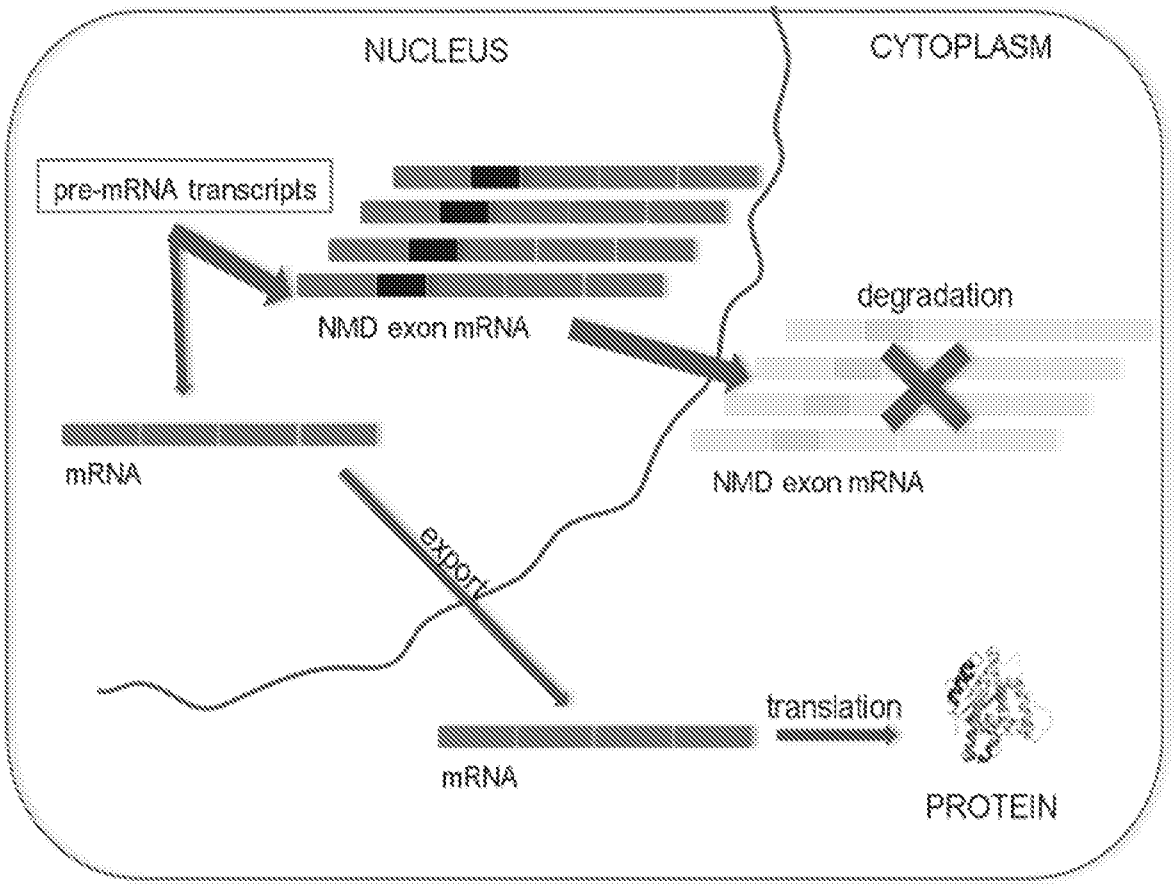

NUCLEUS
CYTOPLASM
pre-mRNA transcripts
NMD exon mRNA
degradation
mRNA
NMD exon mRNA
export
translation
mRNA
PROTEIN

FIG. 1A

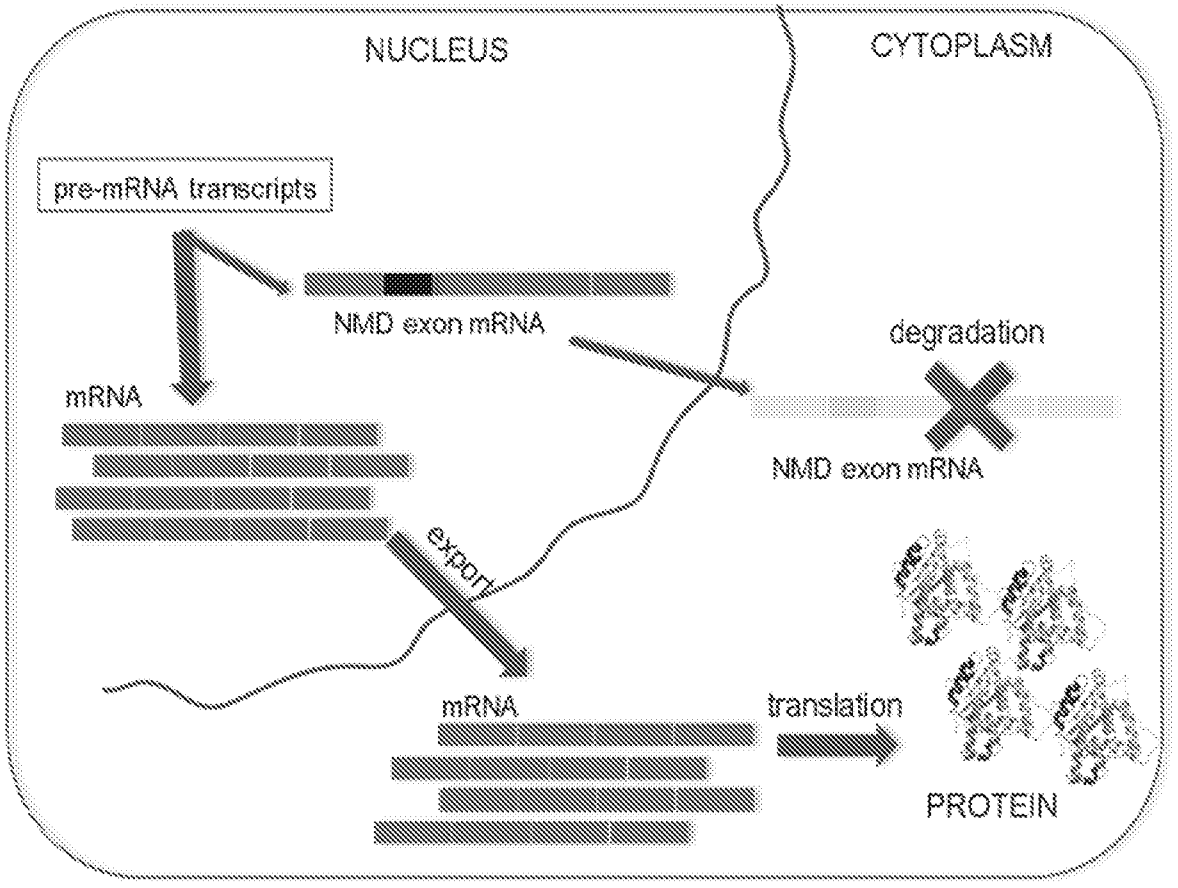

NUCLEUS
CYTOPLASM
pre-mRNA transcripts
NMD exon mRNA
degradation
mRNA
NMD exon mRNA
export
mRNA
translation
PROTEIN

FIG. 1B

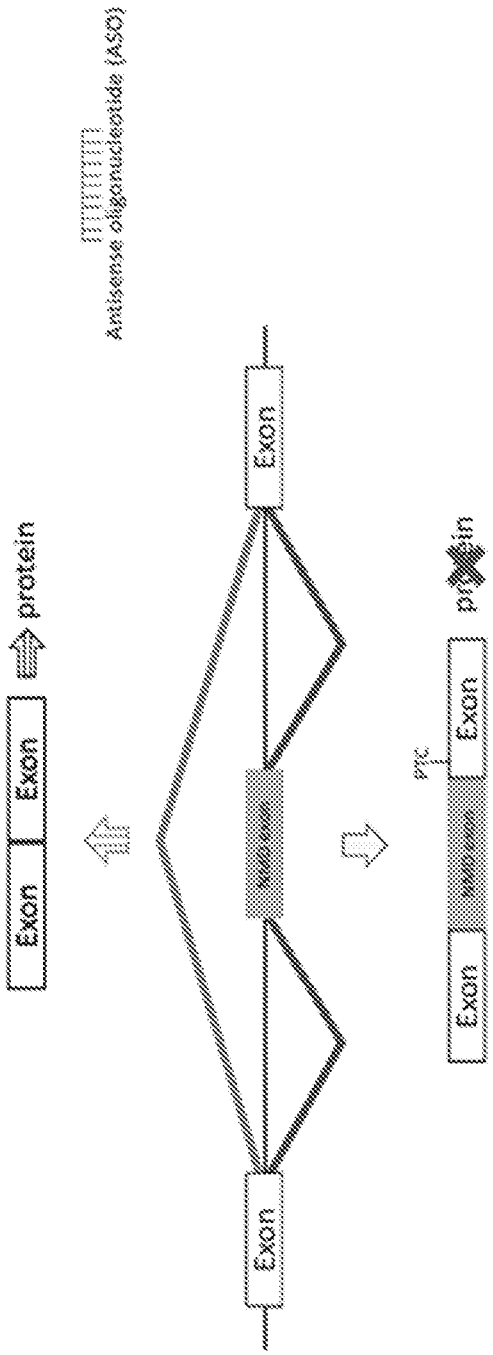

Antisense oligonucleotide (ASO)
Exon
Exon
protein
Exon
Exon
PTC
Exon
Exon

FIG. 1C

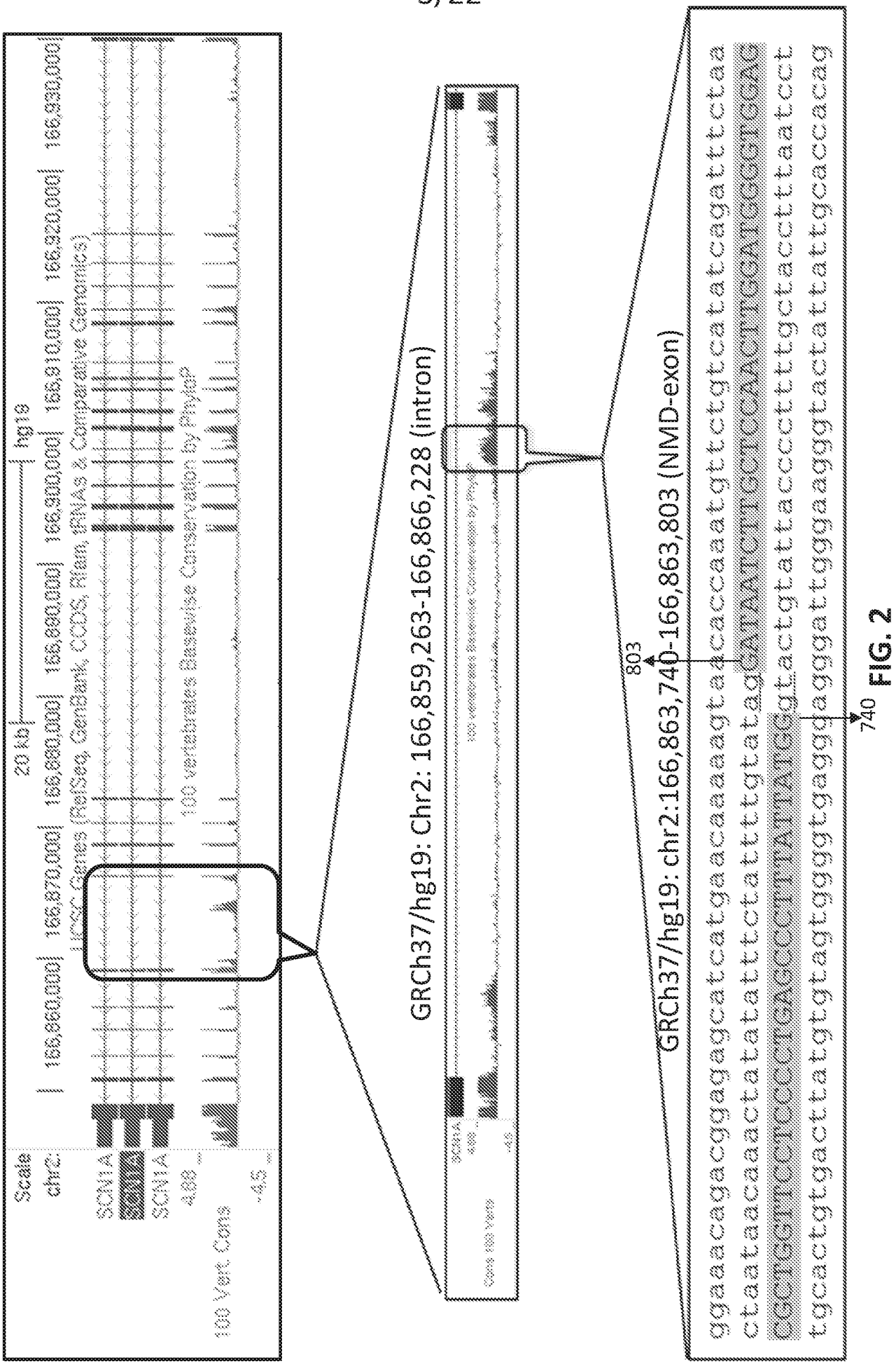

Scale
chr2:
166,860,000
166,870,000
166,880,000
166,890,000
166,900,000
166,910,000
166,920,000
166,930,000
20 kb
hg19
UCSC Genes (RefSeq, GenBank, CCDS, Rfam, tRNAs & Comparative Genomics)
SCN1A
SCN1A
100 vertebrates Basewise Conservation by PhyloP
100 Vert. Cons
400
-45
GRCh37/hg19: Chr2: 166,859,263-166,866,228 (intron)
GRCh37/hg19: chr2:166,863,740-166,863,803 (NMD-exon)
803
740
ggaaacagacggagagcatcatgaacaaaaaagtaacaccaaatgttctgtcatatcagatttctaa
ctaataacaaactatatatttctatttgtatagCATAAATCTTGCTCCAACTTGGATGGGTGGAG
CGCTTGGTTCCTCCCCTGAGCCCTTTTATTATGGgtactgtattaccccctttttgctacctttaatcct
tgcactgtgacttatgtagtgggtgaggaggatttgggaagggtactatttattgcaccacag

FIG. 2

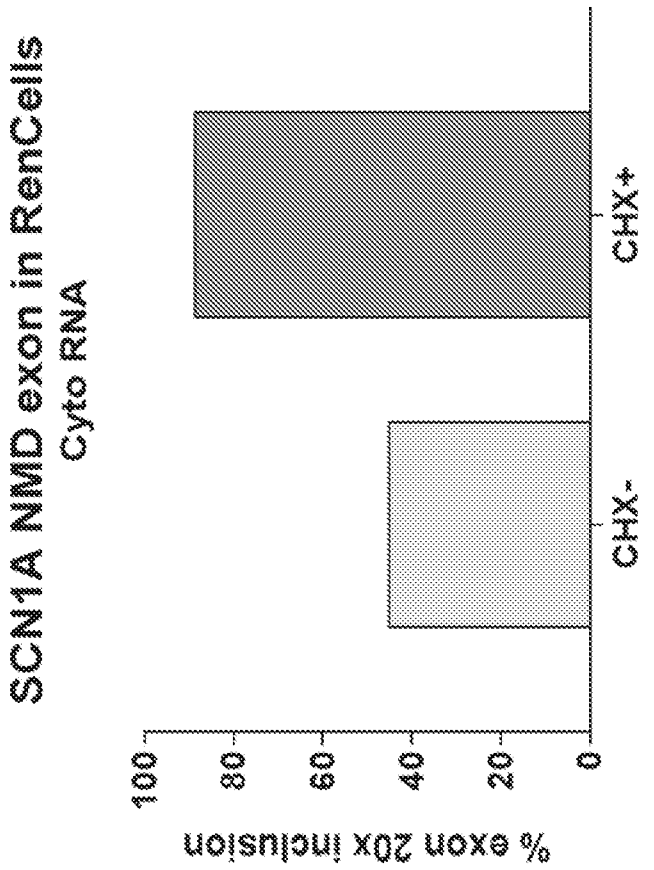

SCN1A NMD exon in RenCells
Cyto RNA
% exon 20x inclusion
100
80
60
40
20
0
CHX+
CHX-

FIG. 3B

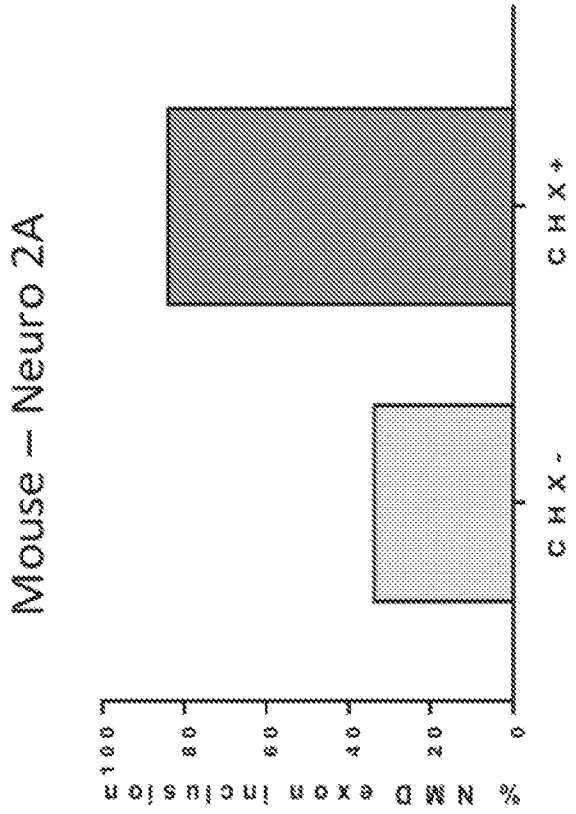

Mouse – Neuro 2A
% NMD exon inclusion
100
80
60
40
20
0
CHX+
CHX-

FIG. 3A

aacagacggagagcatcatgaacaaaagtaacaccaaatgtctgtcatatcagatttctaactaataacaaactatatttctatttgtatag

ttctactttgtatagcgtaaactttcaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaaactgtattacco

gtactgtatttaccccttttgctacctttaatcctgcactgtgtagtgggtgaggaggattgggaaggtactattattgcaccacag

FIG. 4

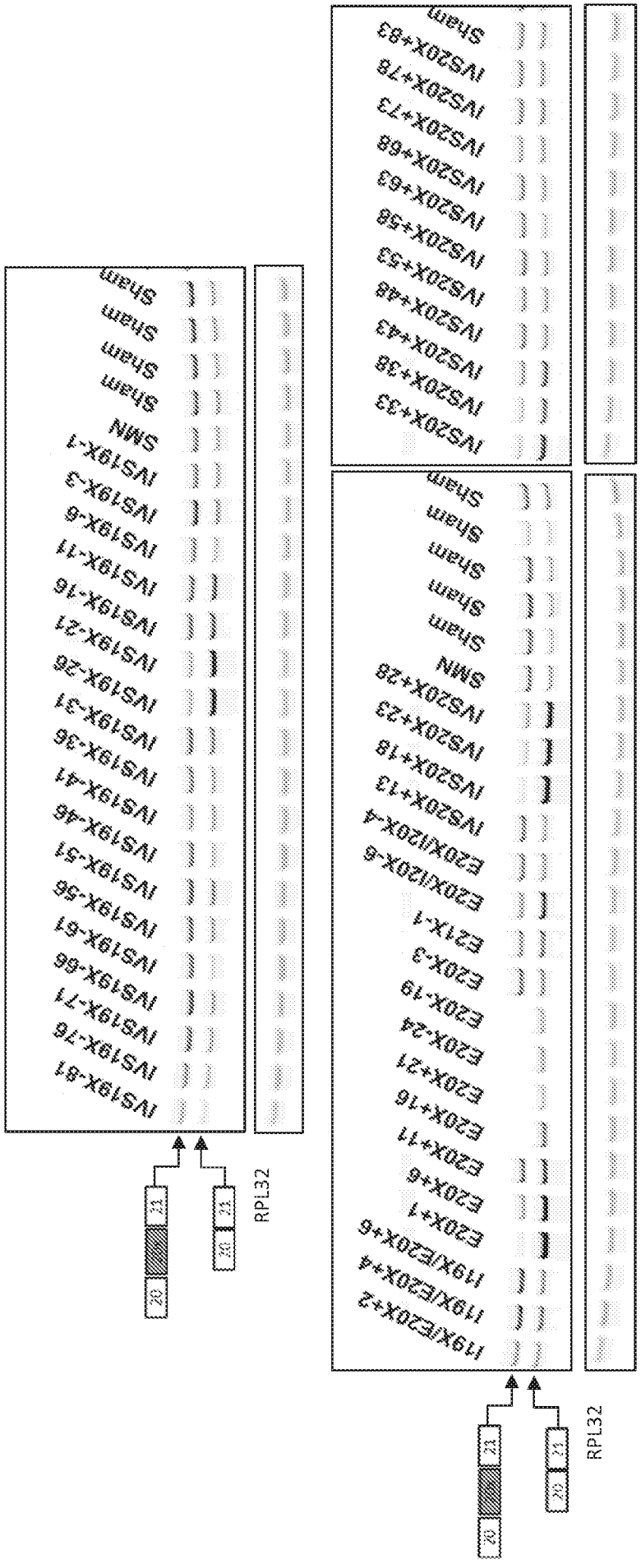

Sham
IVS20X+83
IVS20X+78
IVS20X+73
IVS20X+68
IVS20X+63
IVS20X+58
IVS20X+53
IVS20X+48
IVS20X+43
IVS20X+38
IVS20X+33
Sham
Sham
Sham
Sham
SMN
IVS19X-1
IVS19X-3
IVS19X-6
IVS19X-11
IVS19X-16
IVS19X-21
IVS19X-26
IVS19X-31
IVS19X-36
IVS19X-41
IVS19X-46
IVS19X-51
IVS19X-56
IVS19X-61
IVS19X-66
IVS19X-71
IVS19X-76
IVS19X-81
RPL32
21
20
21
20
Sham
Sham
Sham
Sham
Sham
SMN
IVS20X+28
IVS20X+23
IVS20X+18
IVS20X+13
E20X/I20X-4
E20X/I20X-6
E21X-1
E20X-3
E20X-19
E20X-24
E20X+21
E20X+16
E20X+11
E20X+6
E20X+1
I19X/E20X+6
I19X/E20X+4
I19X/E20X+2
RPL32
21
20
21
20

FIG. 5A

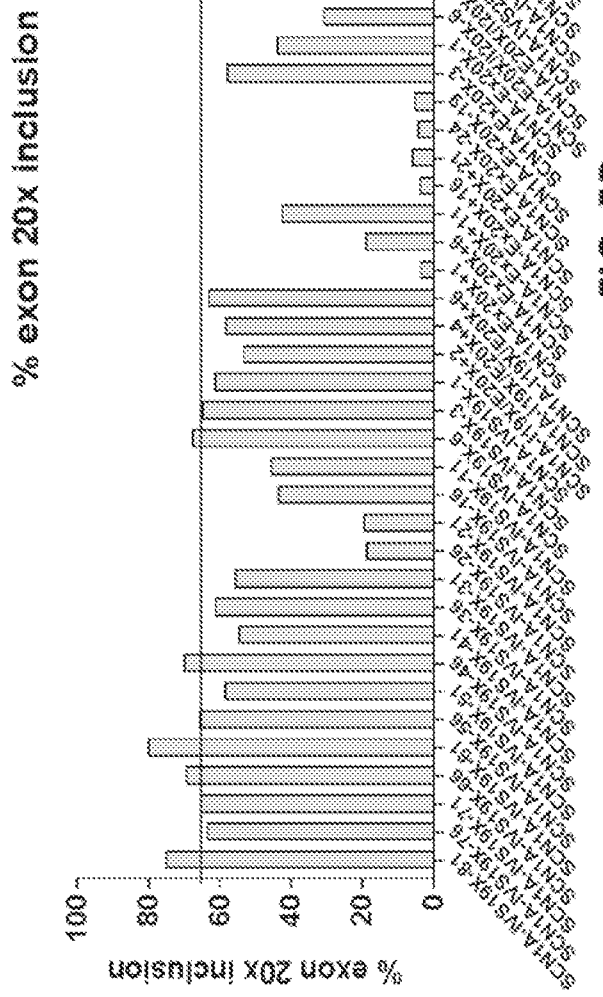

% exon 20x inclusion
% exon 20x inclusion
100
80
60
40
20
0

FIG. 5B

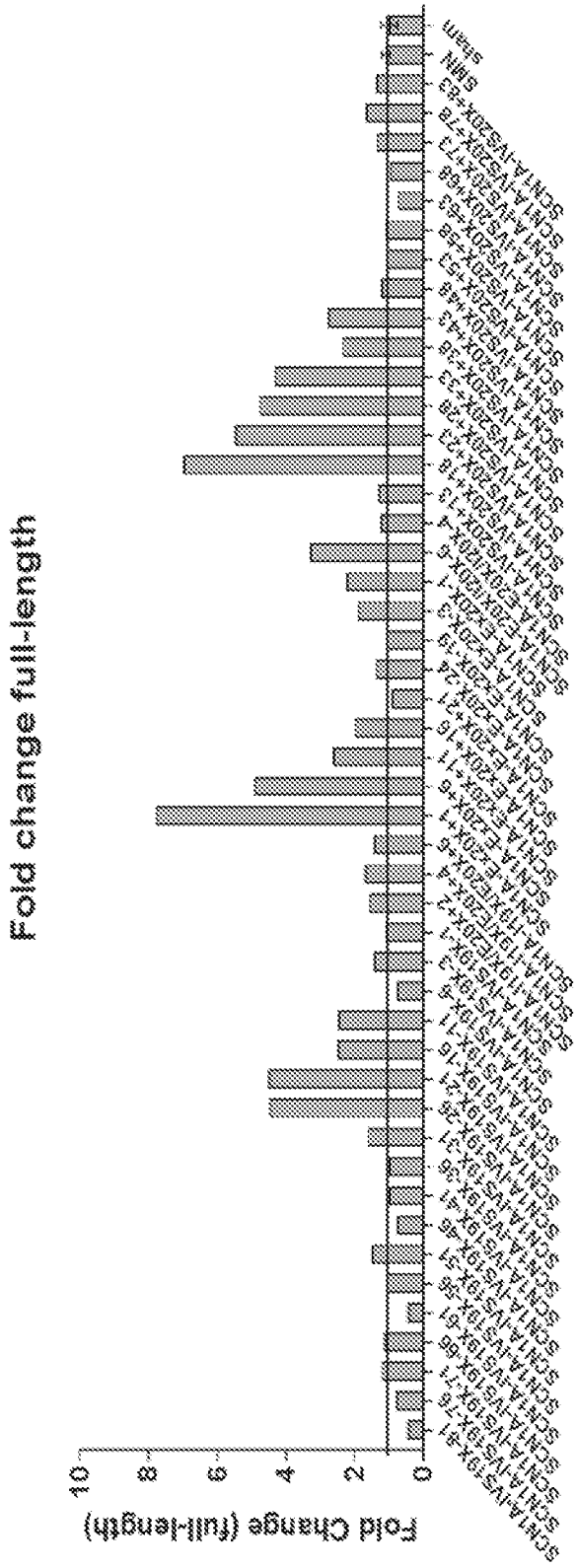

Fold change full-length
Fold Change (full-length)
10
8
6
4
2
0

FIG. 5C

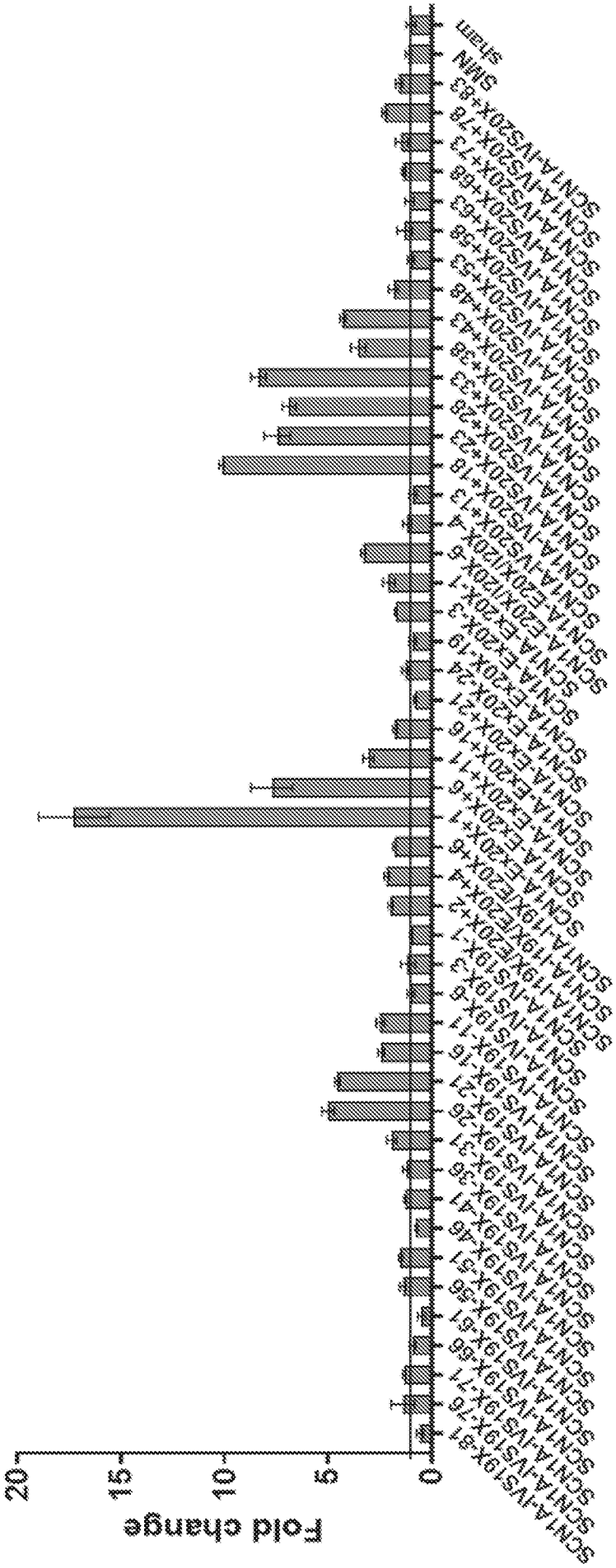

Fold change
0
5
10
15
20
shRNA
SCN1A-IVS19X-61
SCN1A-IVS19X-76
SCN1A-IVS19X-71
SCN1A-IVS19X-66
SCN1A-IVS19X-61
SCN1A-IVS19X-56
SCN1A-IVS19X-51
SCN1A-IVS19X-46
SCN1A-IVS19X-41
SCN1A-IVS19X-36
SCN1A-IVS19X-31
SCN1A-IVS19X-26
SCN1A-IVS19X-21
SCN1A-IVS19X-16
SCN1A-IVS19X-11
SCN1A-IVS19X-6
SCN1A-IVS19X-3
SCN1A-IVS19X-1
SCN1A-IVS19XE20X+2
SCN1A-IVS19XE20X+4
SCN1A-E20X+6
SCN1A-E20X+1
SCN1A-E20X+6
SCN1A-E20X+11
SCN1A-E20X+16
SCN1A-E20X+21
SCN1A-E20X-24
SCN1A-E20X-19
SCN1A-E20X-3
SCN1A-E20X-1
SCN1A-E20X/I20X-6
SCN1A-IVS20X+4
SCN1A-IVS20X+13
SCN1A-IVS20X+18
SCN1A-IVS20X+23
SCN1A-IVS20X+28
SCN1A-IVS20X+33
SCN1A-IVS20X+38
SCN1A-IVS20X+43
SCN1A-IVS20X+48
SCN1A-IVS20X+53
SCN1A-IVS20X+58
SCN1A-IVS20X+63
SCN1A-IVS20X+68
SCN1A-IVS20X+73
SCN1A-IVS20X+78
SCN1A-IVS20X+83

FIG. 6

Sodium voltage-gaited channel alpha subunit

Gene	Approved Name	Chromosome	
SCN1A	sodium voltage-gated channel alpha subunit 1	2q24.3	⬅
SCN2A	sodium voltage-gated channel alpha subunit 2	2q24.3	⇦
SCN3A	sodium voltage-gated channel alpha subunit 3	2q24.3	⇦
SCN7A	sodium voltage-gated channel alpha subunit 7	2q24.3	X
SCN8A	sodium voltage-gated channel alpha subunit 8	12q13.13	⇦
SCN9A	sodium voltage-gated channel alpha subunit 9	2q24.3	⇦
SCN10A	sodium voltage-gated channel alpha subunit 10	3p22.2	X
SCN11A	sodium voltage-gated channel alpha subunit 11	3p22.2	X

X: no expression detected

FIG. 7A

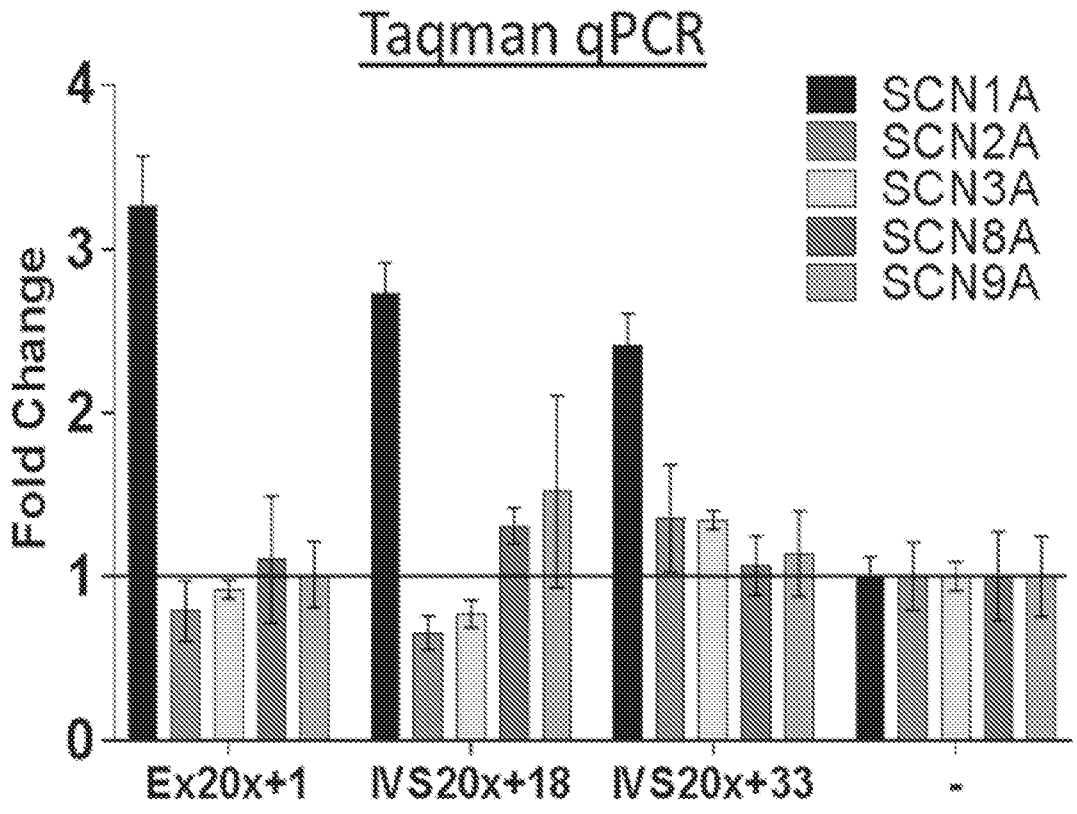


FIG. 7B

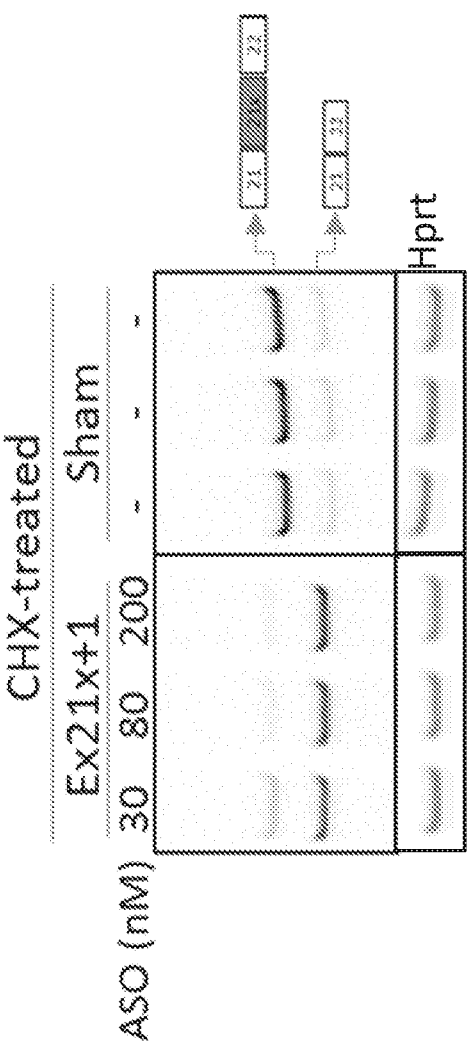


FIG. 8A

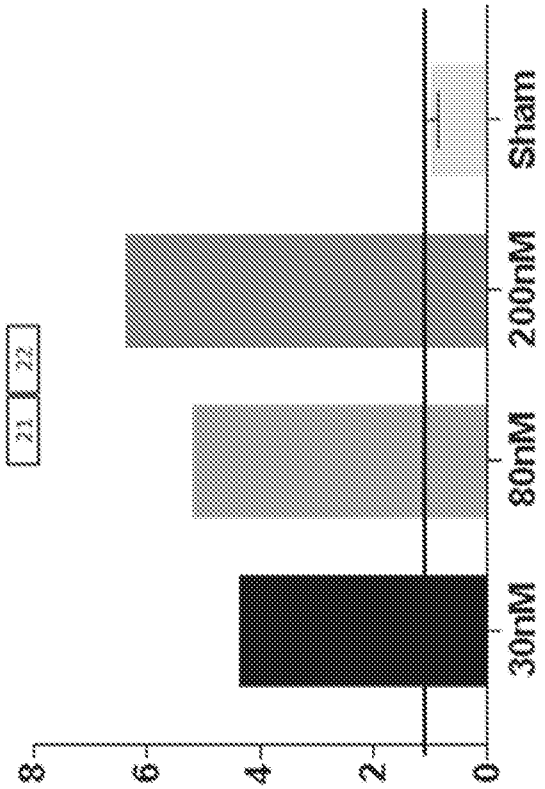


FIG. 8C

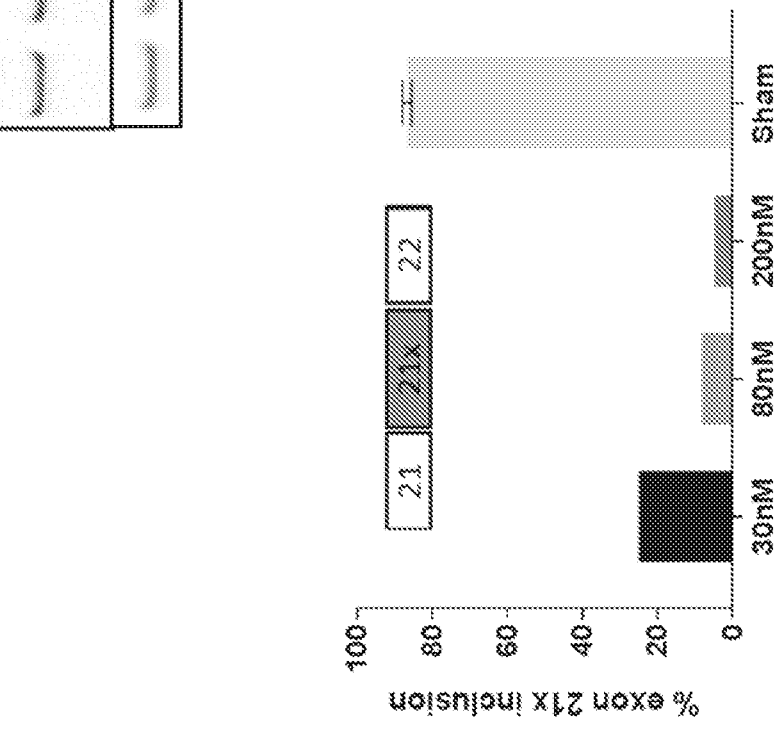


FIG. 8B

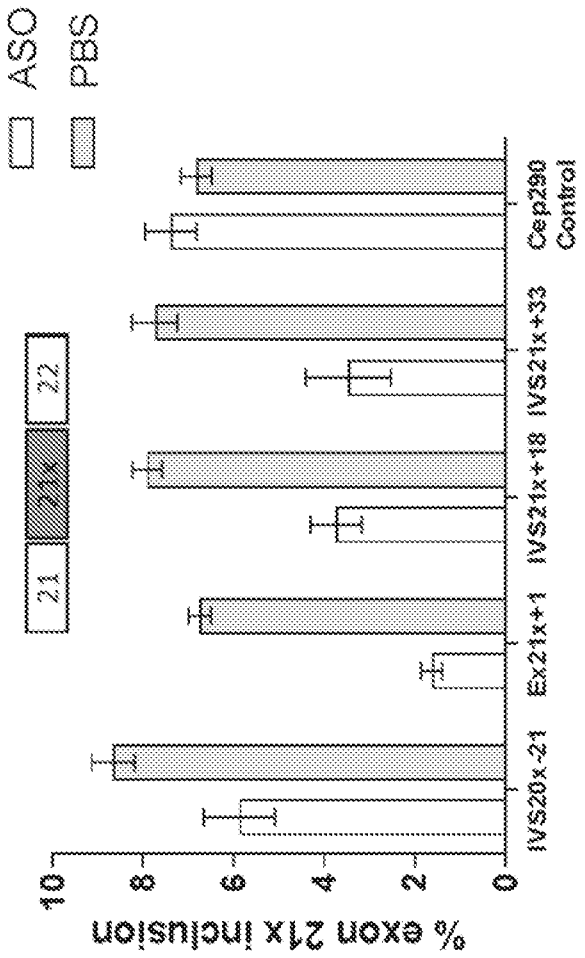


FIG. 9B

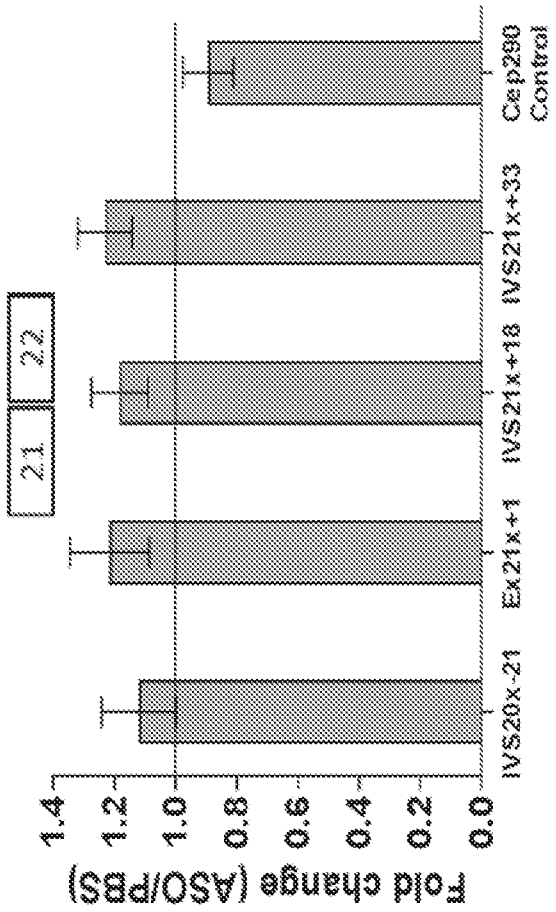


FIG. 9C

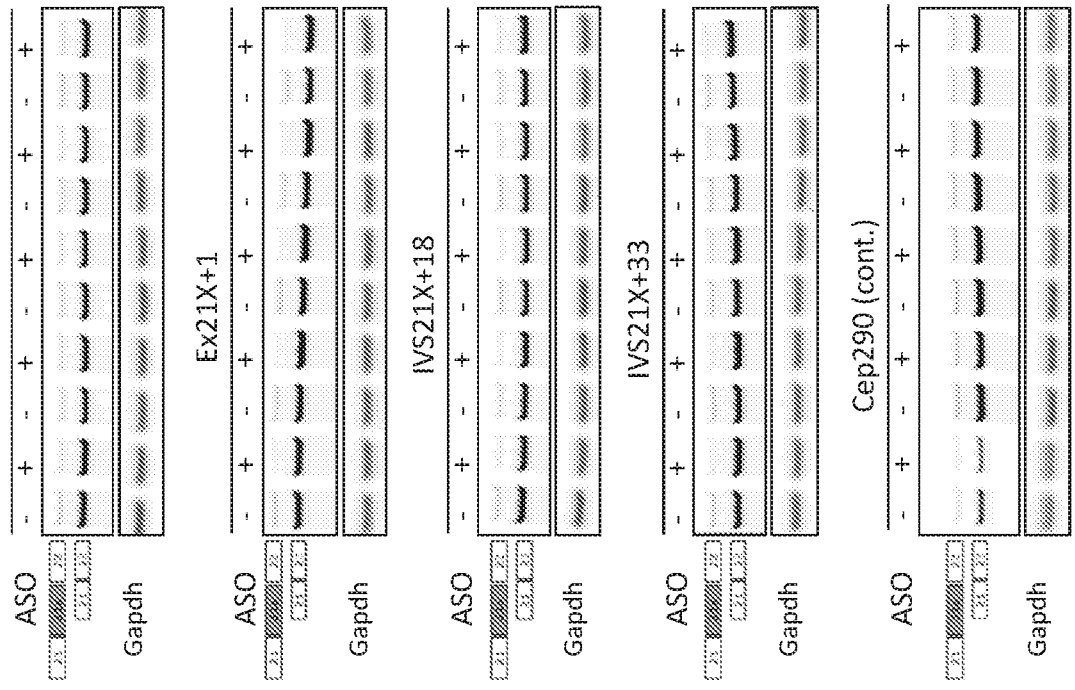


FIG. 9A

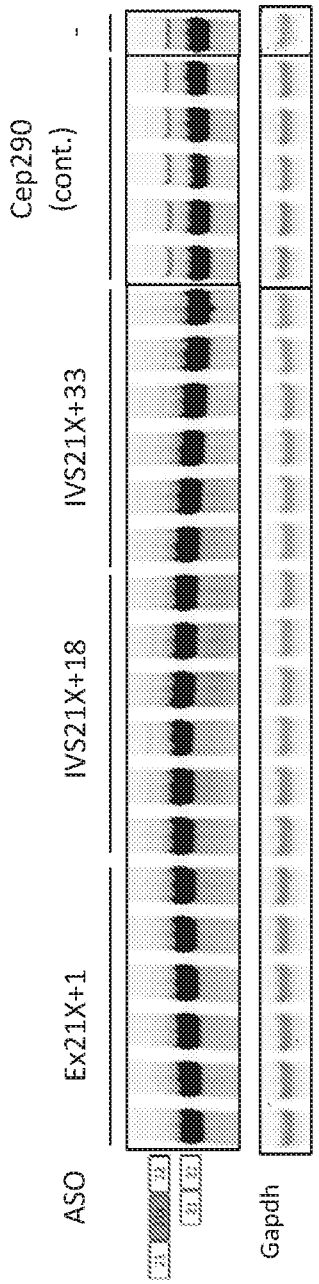


FIG. 10A

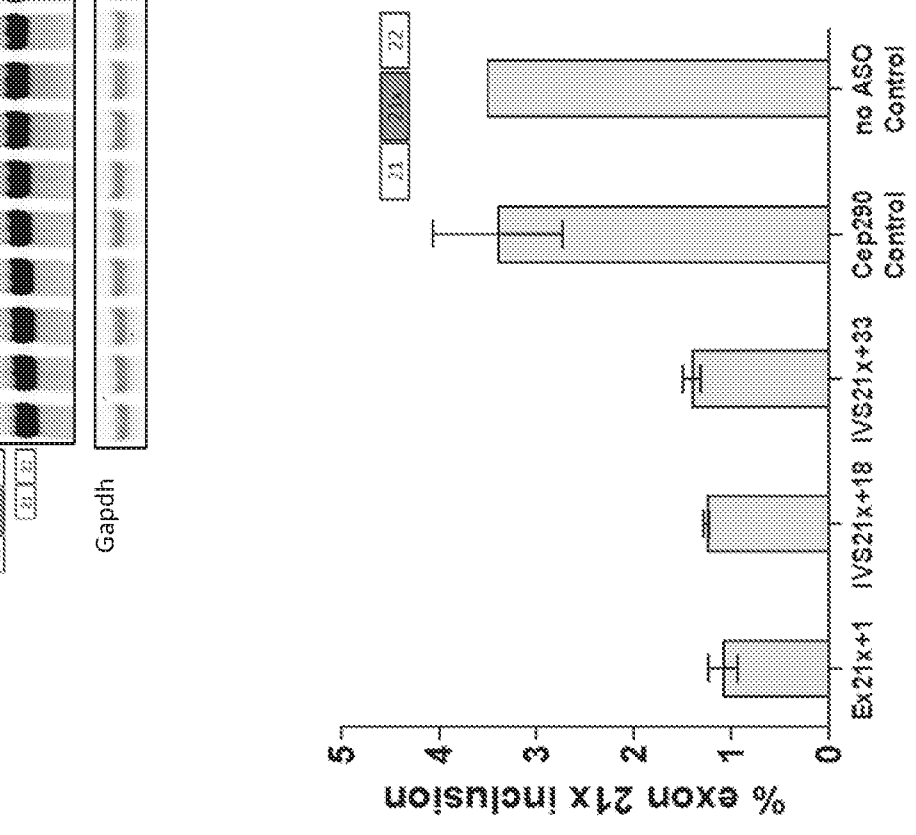


FIG. 10B

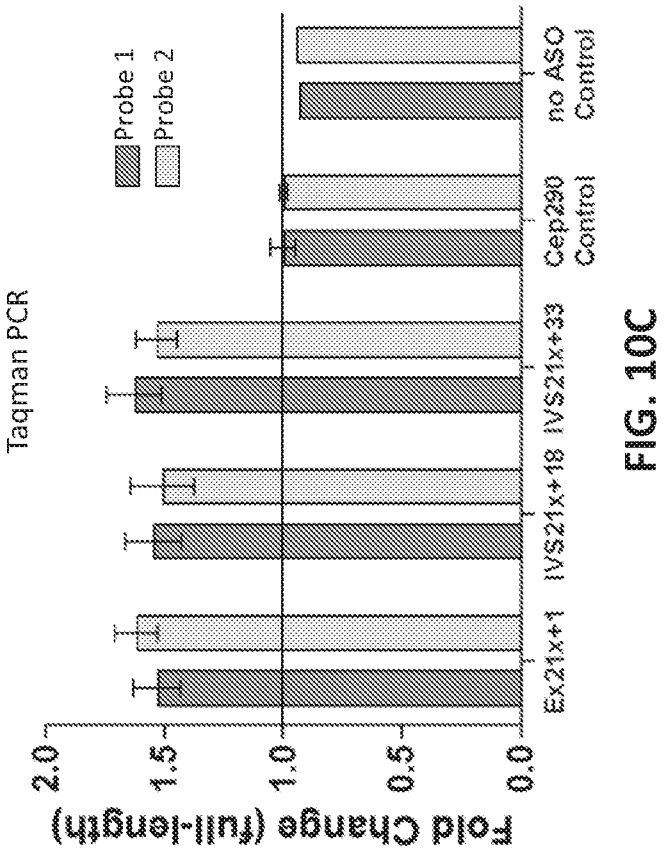


FIG. 10C

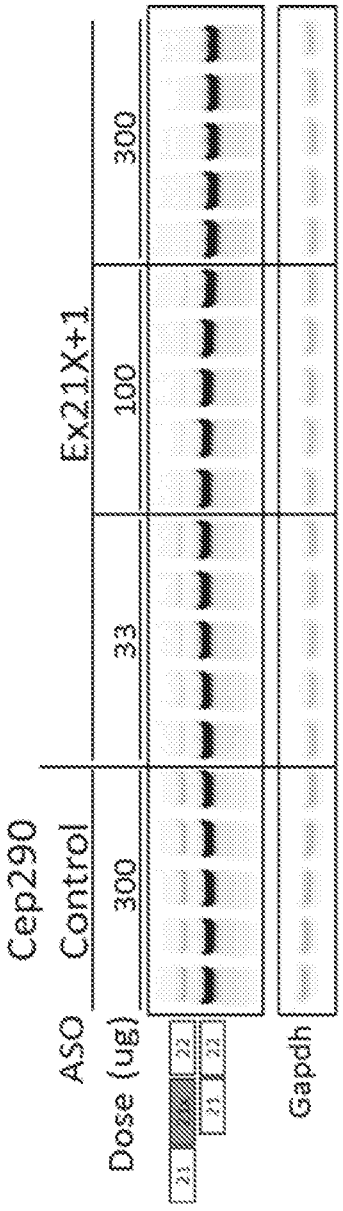


FIG. 11A

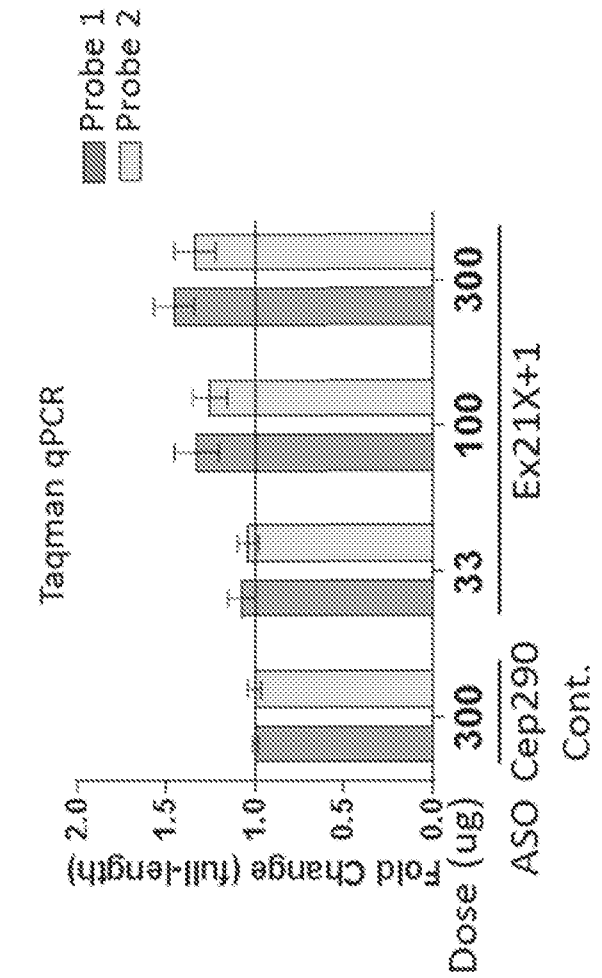


FIG. 11C

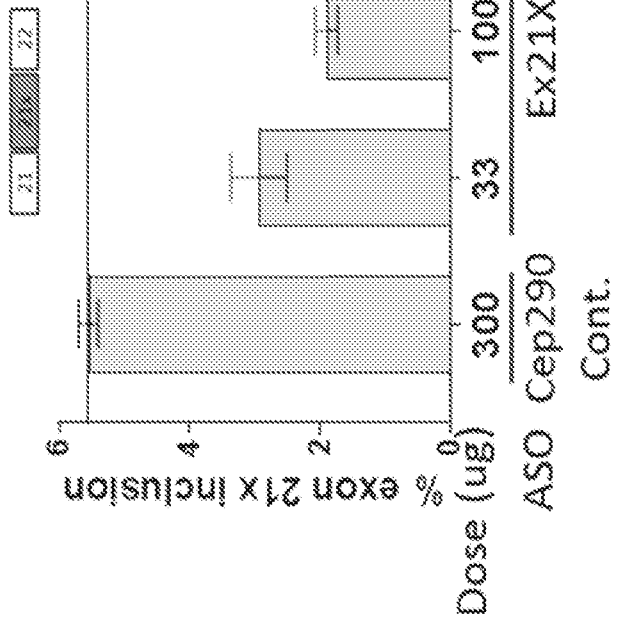


FIG. 11B

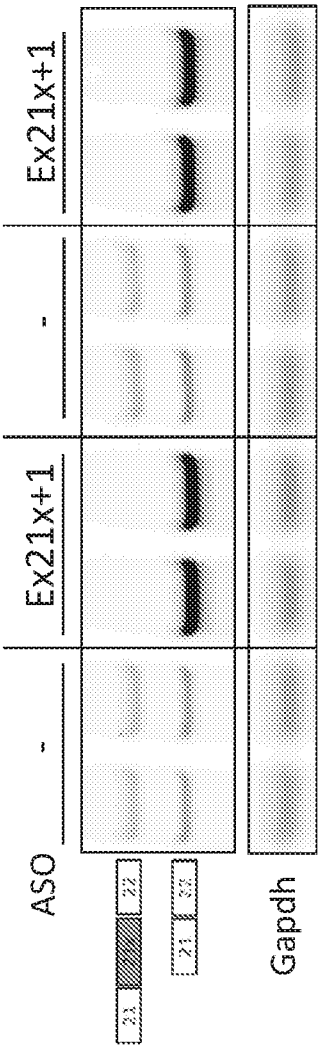


FIG. 12A

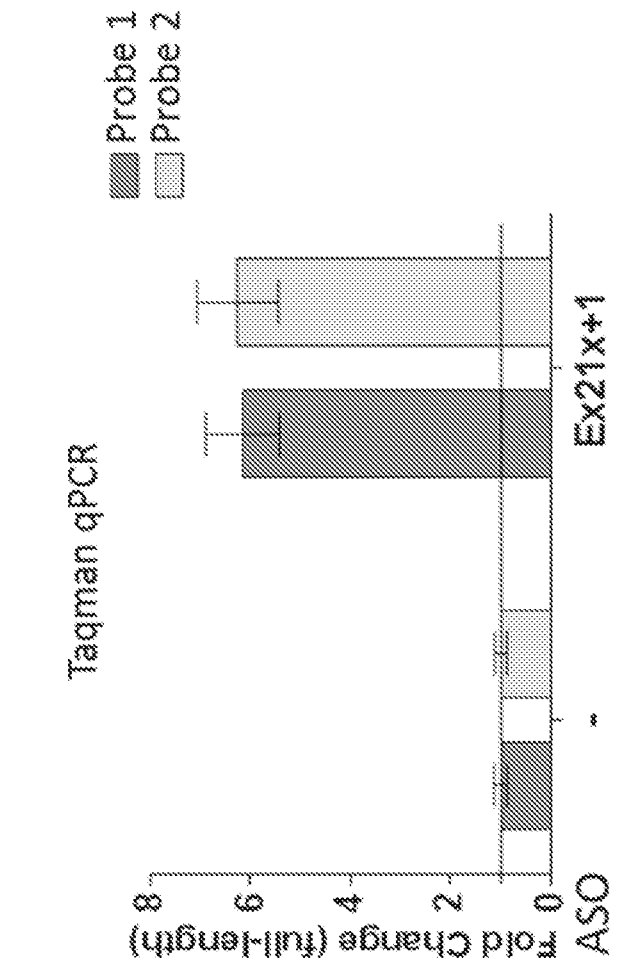


FIG. 12C

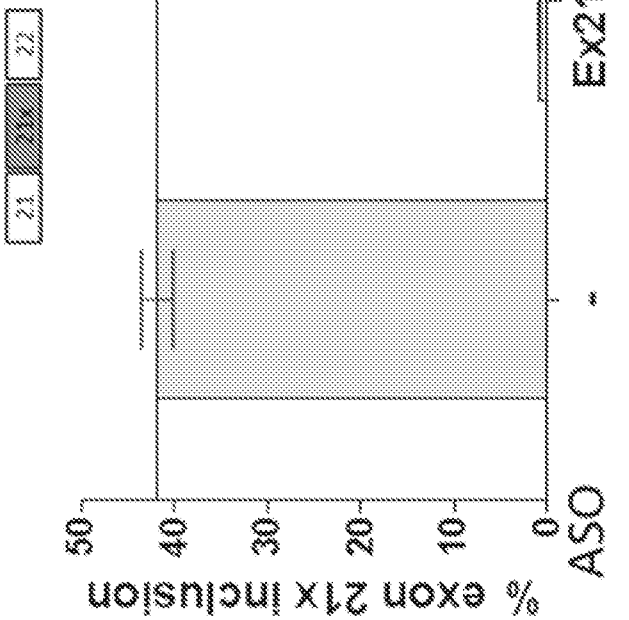


FIG. 12B

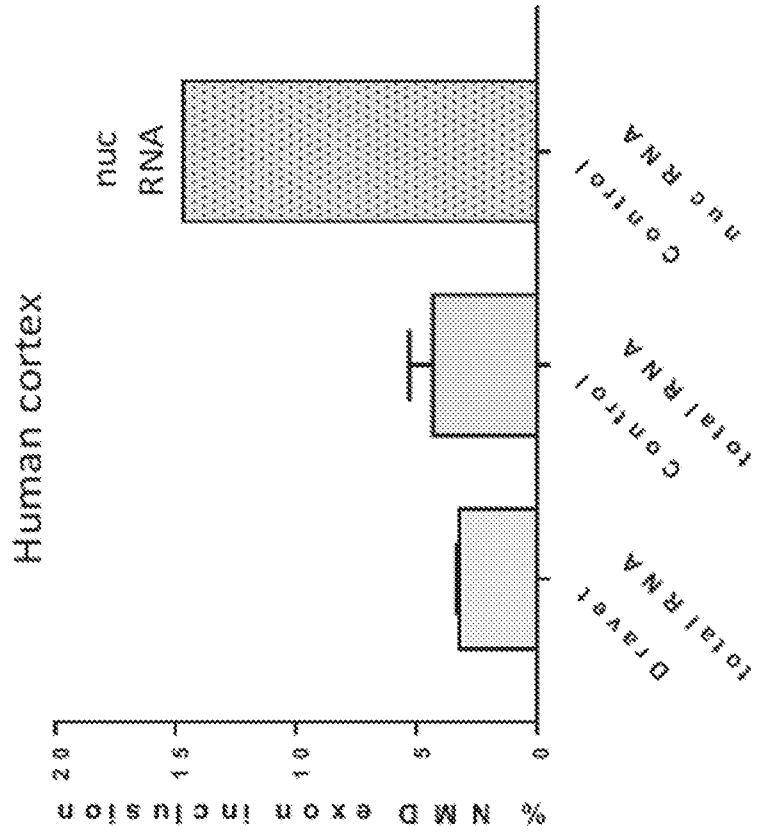


FIG. 13B

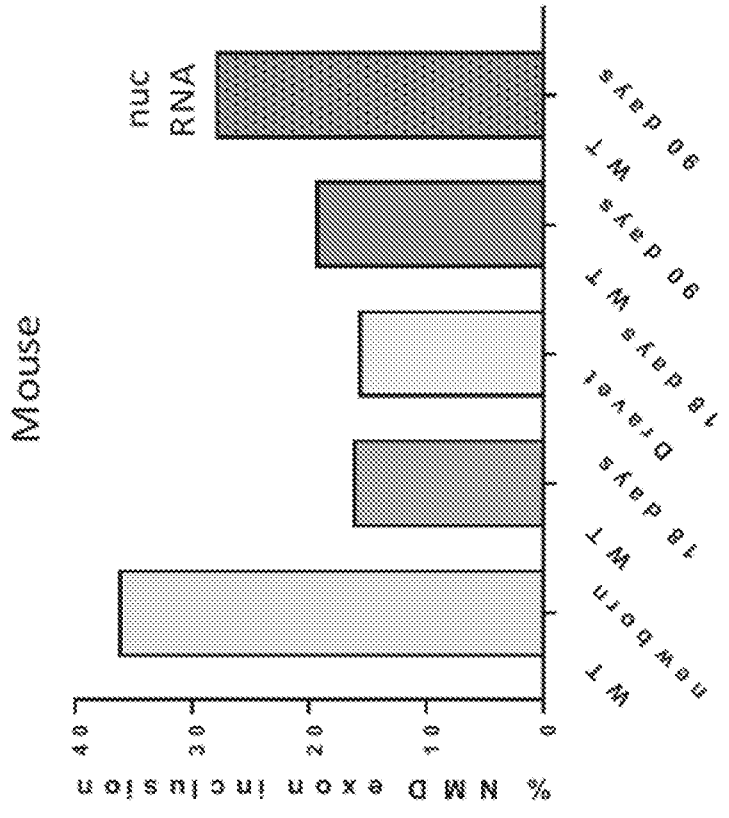


FIG. 13A

Decrease in NMD exon – RT-PCR (SYBR-safe)

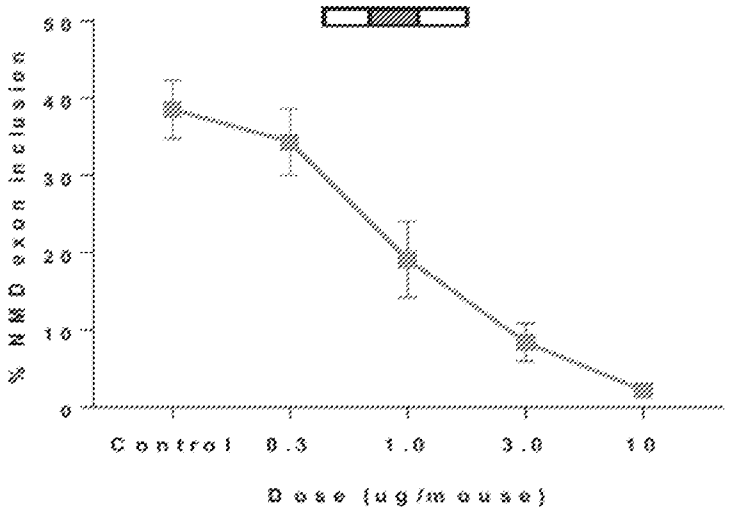


FIG. 14A

Scn1a mRNA increase – TaqmanqPCR

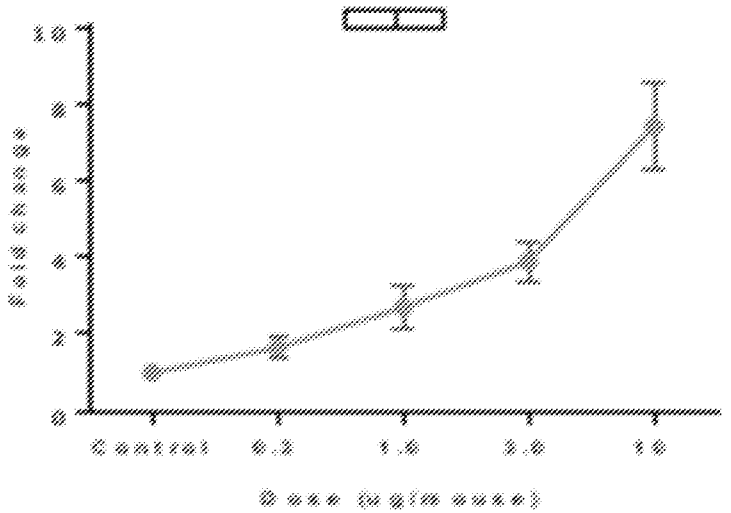


FIG. 14B

Increase of Nav 1.1 protein

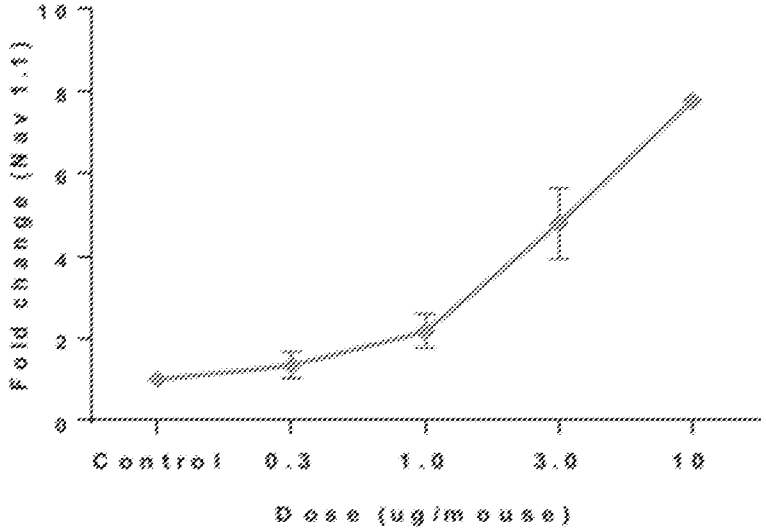


FIG. 14C

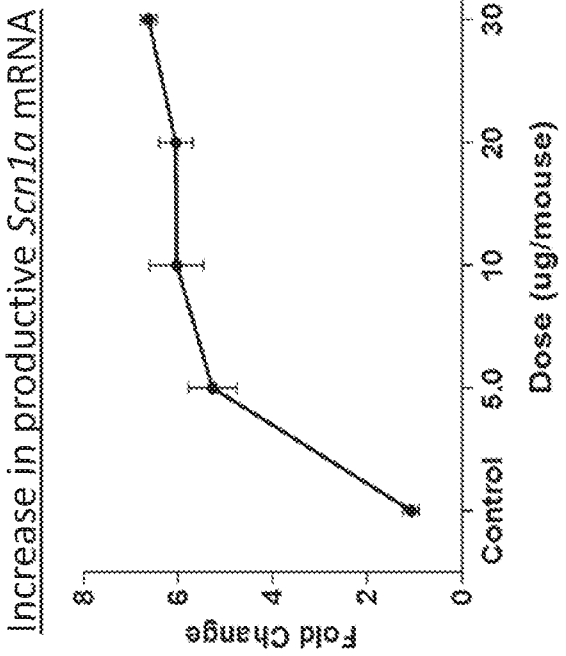


FIG. 15B

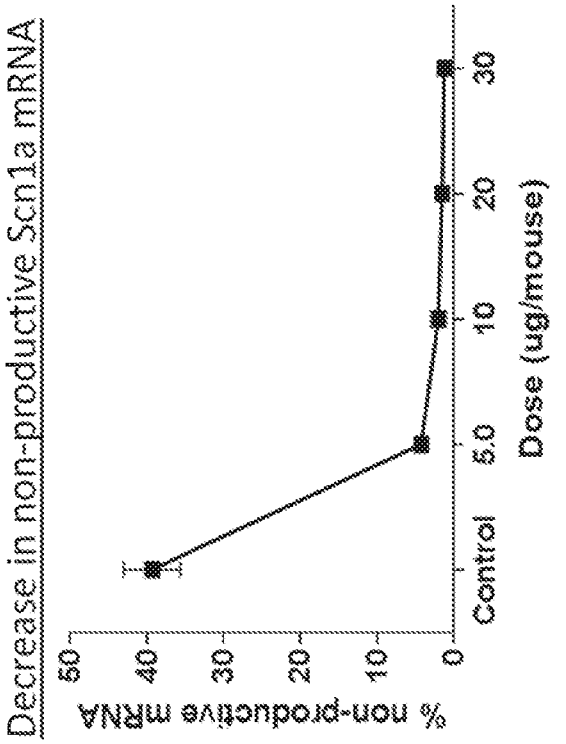


FIG. 15A

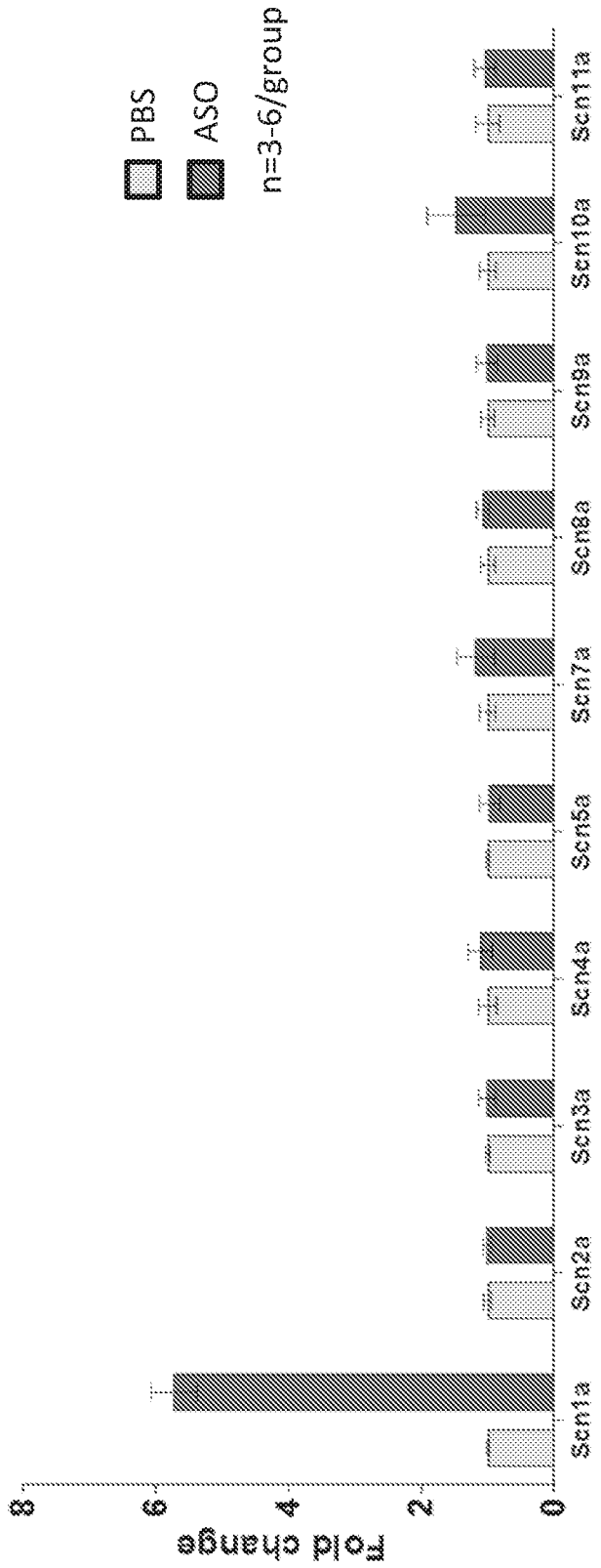

PBS
ASO
n=3-6/group
Fold change
0
2
4
6
8
Scn1a
Scn2a
Scn3a
Scn4a
Scn5a
Scn7a
Scn8a
Scn9a
Scn10a
Scn11a

FIG. 16

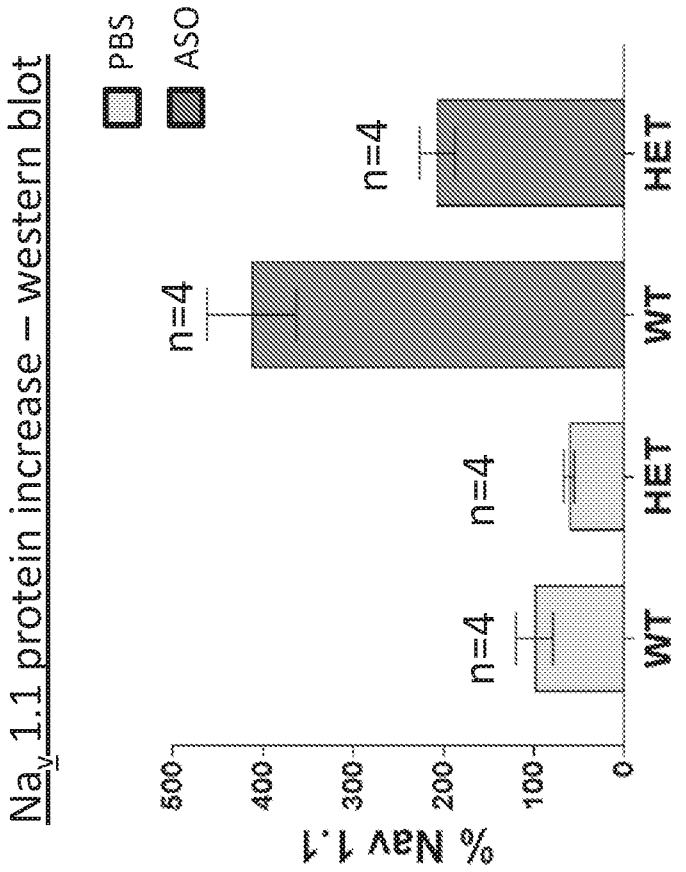


FIG. 17B

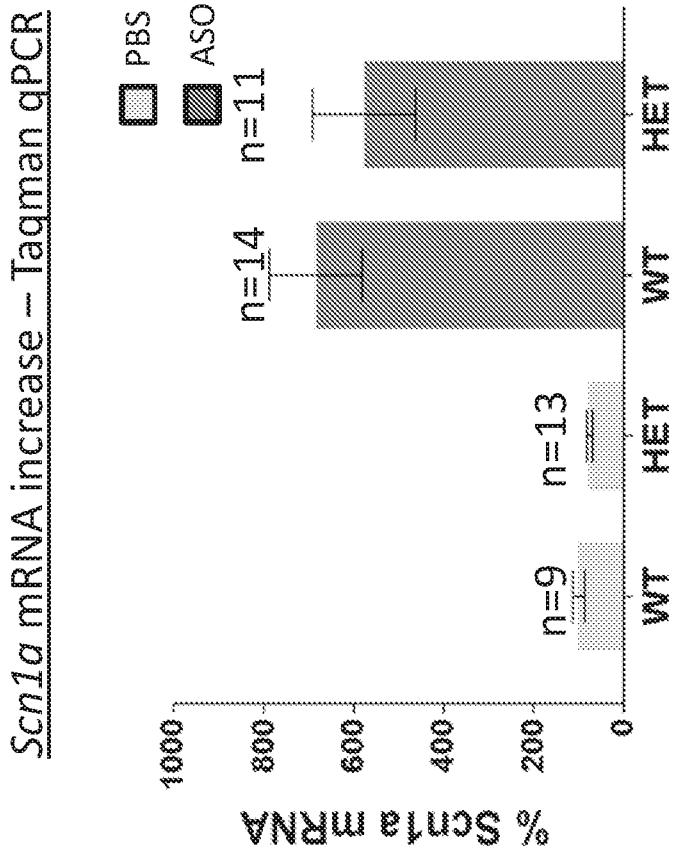


FIG. 17A

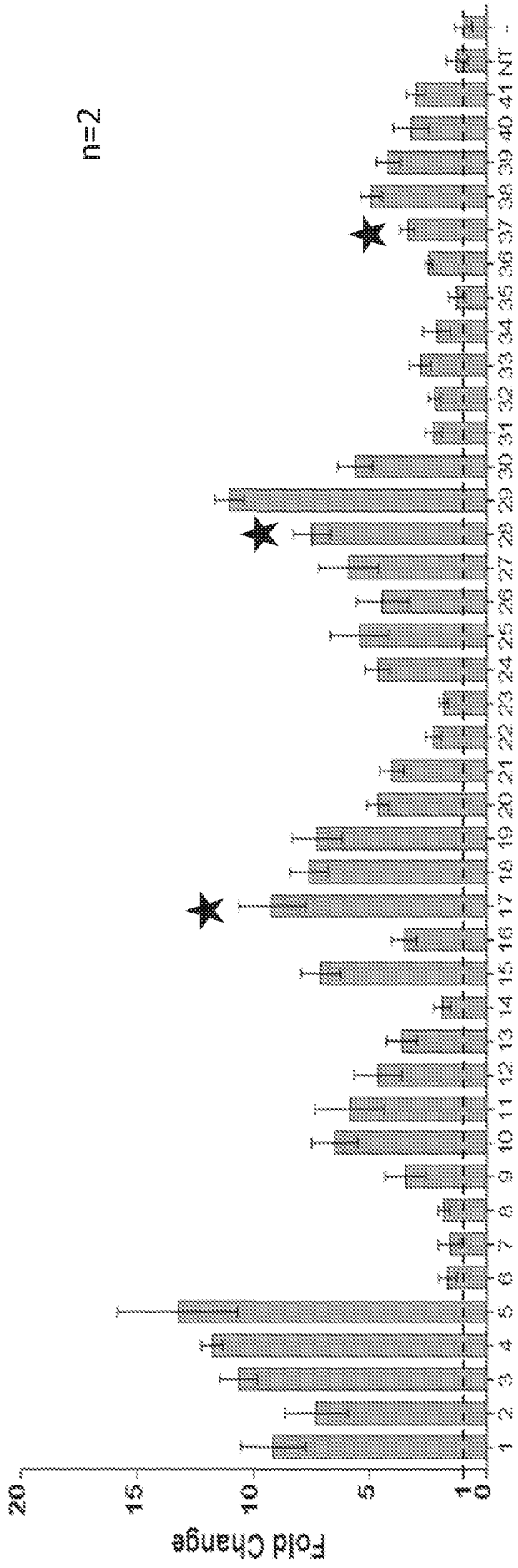

n=2
Fold Change
20
15
10
5
1
0
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28 29 30 31 32 33 34 35 36 37 38 39 40 41 NT -

FIG. 18

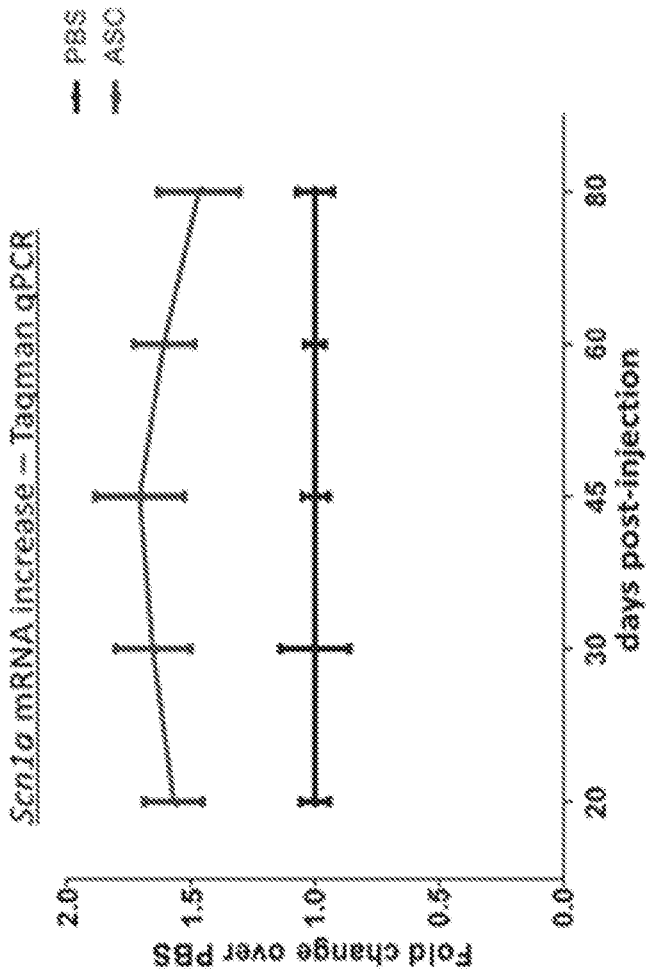

PBS
ASO
Scn1a mRNA increase – Taqman qPCR
Fold change over PBS
2.0
1.5
1.0
0.5
0.0
20
30
45
60
80
days post-injection

FIG. 19

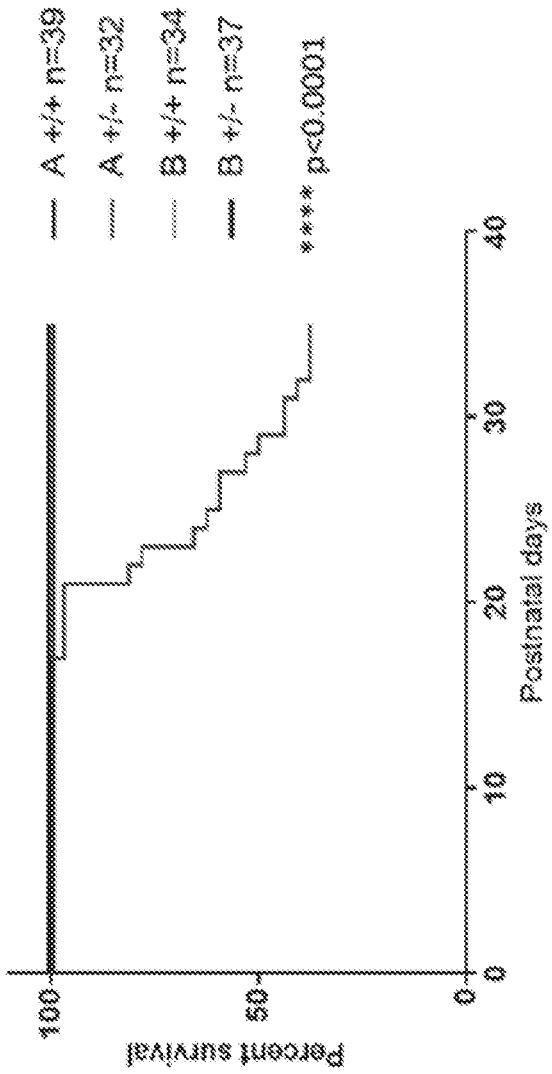

A +/+ n=39
A +/- n=32
B +/+ n=34
B +/- n=37
**** p<0.0001
Percent survival
100
50
0
0
10
20
30
40
Postnatal days

FIG. 20