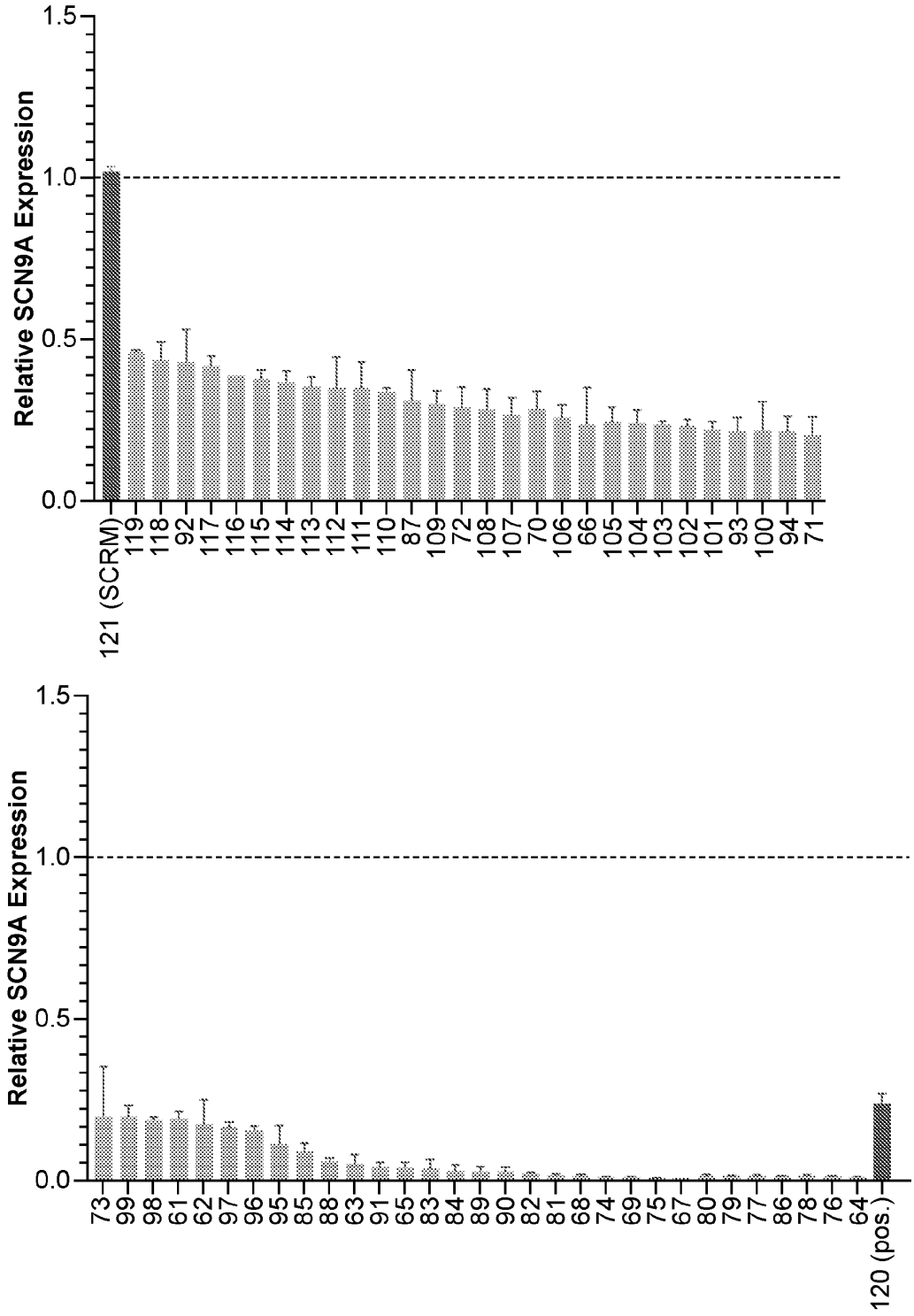

Relative SCN9A Expression
1.5
1.0
0.5
0.0
121 (SCRM)
119
118
92
117
116
115
114
113
112
111
110
87
109
72
108
107
70
106
66
105
104
103
102
101
93
100
94
71
Relative SCN9A Expression
1.5
1.0
0.5
0.0
73
99
98
61
62
97
96
95
85
88
63
91
65
83
84
89
90
82
81
68
74
69
75
67
80
79
77
86
78
76
64
120 (pos.)

FIG. 1

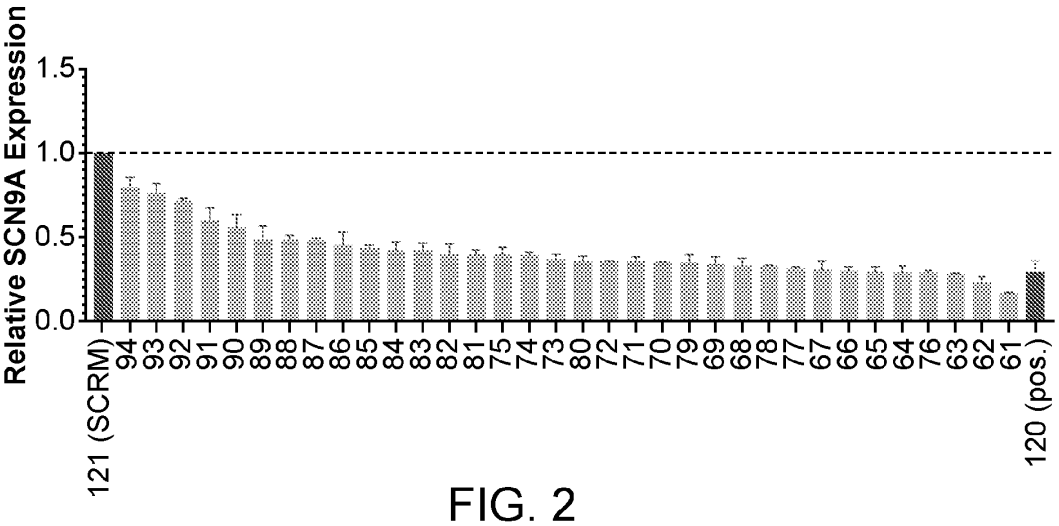

Relative SCN9A Expression
1.5
1.0
0.5
0.0
121 (SCRM)
94
93
92
91
90
89
88
87
86
85
84
83
82
81
75
74
73
80
72
71
70
79
69
68
78
77
67
66
65
64
76
63
62
61
120 (pos.)

FIG. 2

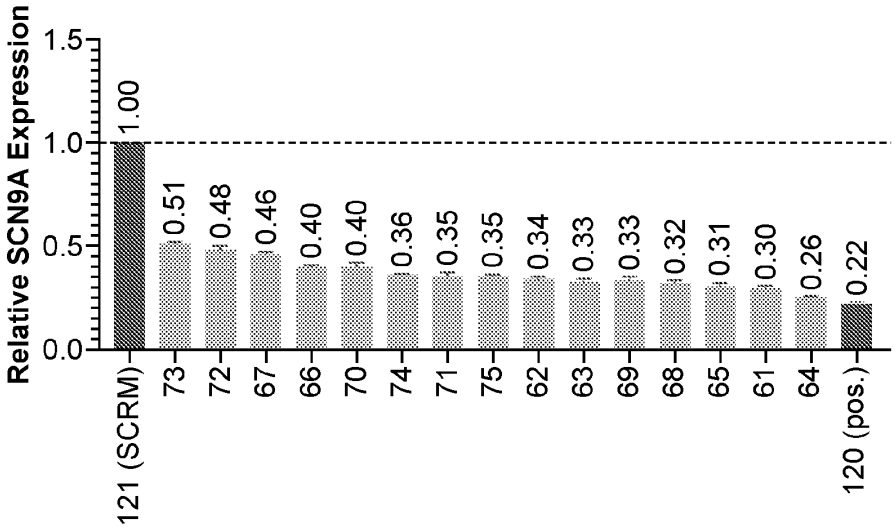

Relative SCN9A Expression
1.5
1.0
0.5
0.0
1.00
0.51
0.48
0.46
0.40
0.40
0.36
0.35
0.35
0.34
0.33
0.33
0.32
0.31
0.30
0.26
0.22
121 (SCRM)
73
72
67
66
70
74
71
75
62
63
69
68
65
61
64
120 (pos.)

FIG. 3

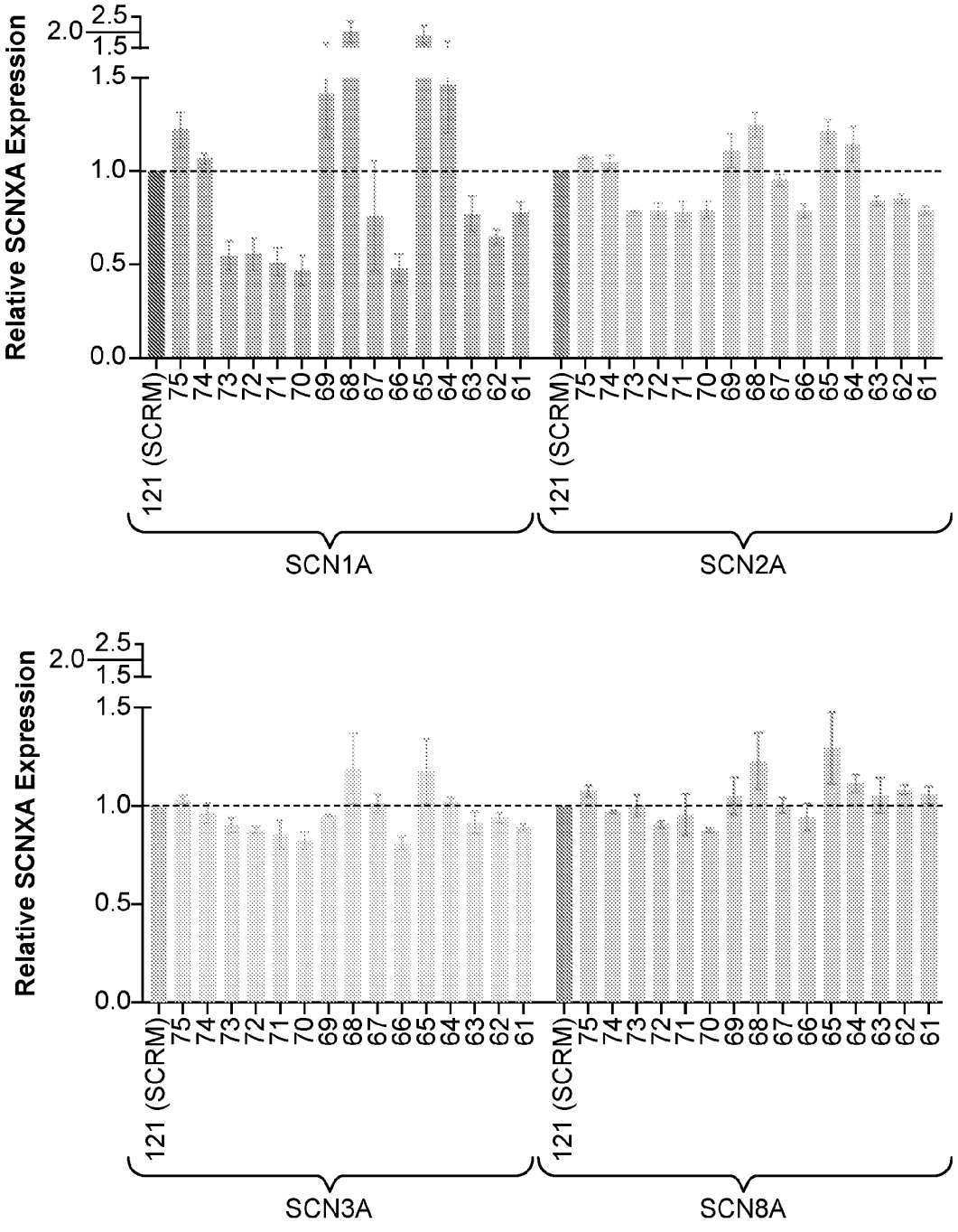

Relative SCNXA Expression
2.0
2.5
1.5
1.5
1.0
0.5
0.0
121 (SCRM)
75
74
73
72
71
70
69
68
67
66
65
64
63
62
61
SCN1A
SCN2A
SCN3A
SCN8A

FIG. 4

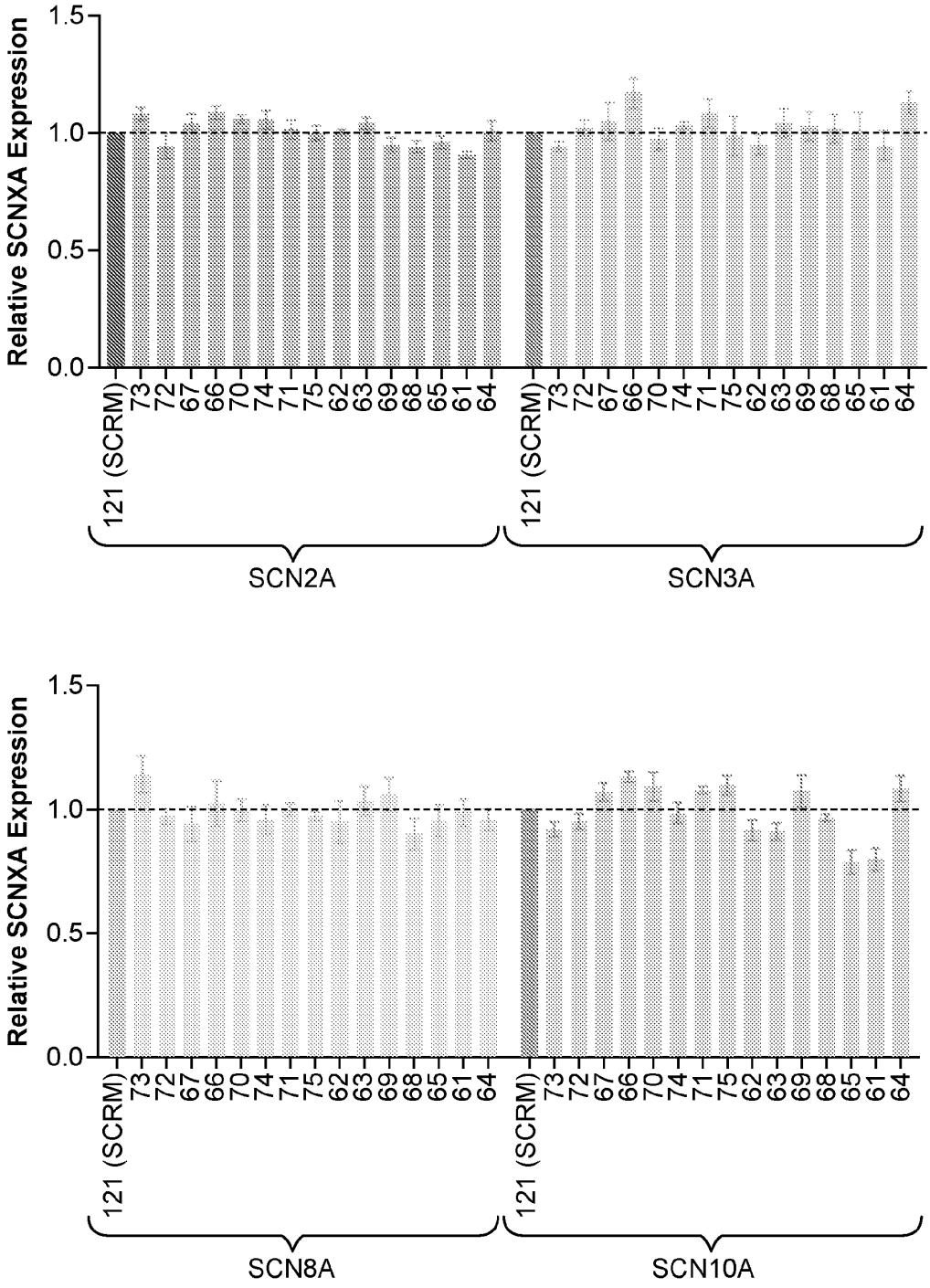

Relative SCNXA Expression
1.5
1.0
0.5
0.0
121 (SCRM)
73
72
67
66
70
74
71
75
62
63
69
68
65
61
64
SCN2A
SCN3A
SCN8A
SCN10A

FIG. 5

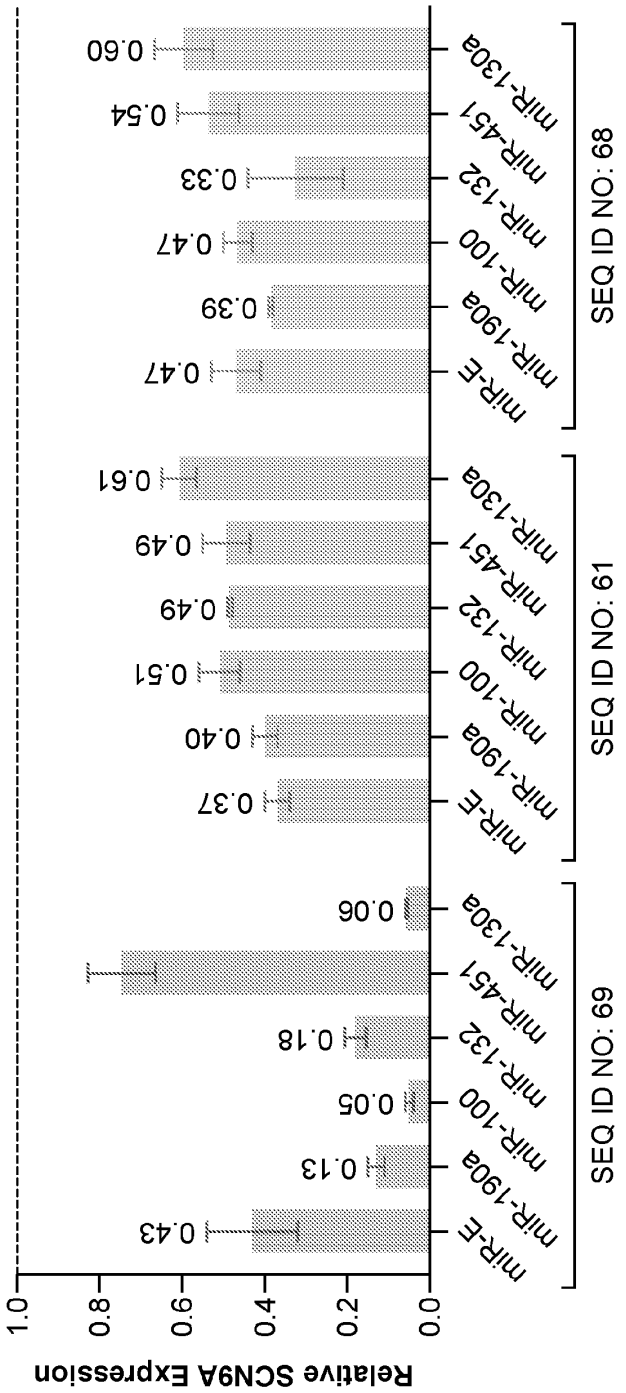

Relative SCN9A Expression
1.0
0.8
0.6
0.4
0.2
0.0
0.43
0.13
0.05
0.18
0.06
0.37
0.40
0.51
0.49
0.49
0.61
0.47
0.39
0.47
0.33
0.54
0.60
miR-E
miR-190a
miR-100
miR-132
miR-451
miR-130a
SEQ ID NO: 69
SEQ ID NO: 61
SEQ ID NO: 68

FIG. 6

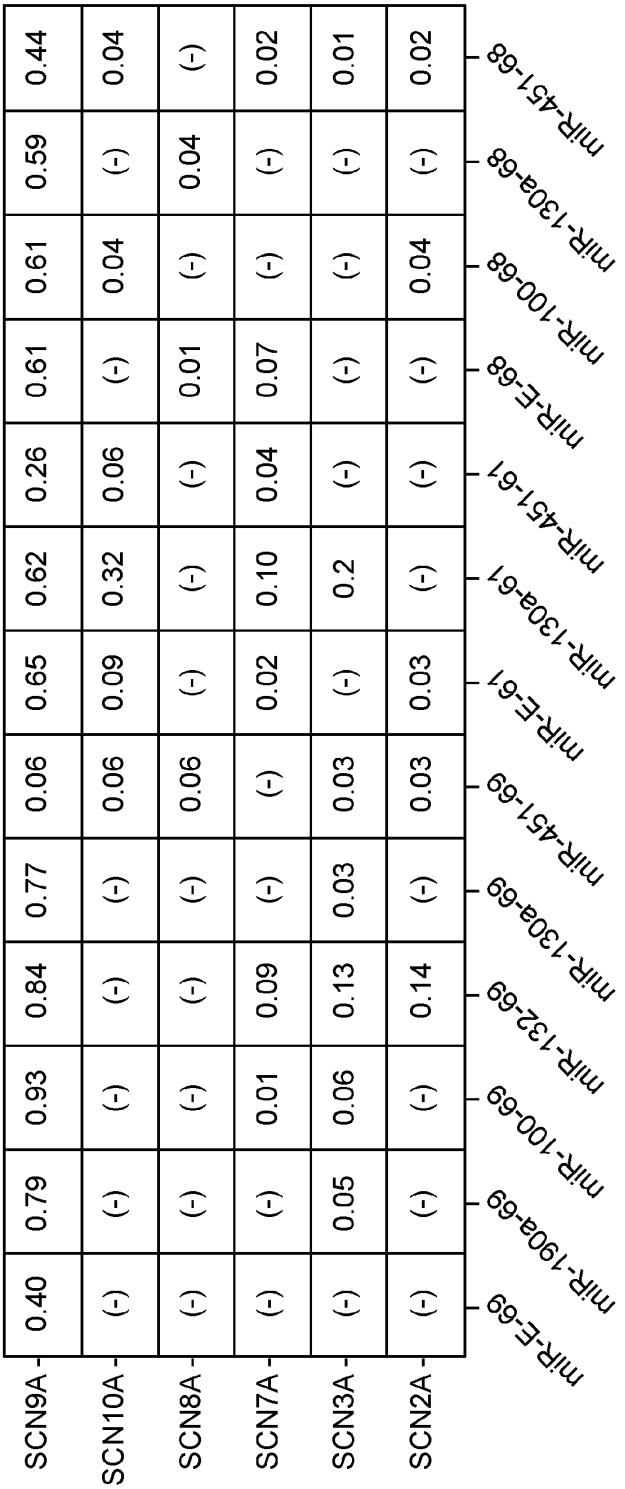

miR-E-69
miR-190a-69
miR-100-69
miR-132-69
miR-130a-69
miR-451-69
miR-E-67
miR-130a-67
miR-451-67
miR-E-68
miR-100-68
miR-130a-68
miR-451-68
SCN9A
SCN10A
SCN8A
SCN7A
SCN3A
SCN2A
0.40 0.79 0.93 0.84 0.77 0.06 0.65 0.62 0.26 0.61 0.61 0.59 0.44
(-) (-) (-) (-) (-) 0.06 0.09 0.32 0.06 (-) 0.04 (-) 0.04
(-) (-) (-) (-) (-) 0.06 (-) (-) (-) 0.01 (-) 0.04 (-)
(-) (-) 0.01 0.09 (-) (-) 0.02 0.10 0.04 0.07 (-) (-) 0.02
(-) 0.05 0.06 0.13 0.03 0.03 (-) 0.2 (-) (-) (-) (-) 0.01
(-) (-) (-) 0.14 (-) 0.03 0.03 (-) (-) (-) 0.04 (-) 0.02

FIG. 7

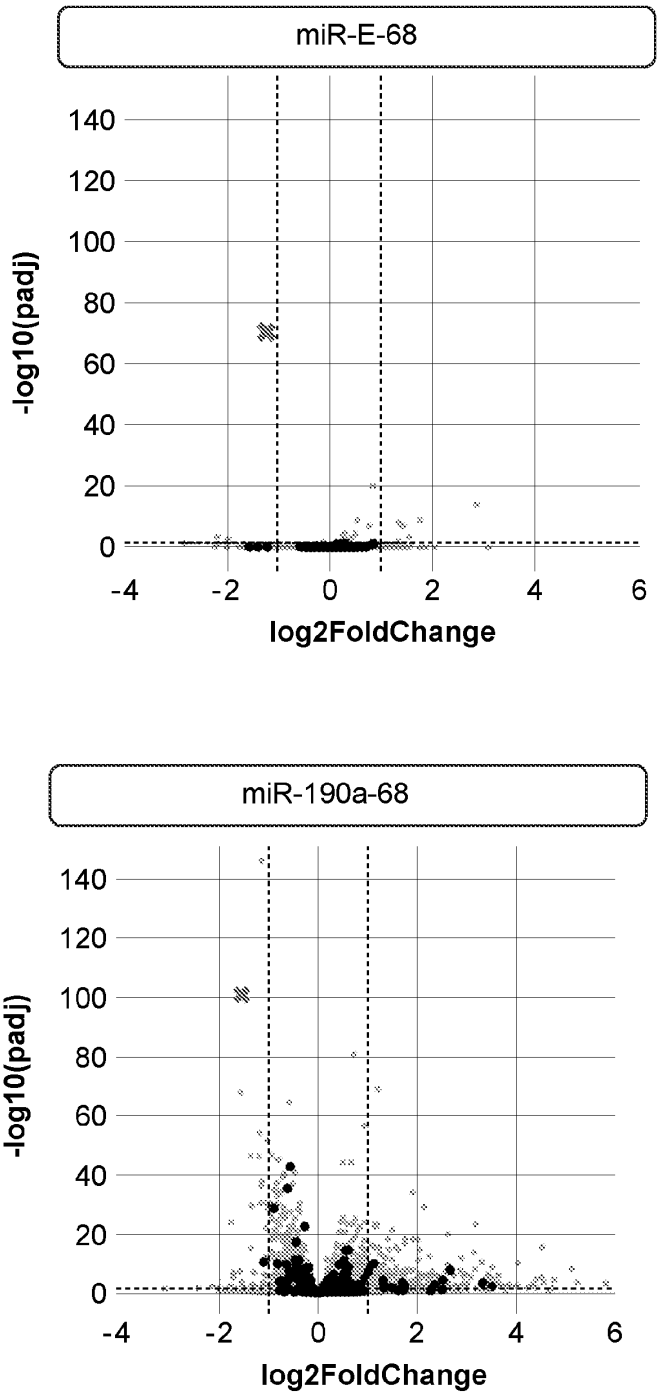

miR-E-68
140
120
100
80
60
40
20
0
-log10(padj)
-4
-2
0
2
4
6
log2FoldChange
miR-190a-68
140
120
100
80
60
40
20
0
-log10(padj)
-4
-2
0
2
4
6
log2FoldChange

FIG. 8A

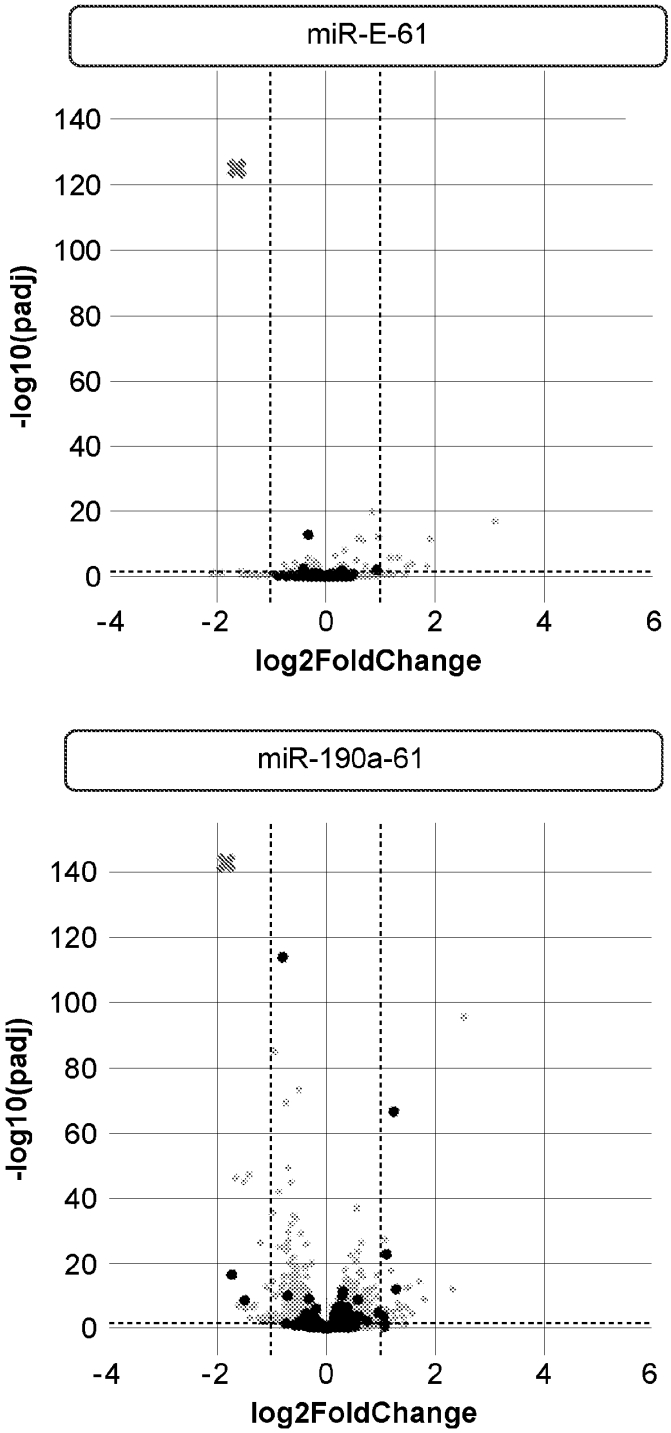

miR-E-61
140
120
100
80
60
40
20
0
-log10(padj)
-4
-2
0
2
4
6
log2FoldChange
miR-190a-61
140
120
100
80
60
40
20
0
-log10(padj)
-4
-2
0
2
4
6
log2FoldChange

FIG. 8B

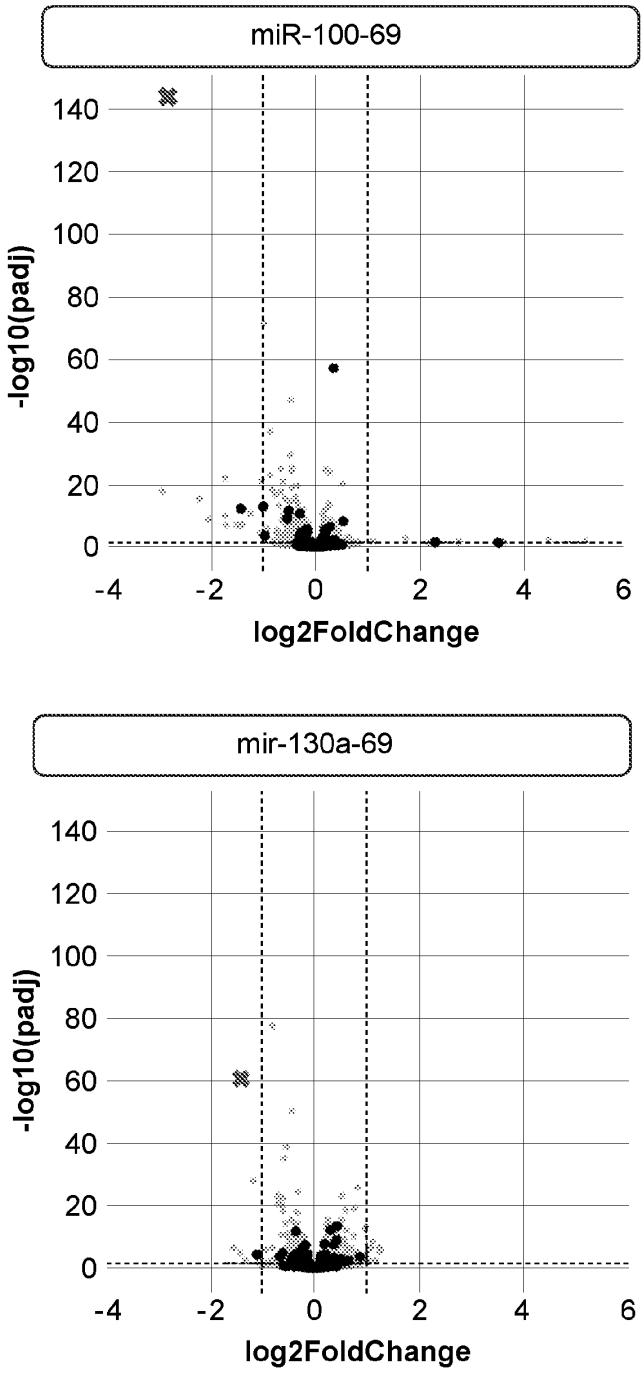

miR-100-69
140
120
100
80
60
40
20
0
-log10(padj)
-4
-2
0
2
4
6
log2FoldChange
mir-130a-69
140
120
100
80
60
40
20
0
-log10(padj)
-4
-2
0
2
4
6
log2FoldChange

FIG. 8C

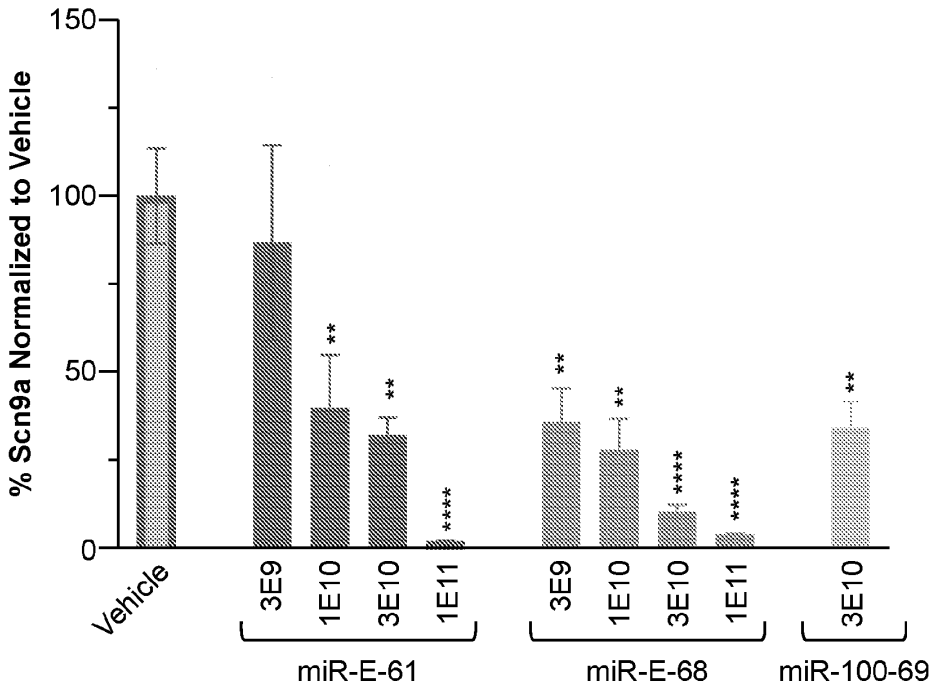

% Scn9a Normalized to Vehicle
150
100
50
0
Vehicle
3E9
1E10
3E10
1E11
3E9
1E10
3E10
1E11
3E10
**
**
****
**
**
****
**
miR-E-61
miR-E-68
miR-100-69

FIG. 9A

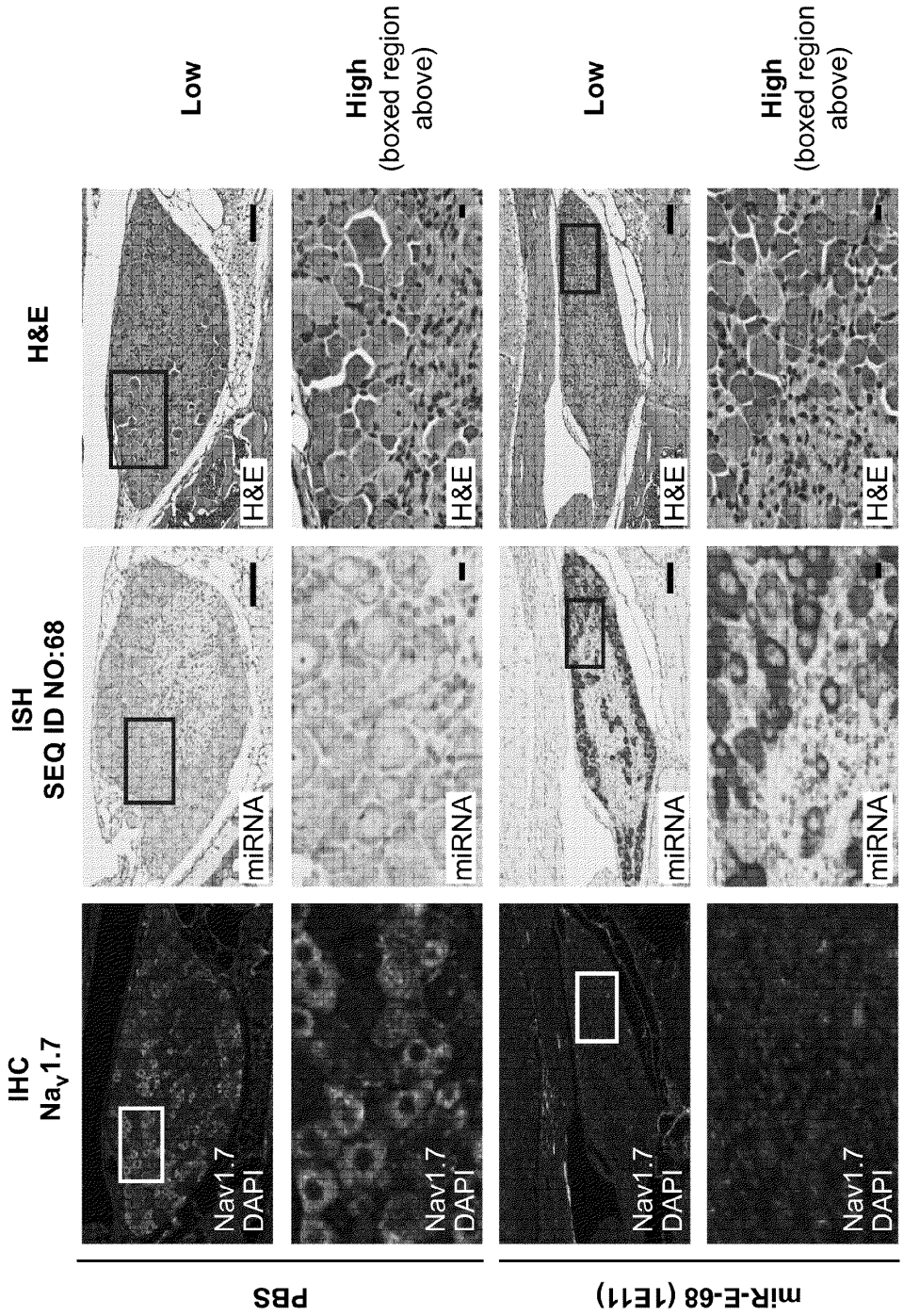

IHC
Nav1.7
ISH
SEQ ID NO:68
H&E
Low
High
(boxed region
above)
Low
High
(boxed region
above)
Nav1.7
DAPI
miRNA
H&E
PBS
miR-E-68 (1E11)

FIG. 9B

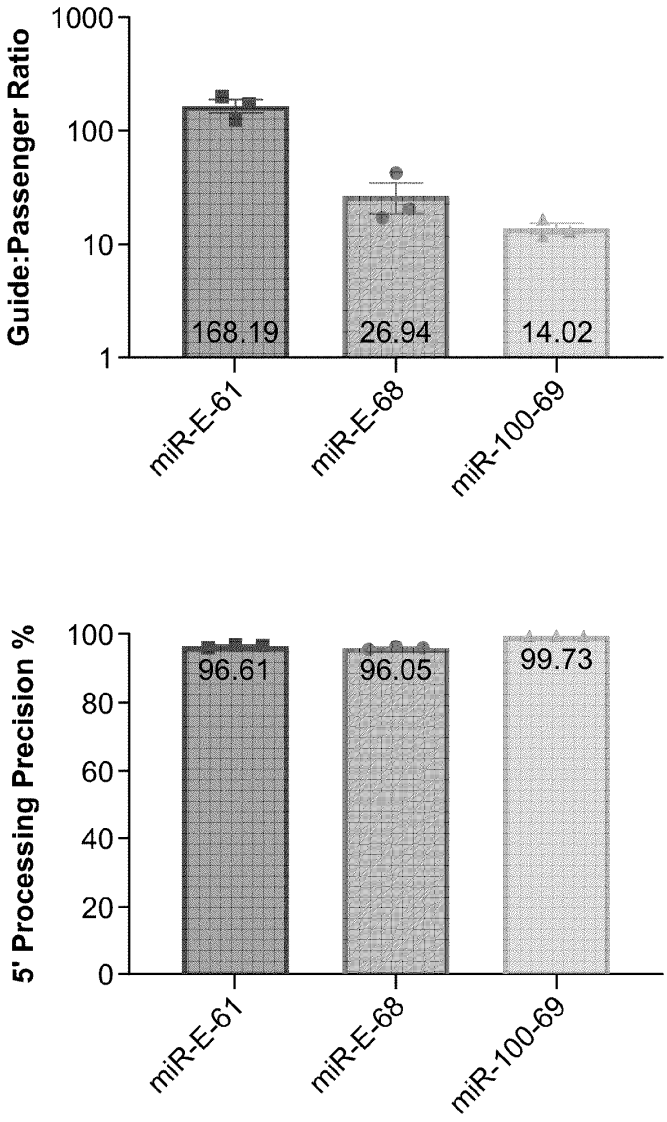

Guide:Passenger Ratio
1000
100
10
1
168.19
26.94
14.02
miR-E-61
miR-E-68
miR-100-69
5' Processing Precision %
100
80
60
40
20
0
96.61
96.05
99.73
miR-E-61
miR-E-68
miR-100-69

FIG. 9C

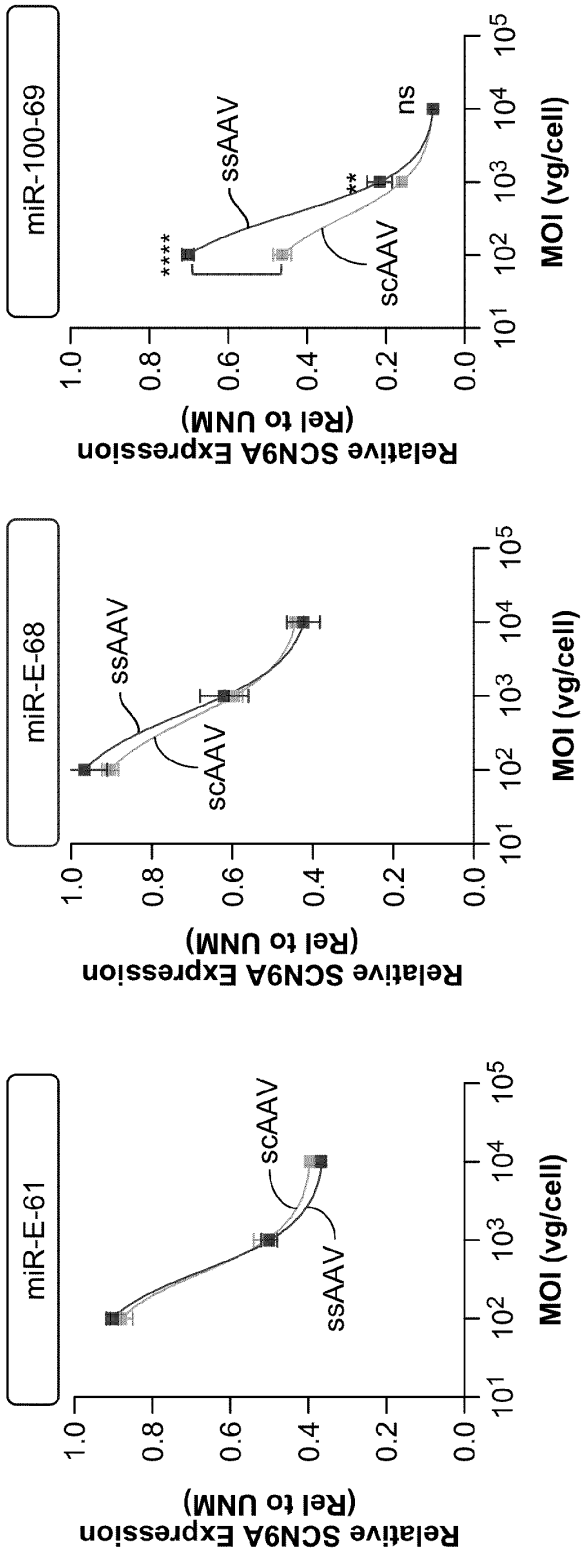

miR-100-69
miR-E-68
miR-E-61
ssAAV
scAAV
****
**
ns
Relative SCN9A Expression
(Rel to UNM)
MOI (vg/cell)
0.0
0.2
0.4
0.6
0.8
1.0
10^1
10^2
10^3
10^4
10^5

FIG. 10

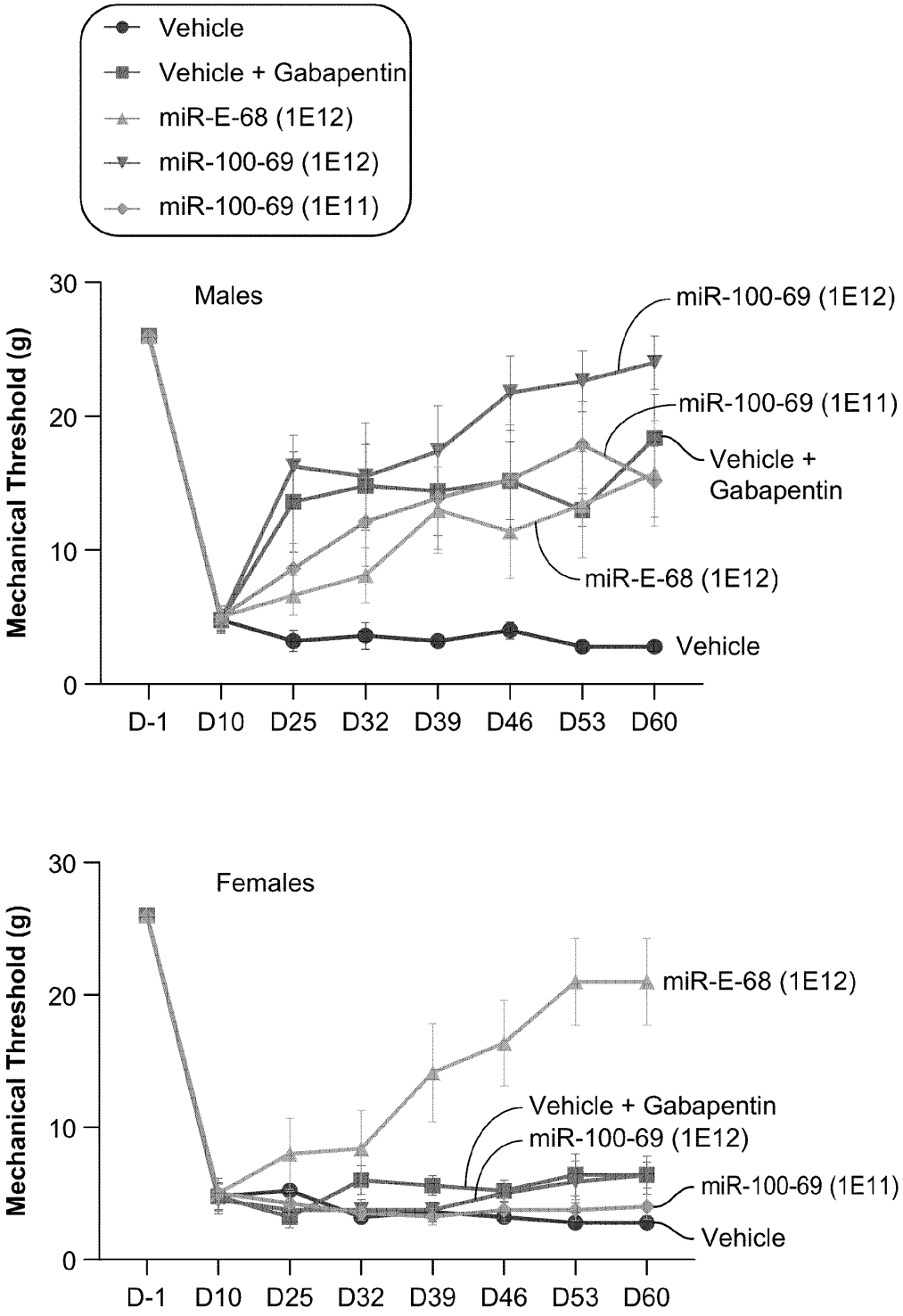

Vehicle
Vehicle + Gabapentin
miR-E-68 (1E12)
miR-100-69 (1E12)
miR-100-69 (1E11)
Males
Mechanical Threshold (g)
30
20
10
0
D-1
D10
D25
D32
D39
D46
D53
D60
miR-100-69 (1E12)
miR-100-69 (1E11)
Vehicle + Gabapentin
miR-E-68 (1E12)
Vehicle
Females
Mechanical Threshold (g)
30
20
10
0
D-1
D10
D25
D32
D39
D46
D53
D60
miR-E-68 (1E12)
Vehicle + Gabapentin
miR-100-69 (1E12)
miR-100-69 (1E11)
Vehicle

FIG. 11A

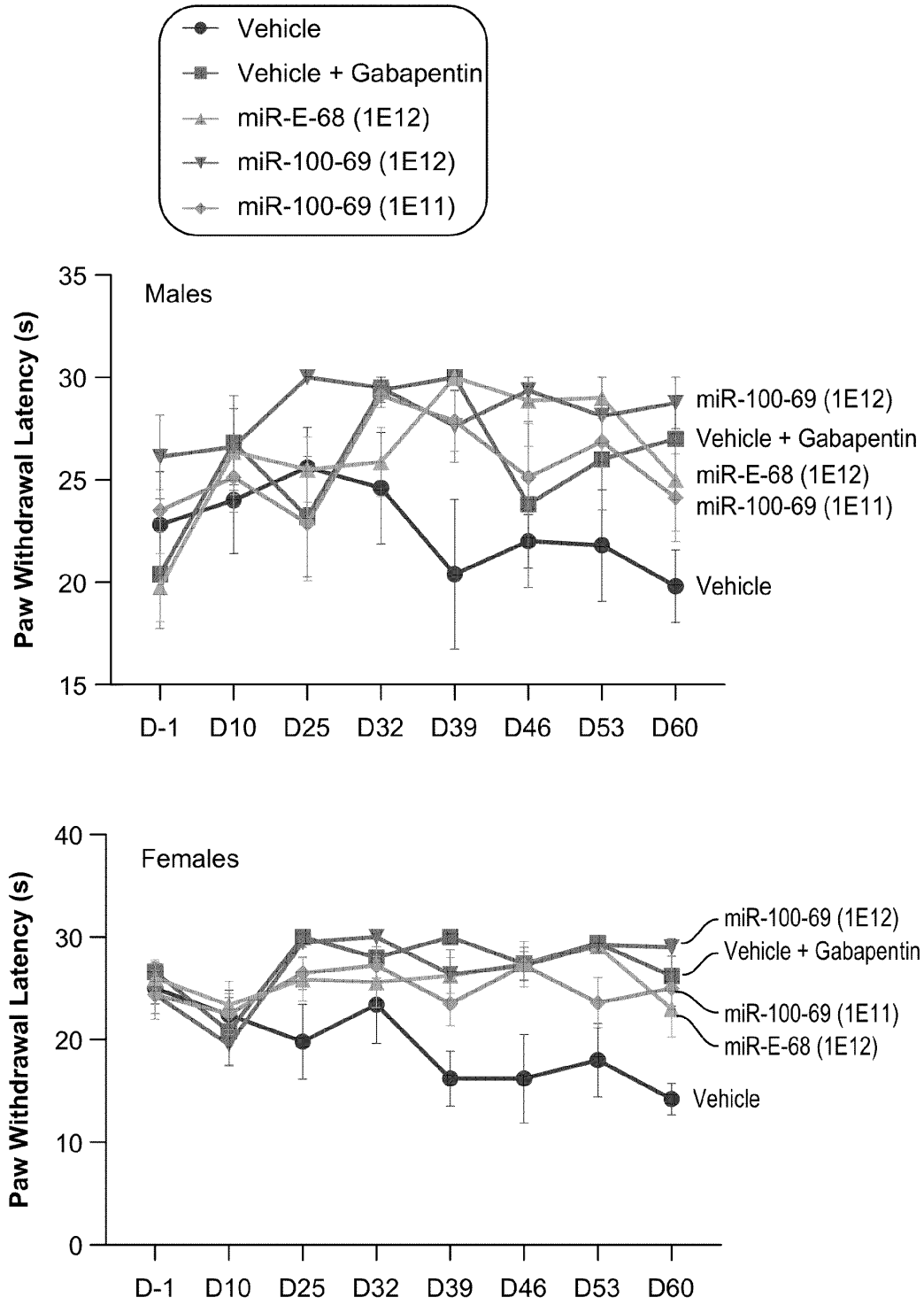

Vehicle
Vehicle + Gabapentin
miR-E-68 (1E12)
miR-100-69 (1E12)
miR-100-69 (1E11)
Males
Paw Withdrawal Latency (s)
35
30
25
20
15
D-1
D10
D25
D32
D39
D46
D53
D60
miR-100-69 (1E12)
Vehicle + Gabapentin
miR-E-68 (1E12)
miR-100-69 (1E11)
Vehicle
Females
Paw Withdrawal Latency (s)
40
30
20
10
0
D-1
D10
D25
D32
D39
D46
D53
D60
miR-100-69 (1E12)
Vehicle + Gabapentin
miR-100-69 (1E11)
miR-E-68 (1E12)
Vehicle

FIG. 11B

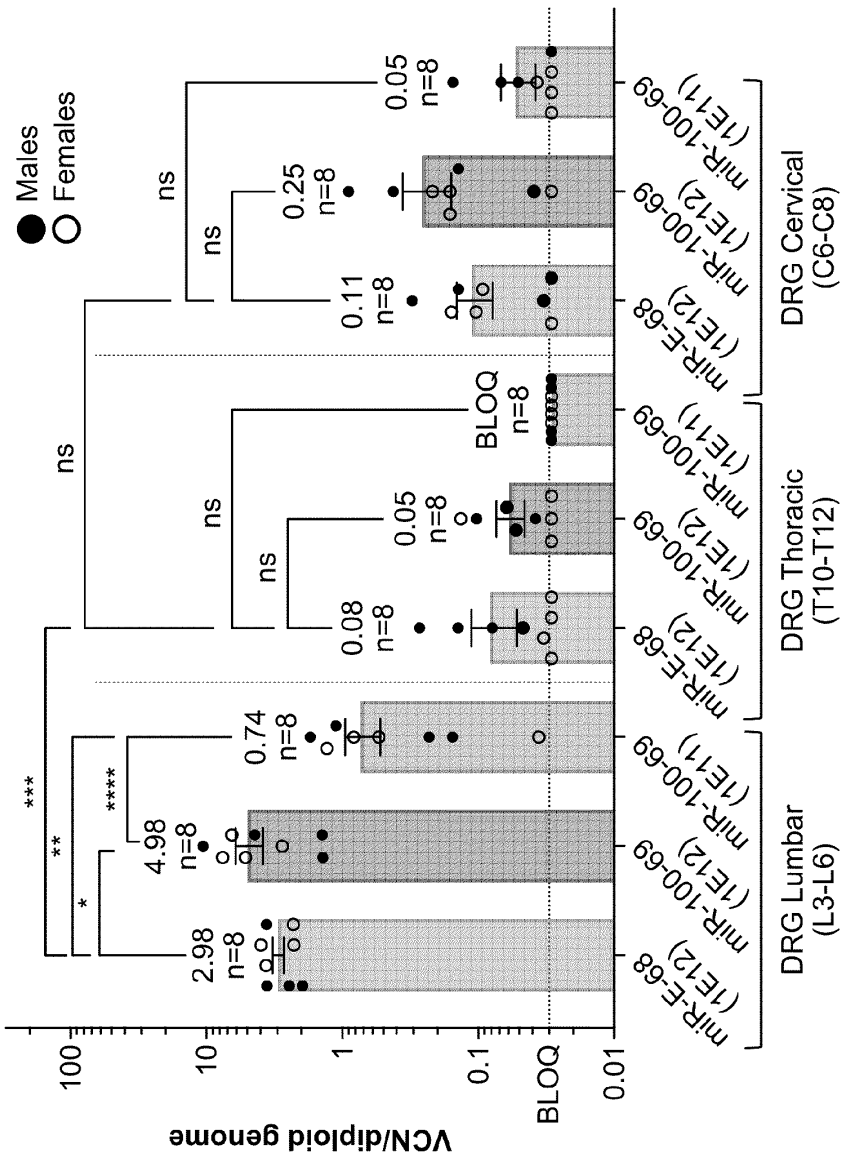

Males
Females
VCN/diploid genome
100
10
1
0.1
0.01
BLOQ
***
**
*
****
ns
2.98 n=8
4.98 n=8
0.74 n=8
0.08 n=8
0.05 n=8
BLOQ n=8
0.11 n=8
0.25 n=8
0.05 n=8
miR-E-68 (1E12)
miR-100-69 (1E12)
miR-100-69 (1E11)
DRG Lumbar (L3-L6)
DRG Thoracic (T10-T12)
DRG Cervical (C6-C8)

FIG. 12A

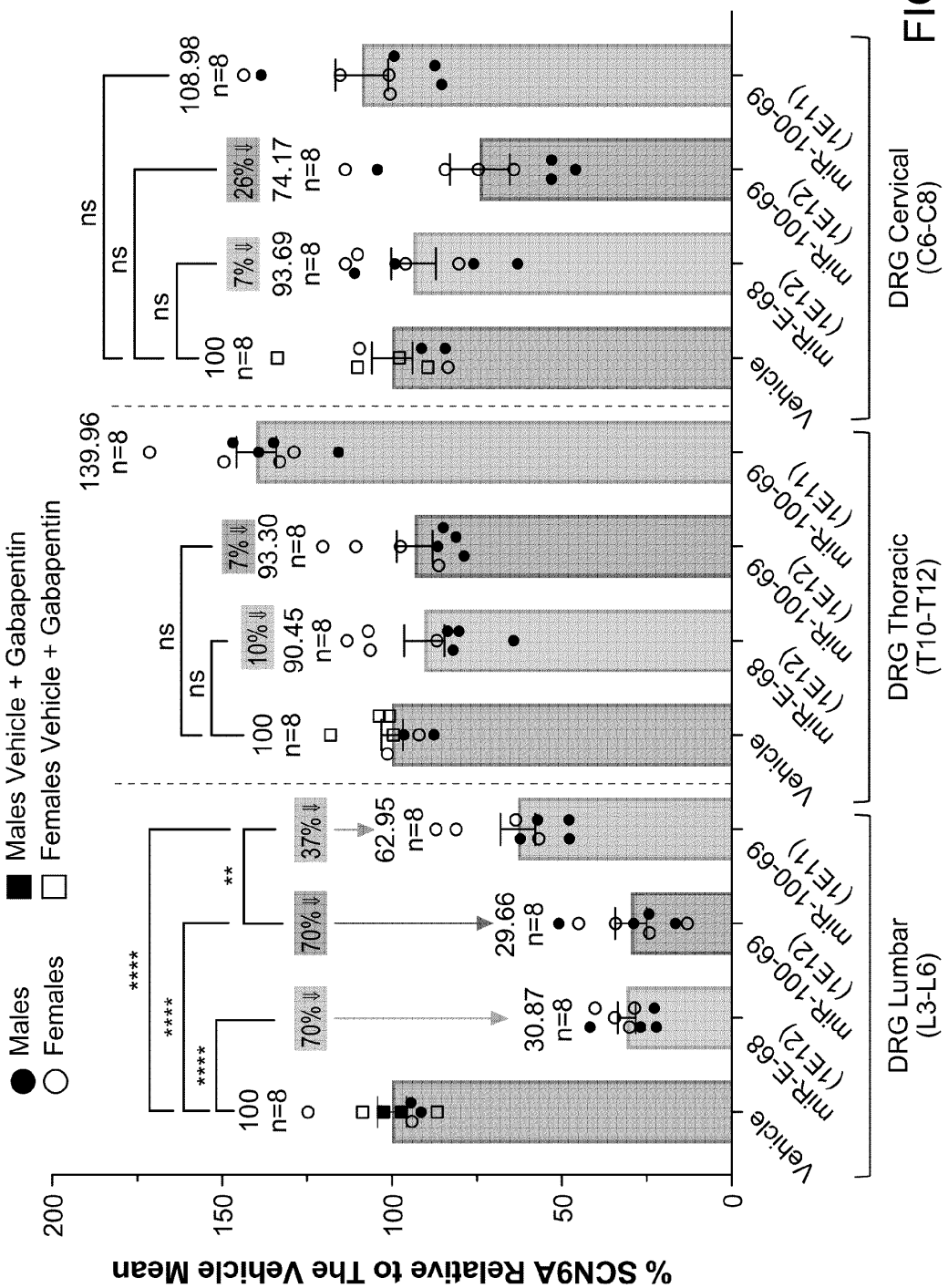

% SCN9A Relative to The Vehicle Mean
200
150
100
50
0
Males
Females
Males Vehicle + Gabapentin
Females Vehicle + Gabapentin
****
****
****
**
ns
ns
ns
ns
ns
ns
100 n=8
70% ⇓
30.87 n=8
70% ⇓
29.66 n=8
37% ⇓
62.95 n=8
100 n=8
10% ⇓
90.45 n=8
7% ⇓
93.30 n=8
139.96 n=8
100 n=8
7% ⇓
93.69 n=8
26% ⇓
74.17 n=8
108.98 n=8
Vehicle
miR-E-68 (1E12)
miR-100-69 (1E12)
miR-100-69 (1E11)
DRG Lumbar (L3-L6)
DRG Thoracic (T10-T12)
DRG Cervical (C6-C8)

FIG. 12B

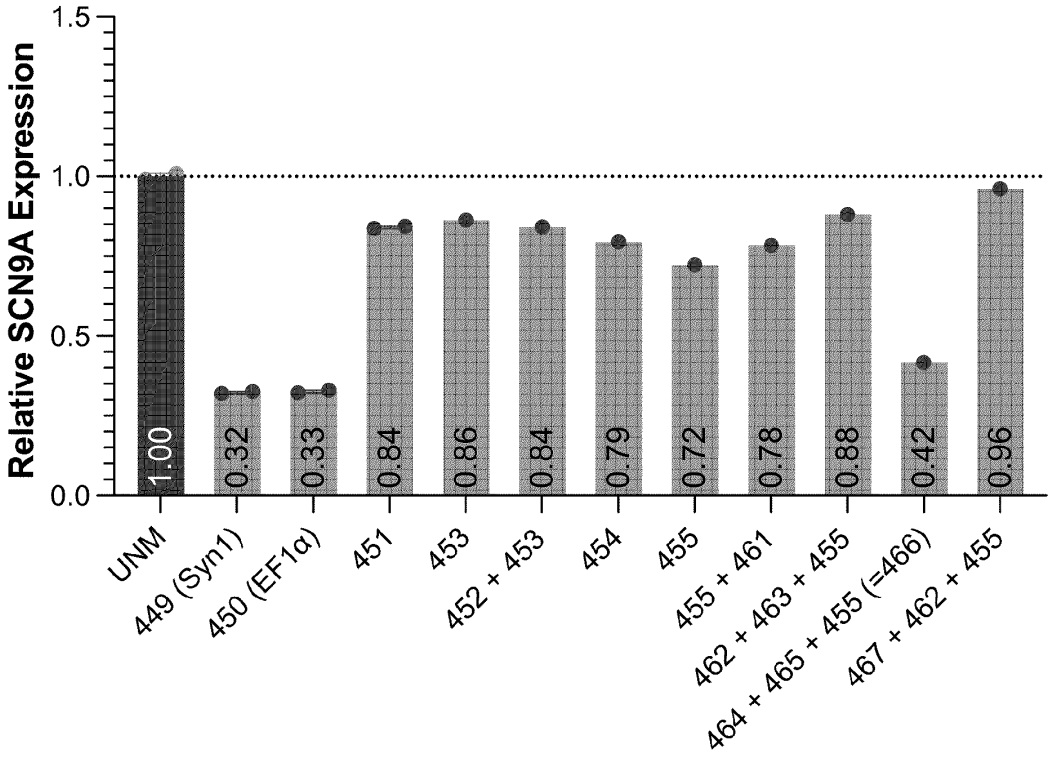

Relative SCN9A Expression
1.5
1.0
0.5
0.0
1.00
0.32
0.33
0.84
0.86
0.84
0.79
0.72
0.78
0.88
0.42
0.96
UNM
449 (Syn1)
450 (EF1α)
451
453
452 + 453
454
455
455 + 461
462 + 463 + 455
464 + 465 + 455 (=466)
467 + 462 + 455

FIG. 13

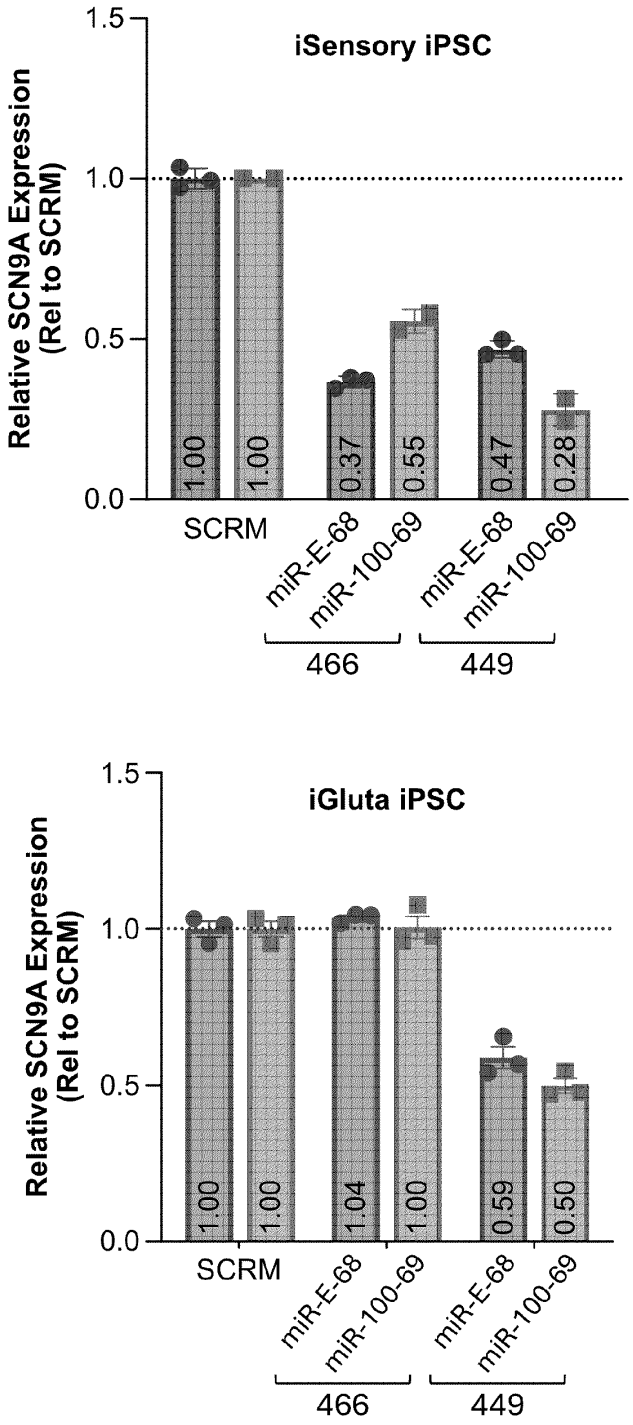

iSensory iPSC
Relative SCN9A Expression (Rel to SCRM)
1.5
1.0
0.5
0.0
1.00
1.00
0.37
0.55
0.47
0.28
SCRM
miR-E-68
miR-100-69
miR-E-68
miR-100-69
466
449
iGluta iPSC
Relative SCN9A Expression (Rel to SCRM)
1.5
1.0
0.5
0.0
1.00
1.00
1.04
1.00
0.59
0.50
SCRM
miR-E-68
miR-100-69
miR-E-68
miR-100-69
466
449

FIG. 14

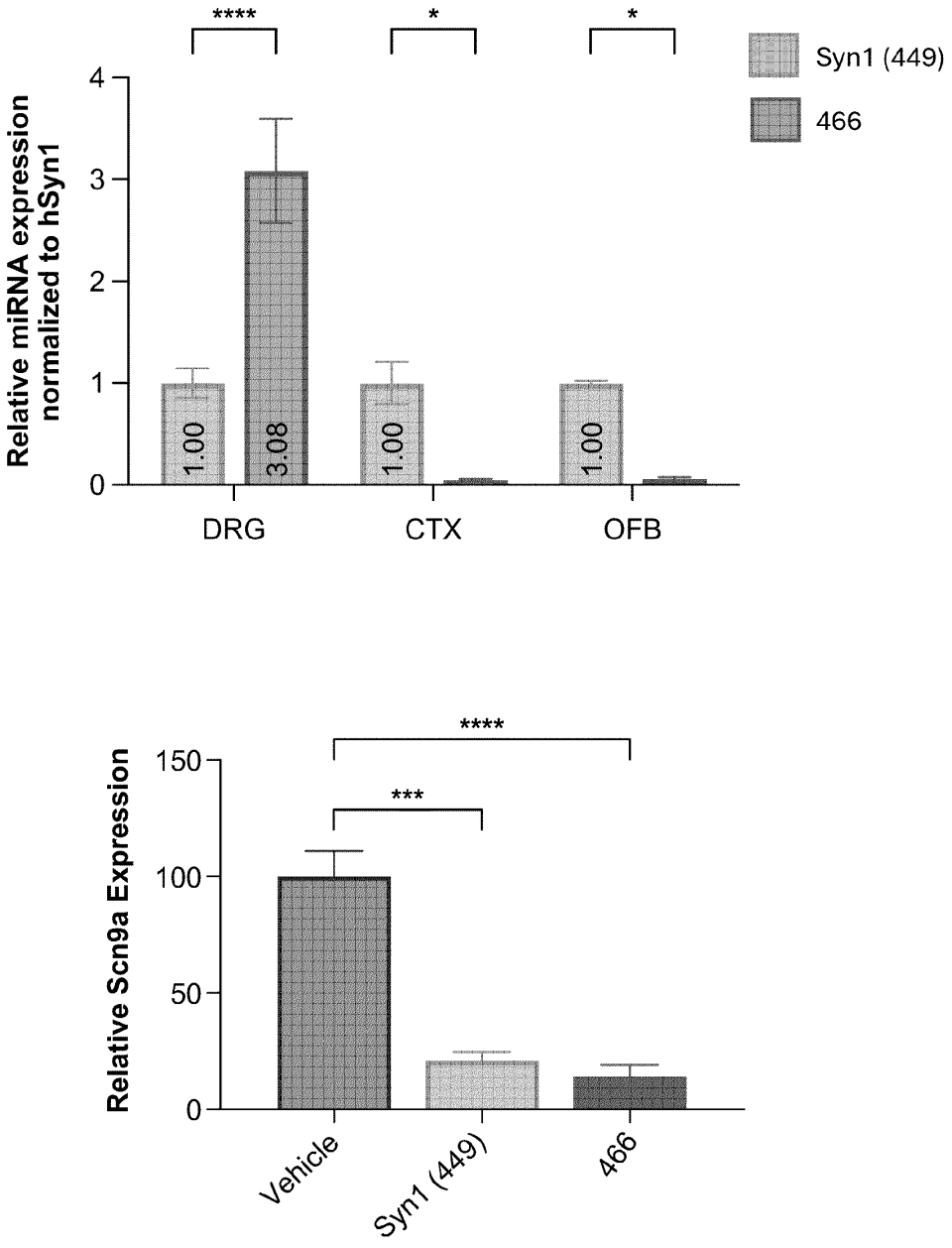

****
*
*
Syn1 (449)
466
Relative miRNA expression normalized to hSyn1
4
3
2
1
0
1.00
3.08
1.00
1.00
DRG
CTX
OFB
****
***
Relative Scn9a Expression
150
100
50
0
Vehicle
Syn1 (449)
466

FIG. 15

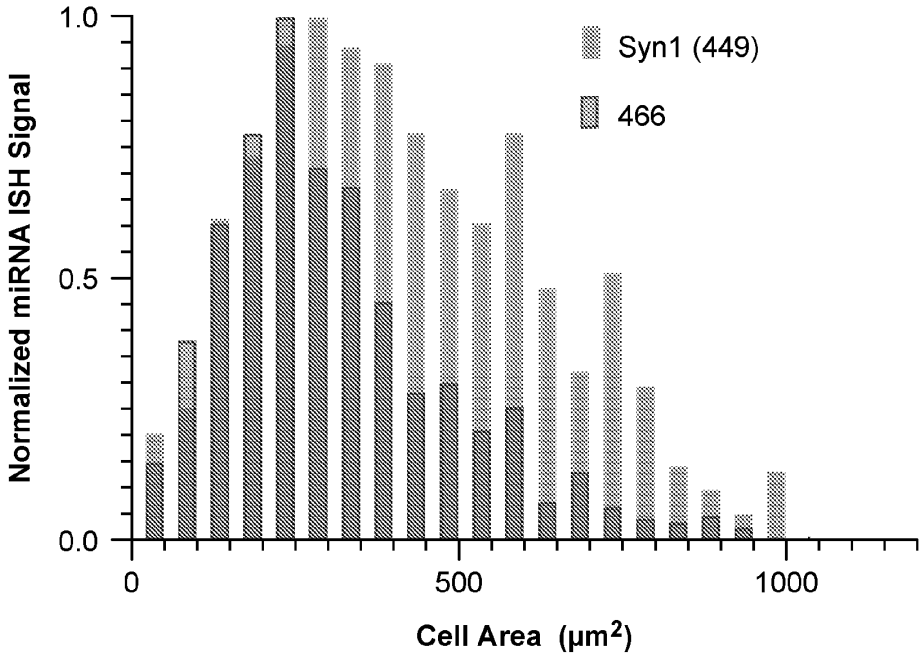

1.0
0.5
0.0
Normalized miRNA ISH Signal
Syn1 (449)
466
0
500
1000
Cell Area (μm2)

FIG. 16

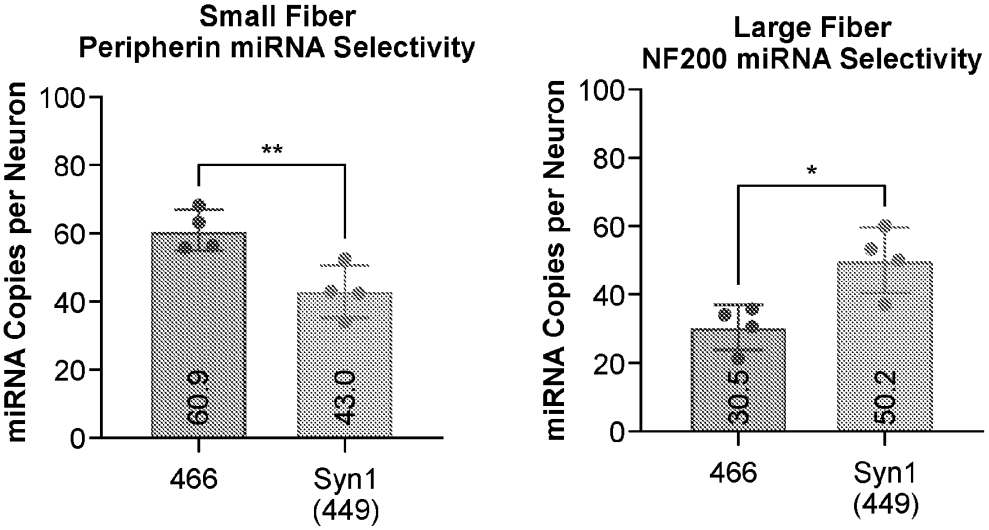

Small Fiber
Peripherin miRNA Selectivity
miRNA Copies per Neuron
100
80
60
40
20
0
**
60.9
43.0
466
Syn1
(449)
Large Fiber
NF200 miRNA Selectivity
miRNA Copies per Neuron
100
80
60
40
20
0
*
30.5
50.2
466
Syn1
(449)

FIG. 17

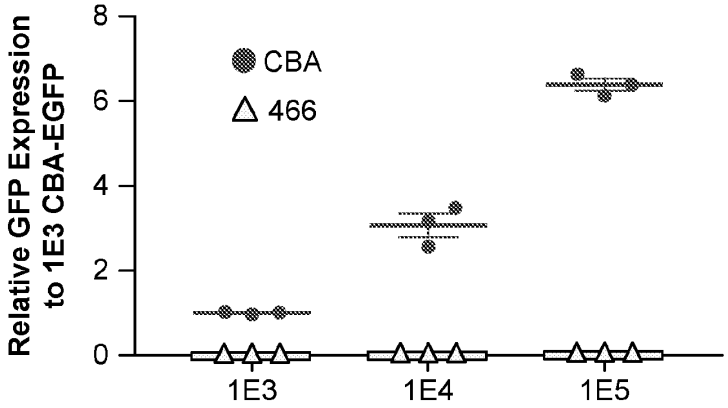

Relative GFP Expression
to 1E3 CBA-EGFP
8
6
4
2
0
CBA
466
1E3
1E4
1E5

FIG. 18