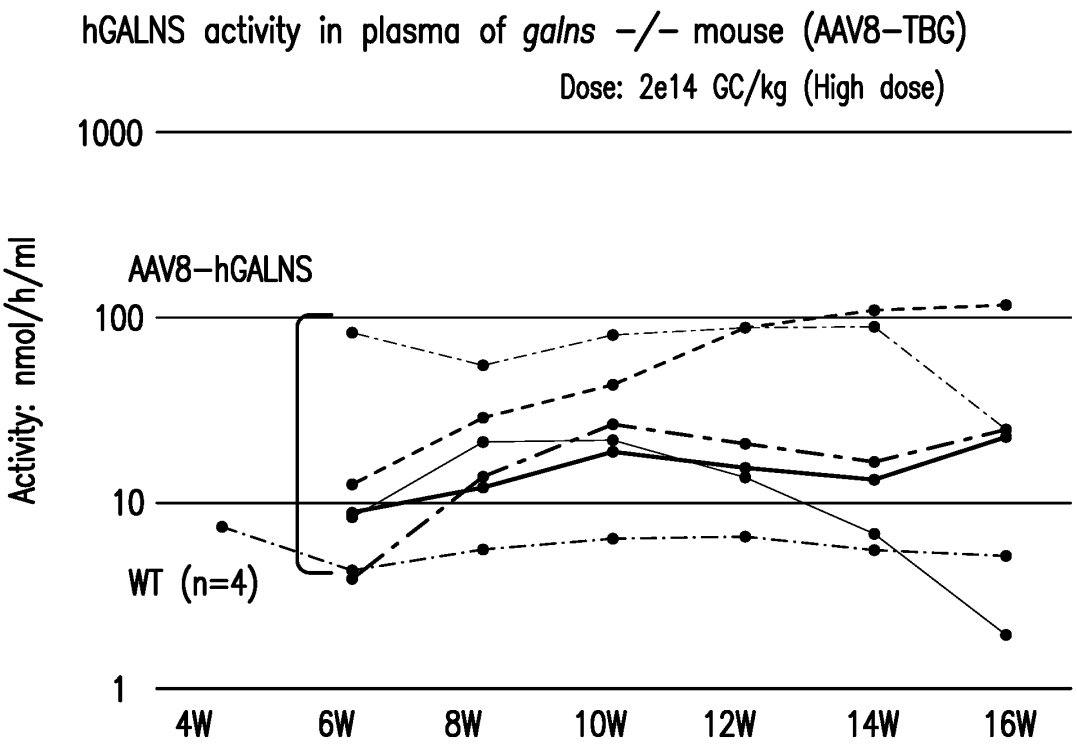

hGALNS activity in plasma of galns −/− mouse (AAV8−TBG)
Dose: 2e14 GC/kg (High dose)
1000
AAV8−hGALNS
100
10
WT (n=4)
1
Activity: nmol/h/ml
4W
6W
8W
10W
12W
14W
16W

FIG. 39

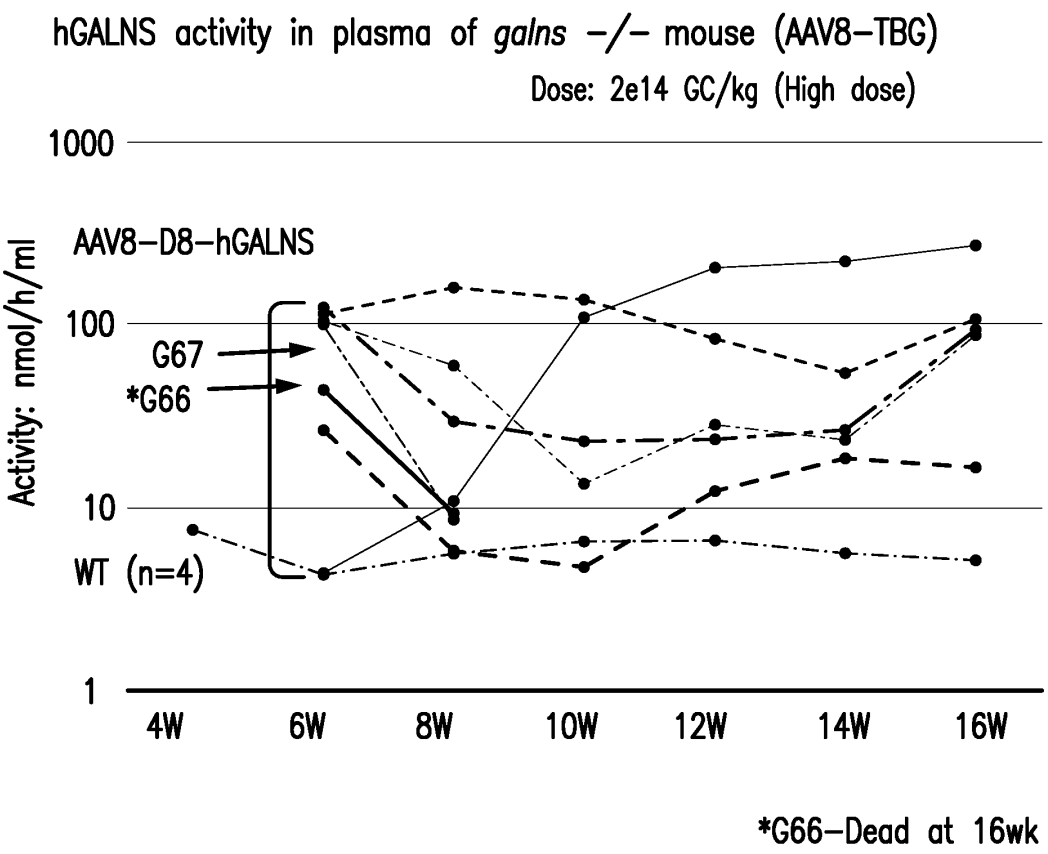

hGALNS activity in plasma of galns −/− mouse (AAV8−TBG)
Dose: 2e14 GC/kg (High dose)
1000
100
10
1
Activity: nmol/h/ml
AAV8−D8−hGALNS
G67
*G66
WT (n=4)
4W
6W
8W
10W
12W
14W
16W
*G66−Dead at 16wk

FIG. 40

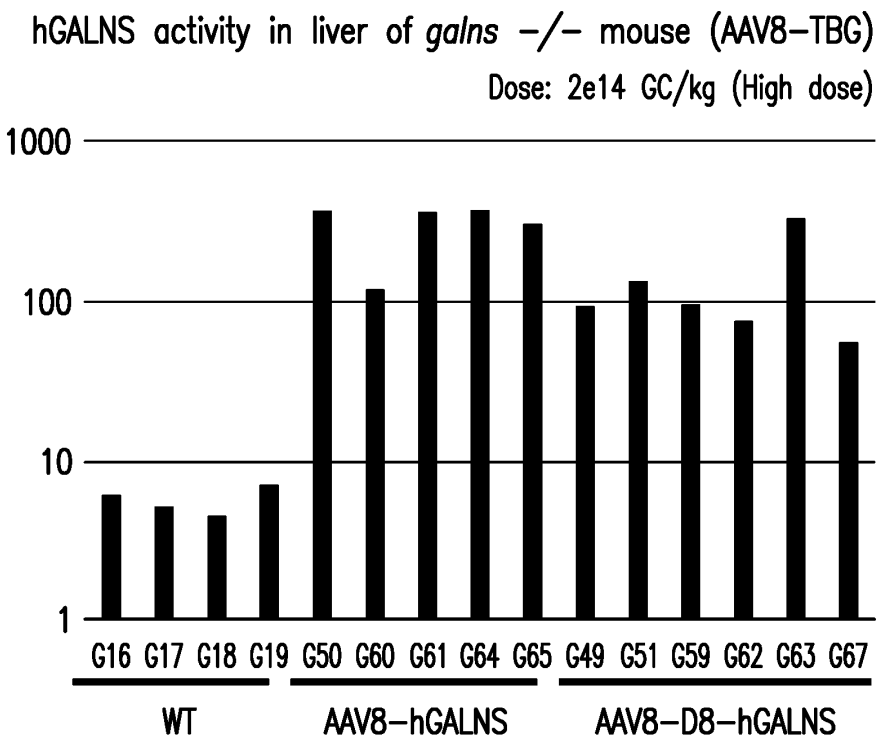

hGALNS activity in liver of galns −/− mouse (AAV8−TBG)
Dose: 2e14 GC/kg (High dose)
1000
100
10
1
G16 G17 G18 G19 G50 G60 G61 G64 G65 G49 G51 G59 G62 G63 G67
WT
AAV8−hGALNS
AAV8−D8−hGALNS

FIG. 41