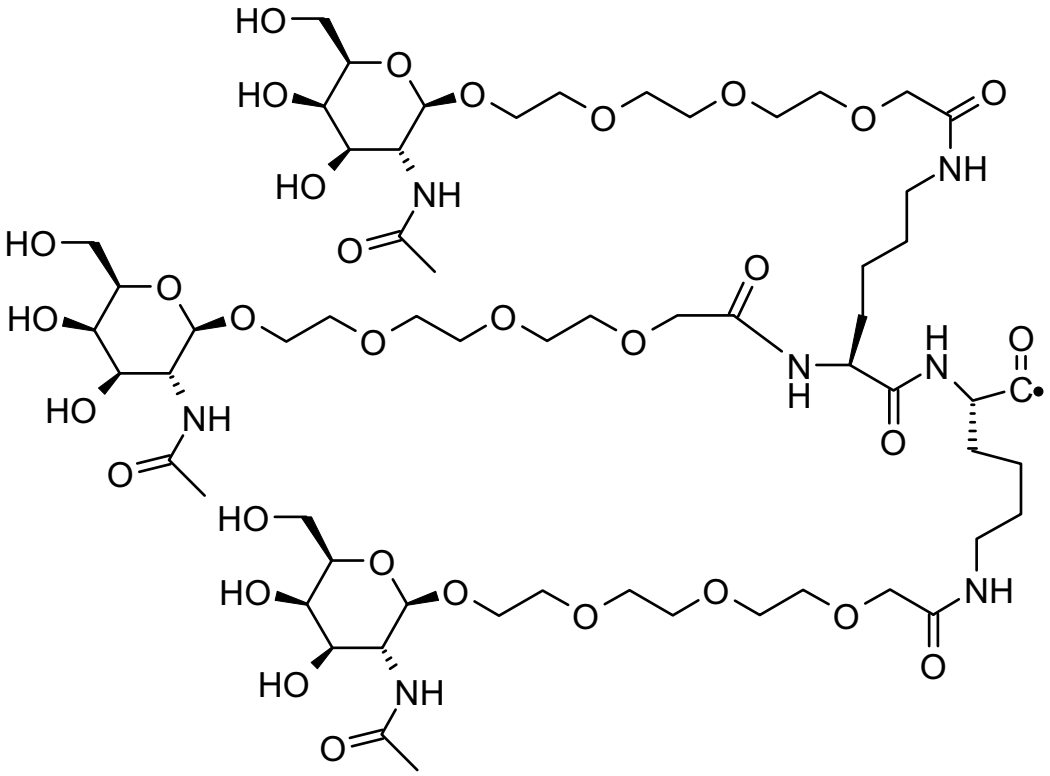

HO
O
HO
O
HO
NH
O
O
O
O
O
O
NH
HO
O
HO
O
HO
NH
O
O
O
O
O
N
H
O
H
N
O
O
C•
HO
O
HO
O
HO
NH
O
O
O
O
O
NH
O

FIG. 1

FIG. 2

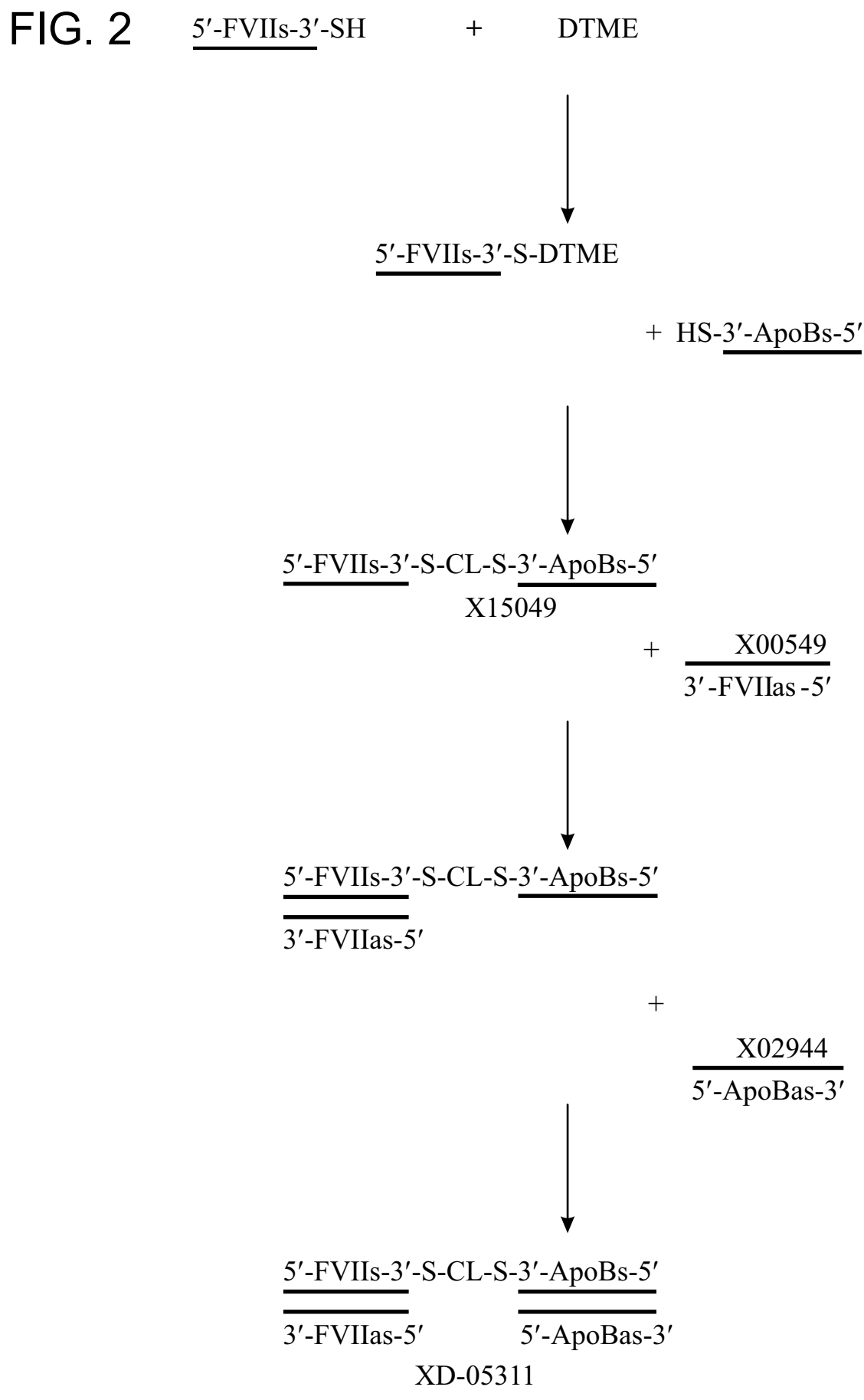

5′-FVIIs-3′-SH + DTME
5′-FVIIs-3′-S-DTME
+ HS-3′-ApoBs-5′
5′-FVIIs-3′-S-CL-S-3′-ApoBs-5′
X15049
+ X00549
3′-FVIIas-5′
5′-FVIIs-3′-S-CL-S-3′-ApoBs-5′
3′-FVIIas-5′
+
X02944
5′-ApoBas-3′
5′-FVIIs-3′-S-CL-S-3′-ApoBs-5′
3′-FVIIas-5′ 5′-ApoBas-3′
XD-05311

FIG. 3

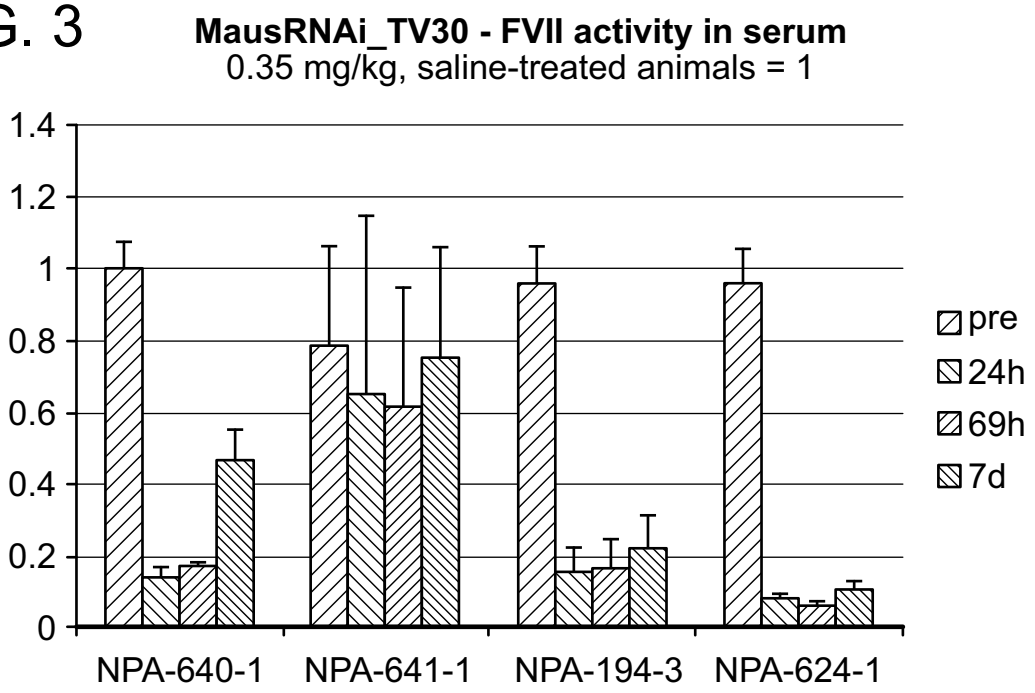


FIG. 4

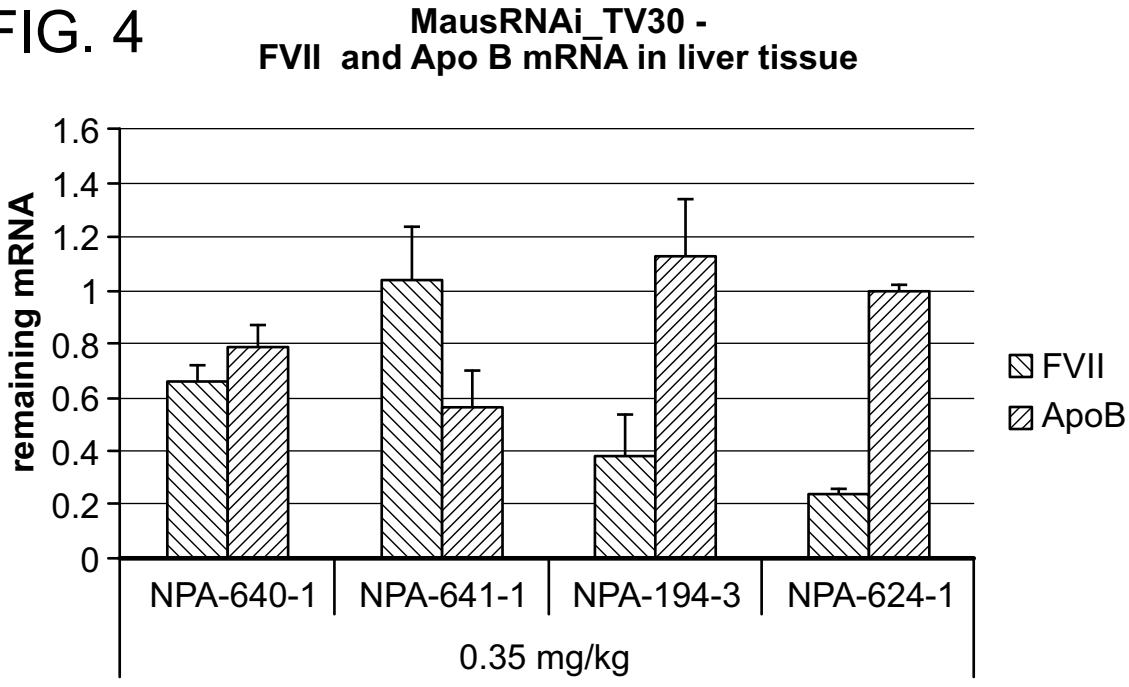


FIG. 5

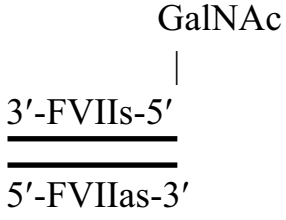


FIG. 6

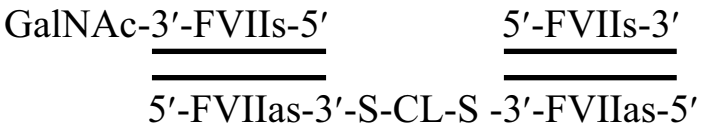

GalNAc-3′-FVIIs-5′
5′-FVIIs-3′
5′-FVIIas-3′-S-CL-S -3′-FVIIas-5′

FIG. 7

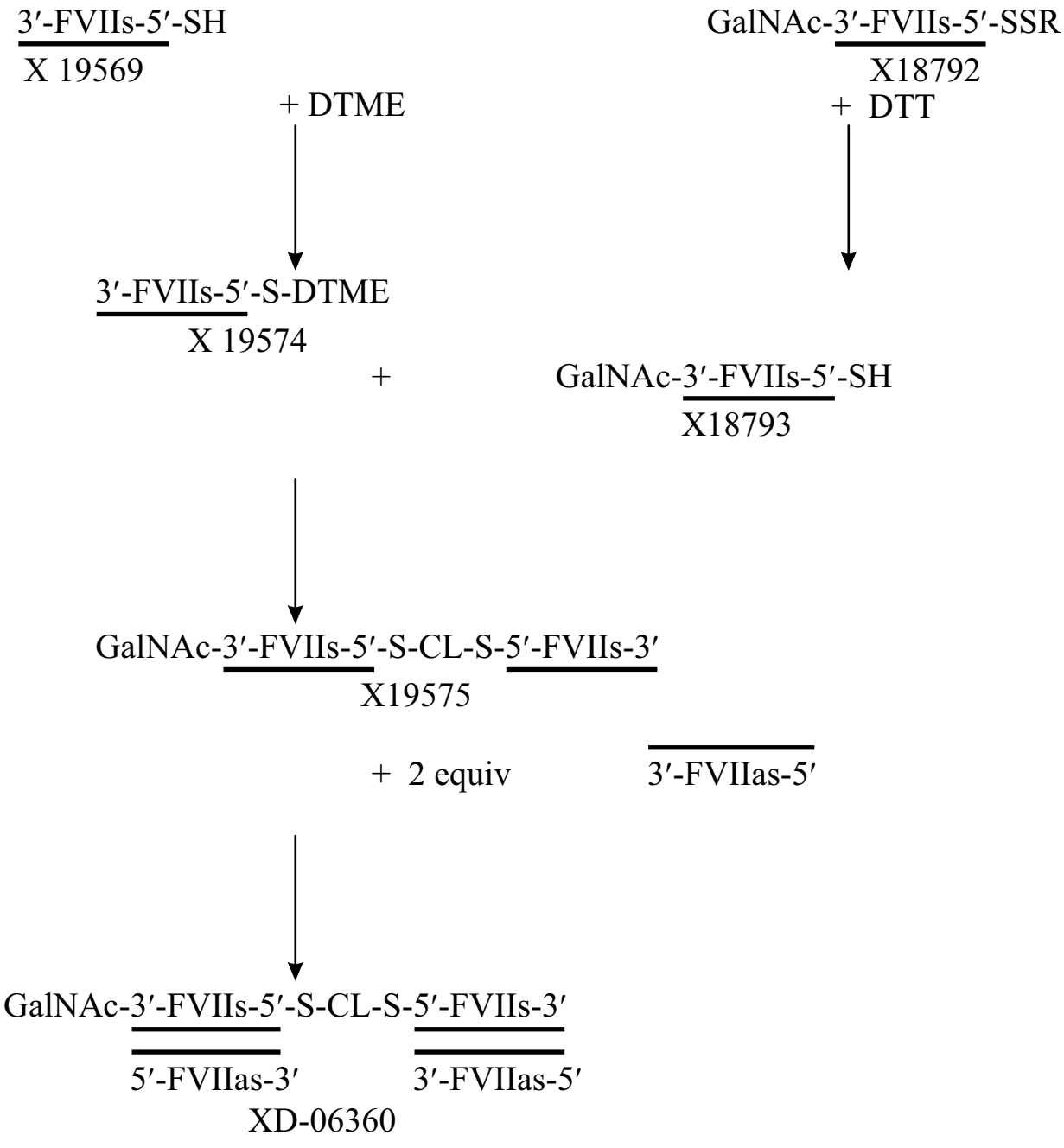

3′-FVIIs-5′-SH
X 19569
+ DTME
GalNAc-3′-FVIIs-5′-SSR
X18792
+ DTT
3′-FVIIs-5′-S-DTME
X 19574
+
GalNAc-3′-FVIIs-5′-SH
X18793
GalNAc-3′-FVIIs-5′-S-CL-S-5′-FVIIs-3′
X19575
+ 2 equiv
3′-FVIIas-5′
GalNAc-3′-FVIIs-5′-S-CL-S-5′-FVIIs-3′
5′-FVIIas-3′
3′-FVIIas-5′
XD-06360

FIG. 8

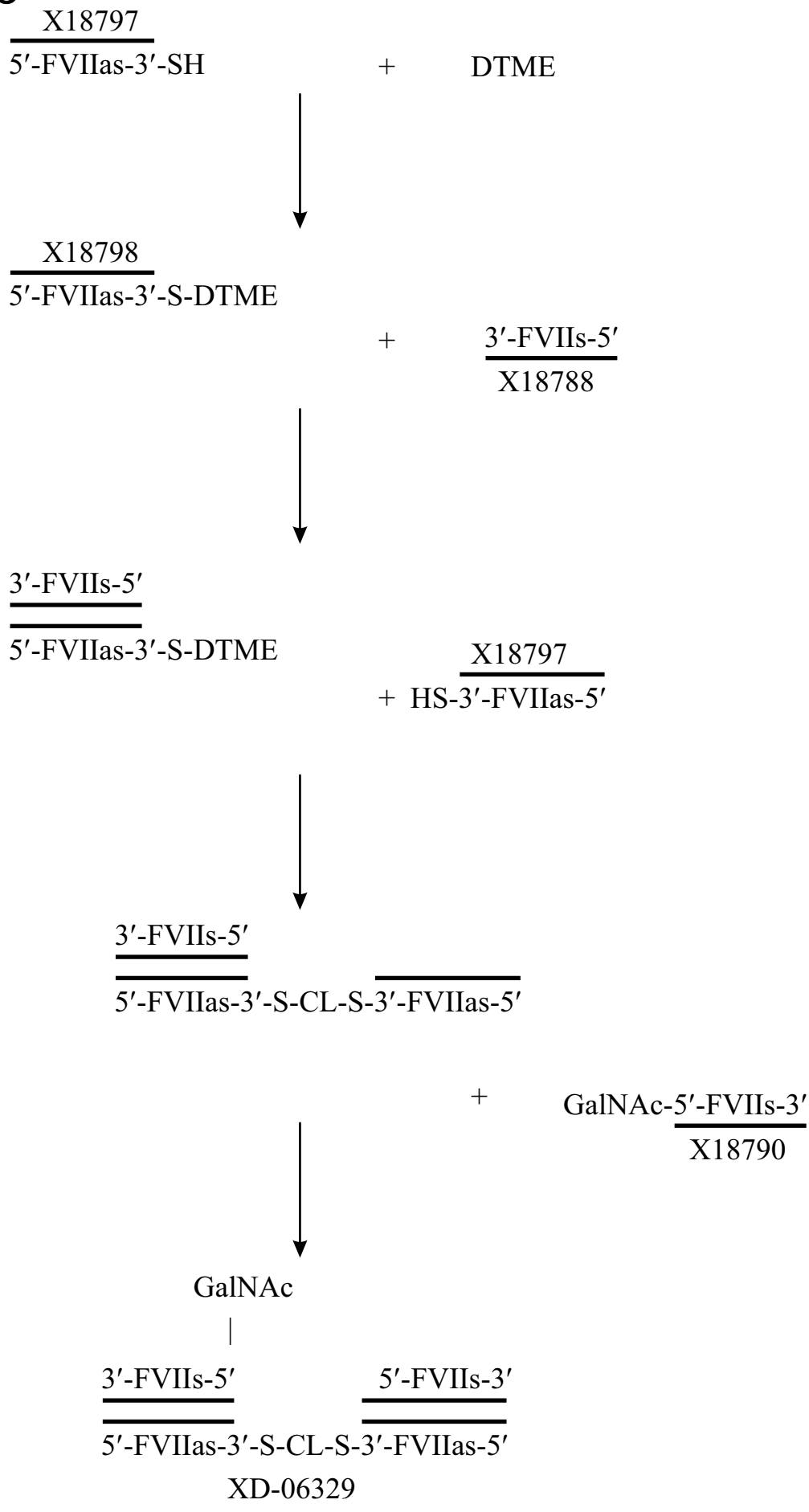

X18797
5′-FVIIas-3′-SH
+ DTME
X18798
5′-FVIIas-3′-S-DTME
+ 3′-FVIIs-5′
X18788
3′-FVIIs-5′
5′-FVIIas-3′-S-DTME
X18797
+ HS-3′-FVIIas-5′
3′-FVIIs-5′
5′-FVIIas-3′-S-CL-S-3′-FVIIas-5′
+ GalNAc-5′-FVIIs-3′
X18790
GalNAc
3′-FVIIs-5′
5′-FVIIs-3′
5′-FVIIas-3′-S-CL-S-3′-FVIIas-5′
XD-06329

FIG. 9

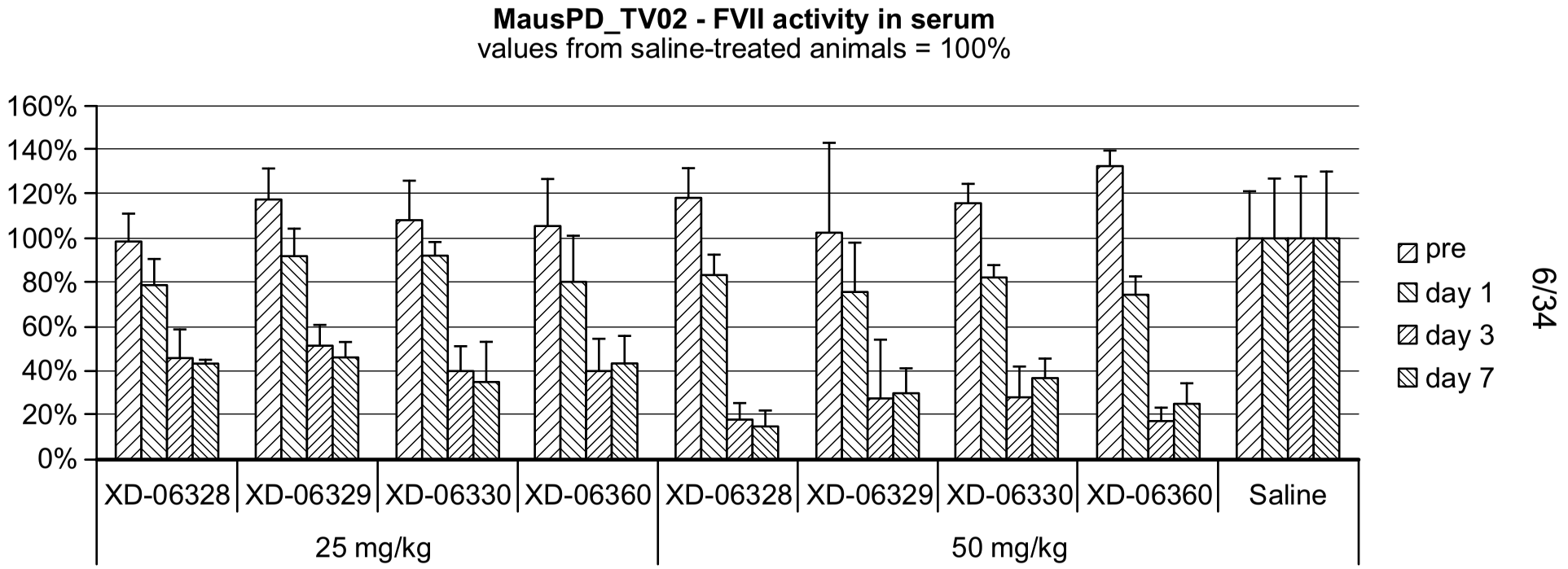

MausPD_TV02 - FVII activity in serum
values from saline-treated animals = 100%
160%
140%
120%
100%
80%
60%
40%
20%
0%
XD-06328
XD-06329
XD-06330
XD-06360
XD-06328
XD-06329
XD-06330
XD-06360
Saline
25 mg/kg
50 mg/kg
pre
day 1
day 3
day 7

FIG. 10A

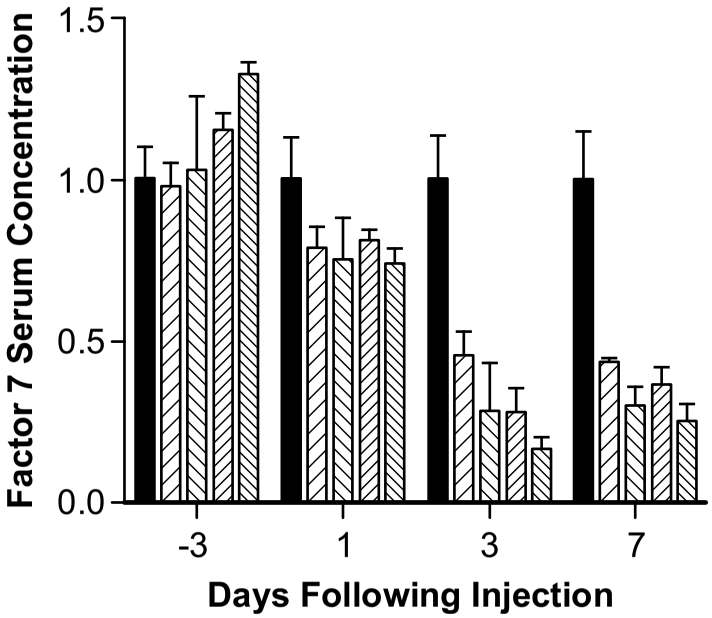


FIG. 10B

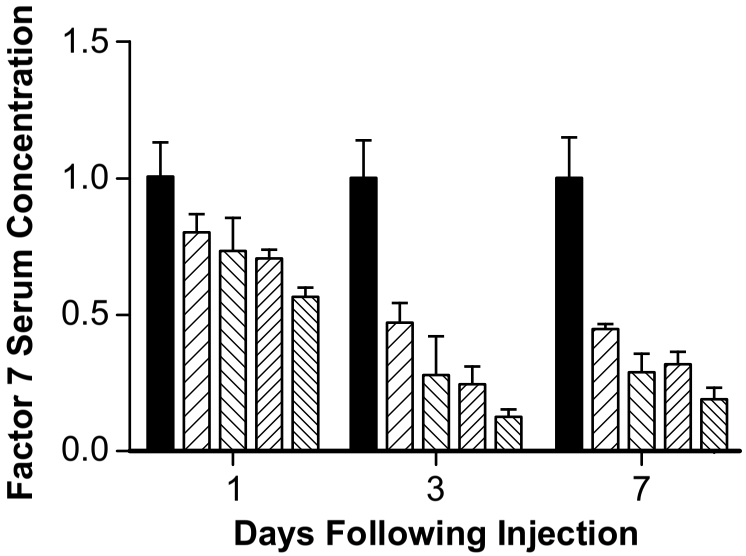
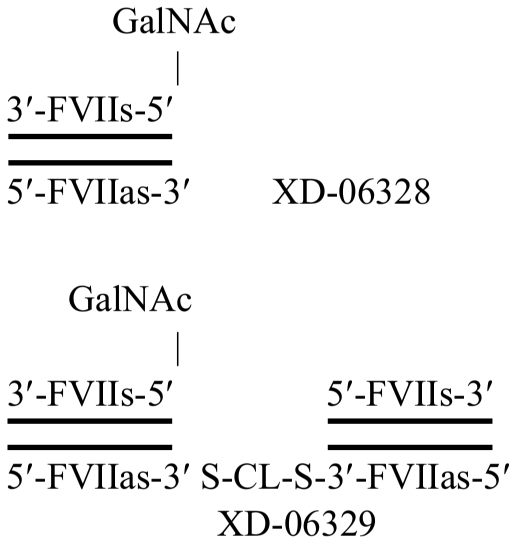
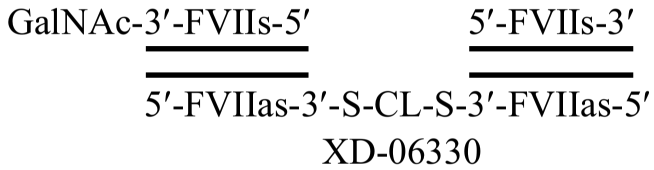


FIG. 10C





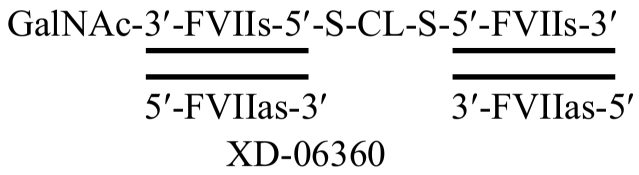
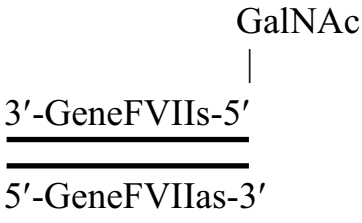
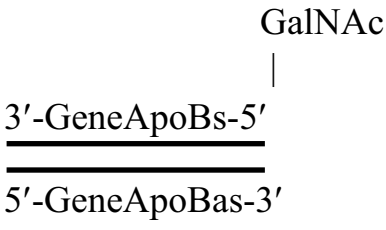


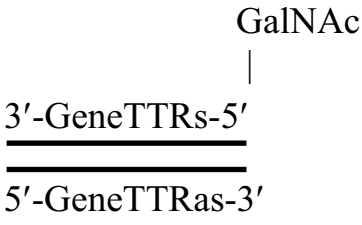
FIG. 11



(XD-06328)



(XD-06728)



(XD-06386)

FIG. 12

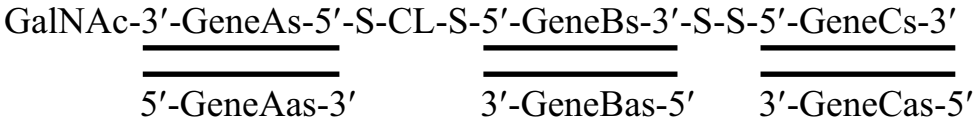


FIG. 13

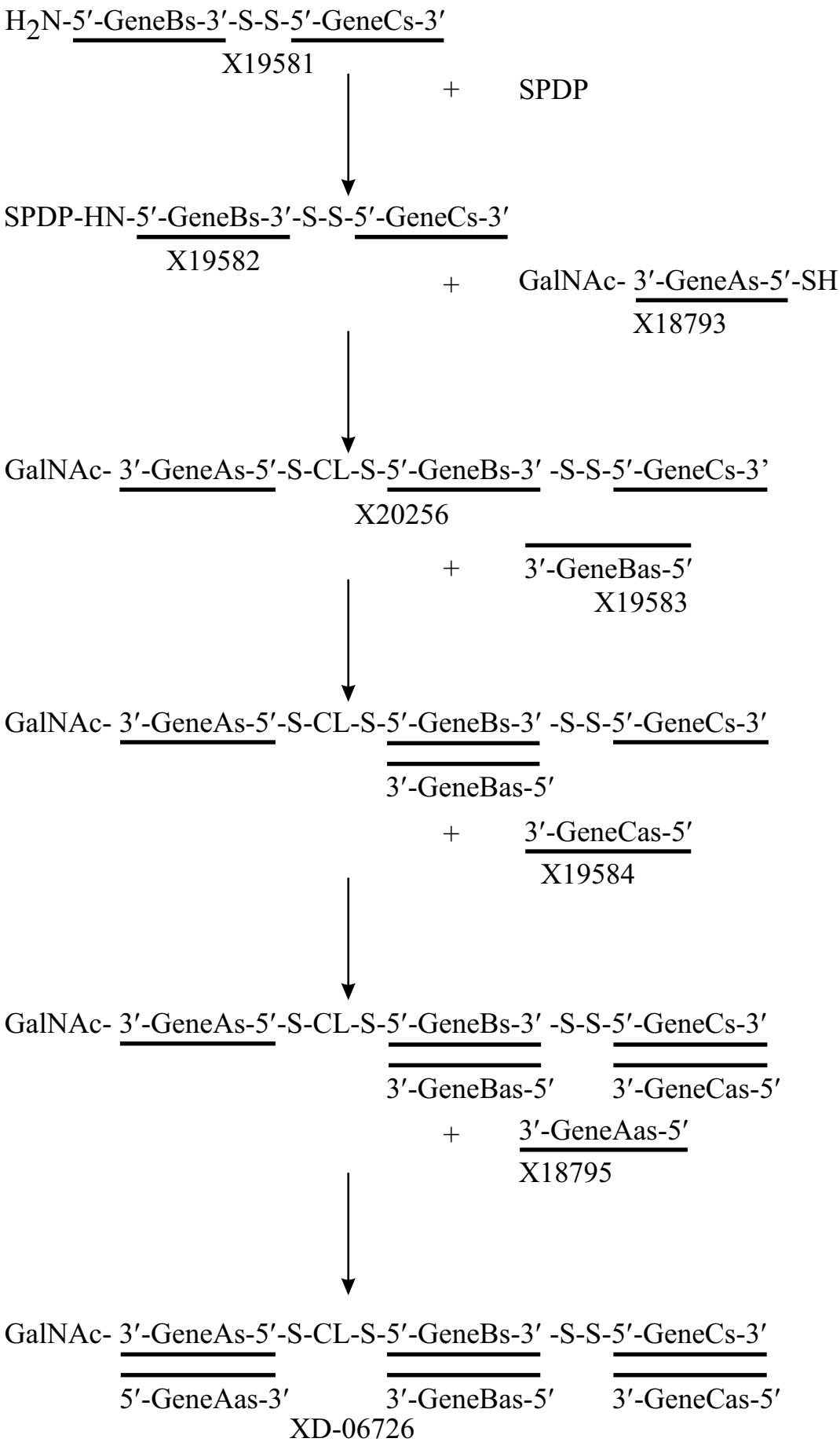

H2N-5′-GeneBs-3′-S-S-5′-GeneCs-3′
X19581
+ SPDP
SPDP-HN-5′-GeneBs-3′-S-S-5′-GeneCs-3′
X19582
+ GalNAc- 3′-GeneAs-5′-SH
X18793
GalNAc- 3′-GeneAs-5′-S-CL-S-5′-GeneBs-3′ -S-S-5′-GeneCs-3’
X20256
+ 3′-GeneBas-5′
X19583
GalNAc- 3′-GeneAs-5′-S-CL-S-5′-GeneBs-3′ -S-S-5′-GeneCs-3′
3′-GeneBas-5′
+ 3′-GeneCas-5′
X19584
GalNAc- 3′-GeneAs-5′-S-CL-S-5′-GeneBs-3′ -S-S-5′-GeneCs-3′
3′-GeneBas-5′ 3′-GeneCas-5′
+ 3′-GeneAas-5′
X18795
GalNAc- 3′-GeneAs-5′-S-CL-S-5′-GeneBs-3′ -S-S-5′-GeneCs-3′
5′-GeneAas-3′ 3′-GeneBas-5′ 3′-GeneCas-5′
XD-06726

FIG. 14

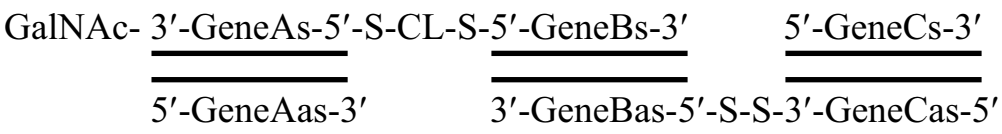

GalNAc- 3′-GeneAs-5′-S-CL-S-5′-GeneBs-3′ 5′-GeneCs-3′
5′-GeneAas-3′ 3′-GeneBas-5′-S-S-3′-GeneCas-5′

FIG. 15

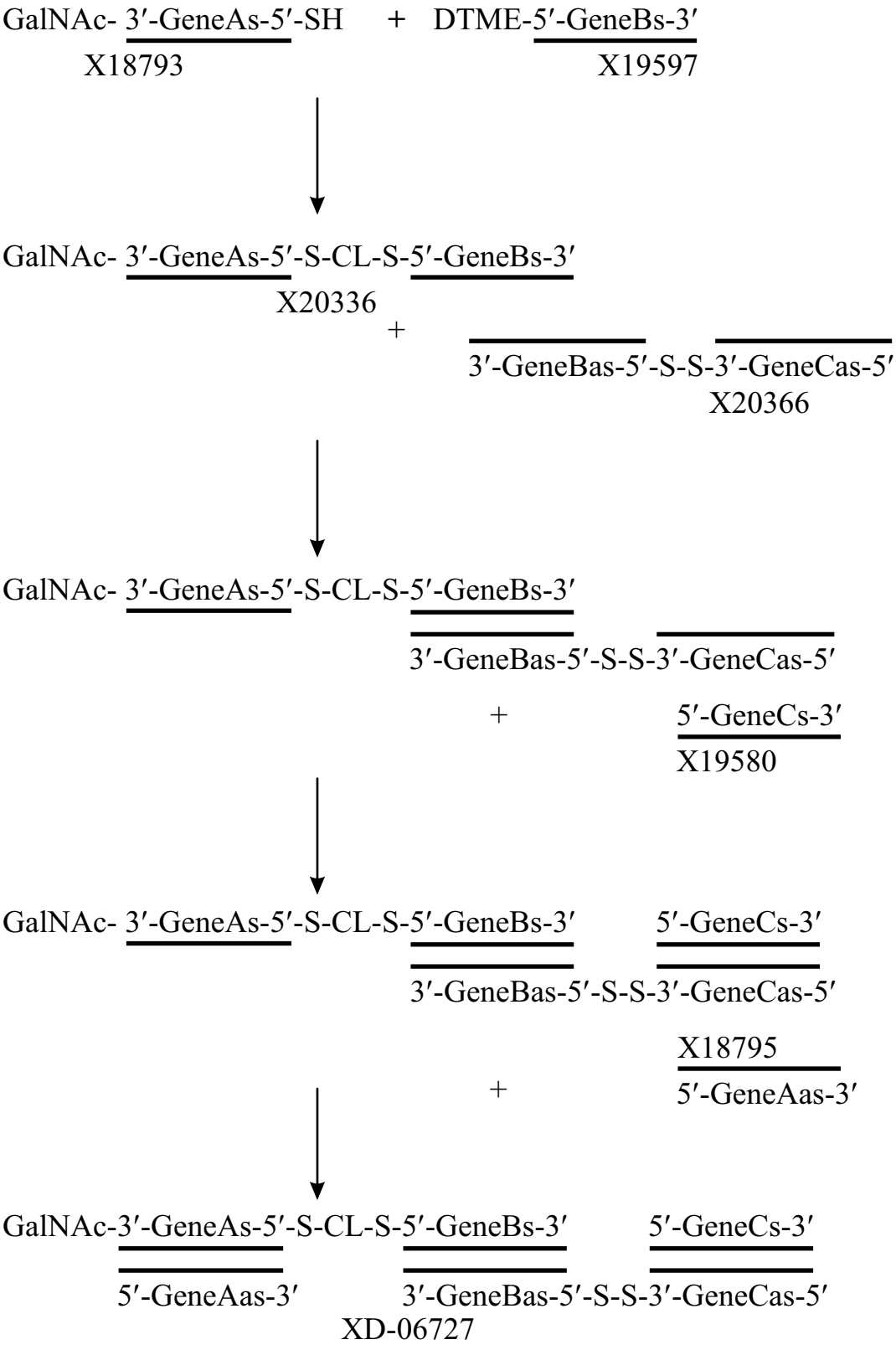

GalNAc- 3′-GeneAs-5′-SH + DTME-5′-GeneBs-3′
X18793 X19597
GalNAc- 3′-GeneAs-5′-S-CL-S-5′-GeneBs-3′
X20336
+
3′-GeneBas-5′-S-S-3′-GeneCas-5′
X20366
GalNAc- 3′-GeneAs-5′-S-CL-S-5′-GeneBs-3′
3′-GeneBas-5′-S-S-3′-GeneCas-5′
+ 5′-GeneCs-3′
X19580
GalNAc- 3′-GeneAs-5′-S-CL-S-5′-GeneBs-3′ 5′-GeneCs-3′
3′-GeneBas-5′-S-S-3′-GeneCas-5′
X18795
+ 5′-GeneAas-3′
GalNAc-3′-GeneAs-5′-S-CL-S-5′-GeneBs-3′ 5′-GeneCs-3′
5′-GeneAas-3′ 3′-GeneBas-5′-S-S-3′-GeneCas-5′
XD-06727

FIG. 16

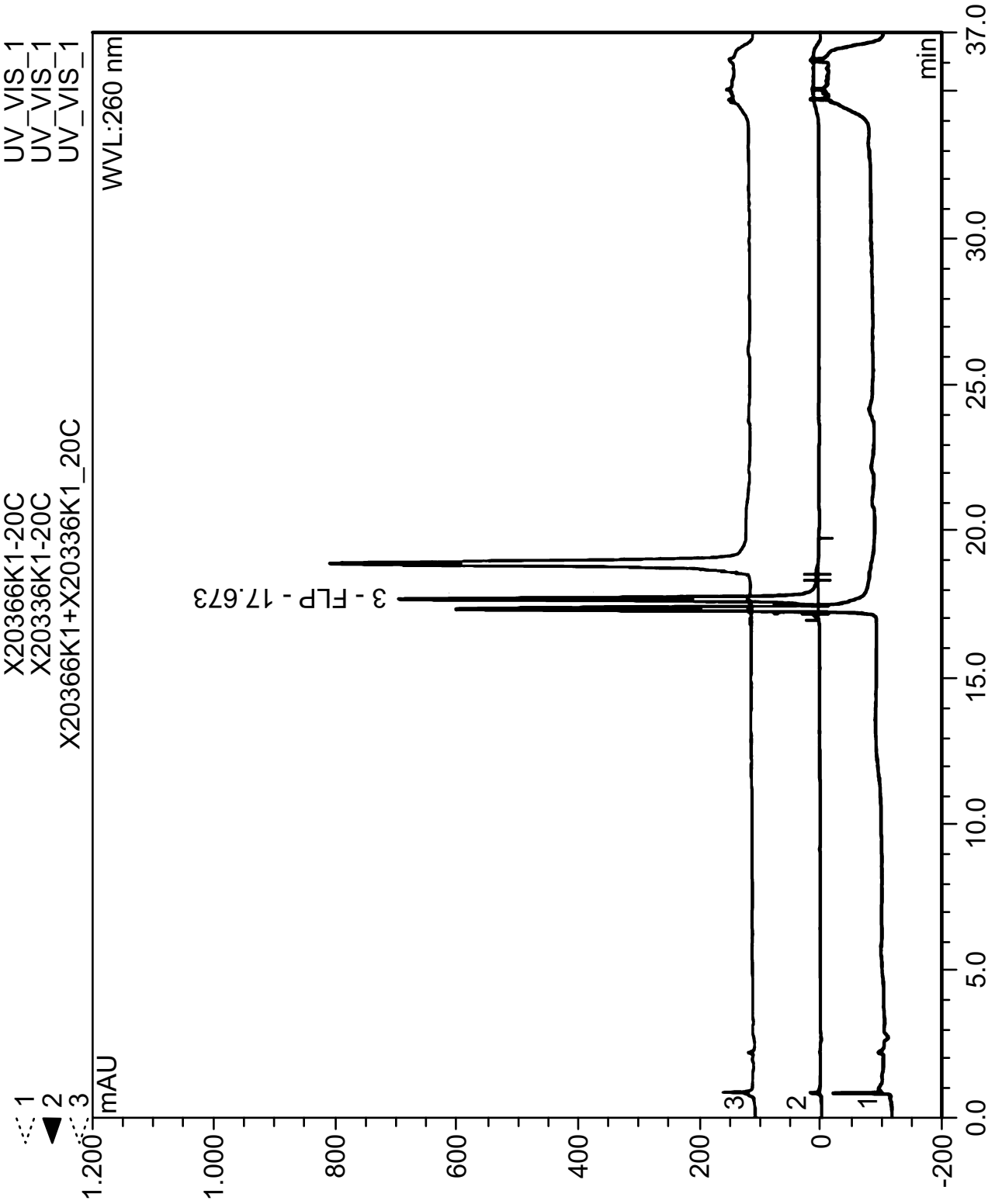

X20366K1-20C
X20336K1-20C
X20366K1+X20336K1_20C
UV_VIS_1
UV_VIS_1
UV_VIS_1
WVL:260 nm
mAU
min
3 - FLP - 17.673
1.200
1.000
800
600
400
200
0
-200
0.0
5.0
10.0
15.0
20.0
25.0
30.0
37.0

FIG. 17

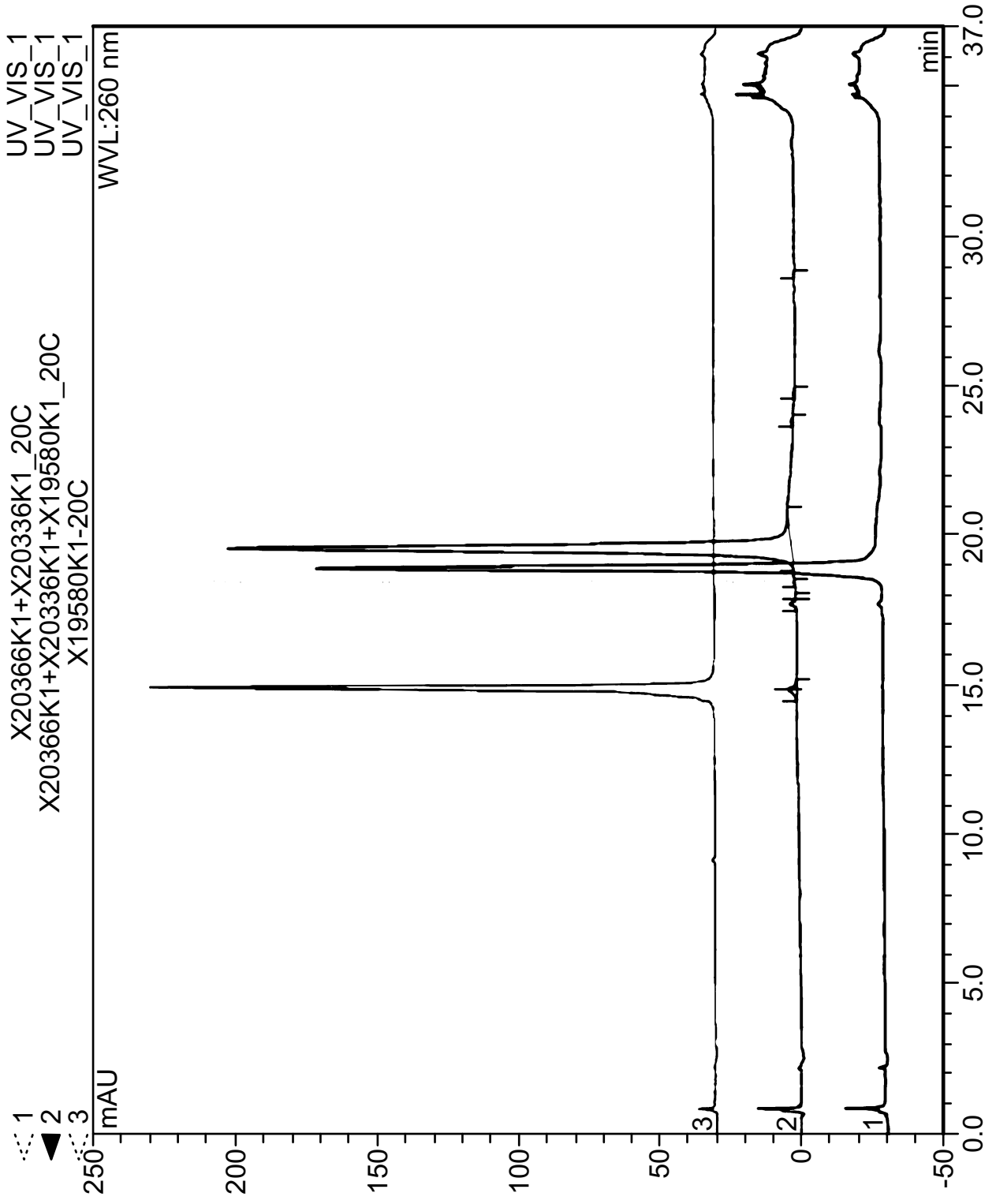

X20366K1+X20336K1_20C
X20366K1+X20336K1+X19580K1_20C
X19580K1-20C
UV_VIS_1
UV_VIS_1
UV_VIS_1
WVL:260 nm
mAU
min
250
200
150
100
50
0
-50
0.0
5.0
10.0
15.0
20.0
25.0
30.0
37.0
1
2
3

FIG. 18

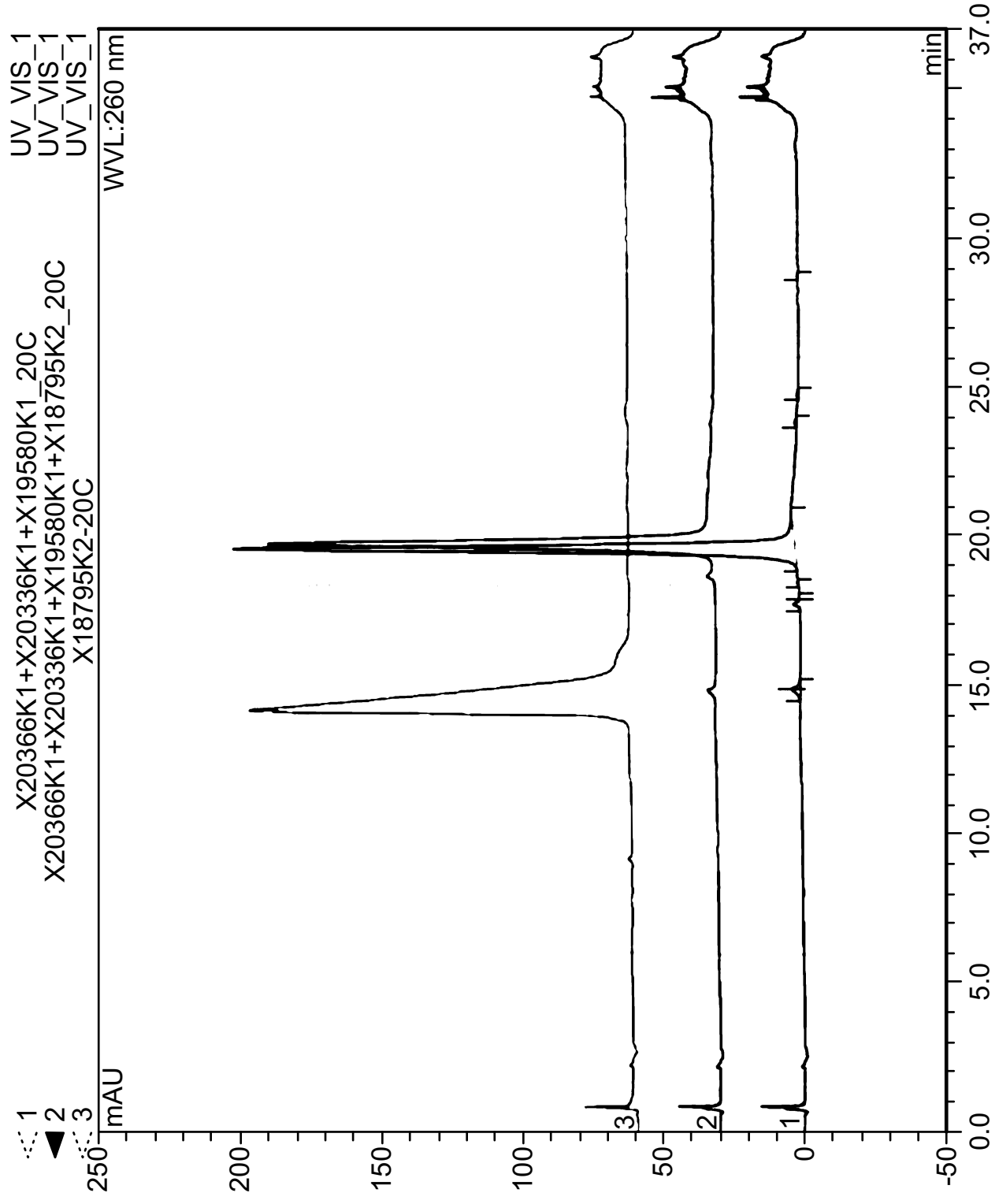

X20366K1+X20336K1+X19580K1_20C
X20366K1+X20336K1+X19580K1+X18795K2_20C
X18795K2-20C
UV_VIS_1
UV_VIS_1
UV_VIS_1
WVL:260 nm
mAU
min
250
200
150
100
50
0
-50
0.0
5.0
10.0
15.0
20.0
25.0
30.0
37.0
1
2
3

FIG. 19A

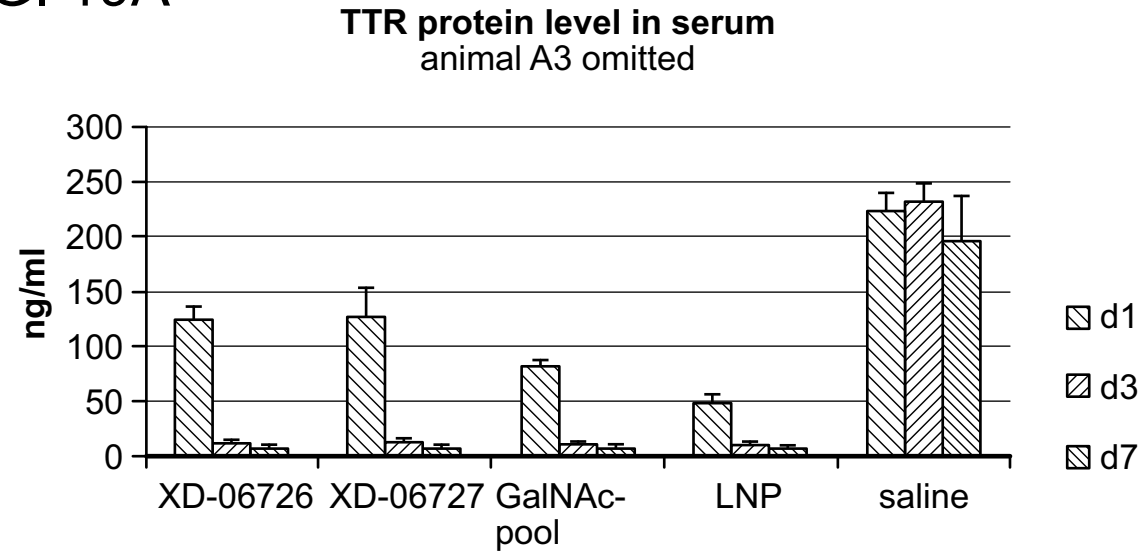


FIG. 19B

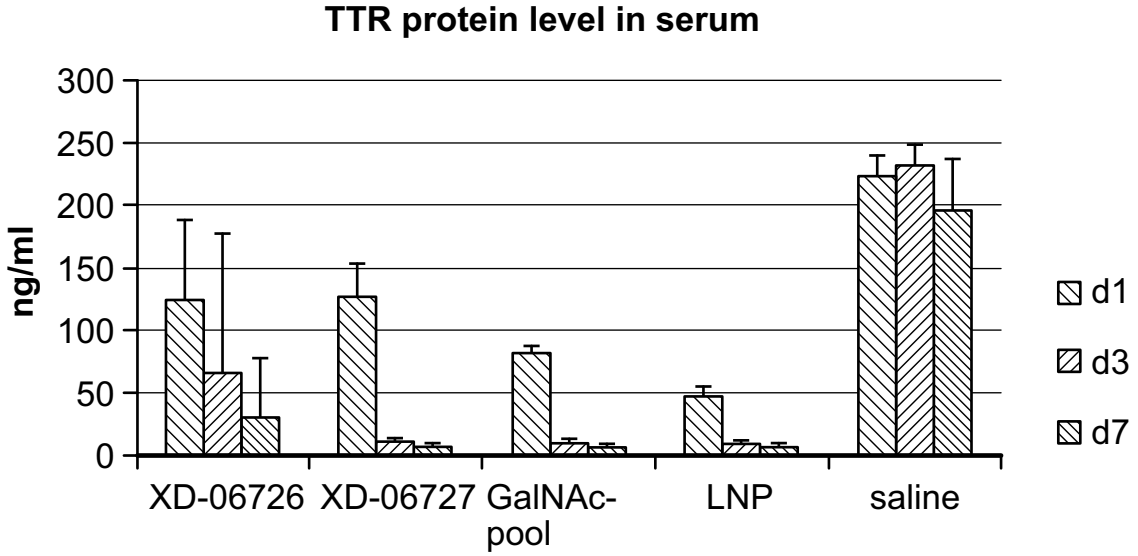


FIG. 20A

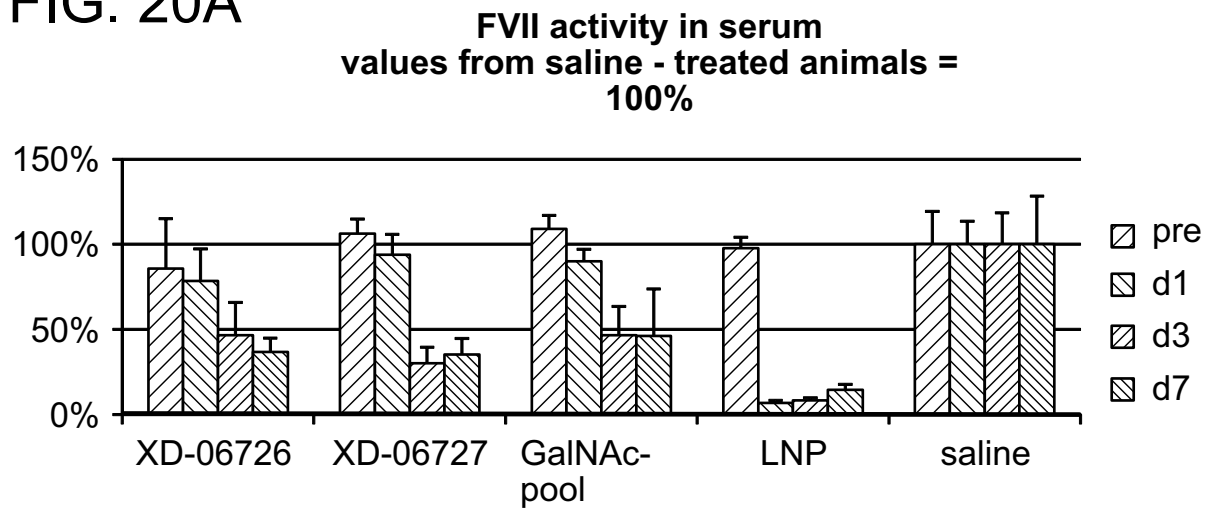


FIG. 20B

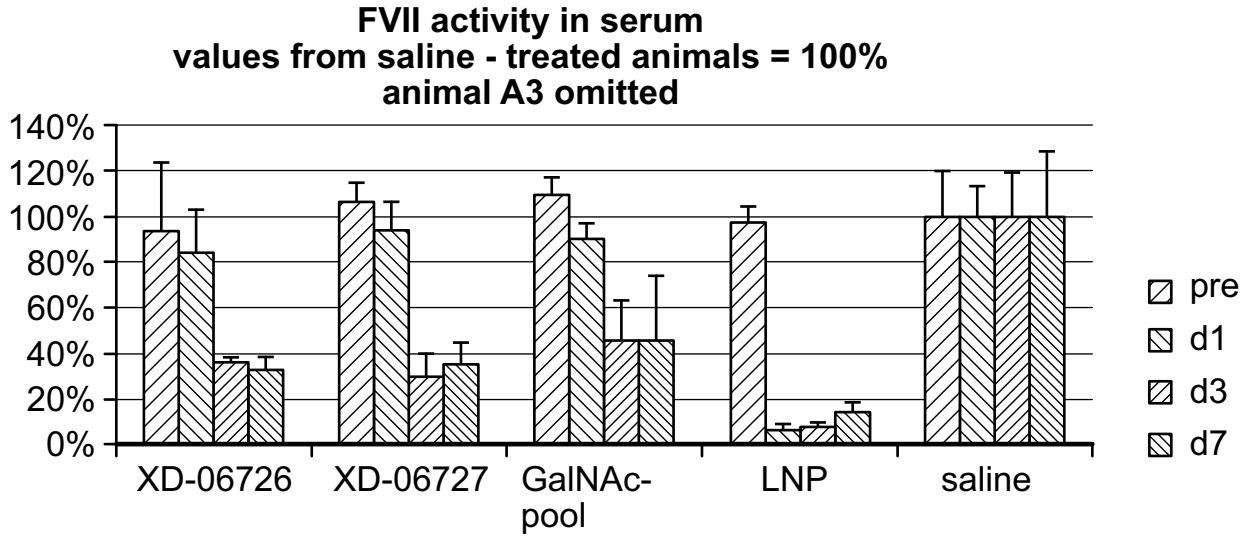


FIG. 21A

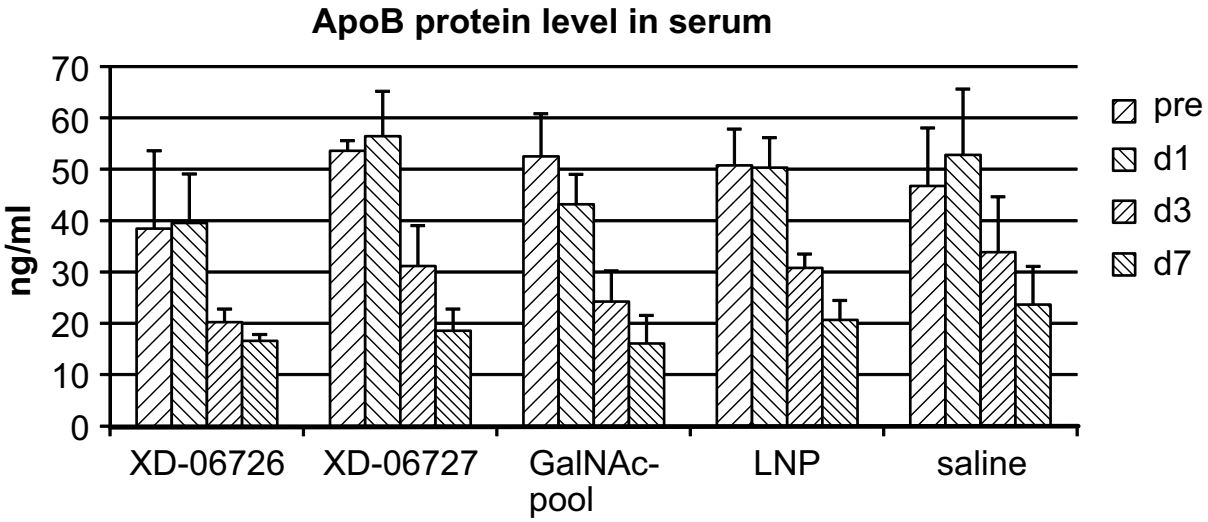


FIG. 21B

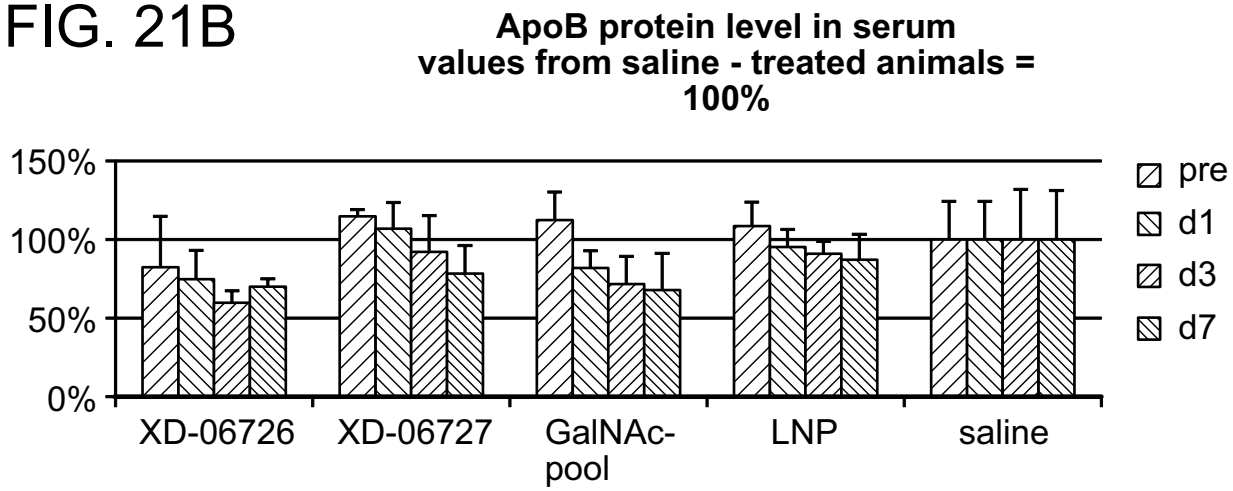


FIG. 22A

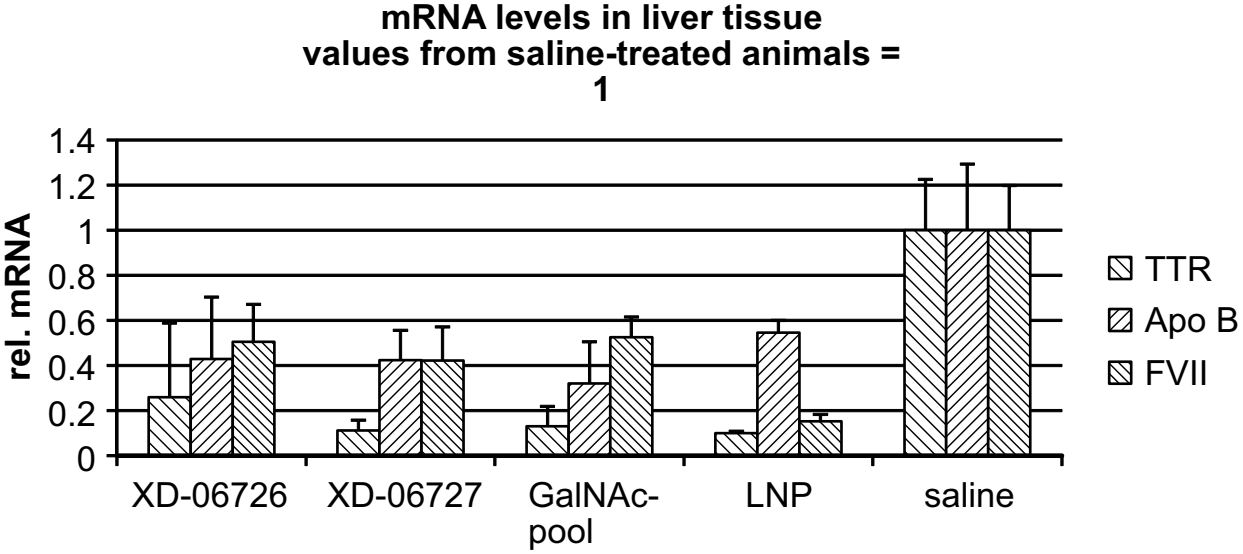


FIG. 22B

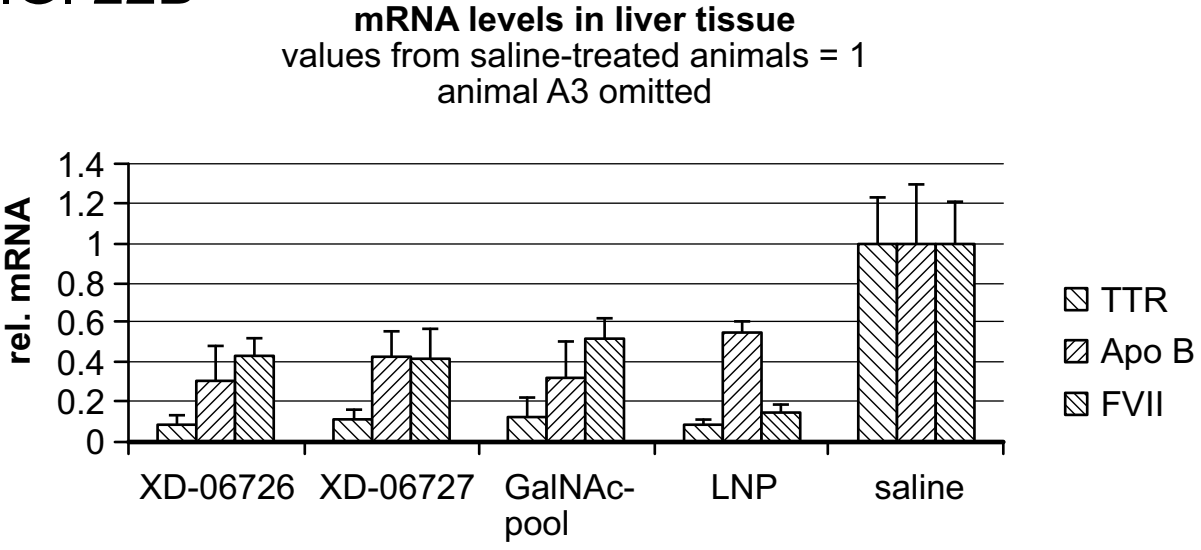


FIG. 23

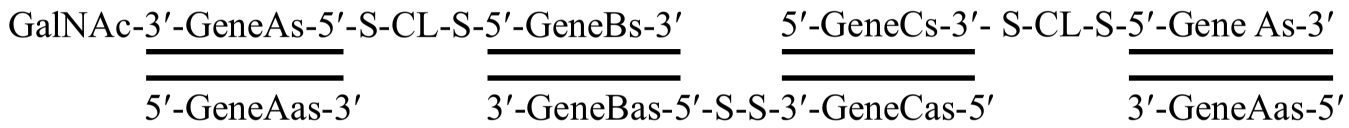

GalNAc-3′-GeneAs-5′-S-CL-S-5′-GeneBs-3′ 5′-GeneCs-3′- S-CL-S-5′-Gene As-3′
5′-GeneAas-3′ 3′-GeneBas-5′-S-S-3′-GeneCas-5′ 3′-GeneAas-5′

FIG. 24

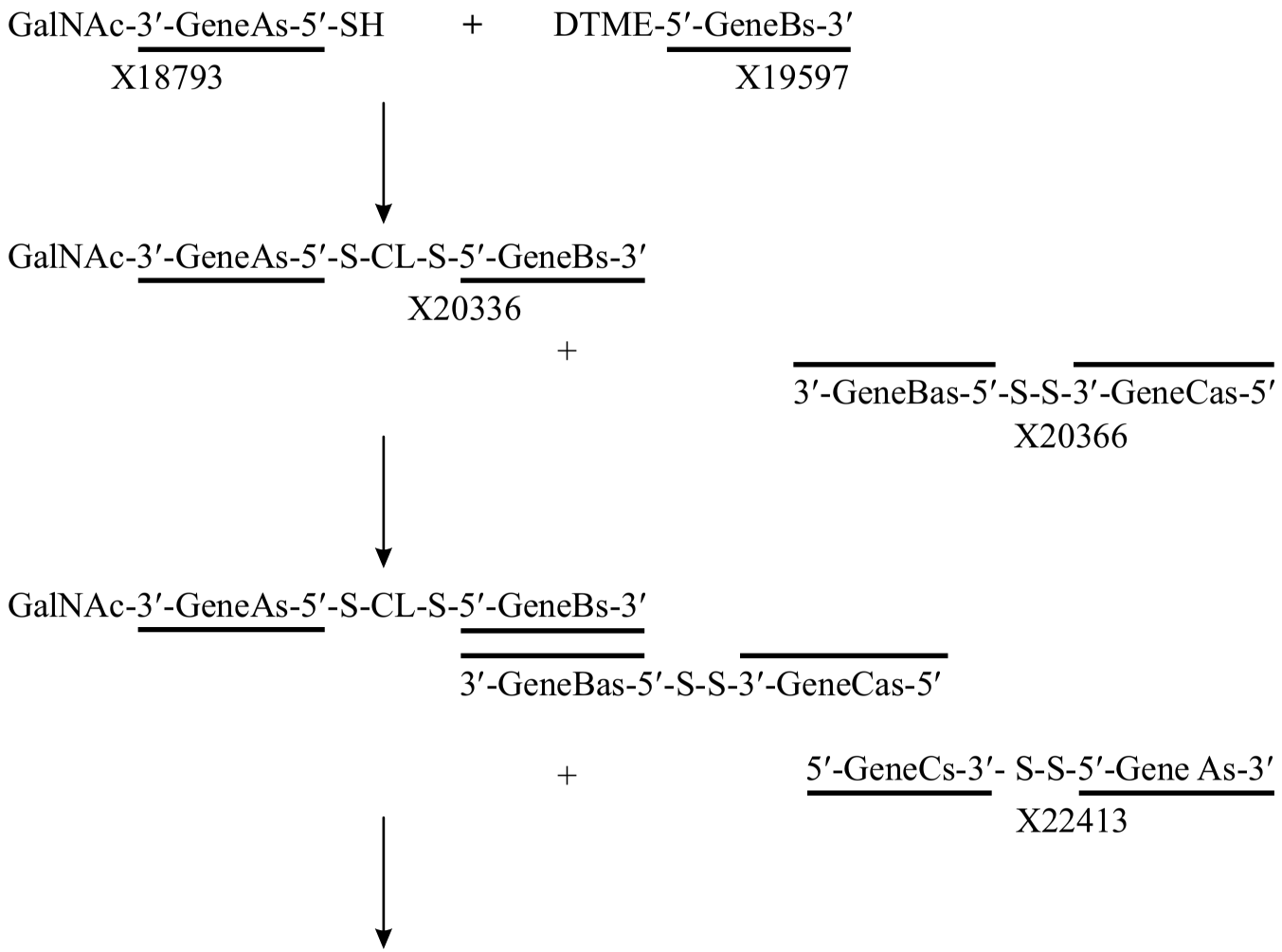
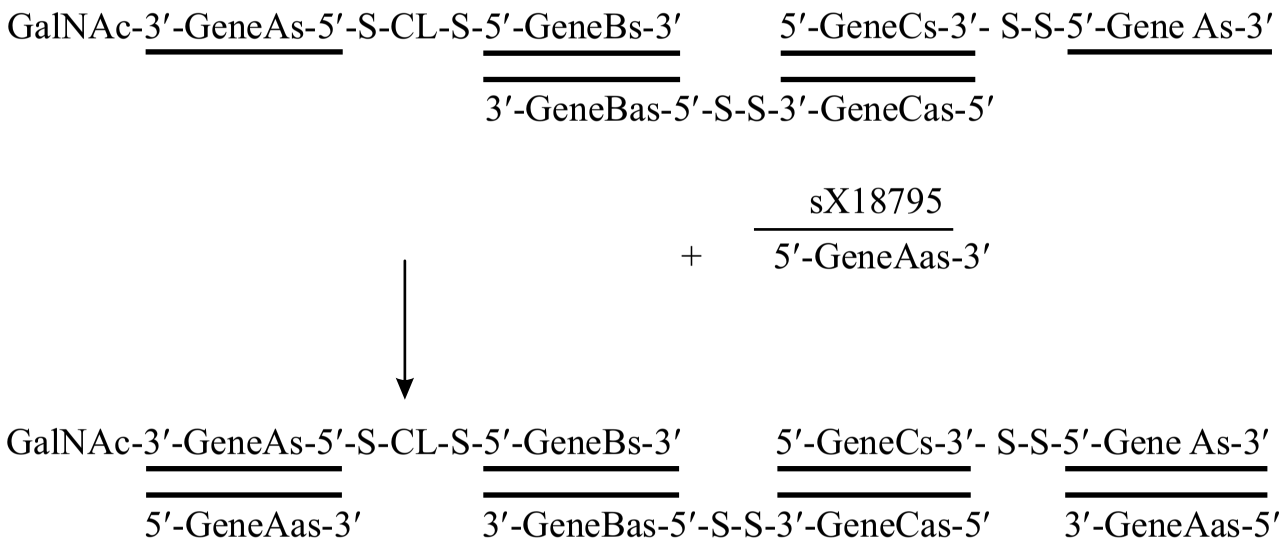

GalNAc-3′-GeneAs-5′-SH
X18793
+
DTME-5′-GeneBs-3′
X19597
GalNAc-3′-GeneAs-5′-S-CL-S-5′-GeneBs-3′
X20336
+
3′-GeneBas-5′-S-S-3′-GeneCas-5′
X20366
GalNAc-3′-GeneAs-5′-S-CL-S-5′-GeneBs-3′
3′-GeneBas-5′-S-S-3′-GeneCas-5′
+
5′-GeneCs-3′- S-S-5′-Gene As-3′
X22413

FIG. 24 (continued)



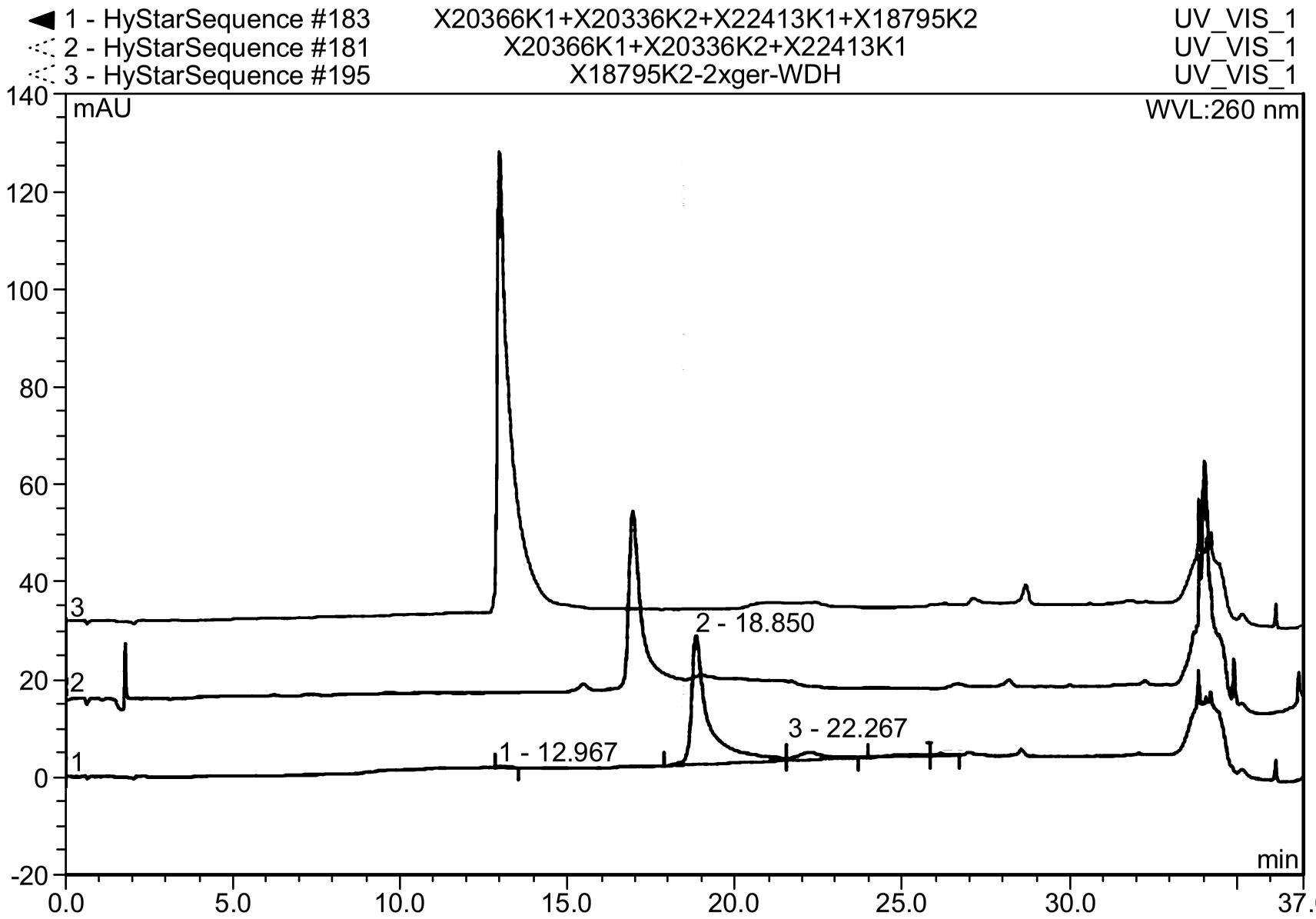

1 - HyStarSequence #183 X20366K1+X20336K2+X22413K1+X18795K2 UV_VIS_1
2 - HyStarSequence #181 X20366K1+X20336K2+X22413K1 UV_VIS_1
3 - HyStarSequence #195 X18795K2-2xger-WDH UV_VIS_1
mAU
WVL:260 nm
140
120
100
80
60
40
20
0
-20
3
2
1
2 - 18.850
3 - 22.267
1 - 12.967
min
0.0
5.0
10.0
15.0
20.0
25.0
30.0
37.0

FIG. 25

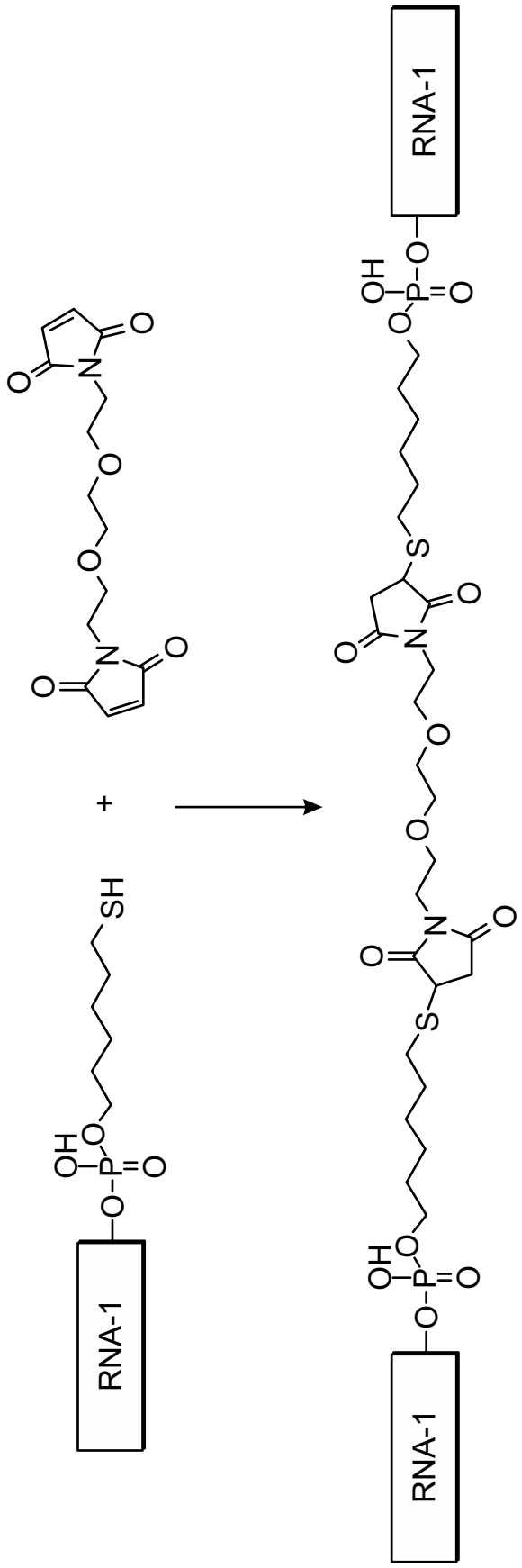


FIG. 26

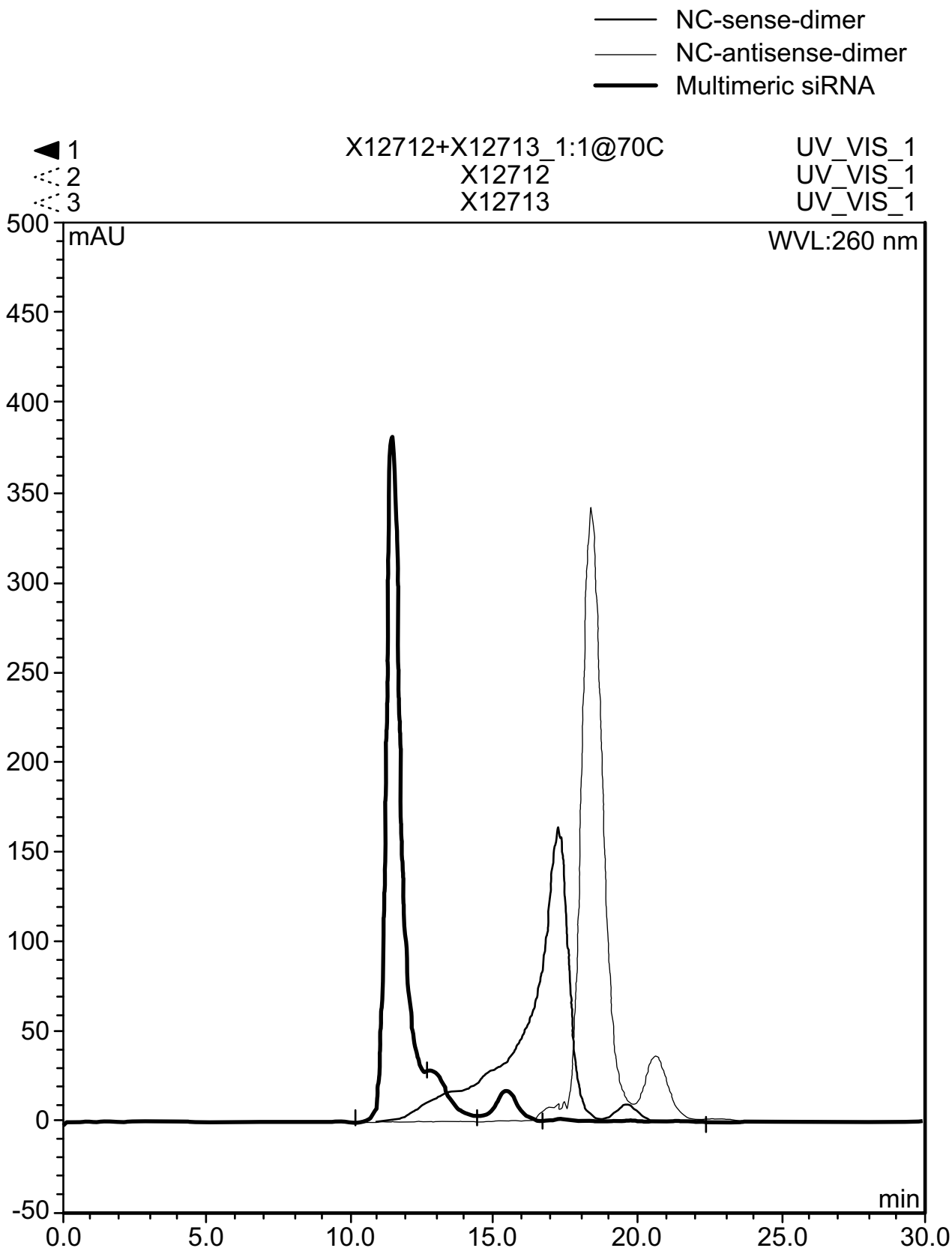

NC-sense-dimer
NC-antisense-dimer
Multimeric siRNA
1 X12712+X12713_1:1@70C UV_VIS_1
2 X12712 UV_VIS_1
3 X12713 UV_VIS_1
mAU
WVL:260 nm
min
500
450
400
350
300
250
200
150
100
50
0
-50
0.0
5.0
10.0
15.0
20.0
25.0
30.0

FIG. 27

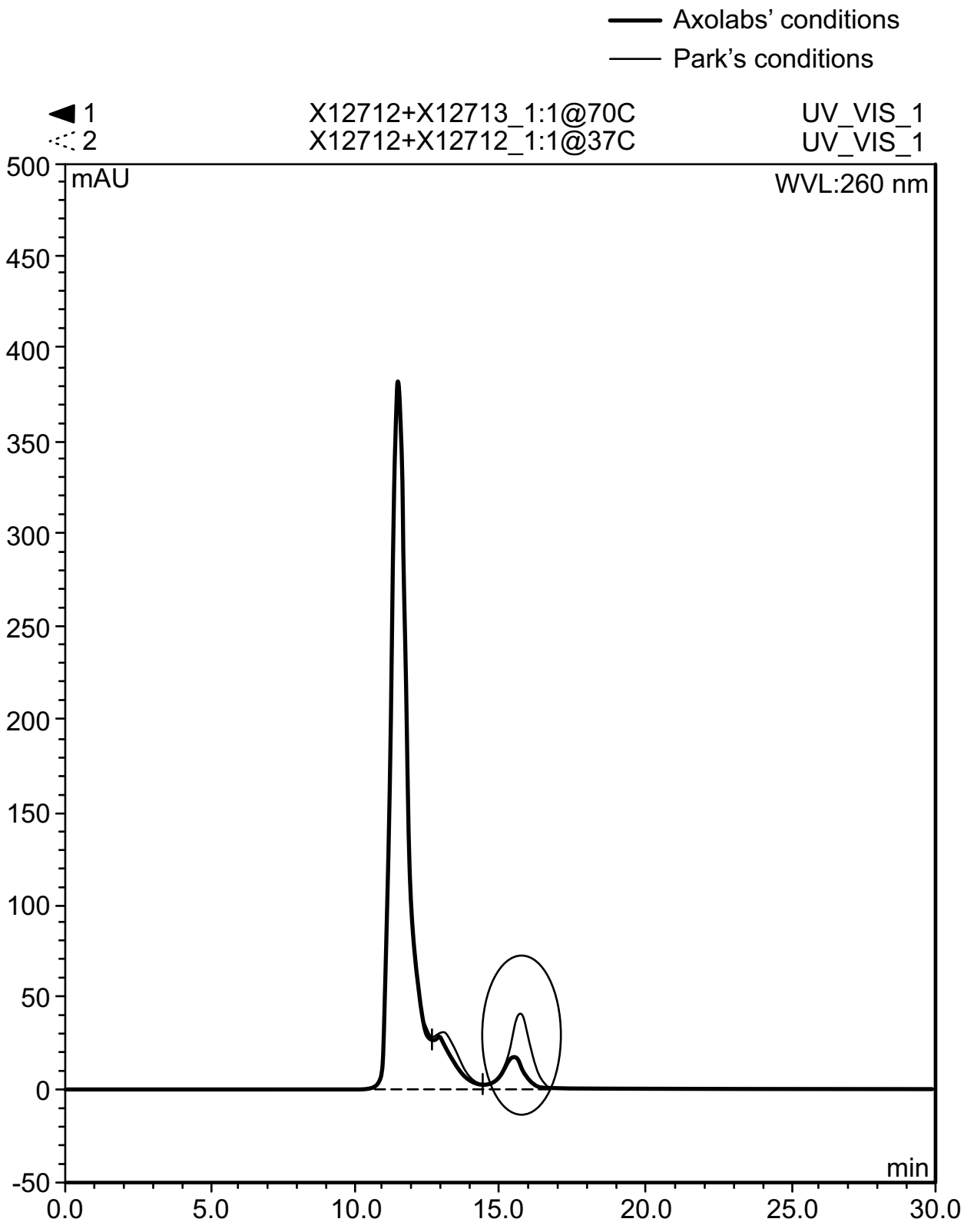

Axolabs' conditions
Park's conditions
1
X12712+X12713_1:1@70C
UV_VIS_1
2
X12712+X12712_1:1@37C
UV_VIS_1
mAU
WVL:260 nm
500
450
400
350
300
250
200
150
100
50
0
-50
min
0.0
5.0
10.0
15.0
20.0
25.0
30.0

FIG. 28

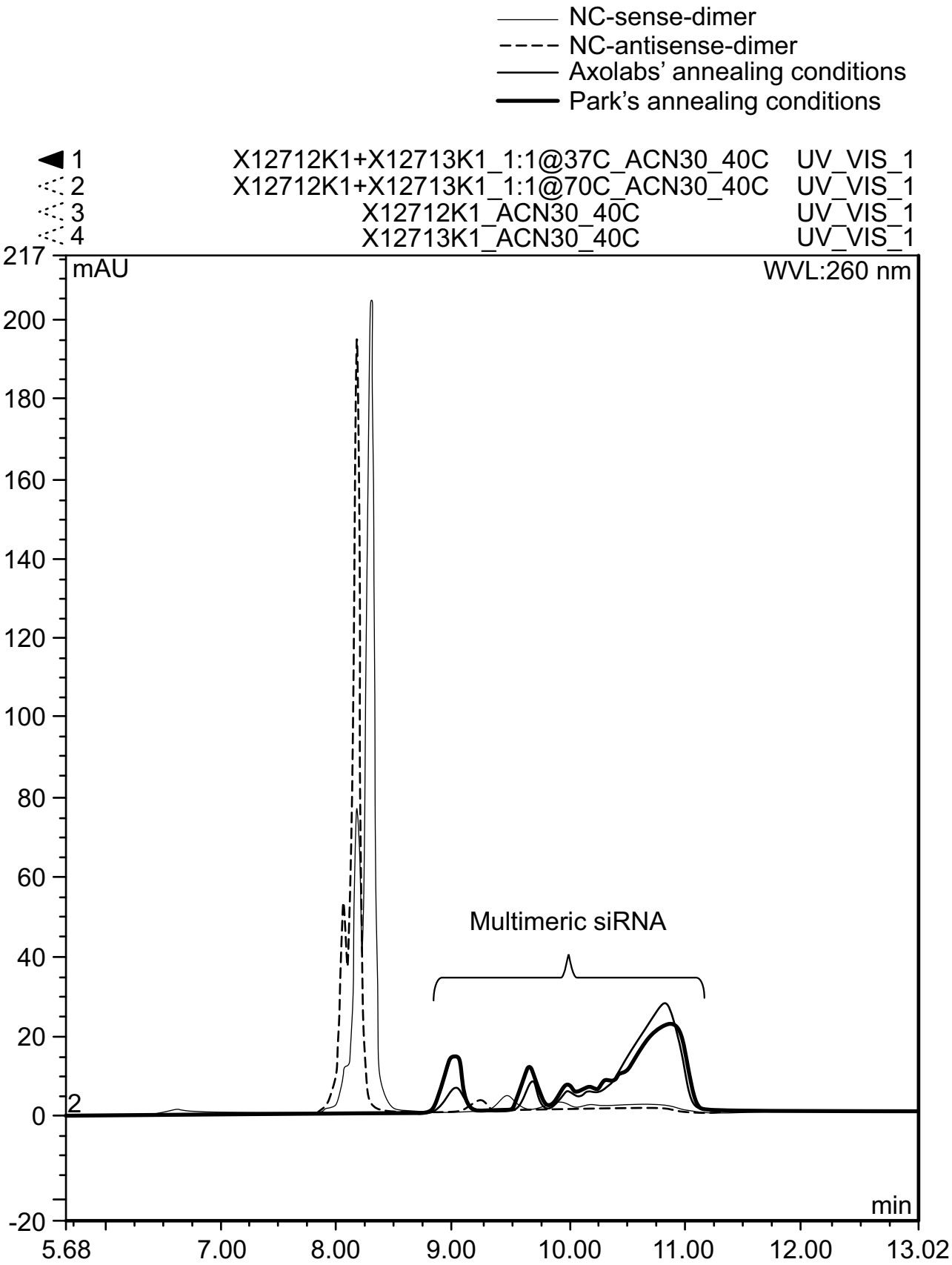

NC-sense-dimer
NC-antisense-dimer
Axolabs' annealing conditions
Park's annealing conditions
1 X12712K1+X12713K1_1:1@37C_ACN30_40C UV_VIS_1
2 X12712K1+X12713K1_1:1@70C_ACN30_40C UV_VIS_1
3 X12712K1_ACN30_40C UV_VIS_1
4 X12713K1_ACN30_40C UV_VIS_1
mAU
WVL:260 nm
Multimeric siRNA
min
217
200
180
160
140
120
100
80
60
40
20
0
-20
5.68
7.00
8.00
9.00
10.00
11.00
12.00
13.02

FIG. 29

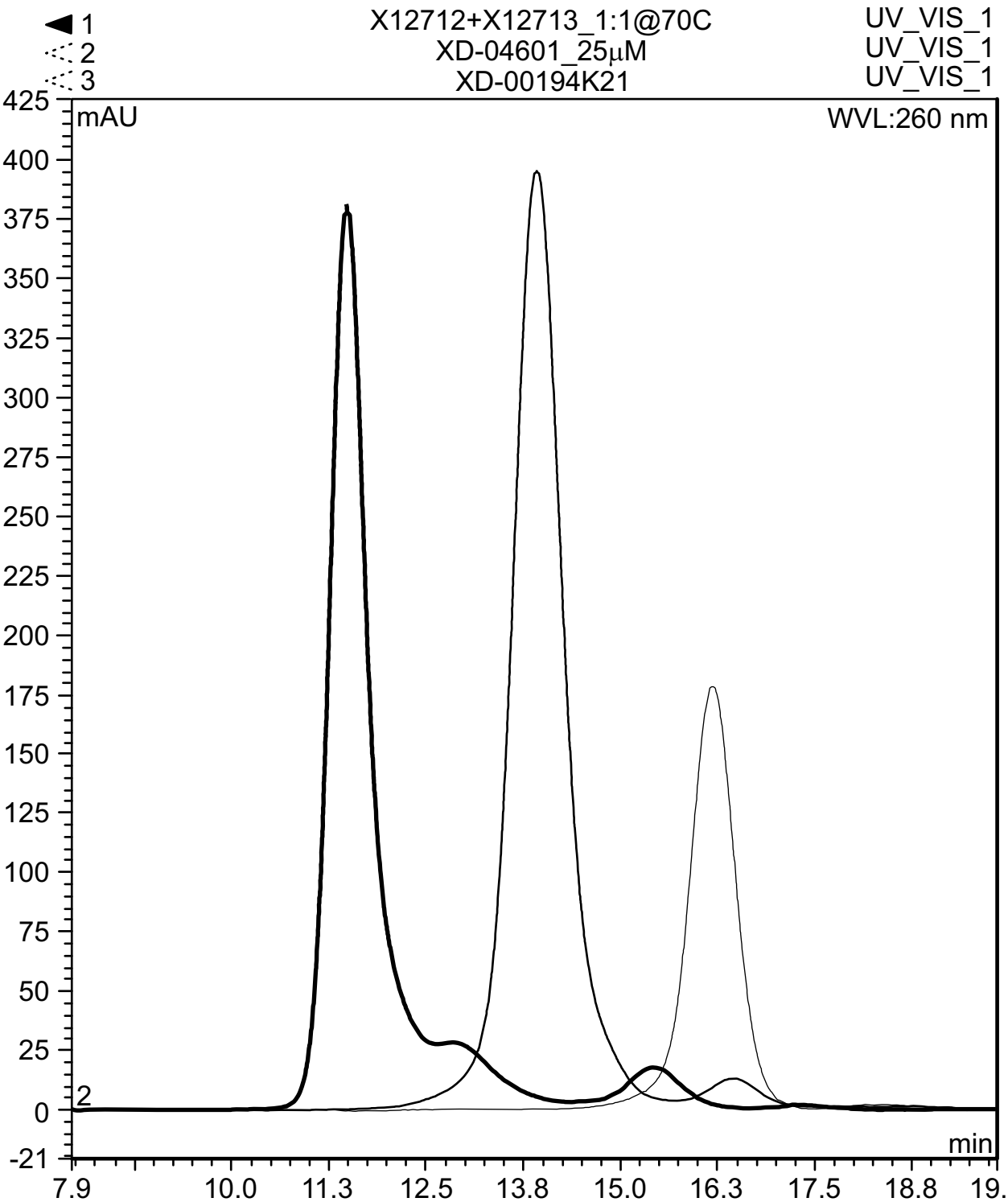

1
2
3
X12712+X12713_1:1@70C
XD-04601_25μM
XD-00194K21
UV_VIS_1
UV_VIS_1
UV_VIS_1
mAU
WVL:260 nm
425
400
375
350
325
300
275
250
225
200
175
150
125
100
75
50
25
0
-21
2
min
7.9
10.0
11.3
12.5
13.8
15.0
16.3
17.5
18.8
19.9

FIG. 30

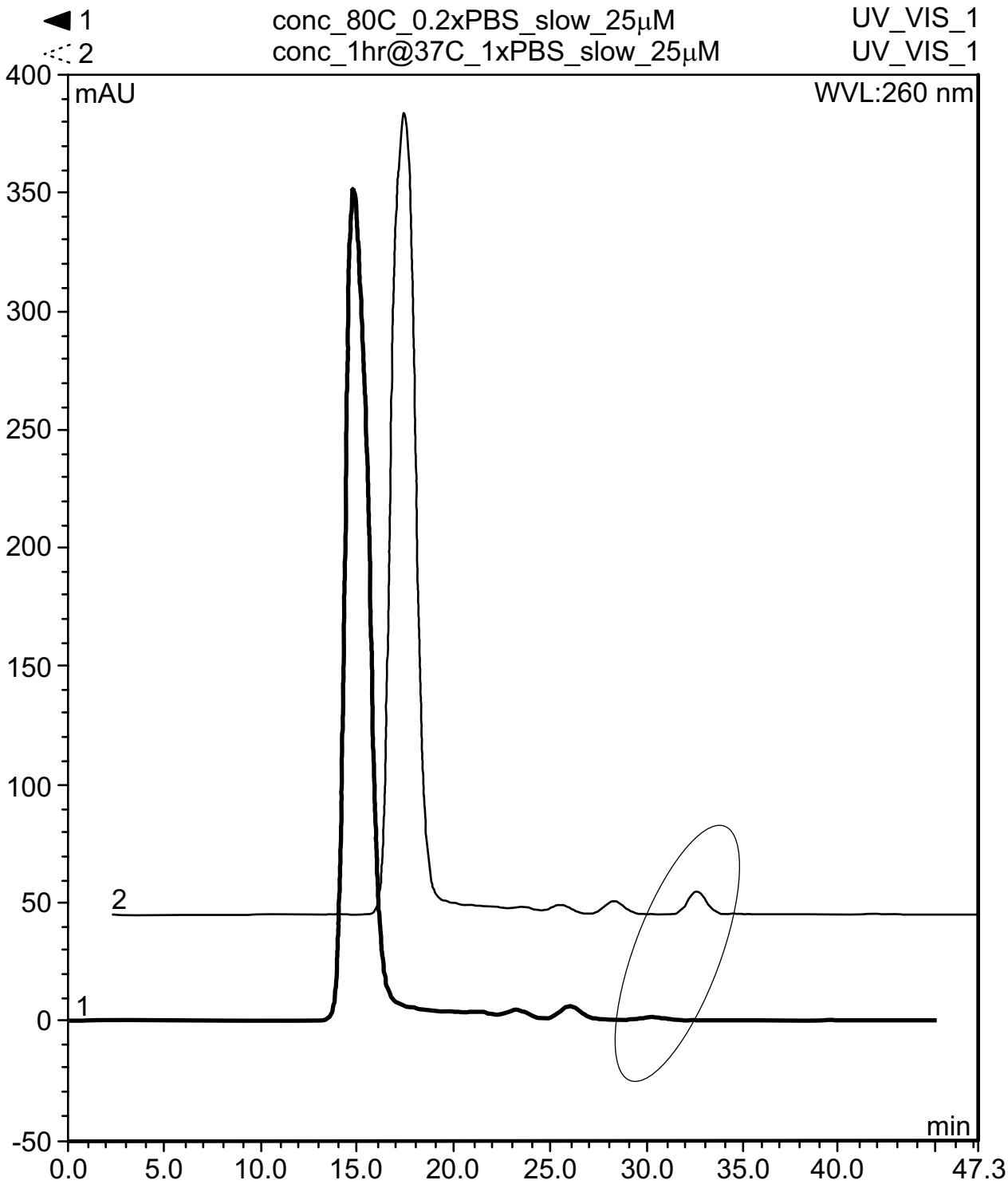

1 conc_80C_0.2xPBS_slow_25μM UV_VIS_1
2 conc_1hr@37C_1xPBS_slow_25μM UV_VIS_1
mAU
WVL:260 nm
400
350
300
250
200
150
100
50
0
-50
2
1
min
0.0
5.0
10.0
15.0
20.0
25.0
30.0
35.0
40.0
47.3

FIG. 31

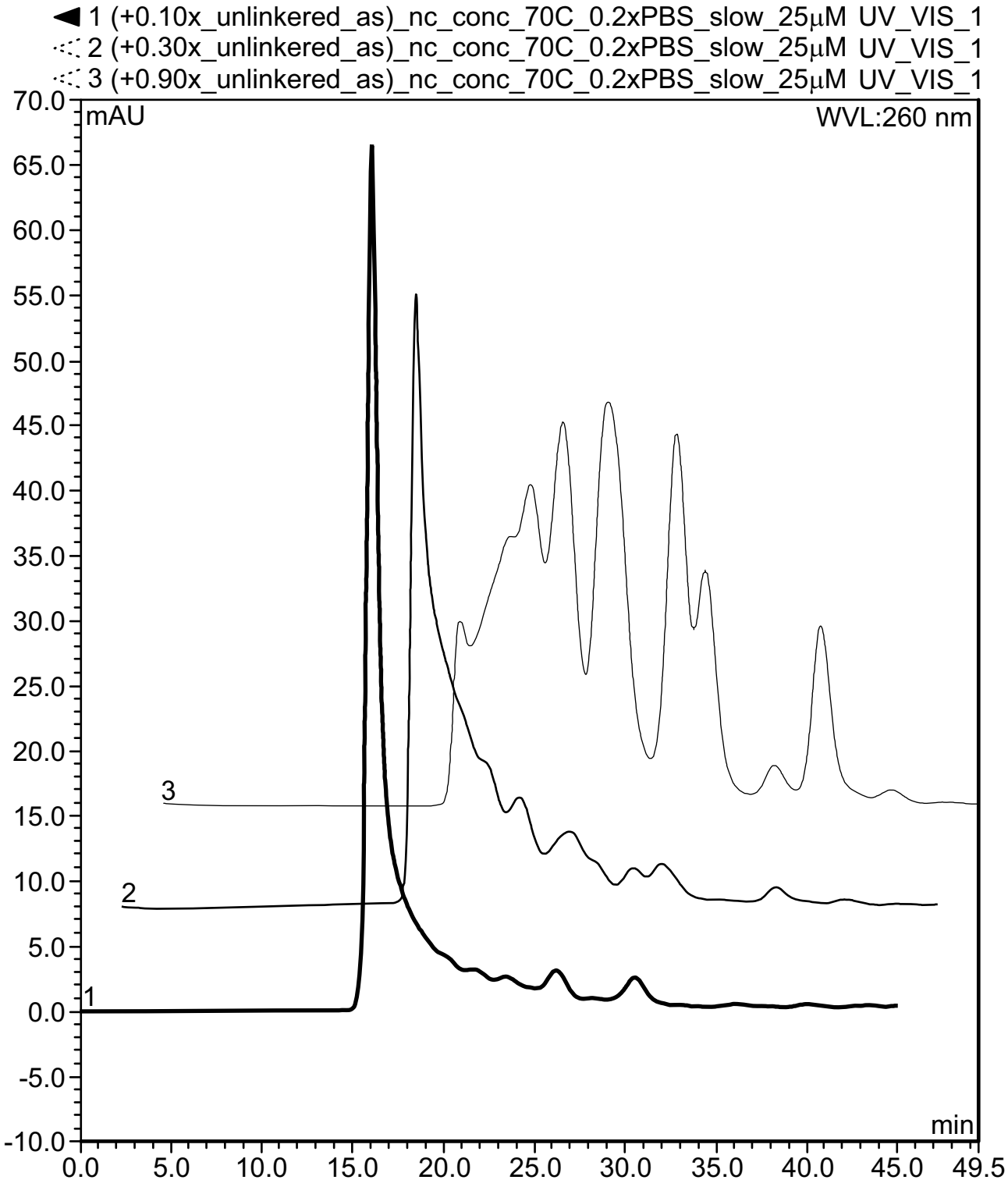

1 (+0.10x_unlinkered_as)_nc_conc_70C_0.2xPBS_slow_25μM UV_VIS_1
2 (+0.30x_unlinkered_as)_nc_conc_70C_0.2xPBS_slow_25μM UV_VIS_1
3 (+0.90x_unlinkered_as)_nc_conc_70C_0.2xPBS_slow_25μM UV_VIS_1
mAU
WVL:260 nm
min
70.0
65.0
60.0
55.0
50.0
45.0
40.0
35.0
30.0
25.0
20.0
15.0
10.0
5.0
0.0
-5.0
-10.0
0.0
5.0
10.0
15.0
20.0
25.0
30.0
35.0
40.0
45.0
49.5
1
2
3

FIG. 32

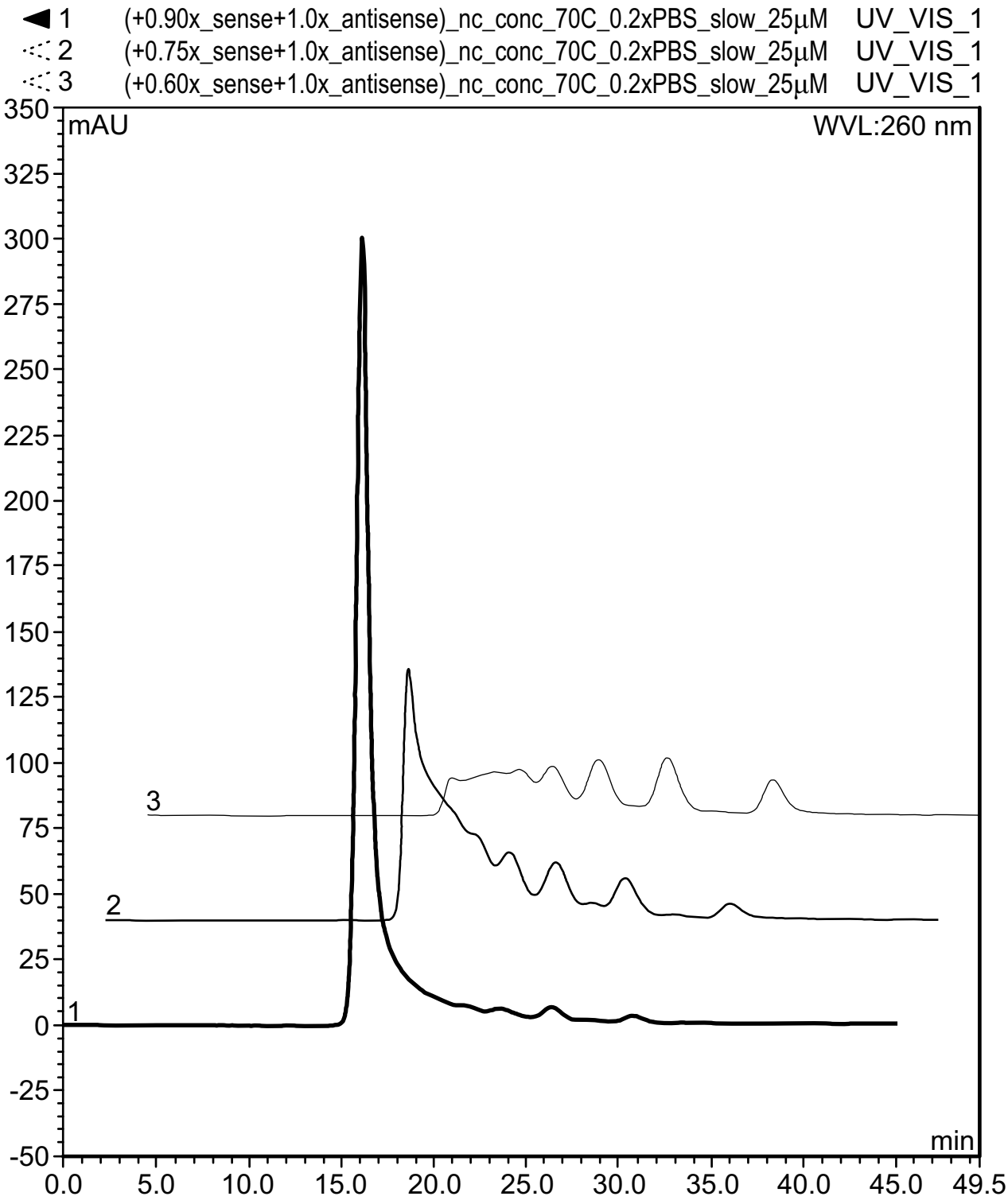

1 (+0.90x_sense+1.0x_antisense)_nc_conc_70C_0.2xPBS_slow_25µM UV_VIS_1
2 (+0.75x_sense+1.0x_antisense)_nc_conc_70C_0.2xPBS_slow_25µM UV_VIS_1
3 (+0.60x_sense+1.0x_antisense)_nc_conc_70C_0.2xPBS_slow_25µM UV_VIS_1
mAU
WVL:260 nm
350
325
300
275
250
225
200
175
150
125
100
75
50
25
0
-25
-50
1
2
3
min
0.0
5.0
10.0
15.0
20.0
25.0
30.0
35.0
40.0
45.0
49.5

FIG. 33

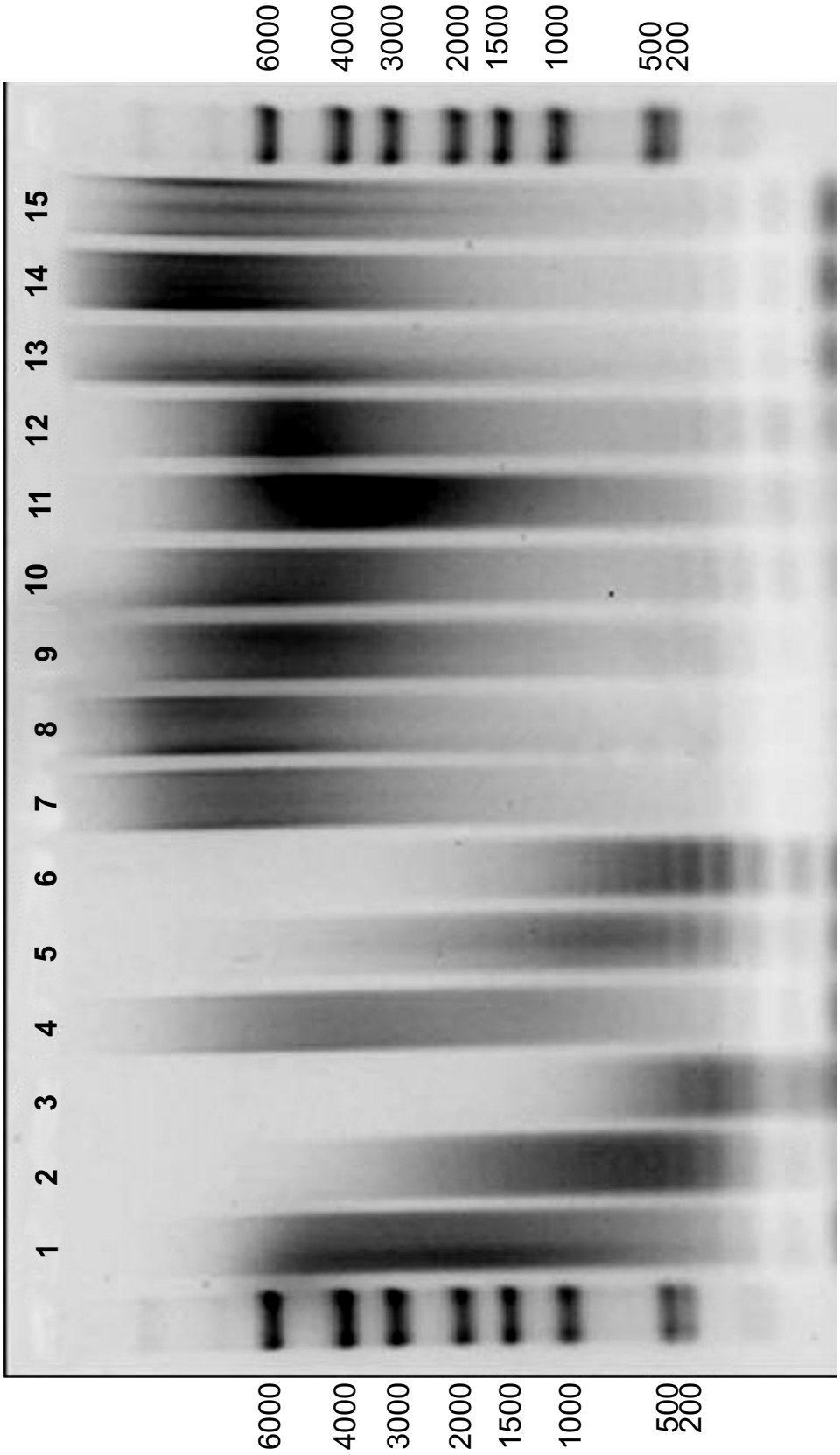

6000
4000
3000
2000
1500
1000
500
200
1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
6000
4000
3000
2000
1500
1000
500
200

FIG. 34A

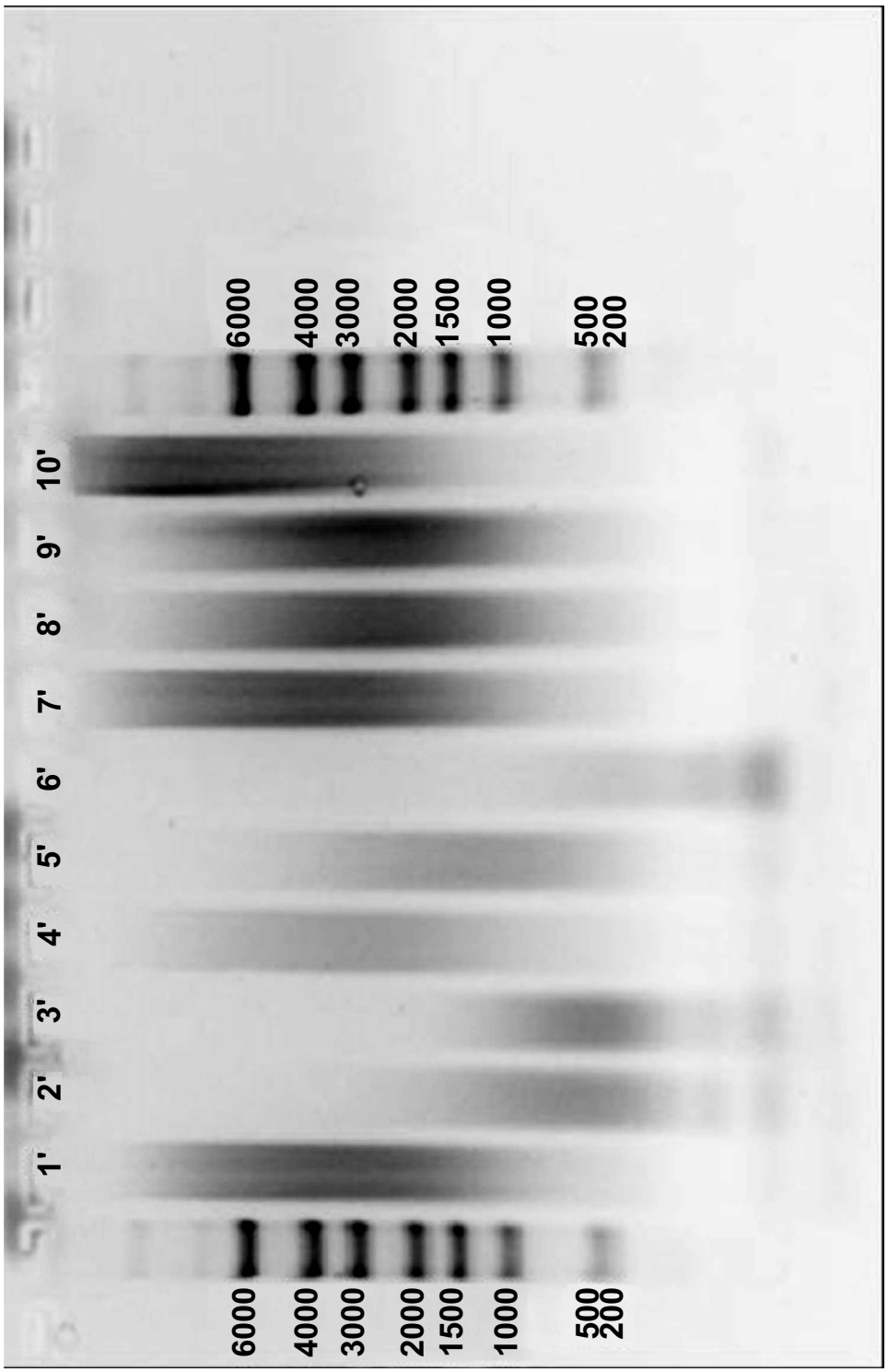

6000
4000
3000
2000
1500
1000
500
200
10'
9'
8'
7'
6'
5'
4'
3'
2'
1'
6000
4000
3000
2000
1500
1000
500
200

FIG. 34B

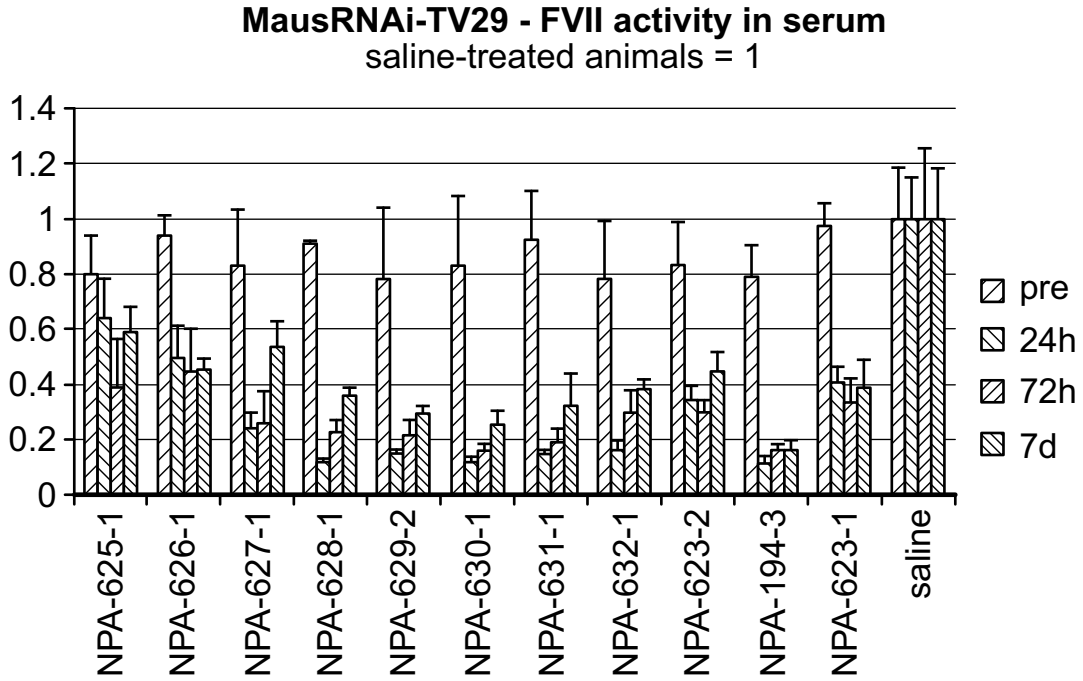

MausRNAi-TV29 - FVII activity in serum
saline-treated animals = 1
1.4
1.2
1
0.8
0.6
0.4
0.2
0
NPA-625-1
NPA-626-1
NPA-627-1
NPA-628-1
NPA-629-2
NPA-630-1
NPA-631-1
NPA-632-1
NPA-623-2
NPA-194-3
NPA-623-1
saline
pre
24h
72h
7d

FIG. 35

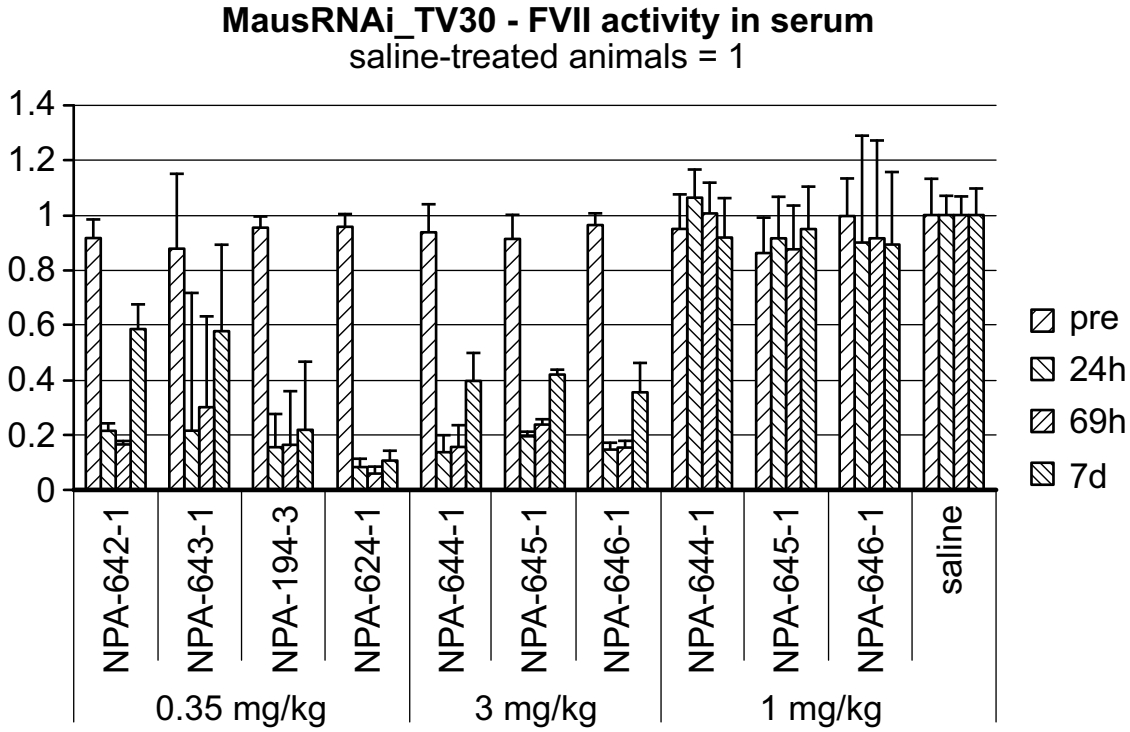

MausRNAi_TV30 - FVII activity in serum
saline-treated animals = 1
1.4
1.2
1
0.8
0.6
0.4
0.2
0
NPA-642-1
NPA-643-1
NPA-194-3
NPA-624-1
NPA-644-1
NPA-645-1
NPA-646-1
NPA-644-1
NPA-645-1
NPA-646-1
saline
0.35 mg/kg
3 mg/kg
1 mg/kg
pre
24h
69h
7d

FIG. 36

FIG. 37

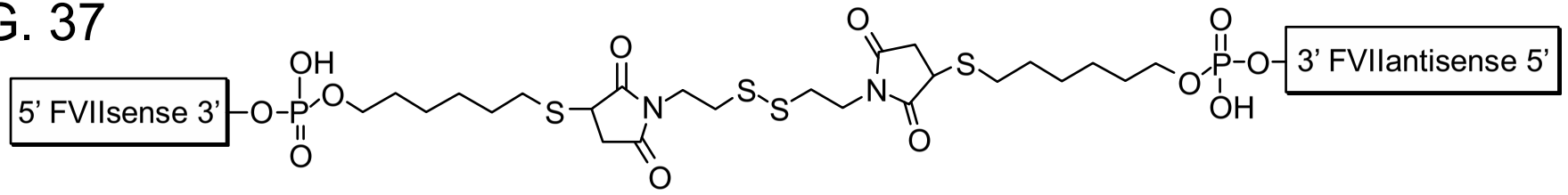
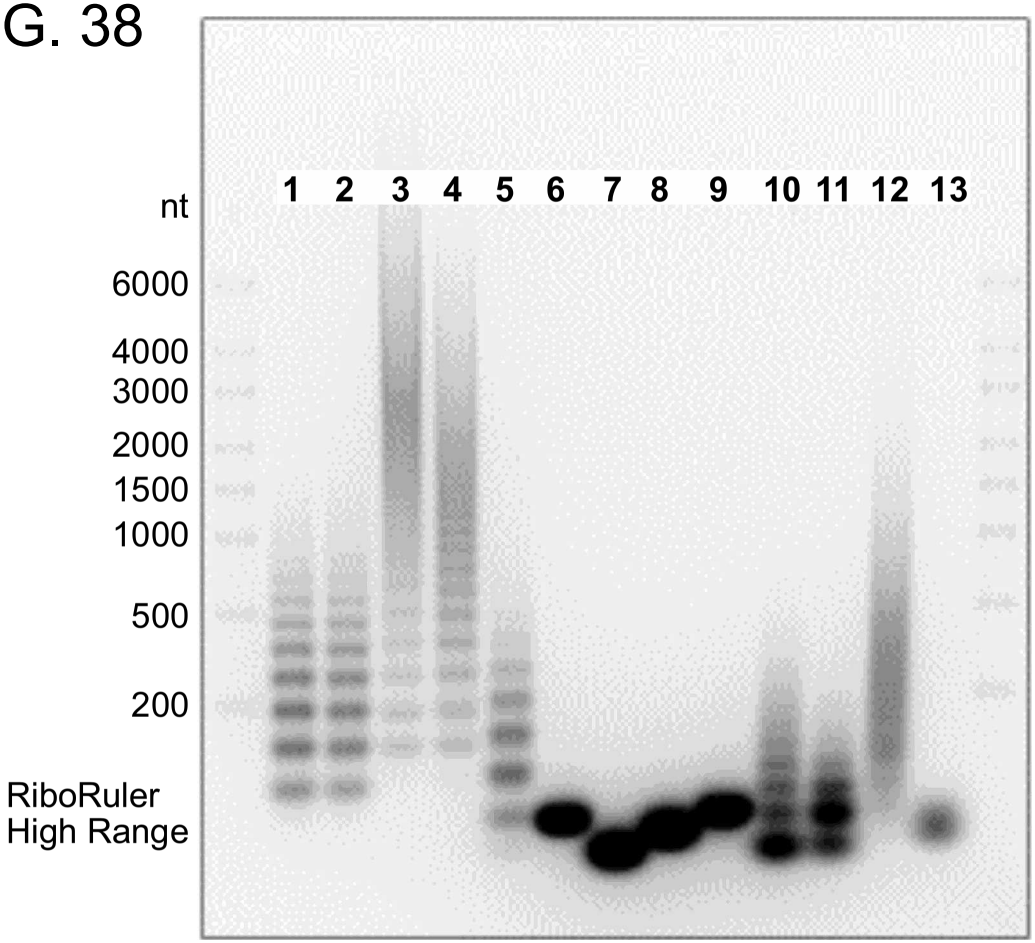


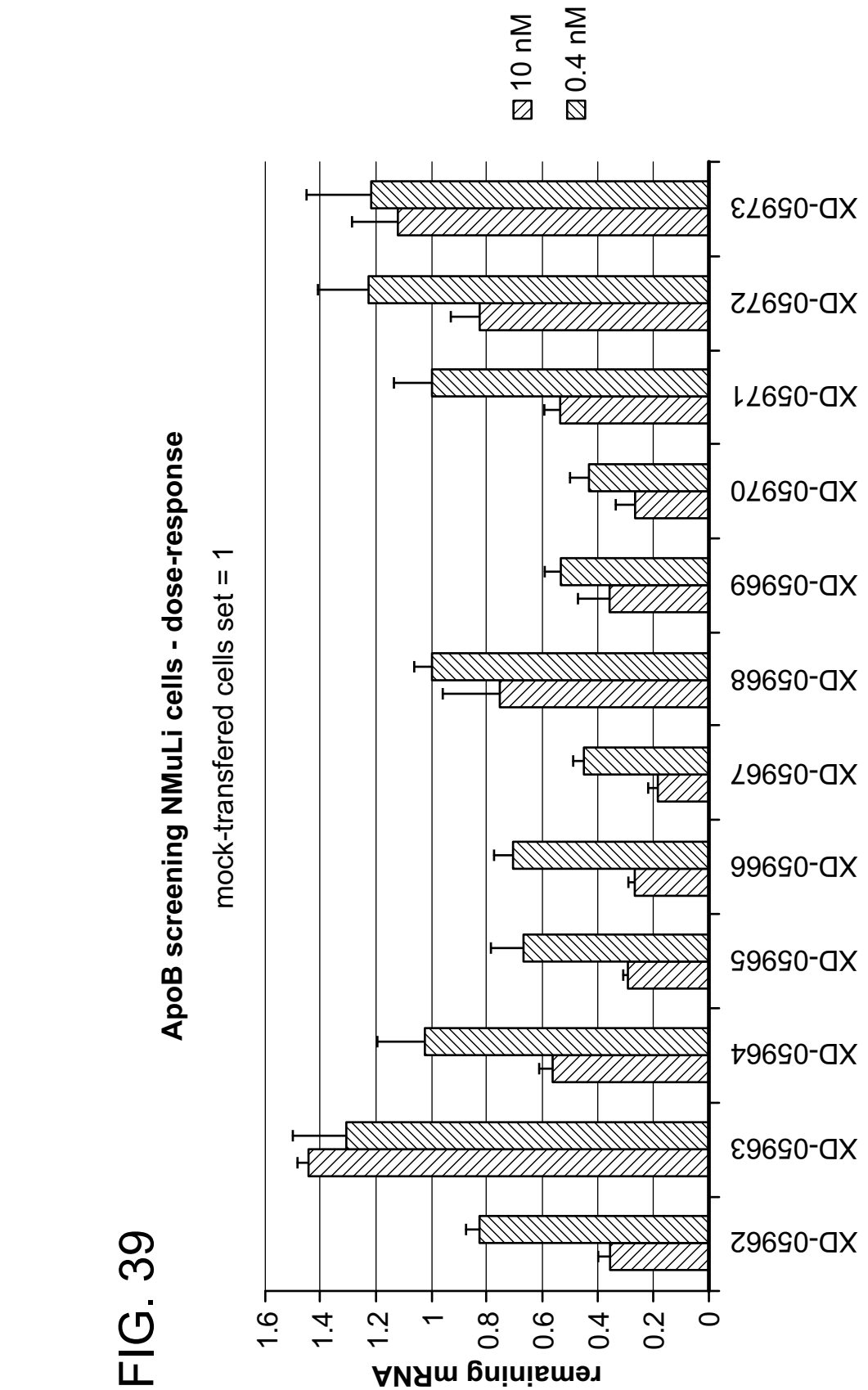
FIG. 38



1. XD-05305 K1: „lane 6" non-cleavable
2. XD-05306 K1: „lane 6" cleavable
3. XD-05307 K1: s-c-s + as-nc-as
4. XD-05308 K1: s-nc-s + as-c-as
5. XD-05309 K1: ⑤ tetrameric siRNA
6. XD-05310 K1: ⑥ dimeric siRNA
7. XD-00376 K3: FVII canonical siRNA
8. XD-00194 K24: F-Luc canonical siRNA
9. XD-05311 K1: FVII-sense-c-ApoB-sense with FVII-antisense and ApoB-antisense
10. XD-05312 K1: FVII-sense-c-FVII antisense with FVII-sense and FVII-antisense (annealed@70°C)
11. XD-05312 K2: FVII-sense-c-FVII-antisense with FVII-sense and FVII-antisense (annealed@90°C)
12. X12714 K1: FVII heterodimer
13. X15049 K1: FVII/ApoB heterodimer

1.5μg/lane
2% agarose gel in 1xTAE
140mA 130min
Gel red staining (1:10000)

FIG. 39


ApoB screening NMuLi cells - dose-response
mock-transfered cells set = 1
remaining mRNA
1.6
1.4
1.2
1
0.8
0.6
0.4
0.2
0
10 nM
0.4 nM
XD-05962
XD-05963
XD-05964
XD-05965
XD-05966
XD-05967
XD-05968
XD-05969
XD-05970
XD-05971
XD-05972
XD-05973