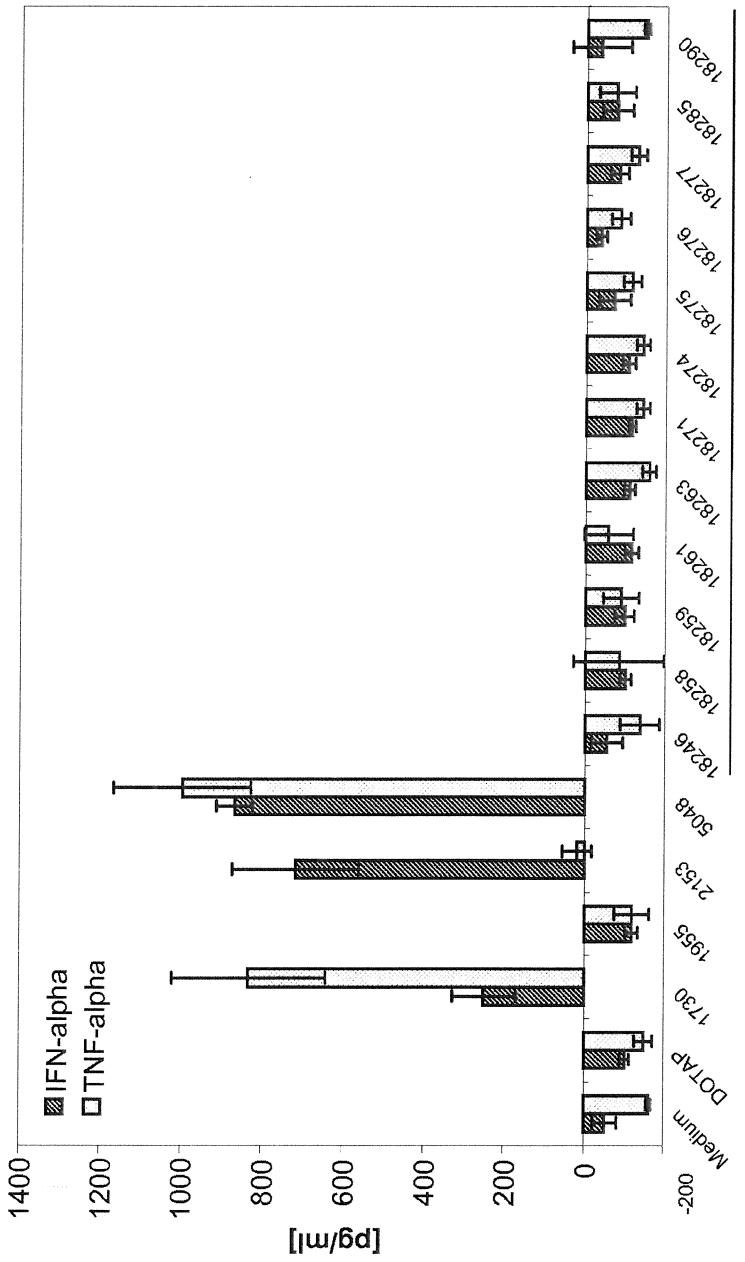

IFN-alpha
TNF-alpha
[pg/ml]
1400
1200
1000
800
600
400
200
0
-200
Medium
DOTAP
1730
1955
2153
5048
18246
18258
18259
18261
18263
18271
18274
18275
18276
18277
18285
18290
candidate siRNAs

FIG. 1

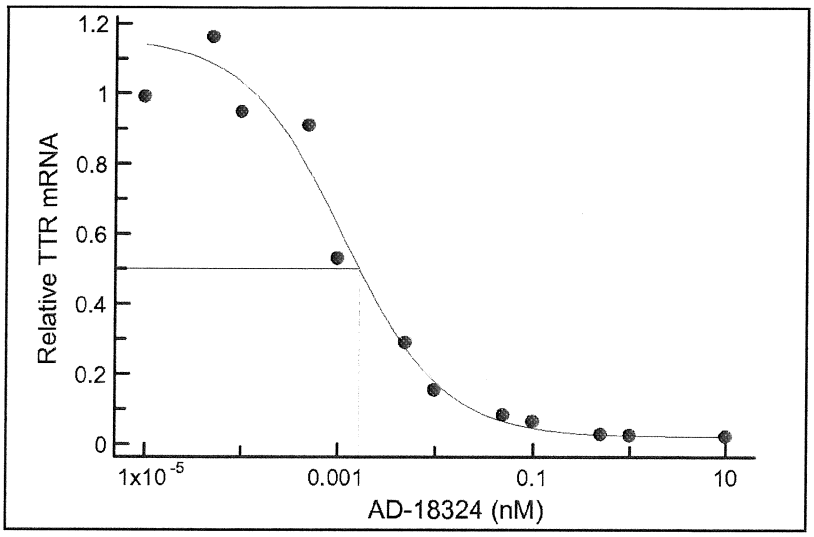

1.2
1
0.8
0.6
0.4
0.2
0
Relative TTR mRNA
1x10 -5
0.001
0.1
10
AD-18324 (nM)

FIG. 2A

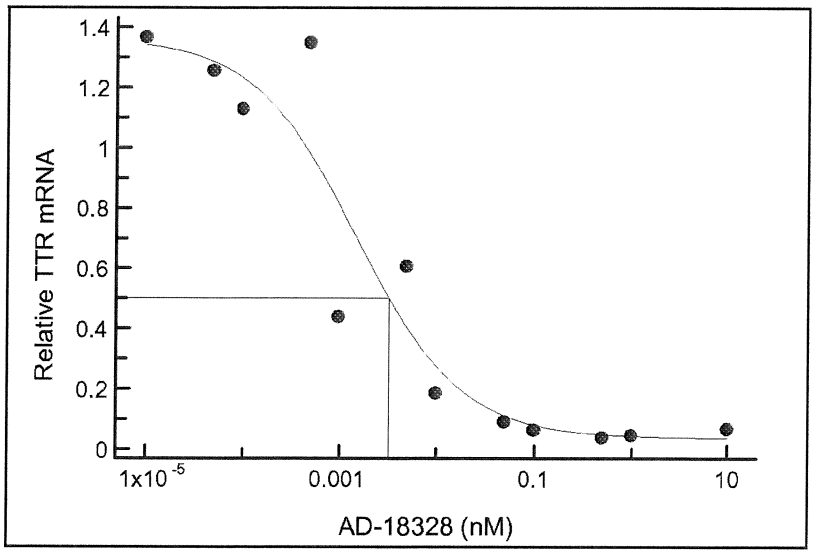

1.4
1.2
1
0.8
0.6
0.4
0.2
0
Relative TTR mRNA
1x10 -5
0.001
0.1
10
AD-18328 (nM)

FIG. 2B

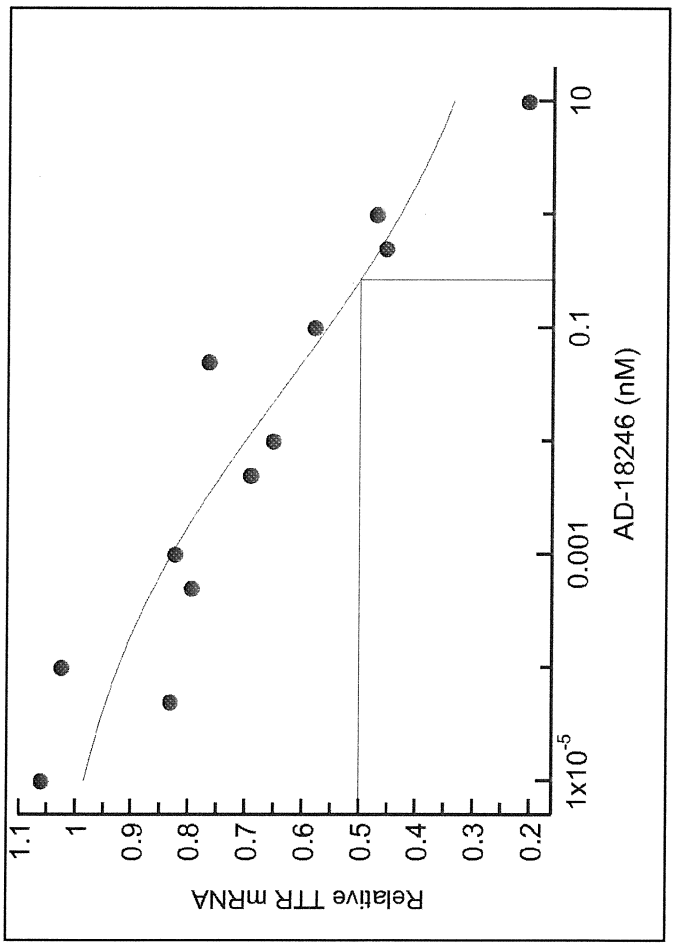

Relative TTR mRNA
1.1
1
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
1x10-5
0.001
0.1
10
AD-18246 (nM)

FIG. 3

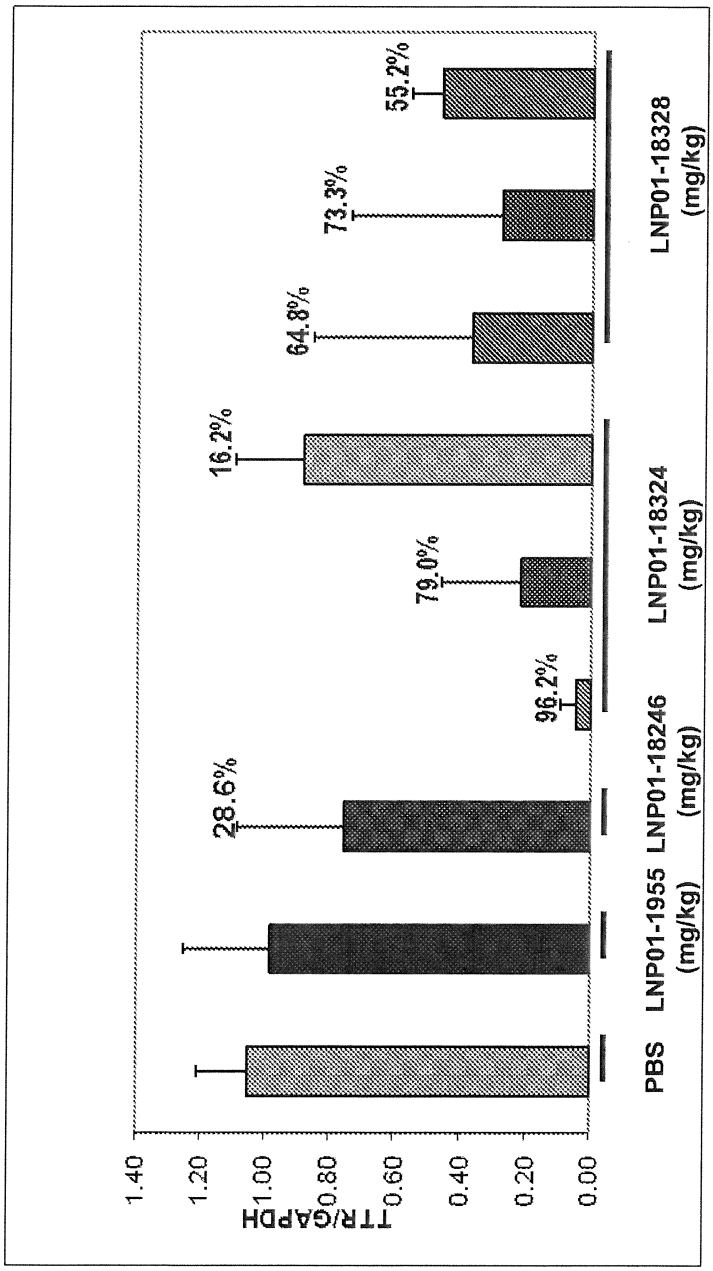

1.40
1.20
1.00
0.80
0.60
0.40
0.20
0.00
TTR/GAPDH
28.6%
16.2%
64.8%
73.3%
55.2%
96.2%
79.0%
PBS
LNP01-1955 (mg/kg)
LNP01-18246 (mg/kg)
LNP01-18324 (mg/kg)
LNP01-18328 (mg/kg)

FIG. 4A

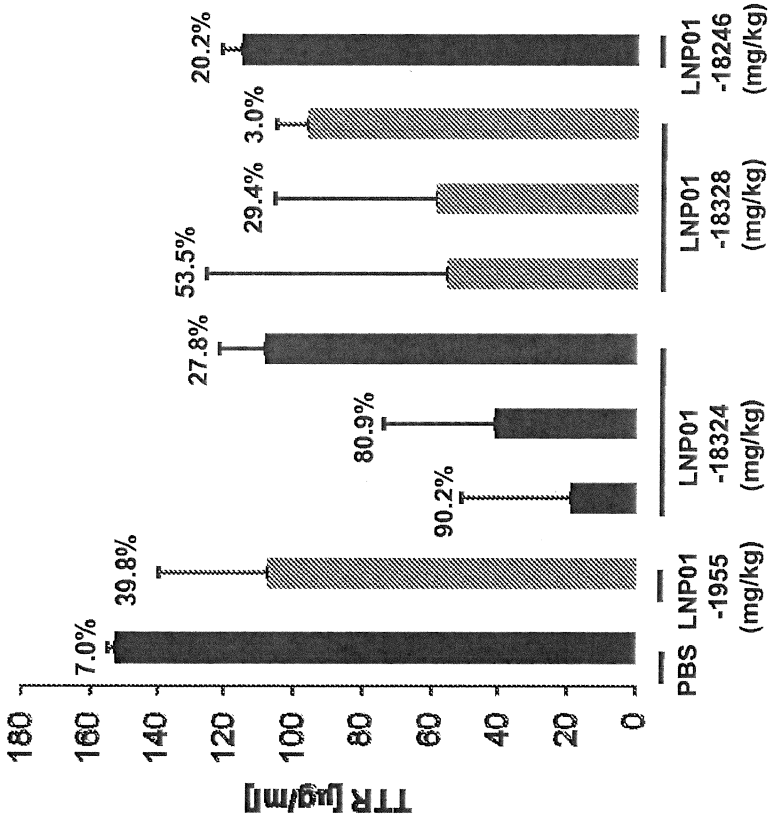

TTR [µg/ml]
180
160
140
120
100
80
60
40
20
0
7.0%
39.8%
90.2%
80.9%
27.8%
53.5%
29.4%
3.0%
20.2%
PBS
LNP01 -1955 (mg/kg)
LNP01 -18324 (mg/kg)
LNP01 -18328 (mg/kg)
LNP01 -18246 (mg/kg)

FIG. 4B

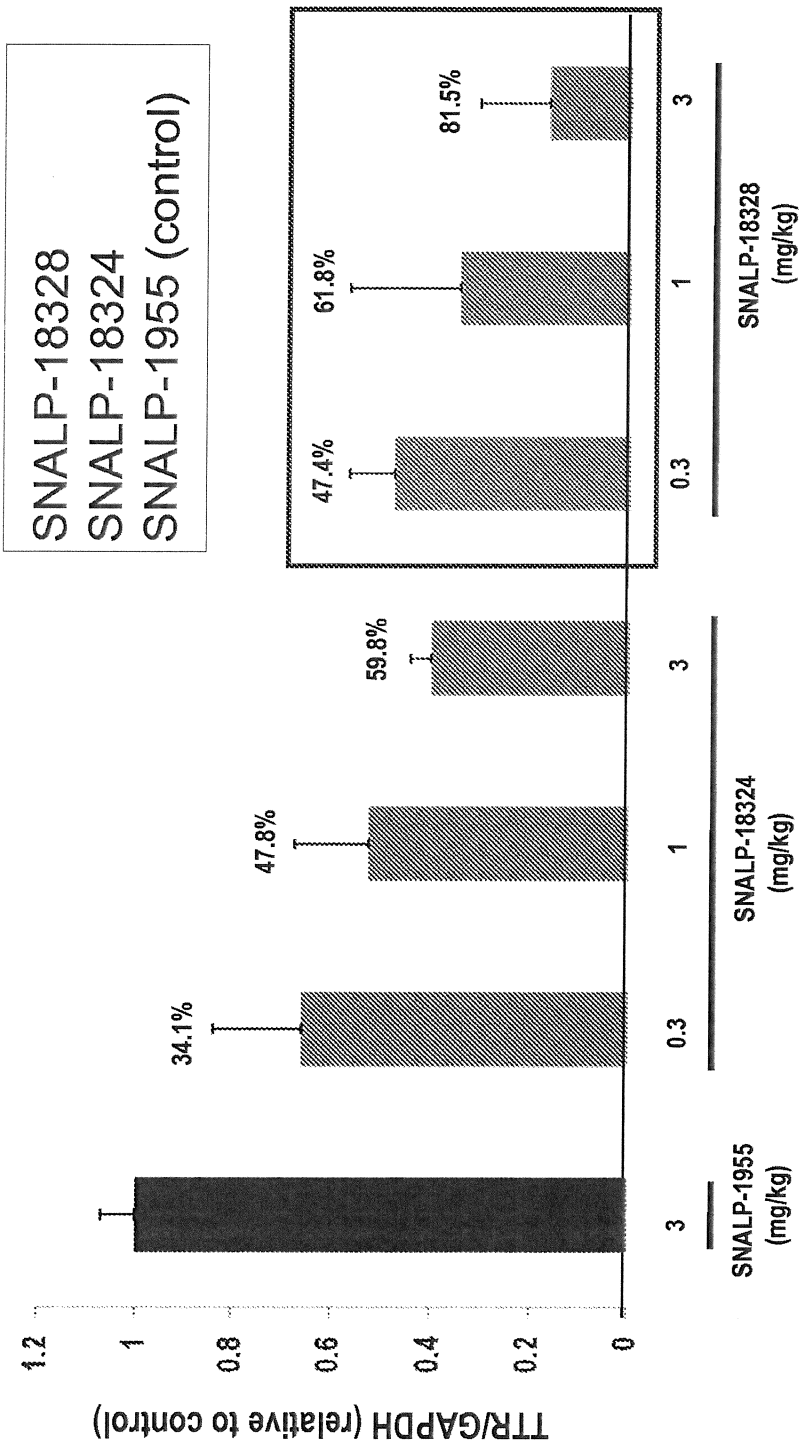

SNALP-18328
SNALP-18324
SNALP-1955 (control)
TTR/GAPDH (relative to control)
1.2
1
0.8
0.6
0.4
0.2
0
34.1%
47.8%
59.8%
47.4%
61.8%
81.5%
3
0.3
1
3
0.3
1
3
SNALP-1955 (mg/kg)
SNALP-18324 (mg/kg)
SNALP-18328 (mg/kg)

FIG. 5

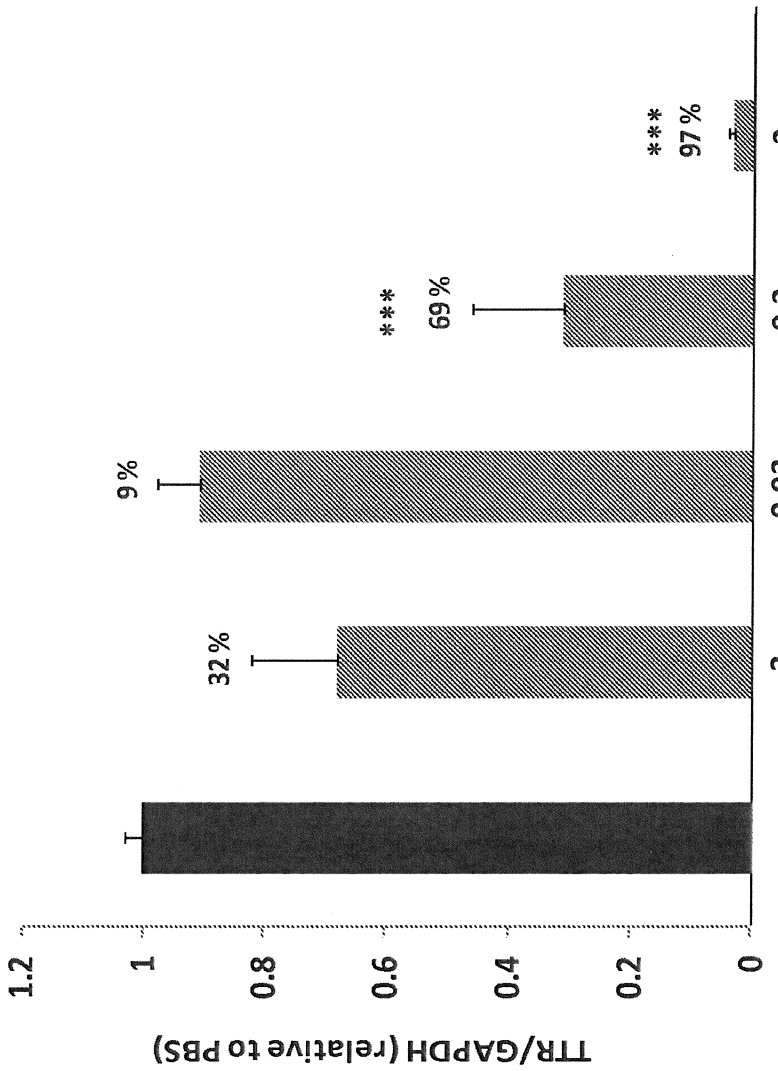

TTR/GAPDH (relative to PBS)
1.2
1
0.8
0.6
0.4
0.2
0
PBS
3
0.03
0.3
3
SNALP-1955 (mg/kg)
SNALP-18328 mg/kg
32 %
9 %
***
69 %
***
97 %

FIG. 6A

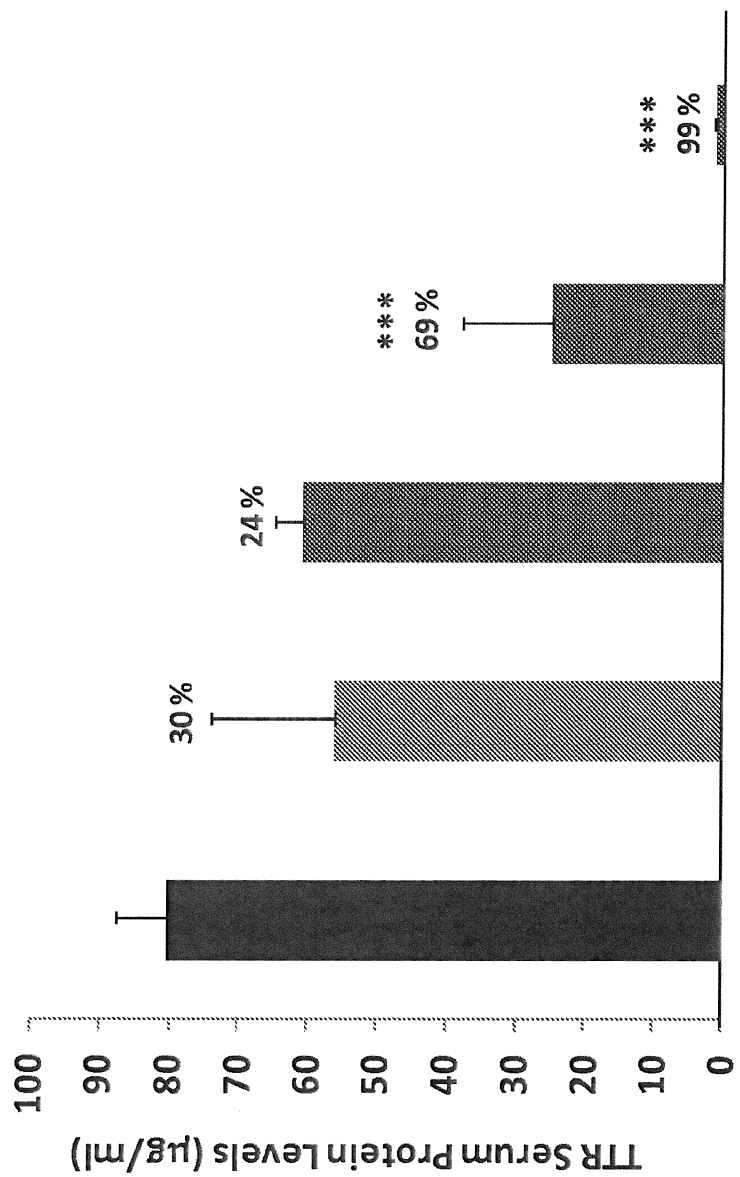

TTR Serum Protein Levels (µg/ml)
100
90
80
70
60
50
40
30
20
10
0
30 %
24 %
***
69 %
***
99 %
PBS
3
0.03
0.3
3
SNALP-1955 (mg/kg)
SNALP-18328 (mg/kg)

FIG. 6B

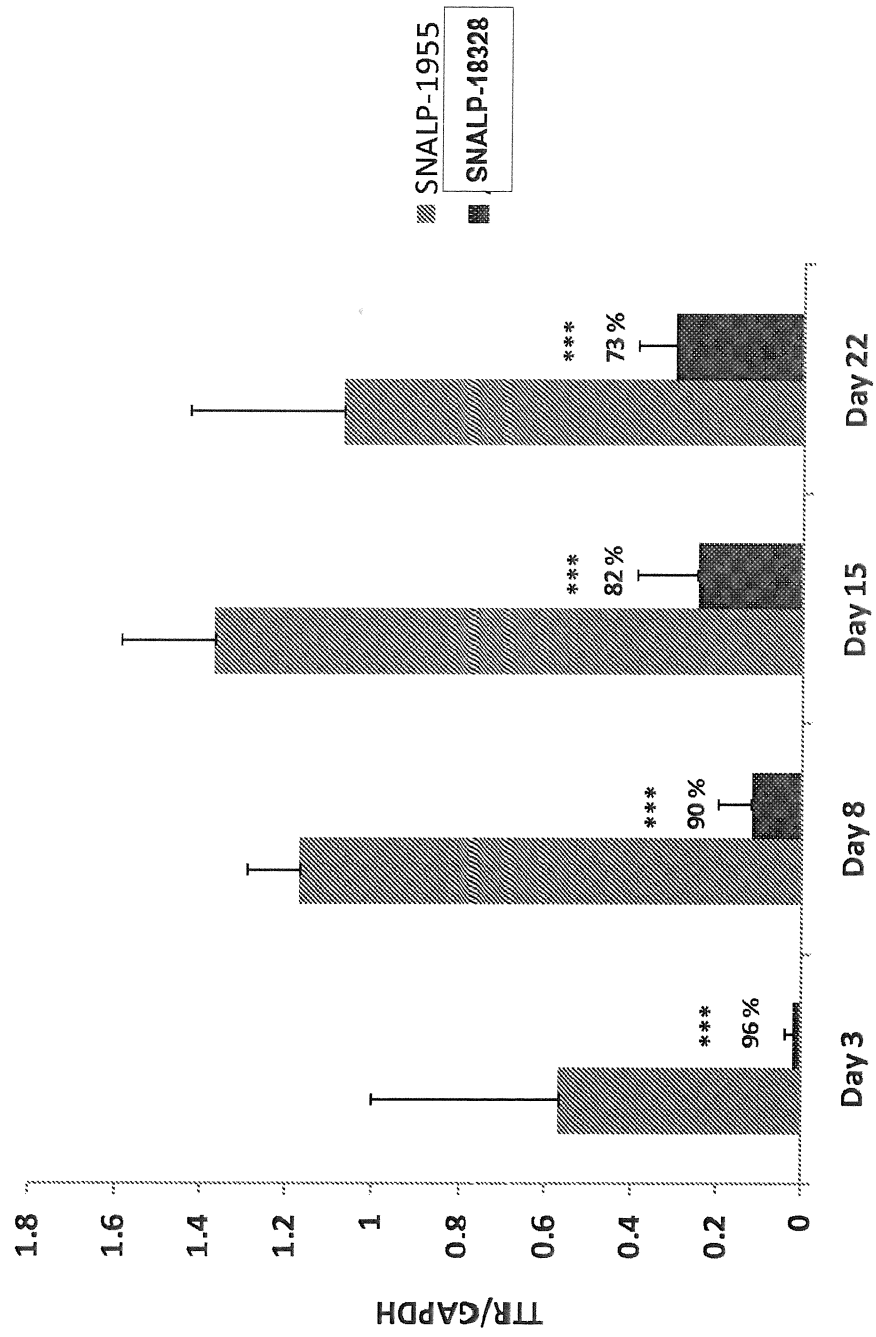

SNALP-1955
SNALP-18328
TTR/GAPDH
1.8
1.6
1.4
1.2
1
0.8
0.6
0.4
0.2
0
Day 3
Day 8
Day 15
Day 22
***
96 %
***
90 %
***
82 %
***
73 %

FIG. 7A

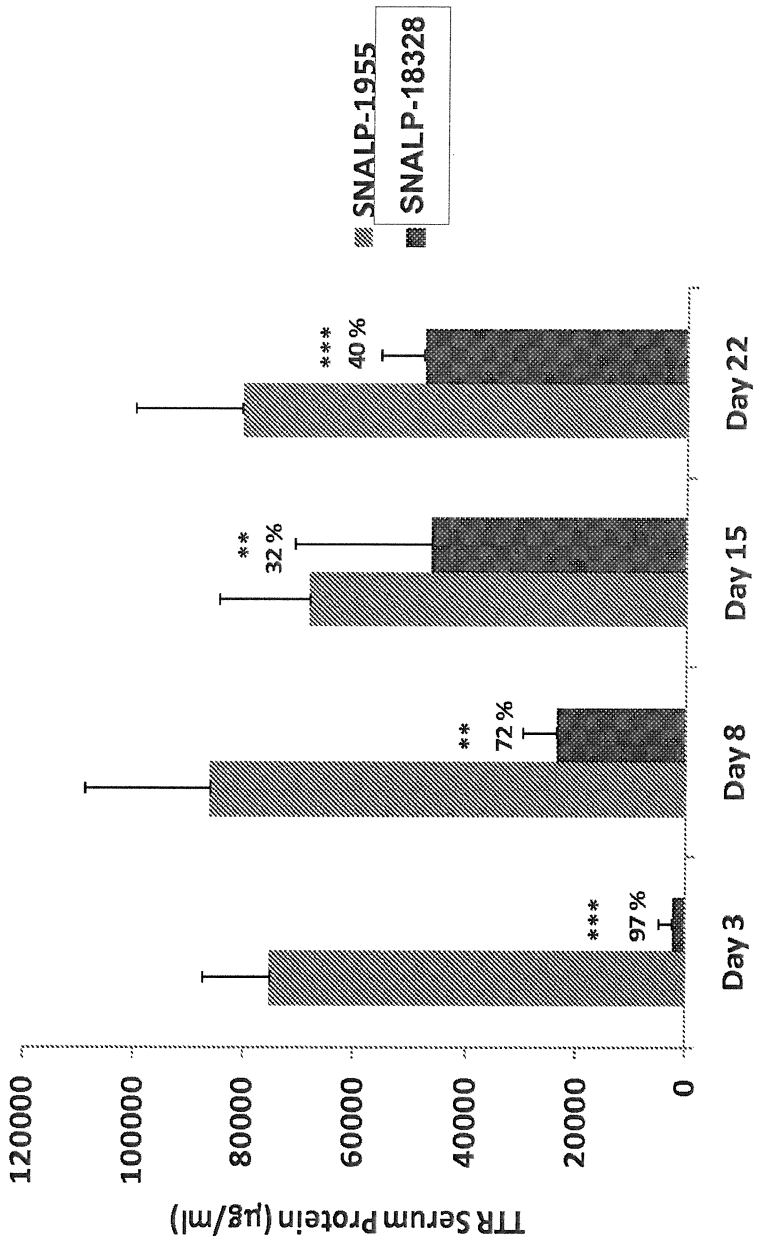

SNALP-1955
SNALP-18328
TTR Serum Protein (µg/ml)
120000
100000
80000
60000
40000
20000
0
Day 3
Day 8
Day 15
Day 22
***
97 %
**
72 %
**
32 %
***
40 %

FIG. 7B

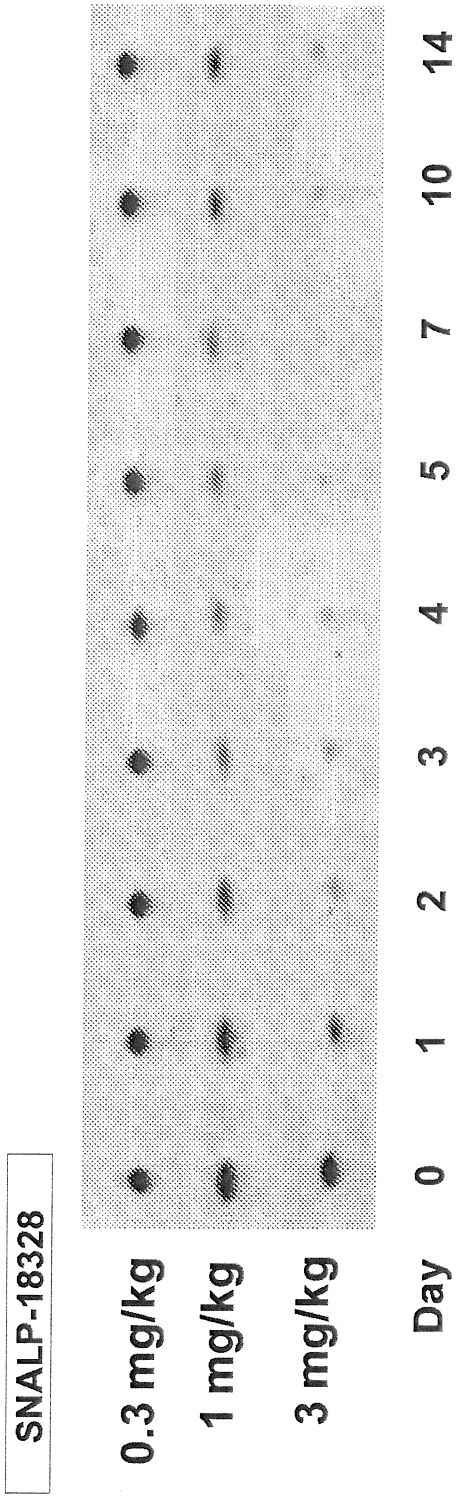


FIG. 8

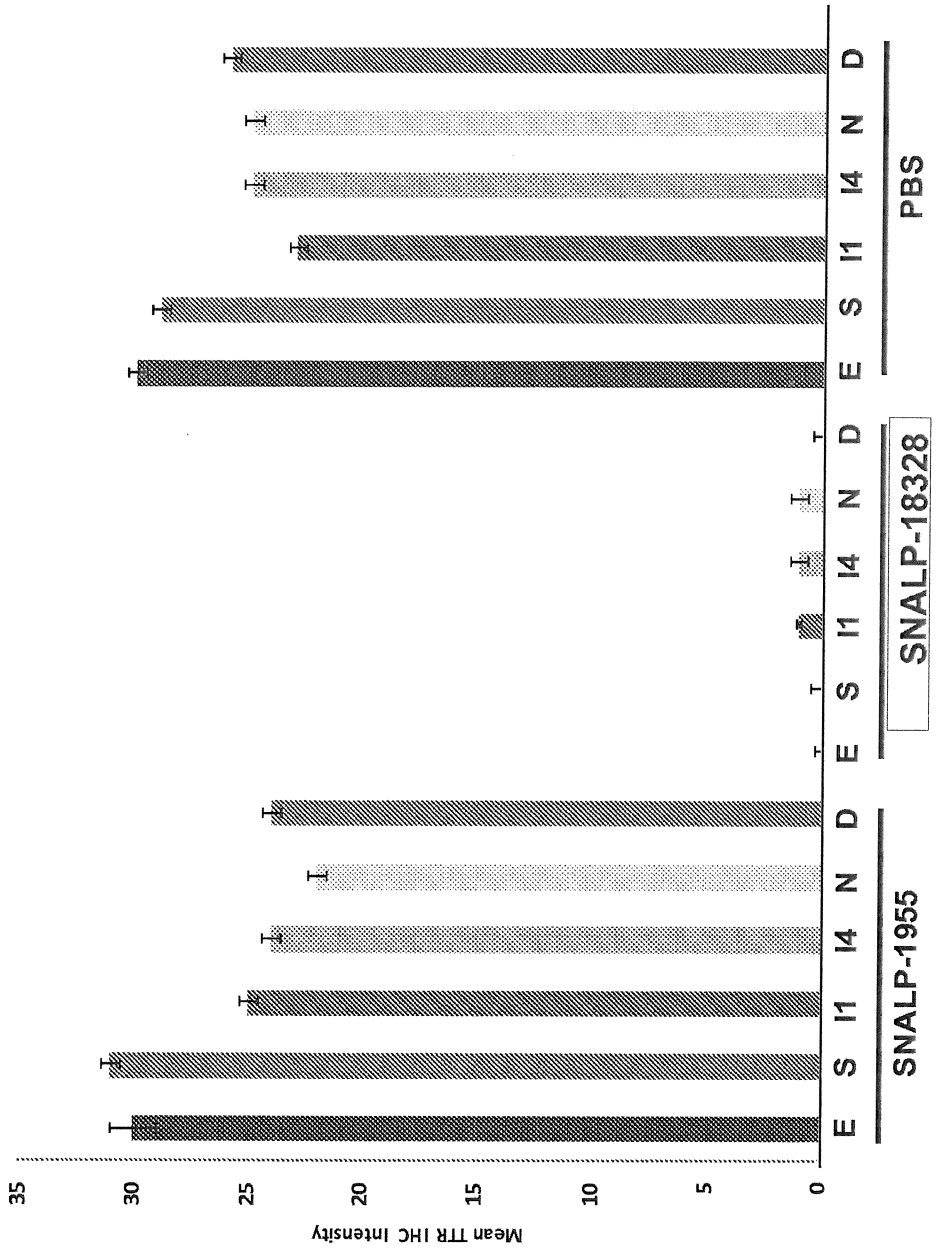

Mean TTR IHC Intensity
0
5
10
15
20
25
30
35
E
S
I1
I4
N
D
SNALP-1955
SNALP-18328
PBS

FIG. 9

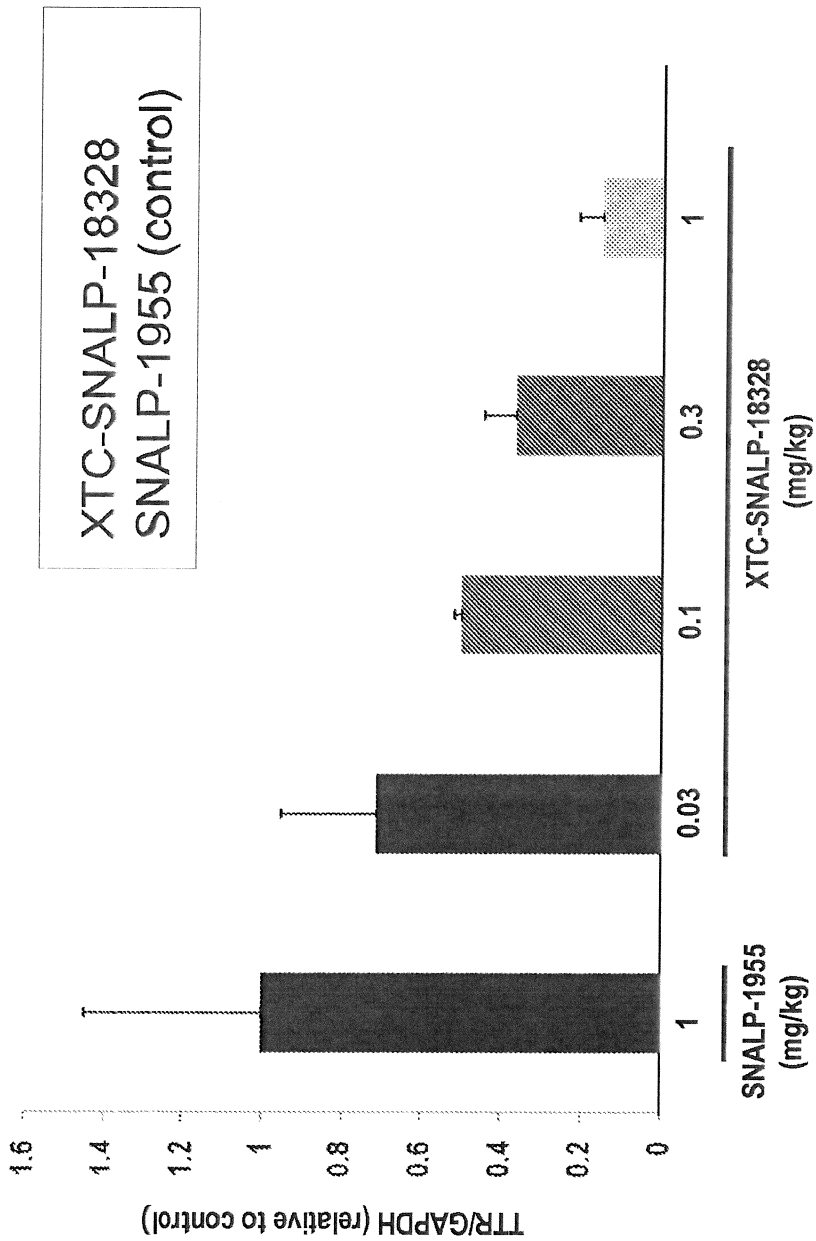

XTC-SNALP-18328
SNALP-1955 (control)
TTR/GAPDH (relative to control)
1.6
1.4
1.2
1
0.8
0.6
0.4
0.2
0
1
0.03
0.1
0.3
1
SNALP-1955 (mg/kg)
XTC-SNALP-18328 (mg/kg)

FIG. 10

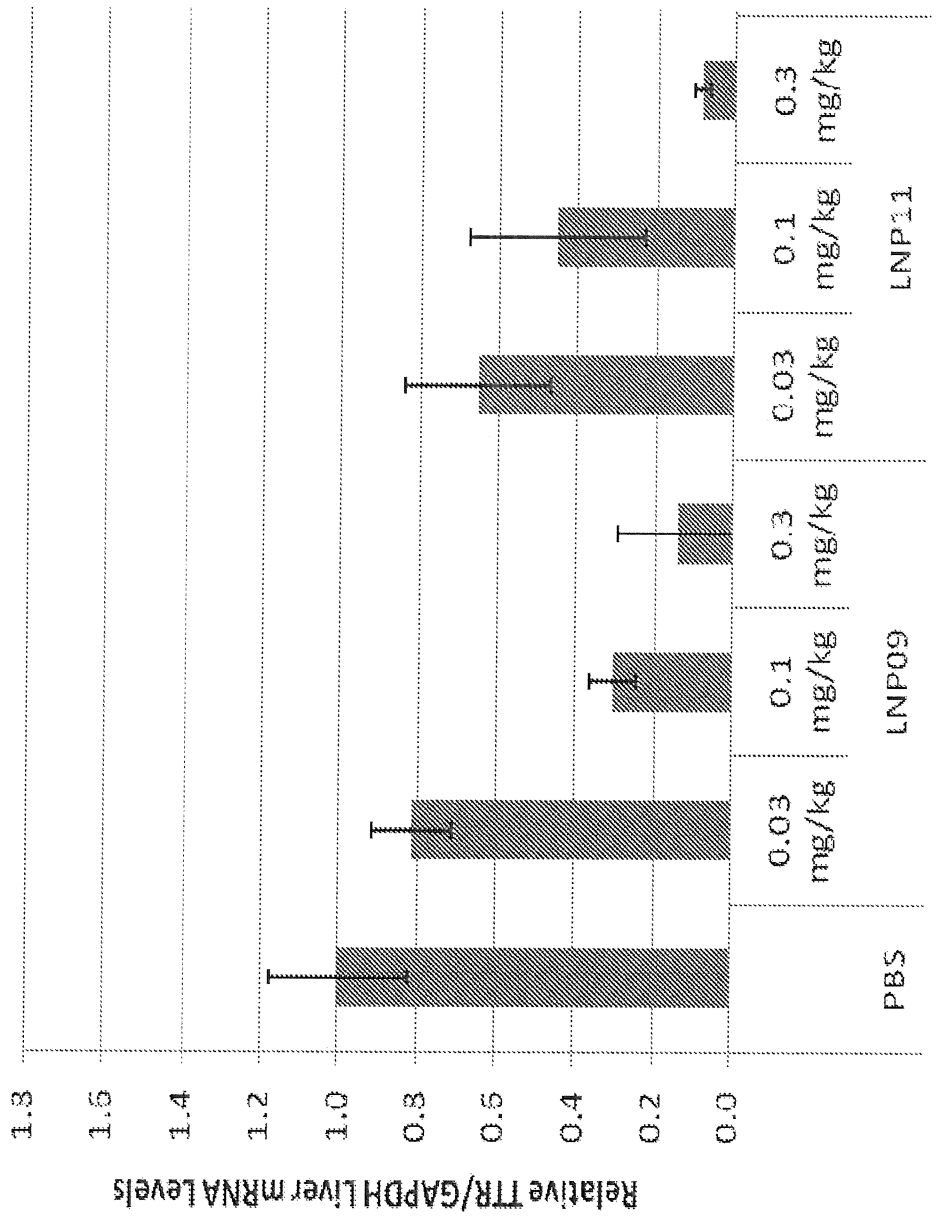

Relative TTR/GAPDH Liver mRNA Levels
1.8
1.6
1.4
1.2
1.0
0.8
0.6
0.4
0.2
0.0
PBS
0.03 mg/kg
0.1 mg/kg
0.3 mg/kg
0.03 mg/kg
0.1 mg/kg
0.3 mg/kg
LNP09
LNP11

FIG. 11A

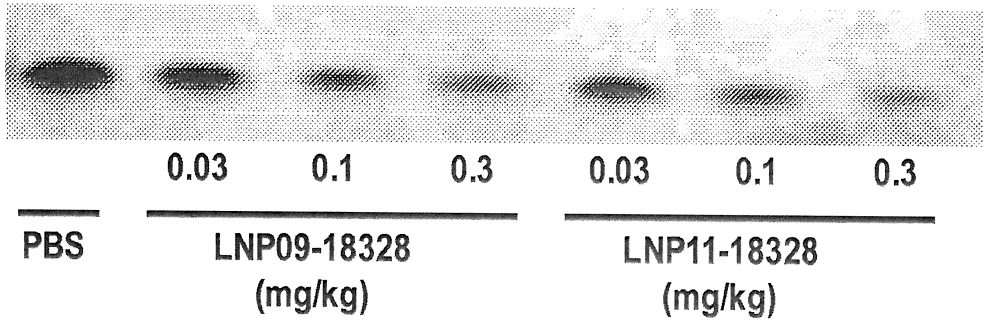
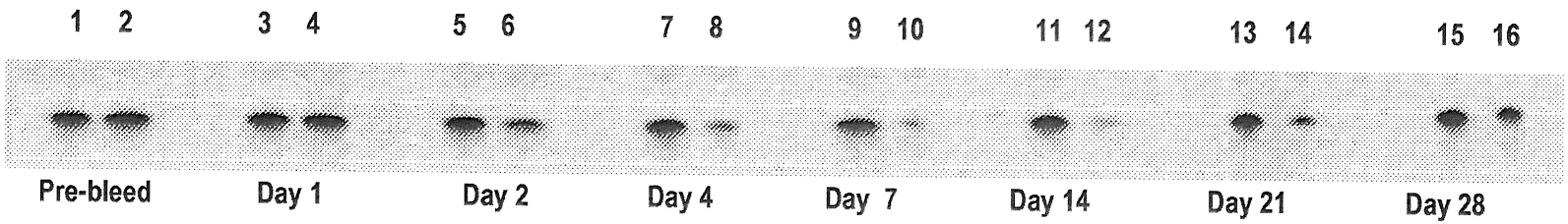


FIG. 11B



Lanes 1, 3, 5, 7, 9, 11, 13, and 15: PBS animals
Lanes 2, 4, 6, 8, 10, 14, and 16: 0.3 mg/kg LNP09-18328

FIG. 11C

gttgactaag tcaataatca gaatcagcag
gtttgcagtc agattggcag ggataagcag
cctagctcag gagaagtgag tataaaagcc
ccaggctggg agcagccatc acagaagtcc
actcattctt ggcaggatgg cttctcatcg
tctgctcctc ctctgccttg ctggactggt
atttgtgtct gaggctggcc ctacgggcac
cggtgaatcc aagtgtcctc tgatggtcaa
agttctagat gctgtccgag gcagtcctgc
catcaatgtg gccgtgcatg tgttcagaaa
ggctgctgat gacacctggg agccatttgc
ctctgggaaa accagtgagt ctggagagct
gcatgggctc acaactgagg aggaatttgt
agaagggata tacaaagtgg aaatagacac
caaatcttac tggaaggcac ttggcatctc
cccattccat gagcatgcag aggtggtatt
cacagccaac gactccggcc cccgccgcta
caccattgcc gccctgctga gcccctactc
ctattccacc acggctgtcg tcaccaatcc
caaggaatga gggacttctc ctccagtgga
cctgaaggac gagggatggg atttcatgta
accaagagta ttccattttt actaaagcag
tgttttcacc tcatatgcta tgttagaagt
ccaggcagag acaataaaac attcctgtga
aaggcacttt tcattccact ttaacttgat
tttttaaatt cccttattgt cccttccaaa
aaaaagagaa tcaaaatttt acaaagaatc
aaaggaattc tagaaagtat ctgggcagaa
cgctaggaga gatccaaatt tccattgtct
tgcaagcaaa gcacgtatta aatatgatct
gcagccatta aaaagacaca ttctgtaaaa
aaaaaaaa (SEQ ID NO: 1331)

FIG. 12

FIG. 13A

ACAGAAGTCCACTCATTCTTGGCAGGATGGCTTCTCATCGTCTGCTCCTCCT
CTGCCTTGCTGGACTGGTATTTGTGTCTGAGGCTGGCCCTACGGGCACCGGT
GAATCCAAGTGTCCTCTGATGGTCAAAGTTCTAGATGCTGTCCGAGGCAGTC
CTGCCATCAATGTGGCCGTGCATGTGTTCAGAAAGGCTGCTGATGACACCTG
GGAGCCATTTGCCTCTGGGAAAACCAGTGAGTCTGGAGAGCTGCATGGGCTC
ACAACTGAGGAGGAATTTGTAGAAGGGATATACAAAGTGGAAATAGACACCA
AATCTTACTGGAAGGCACTTGGCATCTCCCCATTCCATGAGCATGCAGAGGT
GGTATTCACAGCCAACGACTCCGGCCCCCGCCGCTACACCATTGCCGCCCTG
CTGAGCCCCTACTCCTATTCCACCACGGCTGTCGTCACCAATCCCAAGGAAT
GAGGGACTTCTCCTCCAGTGGACCTGAAGGACGAGGGATGGGATTTCATGTA
ACCAAGAGTATTCCATTTTTACTAAAGCAGTGTTTTCACCTCATATGCTATG
TTAGAAGTCCAGGCAGAGACAATAAAACATTCCTGTGAAAGGCACTTTTCAT
TCCAAAAAAAAAAAAAAAAAAAAAA (SEQ ID NO:1329)

FIG. 13B

CCTGACAGGATGGCTTCCCTTCGCCTGTTCCTCCTCTGCCTCGCTGGACTGA
TATTTGCGTCTGAAGCTGGCCCTGGGGGTGCTGGAGAATCCAAGTGTCCTCT
GATGGTCAAAGTCCTGGATGCTGTCCGAGGCAGCCCTGCTGTCGATGTGGCC
GTGAAAGTGTTCAAAAGGACTGCAGACGGAAGCTGGGAGCCGTTTGCCTCTG
GGAAGACCGCCGAGTCTGGAGAGCTGCACGGGCTCACCACAGATGAGAAGTT
CACGGAAGGGGTGTACAGGGTAGAACTGGACACCAAATCATACTGGAAGGCT
CTTGGCATTTCCCCATTCCATGAATACGCAGAGGTGGTTTTCACAGCCAATG
ACTCTGGTCATCGCCACTACACCATCGCAGCCCTGCTCAGCCCGTACTCCTA
CAGCACCACTGCTGTCGTCAGTAACCCCCAGAACTGAGGGACCCAGCCCACG
AGGACCAAGATCTTGCCAAAGCAGTAGCTCCCATTTGTACTGAAACAGTGTT
CTTGCTCTATAAACCGTGTTAGCAACTCGGGAAGATGCCGTGAAACGTTCTT
ATTAAACCACCTTTATTTCATTC
(SEQ ID NO:1330)

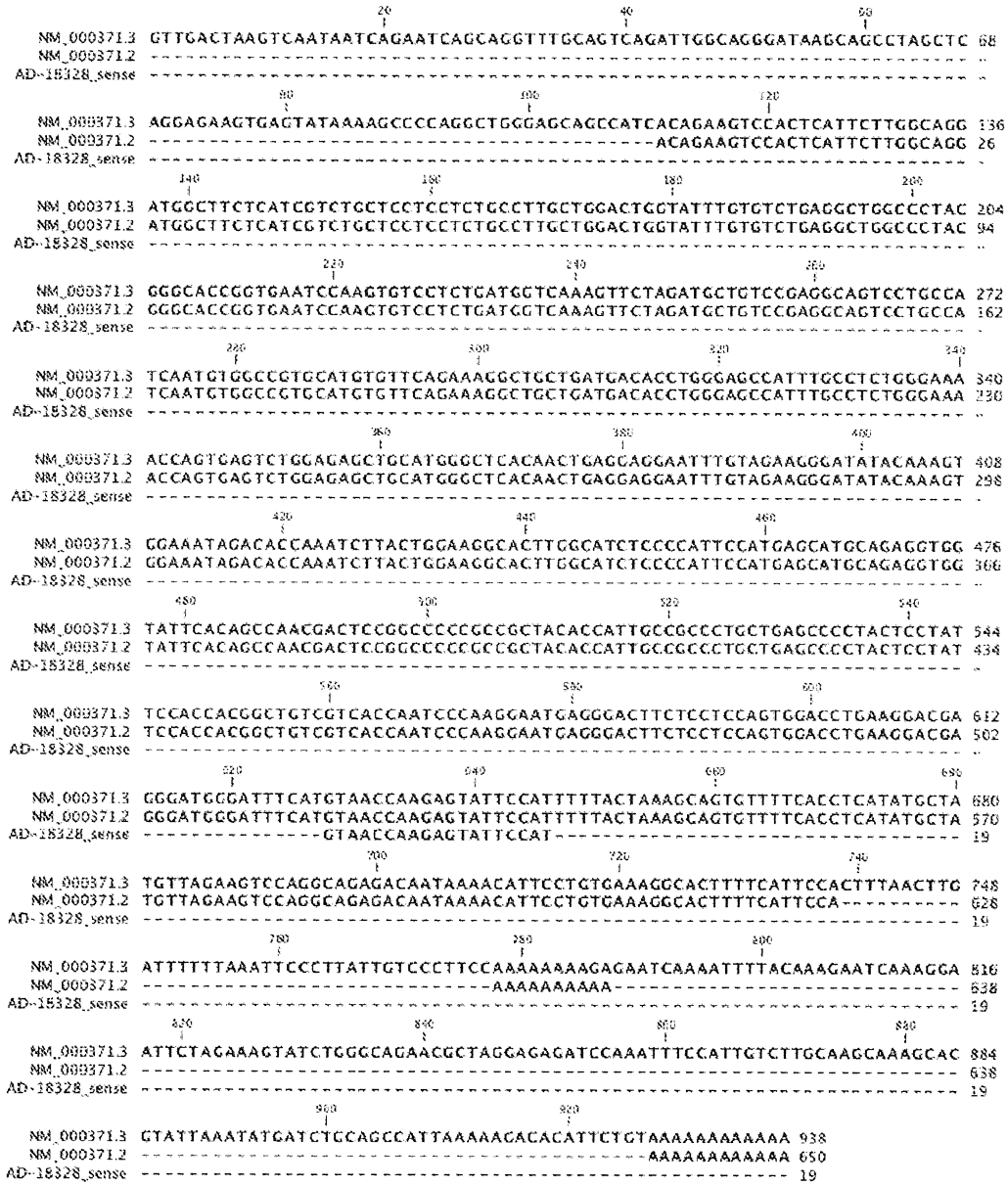

NM_000371.3 GTTGACTAAGTCAATAATCAGAATCAGCAGGTTTGCAGTCAGATTGGCAGGGATAAGCAGCCTAGCTC 68
NM_000371.2 ----------
AD-18328_sense ----------
NM_000371.3 AGGAGAAGTGAGTATAAAAGCCCCAGGCTGGGAGCAGCCATCACAGAAGTCCACTCATTCTTGGCAGG 136
NM_000371.2 ----------ACAGAAGTCCACTCATTCTTGGCAGG 26
AD-18328_sense ----------
NM_000371.3 ATGGCTTCTCATCGTCTGCTCCTCCTCTGCCTTGCTGGACTGGTATTTGTGTCTGAGGCTGGCCCTAC 204
NM_000371.2 ATGGCTTCTCATCGTCTGCTCCTCCTCTGCCTTGCTGGACTGGTATTTGTGTCTGAGGCTGGCCCTAC 94
AD-18328_sense ----------
NM_000371.3 GGGCACCGGTGAATCCAAGTGTCCTCTGATGGTCAAAGTTCTAGATGCTGTCCGAGGCAGTCCTGCCA 272
NM_000371.2 GGGCACCGGTGAATCCAAGTGTCCTCTGATGGTCAAAGTTCTAGATGCTGTCCGAGGCAGTCCTGCCA 162
AD-18328_sense ----------
NM_000371.3 TCAATGTGGCCGTGCATGTGTTCAGAAAGGCTGCTGATGACACCTGGGAGCCATTTGCCTCTGGGAAA 340
NM_000371.2 TCAATGTGGCCGTGCATGTGTTCAGAAAGGCTGCTGATGACACCTGGGAGCCATTTGCCTCTGGGAAA 230
AD-18328_sense ----------
NM_000371.3 ACCAGTGAGTCTGGAGAGCTGCATGGGCTCACAACTGAGGAGGAATTTGTAGAAGGGATATACAAAGT 408
NM_000371.2 ACCAGTGAGTCTGGAGAGCTGCATGGGCTCACAACTGAGGAGGAATTTGTAGAAGGGATATACAAAGT 298
AD-18328_sense ----------
NM_000371.3 GGAAATAGACACCAAATCTTACTGGAAGGCACTTGGCATCTCCCCATTCCATGAGCATGCAGAGGTGG 476
NM_000371.2 GGAAATAGACACCAAATCTTACTGGAAGGCACTTGGCATCTCCCCATTCCATGAGCATGCAGAGGTGG 366
AD-18328_sense ----------
NM_000371.3 TATTCACAGCCAACGACTCCGGCCCCCCGCCGCTACACCATTGCCGCCCTGCTGAGCCCCTACTCCTAT 544
NM_000371.2 TATTCACAGCCAACGACTCCGGCCCCCCGCCGCTACACCATTGCCGCCCTGCTGAGCCCCTACTCCTAT 434
AD-18328_sense ----------
NM_000371.3 TCCACCACGGCTGTCGTCACCAATCCCAAGGAATGAGGGACTTCTCCTCCAGTGGACCTGAAGGACGA 612
NM_000371.2 TCCACCACGGCTGTCGTCACCAATCCCAAGGAATGAGGGACTTCTCCTCCAGTGGACCTGAAGGACGA 502
AD-18328_sense ----------
NM_000371.3 GGGATGGGATTTCATGTAACCAAGAGTATTCCATTTTTACTAAAGCAGTGTTTTCACCTCATATGCTA 680
NM_000371.2 GGGATGGGATTTCATGTAACCAAGAGTATTCCATTTTTACTAAAGCAGTGTTTTCACCTCATATGCTA 570
AD-18328_sense ----------GTAACCAAGAGTATTCCAT---------- 19
NM_000371.3 TGTTAGAAGTCCAGGCAGAGACAATAAAACATTCCTGTGAAAGGCACTTTTCATTCCACTTTAACTTG 748
NM_000371.2 TGTTAGAAGTCCAGGCAGAGACAATAAAACATTCCTGTGAAAGGCACTTTTCATTCCA---------- 628
AD-18328_sense ---------- 19
NM_000371.3 ATTTTTTAAATTCCCTTATTGTCCCTTCCAAAAAAAAAGAGAATCAAAATTTTACAAAGAATCAAAGGA 816
NM_000371.2 ----------AAAAAAAAAA---------- 638
AD-18328_sense ---------- 19
NM_000371.3 ATTCTAGAAAGTATCTGGGCAGAACGCTAGGAGAGATCCAAATTTCCATTGTCTTGCAAGCAAAGCAC 884
NM_000371.2 ---------- 638
AD-18328_sense ---------- 19
NM_000371.3 GTATTAAATATGATCTGCAGCCATTAAAAAGACACATTCTGTAAAAAAAAAAAA 938
NM_000371.2 ----------AAAAAAAAAAAA 650
AD-18328_sense ---------- 19

FIG. 14

Phenotype	Features	Genotypes (associated mutation in TTR)
Familial amyloidotic neuropathy (FAP)	Early: Impotence Sensorimotor polyneuropathy of the legs Carpal tunnel syndrome Autonomic dysfunction Constipation/diarrhea Late: Cardiomyopathy Vitreous opacities Nephropathy	V28M L58H V30M L58R K70N Y78F I84S Y114H V30A K35N G47V S50R T60A Y114C
Familial amyloidotic cardiomyopathy (FAC)	Cardiomegaly Congestive heart failure Conduction abnormalities, arrhythmias Angina Sudden death	D18N D18E V20I P24S E42D A45T T49P S50I H56R I68L A81T Q92K R103S L111M V122I T60A
CNS amyloidosis (CNSA)	Dementia, ataxia, spasticity, seizures, hemorrhage (intracerebellar and/or subarachnoid), psychosis, hydrocephalus	L12P D18G A25T V30G A36P G53E F64S Y69H Y114C

FIG. 15

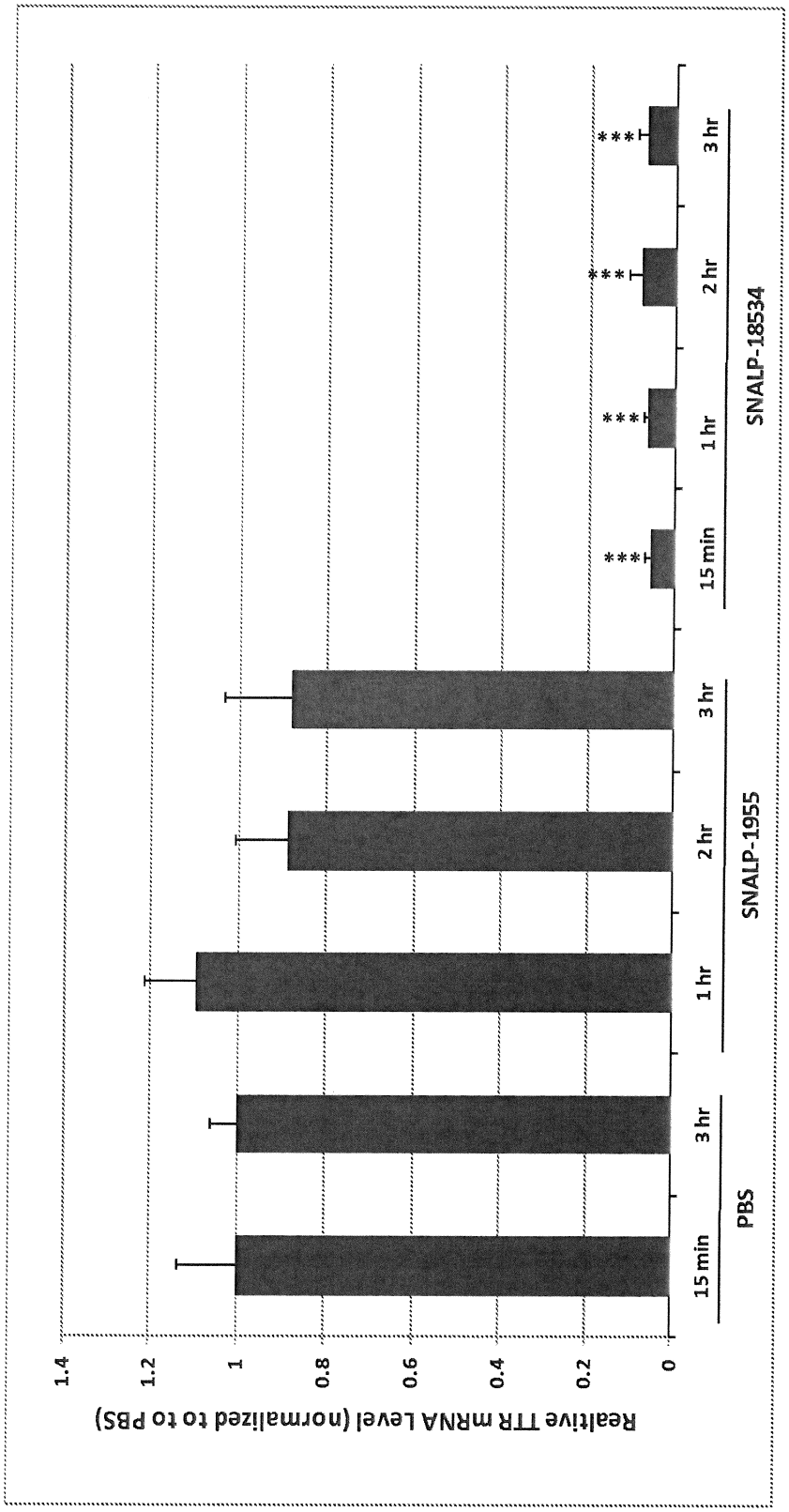

Realtive TTR mRNA Level (normalized to to PBS)
1.4
1.2
1
0.8
0.6
0.4
0.2
0
15 min
3 hr
1 hr
2 hr
3 hr
15 min
1 hr
2 hr
3 hr
PBS
SNALP-1955
SNALP-18534
***
***
**
***

FIG. 16

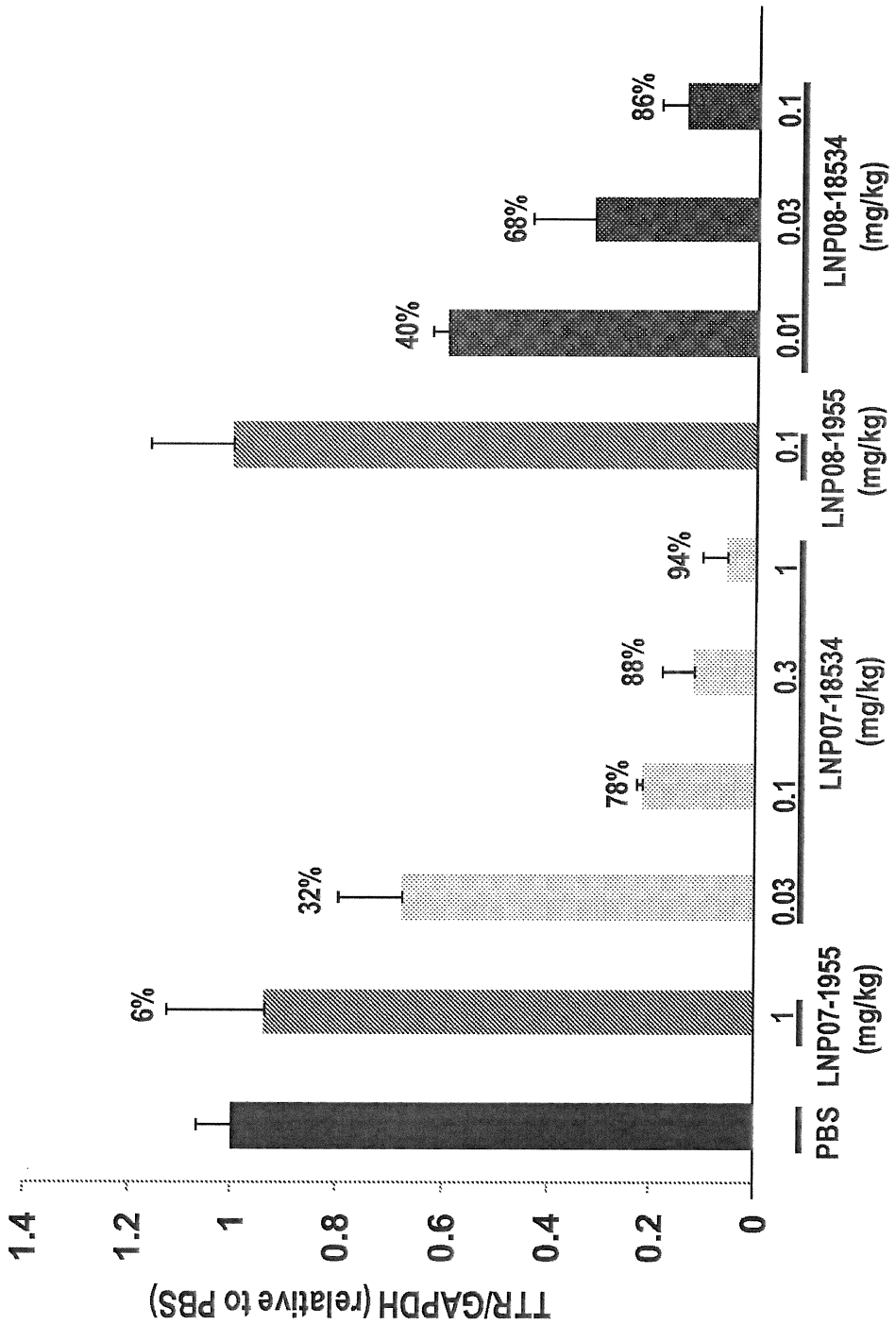

TTR/GAPDH (relative to PBS)
1.4
1.2
1
0.8
0.6
0.4
0.2
0
PBS
LNP07-1955 (mg/kg)
1
6%
LNP07-18534 (mg/kg)
0.03
32%
0.1
78%
0.3
88%
1
94%
LNP08-1955 (mg/kg)
0.1
0.01
40%
LNP08-18534 (mg/kg)
0.03
68%
0.1
86%

FIG. 17

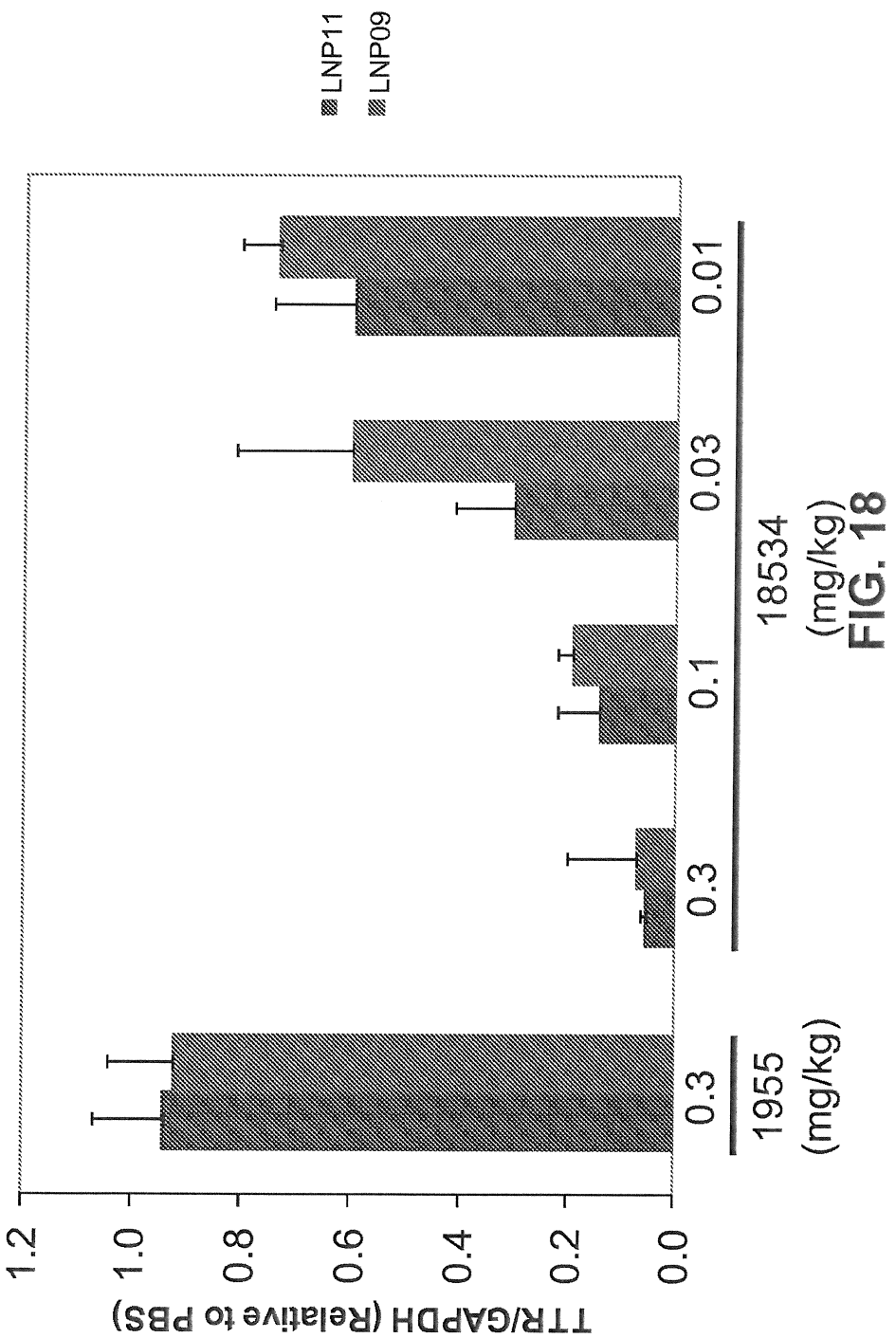

LNP11
LNP09
TTR/GAPDH (Relative to PBS)
1.2
1.0
0.8
0.6
0.4
0.2
0.0
0.3
0.3
0.1
0.03
0.01
1955
(mg/kg)
18534
(mg/kg)

FIG. 18