

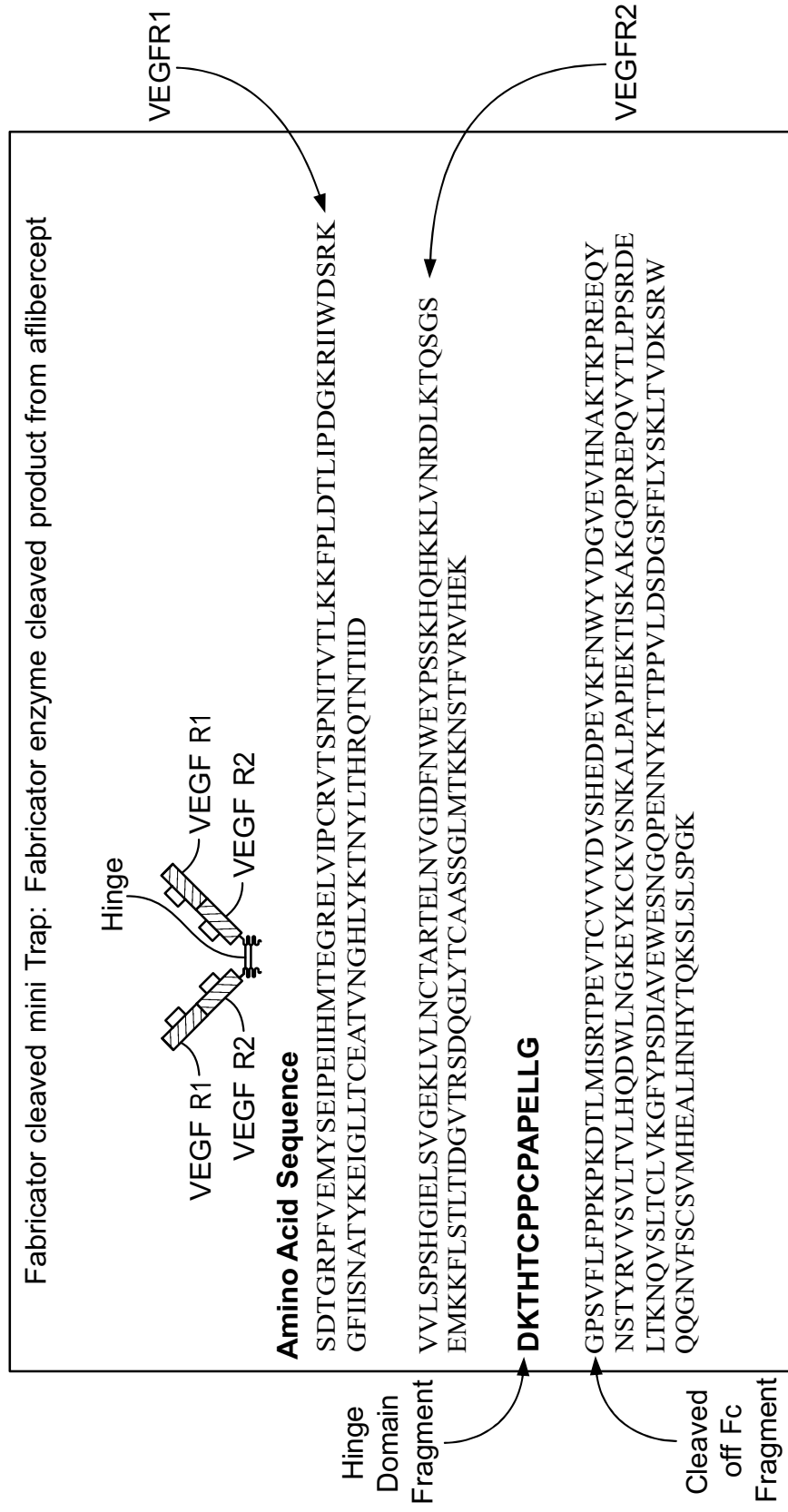


FIG.1

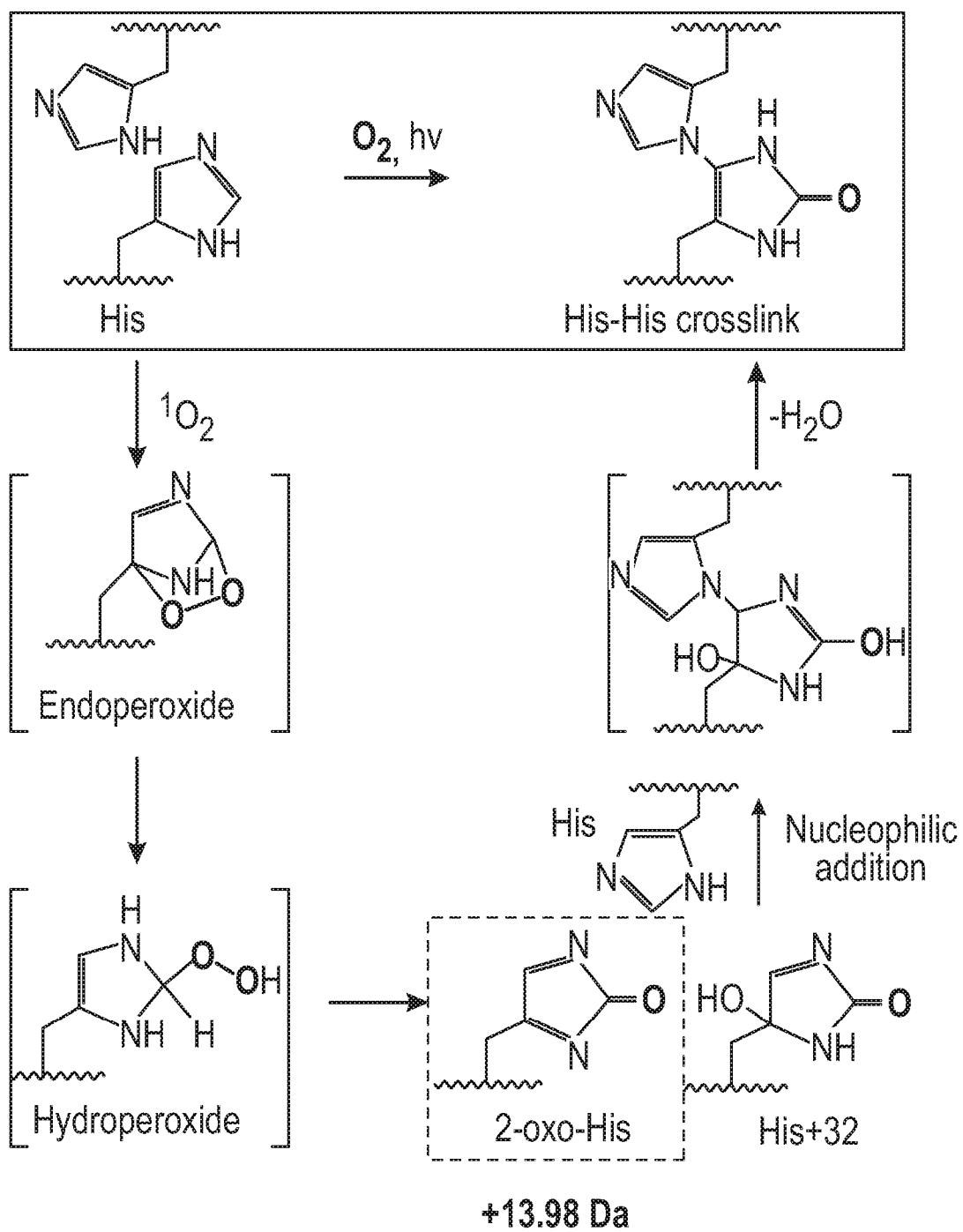
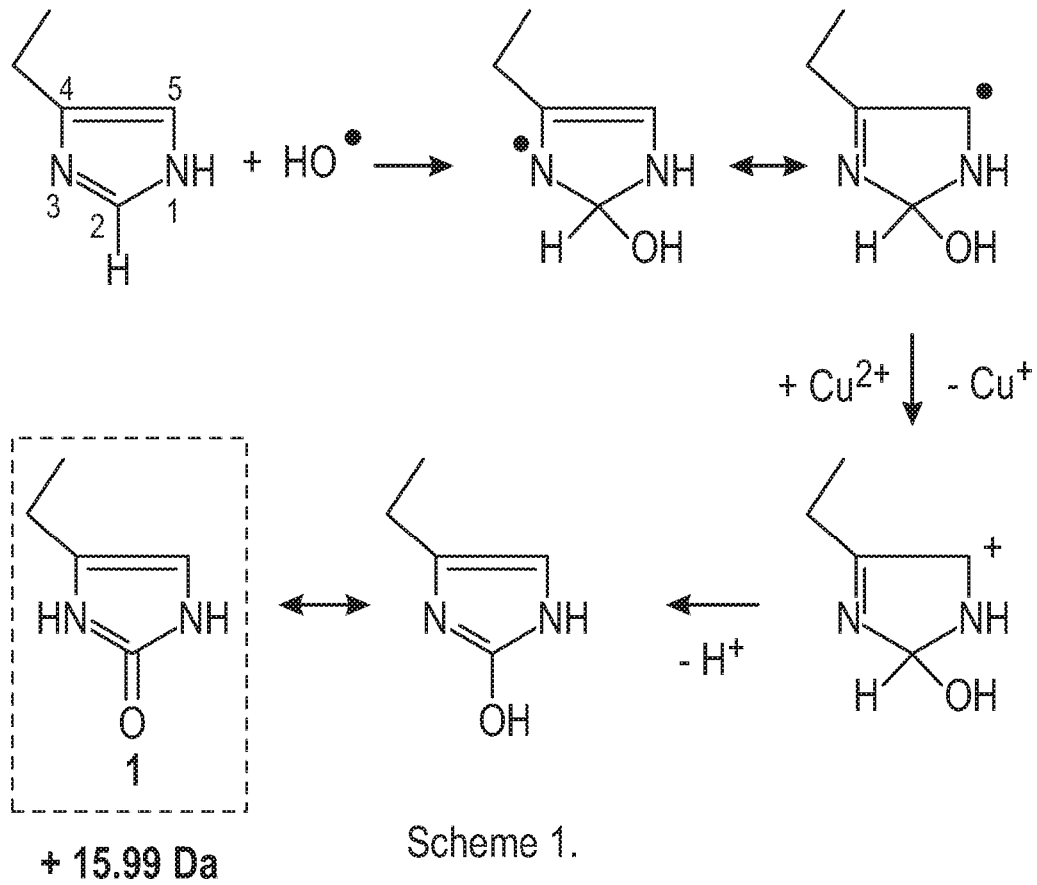


FIG. 2

3/60



Scheme 1.

FIG. 3

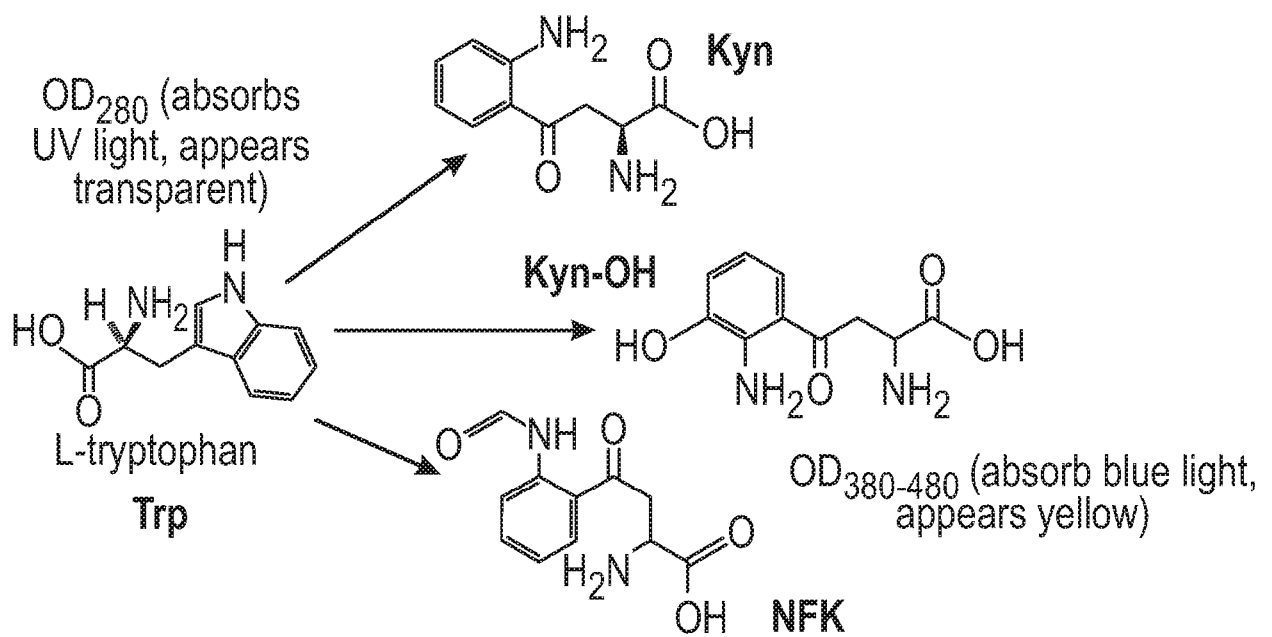
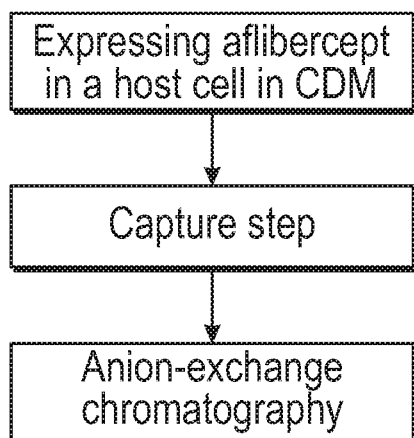
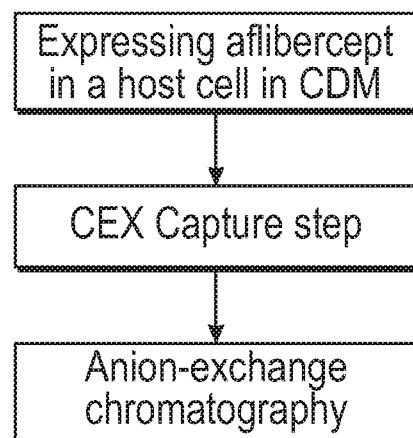
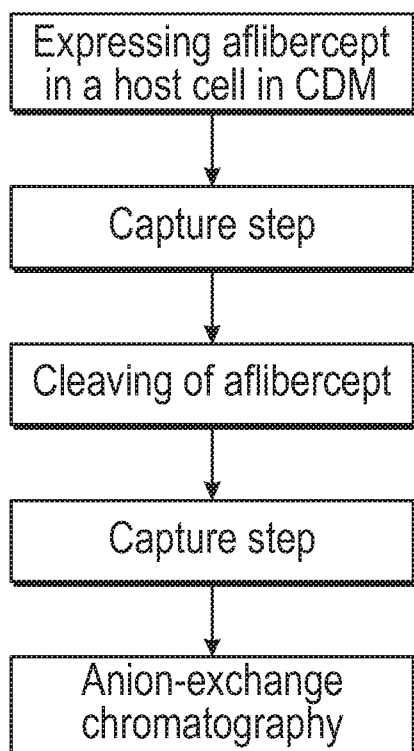
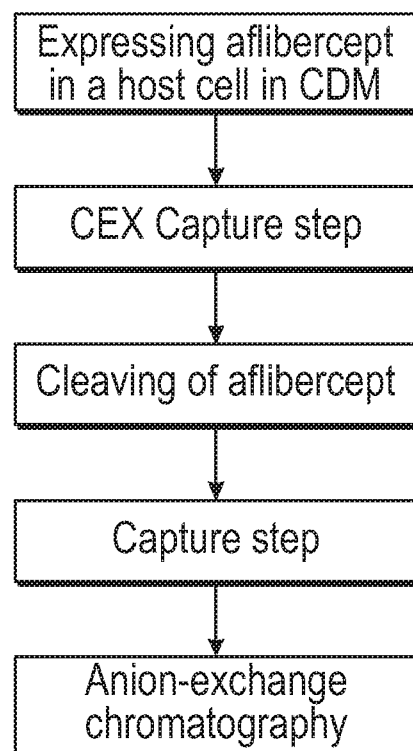


FIG. 4

**FIG. 5****FIG. 7****FIG. 6****FIG. 8**

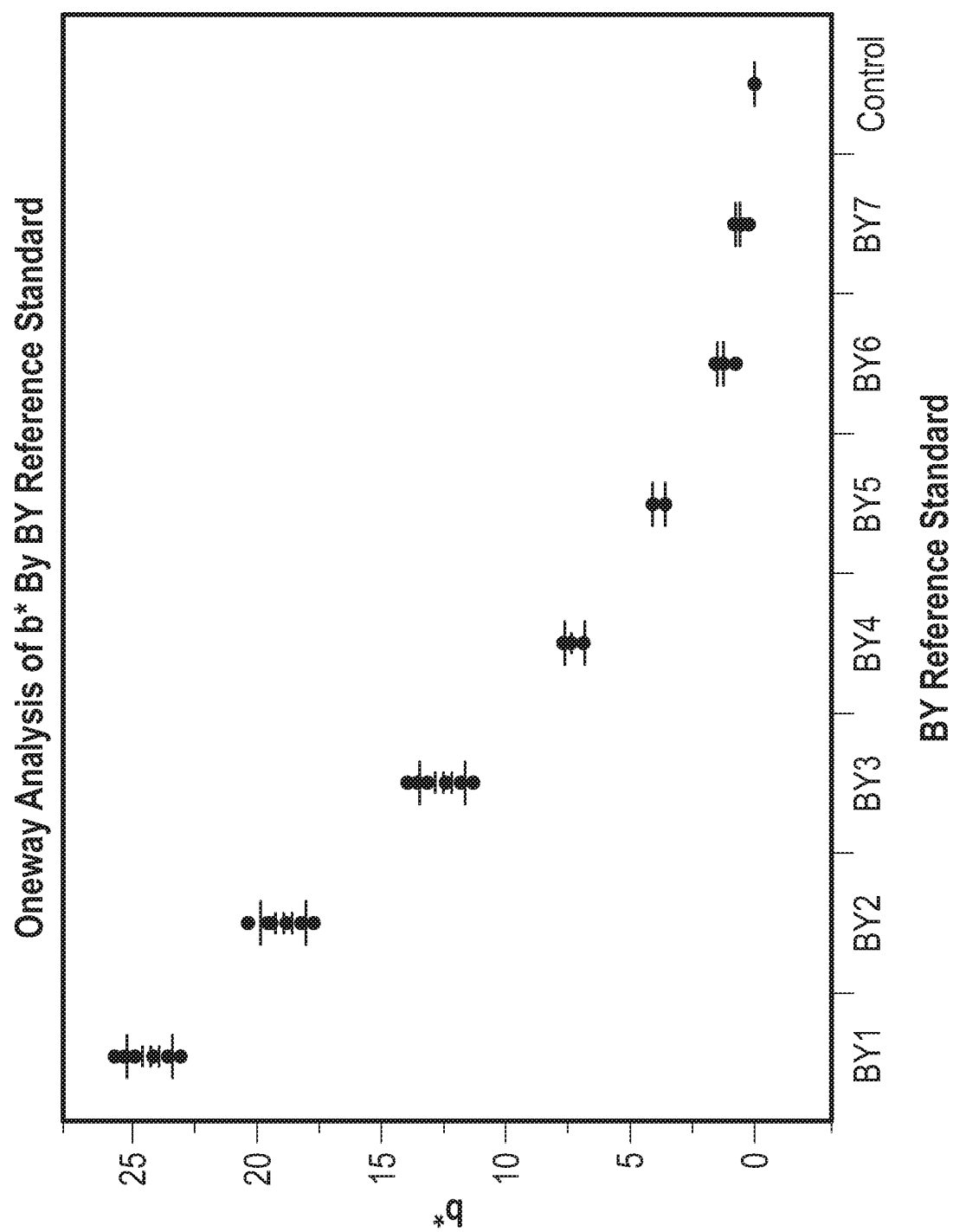


FIG. 9

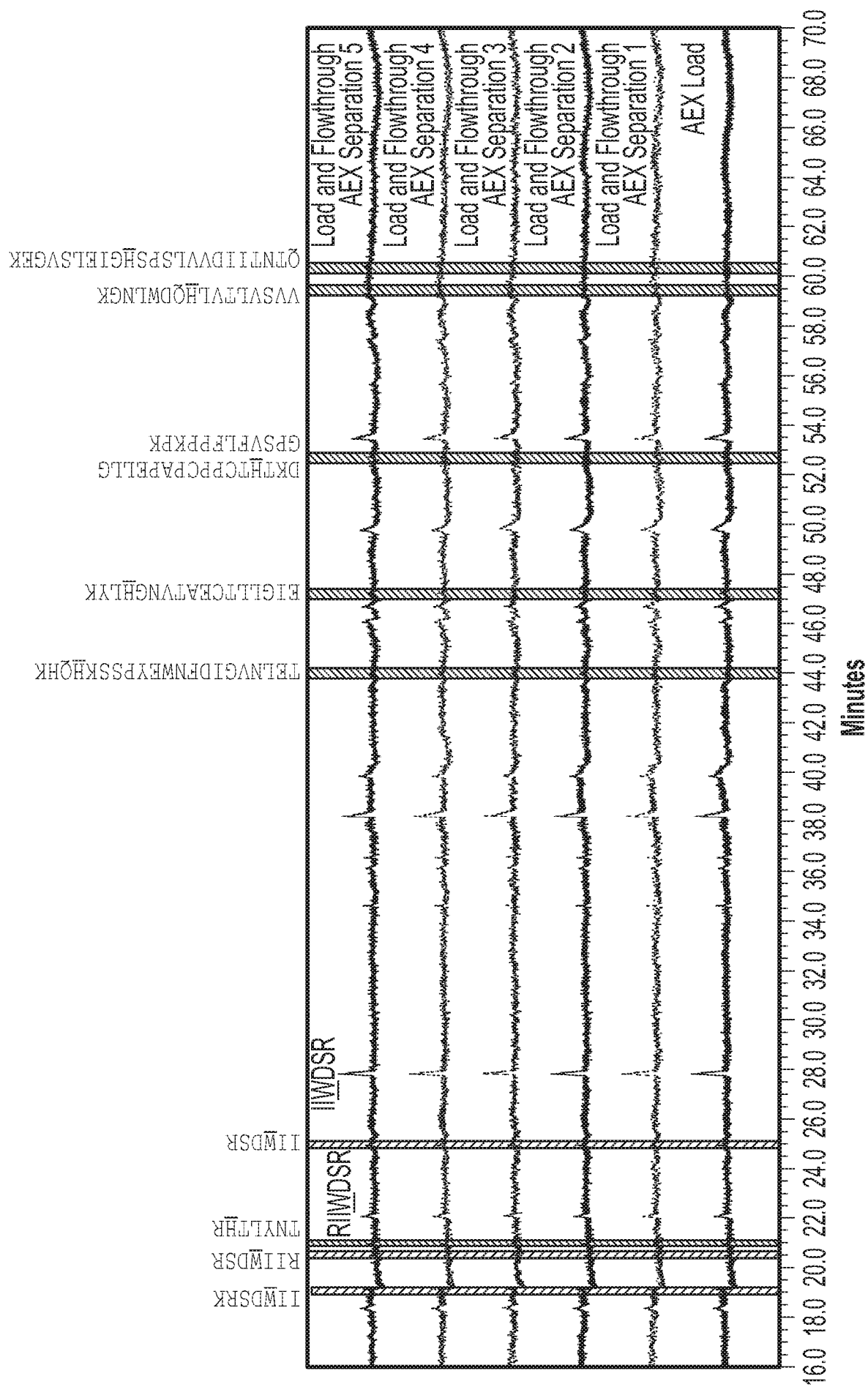


FIG. 10

PTM	Site	Peptide Sequence	Modification	Afilibercept AEX Pool					
				Afilibercept AEX Load	POROS 50 HQ pH 8.4 2.0 mS/cm	QsFF pH 8.0 2.5 mS/cm	POROS 50 HQ pH 8.0 2.5 mS/cm	QsFF pH 7.8 4.0 mS/cm	POROS 50 HQ pH 7.8 4.0 mS/cm
His Oxidation					AEX Separation 1	AEX Separation 2	AEX Separation 3	AEX Separation 4	AEX Separation 5
				3.06	1.05	0.96	1.33	1.35	1.55
	H19	SDTGRPFVEMYSEIPEIIHMTEGR	+14	<0.01%	<0.01%	<0.01%	<0.01%	<0.01%	<0.01%
	H86	EIGLLTC*EATVNGHLYK	+14	0.013%	0.006%	0.007%	0.008%	0.009%	0.010%
	H95	TNYLTHR	+14	<0.01%	<0.01%	<0.01%	<0.01%	<0.01%	<0.01%
	H110	QTNTIIDVVLSPSHGIELSVGEK	+14	<0.01%	<0.01%	<0.01%	<0.01%	<0.01%	<0.01%
	H145	TELVNGIDFNWEYPSSKHQHK	+14	0.067%	0.039%	0.033%	0.035%	0.038%	0.042%
	H209	THTC*PPC*PAPELLGGPSVFLFPKPK	+14	0.056%	0.027%	0.037%	0.040%	0.045%	0.048%
Trp Oxidation	H295	VVSVLTVLHQDWLNGK	+14	<0.01%	<0.01%	<0.01%	<0.01%	<0.01%	<0.01%
			+4	0.012%	0.008%	0.007%	0.009%	0.009%	0.011%
			+16	0.027%	0.017	0.022%	0.021%	0.021%	0.023%
			+32	0.114%	0.102%	0.111%	0.121%	0.114%	0.135%
			+48	<0.01%	<0.01%	<0.01%	<0.01%	<0.01%	<0.01%
			+4	0.069%	0.055%	0.040%	0.036	0.039%	0.038%
			+16	0.131%	0.078%	0.070%	0.089	0.077%	0.081%
			+32	0.086%	0.057%	0.040%	0.059%	0.046%	0.068%

*: ALKYLATED CYSTEINE

FIG. 11

8/60

Full View

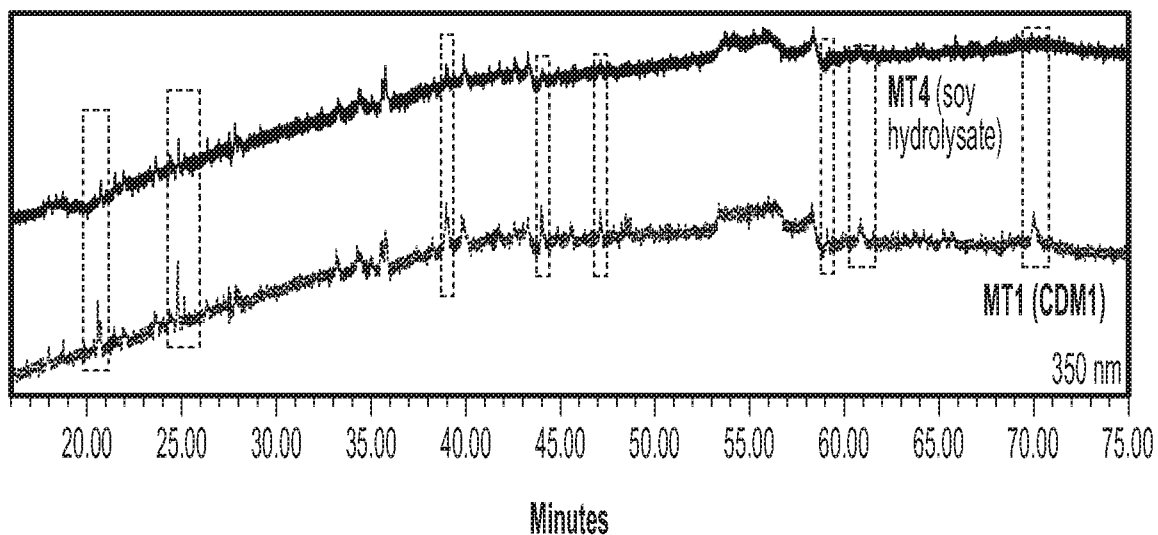


FIG. 12A

16-30 min

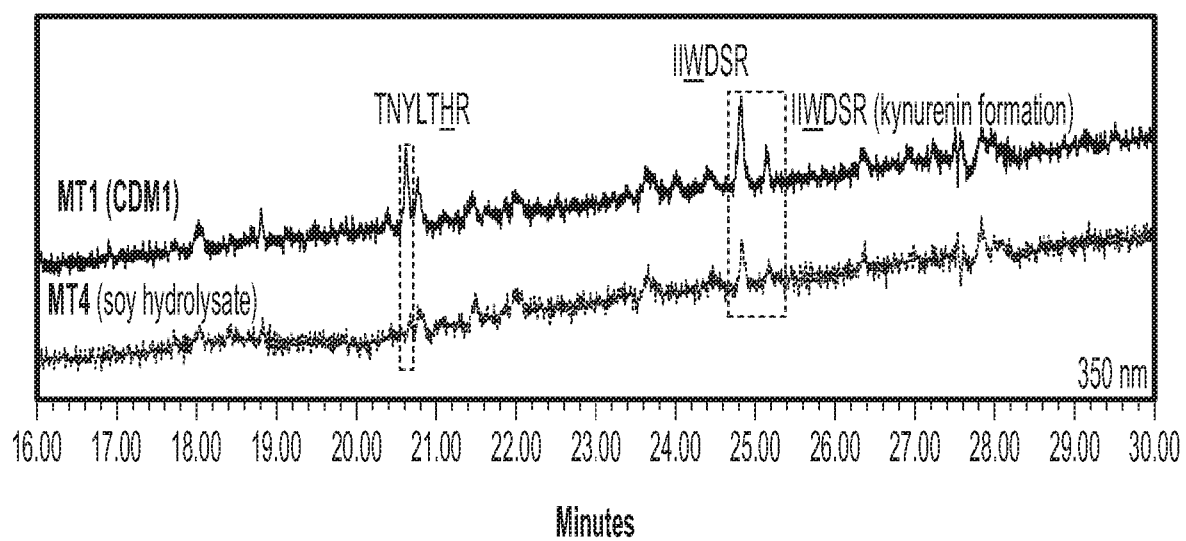


FIG. 12B

9/60

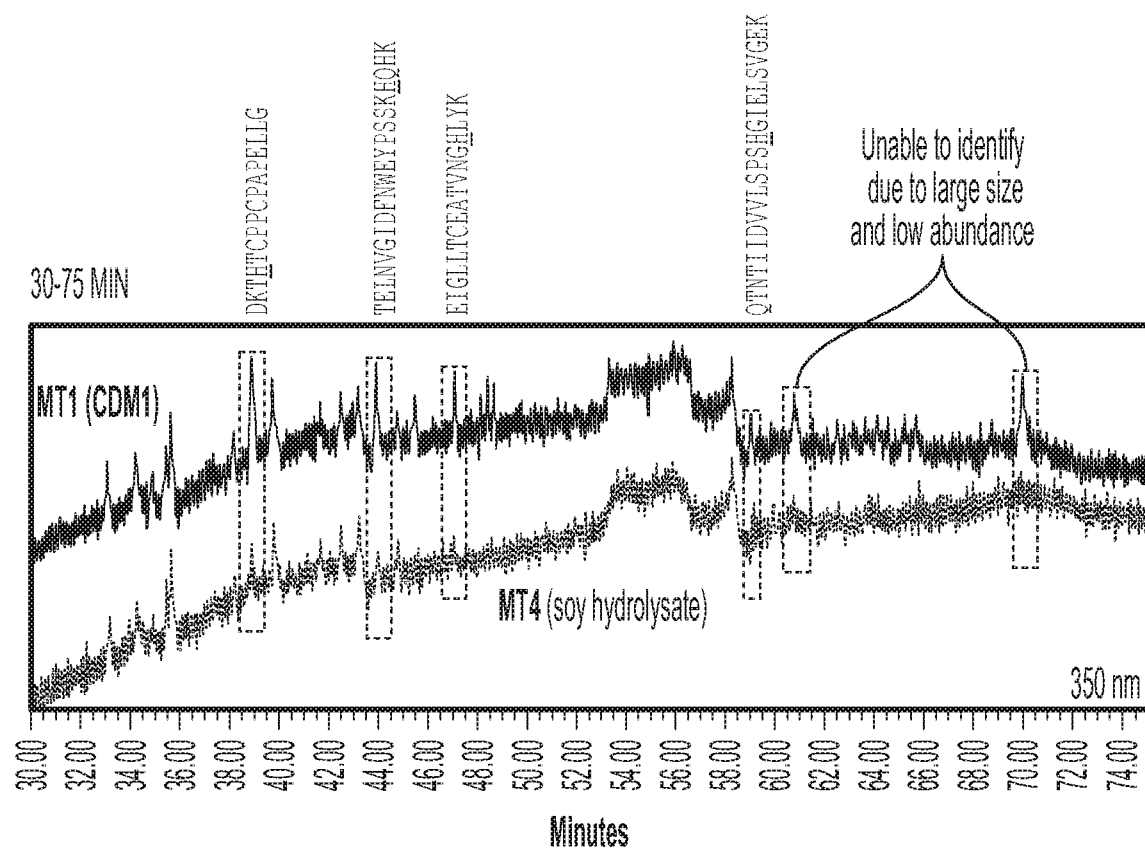


FIG. 12C

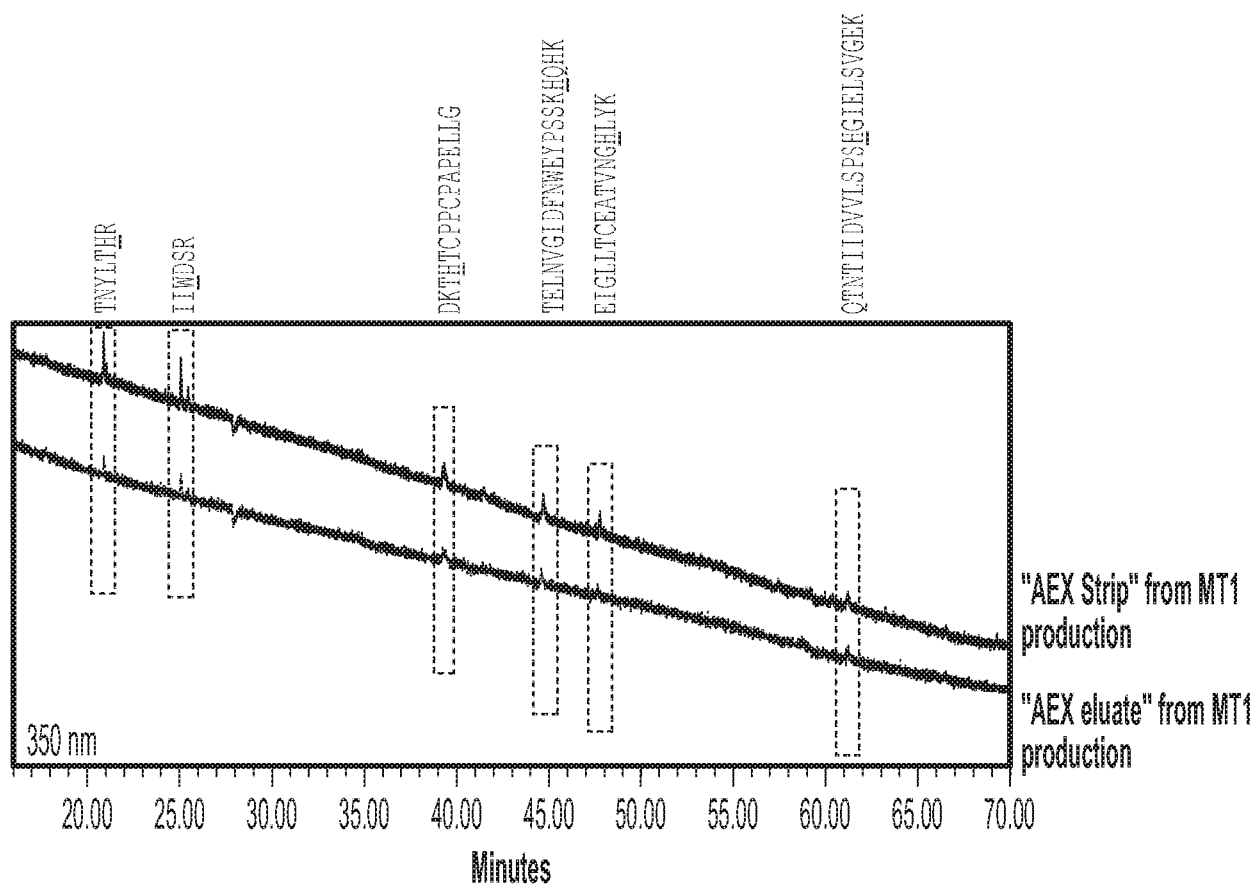


FIG. 13

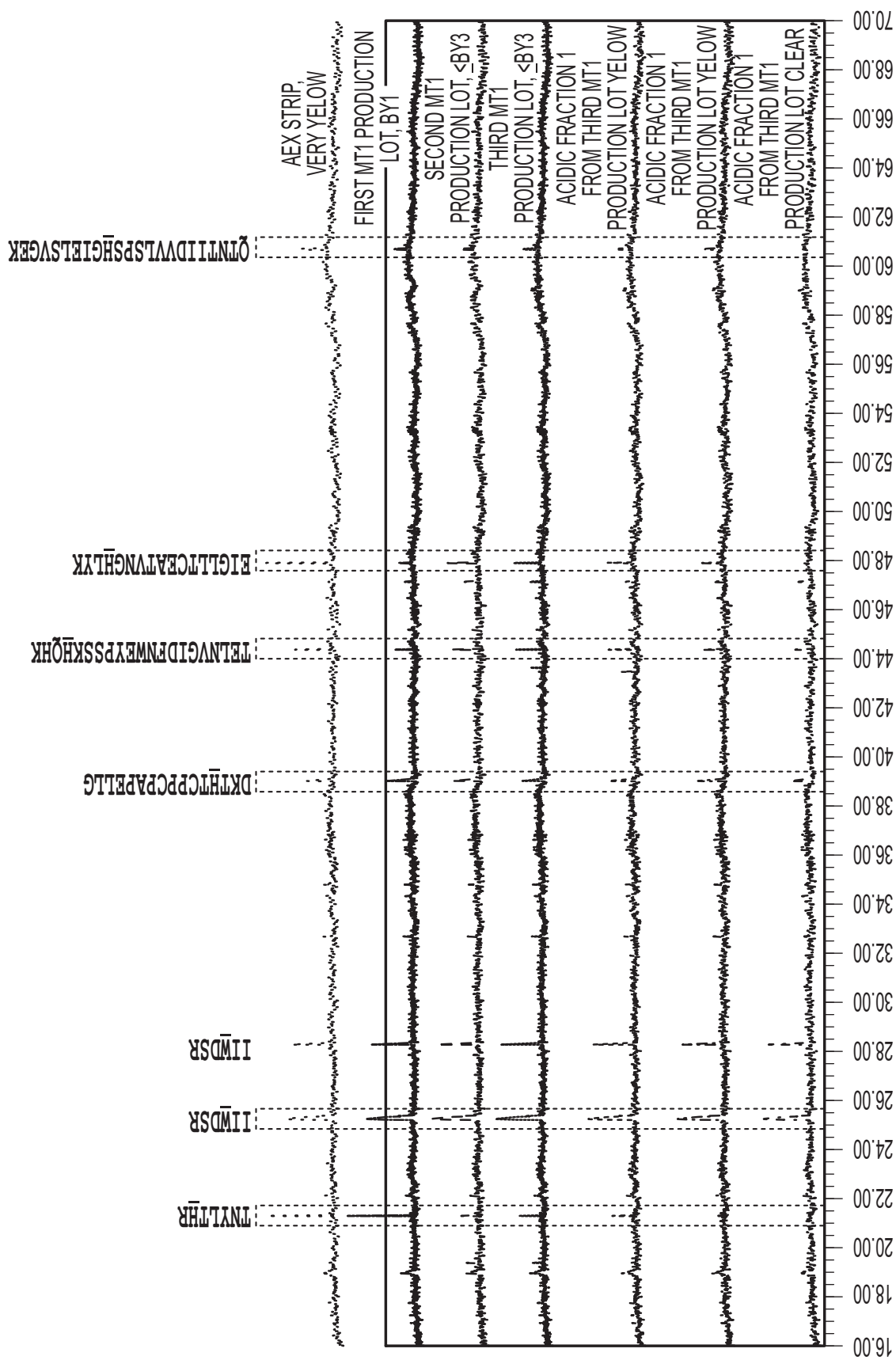


FIG. 14

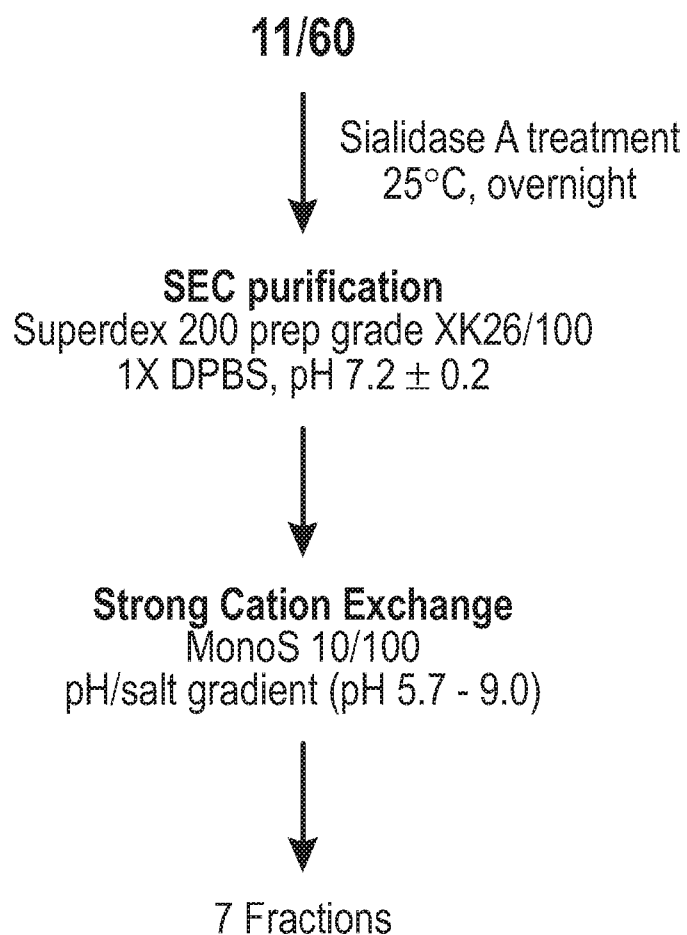


FIG. 15

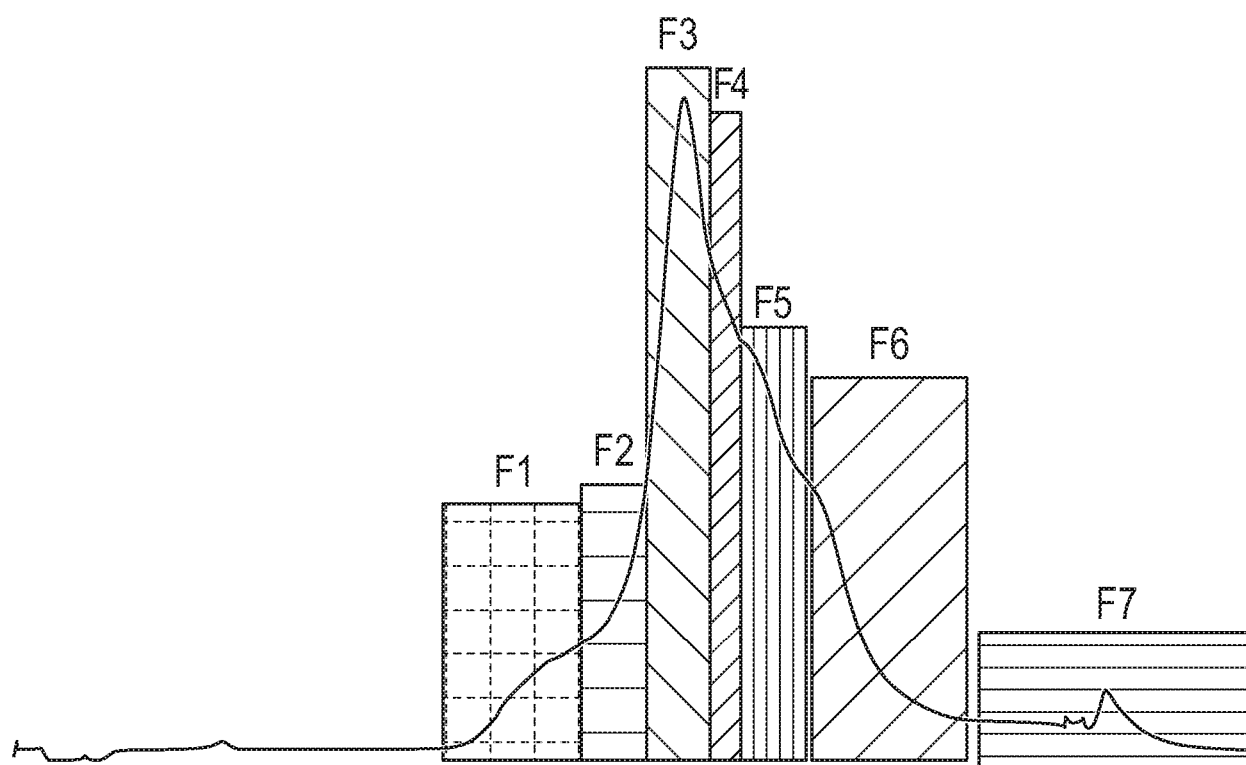


FIG. 16

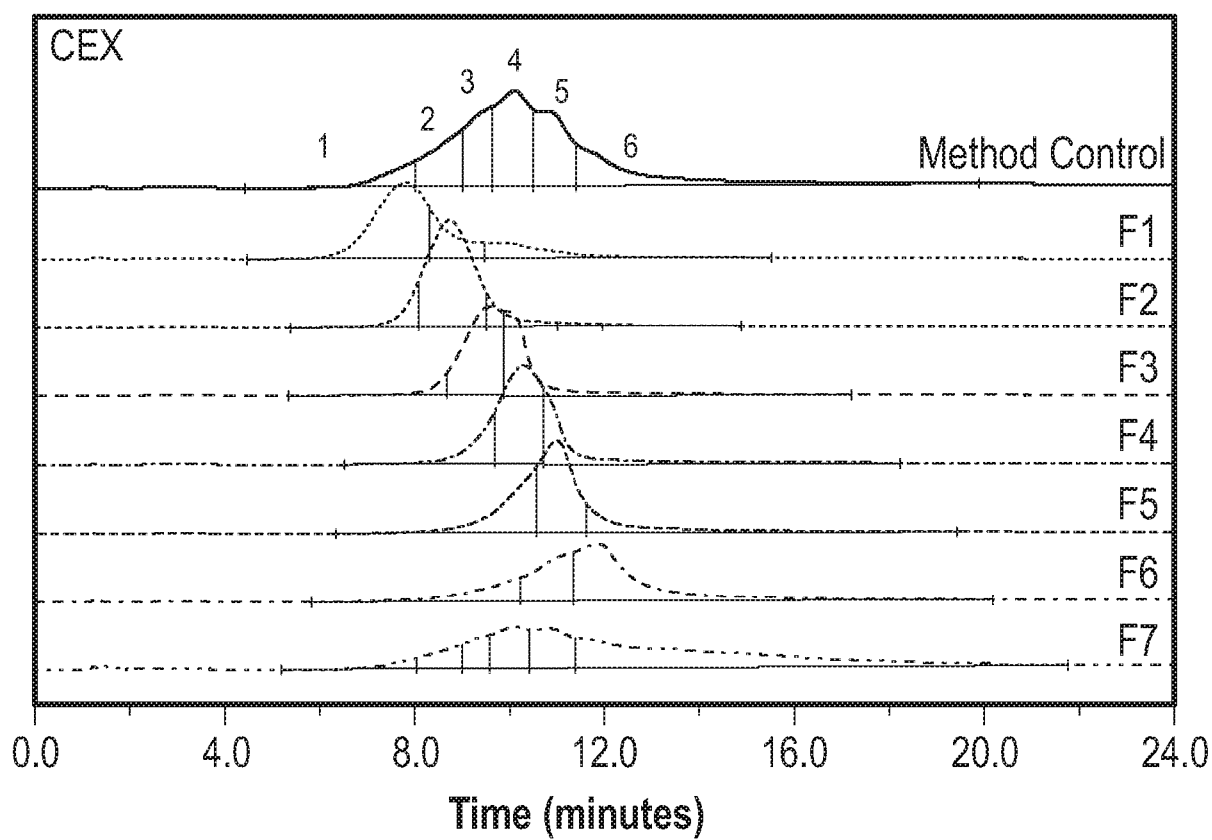


FIG. 17

13/60

MT1 Unfractionated Parental Control

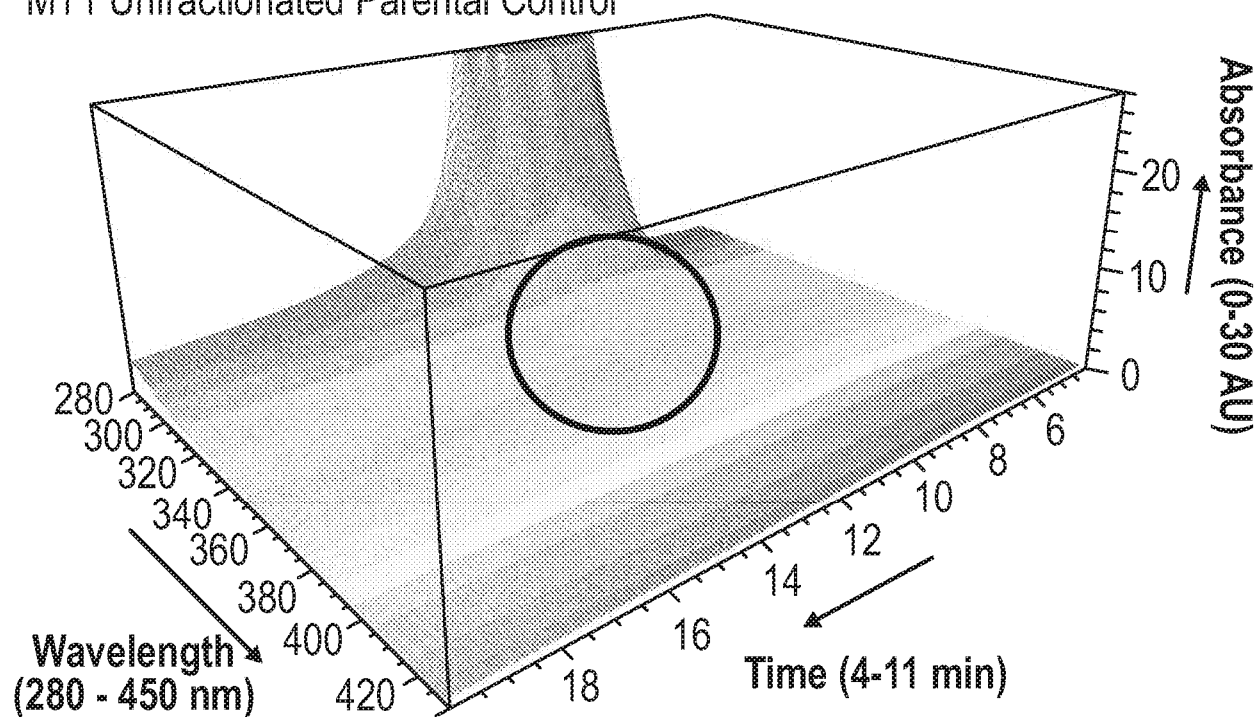


FIG. 18A

MT1 Fraction 1 (F1)

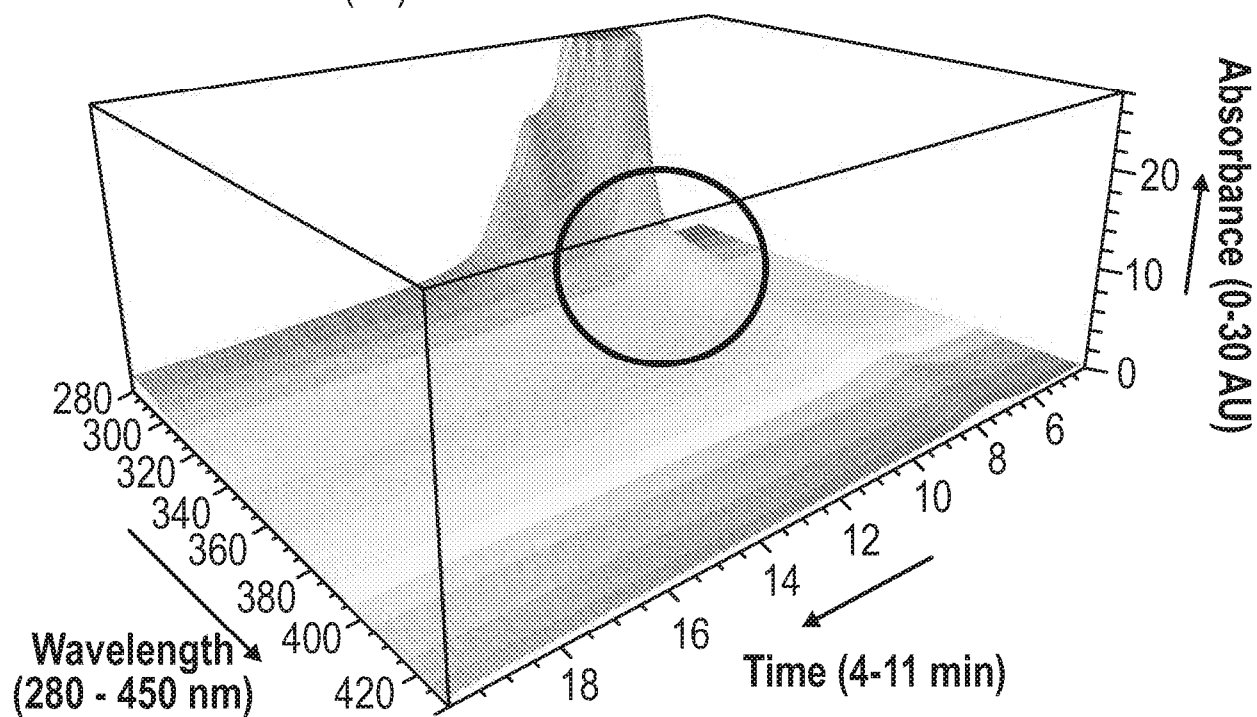


FIG. 18B

14/60

MT1 Fraction 2 (F2)

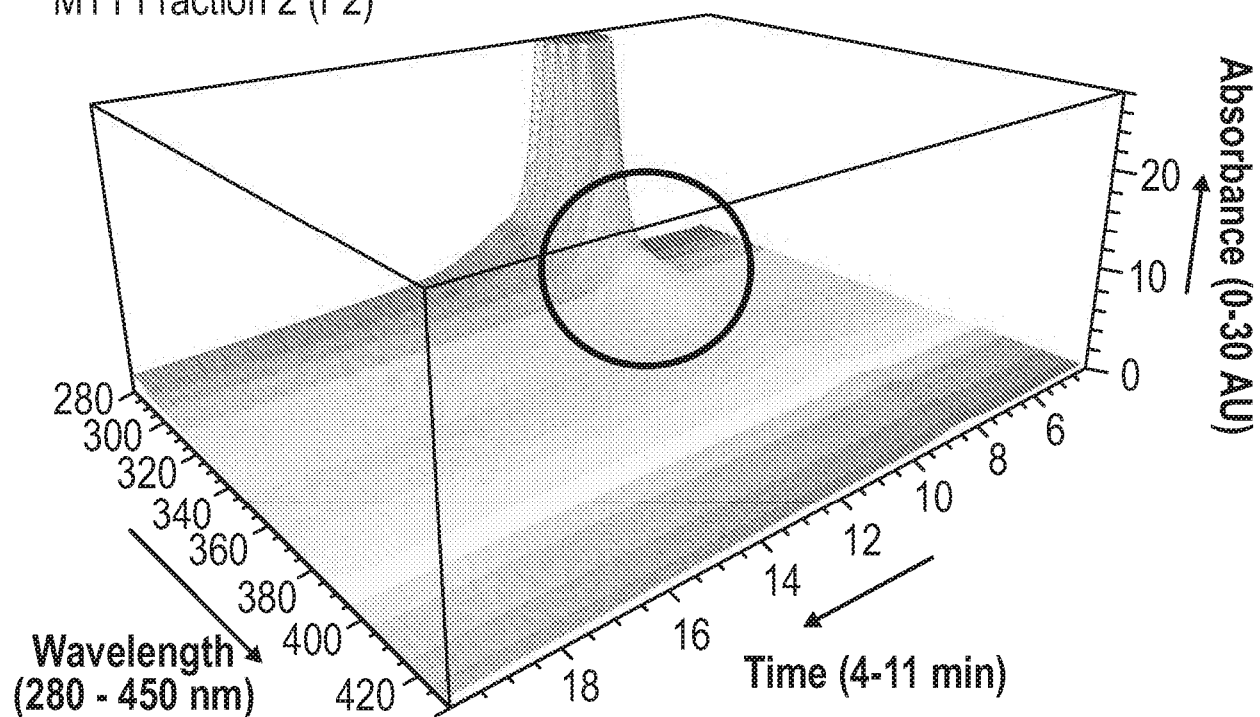


FIG. 18C

MT1 Fraction 3 (F3)

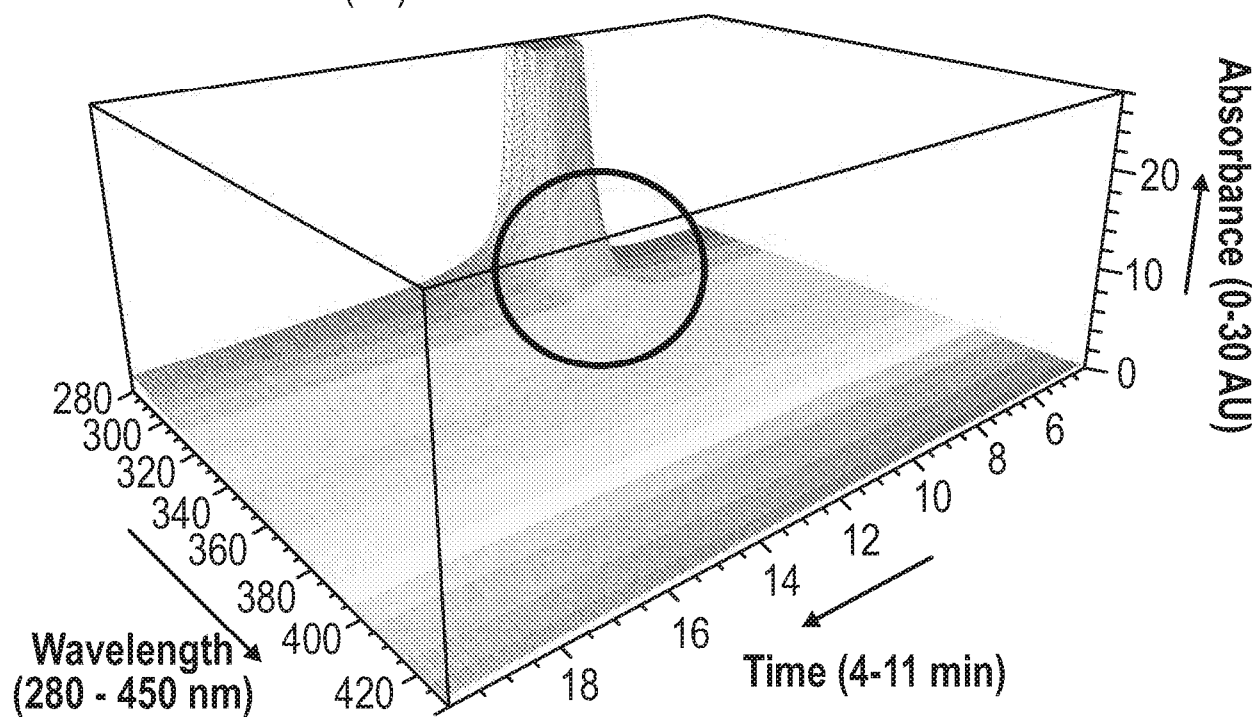


FIG. 18D

15/60

MT1 Fraction 4 (F4)

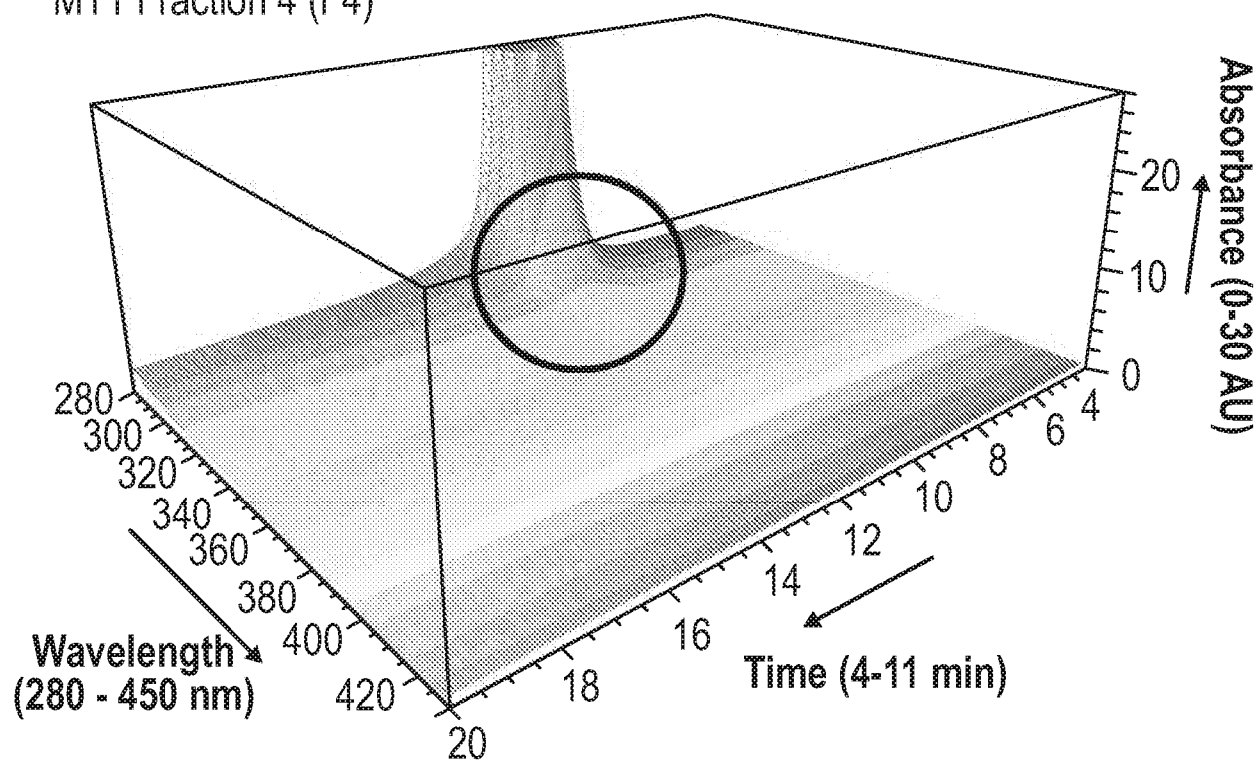


FIG. 18E

MT1 Fraction 5 (F5)

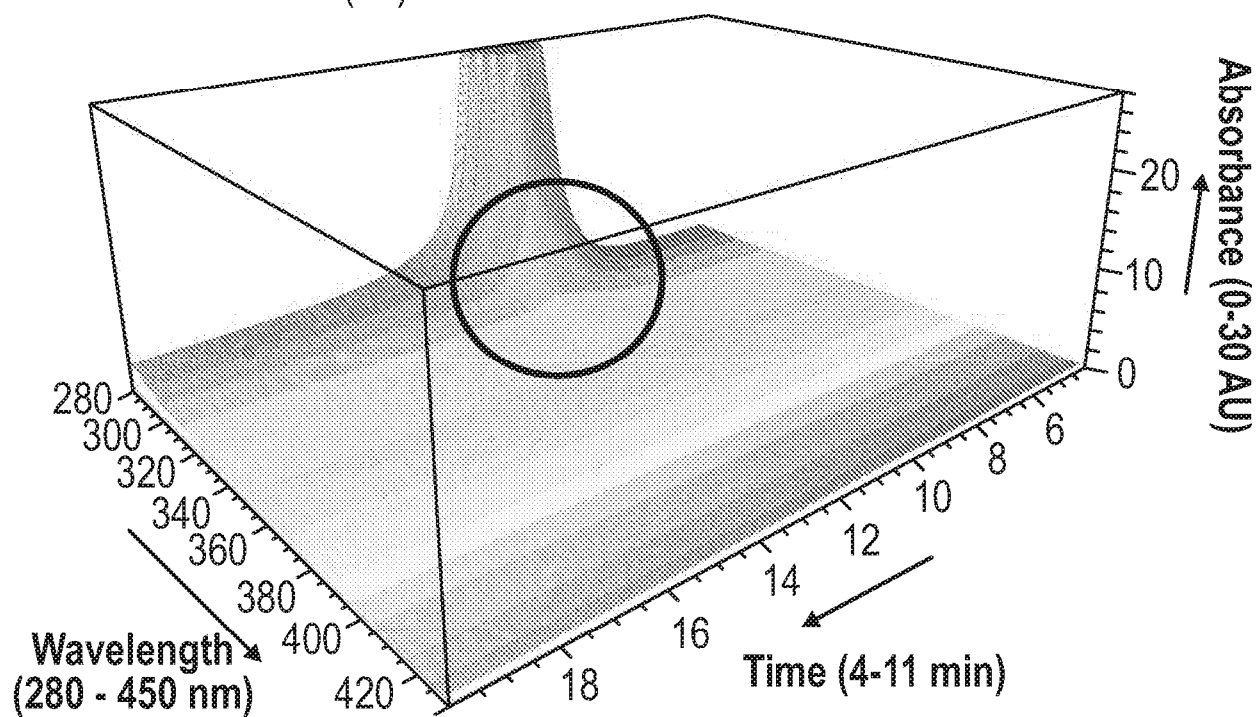


FIG. 18F

16/60

MT1 Fraction 6 (F6)

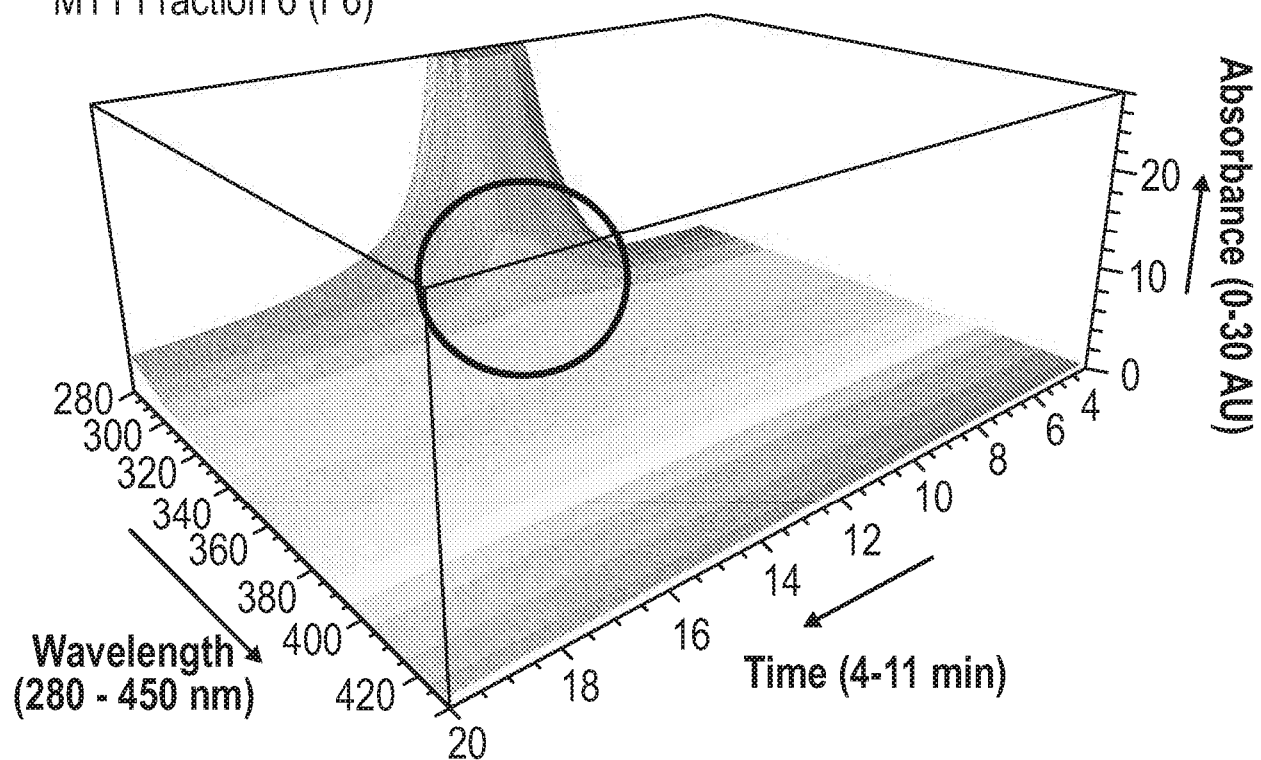


FIG. 18G

MT1 Fraction 7 (F7)

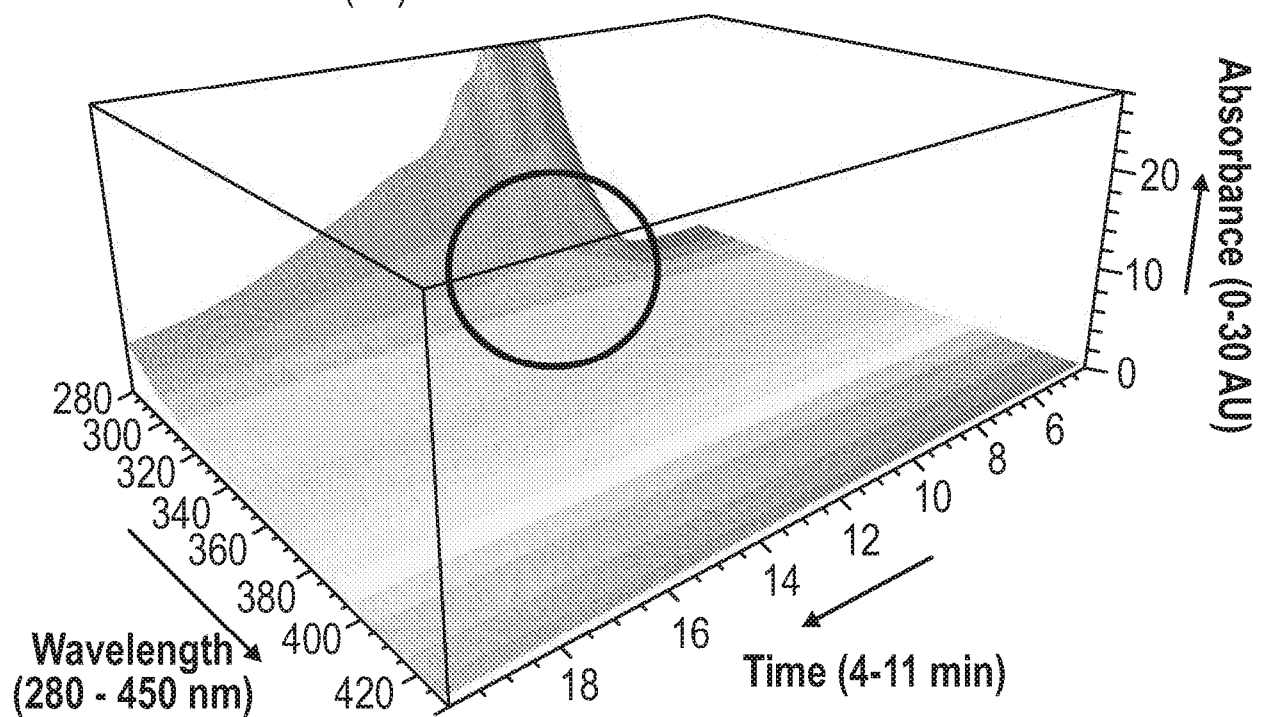


FIG. 18H

iciEF Electropherograms

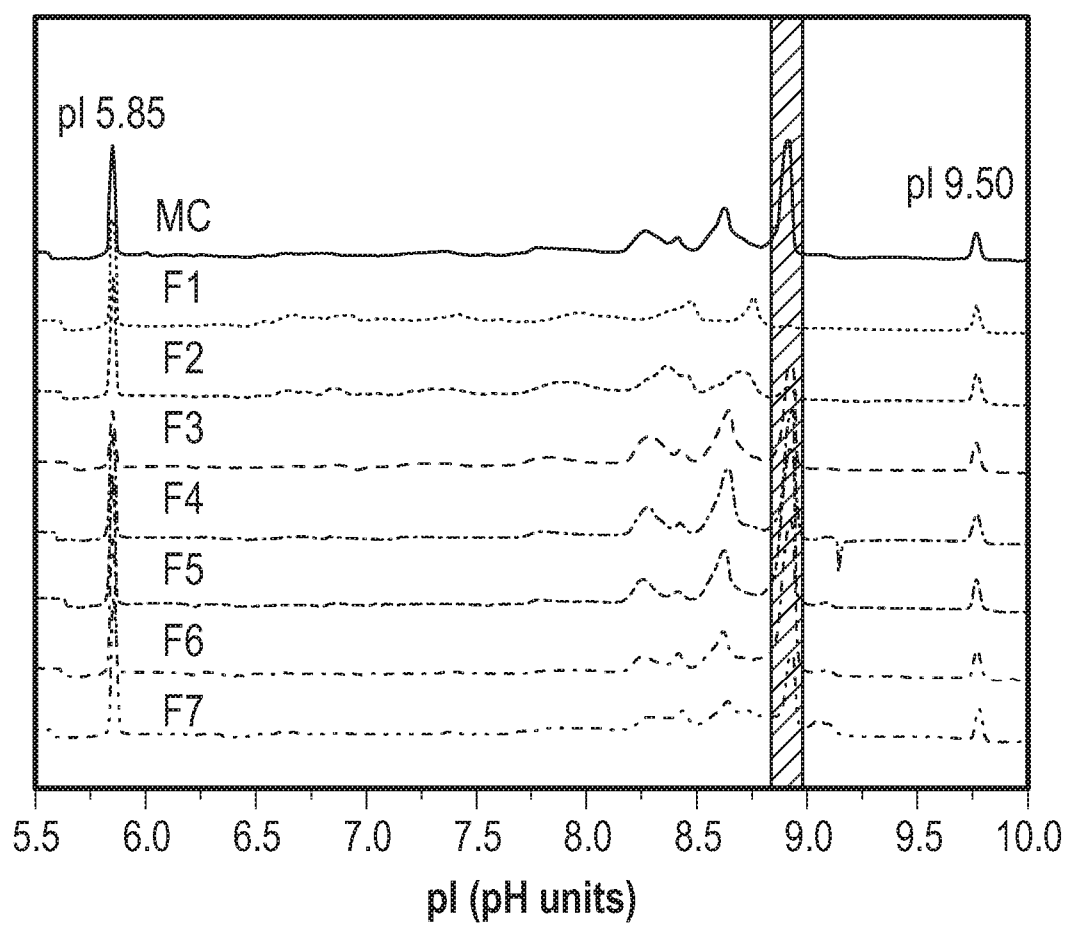


FIG. 19

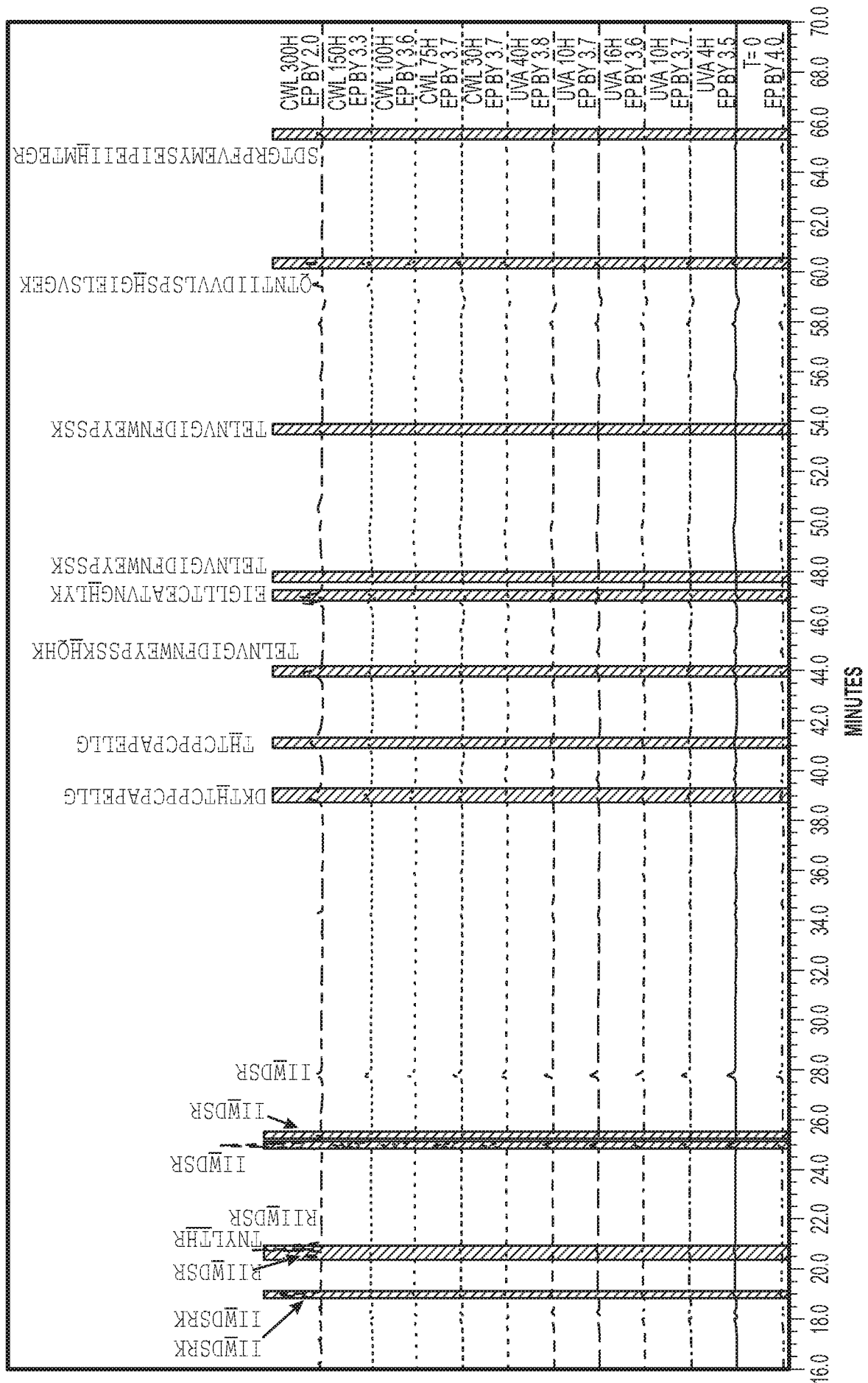


FIG. 20

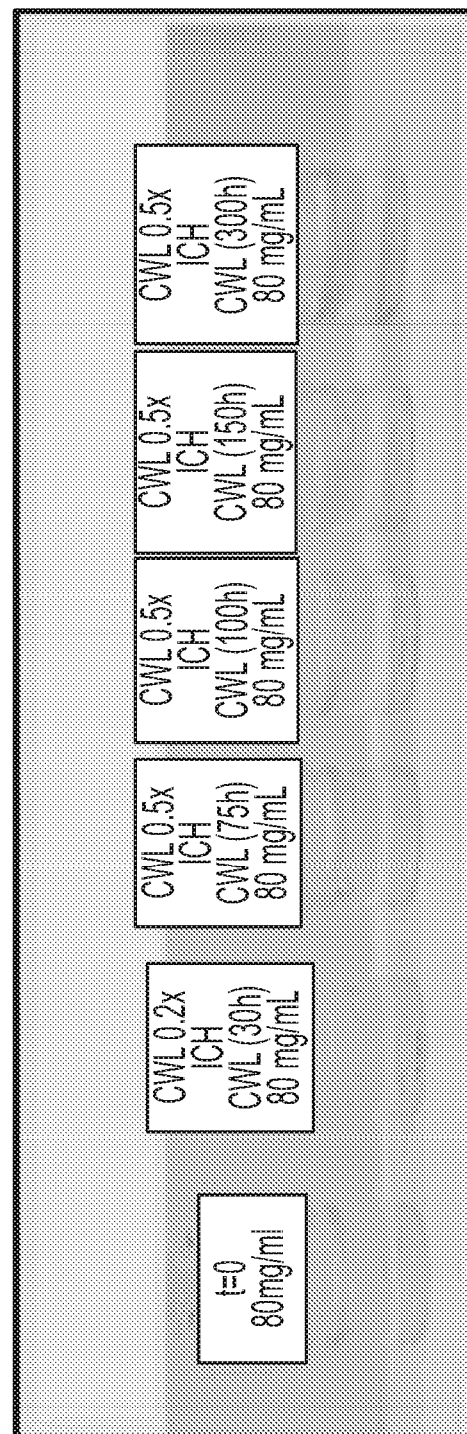
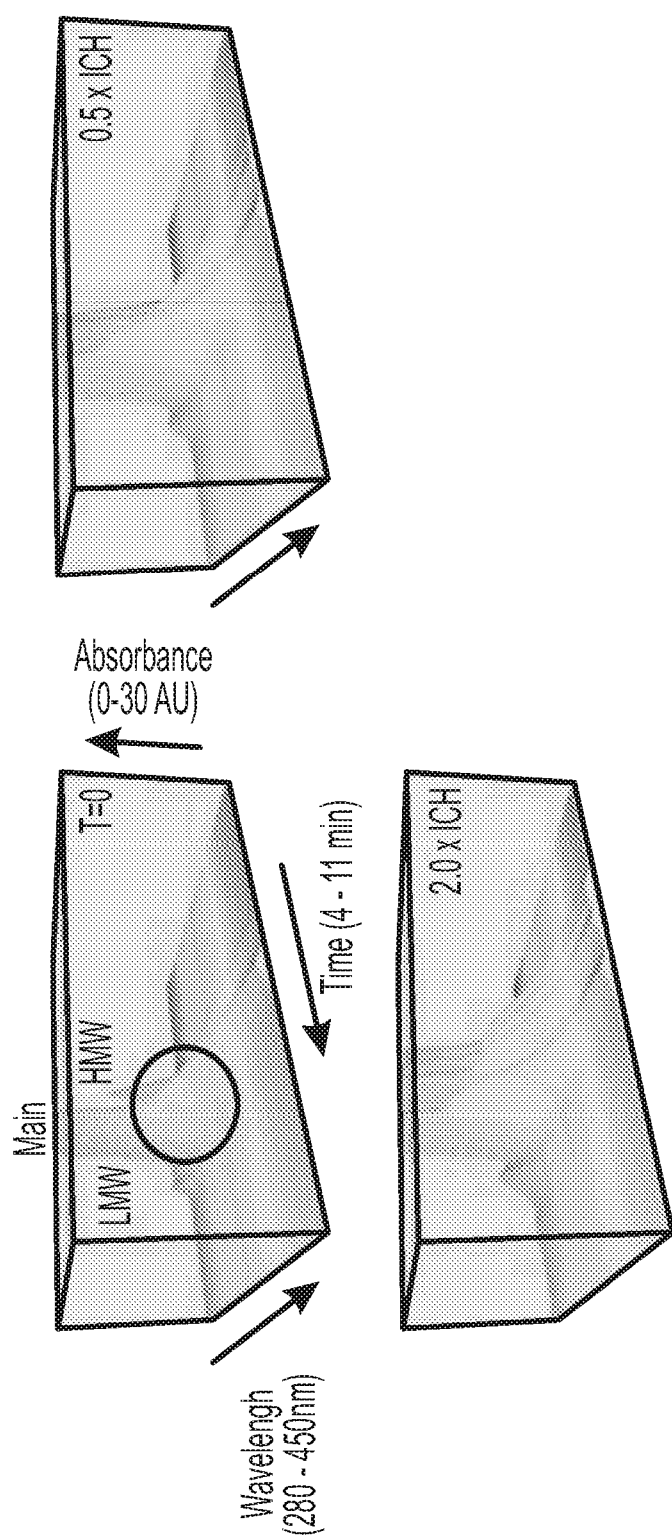


FIG. 21

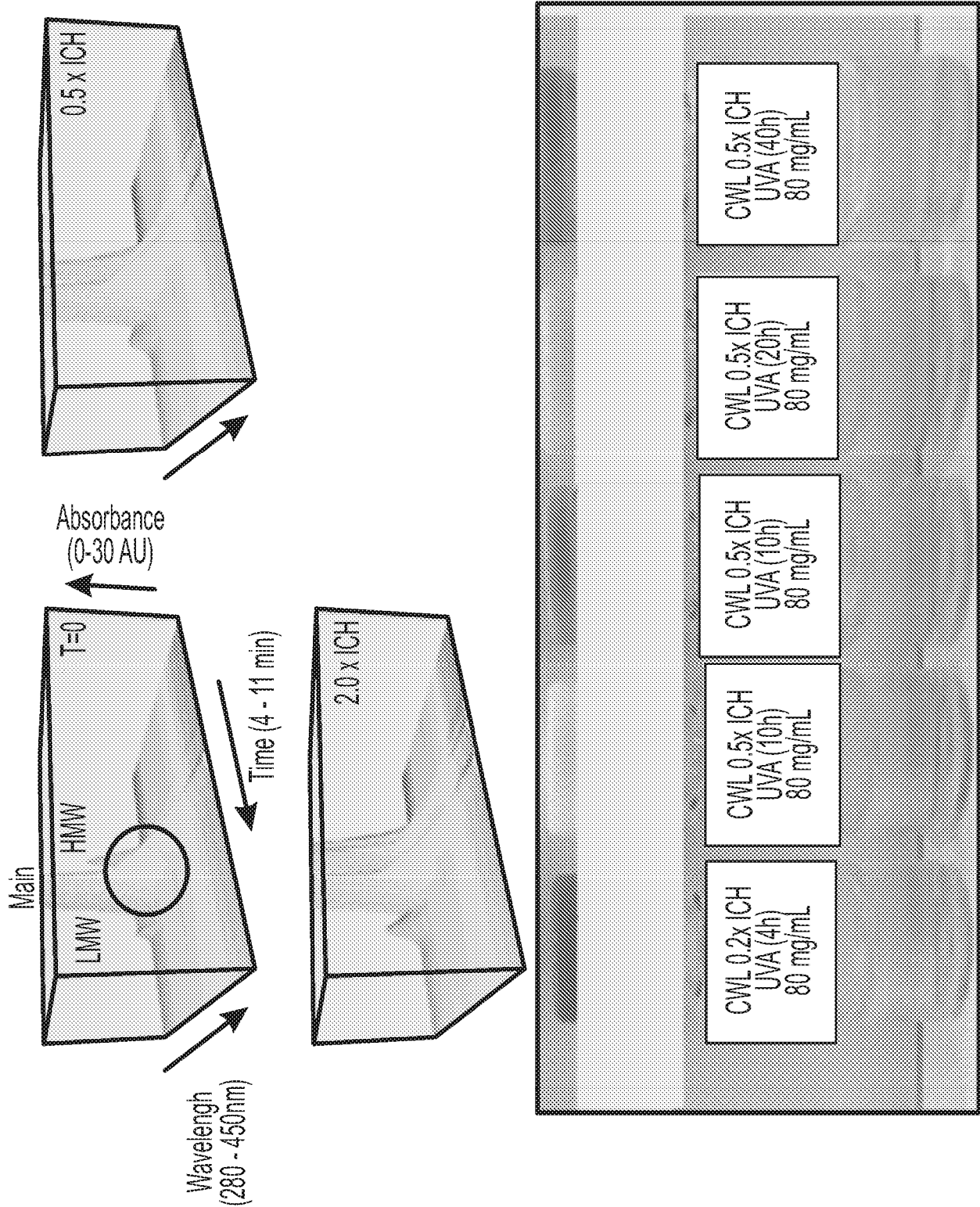


FIG. 22

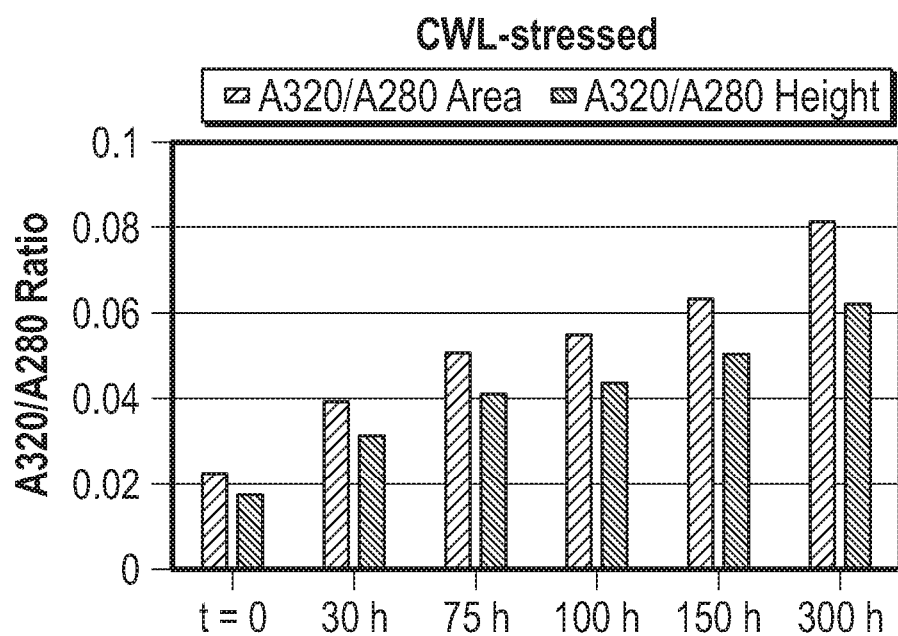


FIG. 23A

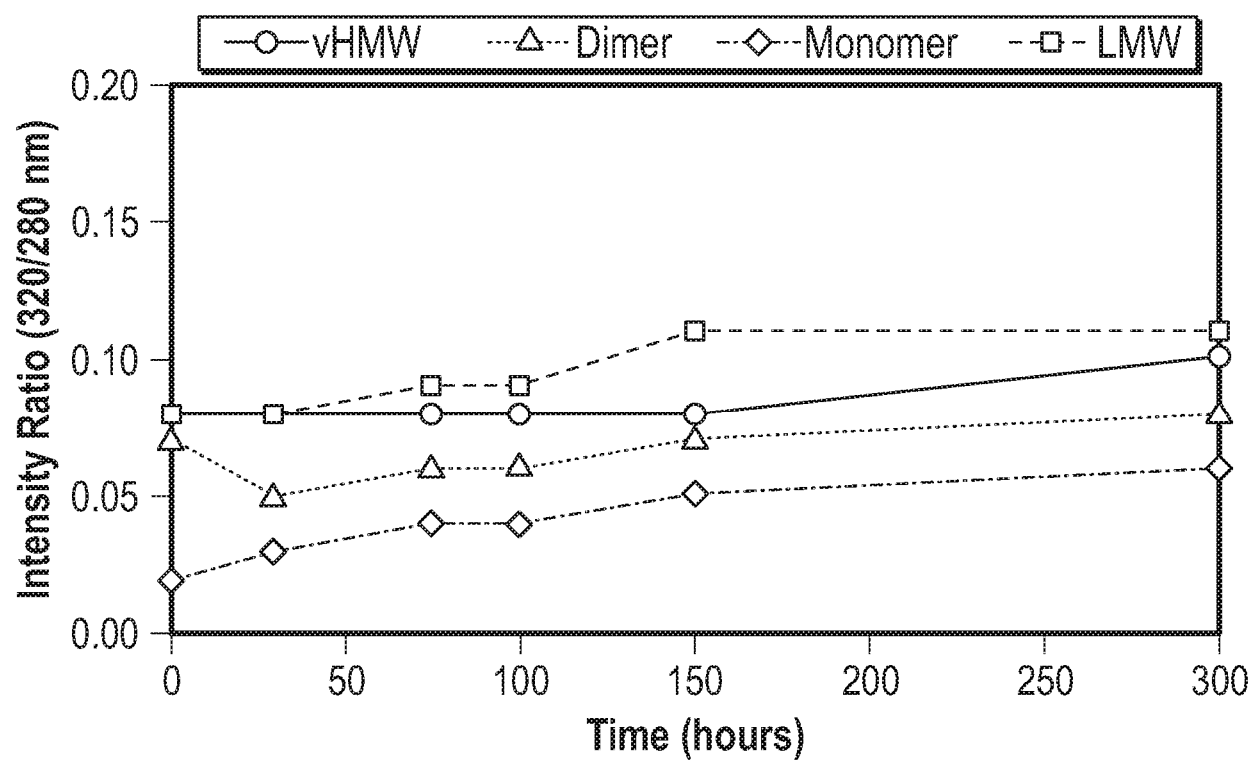


FIG. 23B

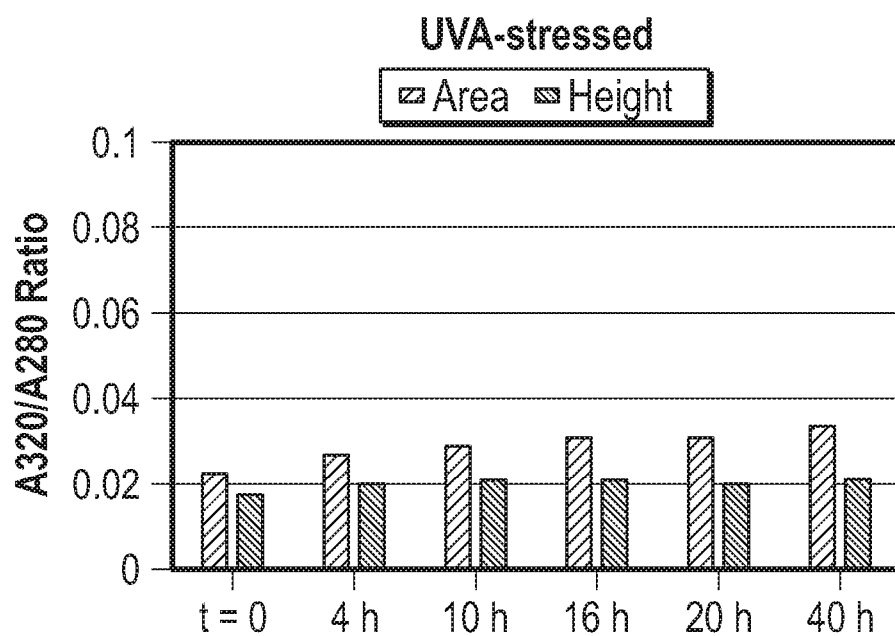


FIG. 24A

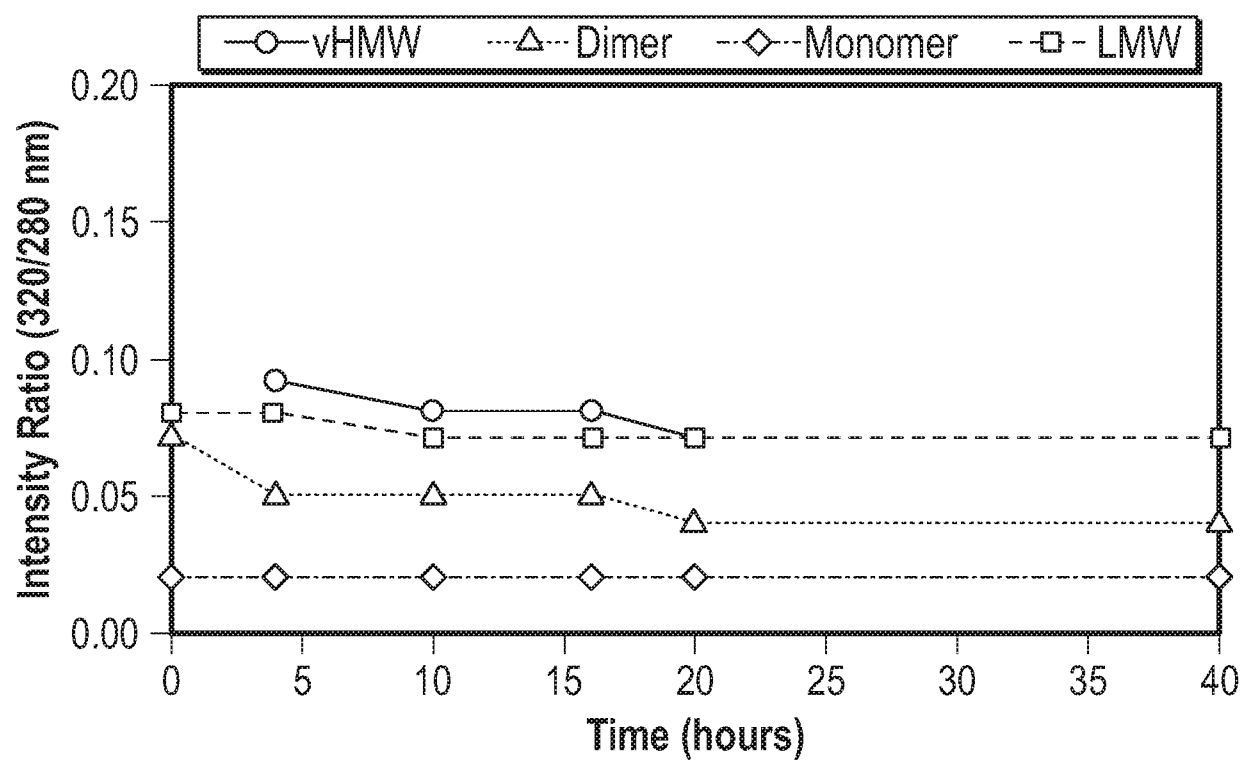


FIG. 24B

Scaled Estimates							
Term	Scaled Estimate				Std Error	t Ratio	Prob> t
Intercept	0.4682071				0.009776	47.89	<.0001*
Cysteine(0,1)	0.3470799				0.009806	35.40	<.0001*
B-Vits Group(0,1)	0.0373611				0.009897	3.78	0.0008*
Iron(0,1)	0.0698208				0.010074	6.93	<.0001*
Zinc(0,1)	0.0538585				0.010255	5.25	<.0001*
Folic Acid(0,1)	0.037899				0.010172	3.73	0.0009*
Cysteine*B-Vits Group	0.0253685				0.009837	2.58	0.0155*
Cysteine*Iron	0.07244				0.01011	7.17	<.0001*
Cysteine*Zinc	0.0339284				0.009973	3.40	0.0020*
Cysteine*Folic Acid	0.0298402				0.010218	2.92	0.0068*
B-Vits Group*Iron	0.0229271				0.009945	2.31	0.0288*
B-Vits Group*Folic Acid	-0.022614				0.010243	-2.21	0.0356*

FIG. 25A

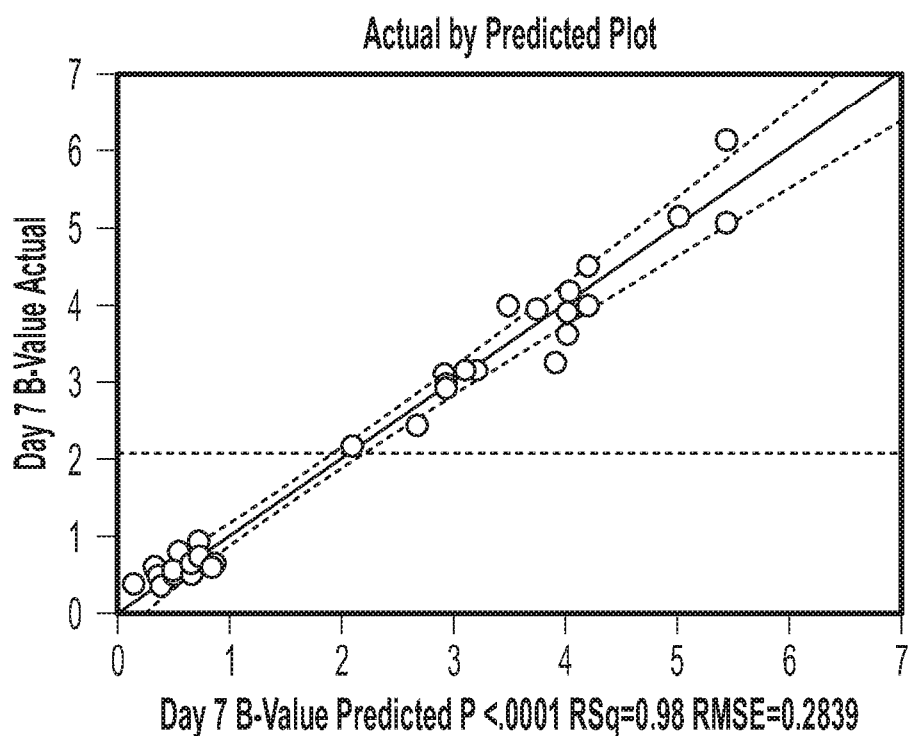


FIG. 25B

24/60

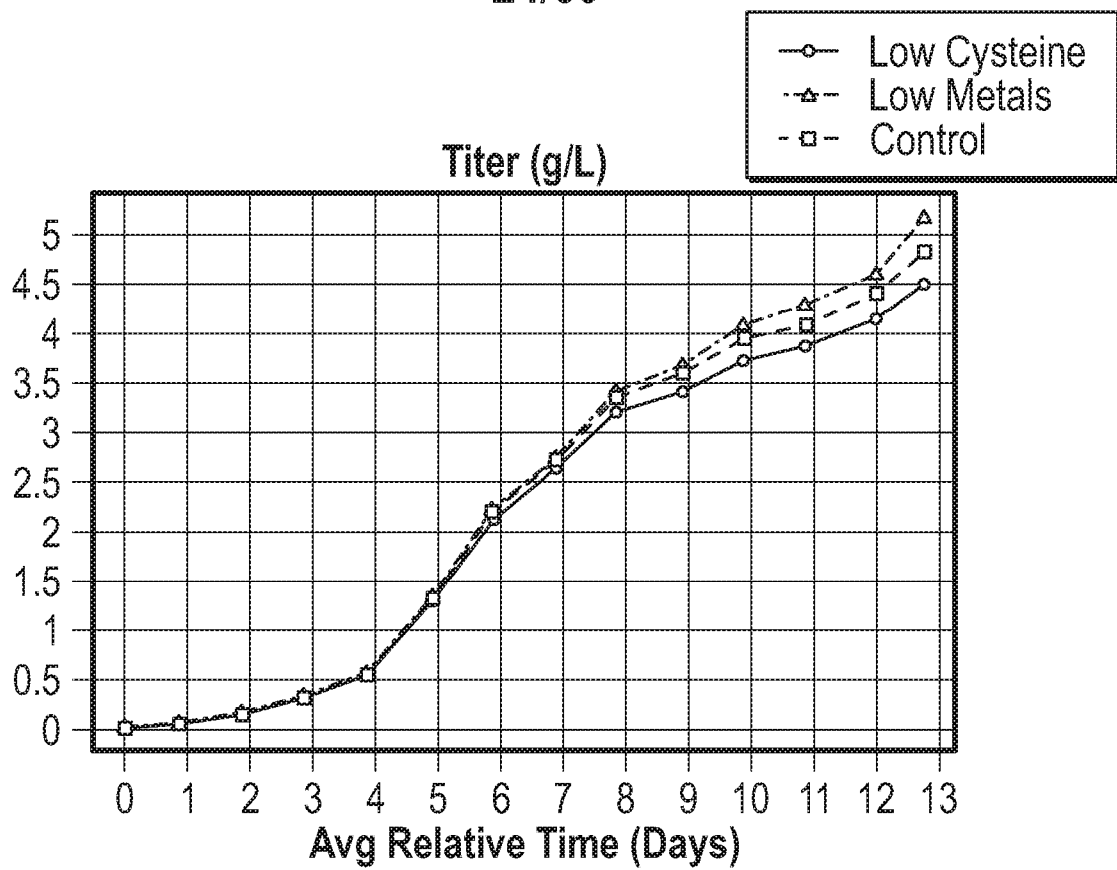


FIG. 26A

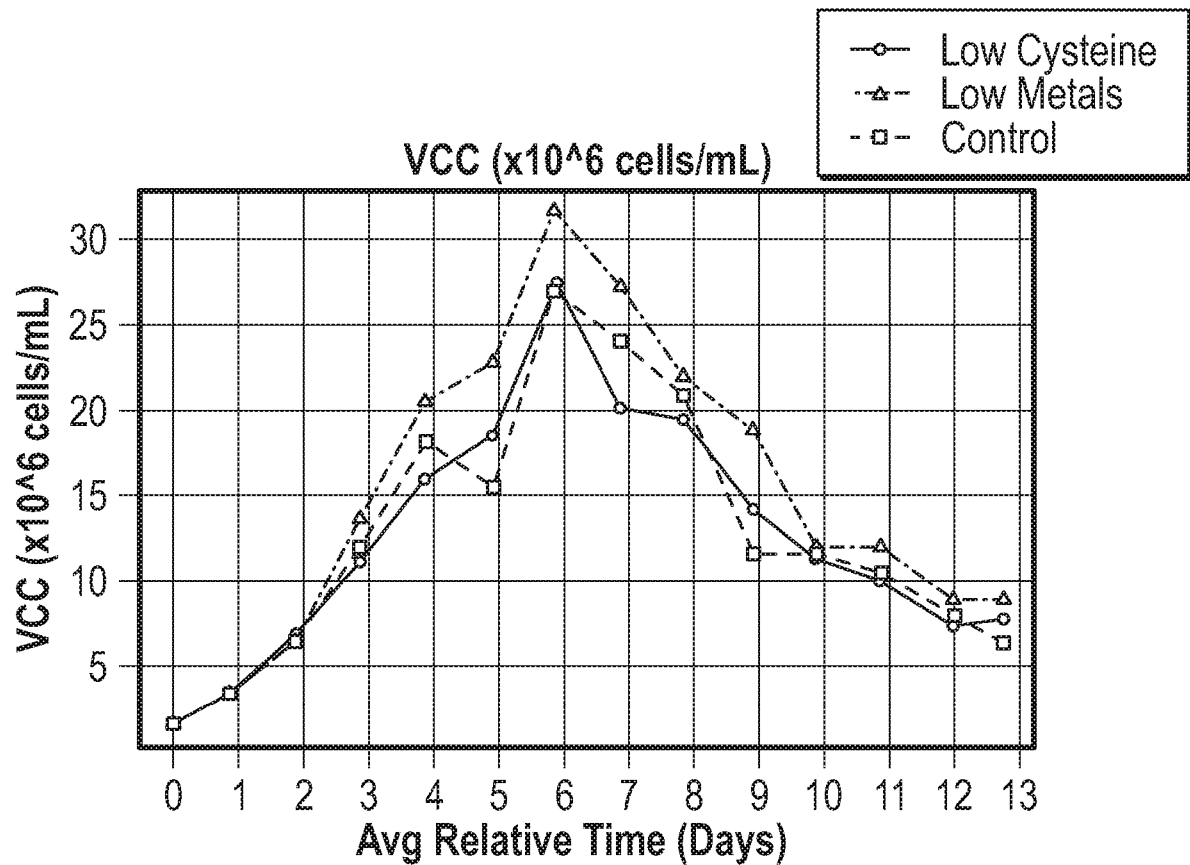


FIG. 26B

25/60

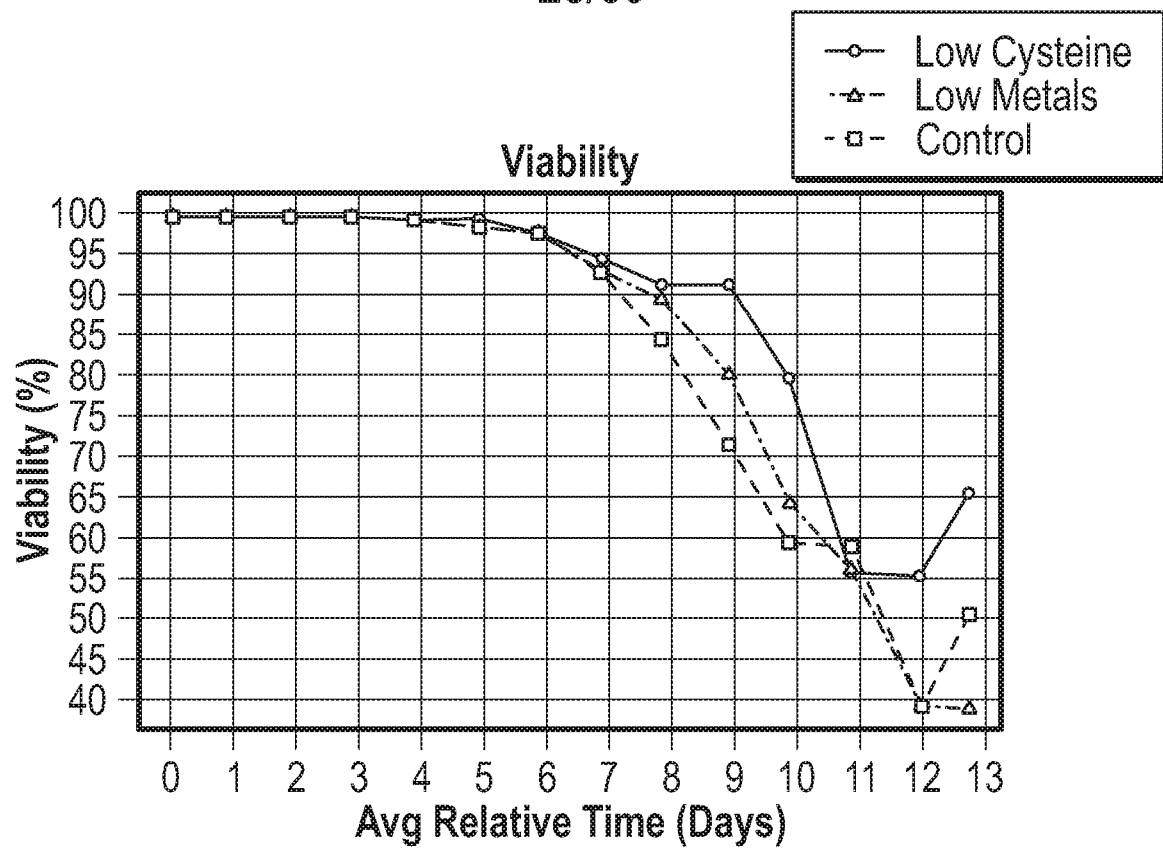


FIG. 26C

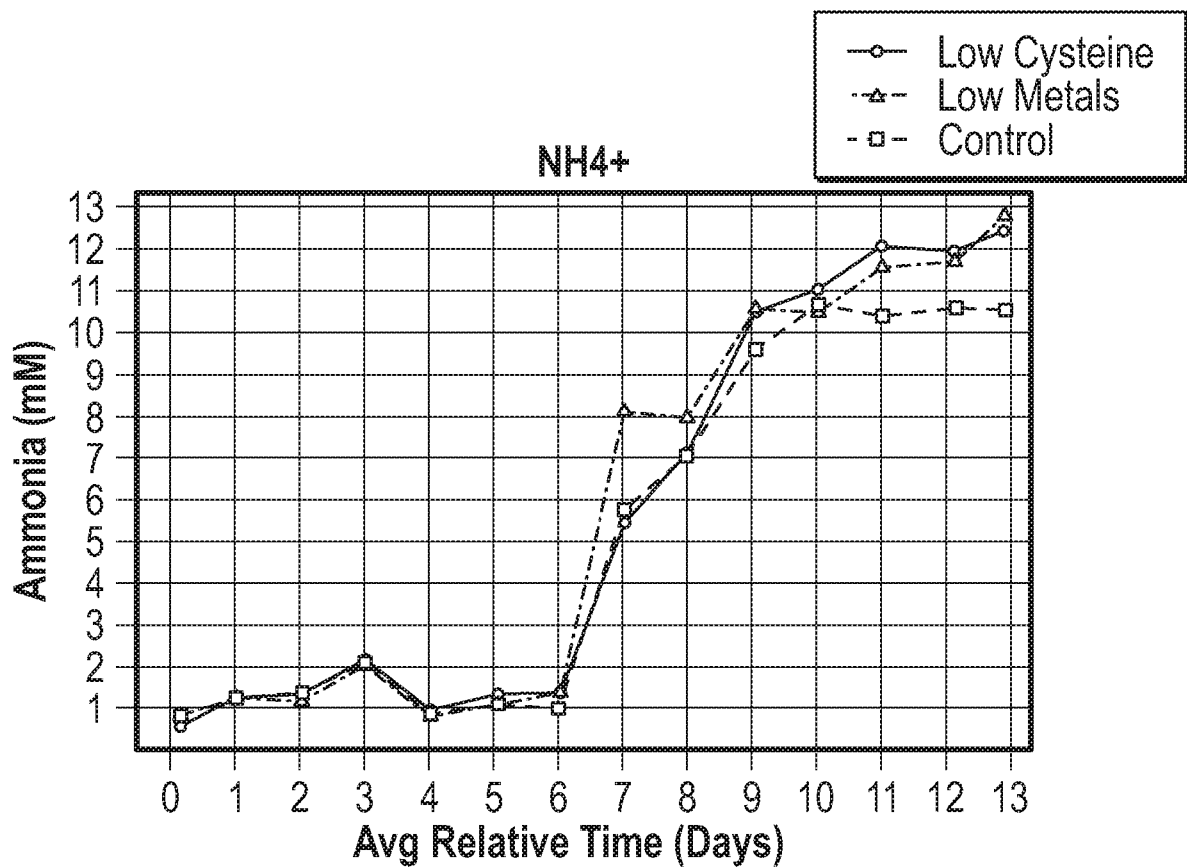


FIG. 26D

26/60

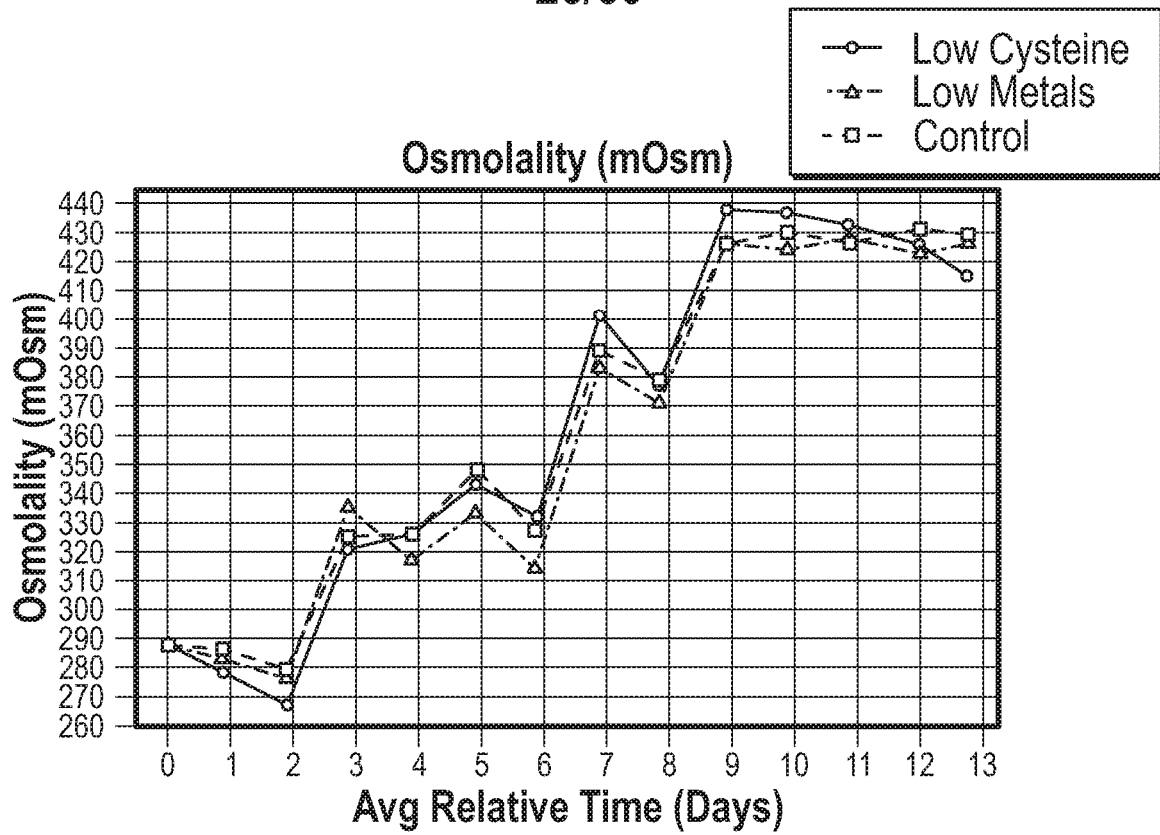


FIG. 26E

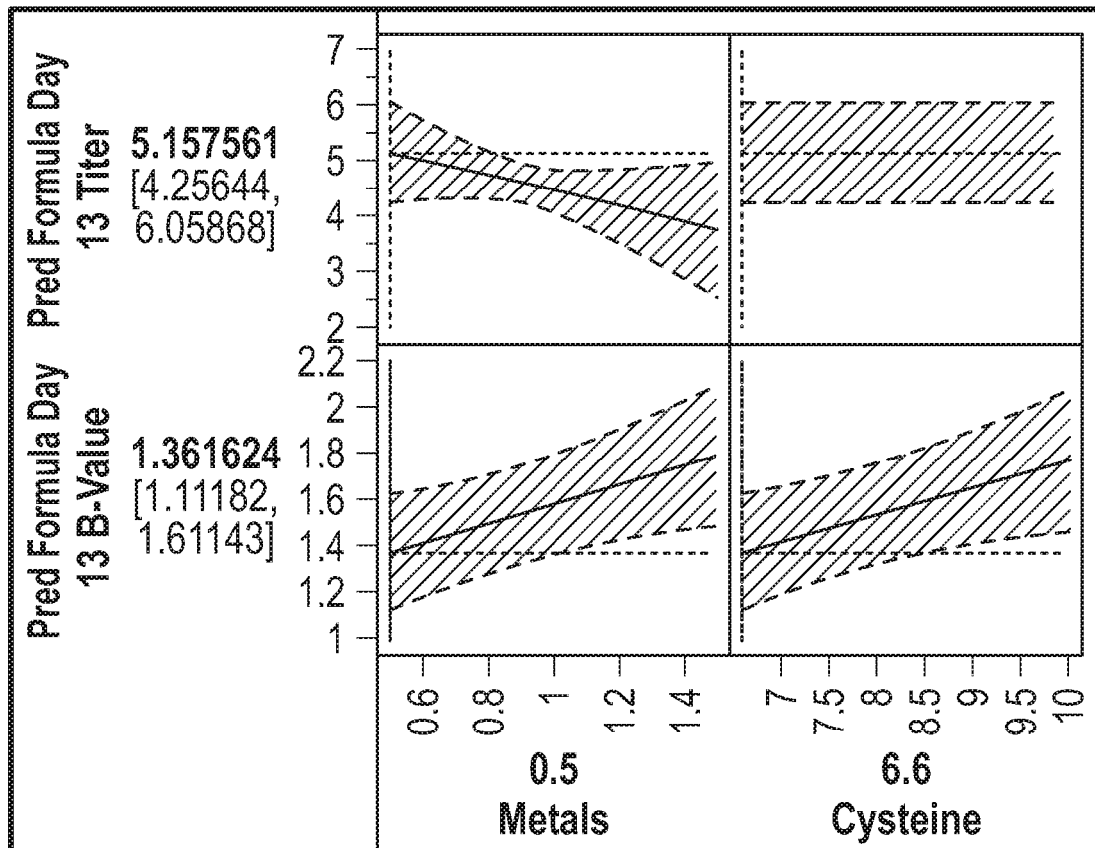


FIG. 27

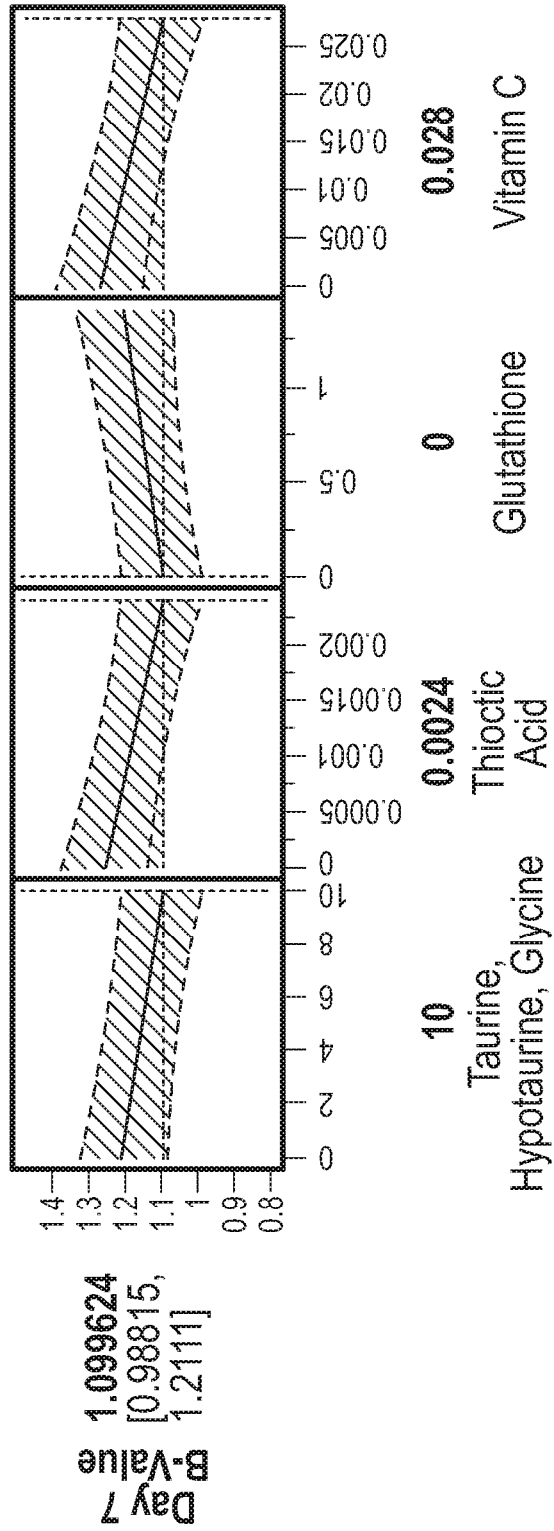


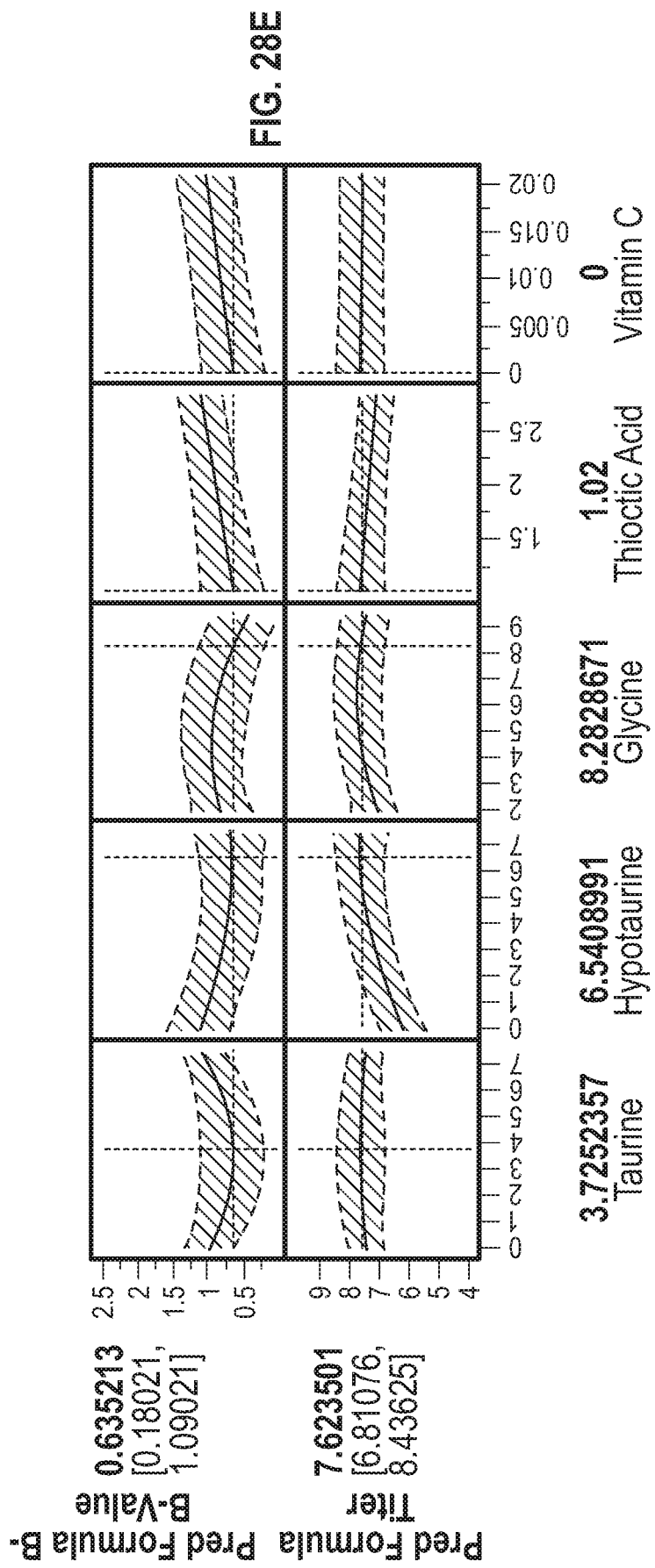
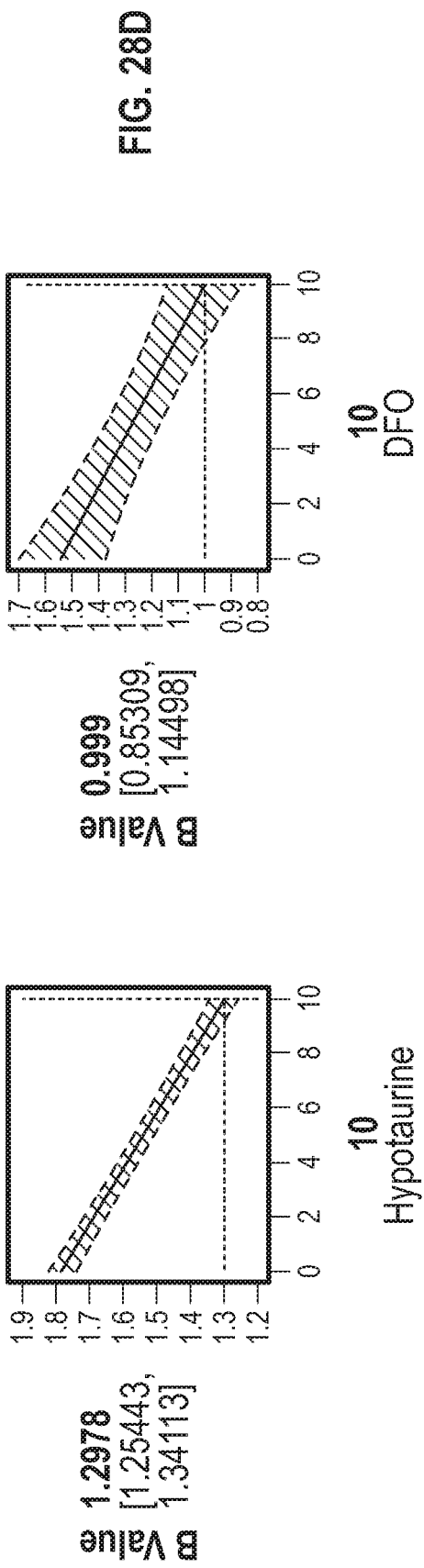
FIG. 28A

Scaled Estimates					
Term	Scaled Estimate	Std Error	t Ratio	Prob> t	
Intercept	1.3997181	0.022019	63.57	<.0001*	
Taurine, Hypotaurine, Glycine(0,1)	-0.153727	0.02201	-6.98	<.0001*	
Thiocetic Acid(0,1)	-0.010756	0.022019	-0.49	0.6293	
Glutathione(0,1)	0.152492	0.022112	6.90	<.0001*	
Vitamin C(0,1)	-0.017603	0.02201	-0.80	0.4311	
Taurine, Hypotaurine, Glycine*Glutathione	-0.101323	0.022186	-4.57	0.0001*	
Thiocetic Acid*Vitamin C	-0.06684	0.02201	-3.04	0.0054*	

FIG. 28B

Term	Scaled Estimate	Std Error	t Ratio	Prob> t
Intercept	1.079804	0.126434	8.54	0.0010*
Taurine (0,1)	-0.030846	0.040585	-0.76	0.4896
Hypotaurine (0,1)	-0.085952	0.044048	-1.95	0.1228
Glycine (0,1)	-0.181863	0.047269	-3.85	0.0183*
Thioctic Acid (0,1)	0.0575148	0.0465	1.24	0.2838
Vitamin C (0,1)	-0.093266	0.0392	-2.38	0.0761
Taurine*Hypotaurine	0.1943093	0.042535	4.57	0.0103*
Taurine*Thioctic Acid	-0.031889	0.044591	-0.72	0.5140
Taurine*Vitamin C	0.0948234	0.046063	2.06	0.1086
Hypotaurine*Thioctic Acid	-0.009982	0.045049	-0.22	0.8355
Hypotaurine*Vitamin C	0.1509187	0.043708	3.45	0.0260*
Thioctic Acid*Vitamin C	-0.176346	0.043781	-4.03	0.0158*
Taurine*Taurine	0.3679087	0.117837	3.12	0.0355*
Hypotaurine*Hypotaurine	0.192703	0.110017	1.75	0.1547
Glycine*Glycine	-0.270392	0.11851	-2.28	0.0846

FIG. 28C



30/60

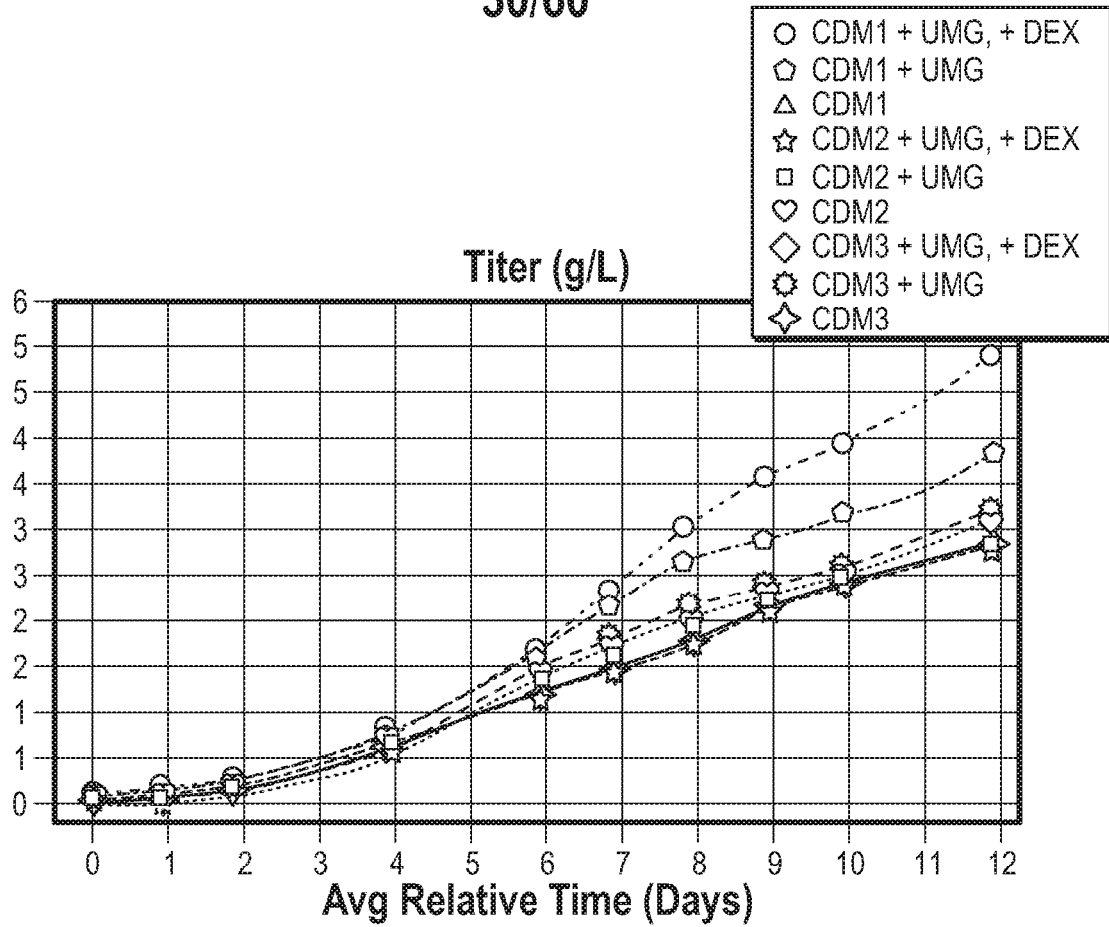


FIG. 29

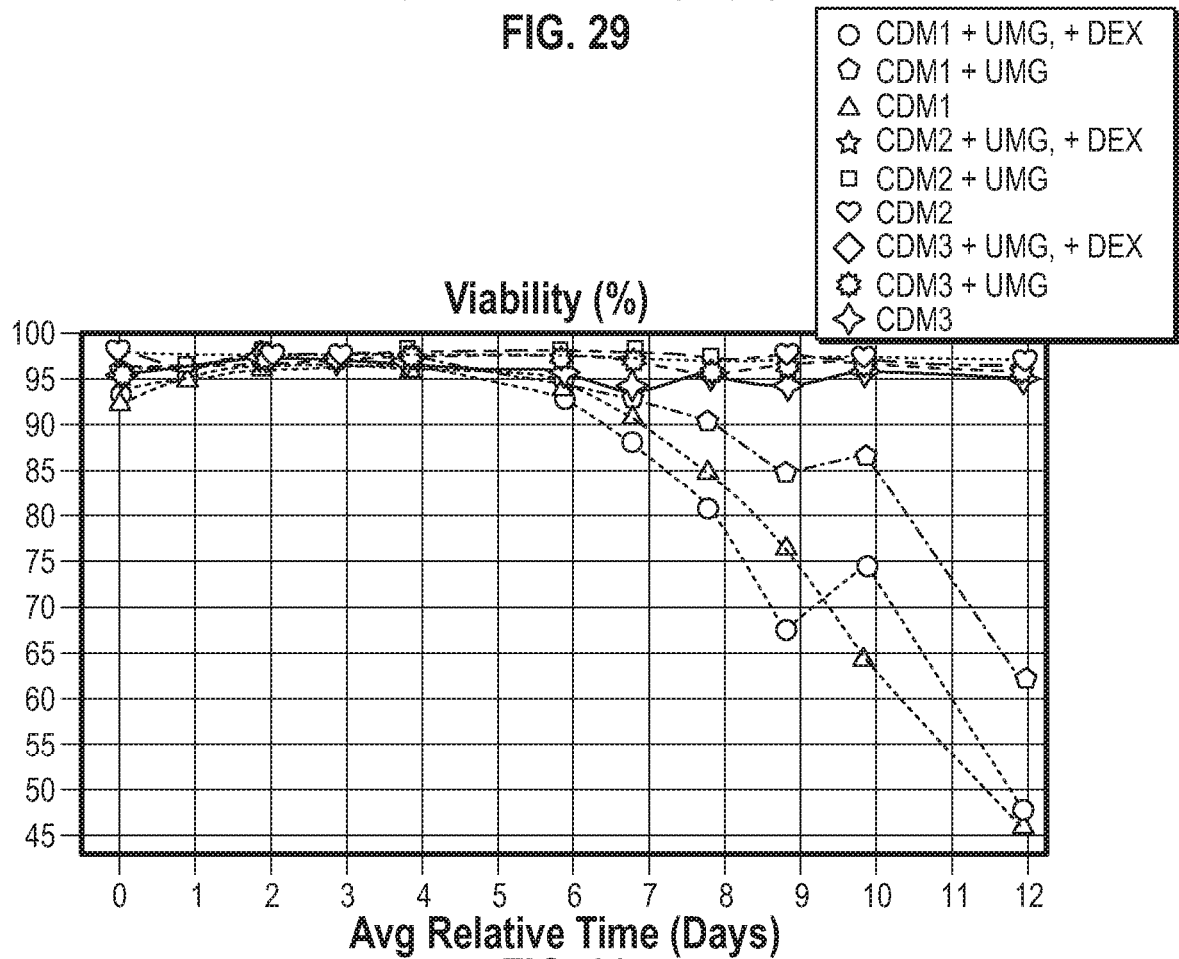


FIG. 30

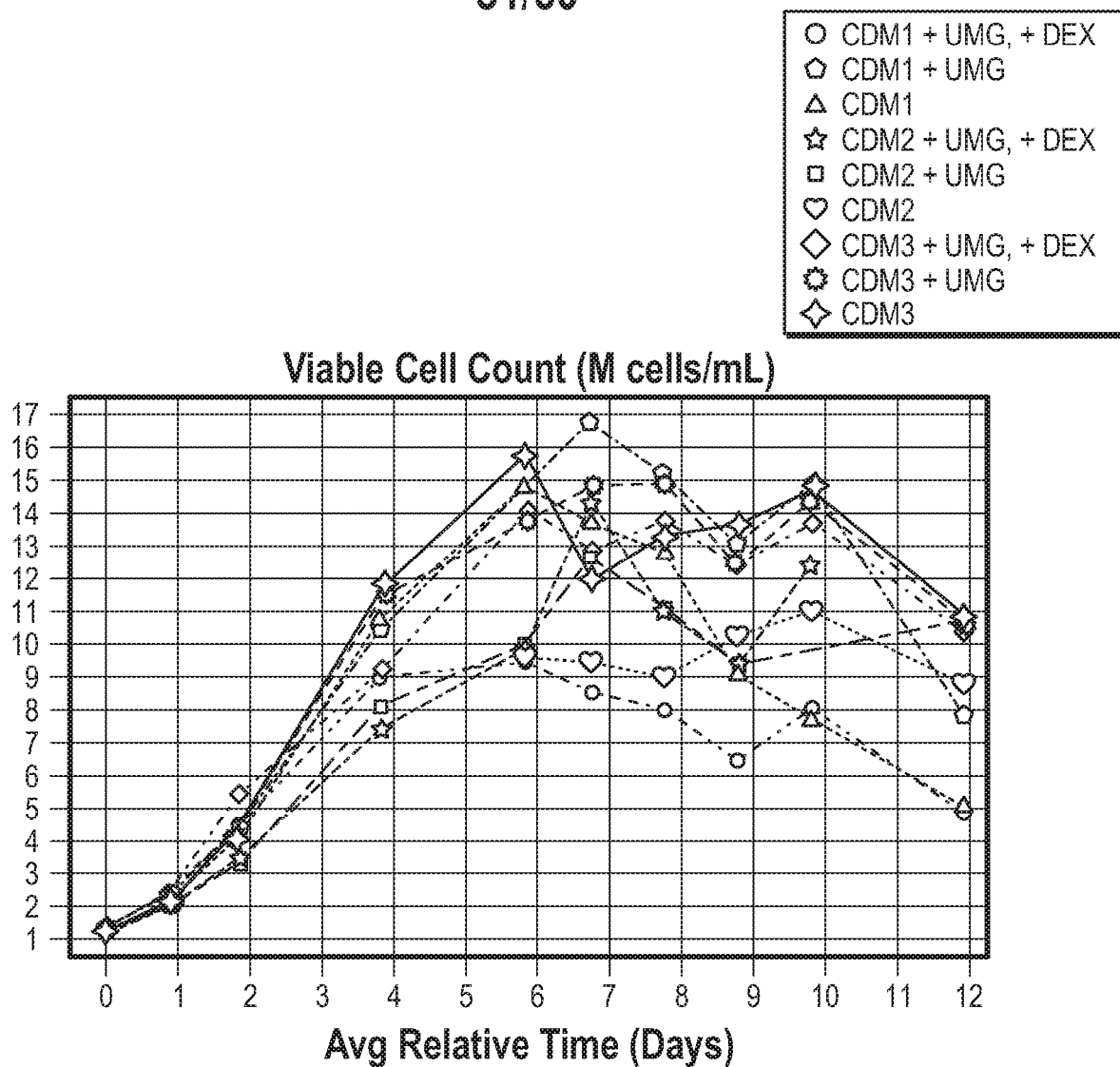


FIG. 31

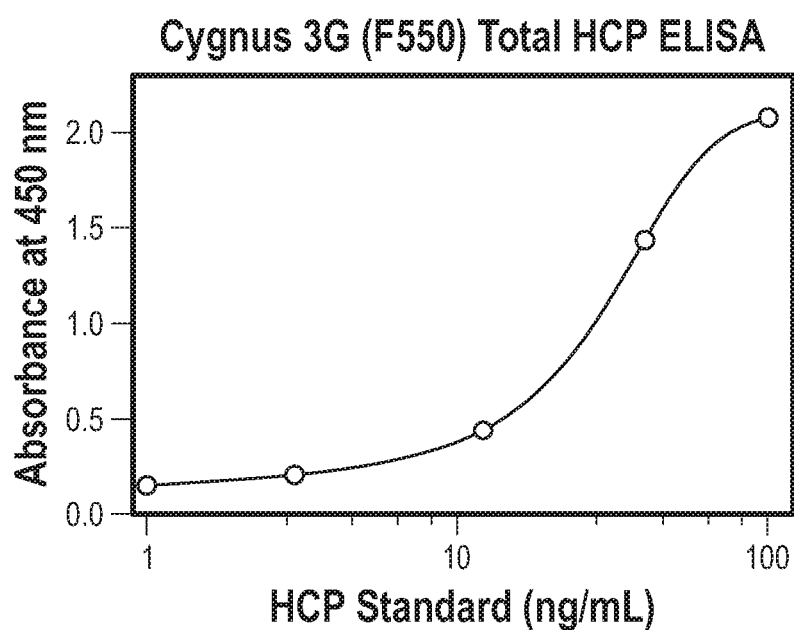


FIG. 32

32/60

Non Reduced

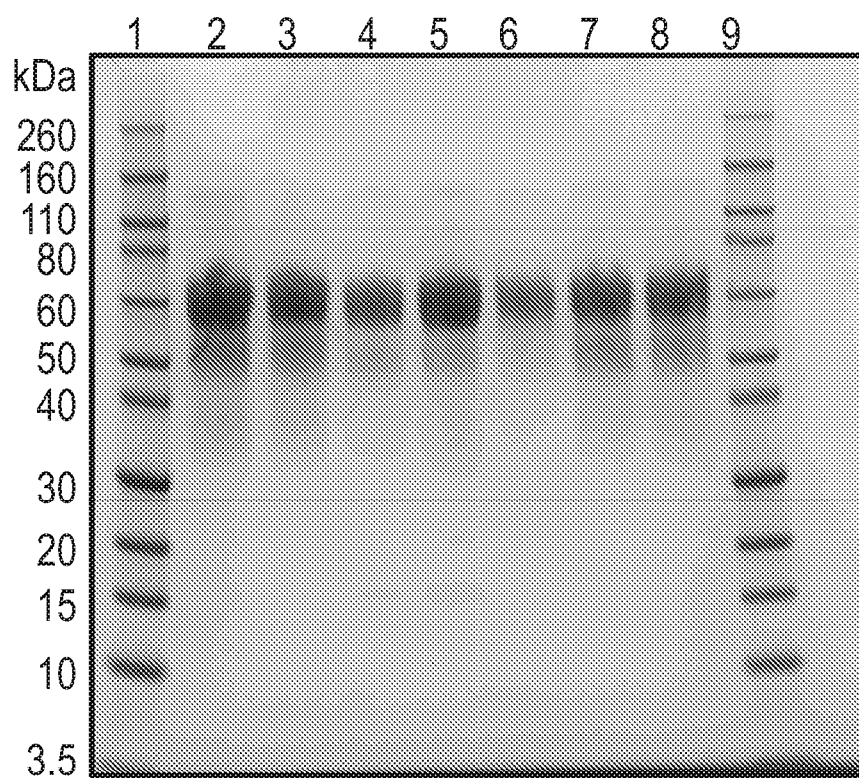


FIG. 33

Reduced

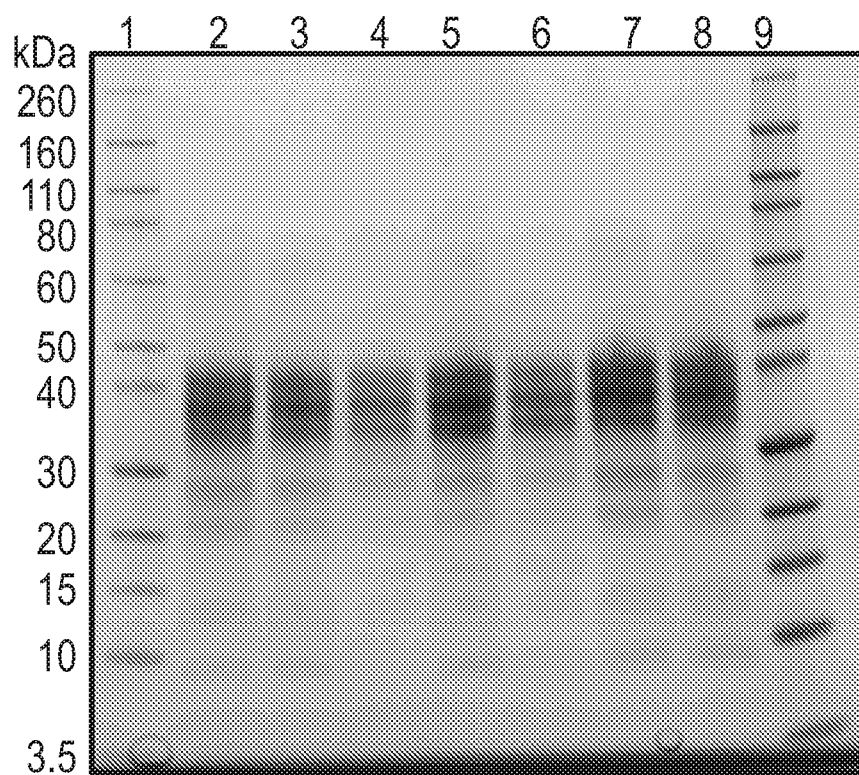


FIG. 34

33/60

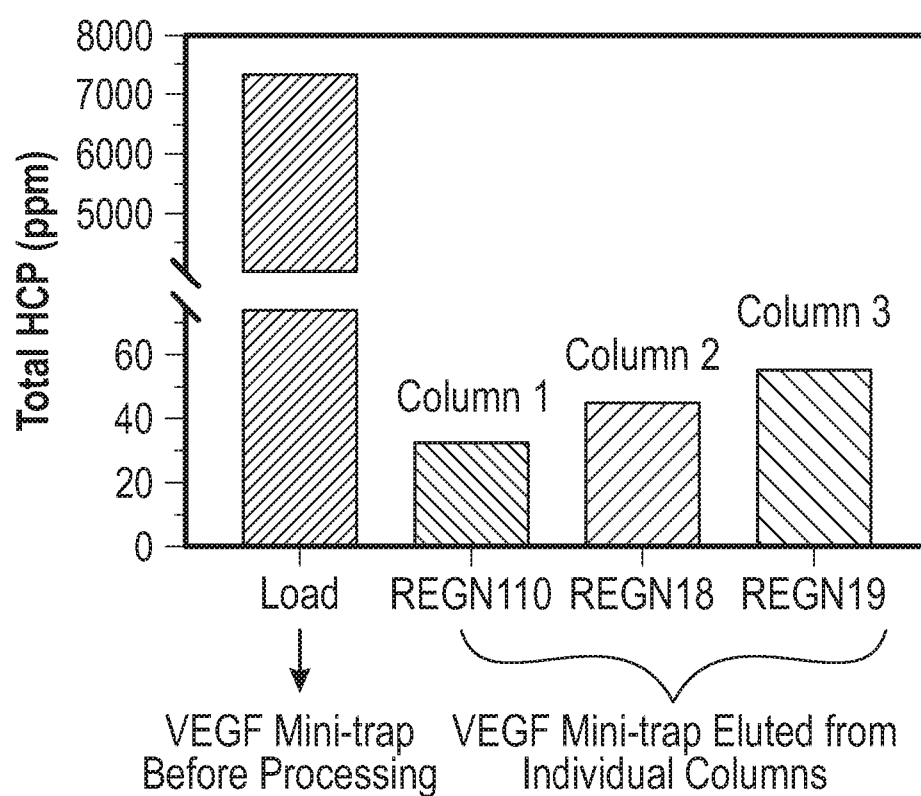


FIG. 35A

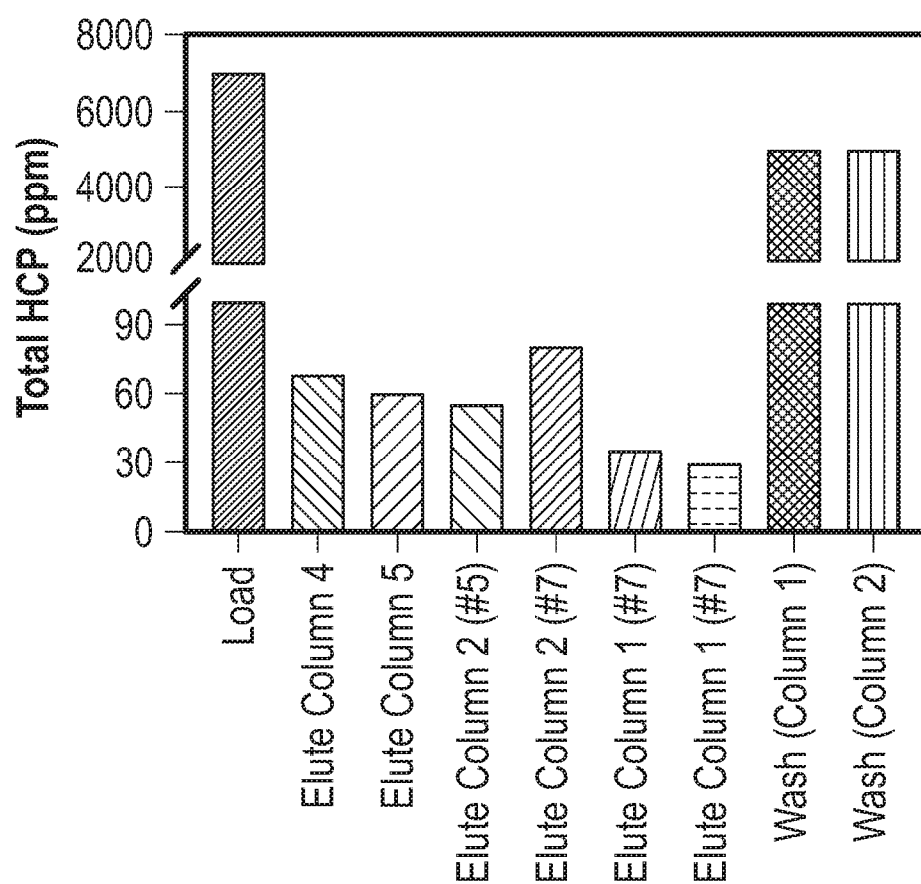


FIG. 35B

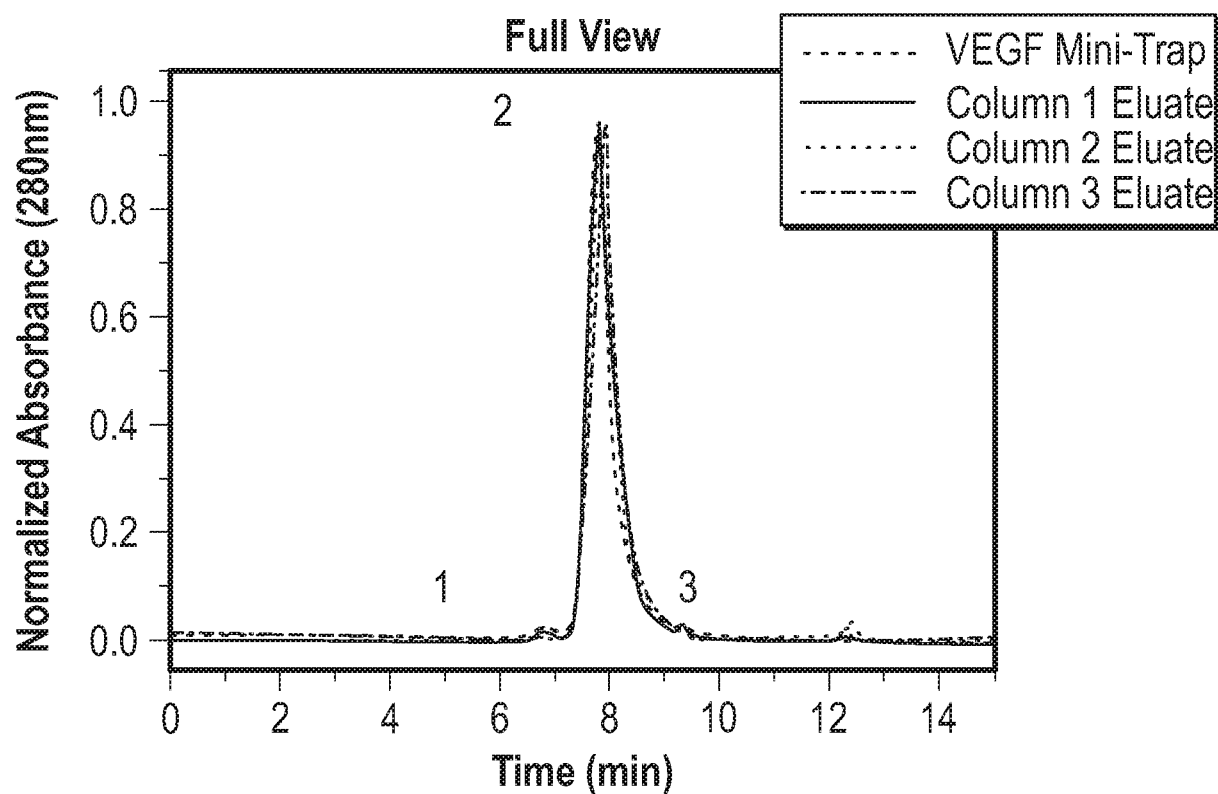


FIG. 36A

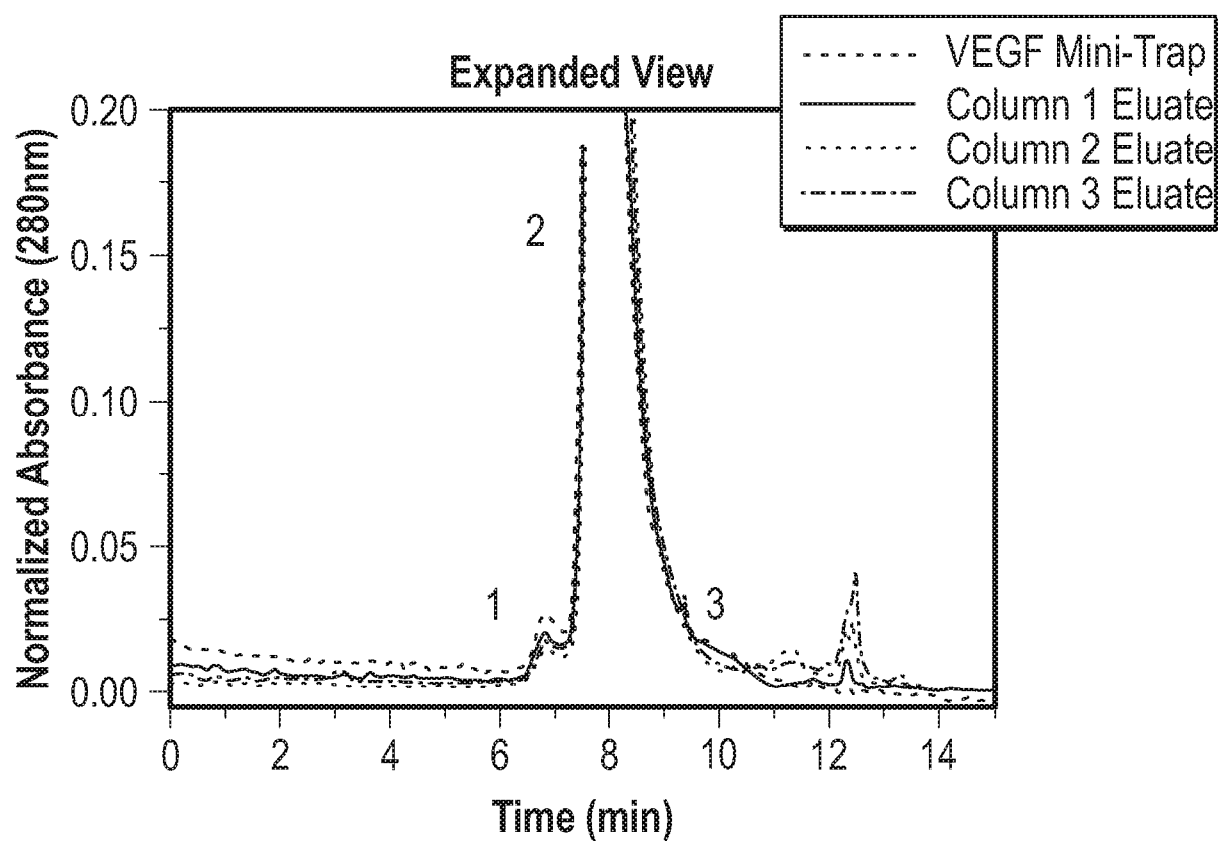


FIG. 36B

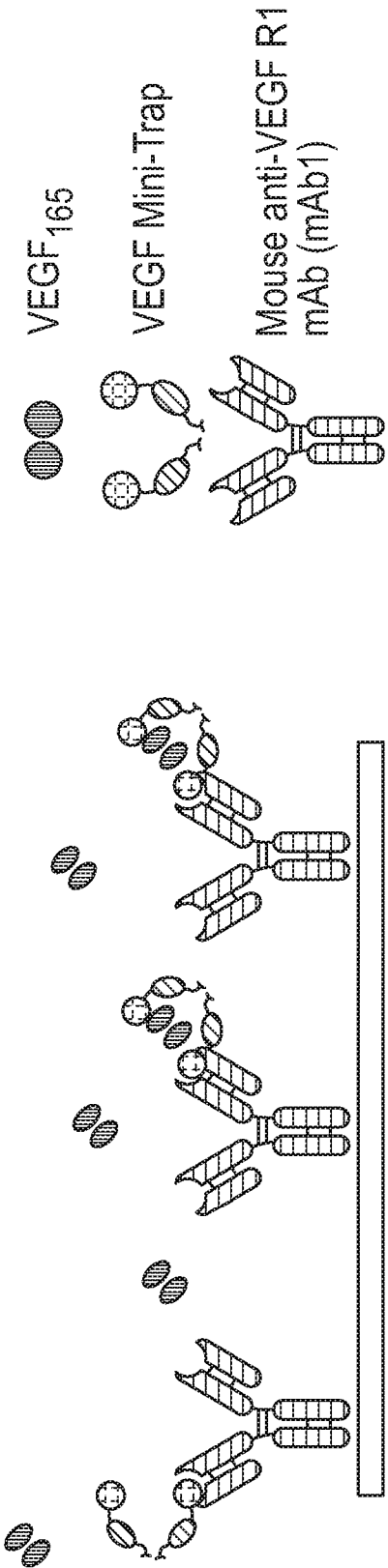
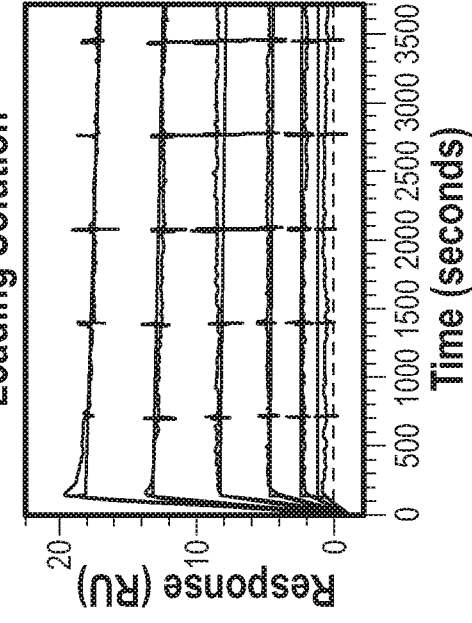
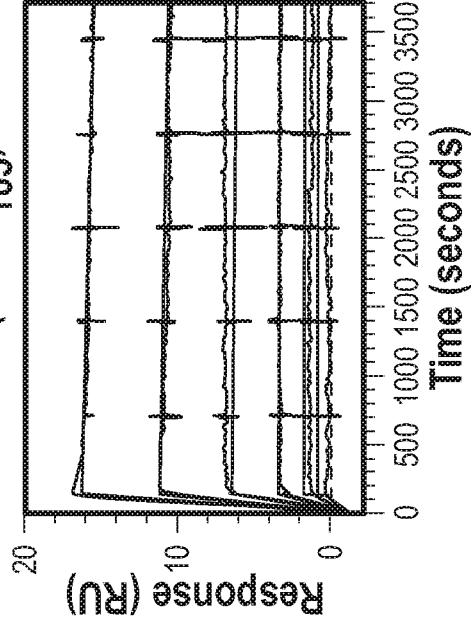


FIG. 37

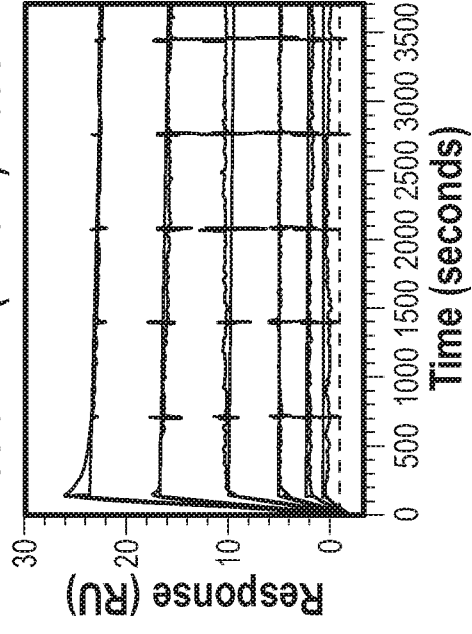
Loading Solution



Column 1 (VEGF₁₆₅) Elution



Column 2 (mAb 1) Elution



36/60

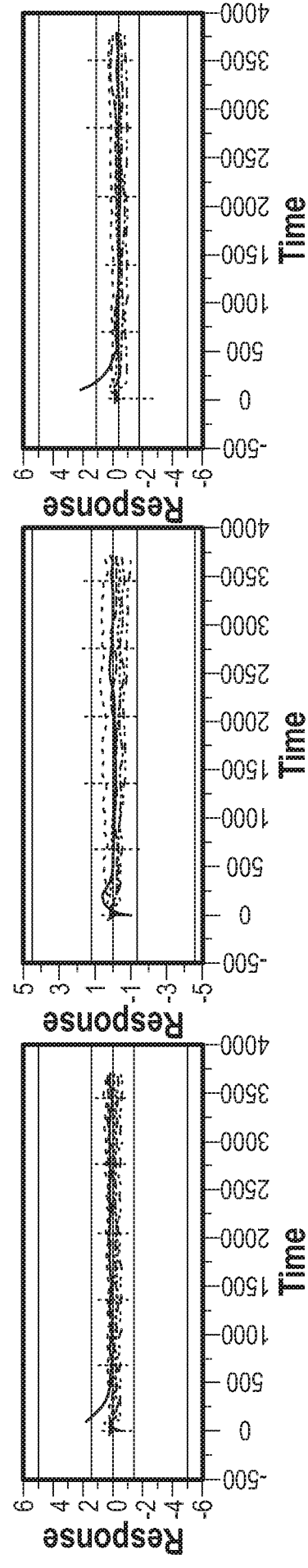


FIG. 38

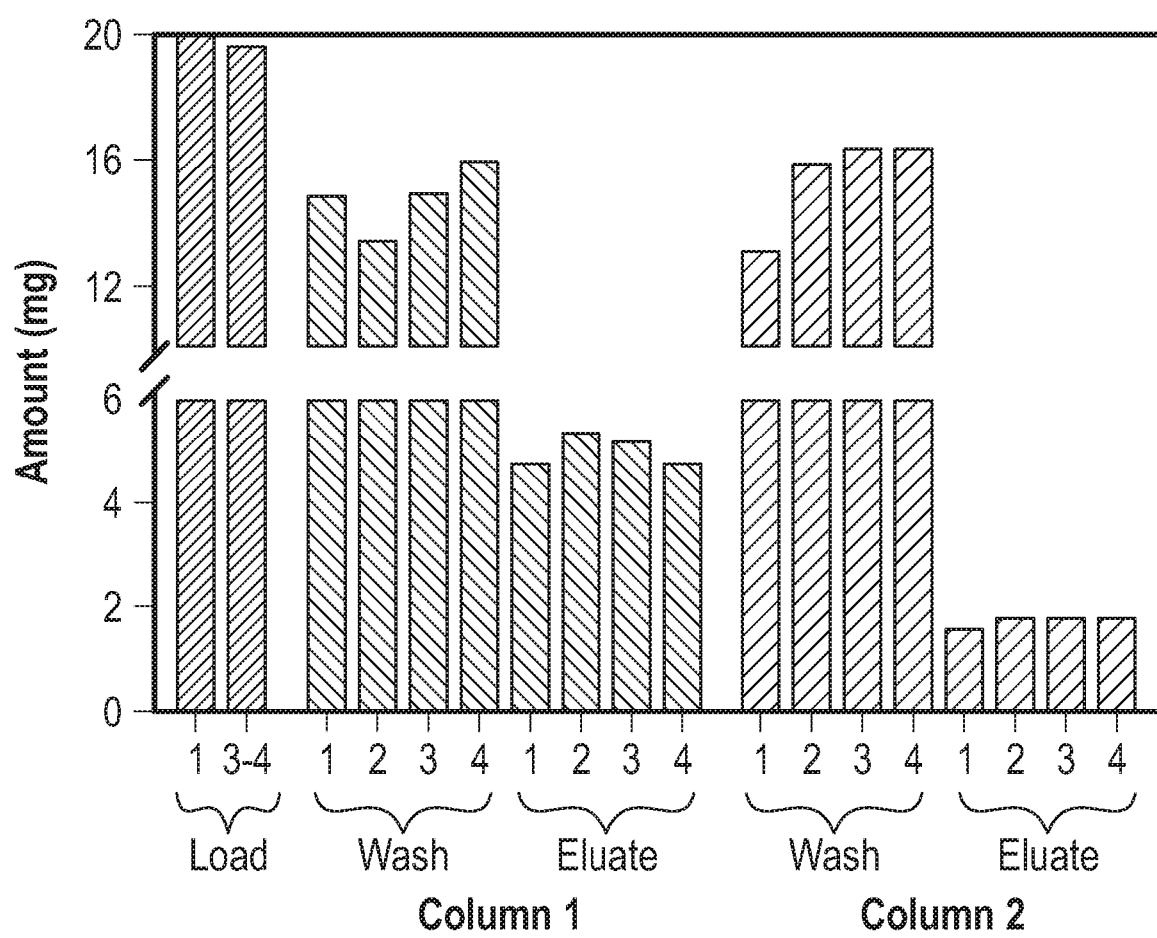


FIG. 39

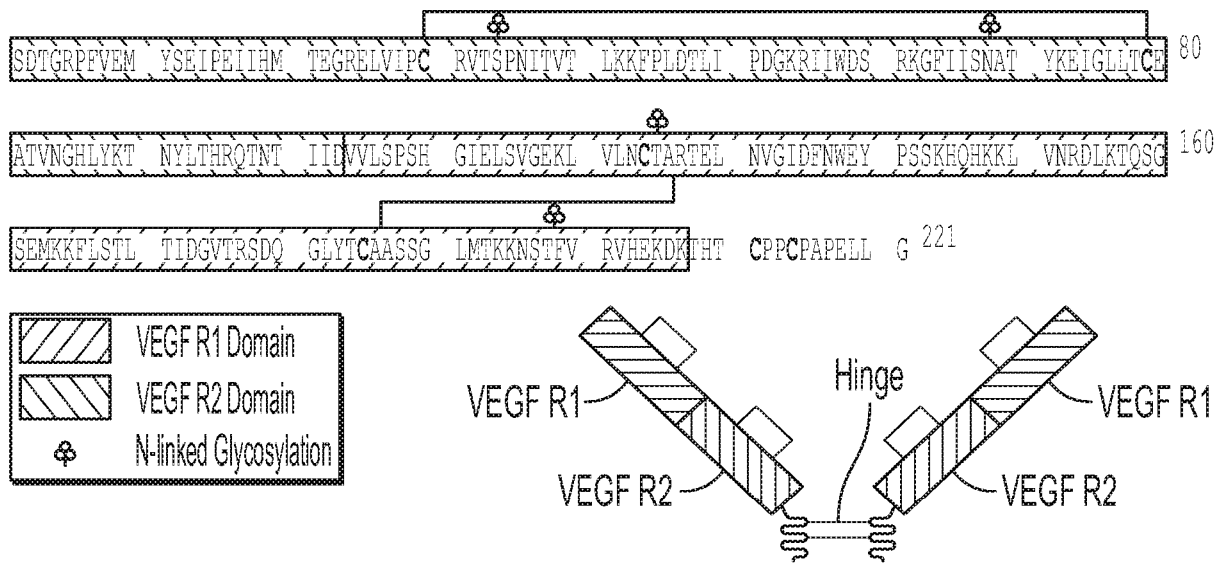


FIG. 40

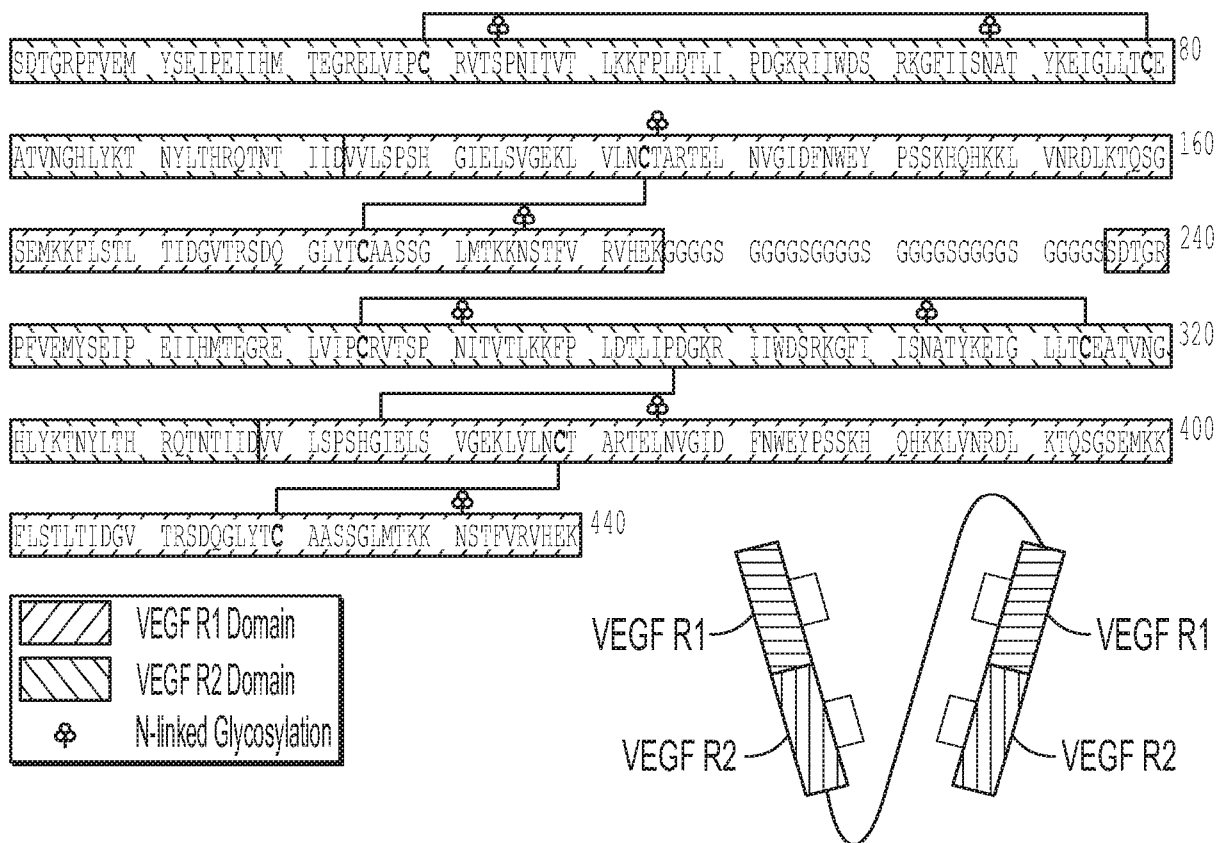
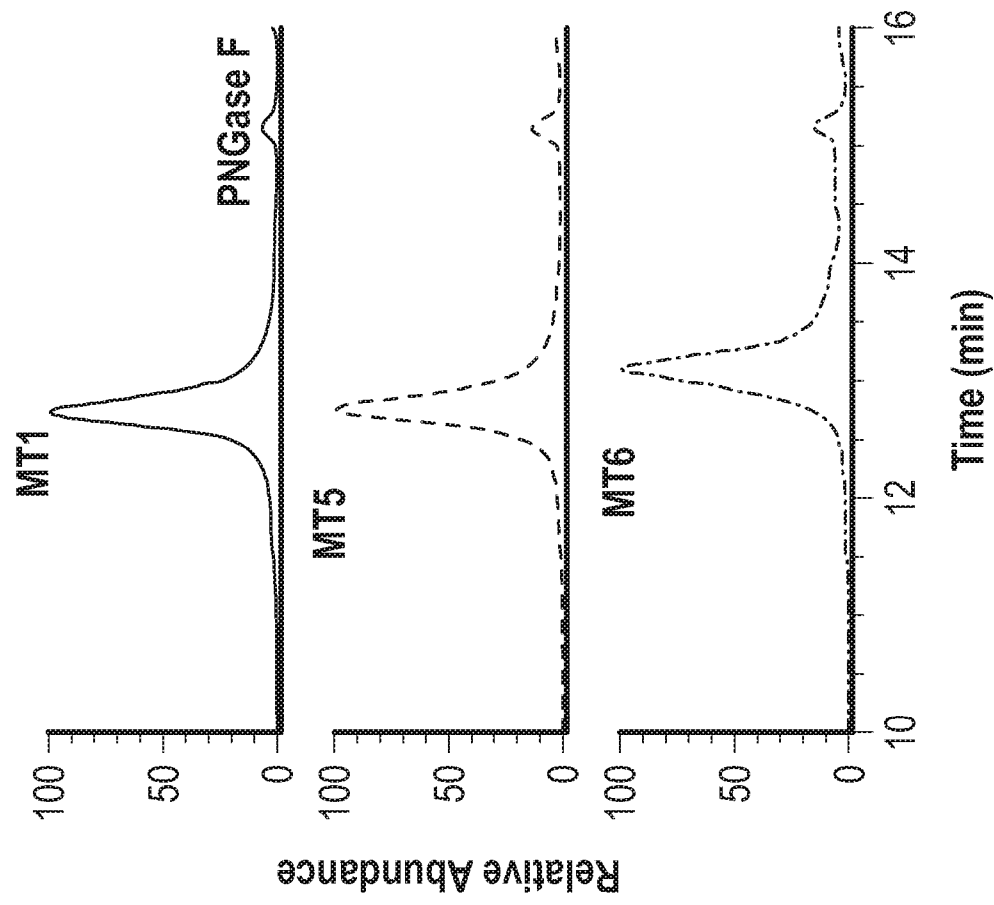


FIG. 41

TICs of Native SEC-MS Analysis



Zoom-in LMW Region:

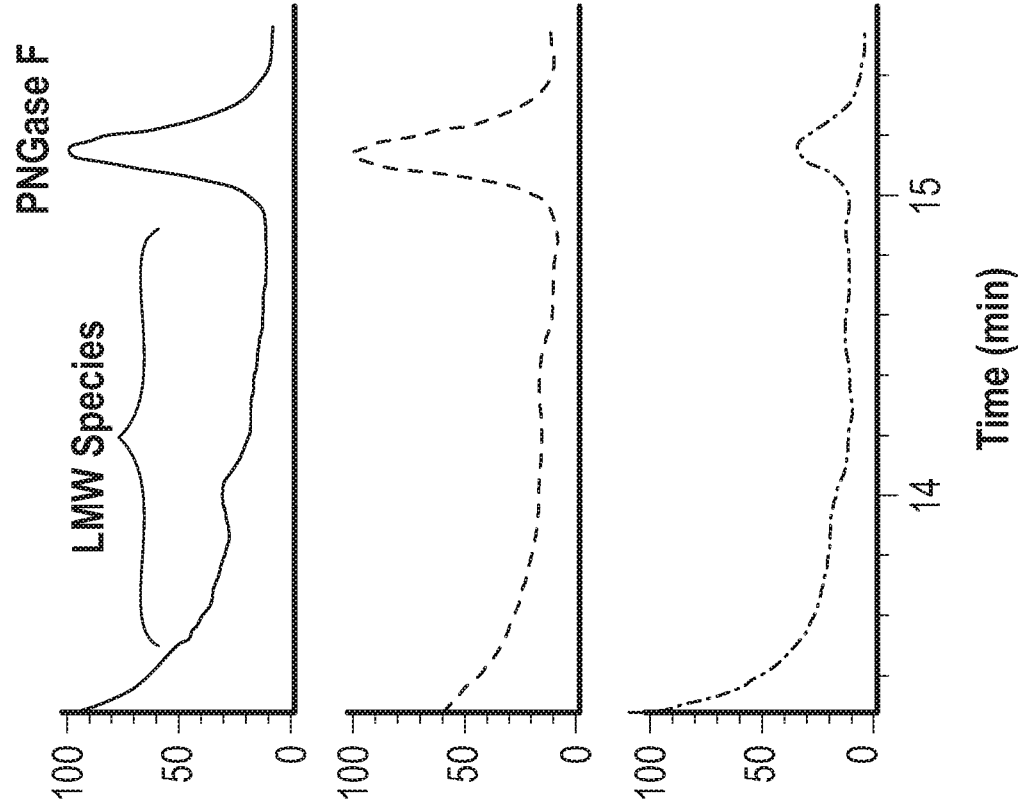


FIG. 42

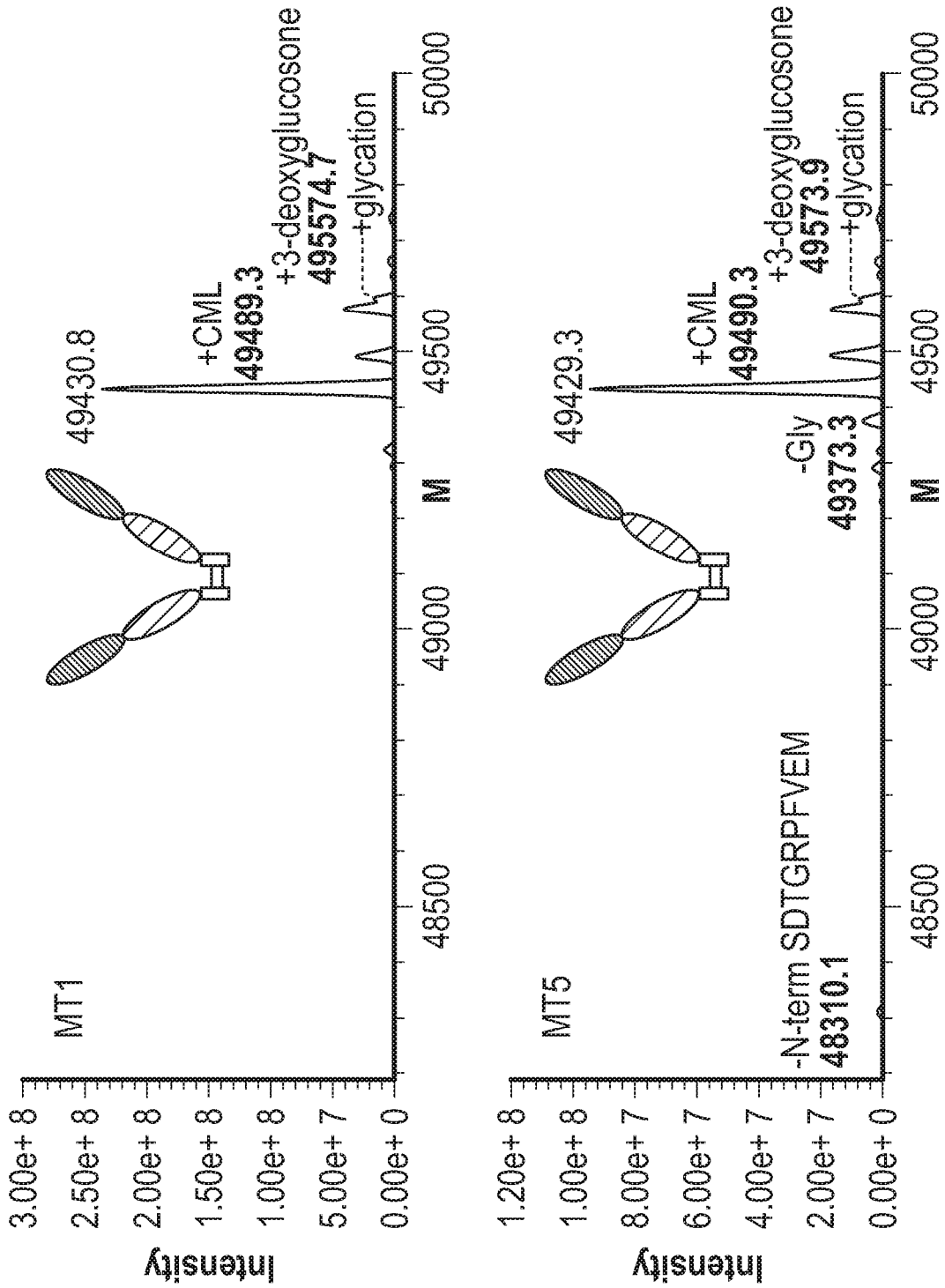


FIG. 43

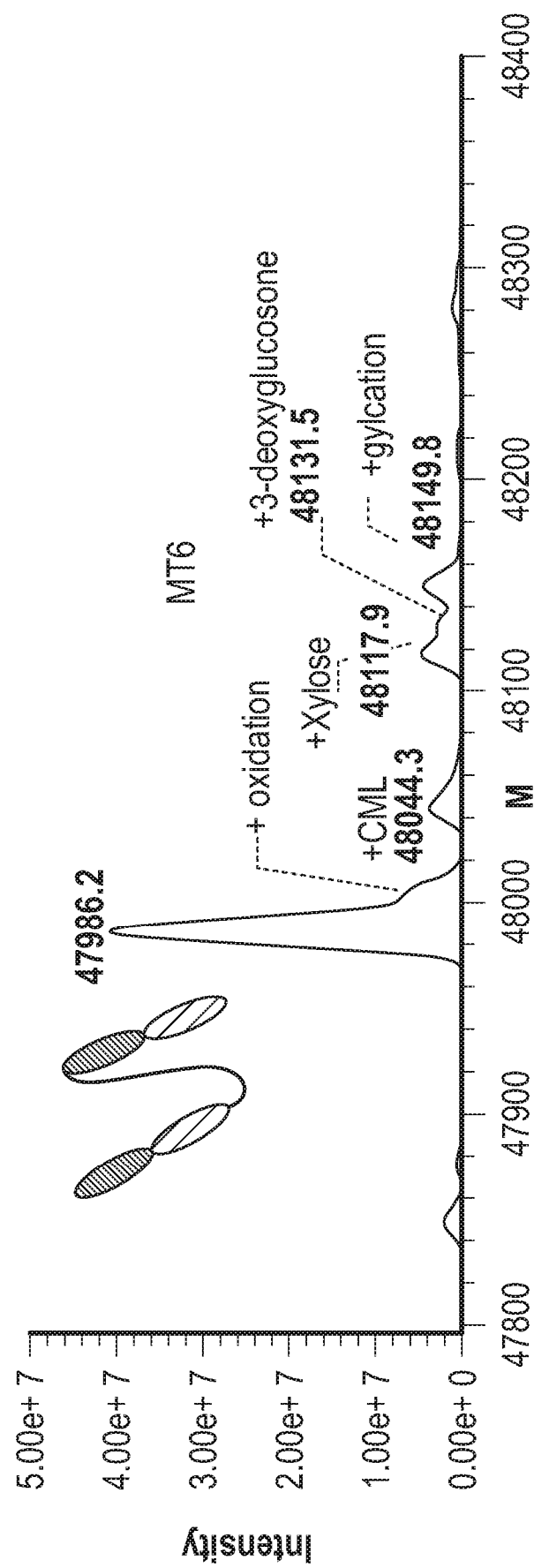


FIG. 44

42/60

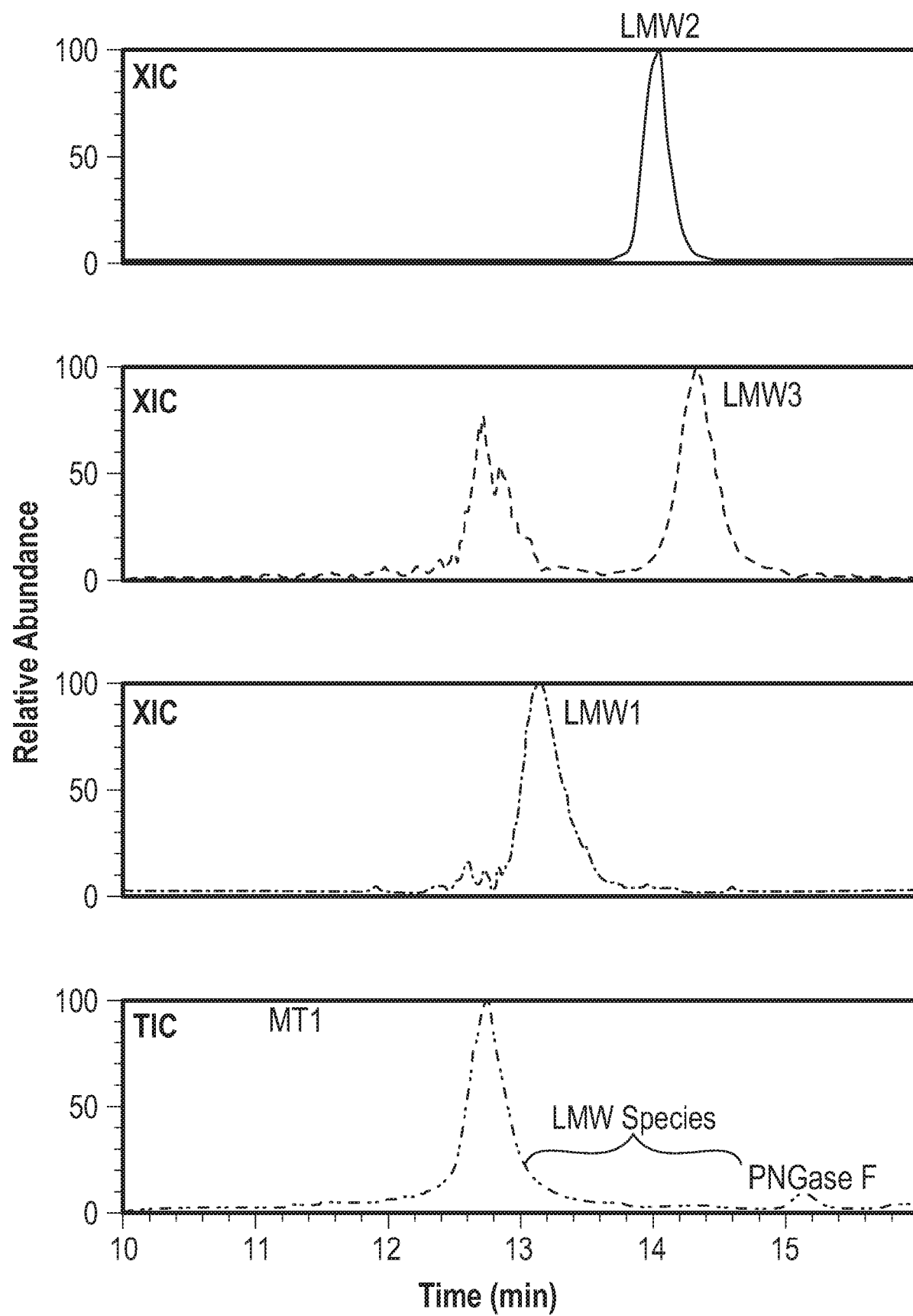


FIG. 45A

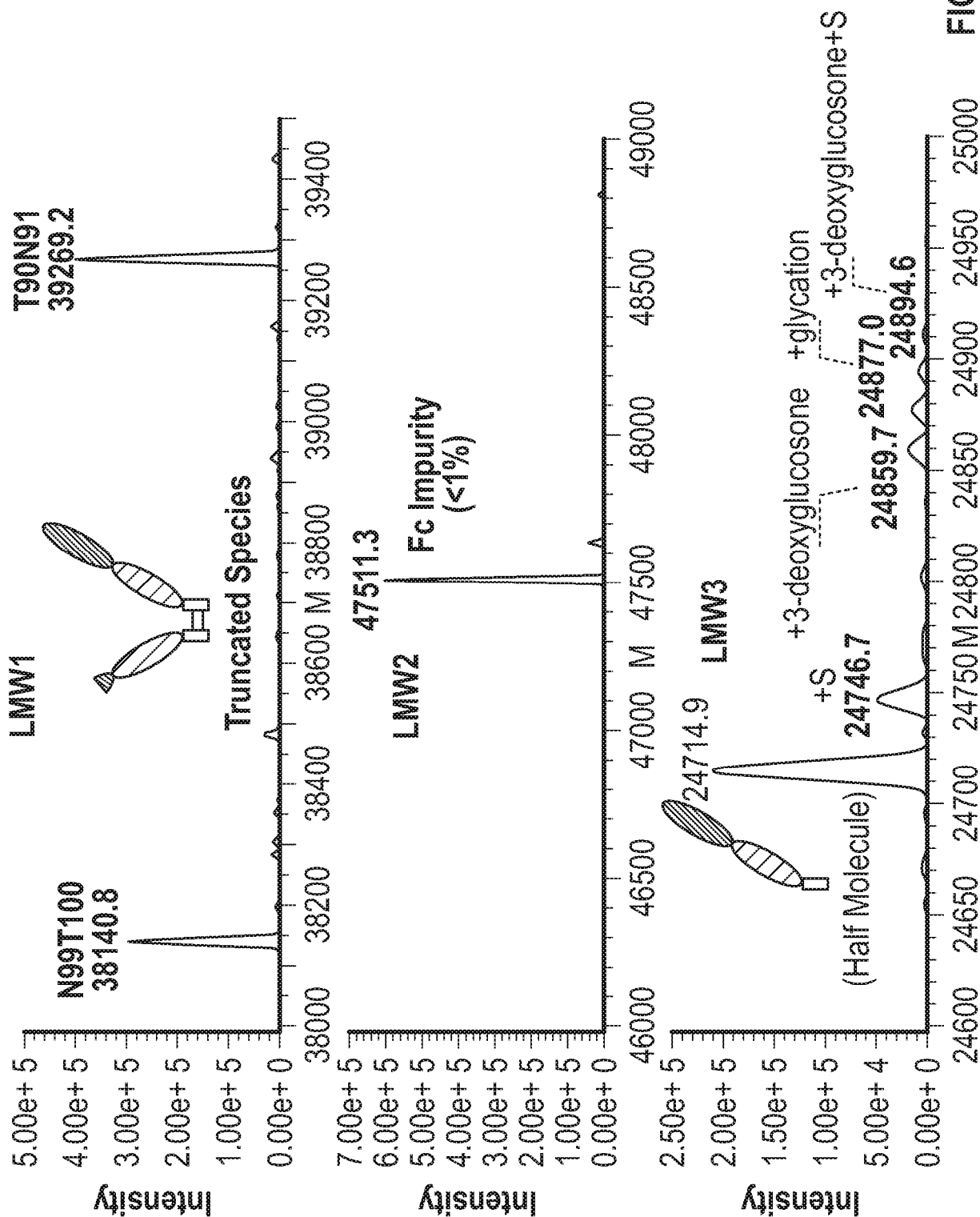


FIG. 45B

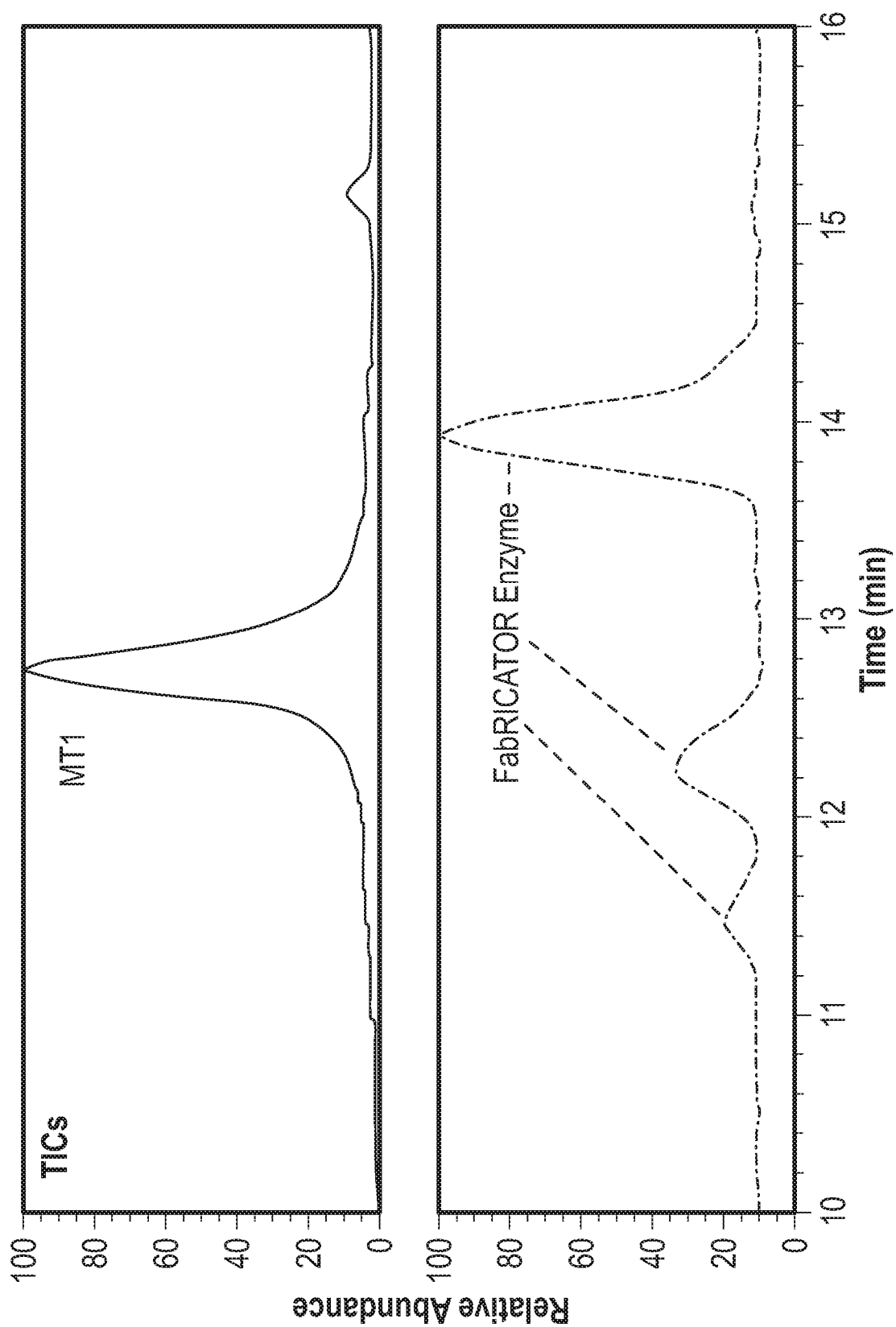


FIG. 46

45/60

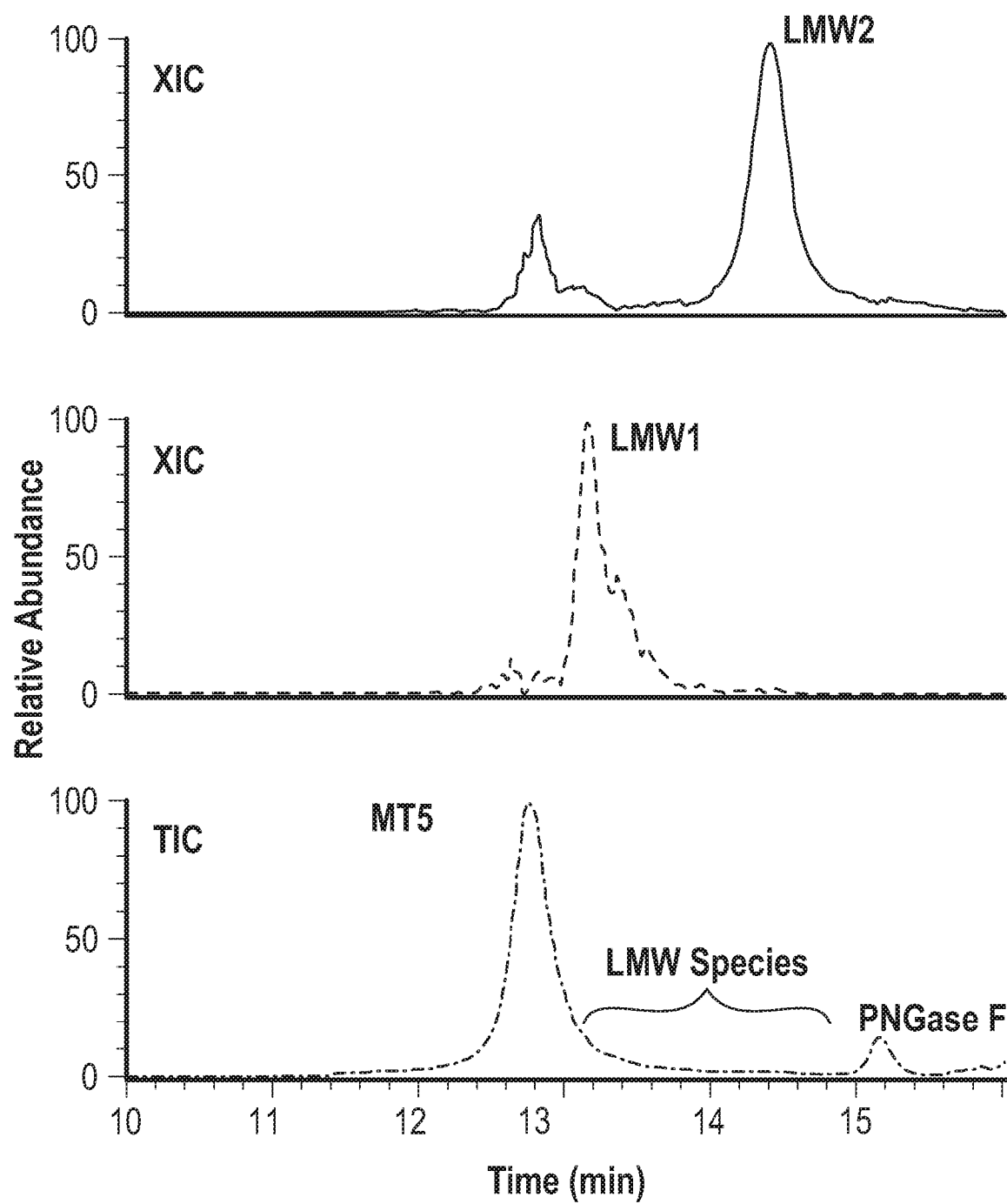


FIG. 47

46/60

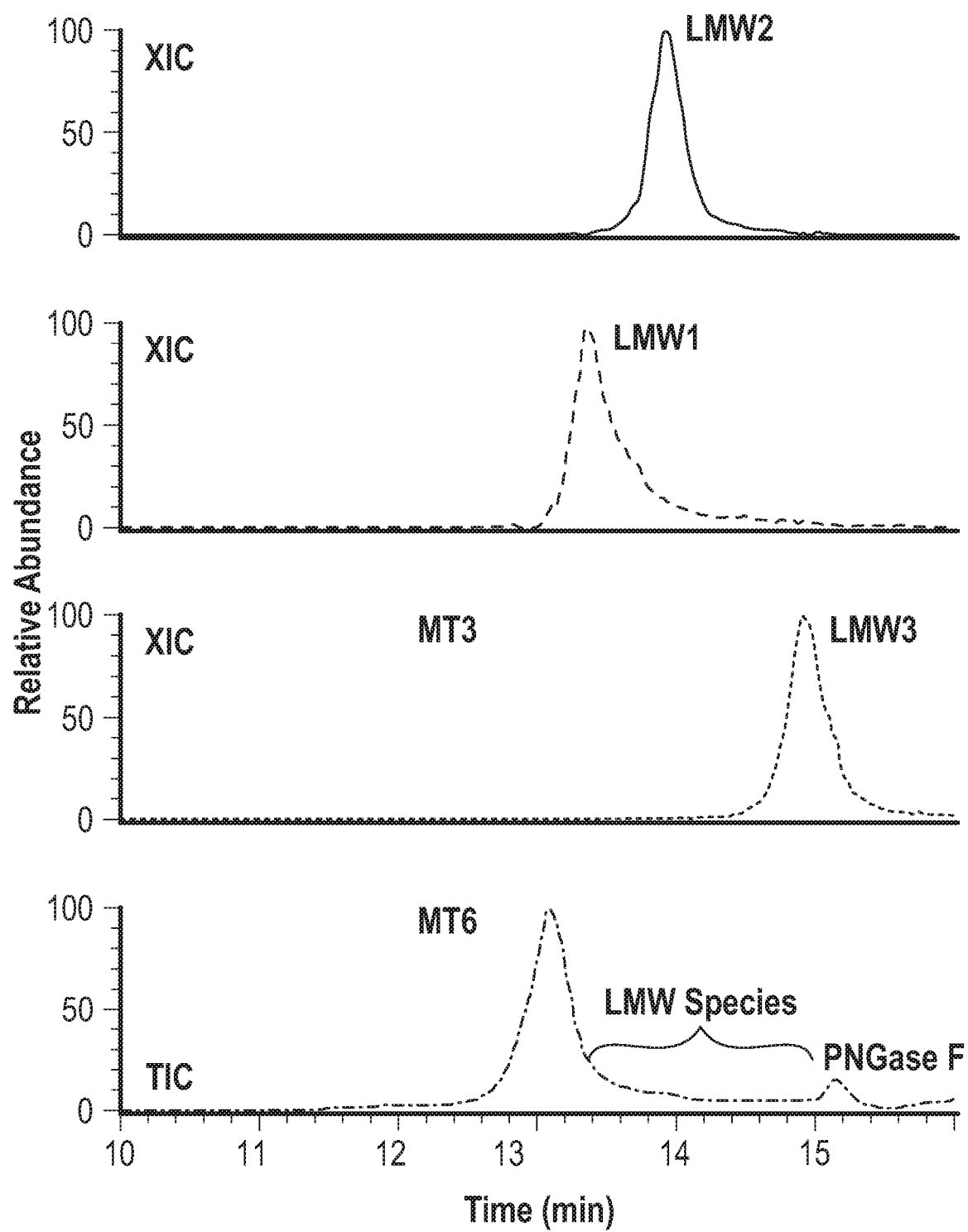


FIG. 48

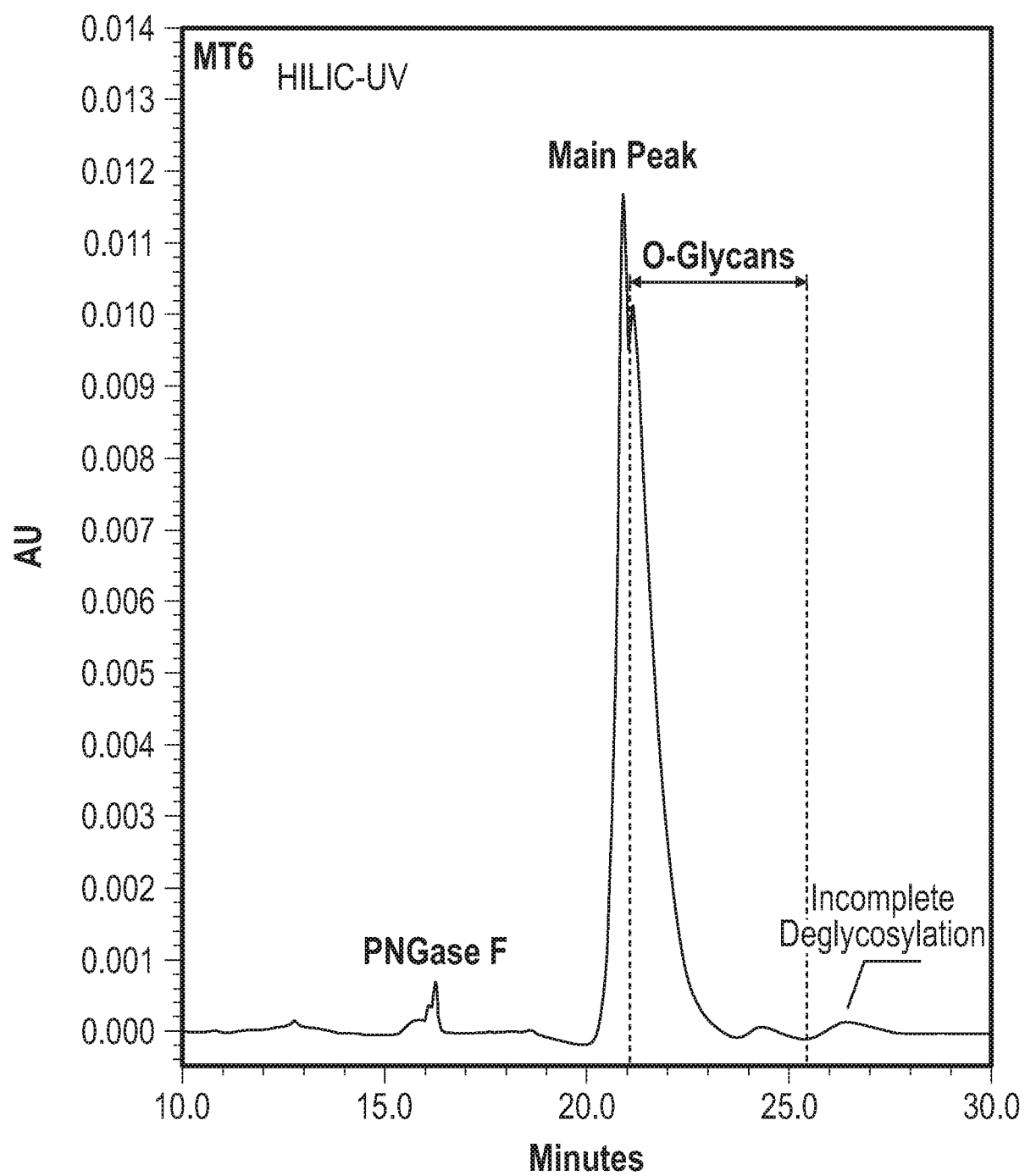
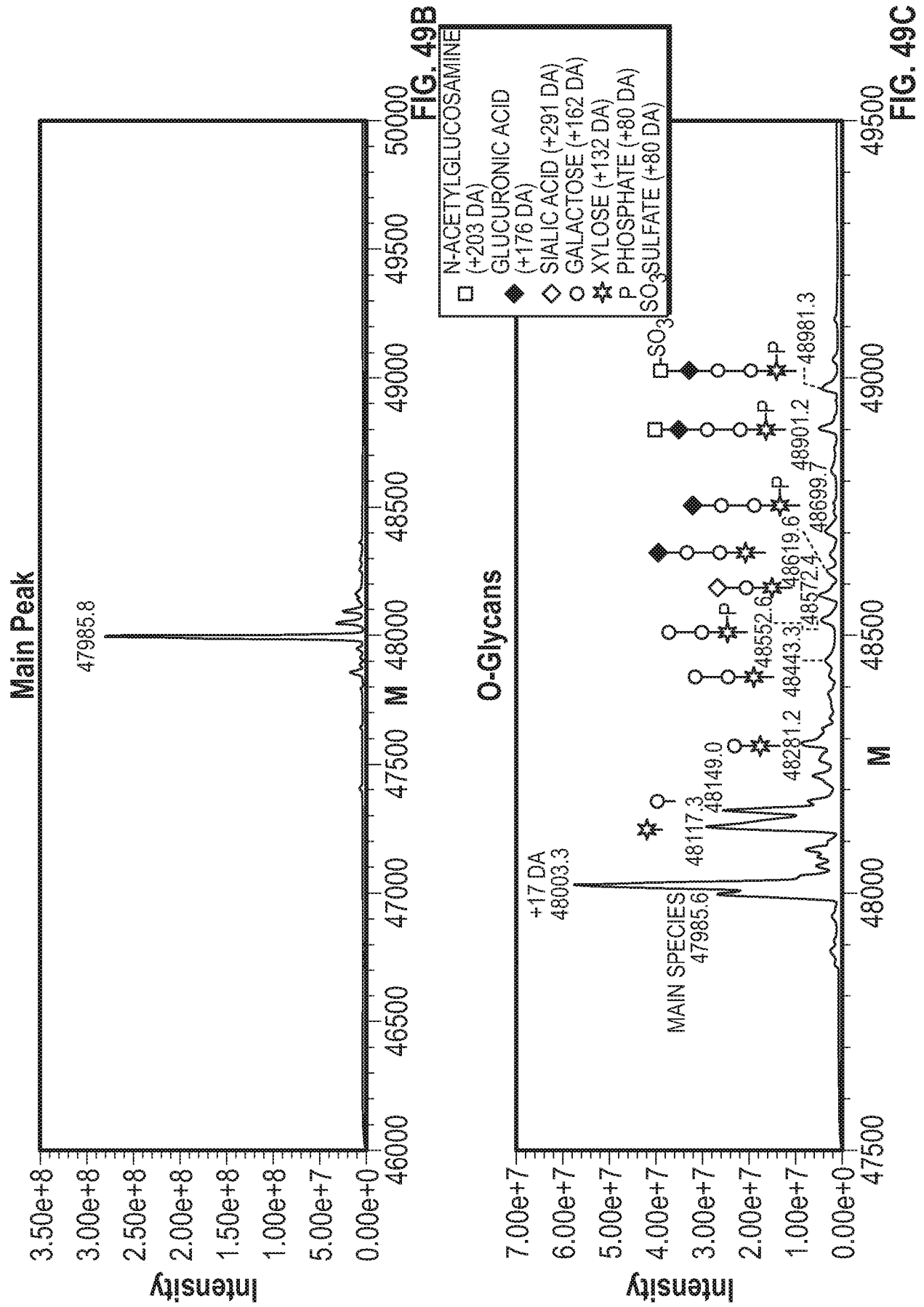


FIG. 49A



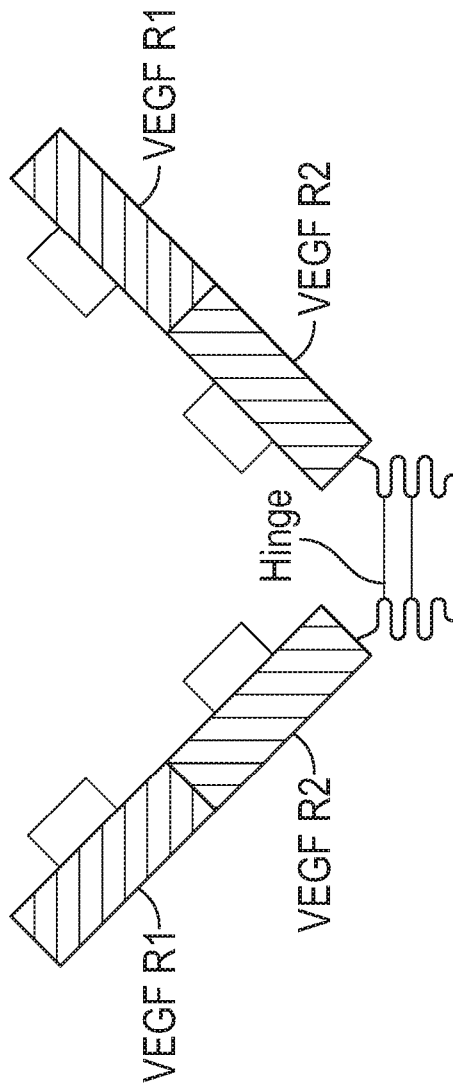
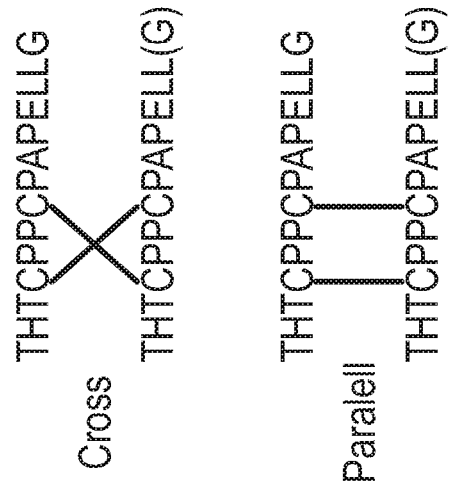


FIG. 50



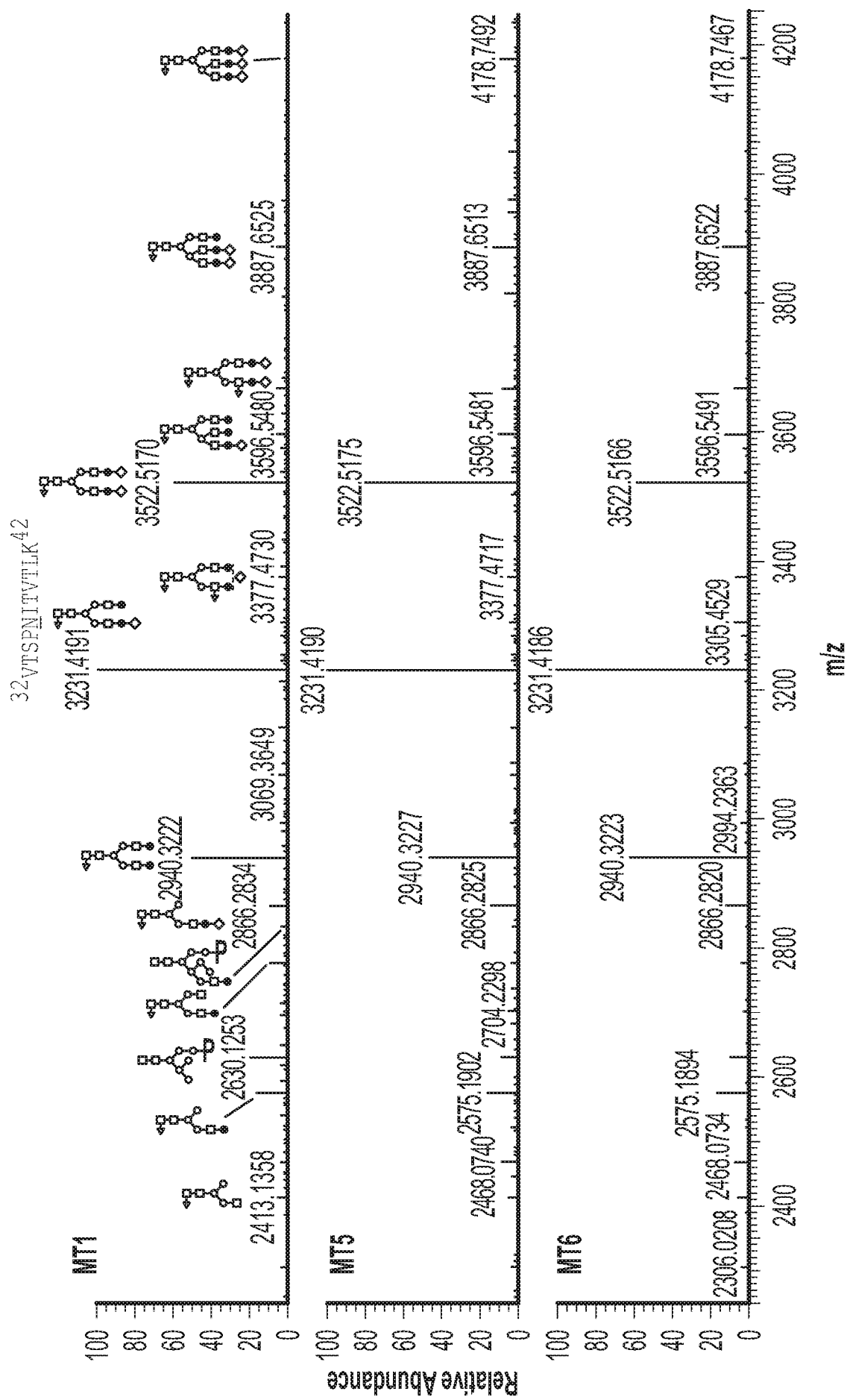


FIG. 51

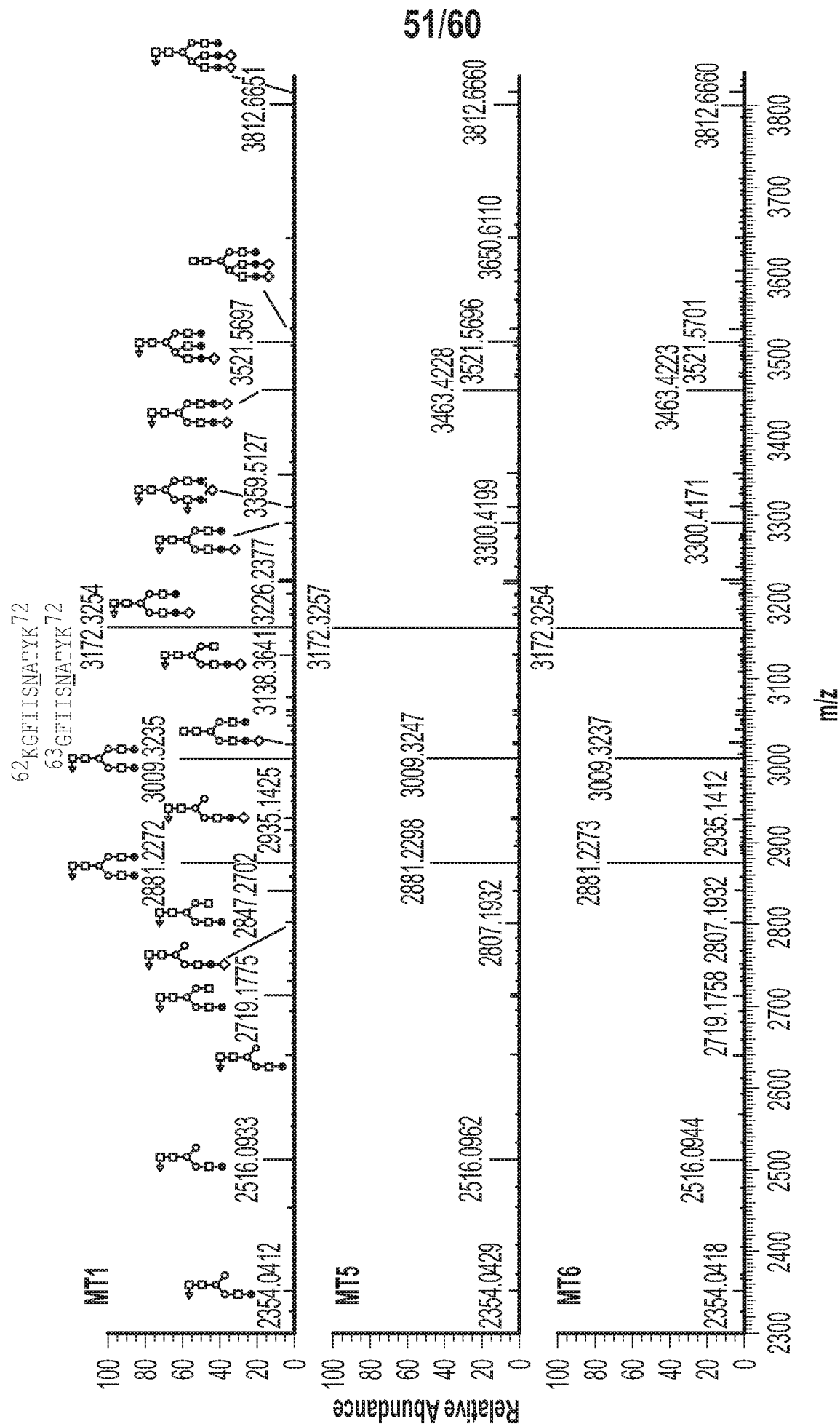


FIG. 52

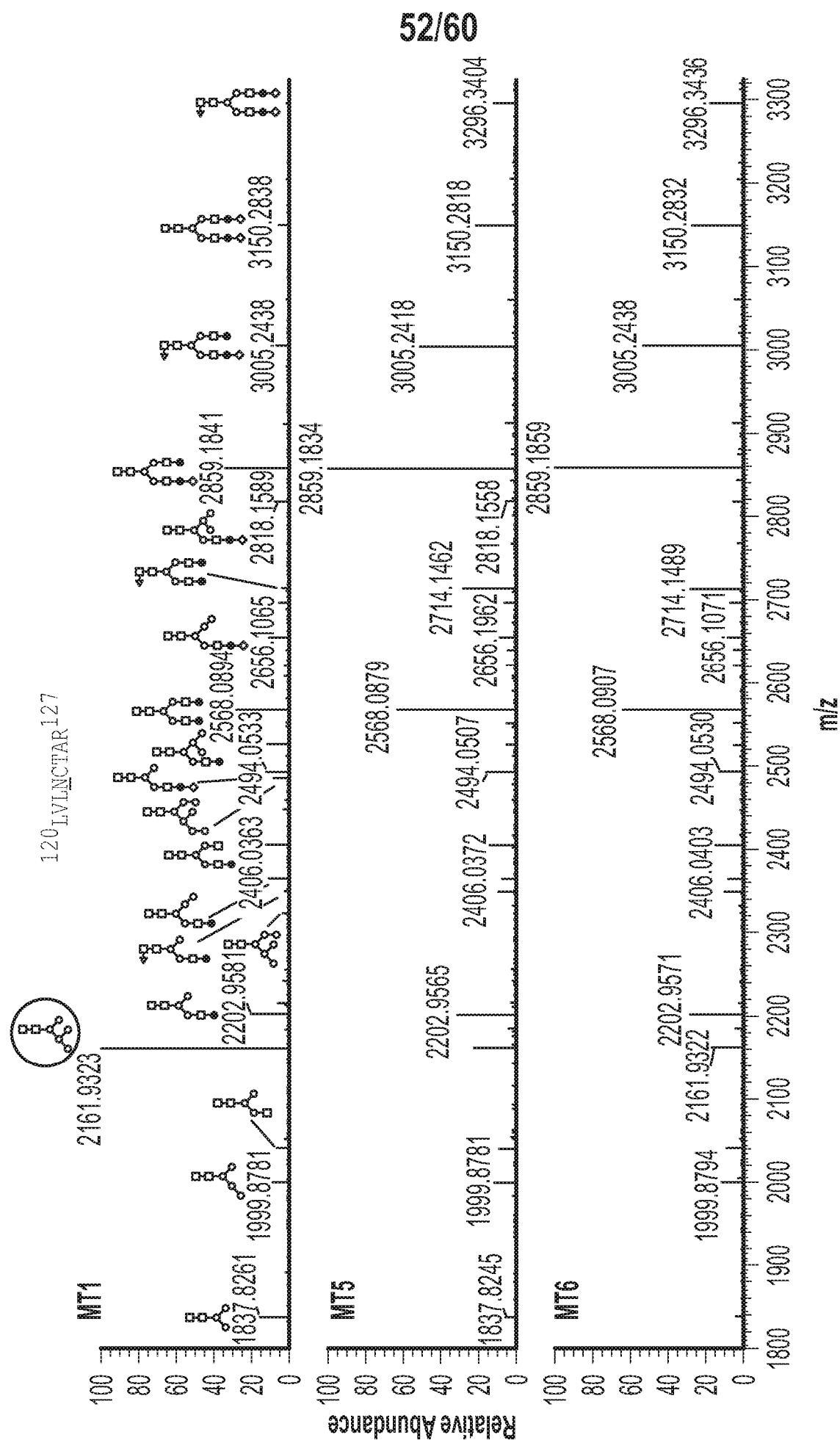


FIG. 53

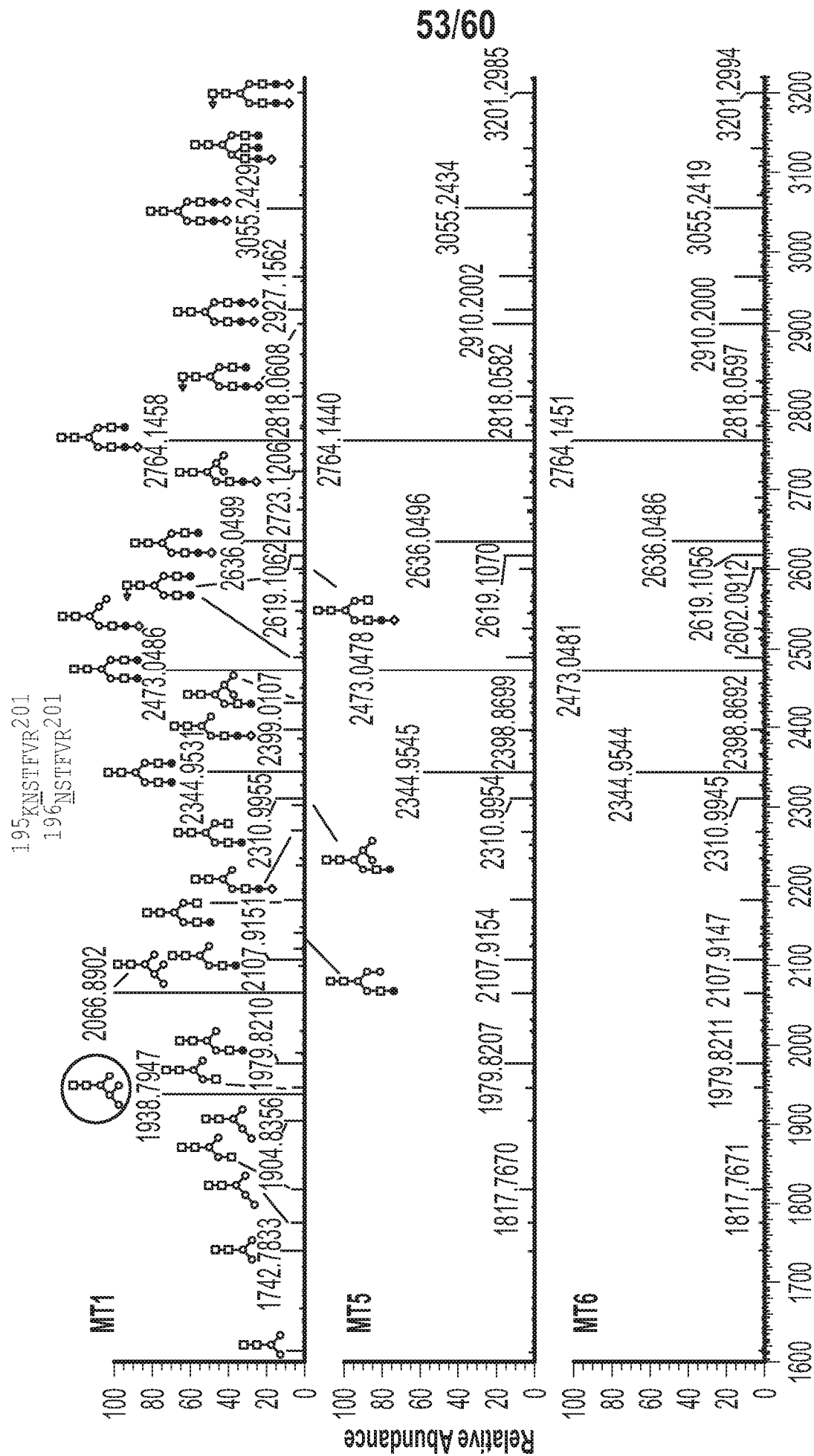


FIG. 54

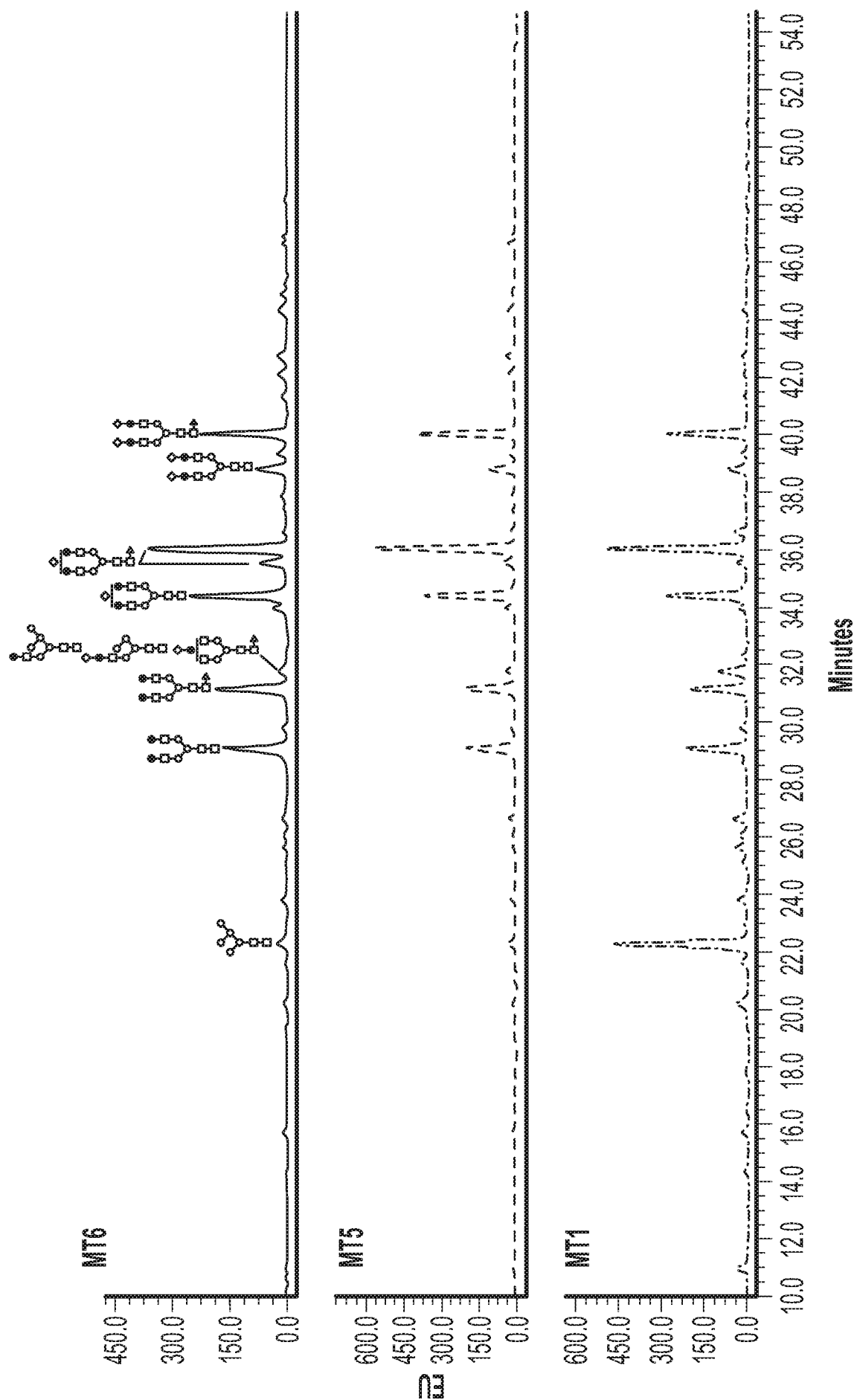


FIG. 55

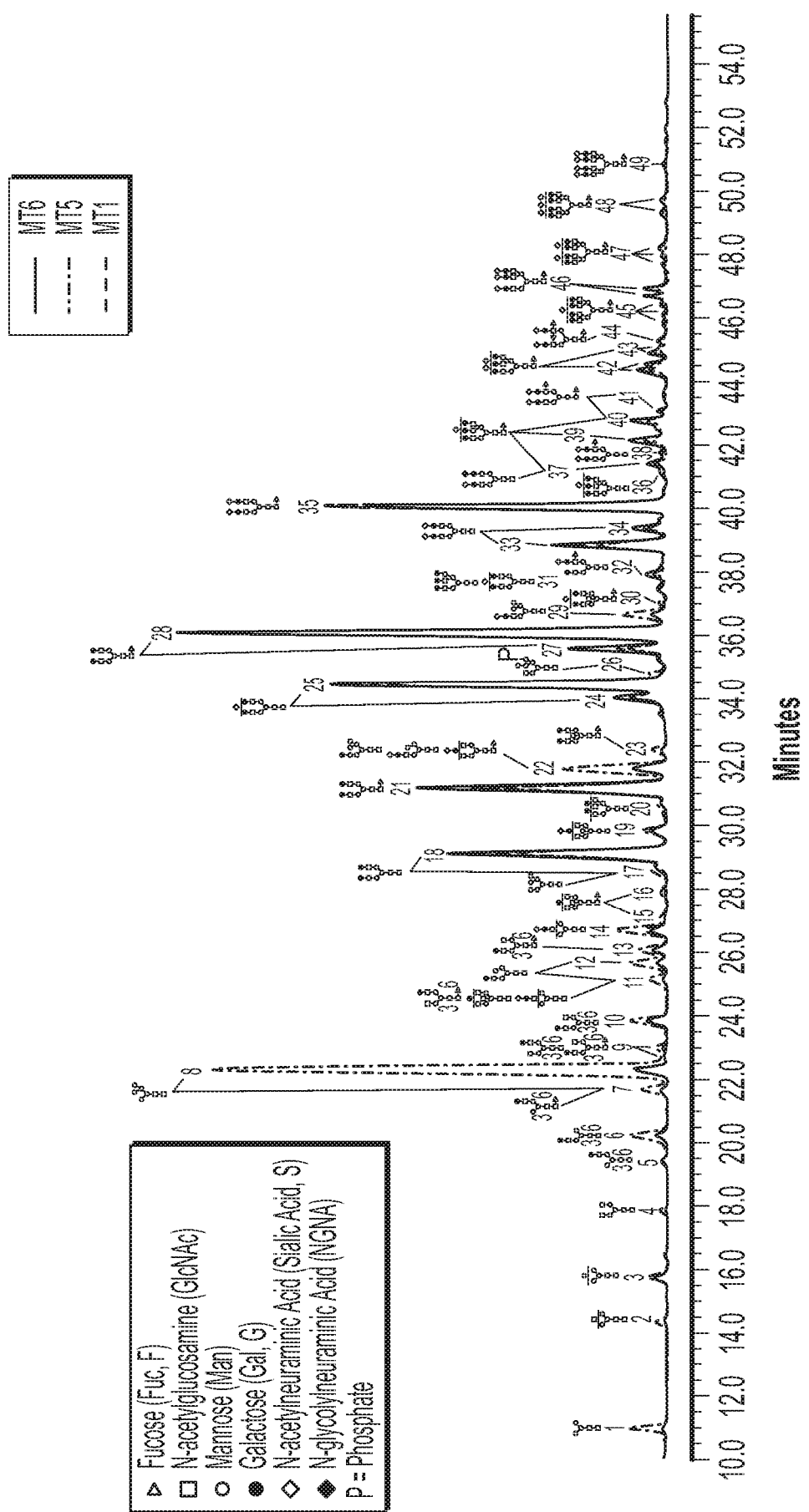
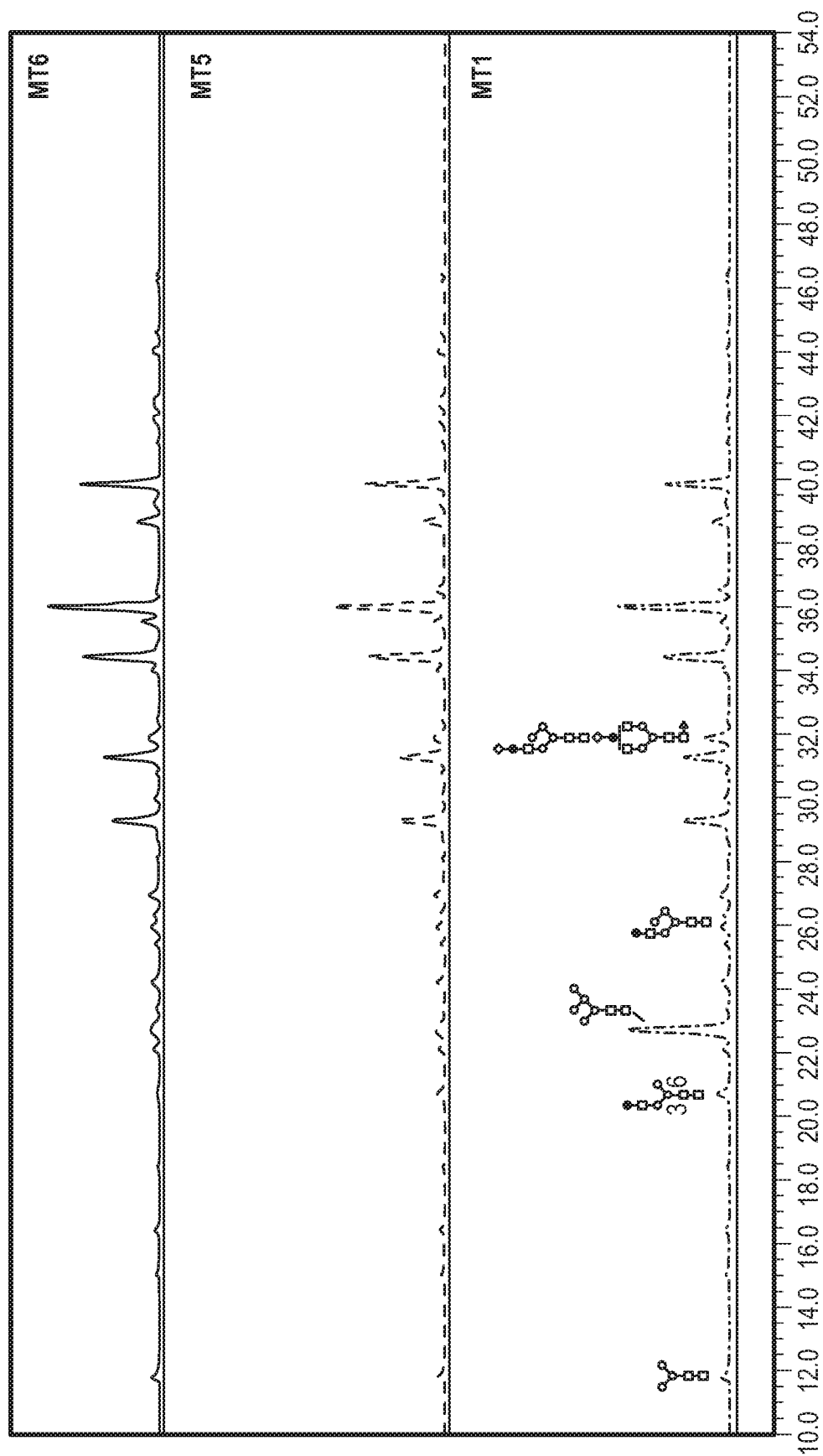


FIG. 56



Minutes

FIG. 57

Peak No.	Abbreviation	Glycan Structure	% Peak Area			Theoretical Mass (Da)
			MT6	MT5	MT1	
1	G0-2GlcNAc		0.11	0.19	0.96	1221.50
2	G0-GlcNAc		0.09	0.11	0.34	1424.58
3	Man4		0.51	0.56	0.62	1383.56
4	G0		0.07	0.05	0.26	1627.66
5	G1-GlcNAc (1-6)		0.26	0.31	0.14	1586.63
6	G1-GlcNAc (1-3)		0.55	0.61	1.76	1586.63
7	G1F-GlcNAc (1-6)		0.34	0.37	0.92	1732.69
8	Man5		1.32	1.63	16.11	1545.61
9	G1F-GlcNAc (1-3)		0.35	0.26	0.45	1732.69
	G1 (1-6)					1789.71
10	G1 (1-3)		0.89	0.88	1.34	1789.71
11	bG1		0.26	0.22	0.78	1992.79
	G1S1-GlcNAc					1877.73
	G1F (1-6)					1935.77
	Man4_G1		0.51	0.46	1.53	1748.69
12						
13	G1F (1-3)		0.67	0.64	1.11	1935.77
14	G1S1-GlcNAc		0.73	1.20	1.86	1877.73
15	bG1F		0.00	0.00	0.32	2138.85

FIG. 58A

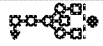
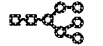
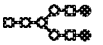
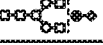
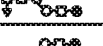
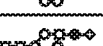
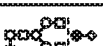
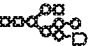
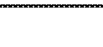
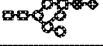
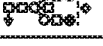
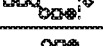
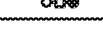
Peak No.	Abbreviation	Glycan Structure	% Peak Area			Theoretical Mass (Da)
			MT6	MT5	MT1	
16	bG1F		0.00	0.00	0.40	2138.85
17	Man6		0.51	0.41	0.85	1707.66
	G2					1951.77
18			9.95	8.85	7.96	
19	G1S1-GlcNAc		0.84	0.89	1.04	2080.81
20	G2+GlcNAc		0.28	0.28	0.57	2154.85
21	G2F		11.24	9.13	7.47	2097.82
22	Man5_G1		1.61	1.82	4.13	1910.74
	Man4_G1S1					2039.78
	G1FS1					2226.87
23	bG2F		0.27	0.32	0.66	2300.90
24	G2S1+Fuc		2.63	2.13	1.75	2242.86
25			13.42	14.46	9.50	
26	Man6_G0+Phos		0.00	0.00	0.53	1787.63
27	G2FS1		4.00	2.43	1.67	2388.92
28			18.85	21.19	15.94	2388.92
29	Man5_G1S1		0.51	0.62	1.70	2201.84
30	bG2FS1		0.00	0.00	0.27	2592.00
31	G2+NGNA		0.44	0.35	0.35	2258.86
	G3					2316.90

FIG. 58B

Peak No.	Abbreviation	Glycan Structure	% Peak Area			Theoretical Mass (Da)
			MT6	MT5	MT1	
32	G2S1+Fuc		0.85	0.59	0.38	2388.92
33	G2S2		4.56	4.36	2.57	2533.96
34			1.31	0.99	0.71	2533.96
35	G2FS2		10.76	13.67	8.33	2680.02
36	G3S1		0.20	0.14	0.11	2607.99
37	G2S1+NGNA		0.87	0.59	0.35	2549.95
	G3FS1					2754.05
38	G2S2+Fuc		0.23	0.13	0.07	2680.02
39	G3FS1		1.61	1.22	0.67	2754.05
40			1.77	1.77	0.95	2754.05
41	G2FS2+Fuc		0.24	0.24	0.15	2826.07
42	G3FS2		1.85	1.59	0.77	3045.15
43			0.78	0.77	0.32	
44	G2FS2+2Fuc		0.34	0.31	0.14	2972.13
45	G4FS1		0.57	0.29	0.12	3119.18
46	G3FS3		1.14	1.43	0.58	3336.24
47	G4FS2		0.89	0.72	0.24	3410.28
48	G4FS3		0.63	0.60	0.18	3701.38
49	G4FS4		0.22	0.23	0.07	3992.47

FIG. 58C

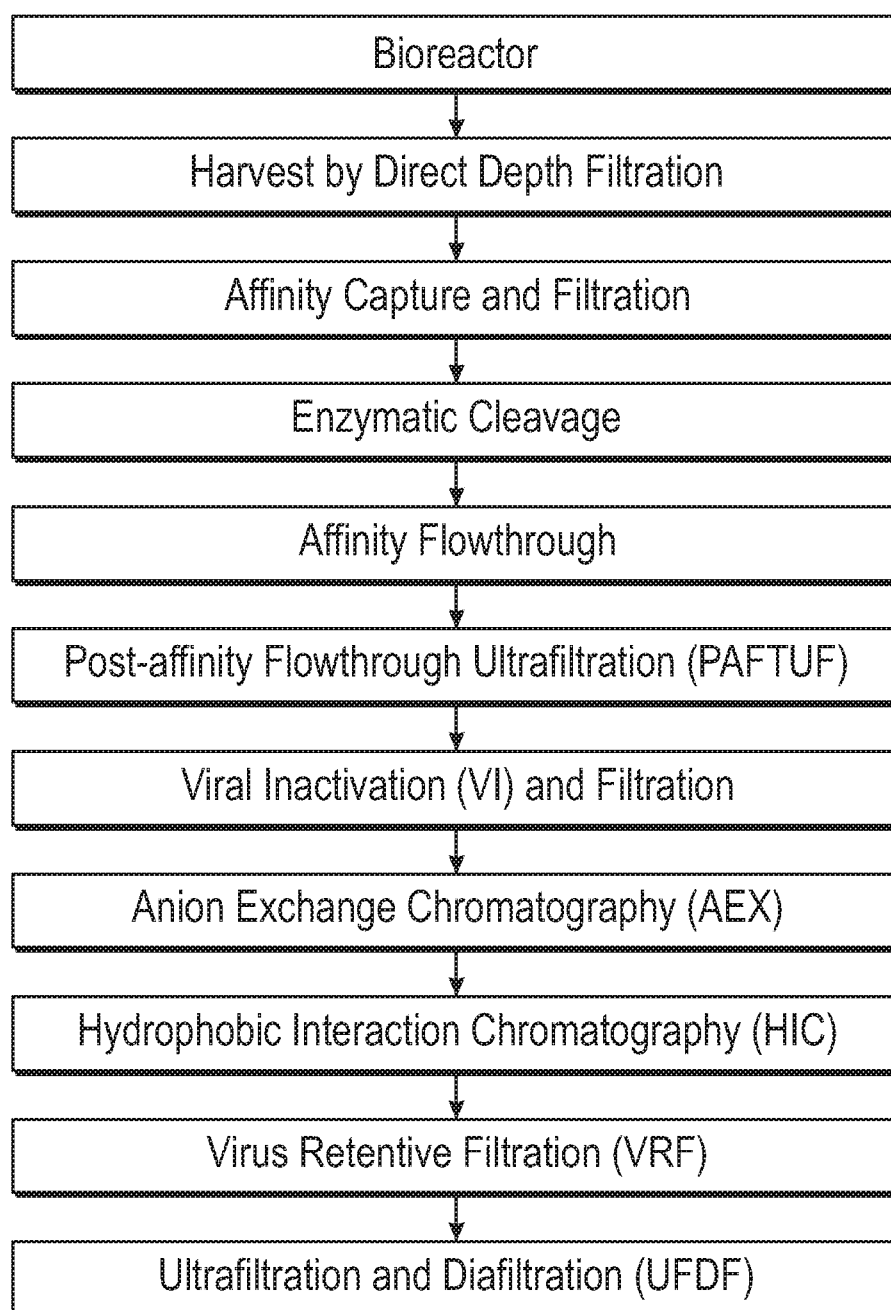


FIG. 59