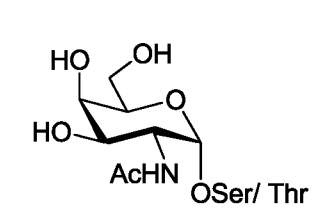
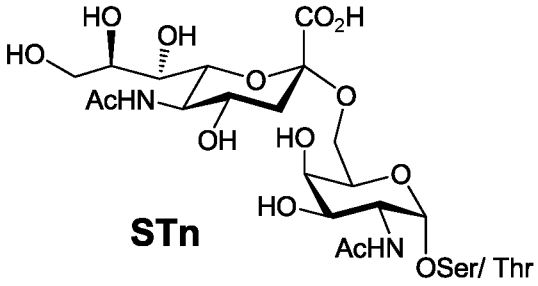
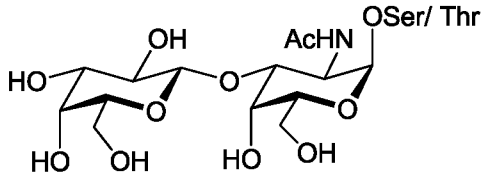
Fig. 1



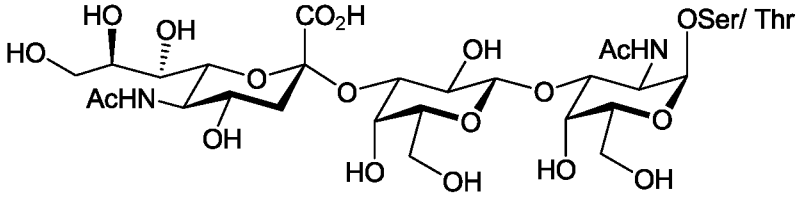
Tn



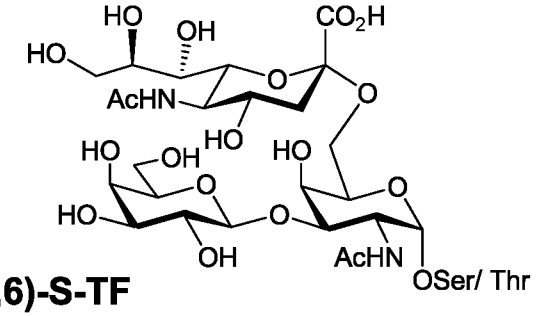
STn



TF



(2,3)-S-TF



(2,6)-S-TF

Fig. 2

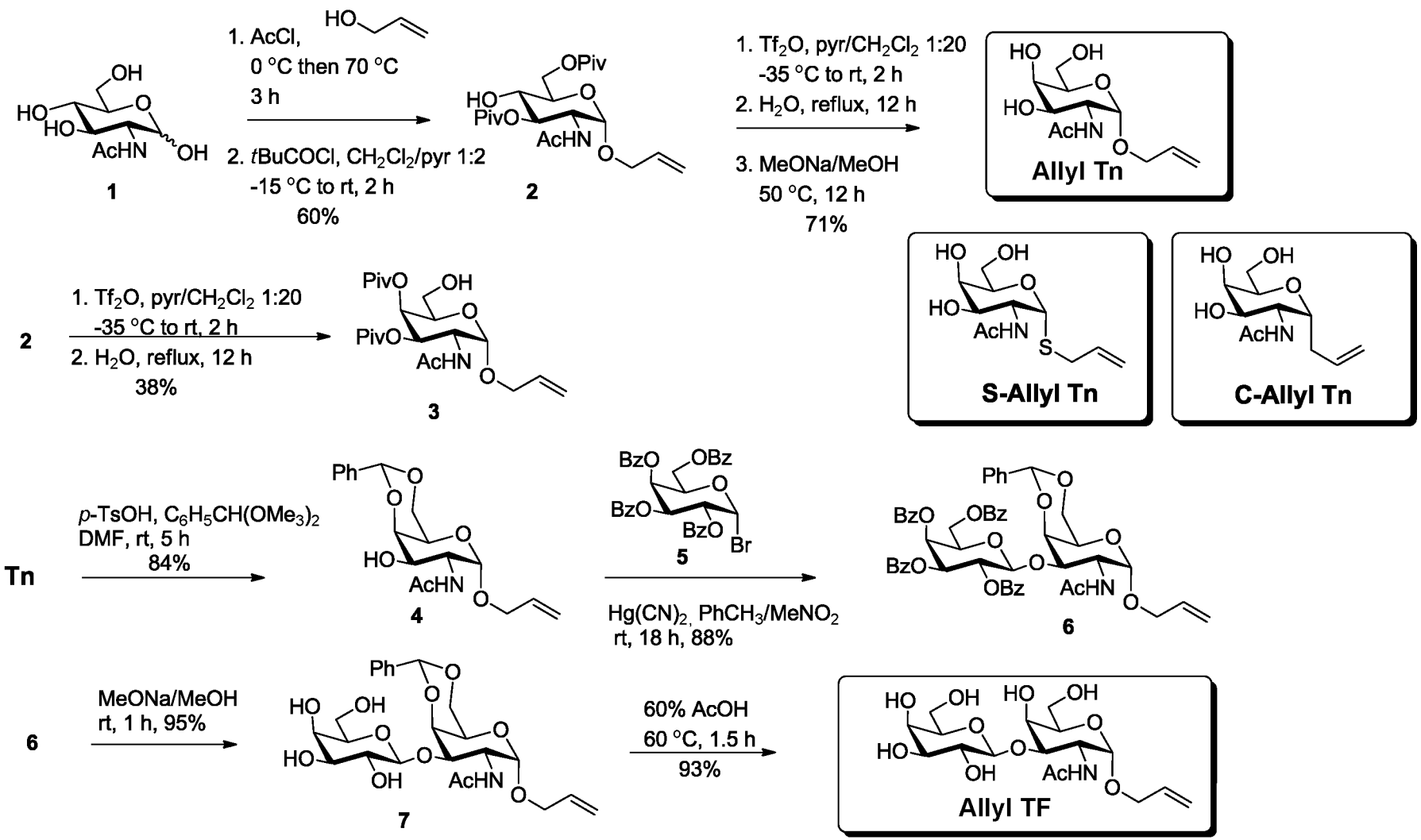

1. AcCl,
0 °C then 70 °C
3 h
2. tBuCOCl, CH2Cl2/pyr 1:2
-15 °C to rt, 2 h
60%
1
2
1. Tf2O, pyr/CH2Cl2 1:20
-35 °C to rt, 2 h
2. H2O, reflux, 12 h
3. MeONa/MeOH
50 °C, 12 h
71%
Allyl Tn
S-Allyl Tn
C-Allyl Tn
1. Tf2O, pyr/CH2Cl2 1:20
-35 °C to rt, 2 h
2. H2O, reflux, 12 h
38%
3
Tn
p-TsOH, C6H5CH(OMe3)2
DMF, rt, 5 h
84%
4
5
Hg(CN)2, PhCH3/MeNO2
rt, 18 h, 88%
6
MeONa/MeOH
rt, 1 h, 95%
7
60% AcOH
60 °C, 1.5 h
93%
Allyl TF

Fig. 3A

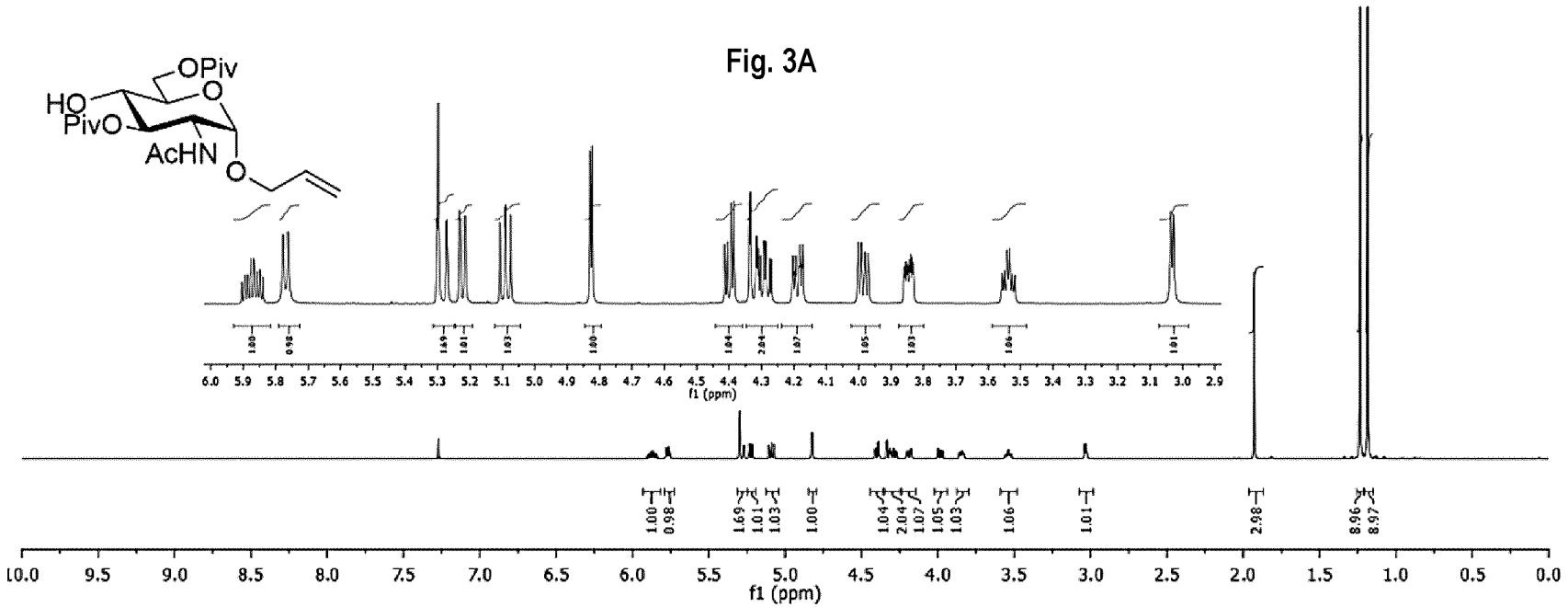


Fig. 3B

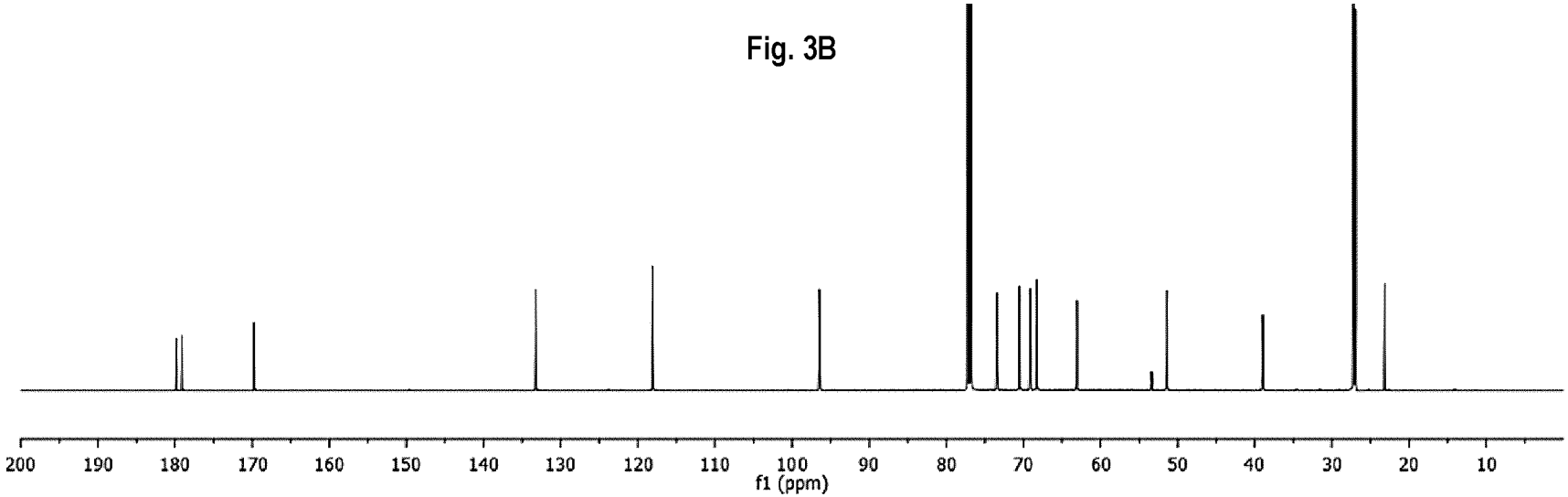


Fig. 3C

Compound Table

Compound Label	RT	Mass	Abund	Formula	Tgt Mass	Diff (ppm)
Cpd 1: C21 H35 N O8	0.1	429.2372	654630	C21 H35 N O8	429.2363	2.16

Compound Label	RT	Algorithm	Mass
Cpd 1: C21 H35 N O8	0.1	Find By Formula	429.2372

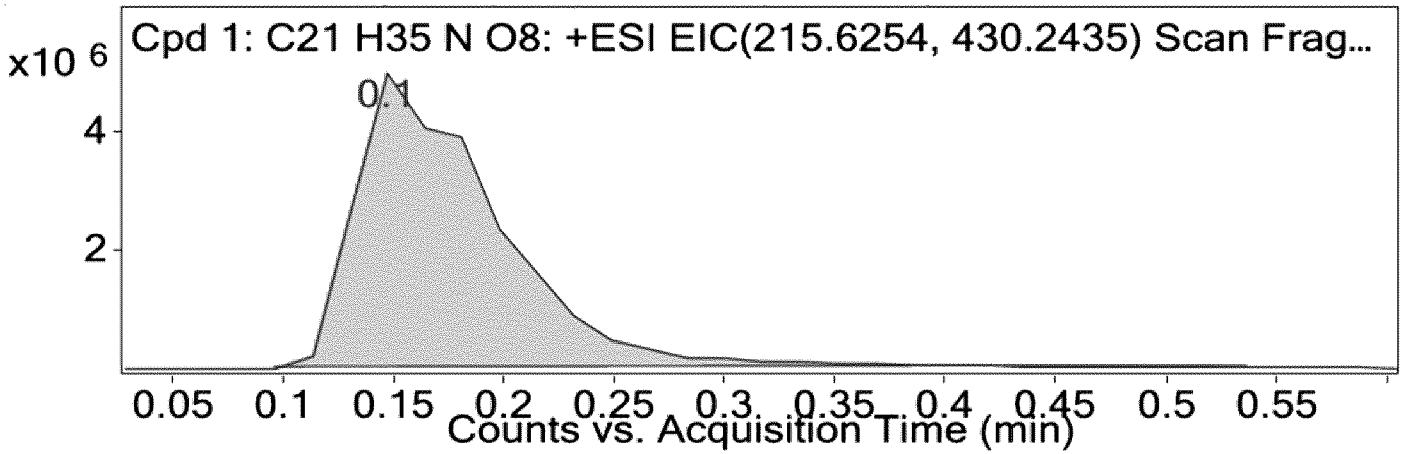
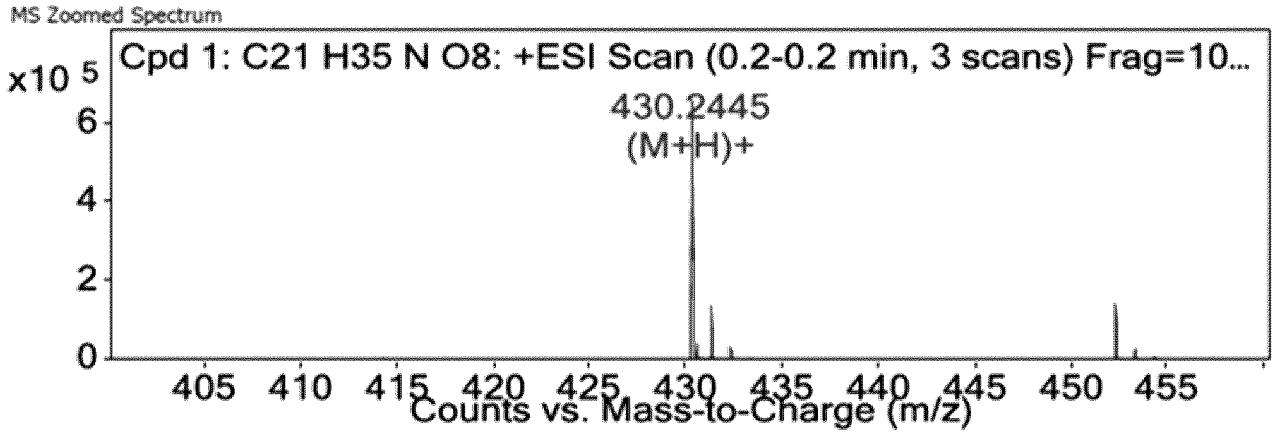


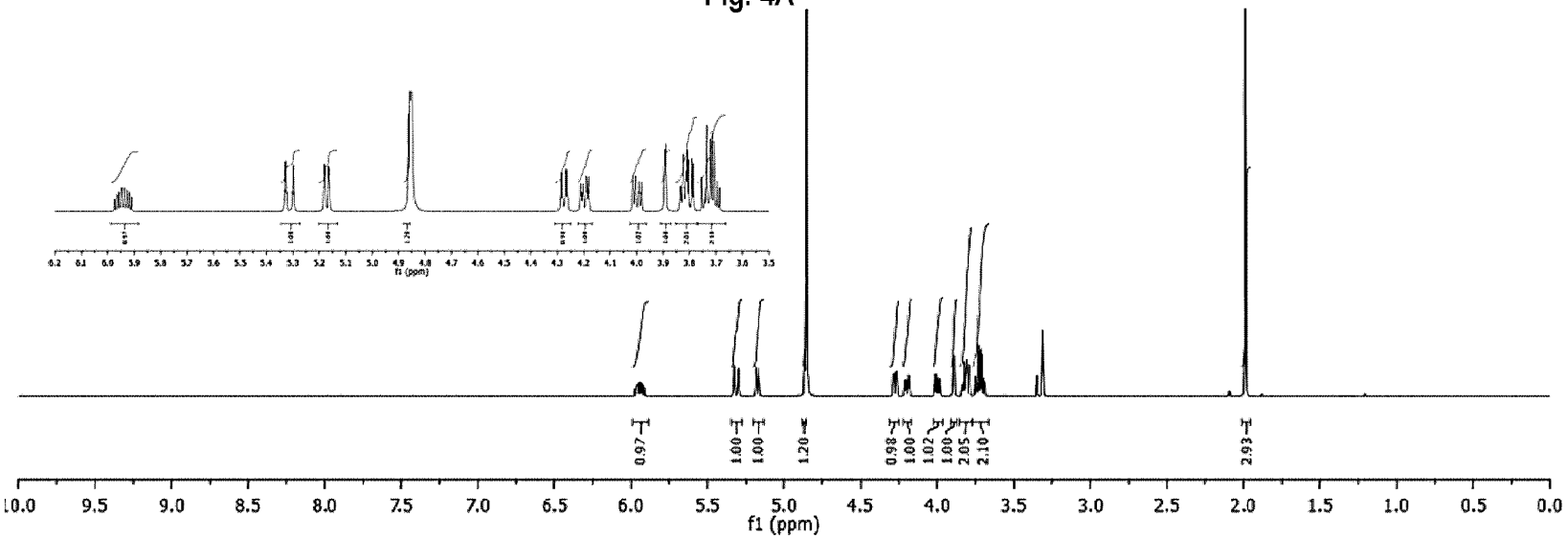
Fig. 3D



MS Spectrum Peak List

m/z	*Calc m/z*	Diff(ppm)	Abund	Formula	Ion
430.2445	430.2435	2.16	655034	C21 H36 N O8	(M+H)+
430.499			41847		
430.6539			3128		
430.7544			3520		
430.8793			2068		
431.0066			1911		
431.2472	431.2469	0.88	142986	C21 H36 N O8	(M+H)+
431.5048			5546		
432.2527	432.2493	7.91	32466	C21 H36 N O8	(M+H)+
433.2576	433.2519	13.25	5076	C21 H36 N O8	(M+H)+

Fig. 4A


0.97
1.00
1.00
1.20
0.98
1.00
1.02
1.00
2.05
2.10
2.93
10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0
f1 (ppm)

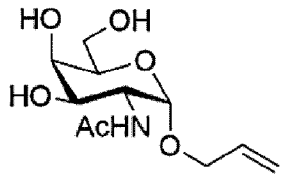

HO
OH
O
HO
AcHN
O

Fig. 4B

200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0
f1 (ppm)

Fig. 4C

Compound Table

Compound Label	RT	Mass	Abund	Formula	Tgt Mass	Diff (ppm)
Cpd 1: C11 H19 N O6	0.1	261.122	4575	C11 H19 N O6	261.1212	3.02

Compound Label	RT	Algorithm	Mass
Cpd 1: C11 H19 N O6	0.1	Find By Formula	261.122

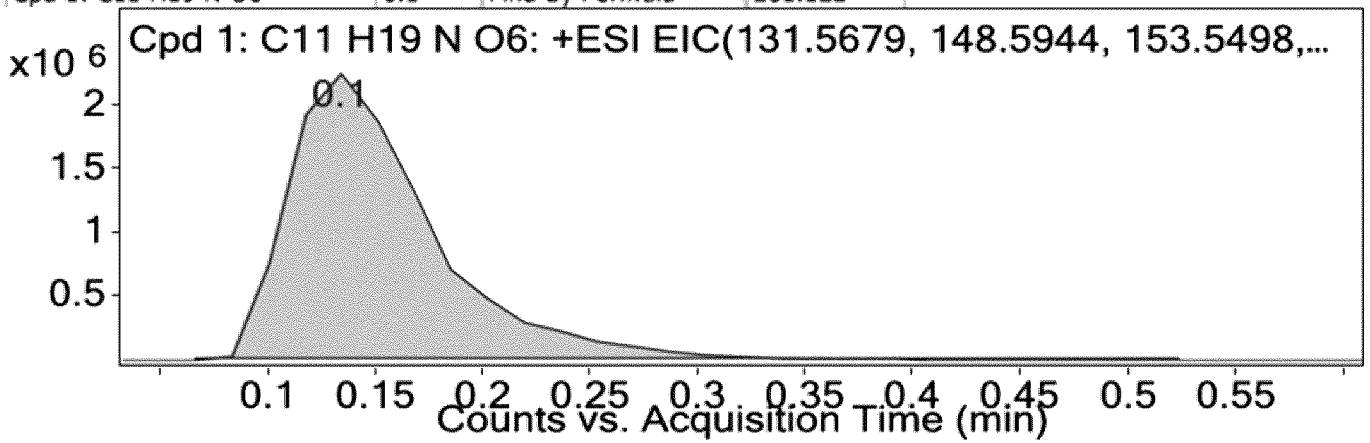
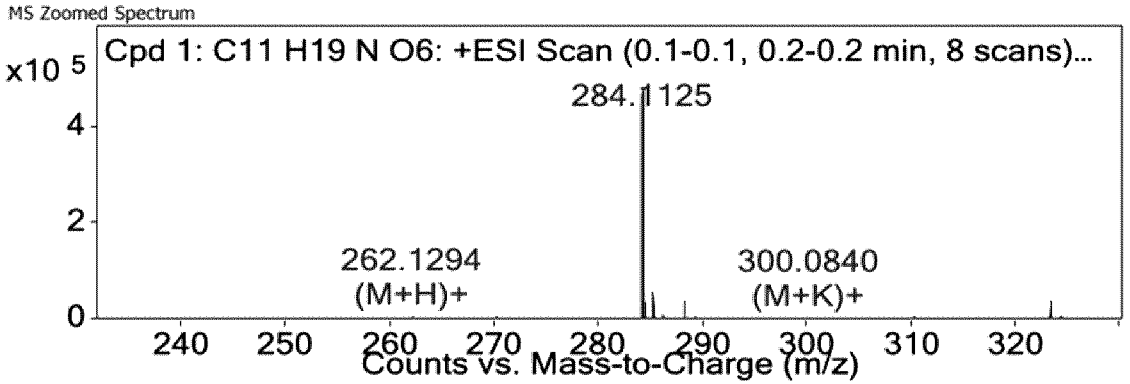


Fig. 4D



MS Spectrum Peak List

m/z	*Calc m/z*	Diff(ppm)	z	Abund	Formula	Ion
262.1294	262.1285	3.29	1	4575	C11 H20 N O6	(M+H)+
281.1036				3013		
282.2803				2279		
284.1125				499971		
284.3188				35672		
284.5274				2930		
285.1149				56535		
286.1165				8170		
288.1456				40848		
300.084	300.0844	-1.48	1	1776	C11 H19 K N O6	(M+K)+

Fig. 5A

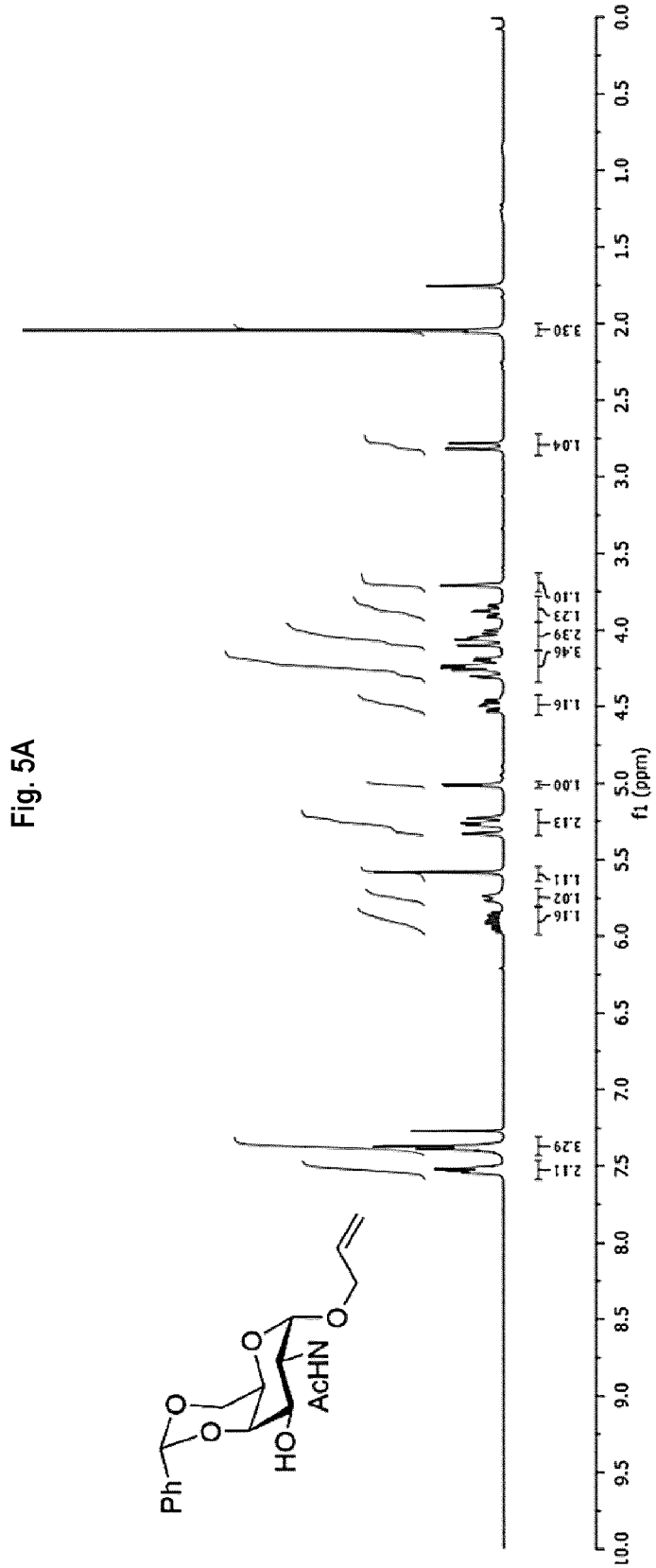

Ph
O
O
O
HO
AcHN
O
3.30
1.04
1.10
1.23
2.39
3.46
1.16
1.00
2.13
1.11
1.02
1.16
3.29
2.11
10.0
9.5
9.0
8.5
8.0
7.5
7.0
6.5
6.0
5.5
5.0
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
f1 (ppm)

Fig. 5B

Compound Table

Compound Label	RT	Mass	Abund	Formula	Tgt Mass	Diff (ppm)
Cpd 1: C18 H23 N O6	0.1	349.1535	144896	C18 H23 N O6	349.1525	2.83

Compound Label	RT	Algorithm	Mass
Cpd 1: C18 H23 N O6	0.1	Find By Formula	349.1535

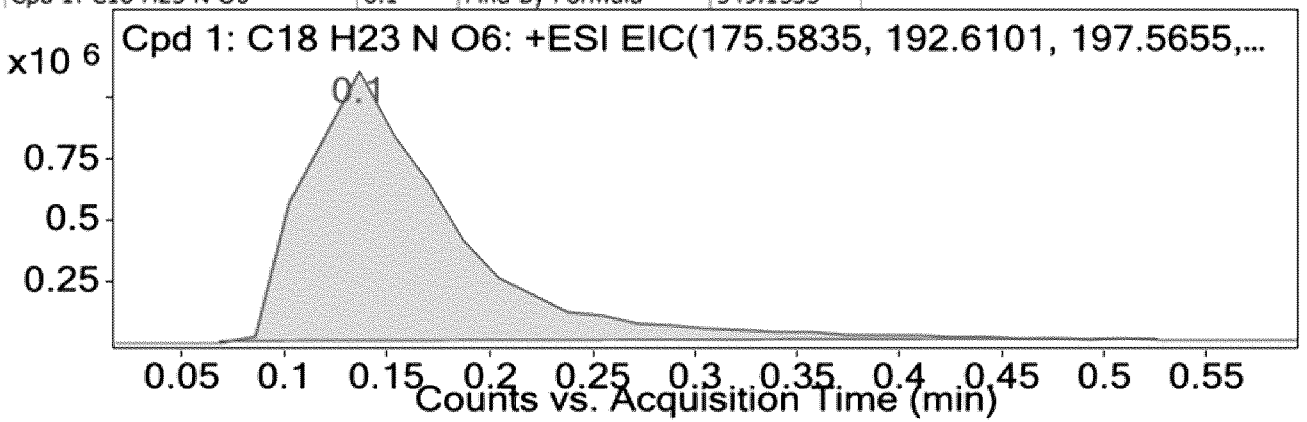
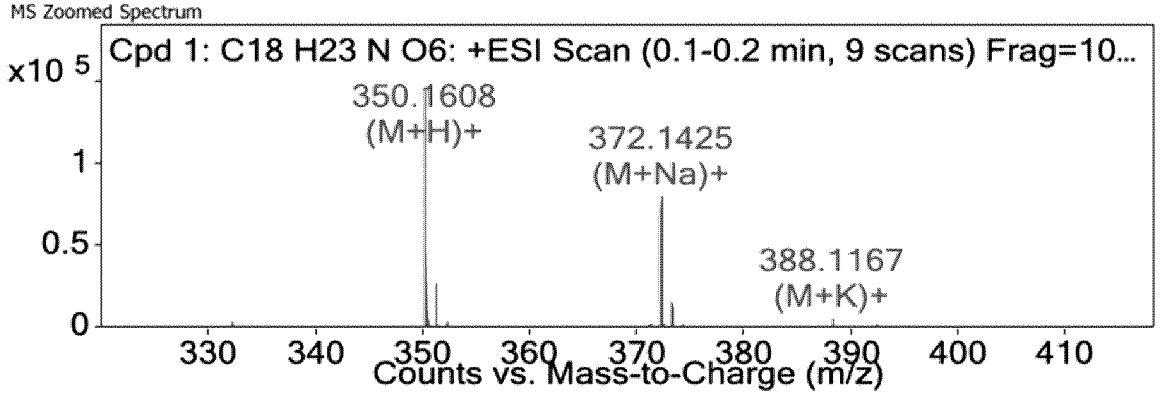


Fig. 5C



MS Spectrum Peak List

m/z	*Calc m/z*	Diff(ppm)	z	Abund	Formula	Ion
292.1189				268697		
292.3287				12192		
293.1218				39788		
294.1243				5059		
350.1608	350.1598	2.94	1	146470	C18 H24 N O6	(M+H)+
351.1639	351.1631	2.17	1	26665	C18 H24 N O6	(M+H)+
352.1666	352.1655	3.25	1	3895	C18 H24 N O6	(M+H)+
372.1425	372.1418	2.07	1	83058	C18 H23 N Na O6	(M+Na)+
373.1457	373.145	1.83	1	15140	C18 H23 N Na O6	(M+Na)+
388.1167	388.1157	2.72	1	5672	C18 H23 K N O6	(M+K)+

Fig. 6A

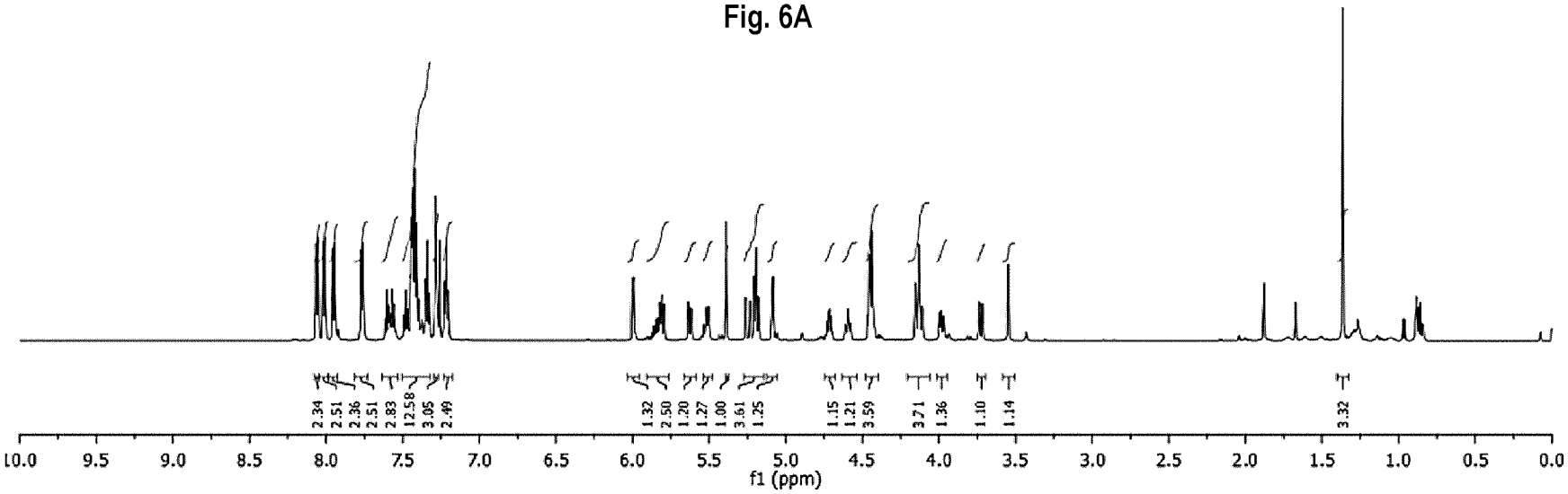

2.34
2.51
2.36
2.51
2.83
12.58
3.05
2.49
1.32
2.50
1.20
1.27
1.00
3.61
1.25
1.15
1.21
3.59
3.71
1.36
1.10
1.14
3.32
10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0
f1 (ppm)

Fig. 6B

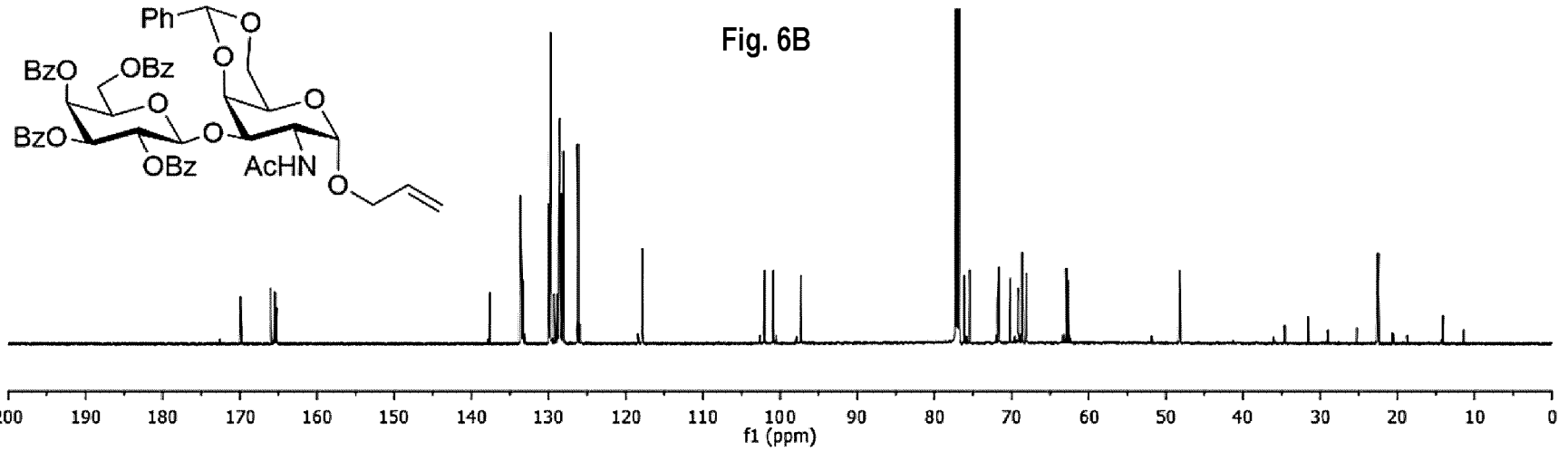

Ph
O
O
BzO
OBz
O
O
BzO
O
OBz
AcHN
O
200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0
f1 (ppm)

Fig. 6C

Compound Table

Compound Label	RT	Mass	Abund	Formula	Tgt Mass	Diff (ppm)
Cpd 1: C52 H49 N O15	0.1	927.306	163842	C52 H49 N O15	927.3102	-4.52

Compound Label	RT	Algorithm	Mass
Cpd 1: C52 H49 N O15	0.1	Find By Formula	927.306

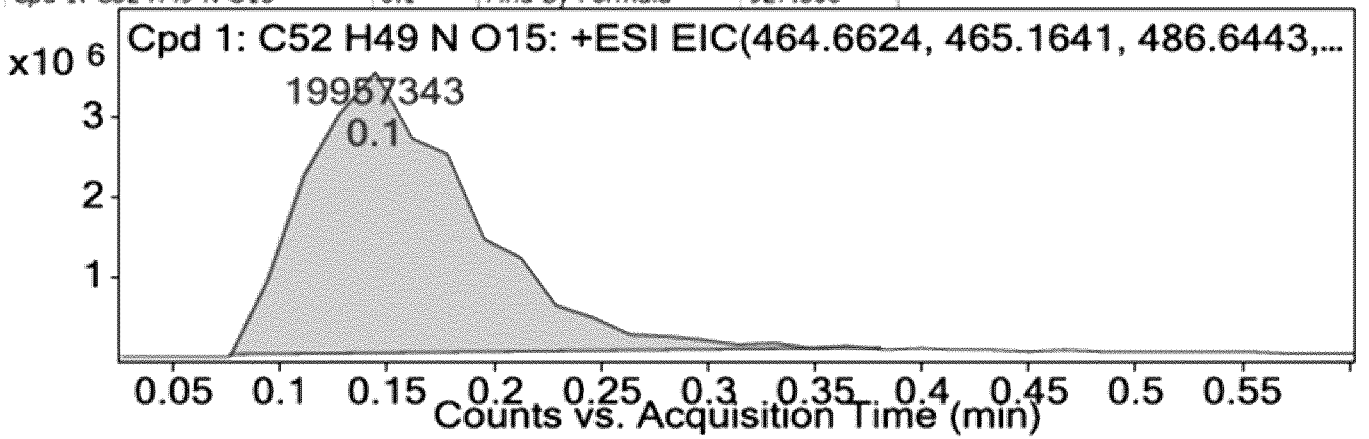
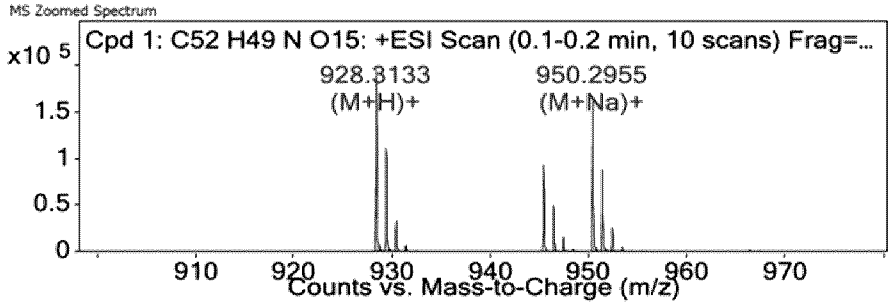


Fig. 6D



MS Spectrum Peak List

m/z	Calc m/z	Diff(ppm)	z	Abund	Formula	Ion
923.5509				21		
923.7411				20		
924.3629				28		
924.7805				22		
925.1704				18		
925.3865				27		
925.7963				28		
926.1317				17		
926.5423				32		
927.3562				40		
928.3133	928.3175	-4.56		206071	C52 H50 N O15	(M+H)+
928.683				6911		
929.3161	929.3208	-5.1		114342	C52 H50 N O15	(M+H)+
929.6901				3637		
930.3182	930.3238	-5.99		33881	C52 H50 N O15	(M+H)+
930.6988				1181		
930.9867				687		
931.3212	931.3266	-5.84		7262	C52 H50 N O15	(M+H)+
931.709				486		
931.9709				379		
932.3217	932.3294	-8.29		1526	C52 H50 N O15	(M+H)+
932.6588				232		
932.7928				212		
933.0294				184		
933.3202	933.3321	-12.71	1	311	C52 H50 N O15	(M+H)+
950.2955	950.2994	-4.13	1	164323	C52 H49 N Na O15	(M+Na)+
951.2986	951.3028	-4.41	1	91619	C52 H49 N Na O15	(M+Na)+
952.3009	952.3058	-5.08	1	27079	C52 H49 N Na O15	(M+Na)+
953.3035	953.3086	-5.37	1	5917	C52 H49 N Na O15	(M+Na)+
954.3025	954.3113	-9.29	1	1274	C52 H49 N Na O15	(M+Na)+

Fig. 7A

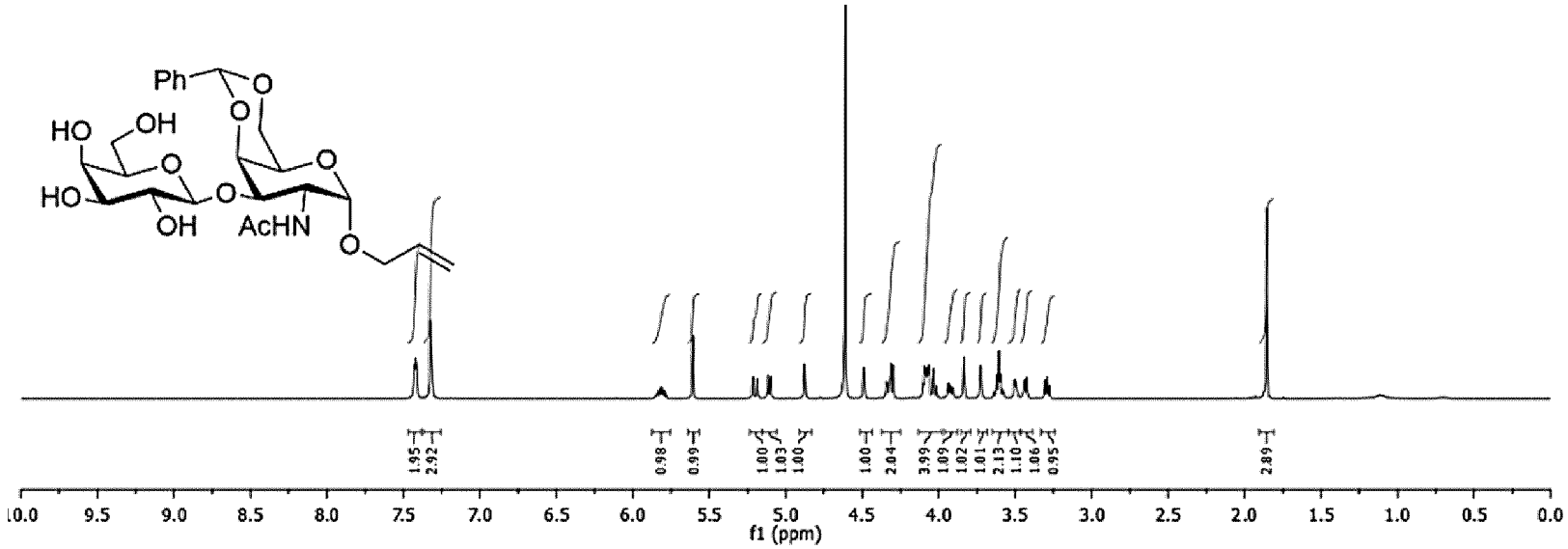

Ph
HO
OH
HO
OH
AcHN
1.95
2.92
0.98
0.99
1.00
1.03
1.00
1.00
2.04
3.99
1.09
1.02
1.01
2.13
1.10
1.06
0.95
2.89
10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0
f1 (ppm)

Fig. 7B

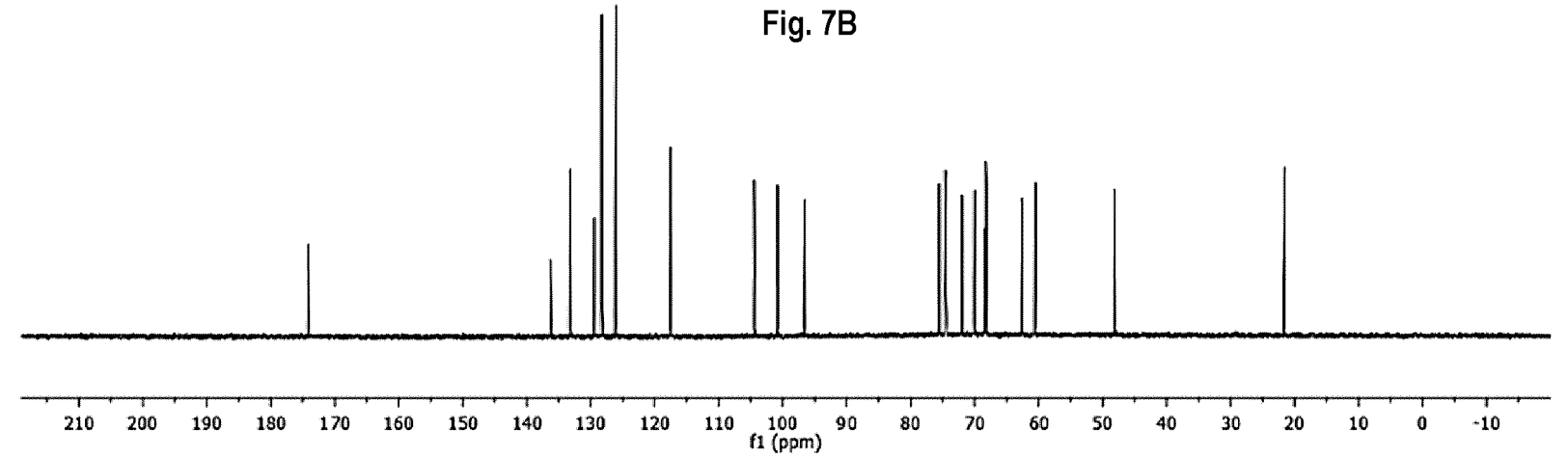

210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10
f1 (ppm)

Fig. 7C

Compound Table

Compound Label	RT	Mass	Abund	Formula	Tgt Mass	Diff (ppm)
Cpd 1: C24 H33 N O11	0.1	511.205	166749	C24 H33 N O11	511.2054	-0.72

Compound Label	RT	Algorithm	Mass
Cpd 1: C24 H33 N O11	0.1	Find By Formula	511.205

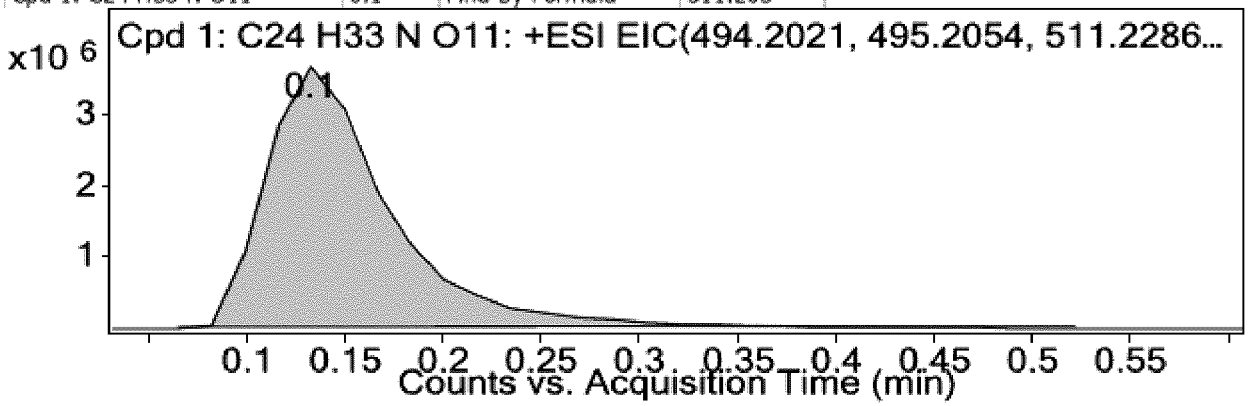
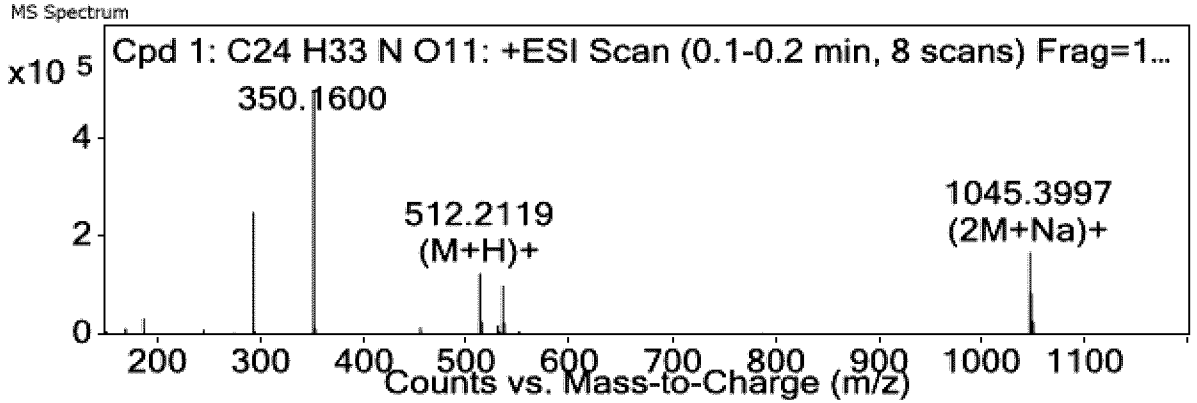


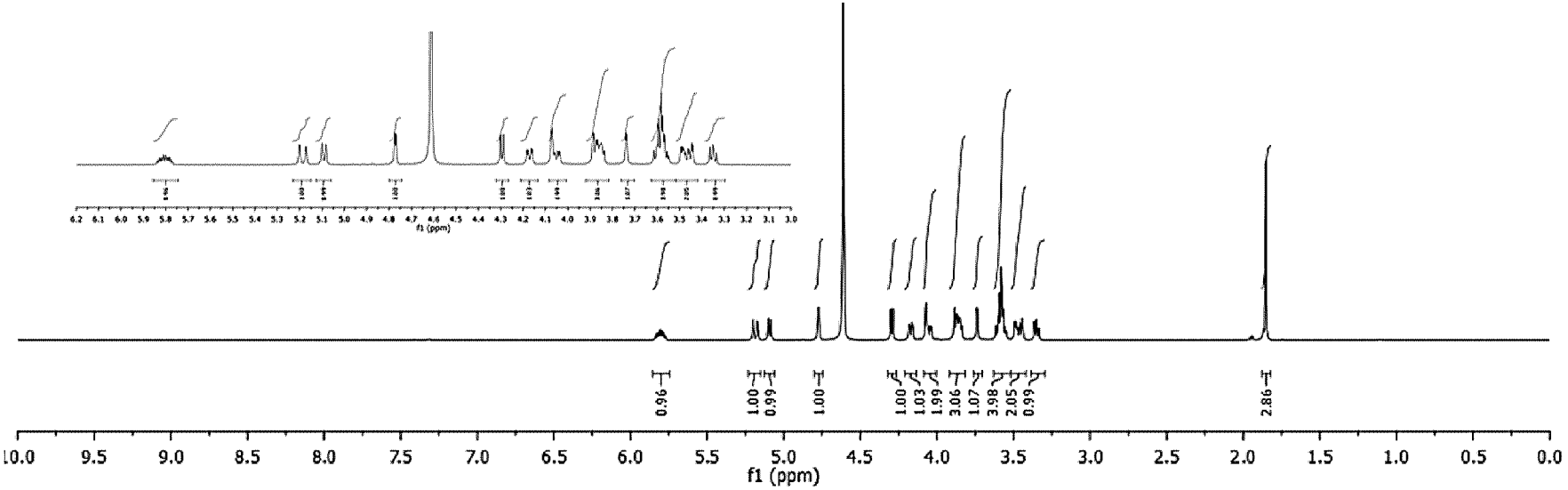
Fig. 7D



MS Spectrum Peak List

m/z	*Calc m/z*	Diff(ppm)	z	Abund	Formula	Ion
350.16				495714		
351.1626				91586		
512.2119	512.2126	-1.39	1	126315	C24 H34 N O11	(M+H)+
529.2383	529.2392	-1.64	1	20391	C24 H37 N2 O11	(M+NH4)+
534.1943	534.1946	-0.47	1	103350	C24 H33 N Na O11	(M+Na)+
550.1667	550.1685	-3.25	1	6050	C24 H33 K N O11	(M+K)+
1023.4185	1023.418	0.53	1	295	C48 H67 N2 O22	(2M+H)+
1040.4393	1040.4445	-5.01	1	1591	C48 H70 N3 O22	(2M+NH4)+
1045.3997	1045.3999	-0.25	1	171974	C48 H66 N2 Na O22	(2M+Na)+
1061.3716	1061.3739	-2.14	1	1026	C48 H66 K N2 O22	(2M+K)+

Fig. 8A


0.96
1.00
0.99
1.00
1.00
1.03
1.99
3.06
1.07
3.98
2.05
0.99
2.86
10.0 9.5 9.0 8.5 8.0 7.5 7.0 6.5 6.0 5.5 5.0 4.5 4.0 3.5 3.0 2.5 2.0 1.5 1.0 0.5 0.0
f1 (ppm)

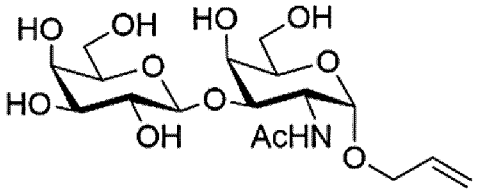

HO
OH
HO
O
O
HO
OH
AcHN
O

Fig. 8B

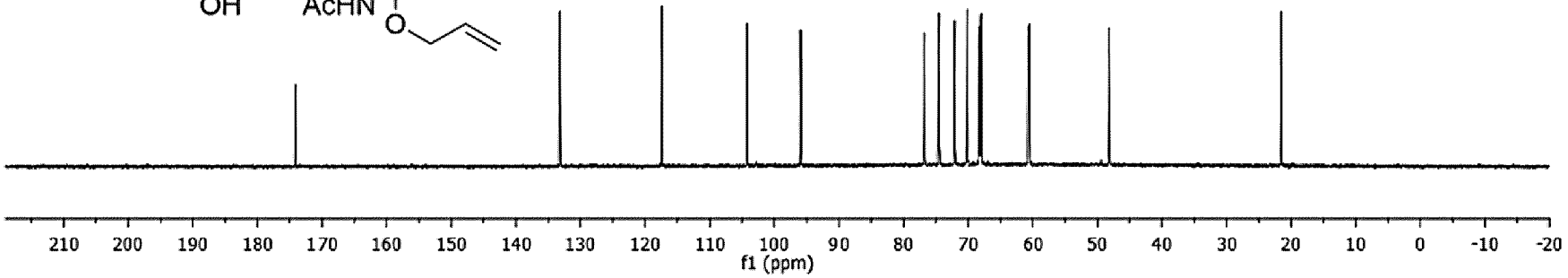

210 200 190 180 170 160 150 140 130 120 110 100 90 80 70 60 50 40 30 20 10 0 -10 -20
f1 (ppm)

Fig. 8C

Compound Table

Compound Label	RT	Mass	Abund	Formula	Tgt Mass	Diff (ppm)
Cpd 1: C17 H29 N O11	0.2	423.1721	82458	C17 H29 N O11	423.1741	-4.71

Compound Label	RT	Algorithm	Mass
Cpd 1: C17 H29 N O11	0.2	Find By Formula	423.1721

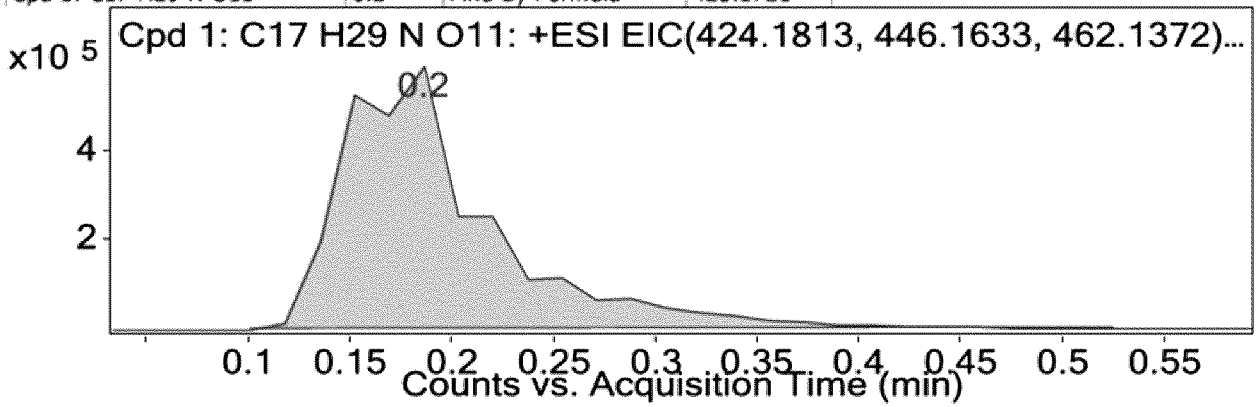
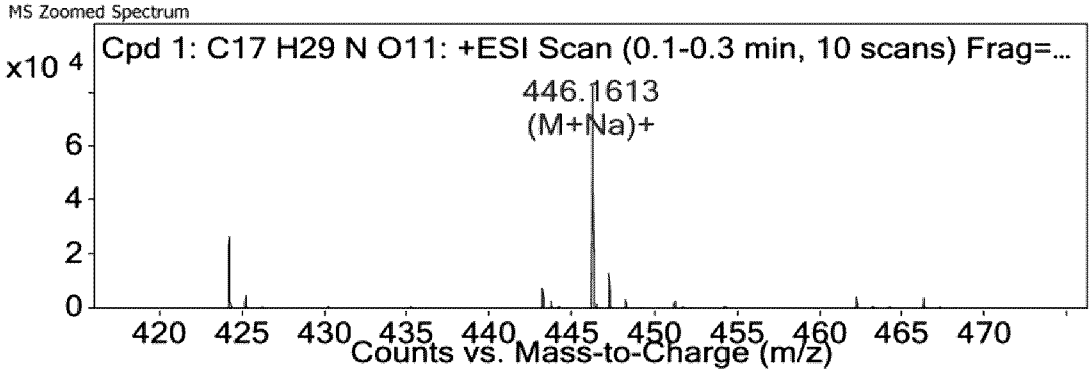


Fig. 8D



MS Spectrum Peak List

m/z	*Calc m/z*	Diff(ppm)	z	Abund	Formula	Ion
446.1613	446.1633	-4.46	1	82596	C17 H29 N Na O11	(M+Na)+
447.1642	447.1666	-5.42	1	14332	C17 H29 N Na O11	(M+Na)+
448.1622	448.1686	-14.24	1	3664	C17 H29 N Na O11	(M+Na)+
869.3348				211251		
869.6971				9148		
869.8102				1879		
870.3378				76033		
870.7046				2436		
871.3364				22835		
872.3365				5226		

--- End Of Report ---

Fig. 9

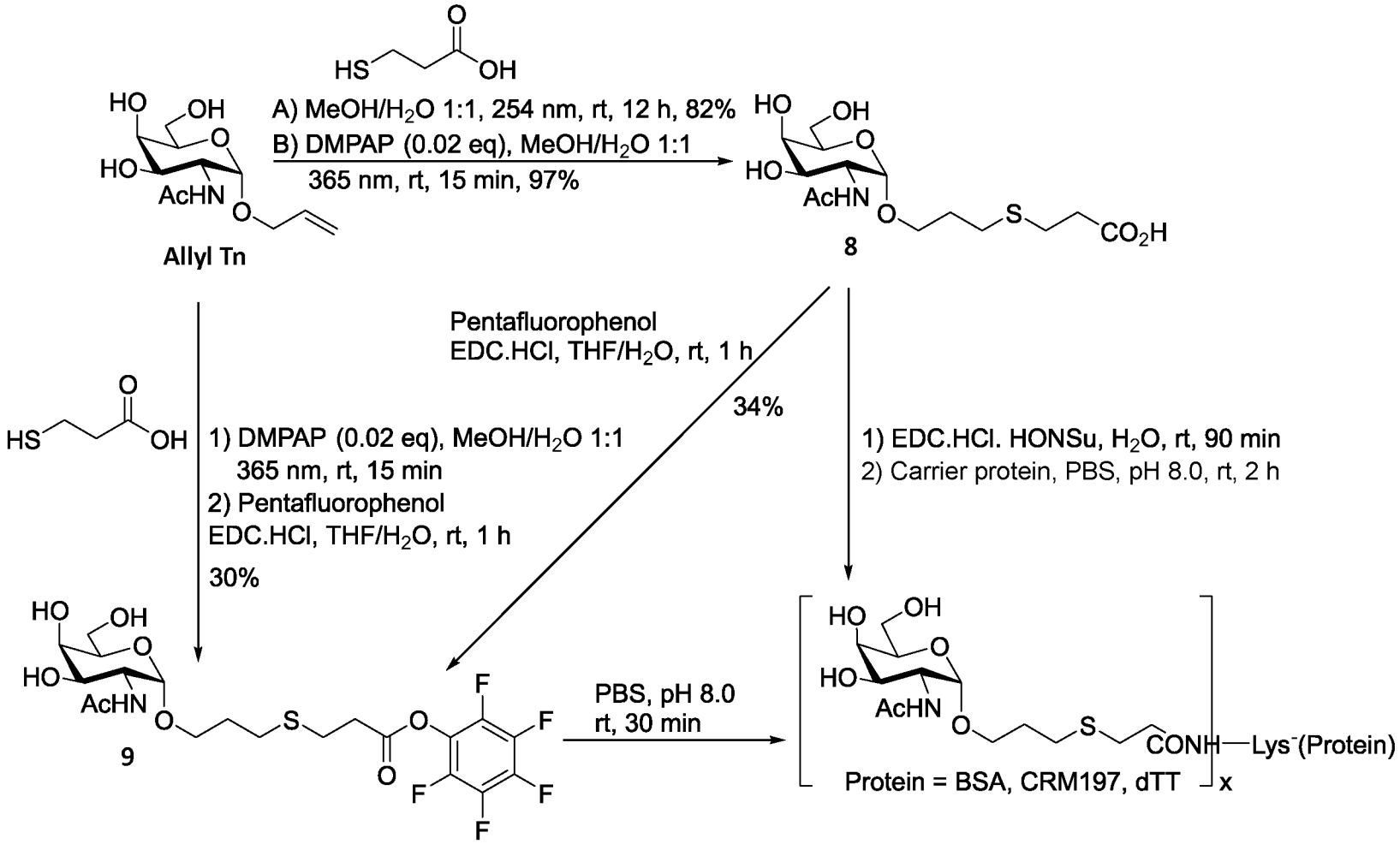

HS
OH
O
A) MeOH/H2O 1:1, 254 nm, rt, 12 h, 82%
B) DMPAP (0.02 eq), MeOH/H2O 1:1
365 nm, rt, 15 min, 97%
HO
OH
HO
AcHN
O
Allyl Tn
8
CO2H
S
Pentafluorophenol
EDC.HCl, THF/H2O, rt, 1 h
34%
1) DMPAP (0.02 eq), MeOH/H2O 1:1
365 nm, rt, 15 min
2) Pentafluorophenol
EDC.HCl, THF/H2O, rt, 1 h
30%
1) EDC.HCl. HONSu, H2O, rt, 90 min
2) Carrier protein, PBS, pH 8.0, rt, 2 h
9
F
PBS, pH 8.0
rt, 30 min
CONH—Lys-(Protein)
Protein = BSA, CRM197, dTT
x

Fig. 10

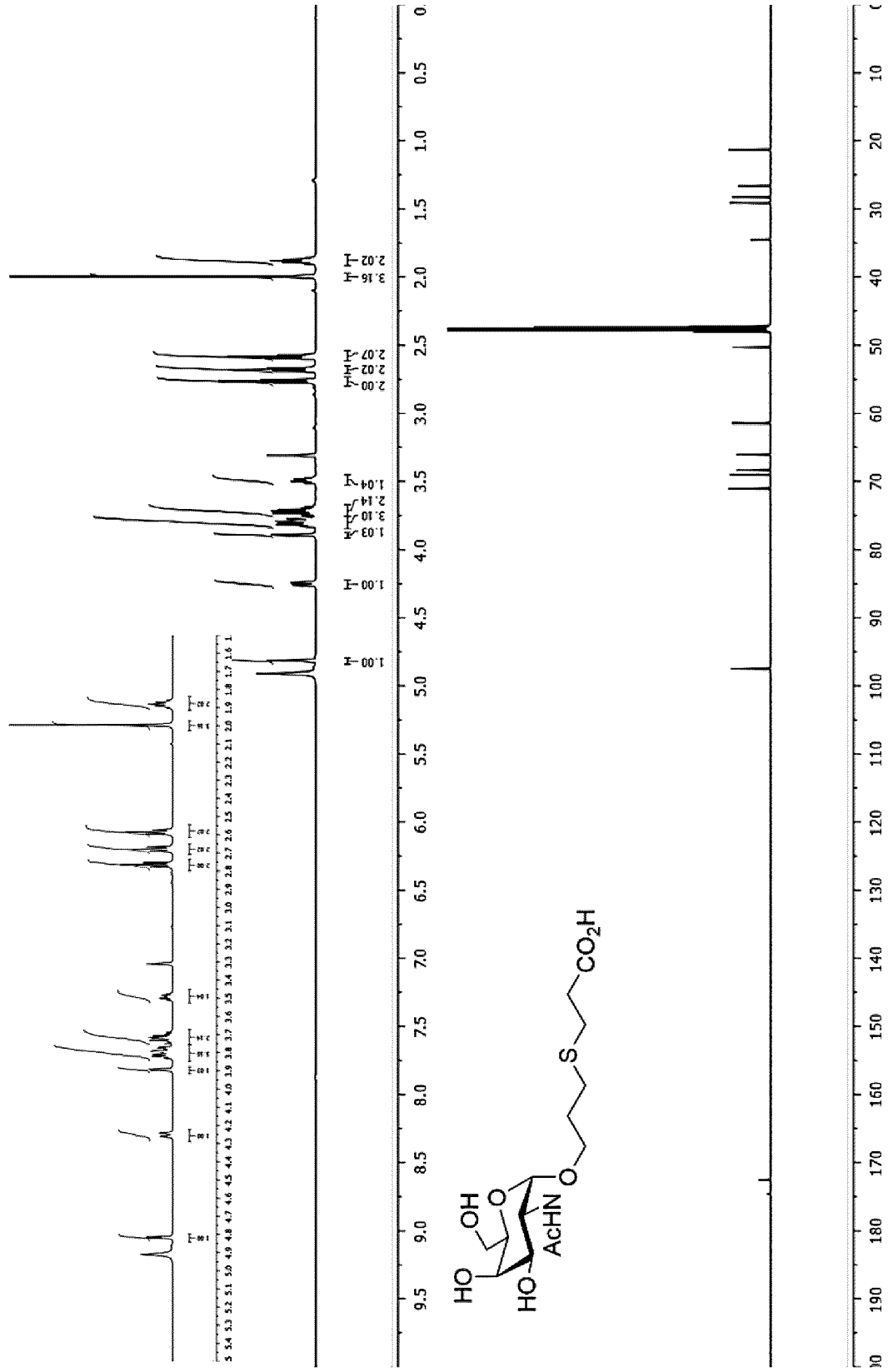


Fig. 11A

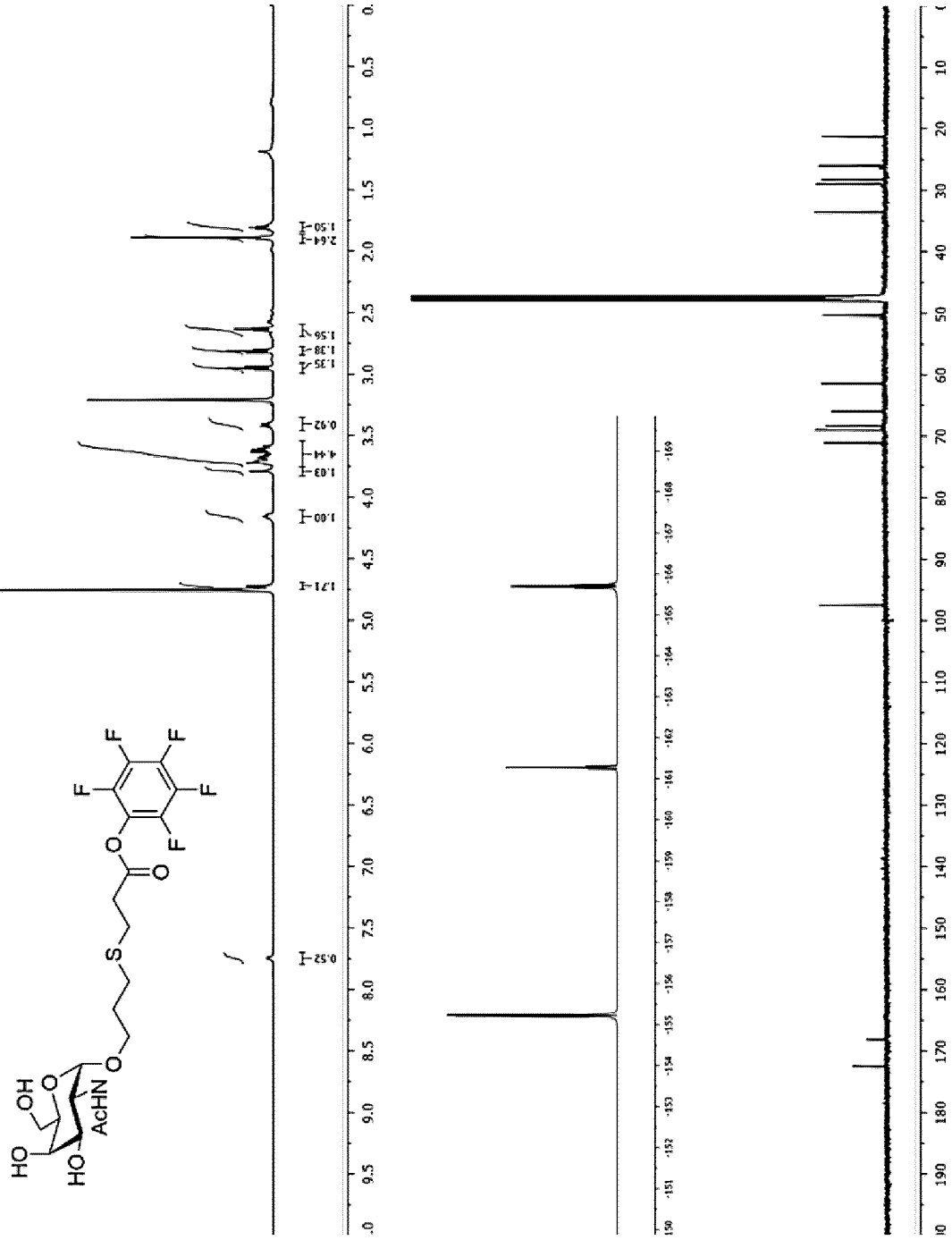
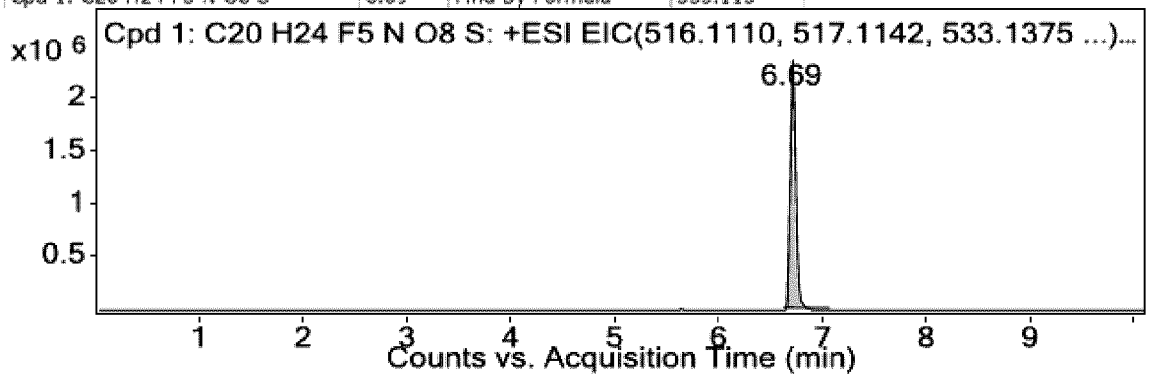


Fig. 11B

Compound Table

Compound Label	RT	Mass	Abund	Formula	Tgt Mass	Diff (ppm)
Cpd 1: C20 H24 F5 N O8 S	6.69	533.115	410940	C20 H24 F5 N O8 S	533.1143	1.29

Compound Label	RT	Algorithm	Mass
Cpd 1: C20 H24 F5 N O8 S	6.69	Find By Formula	533.115



MS Spectrum

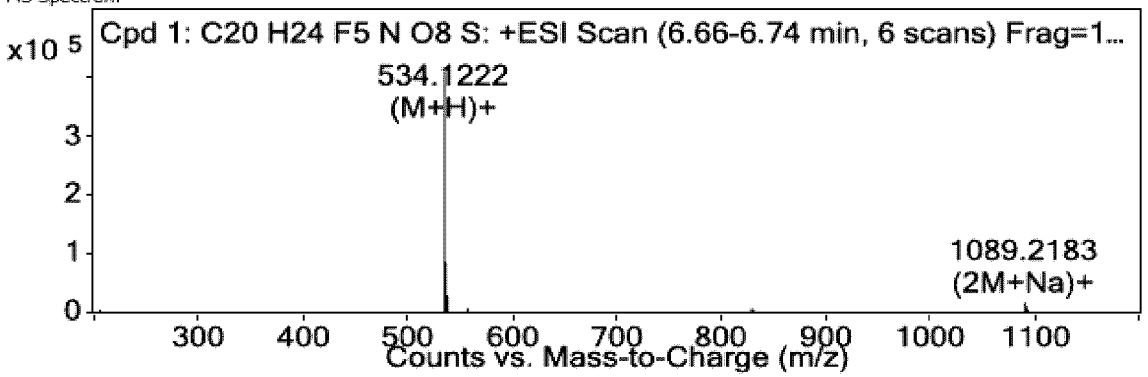


Fig. 12

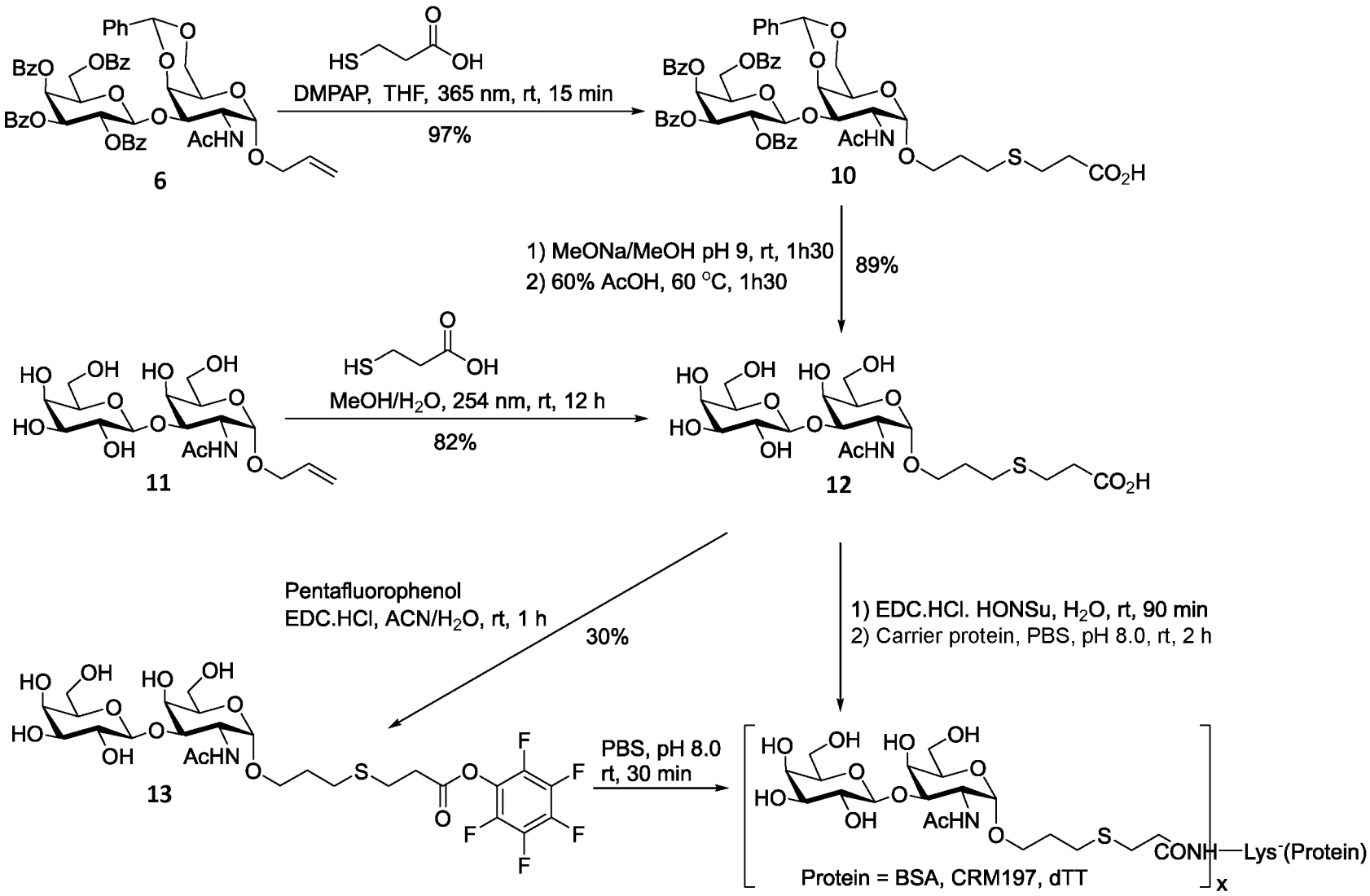

Ph
BzO
OBz
AcHN
6
HS
OH
DMPAP, THF, 365 nm, rt, 15 min
97%
10
CO2H
1) MeONa/MeOH pH 9, rt, 1h30
2) 60% AcOH, 60 °C, 1h30
89%
HO
11
MeOH/H2O, 254 nm, rt, 12 h
82%
12
Pentafluorophenol
EDC.HCl, ACN/H2O, rt, 1 h
30%
1) EDC.HCl. HONSu, H2O, rt, 90 min
2) Carrier protein, PBS, pH 8.0, rt, 2 h
13
F
PBS, pH 8.0
rt, 30 min
CONH—Lys-(Protein)
Protein = BSA, CRM197, dTT
x

Fig. 13

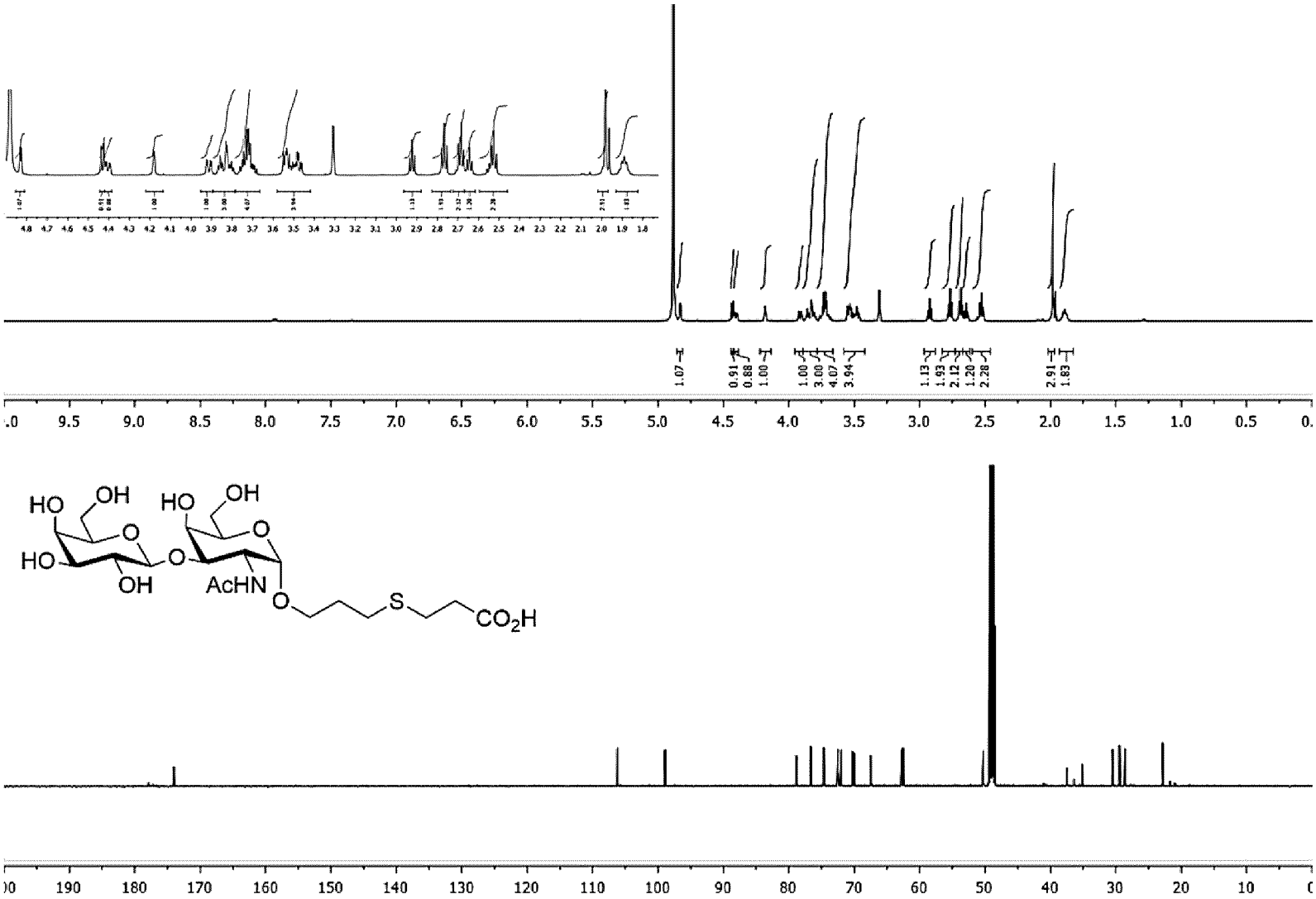


Fig. 14

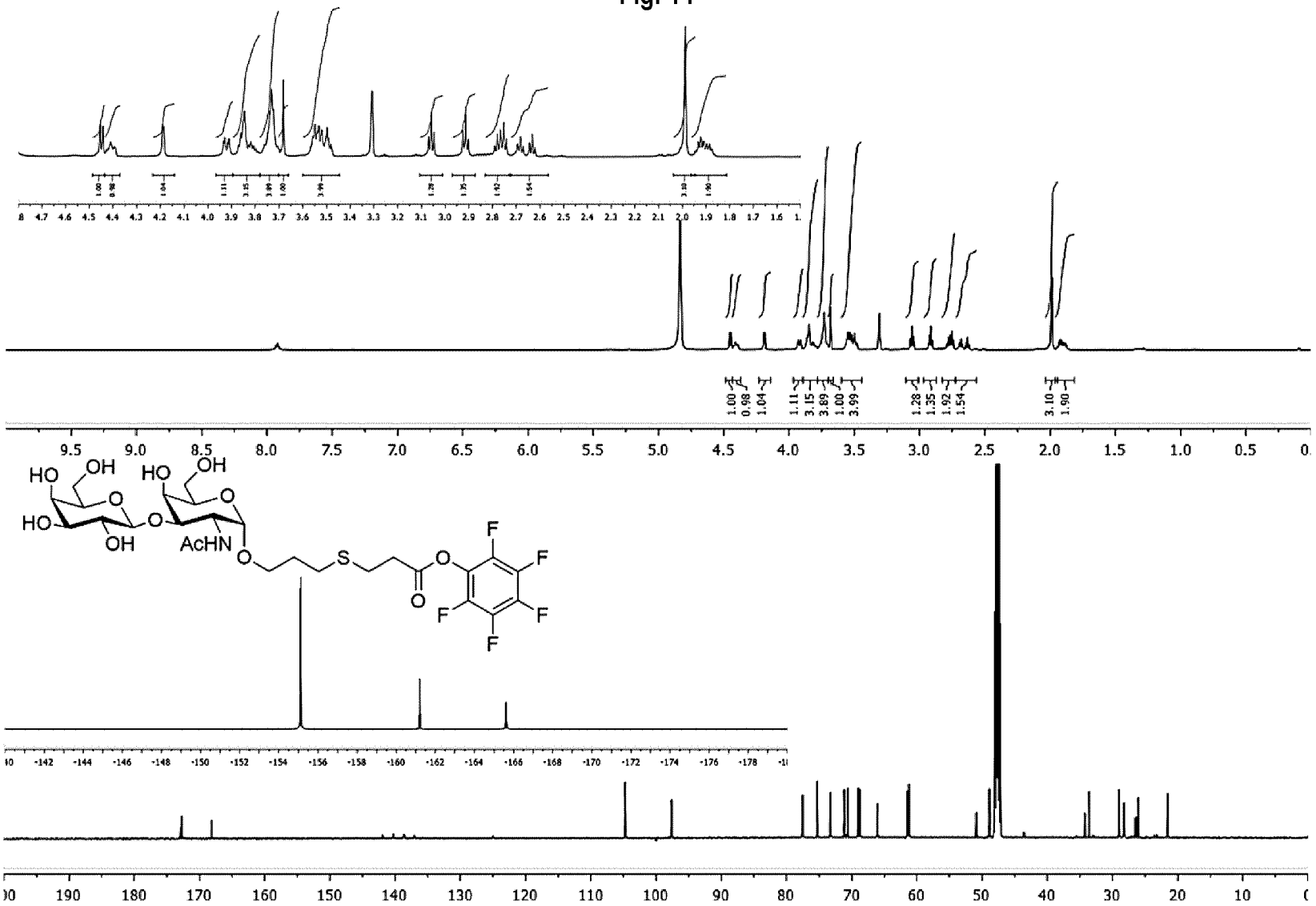


Fig. 15A

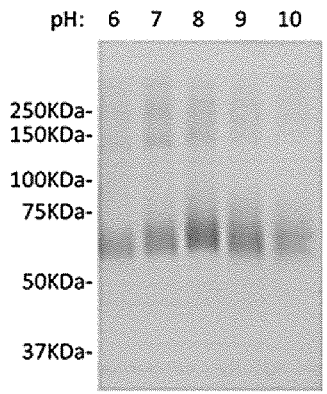


Fig. 15B

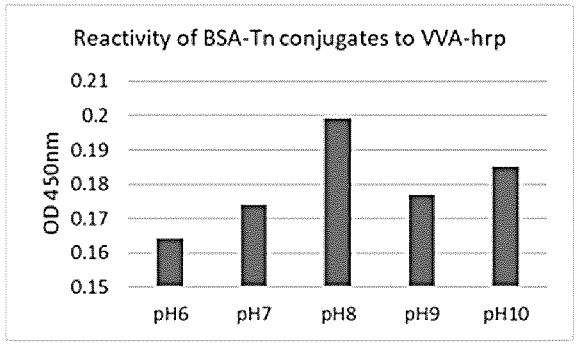


Fig. 15C

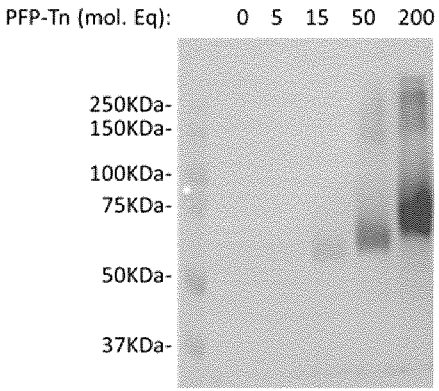


Fig. 15D

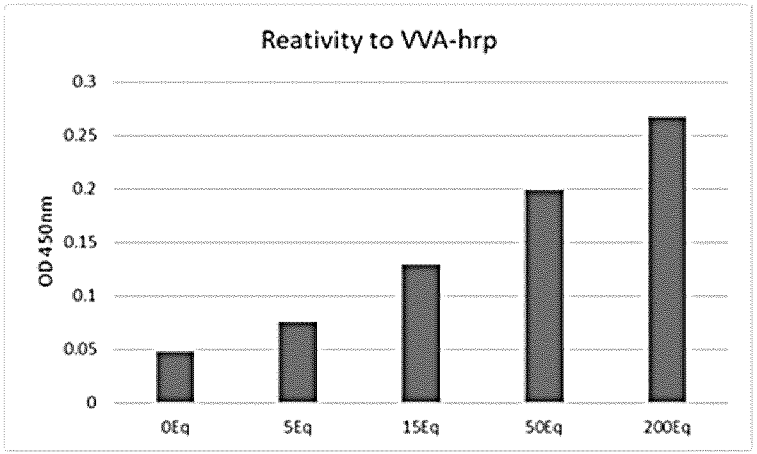


Fig. 15E

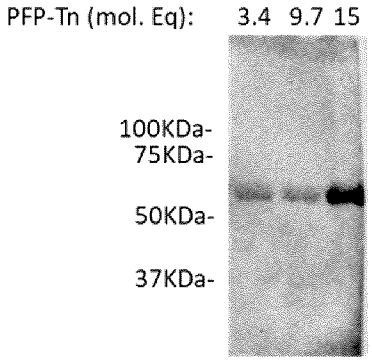


Fig. 16A

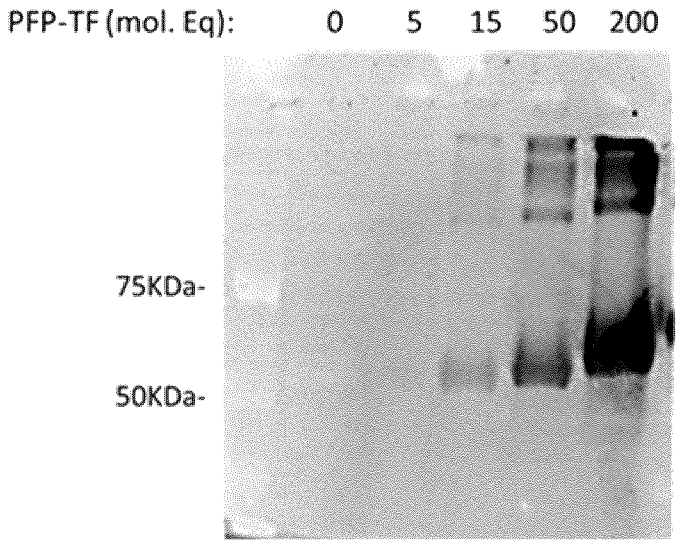


Fig. 16B

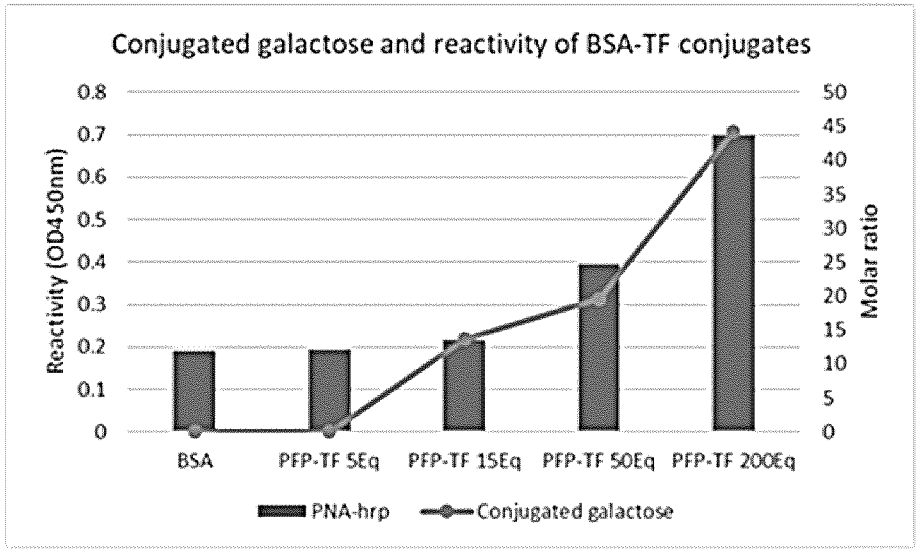


Fig. 17A

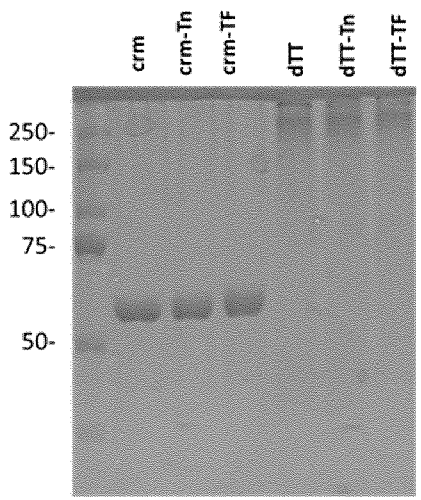


Fig. 17B

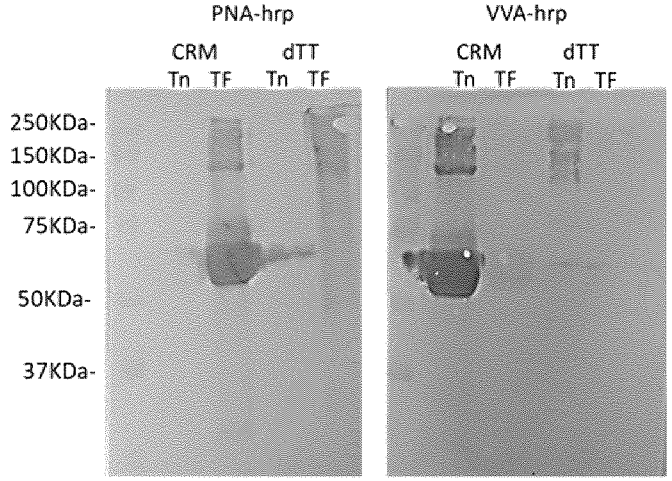


Fig. 17C

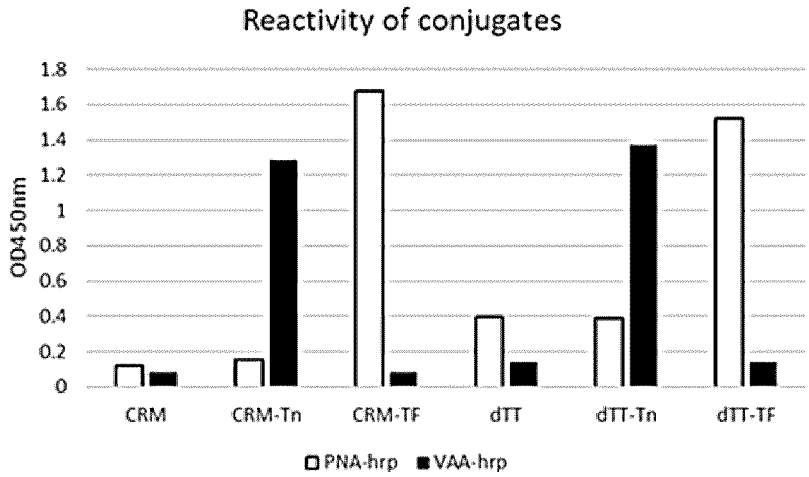


Fig. 18

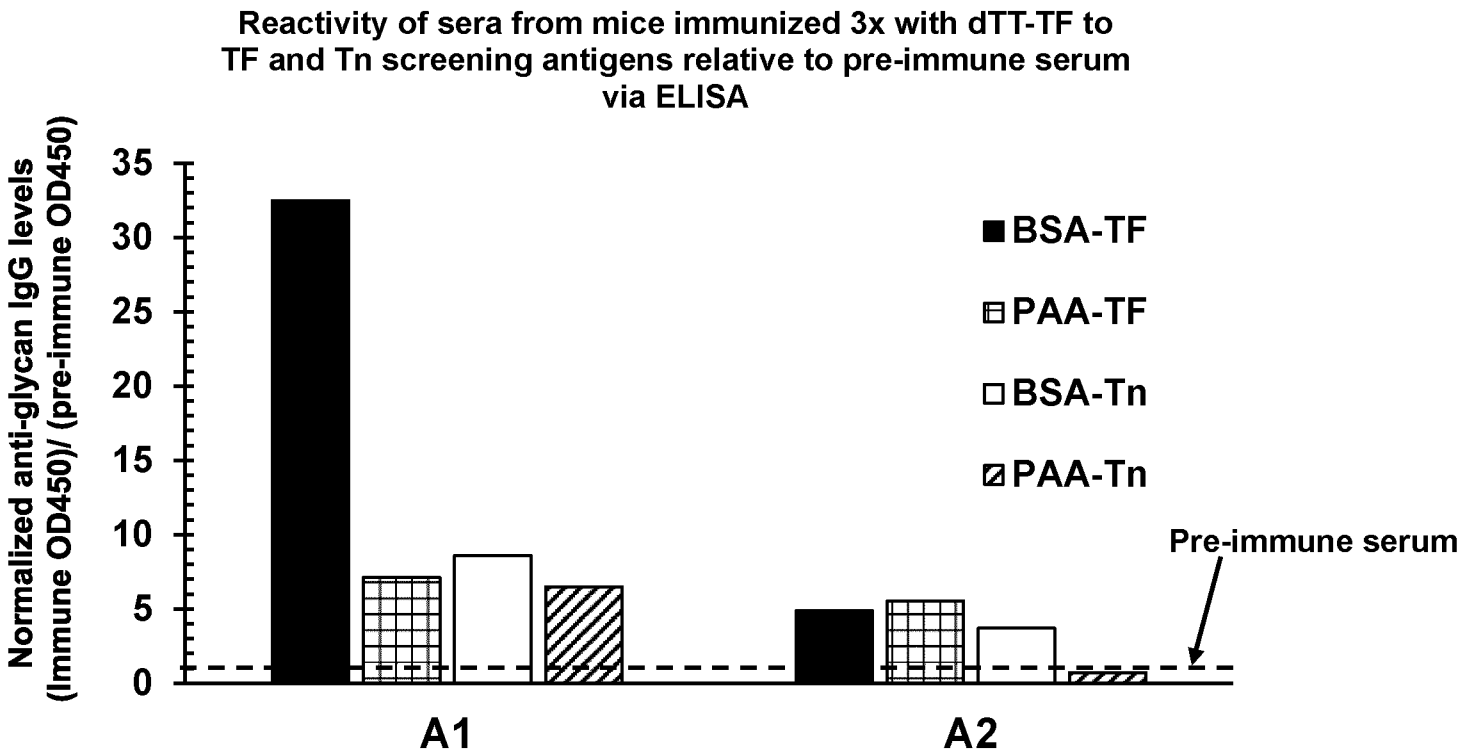

Reactivity of sera from mice immunized 3x with dTT-TF to TF and Tn screening antigens relative to pre-immune serum via ELISA
Normalized anti-glycan IgG levels (Immune OD450)/ (pre-immune OD450)
35
30
25
20
15
10
5
0
BSA-TF
PAA-TF
BSA-Tn
PAA-Tn
Pre-immune serum
A1
A2

Fig. 19A

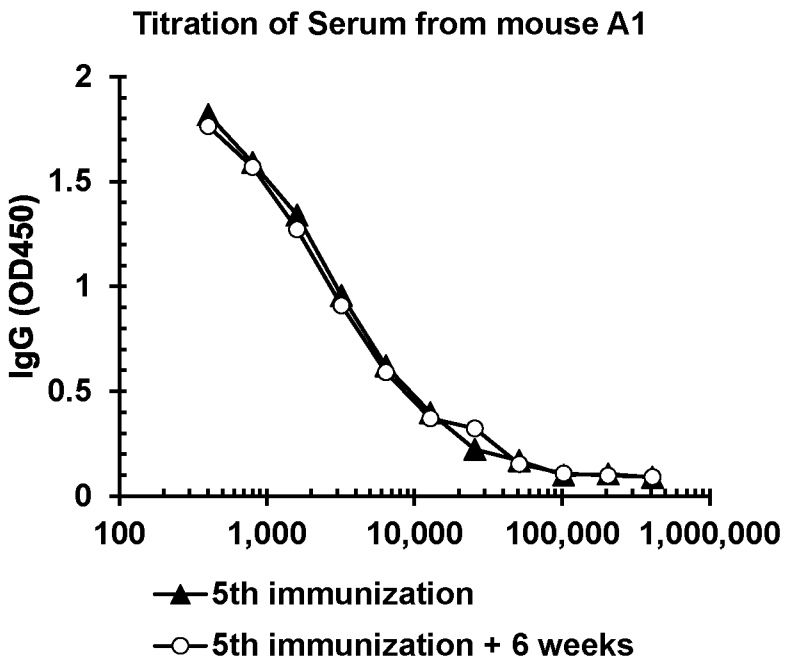


Fig. 19B

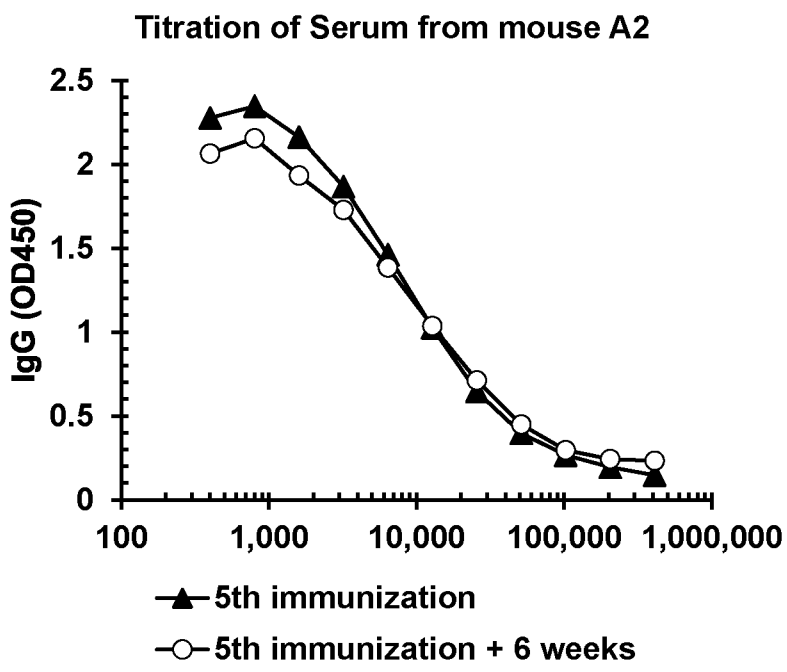


Fig. 20

Relative abundance of all O-glycosylated forms of peptide VQPTESIVR (SEQ ID NO: 3) amongst proteins migrating above 75 kDa

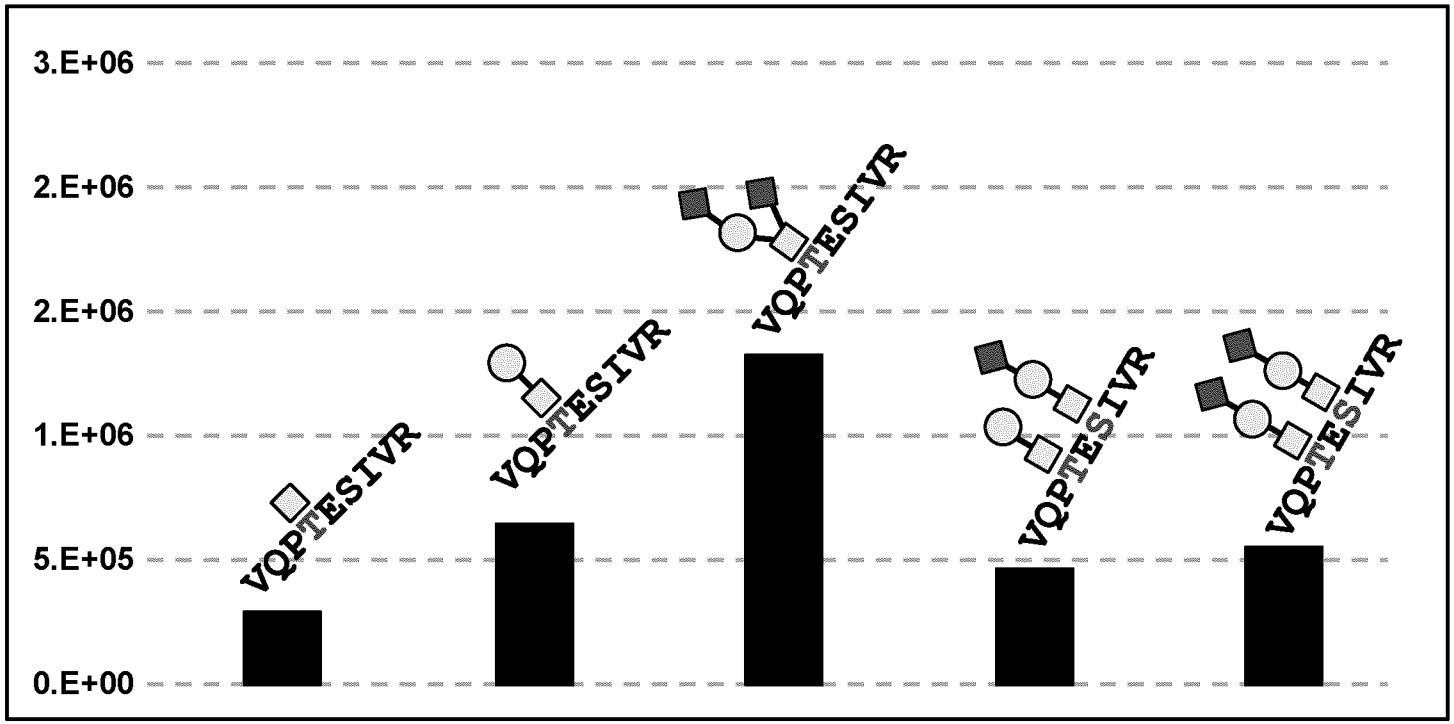


Fig. 21

Relative abundance of all O-glycosylated forms of peptide VQPTESIVR (SEQ ID NO: 3) amongst proteins migrating between 75-100 kDa

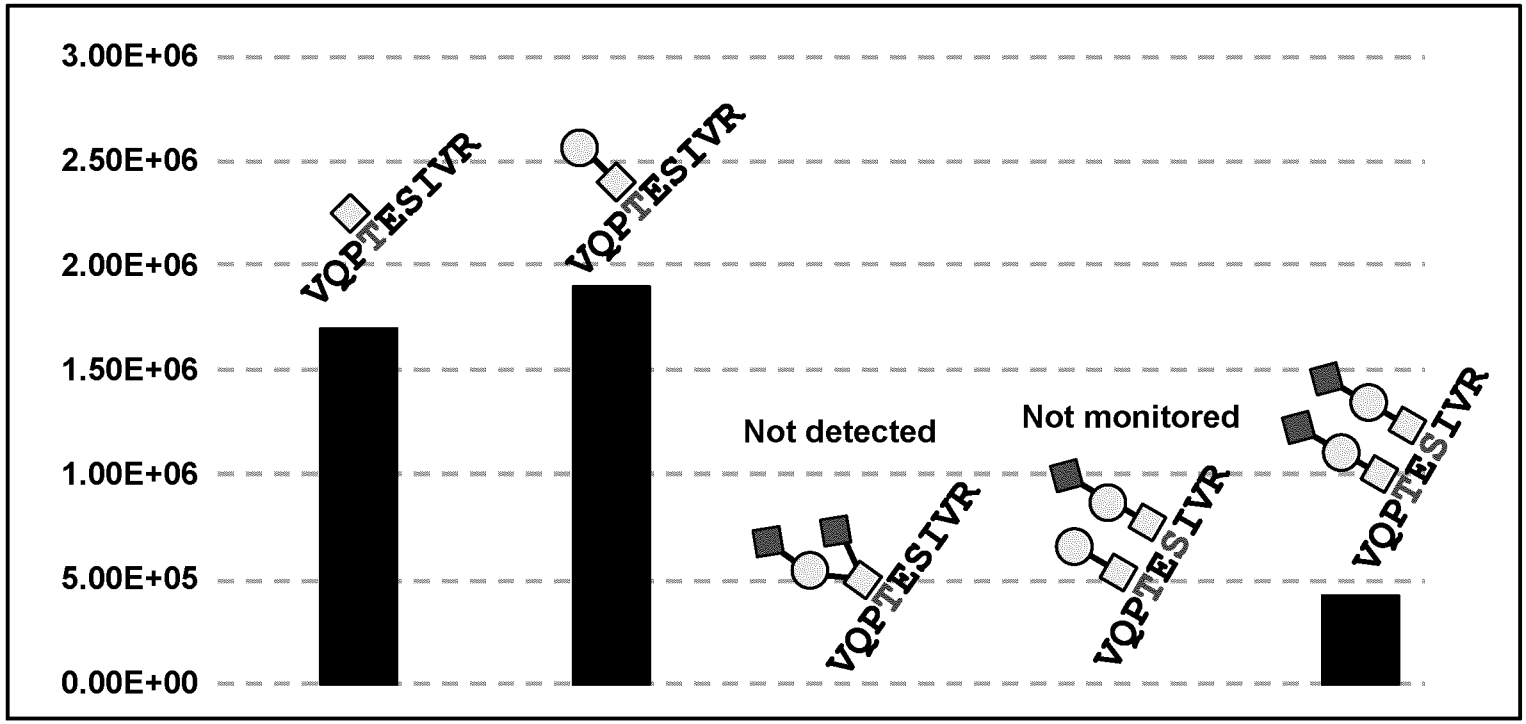


Fig. 22

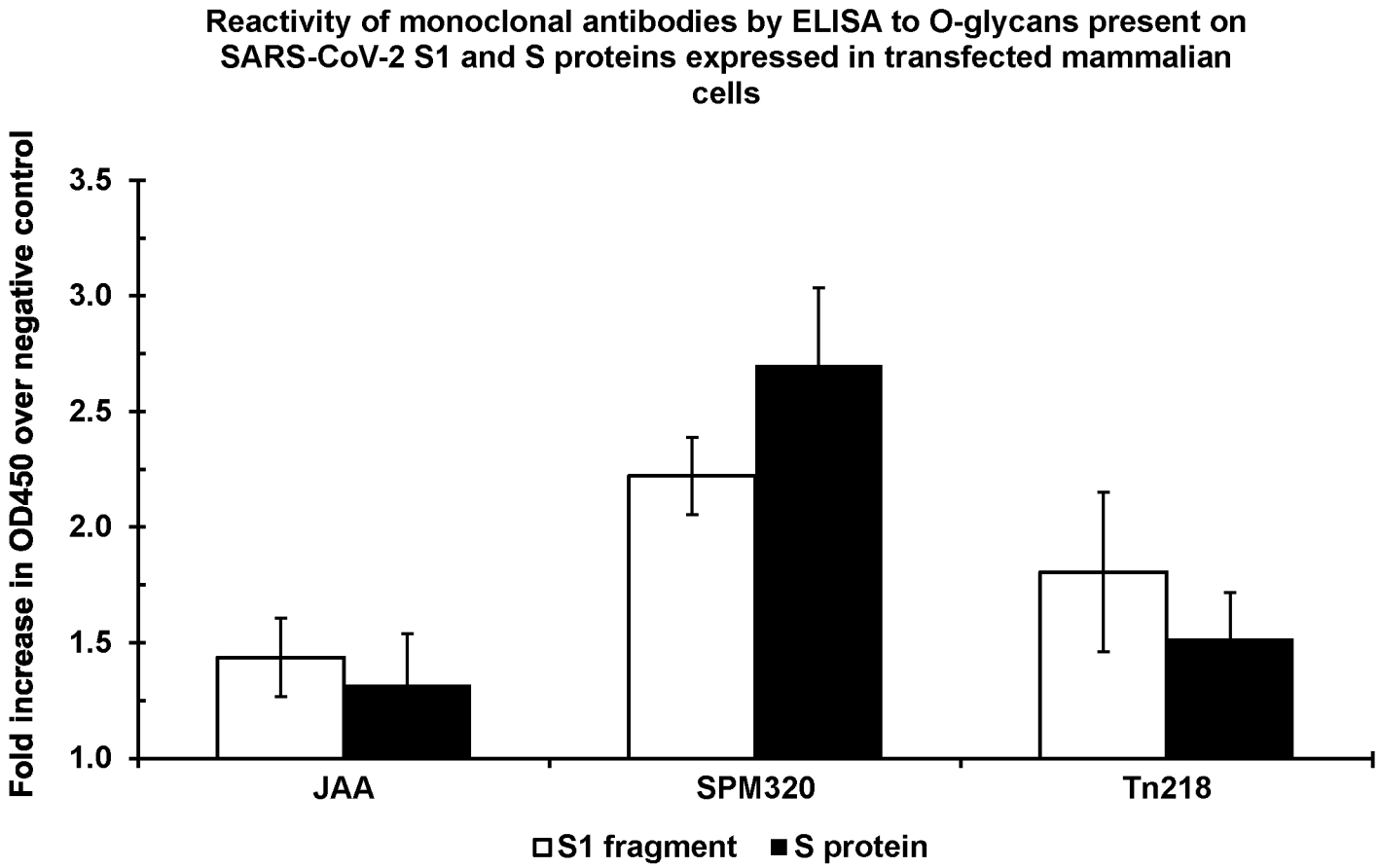

Reactivity of monoclonal antibodies by ELISA to O-glycans present on SARS-CoV-2 S1 and S proteins expressed in transfected mammalian cells
Fold increase in OD450 over negative control
3.5
3.0
2.5
2.0
1.5
1.0
JAA
SPM320
Tn218
S1 fragment
S protein

Fig. 23

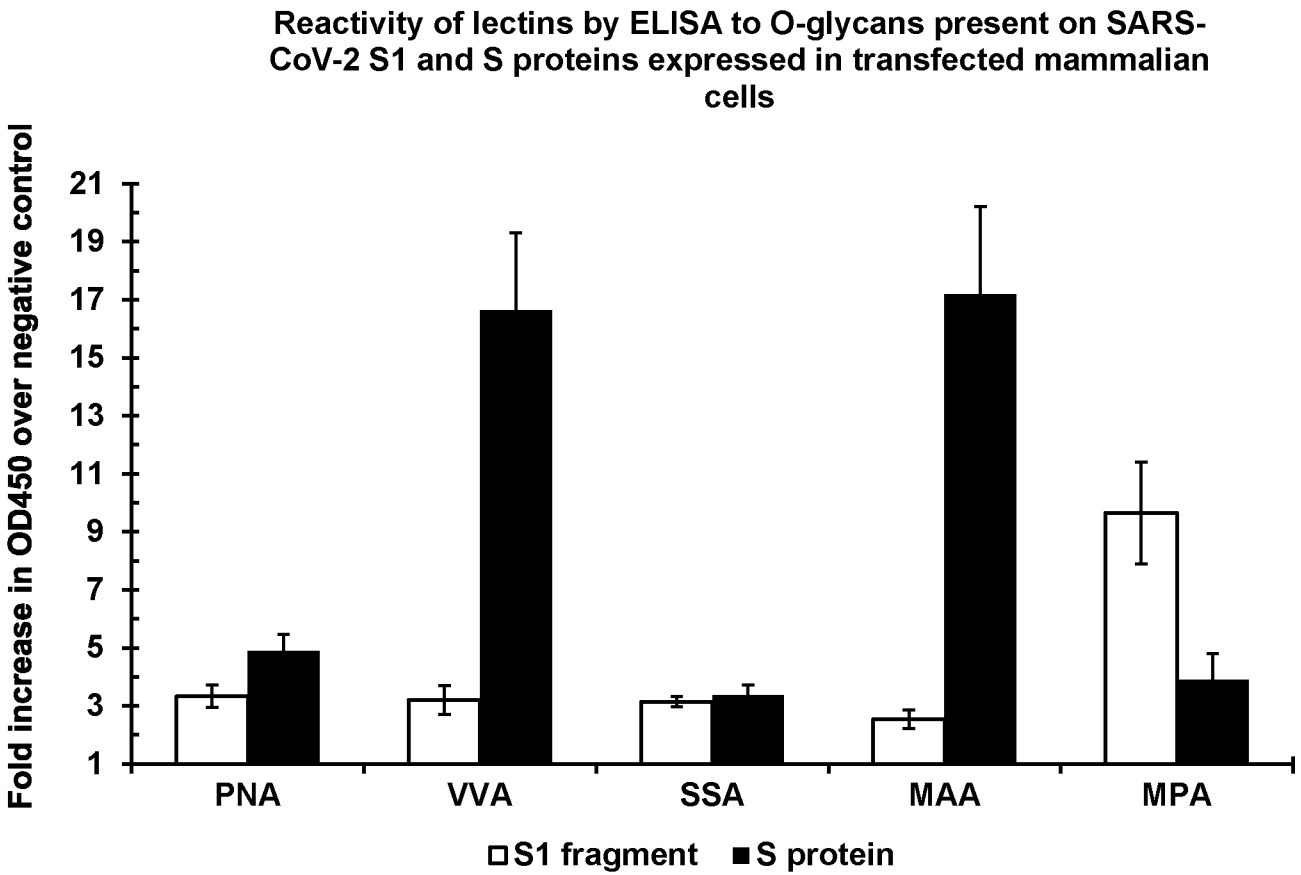
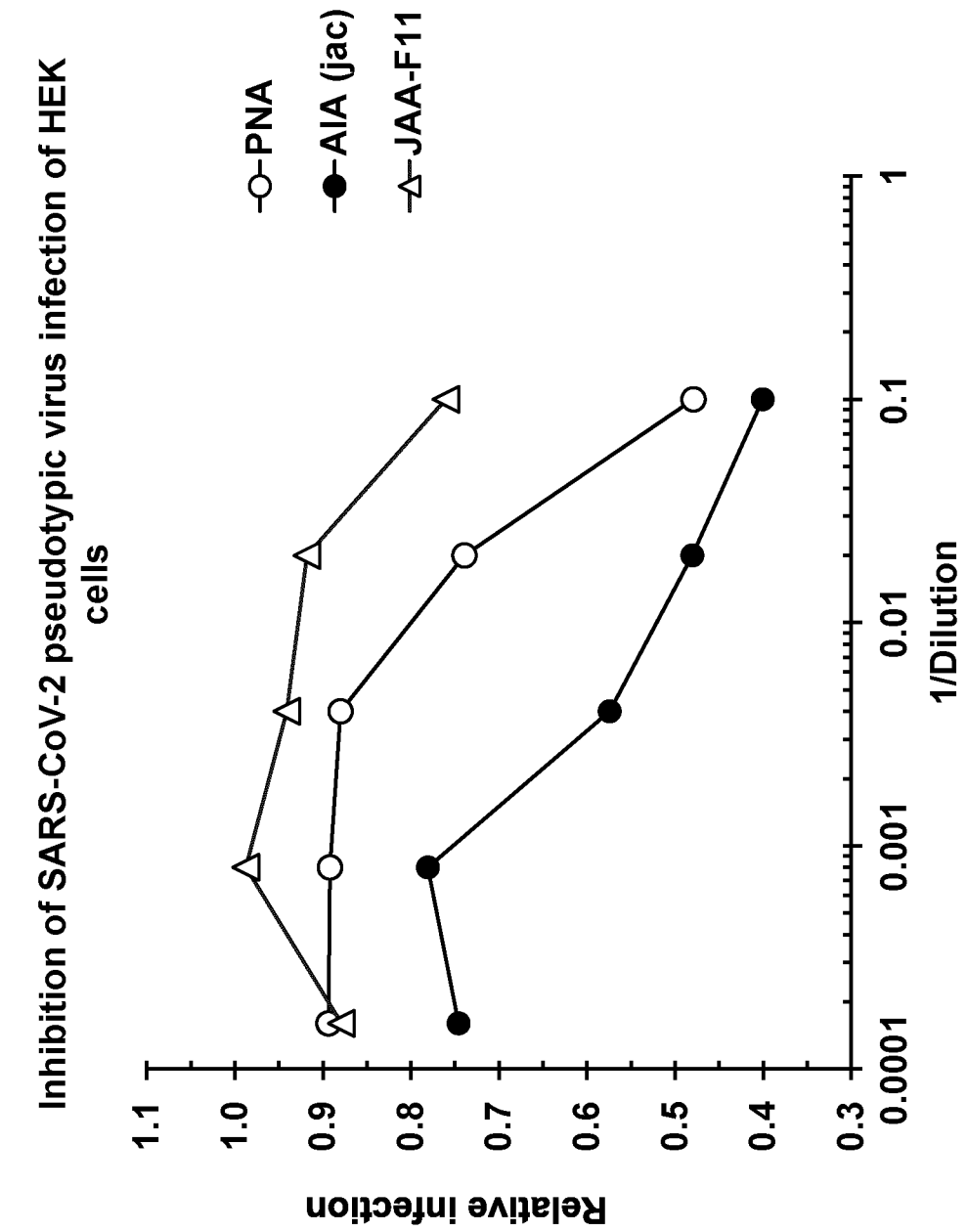

Reactivity of lectins by ELISA to O-glycans present on SARS-CoV-2 S1 and S proteins expressed in transfected mammalian cells
Fold increase in OD450 over negative control
21
19
17
15
13
11
9
7
5
3
1
PNA
VVA
SSA
MAA
MPA
S1 fragment
S protein

Fig. 24


Inhibition of SARS-CoV-2 pseudotypic virus infection of HEK cells
PNA
AIA (jac)
JAA-F11
Relative infection
1.1
1.0
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.0001
0.001
0.01
0.1
1
1/Dilution