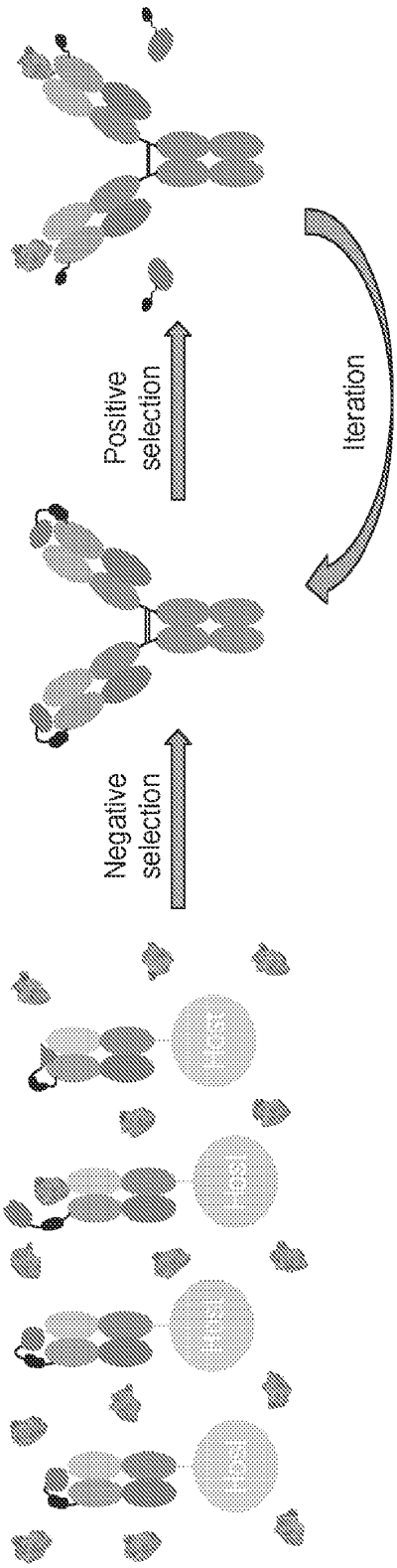


FIG. 1

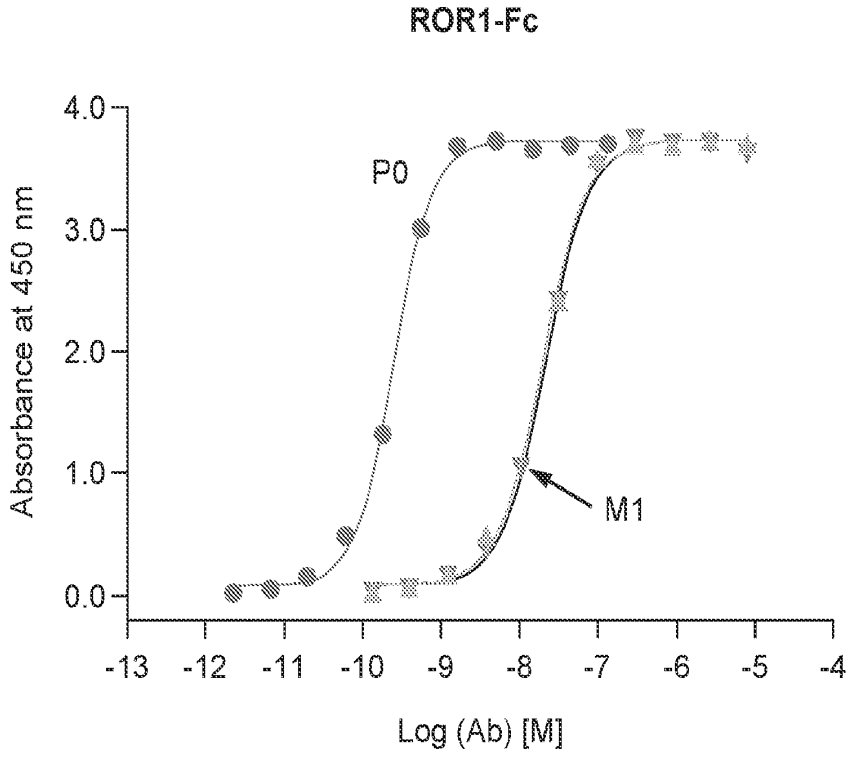

ROR1-Fc
Absorbance at 450 nm
4.0
3.0
2.0
1.0
0.0
P0
M1
-13
-12
-11
-10
-9
-8
-7
-6
-5
-4
Log (Ab) [M]

FIG. 2A

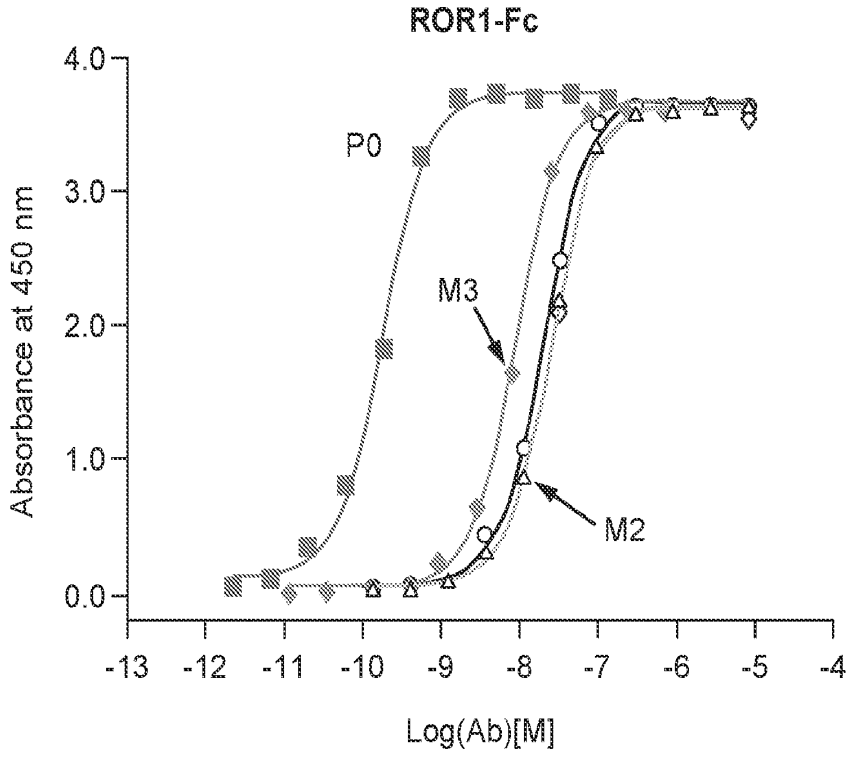

ROR1-Fc
Absorbance at 450 nm
4.0
3.0
2.0
1.0
0.0
P0
M3
M2
-13
-12
-11
-10
-9
-8
-7
-6
-5
-4
Log(Ab)[M]

FIG. 2B

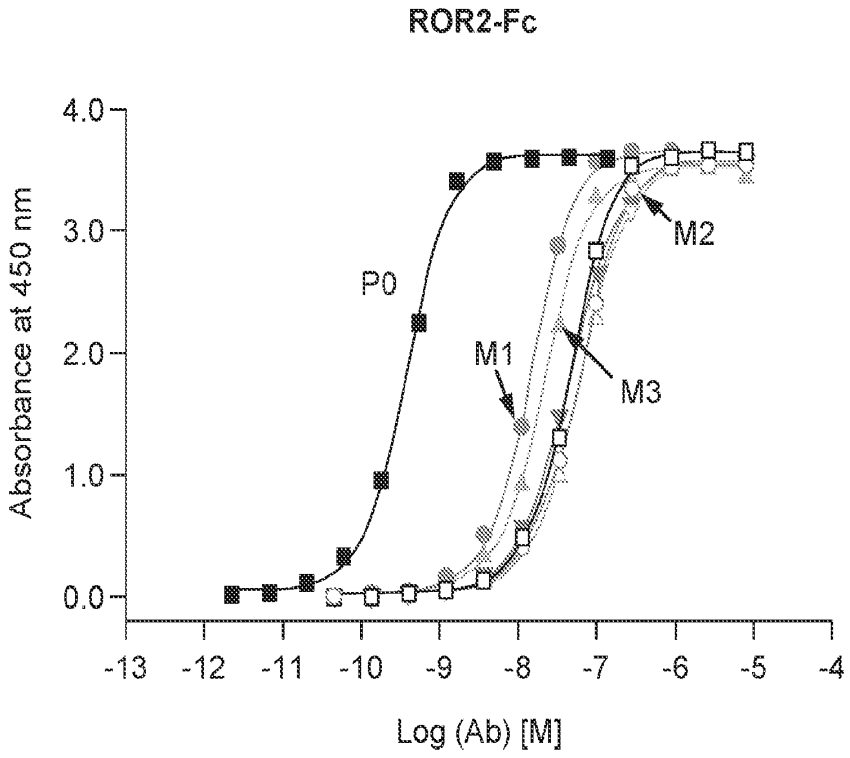

ROR2-Fc
Absorbance at 450 nm
4.0
3.0
2.0
1.0
0.0
P0
M1
M2
M3
-13
-12
-11
-10
-9
-8
-7
-6
-5
-4
Log (Ab) [M]

FIG. 2C

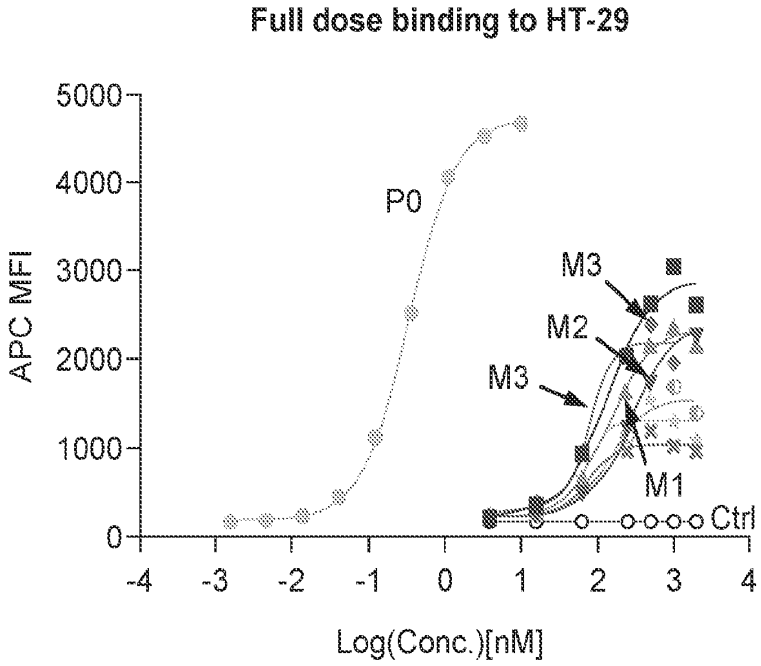

Full dose binding to HT-29
APC MFI
5000
4000
3000
2000
1000
0
-4
-3
-2
-1
0
1
2
3
4
Log(Conc.)[nM]
P0
M3
M2
M3
M1
Ctrl

FIG. 2D

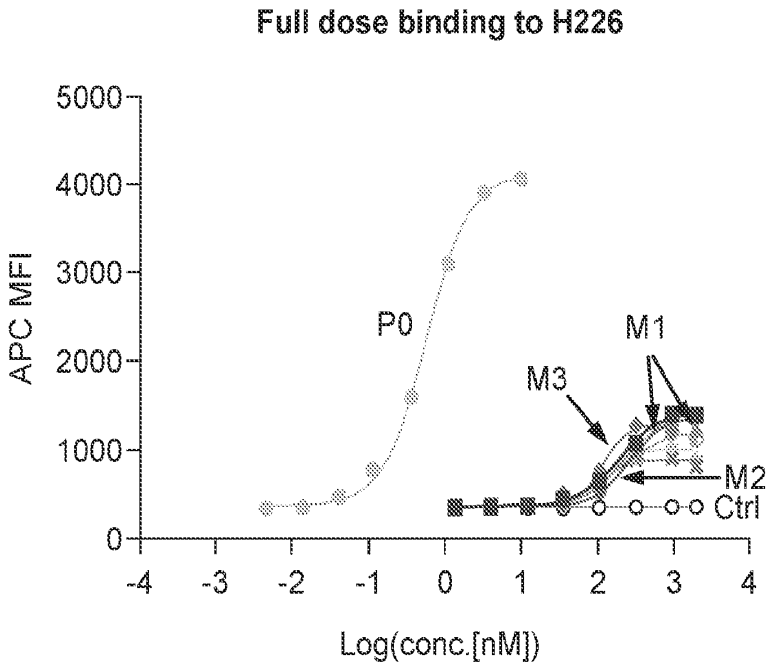

Full dose binding to H226
APC MFI
5000
4000
3000
2000
1000
0
-4
-3
-2
-1
0
1
2
3
4
Log(conc.[nM])
P0
M1
M3
M2
Ctrl

FIG. 2E

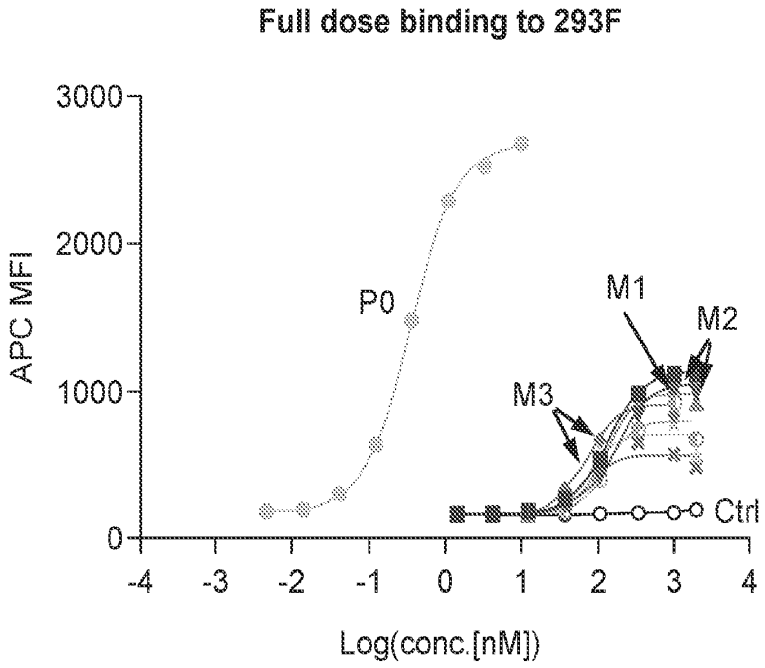

Full dose binding to 293F
3000
2000
1000
0
APC MFI
P0
M1
M2
M3
Ctrl
-4
-3
-2
-1
0
1
2
3
4
Log(conc.[nM])

FIG. 2F

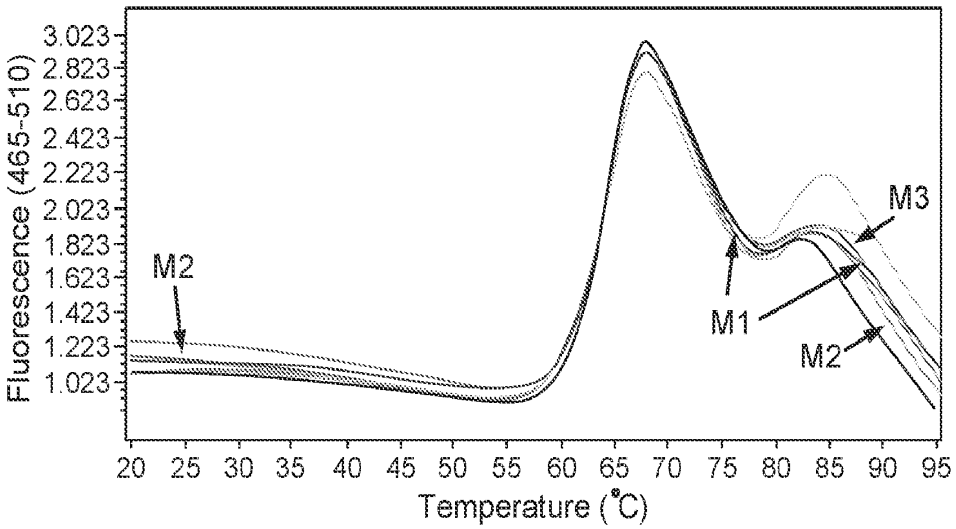

Fluorescence (465-510)
3.023
2.823
2.623
2.423
2.223
2.023
1.823
1.623
1.423
1.223
1.023
M2
M3
M1
M2
20 25 30 35 40 45 50 55 60 65 70 75 80 85 90 95
Temperature (°C)

FIG. 3A

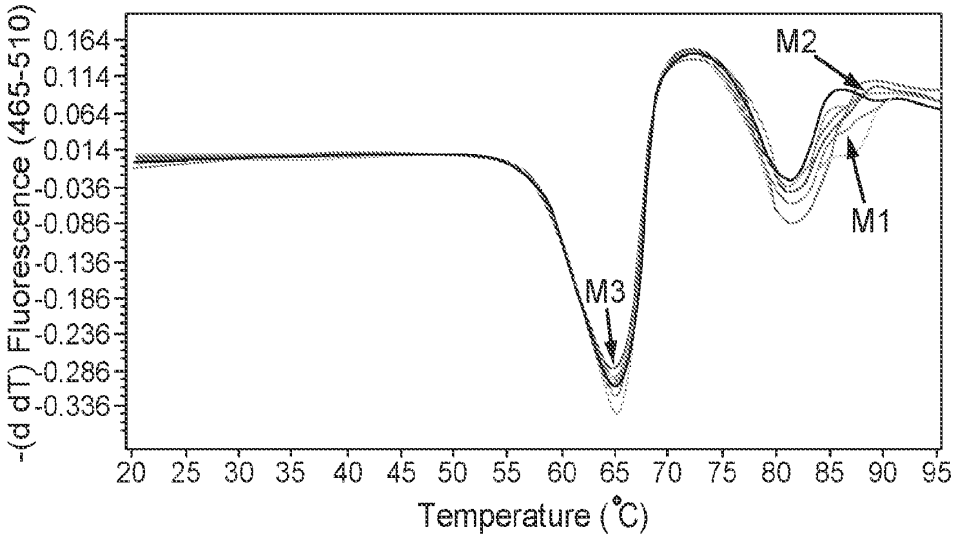

-(d dT) Fluorescence (465-510)
0.164
0.114
0.064
0.014
-0.036
-0.086
-0.136
-0.186
-0.236
-0.286
-0.336
M2
M1
M3
20 25 30 35 40 45 50 55 60 65 70 75 80 85 90 95
Temperature (°C)

FIG. 3B

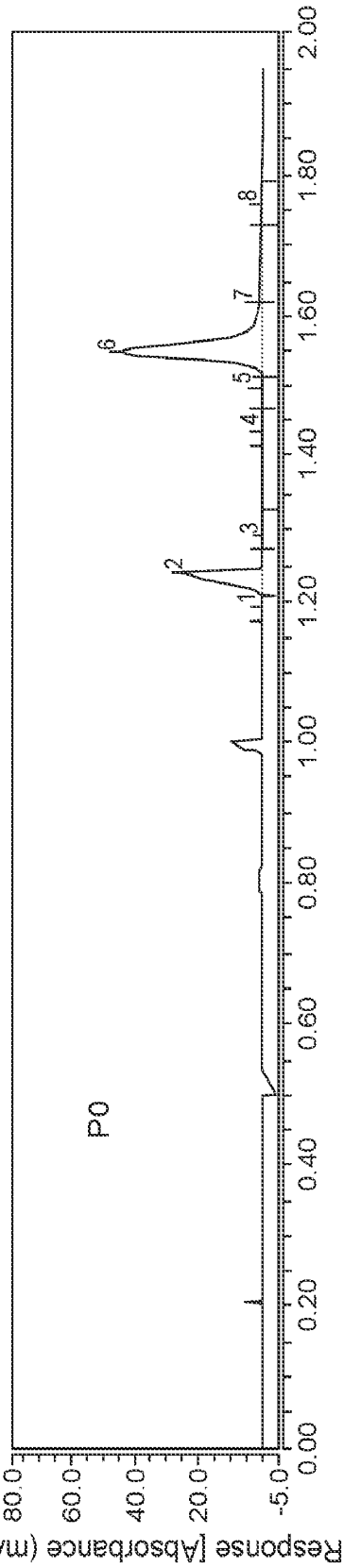

P0
Response [Absorbance (mAU)]
80.0
60.0
40.0
20.0
-5.0
0.00
0.20
0.40
0.60
0.80
1.00
1.20
1.40
1.60
1.80
2.00
1
2
3
4
5
6
7
8

FIG. 4A

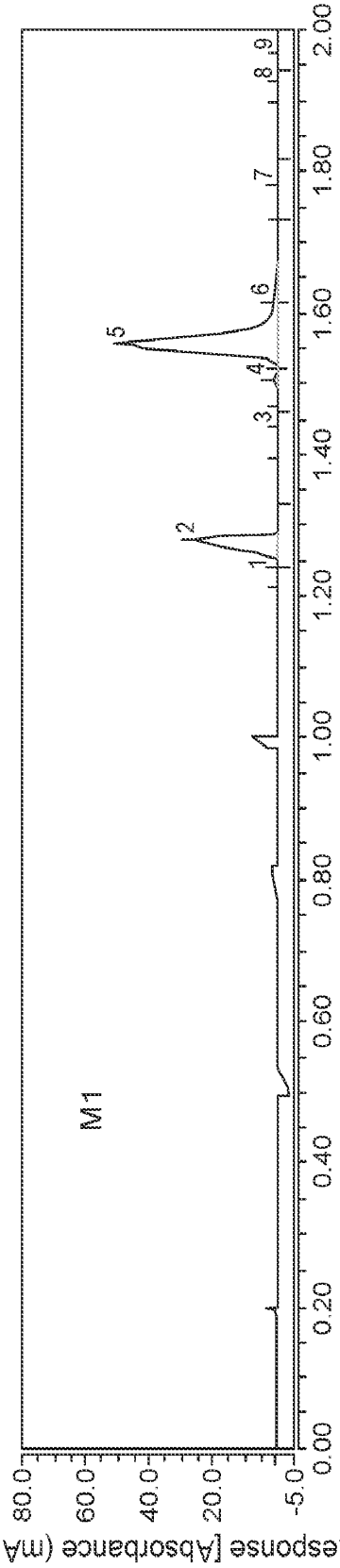

M1
Response [Absorbance (mAU)]
80.0
60.0
40.0
20.0
-5.0
0.00
0.20
0.40
0.60
0.80
1.00
1.20
1.40
1.60
1.80
2.00
1
2
3
4
5
6
7
8
9

FIG. 4B

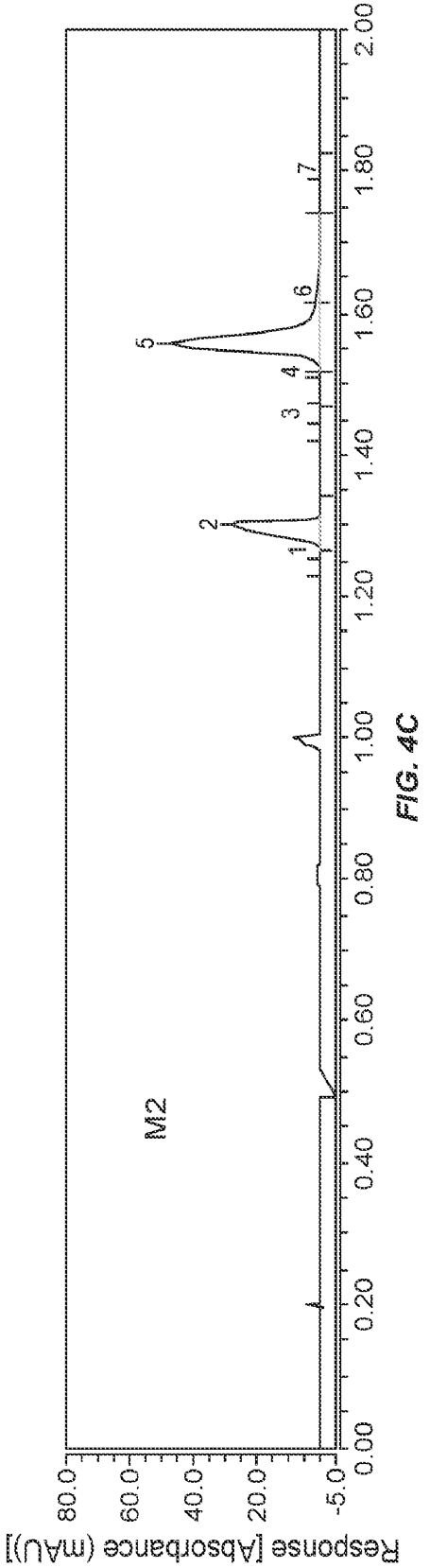

M2
Response [Absorbance (mAU)]
80.0
60.0
40.0
20.0
-5.0
0.00
0.20
0.40
0.60
0.80
1.00
1.20
1.40
1.60
1.80
2.00
1
2
3
4
5
6
7

FIG. 4C

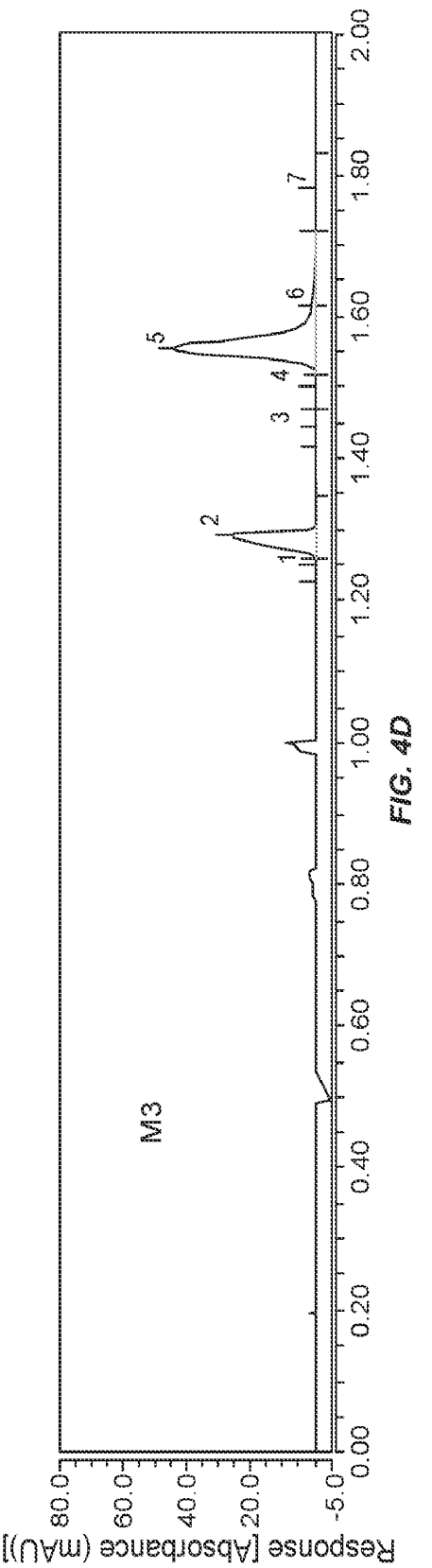

M3
Response [Absorbance (mAU)]
80.0
60.0
40.0
20.0
-5.0
0.00
0.20
0.40
0.60
0.80
1.00
1.20
1.40
1.60
1.80
2.00
1
2
3
4
5
6
7

FIG. 4D

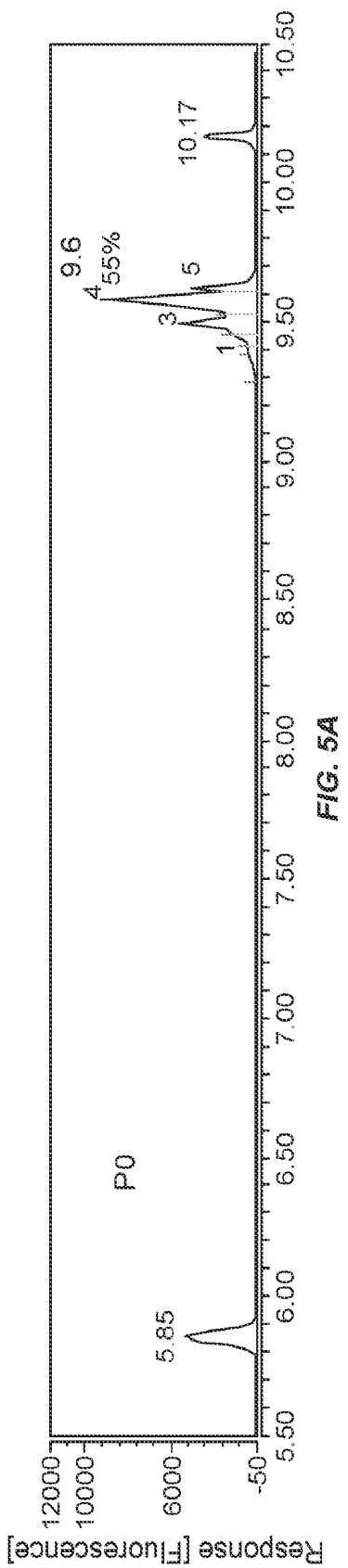

P0
5.85
9.6
55%
10.17
1
2
3
4
5
Response [Fluorescence]
12000
10000
6000
-50
5.50
6.00
6.50
7.00
7.50
8.00
8.50
9.00
9.50
10.00
10.50

FIG. 5A

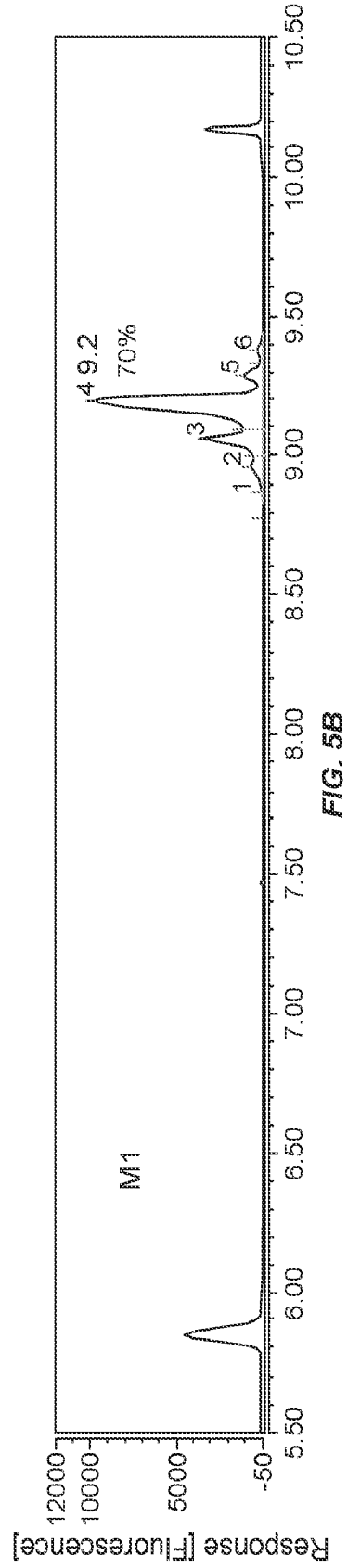

M1
9.2
70%
1
2
3
4
5
6
Response [Fluorescence]
12000
10000
5000
-50
5.50
6.00
6.50
7.00
7.50
8.00
8.50
9.00
9.50
10.00
10.50

FIG. 5B

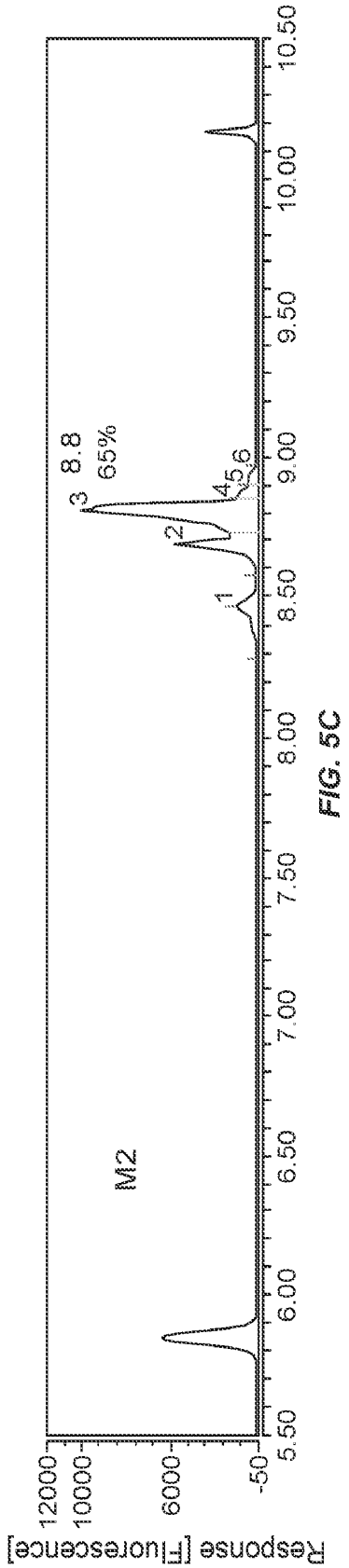

M2
8.8
65%
1
2
3
4
5,6
Response [Fluorescence]
12000
10000
6000
-50
5.50
6.00
6.50
7.00
7.50
8.00
8.50
9.00
9.50
10.00
10.50

FIG. 5C

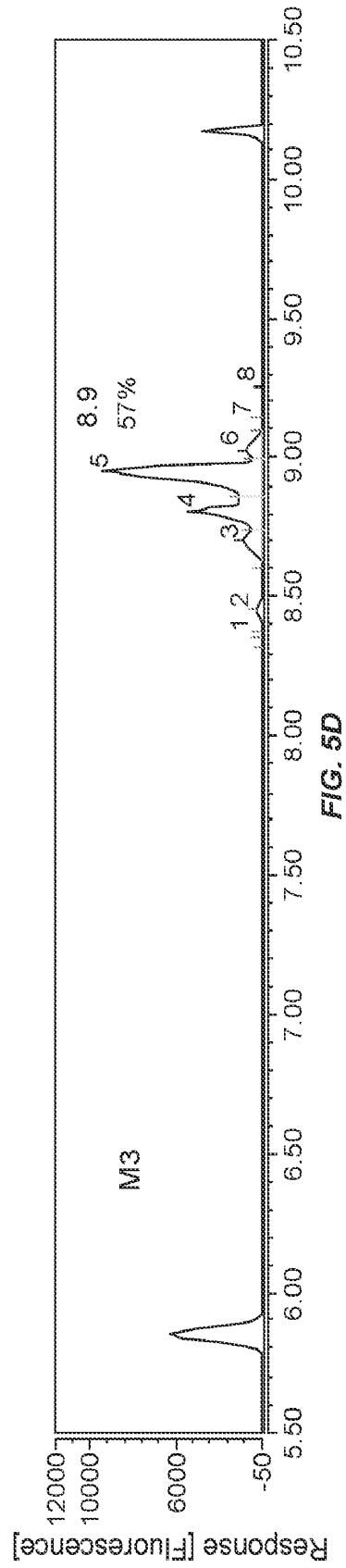

M3
8.9
57%
1
2
3
4
5
6
7
8
Response [Fluorescence]
12000
10000
6000
-50
5.50
6.00
6.50
7.00
7.50
8.00
8.50
9.00
9.50
10.00
10.50

FIG. 5D

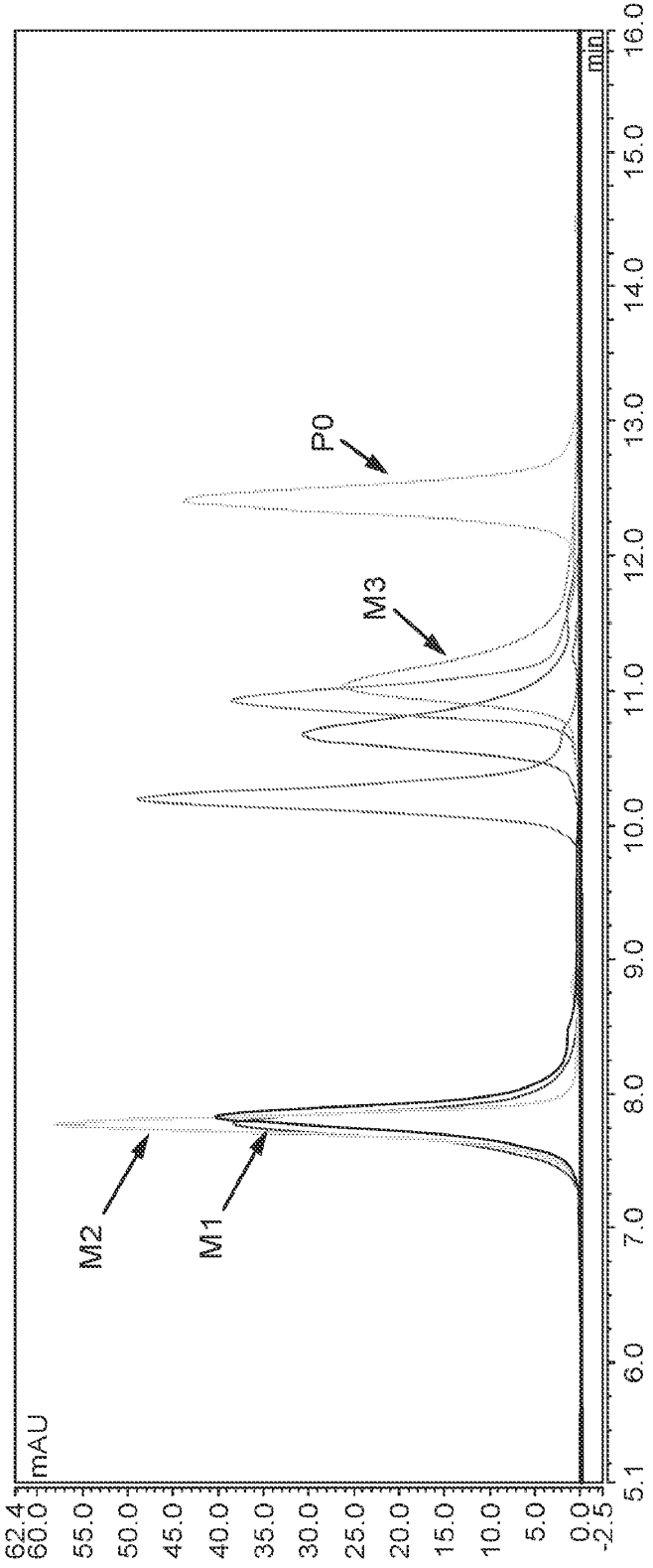

mAU
62.4
60.0
55.0
50.0
45.0
40.0
35.0
30.0
25.0
20.0
15.0
10.0
5.0
0.0
-2.5
min
5.1
6.0
7.0
8.0
9.0
10.0
11.0
12.0
13.0
14.0
15.0
16.0
M2
M1
M3
P0

FIG. 6

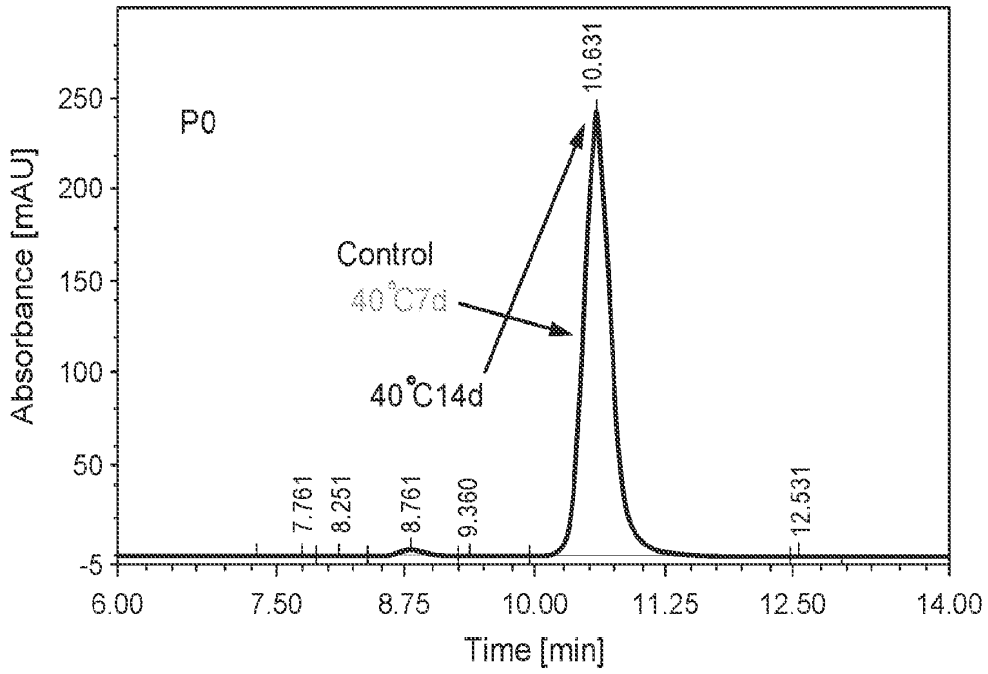

P0
Control
40°C7d
40°C14d
10.631
7.761
8.251
8.761
9.360
12.531
Absorbance [mAU]
Time [min]
250
200
150
100
50
-5
6.00
7.50
8.75
10.00
11.25
12.50
14.00

FIG. 7A

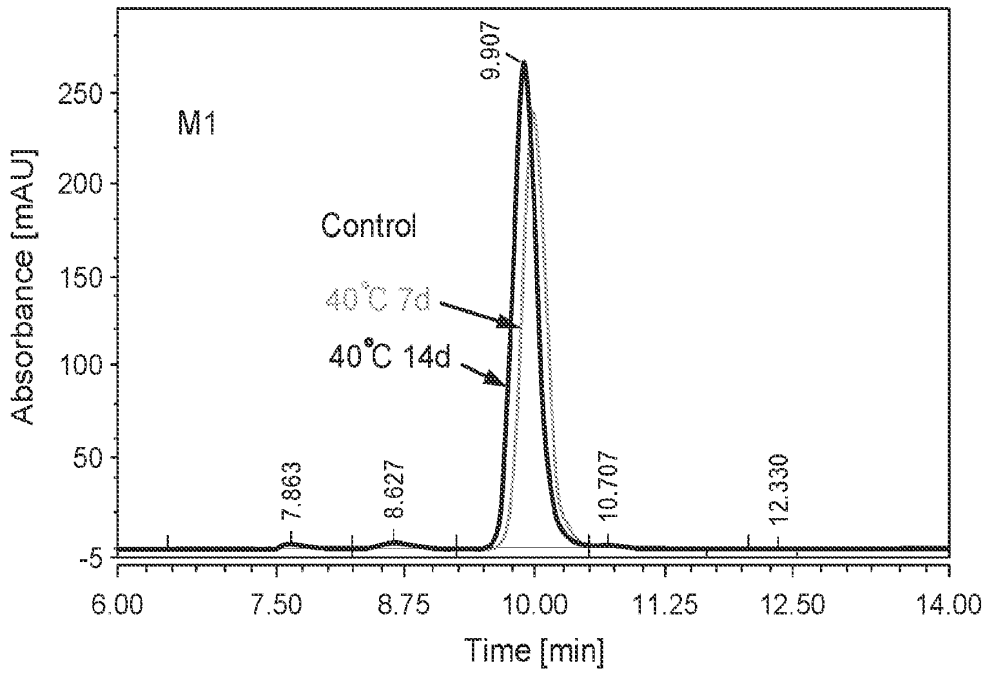

M1
Control
40°C 7d
40°C 14d
9.907
7.863
8.627
10.707
12.330
Absorbance [mAU]
Time [min]
250
200
150
100
50
-5
6.00
7.50
8.75
10.00
11.25
12.50
14.00

FIG. 7B

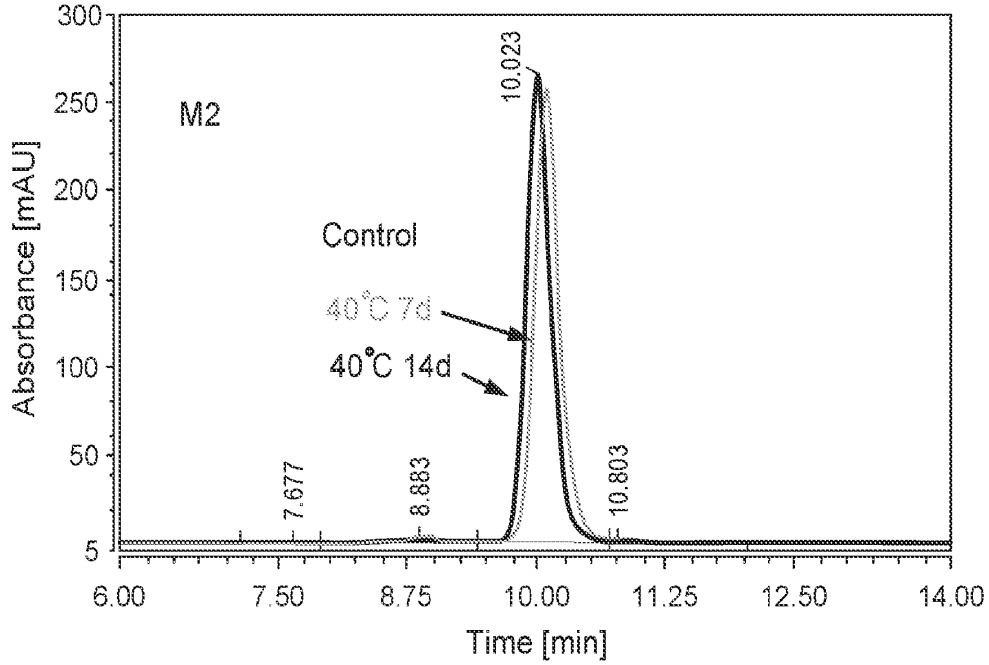

M2
Control
40°C 7d
40°C 14d
10.023
7.677
8.883
10.803
Absorbance [mAU]
Time [min]
300
250
200
150
100
50
5
6.00
7.50
8.75
10.00
11.25
12.50
14.00

FIG. 7C

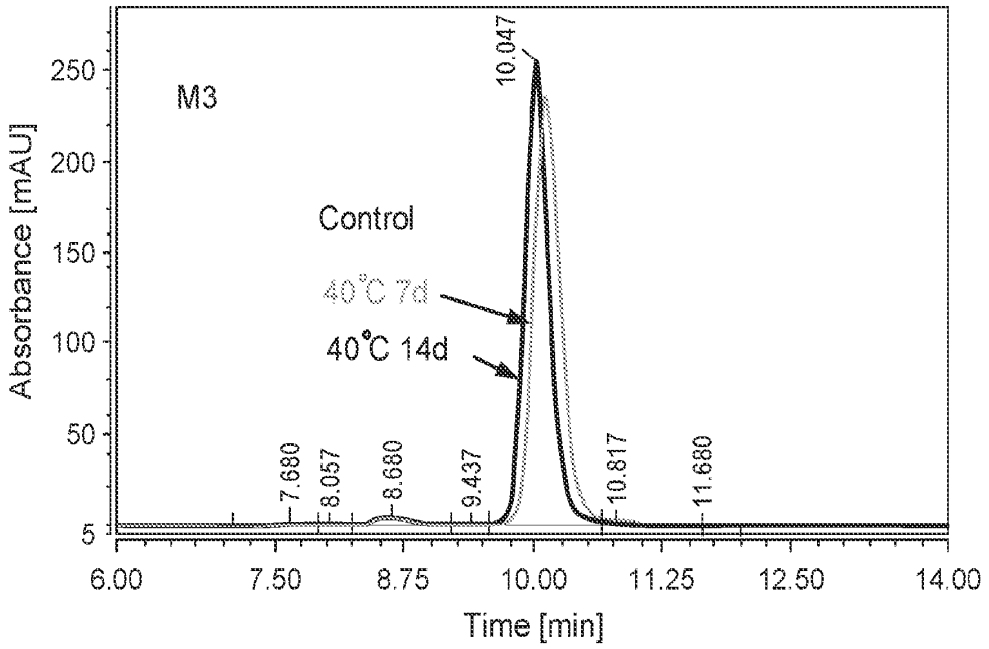

M3
Control
40°C 7d
40°C 14d
10.047
7.680
8.057
8.680
9.437
10.817
11.680
Absorbance [mAU]
Time [min]
250
200
150
100
50
5
6.00
7.50
8.75
10.00
11.25
12.50
14.00

FIG. 7D

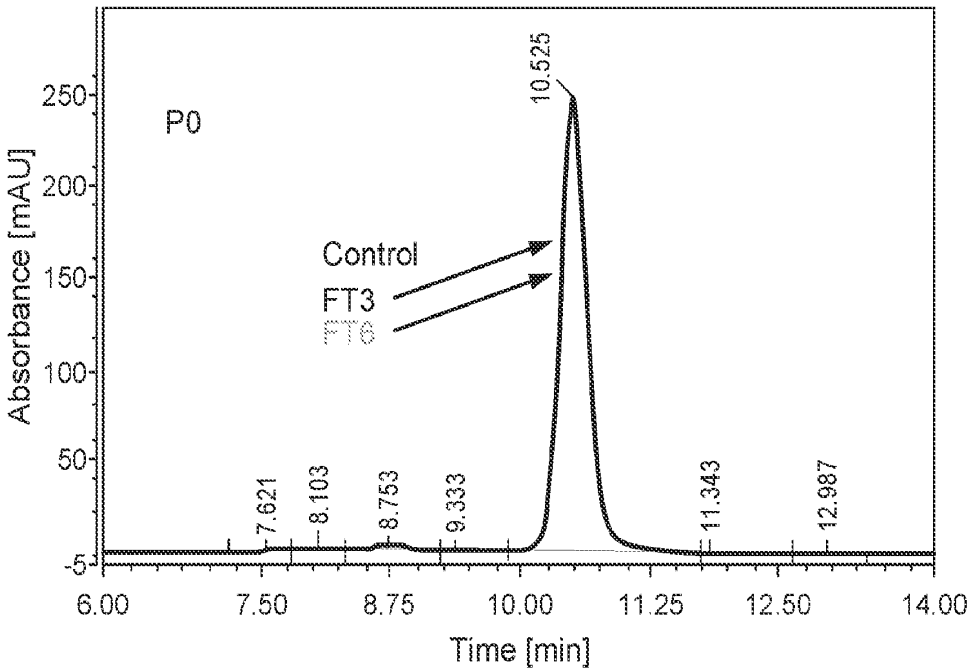

P0
Control
FT3
FT6
10.525
7.621
8.103
8.753
9.333
11.343
12.987
Absorbance [mAU]
250
200
150
100
50
-5
6.00
7.50
8.75
10.00
11.25
12.50
14.00
Time [min]

FIG. 8A

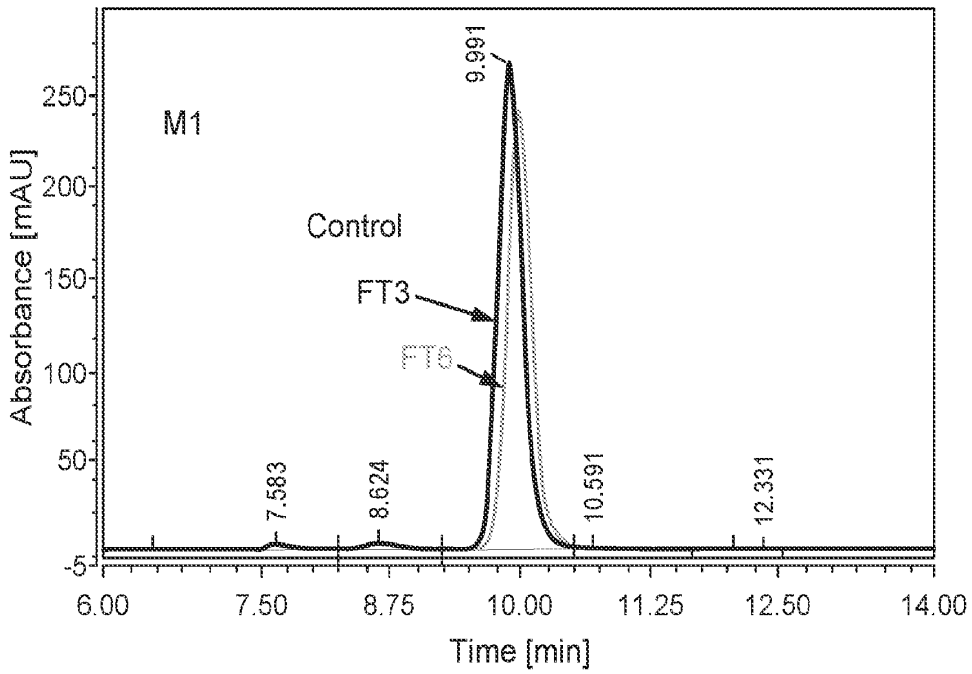

M1
Control
FT3
FT6
9.991
7.583
8.624
10.591
12.331
Absorbance [mAU]
250
200
150
100
50
-5
6.00
7.50
8.75
10.00
11.25
12.50
14.00
Time [min]

FIG. 8B

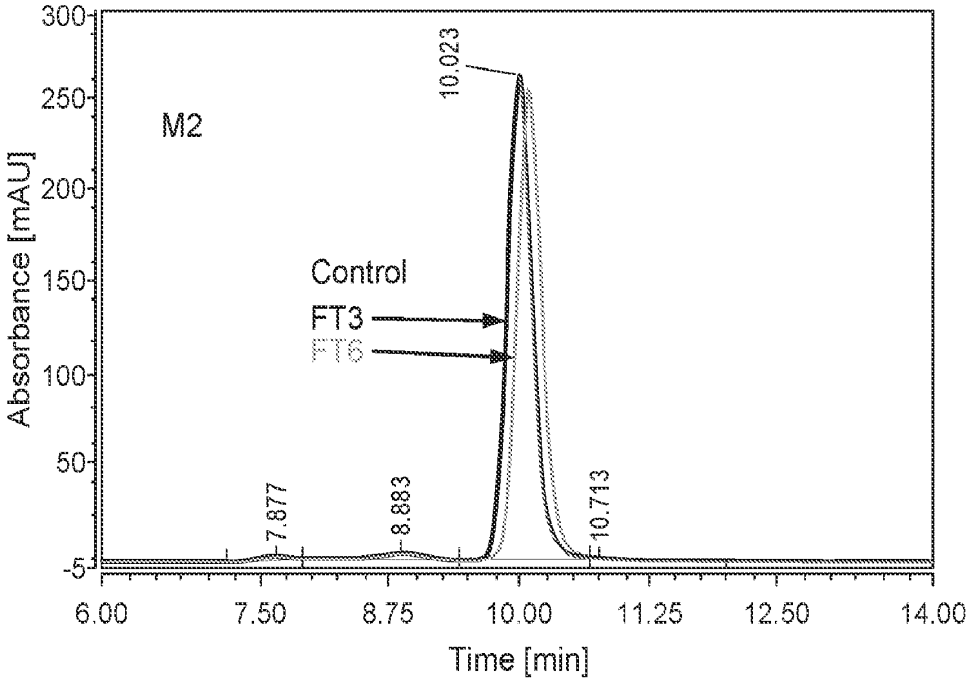

M2
Control
FT3
FT6
10.023
7.877
8.883
10.713
Absorbance [mAU]
Time [min]
300
250
200
150
100
50
-5
6.00
7.50
8.75
10.00
11.25
12.50
14.00

FIG. 8C

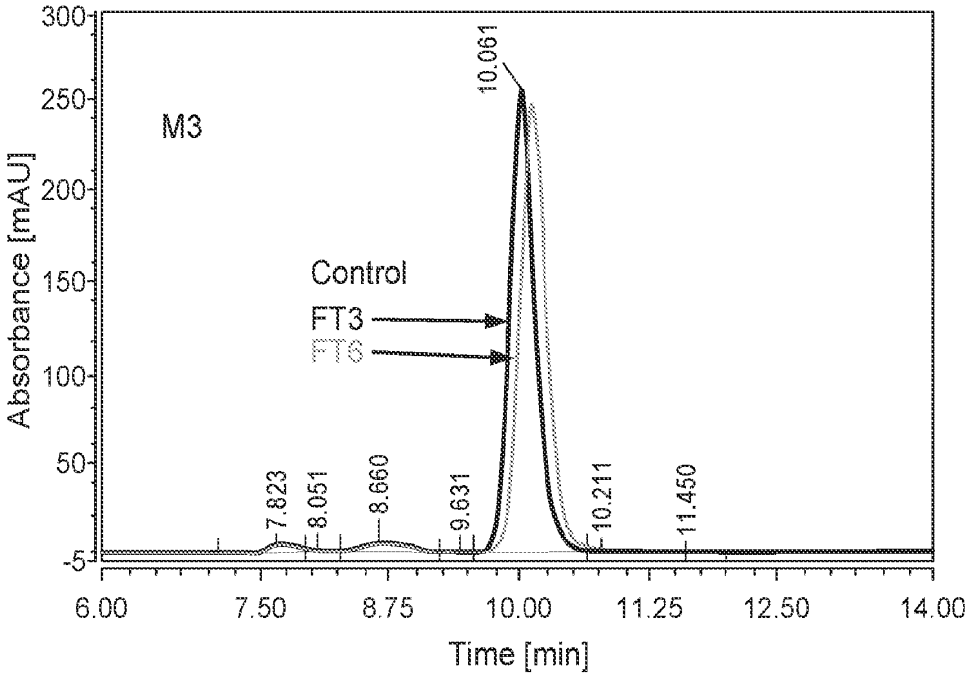

M3
Control
FT3
FT6
10.061
7.823
8.051
8.660
9.631
10.211
11.450
Absorbance [mAU]
Time [min]
300
250
200
150
100
50
-5
6.00
7.50
8.75
10.00
11.25
12.50
14.00

FIG. 8D

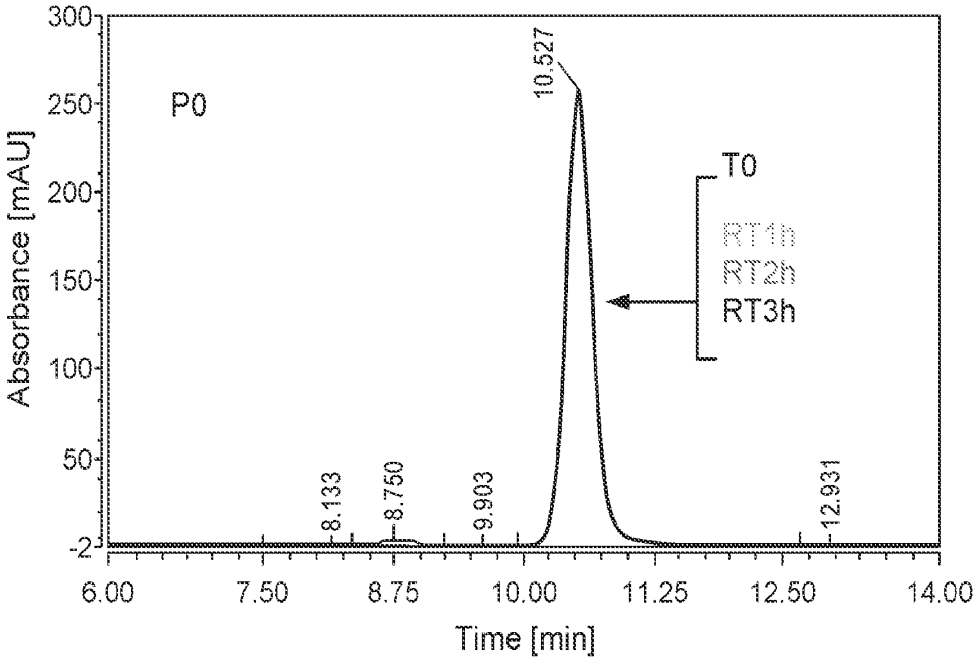

P0
10.527
T0
RT1h
RT2h
RT3h
8.133
8.750
9.903
12.931
Absorbance [mAU]
300
250
200
150
100
50
-2
6.00
7.50
8.75
10.00
11.25
12.50
14.00
Time [min]

FIG. 9A

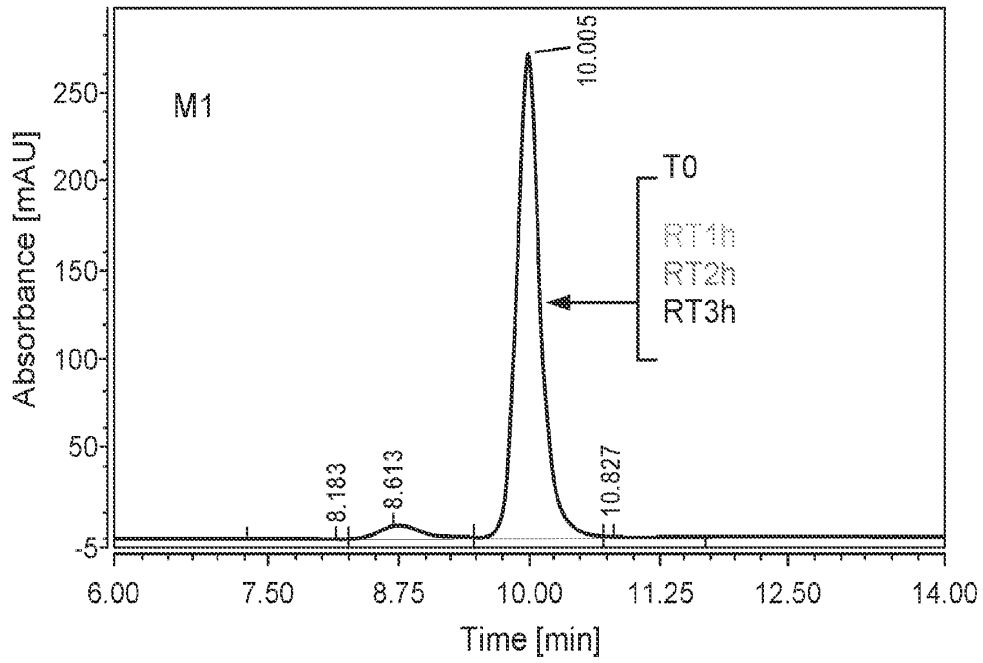

M1
10.005
T0
RT1h
RT2h
RT3h
8.183
8.613
10.827
Absorbance [mAU]
250
200
150
100
50
-5
6.00
7.50
8.75
10.00
11.25
12.50
14.00
Time [min]

FIG. 9B

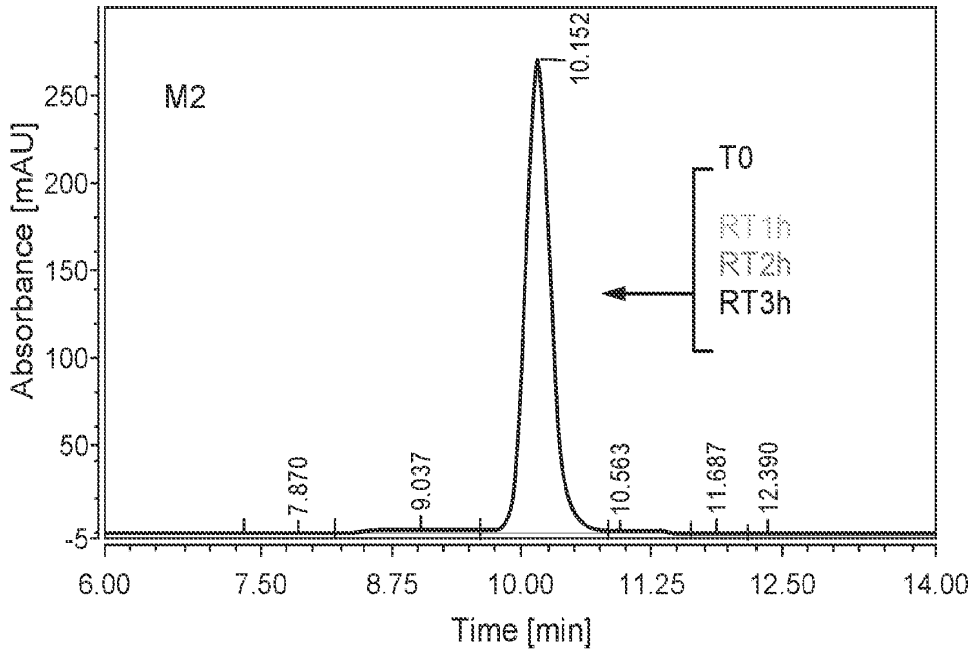

M2
10.152
T0
RT1h
RT2h
RT3h
7.870
9.037
10.563
11.687
12.390
Absorbance [mAU]
250
200
150
100
50
-5
6.00
7.50
8.75
10.00
11.25
12.50
14.00
Time [min]

FIG. 9C

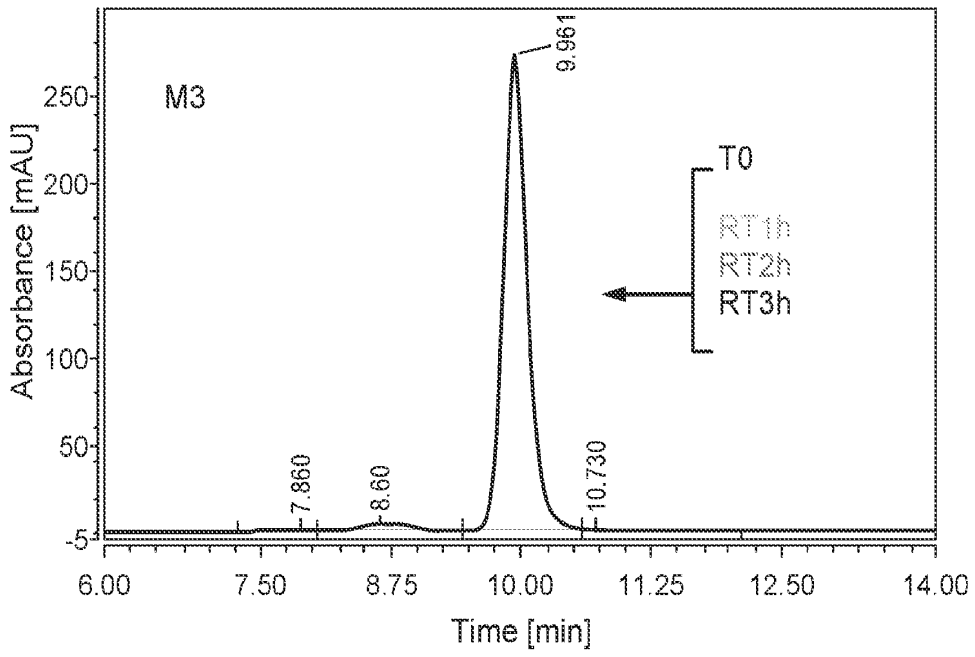

M3
9.961
T0
RT1h
RT2h
RT3h
7.860
8.60
10.730
Absorbance [mAU]
250
200
150
100
50
-5
6.00
7.50
8.75
10.00
11.25
12.50
14.00
Time [min]

FIG. 9D

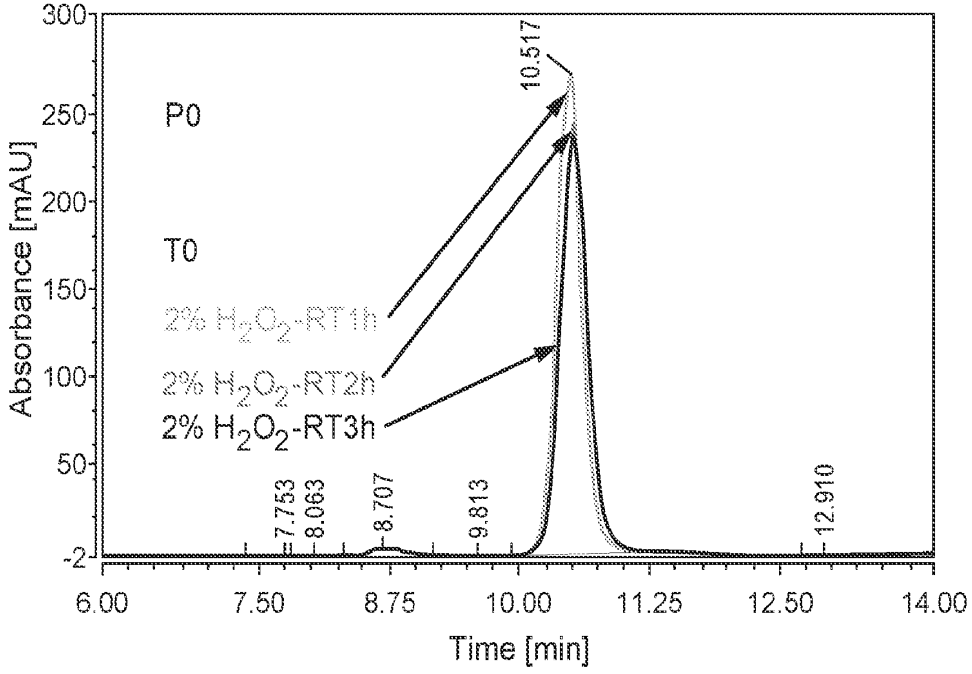


FIG. 10A

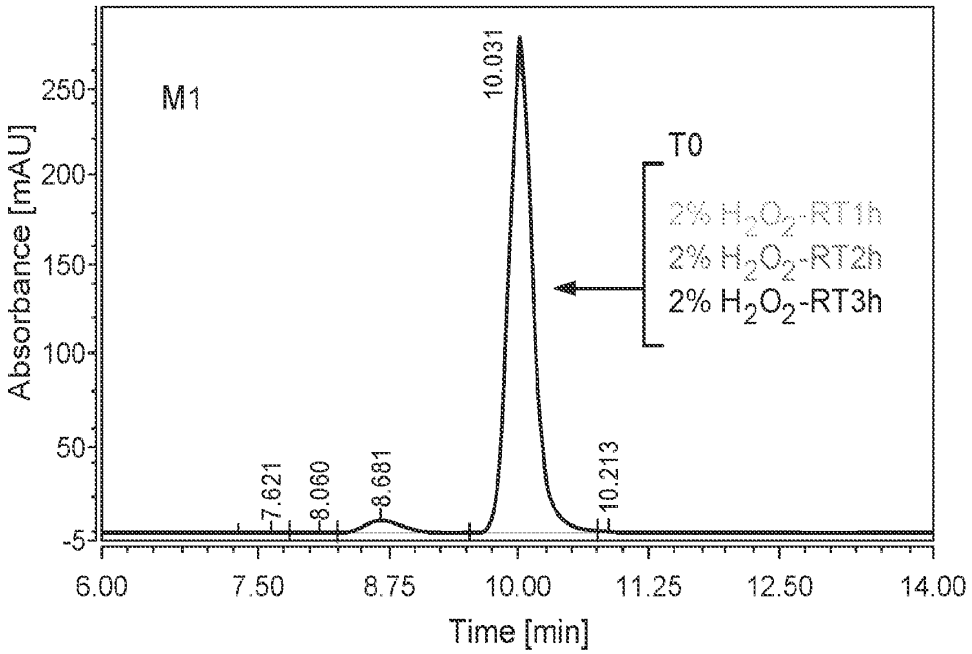


FIG. 10B

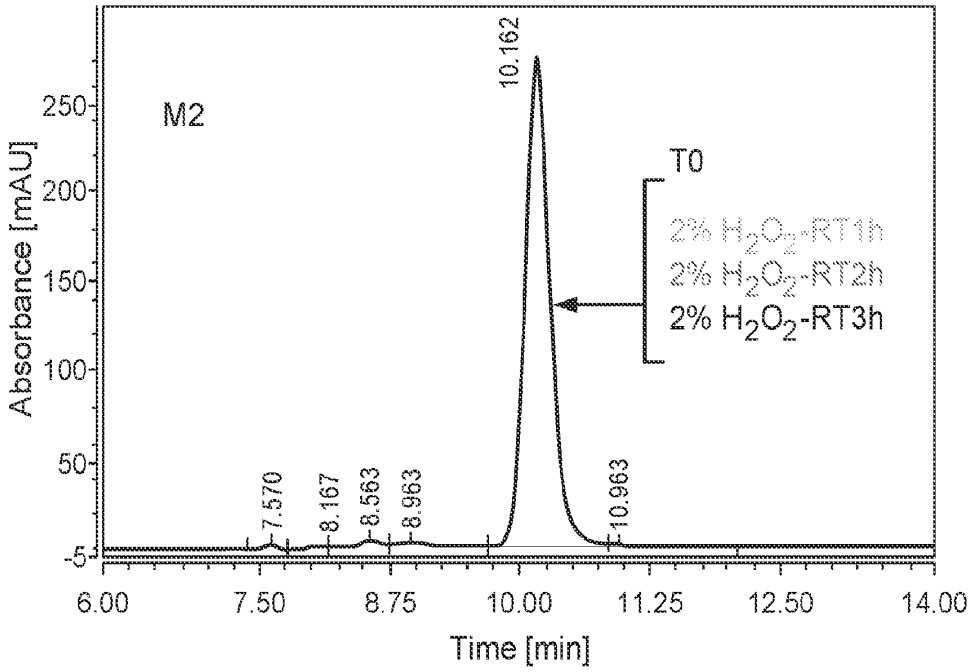


FIG. 10C

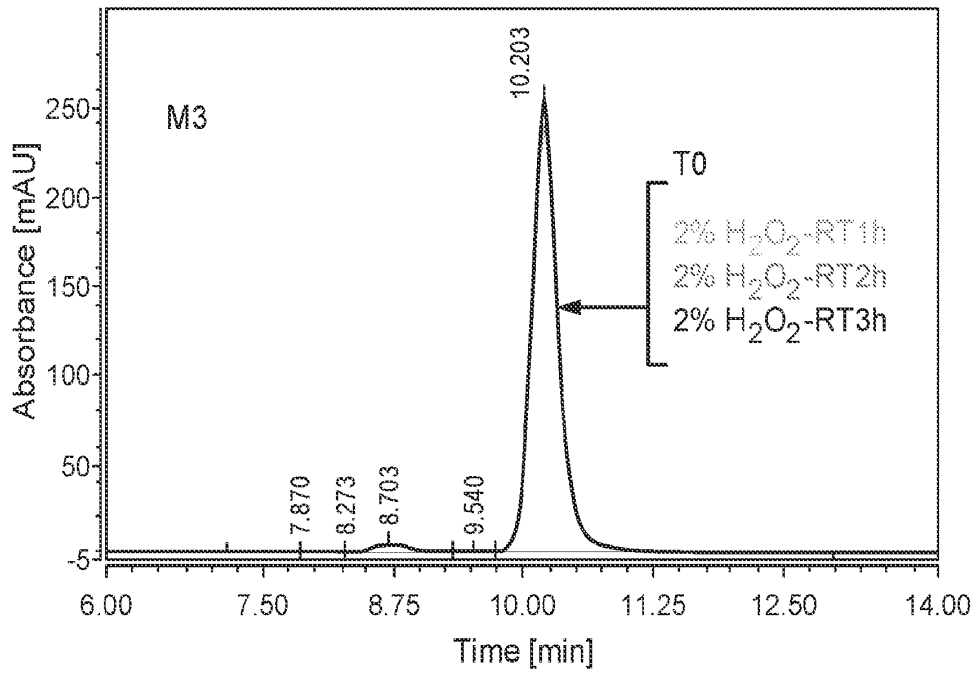


FIG. 10D

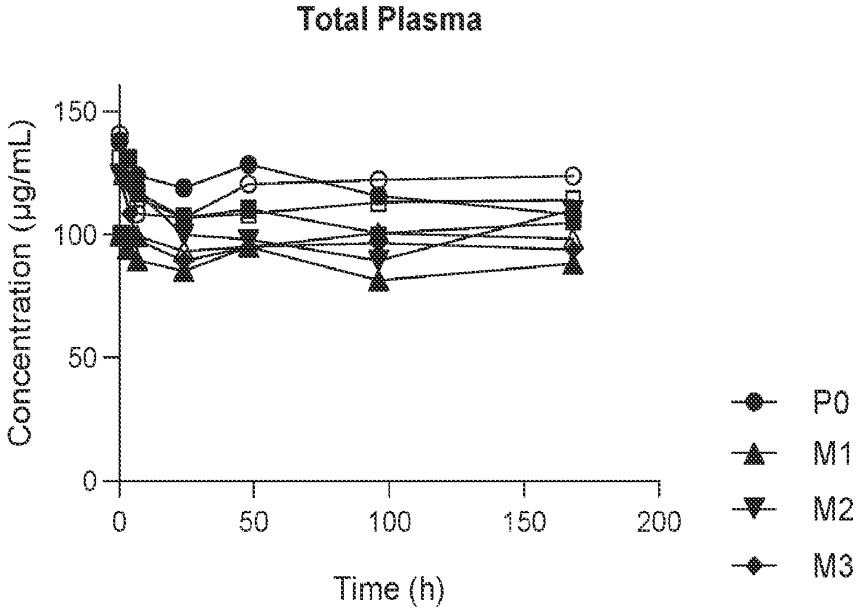


FIG. 11A

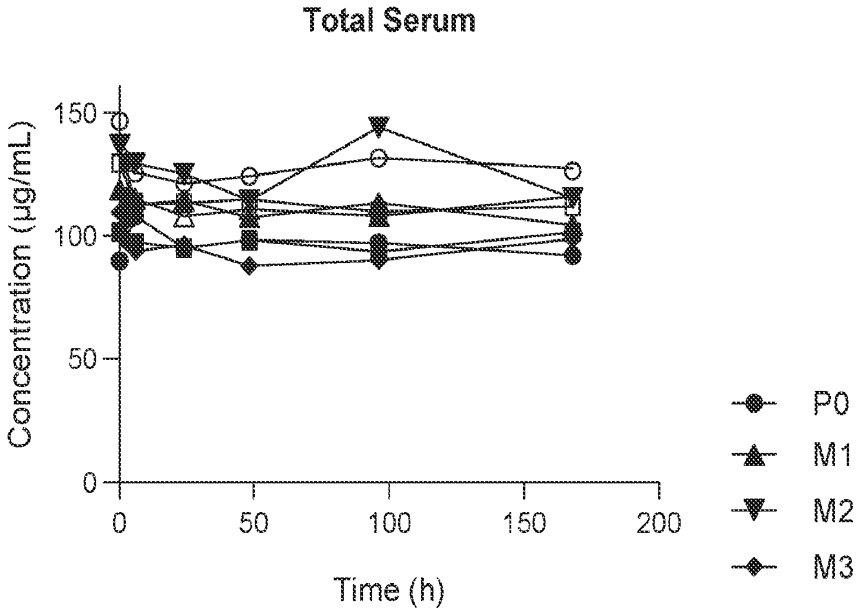


FIG. 11B

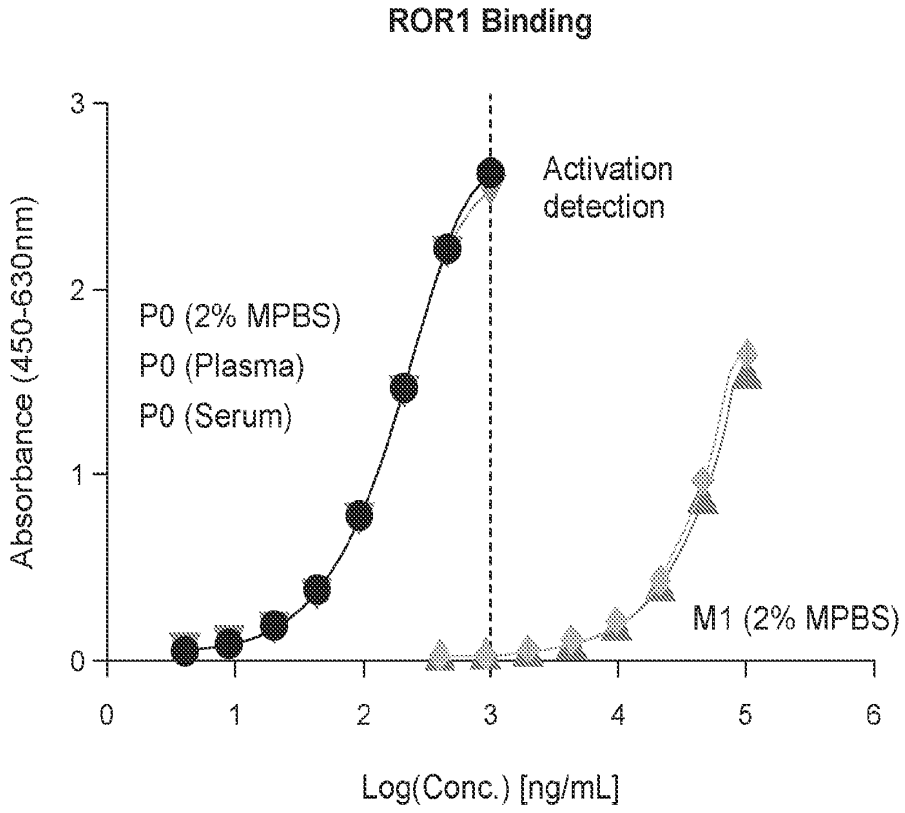
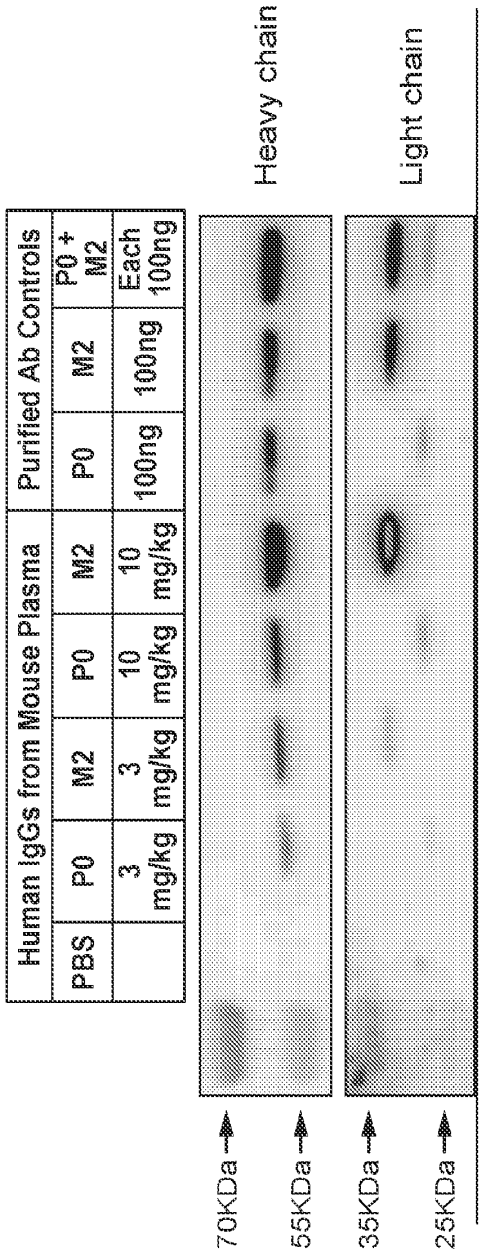

ROR1 Binding
Absorbance (450-630nm)
Log(Conc.) [ng/mL]
Activation detection
P0 (2% MPBS)
P0 (Plasma)
P0 (Serum)
M1 (2% MPBS)
0
1
2
3
4
5
6

FIG. 11C



Human IgGs from Mouse Plasma					Purified Ab Controls		
PBS	P0	M2	P0	M2	P0	M2	P0 + M2
	3 mg/kg	3 mg/kg	10 mg/kg	10 mg/kg	100ng	100ng	Each 100ng

FIG. 12

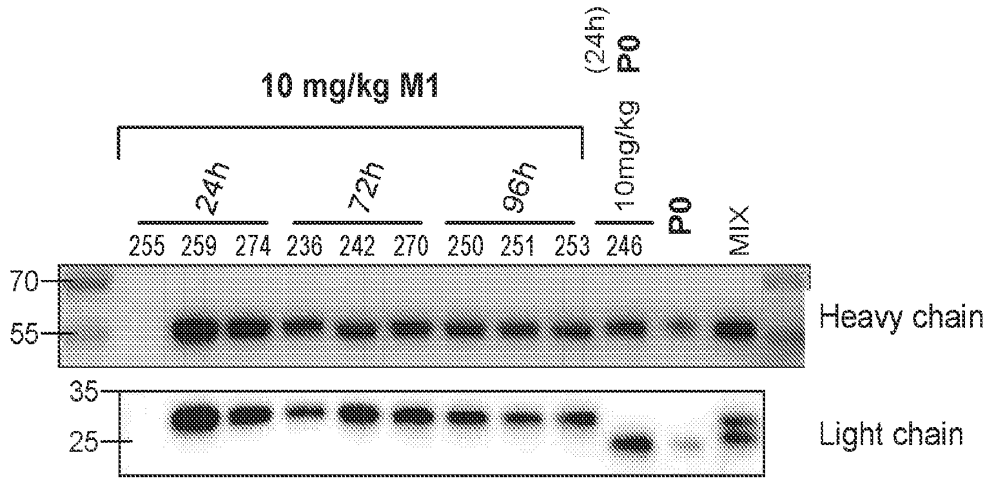


FIG. 13A

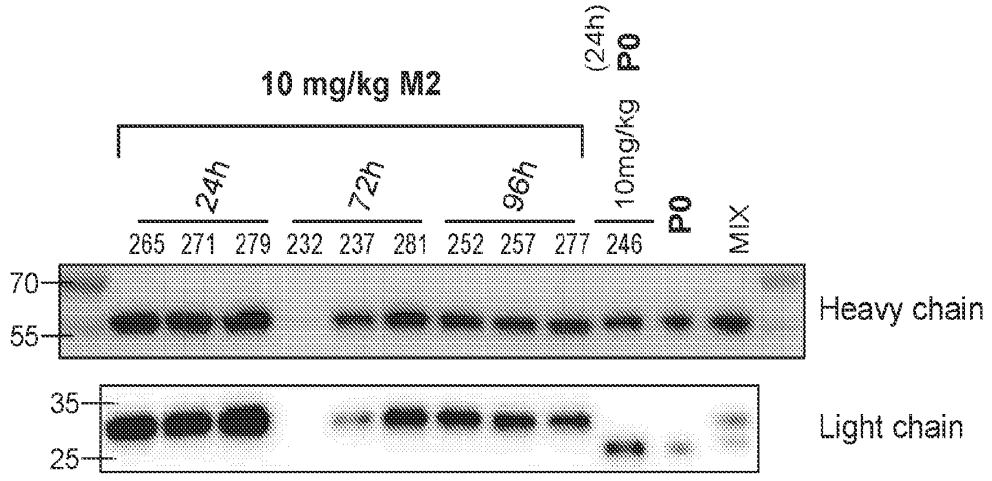


FIG. 13B

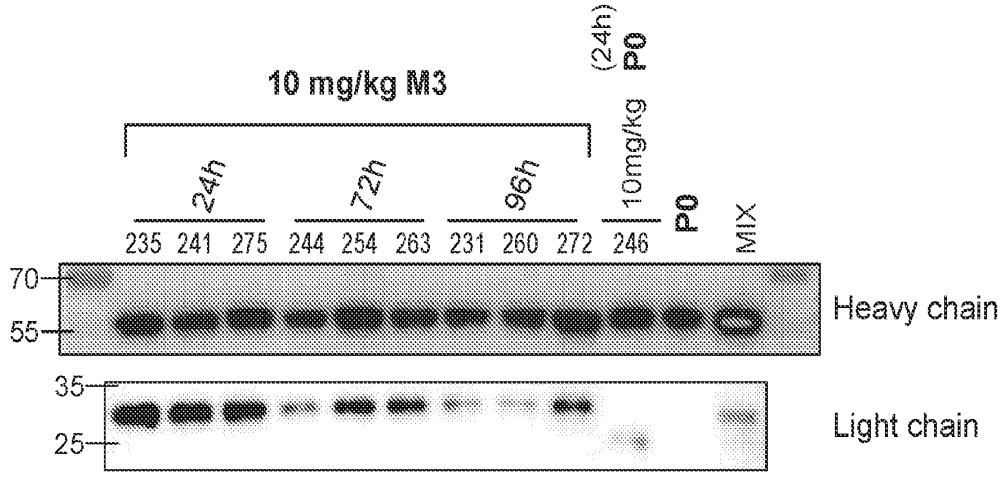


FIG. 13C

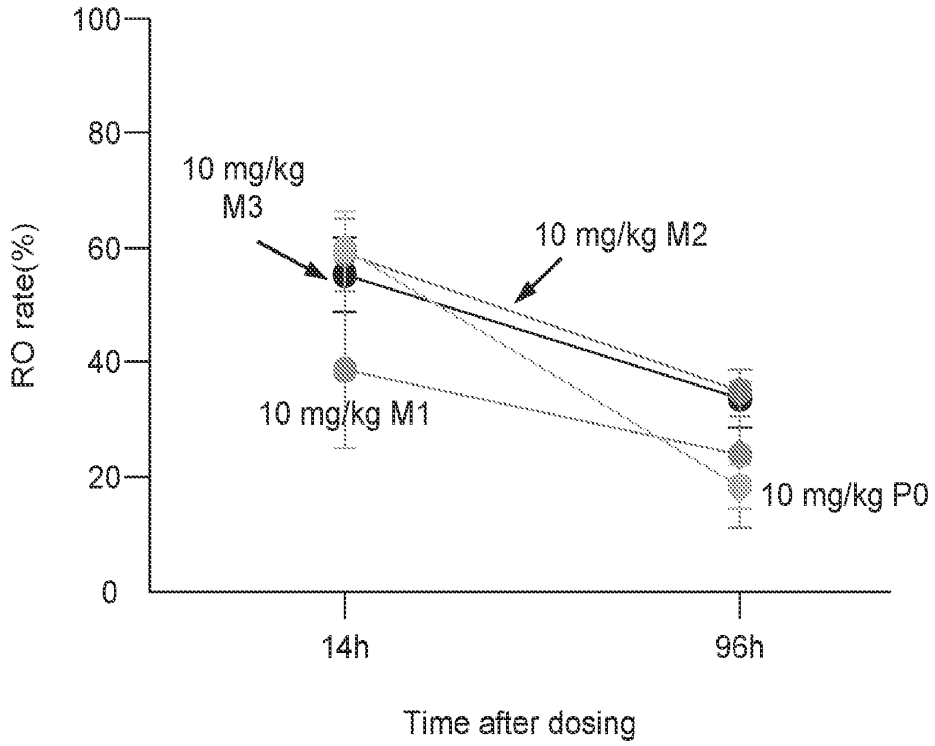

100
80
60
40
20
0
RO rate(%)
10 mg/kg
M3
10 mg/kg M2
10 mg/kg M1
10 mg/kg P0
14h
96h
Time after dosing

FIG. 14

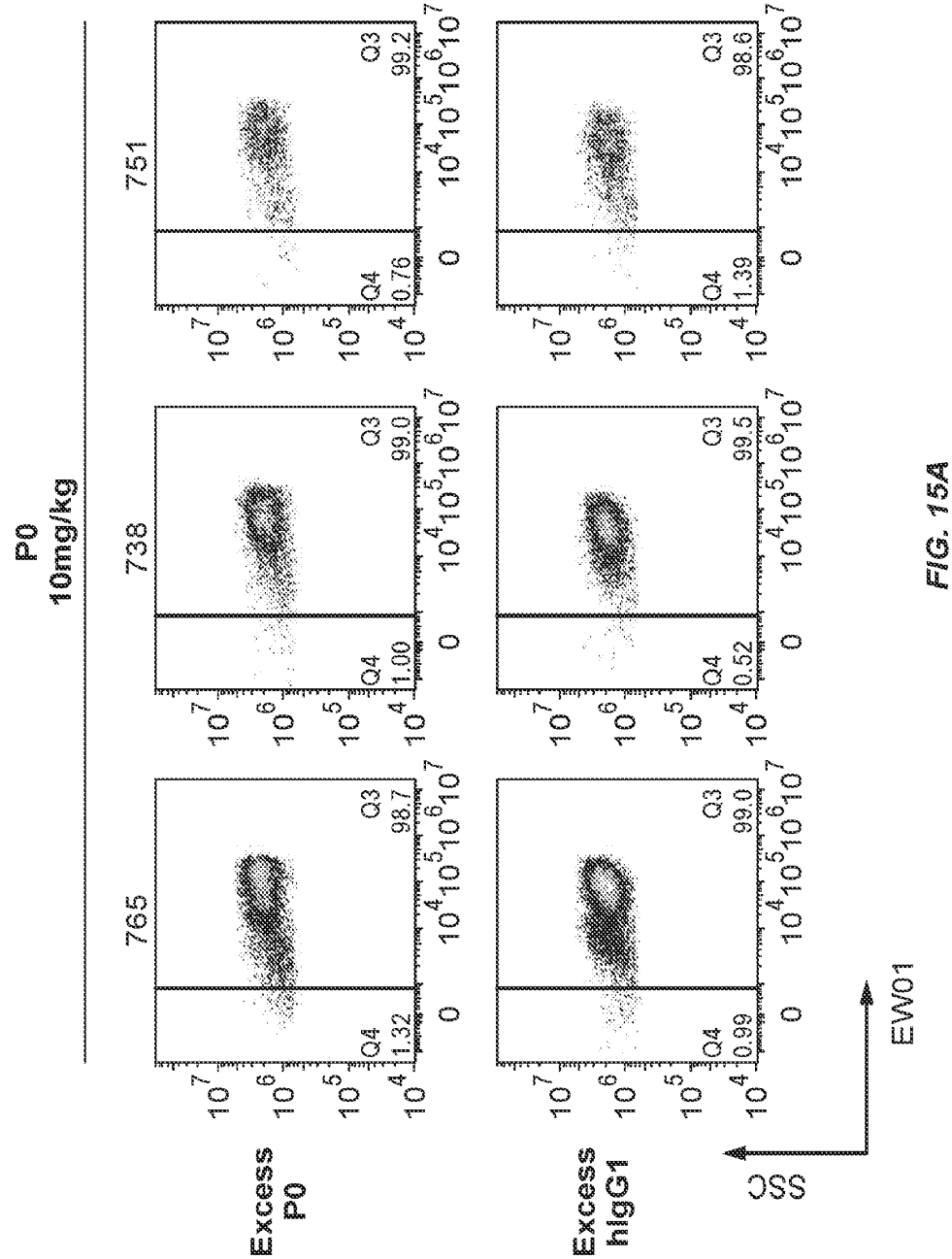

P0
10mg/kg
765
738
751
Excess
P0
Excess
hIgG1
SSC
EW01
Q3
Q4
98.7
1.32
99.0
1.00
99.2
0.76
99.0
0.99
99.5
0.52
98.6
1.39

FIG. 15A

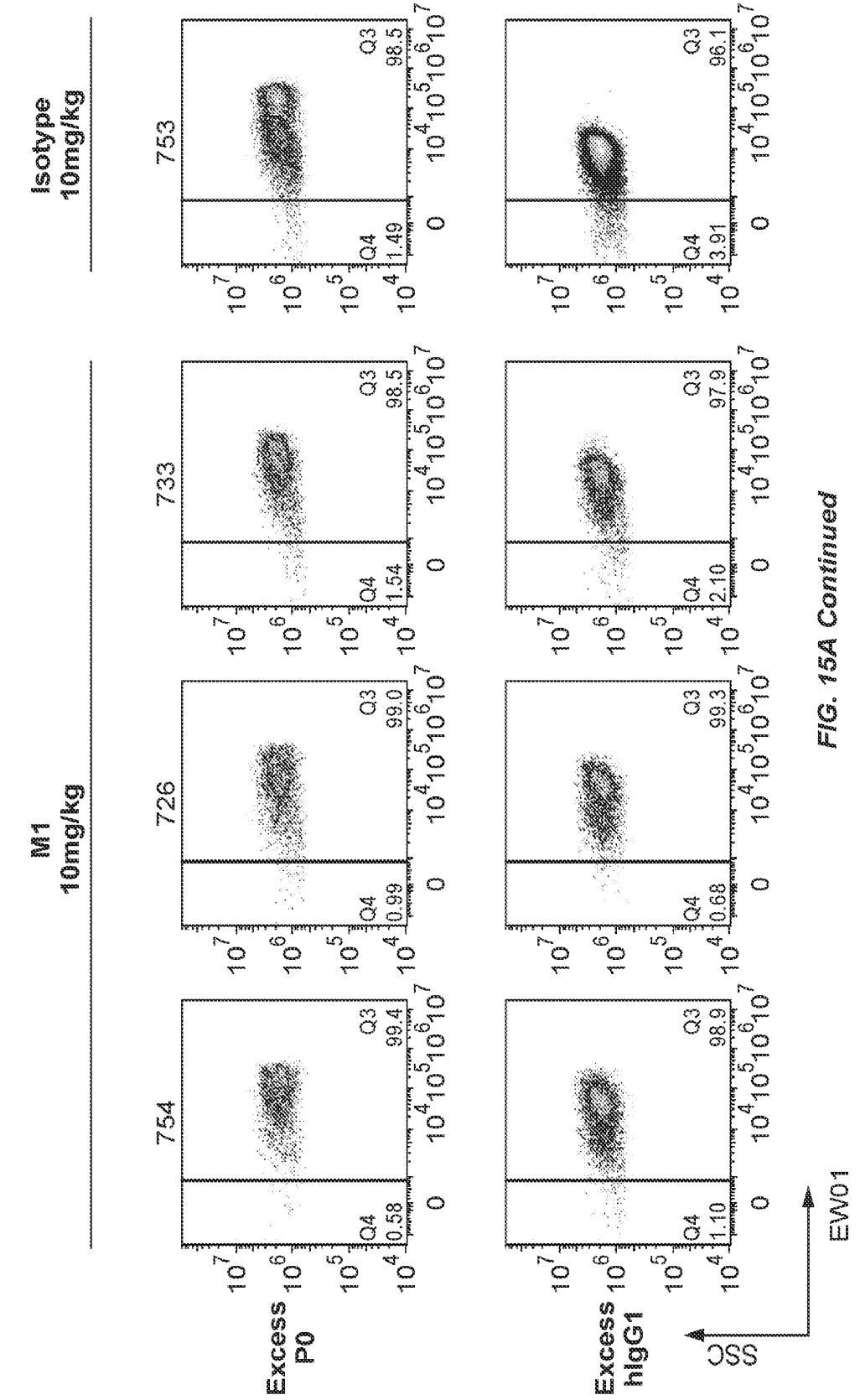


FIG. 15A Continued

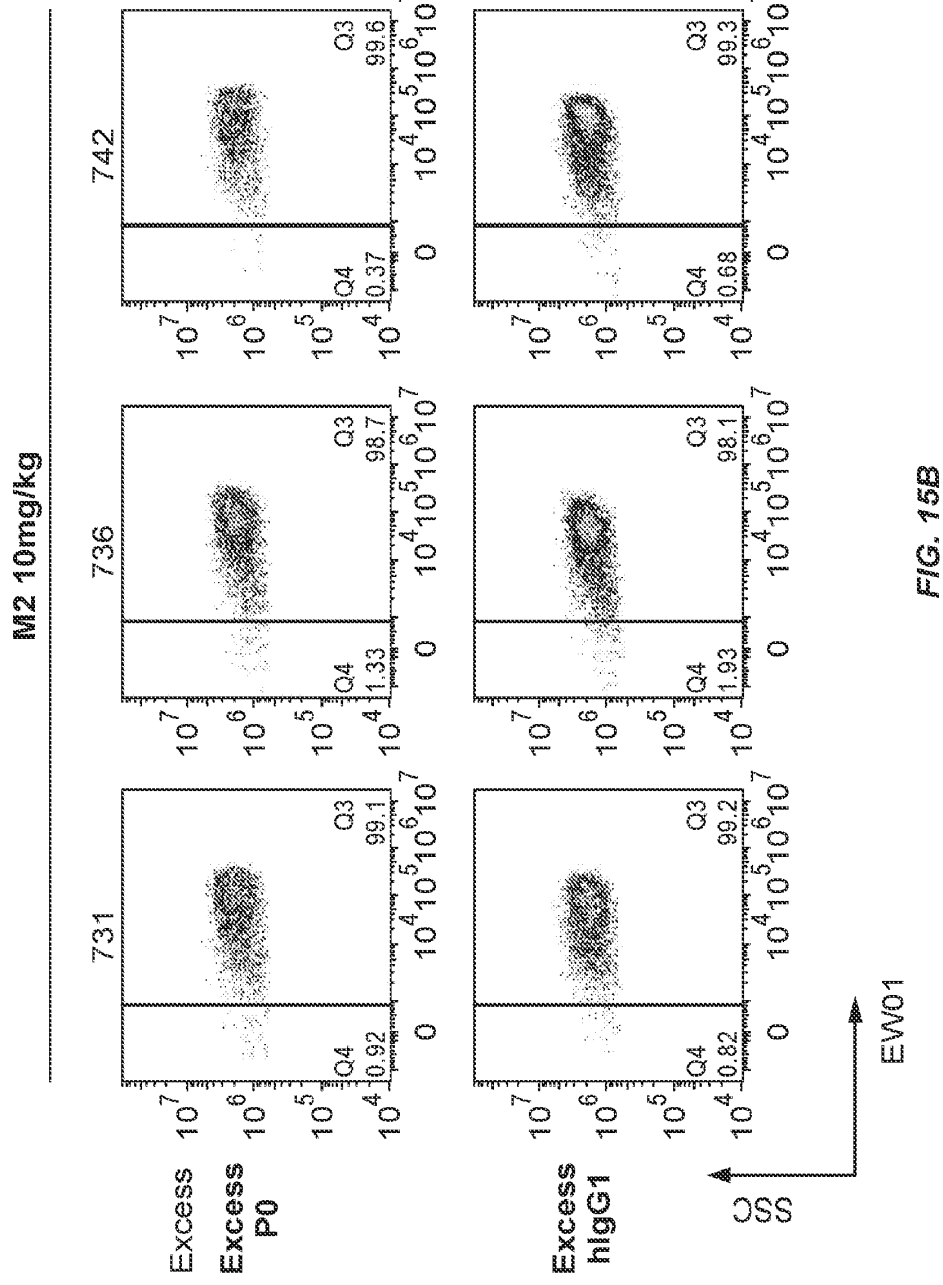

M2 10mg/kg
731
736
742
Excess
Excess P0
Excess hIgG1
Q4 0.92
Q3 99.1
Q4 1.33
Q3 98.7
Q4 0.37
Q3 99.6
Q4 0.82
Q3 99.2
Q4 1.93
Q3 98.1
Q4 0.68
Q3 99.3
SSC
FWD1

FIG. 15B

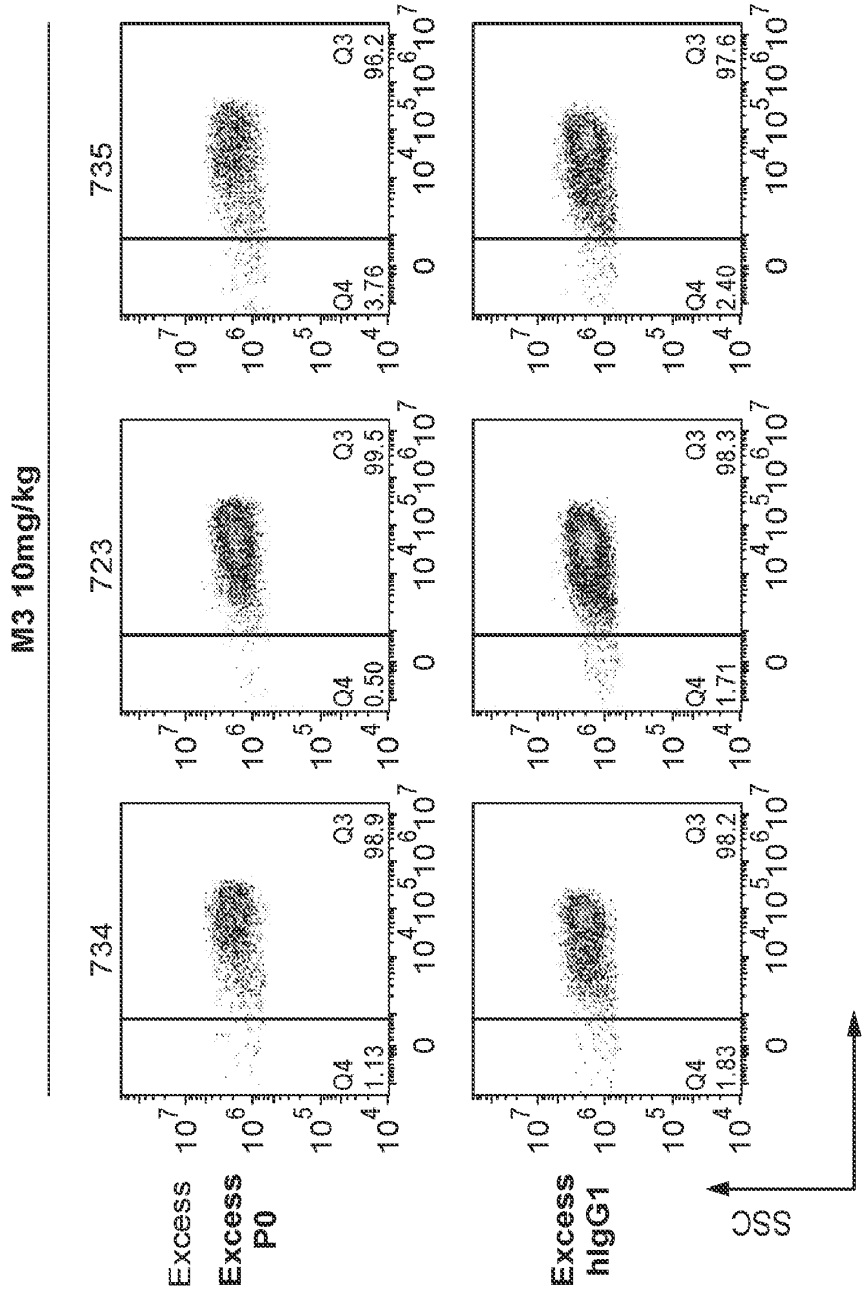


FIG. 15B Continued

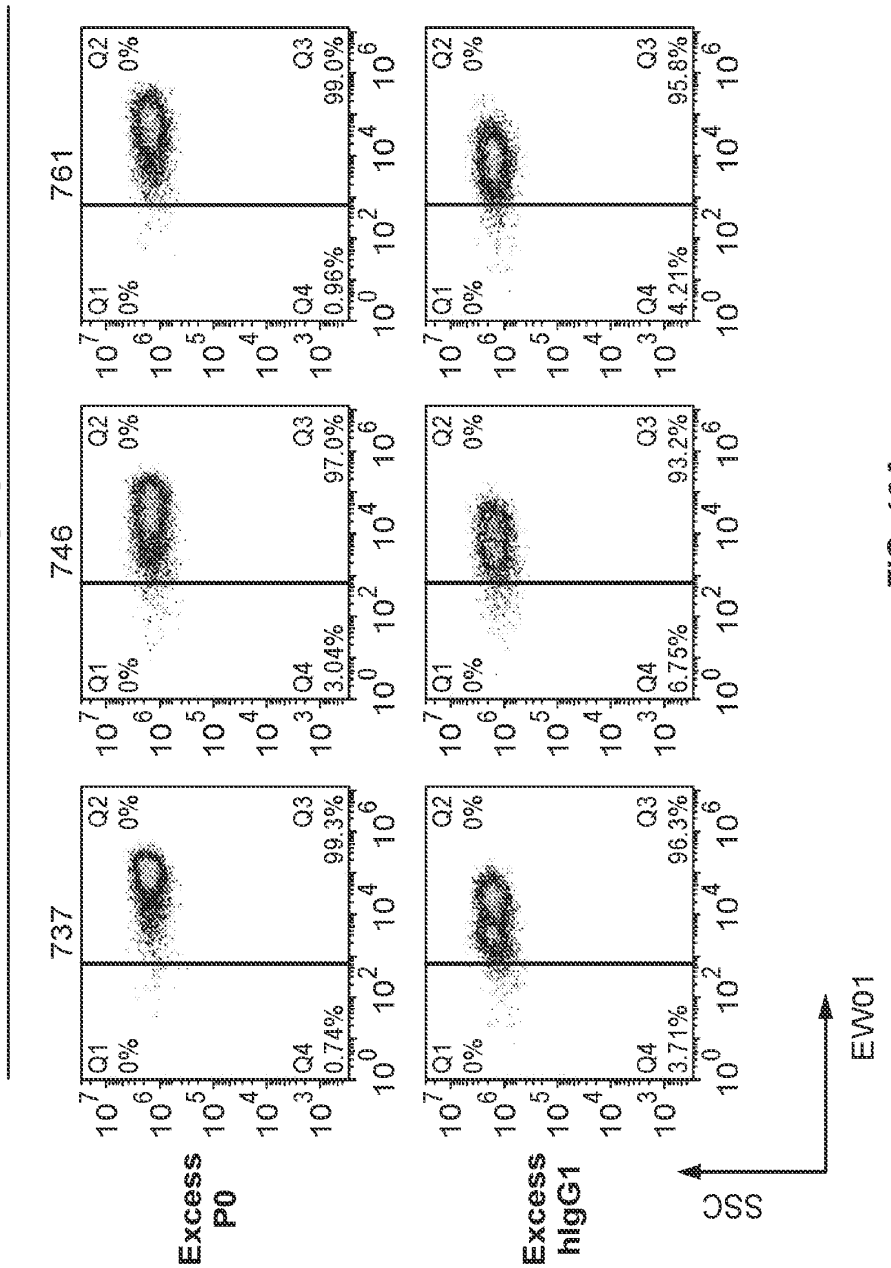

P0
10mg/kg
737
746
761
Excess P0
Excess hIgG1
Q1 0%
Q2 0%
Q3 99.3%
Q4 0.74%
Q3 97.0%
Q4 3.04%
Q3 99.0%
Q4 0.96%
Q3 96.3%
Q4 3.71%
Q3 93.2%
Q4 6.75%
Q3 95.8%
Q4 4.21%
SSC
EW01

FIG. 16A

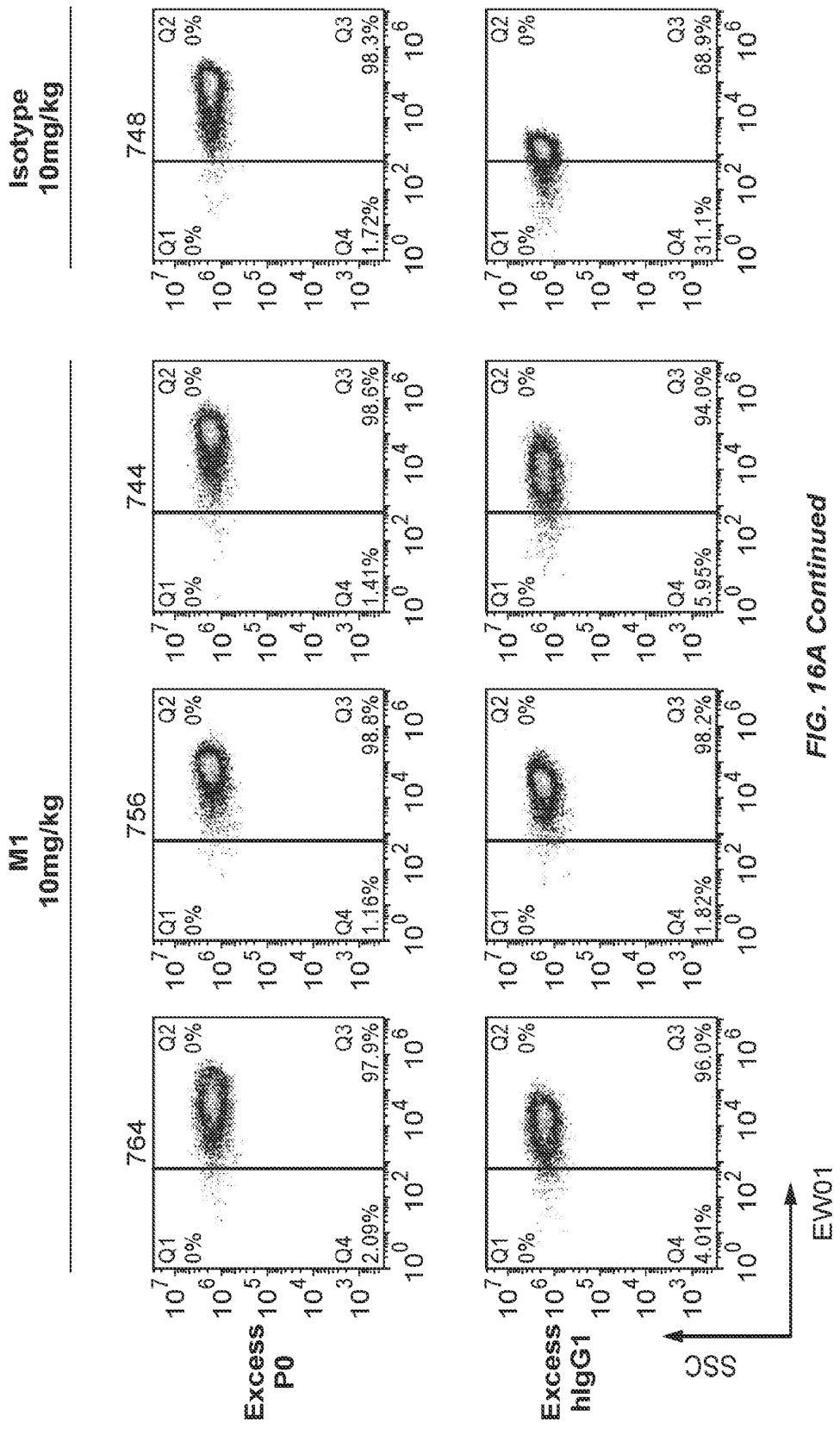


FIG. 16A Continued

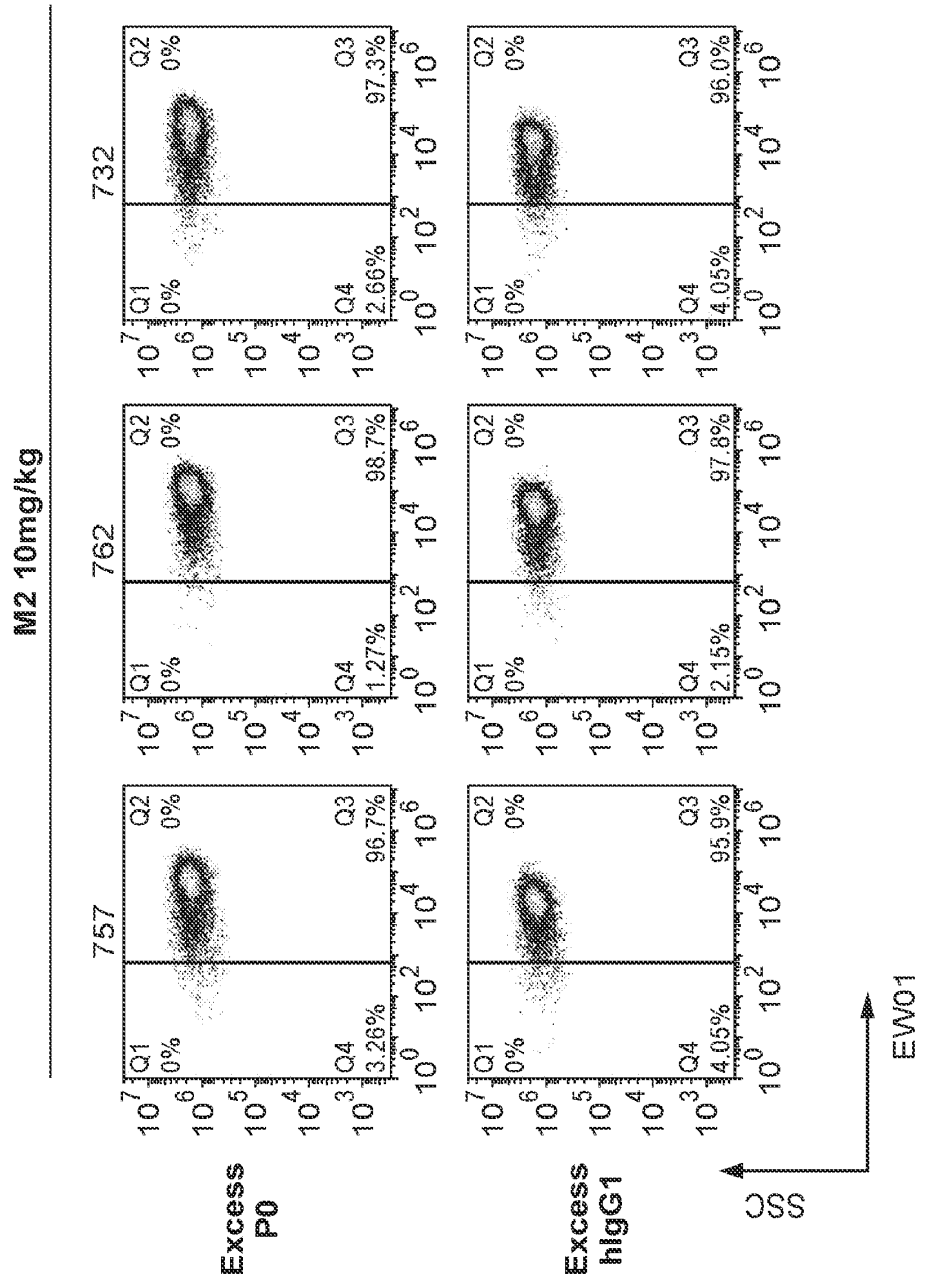

M2 10mg/kg
757
762
732
Excess P0
Excess hIgG1
Q1 0%
Q2 0%
Q3 96.7%
Q4 3.26%
Q3 98.7%
Q4 1.27%
Q3 97.3%
Q4 2.66%
Q3 95.9%
Q4 4.05%
Q3 97.8%
Q4 2.15%
Q3 96.0%
Q4 4.05%
SSC
FWD1

FIG. 16B

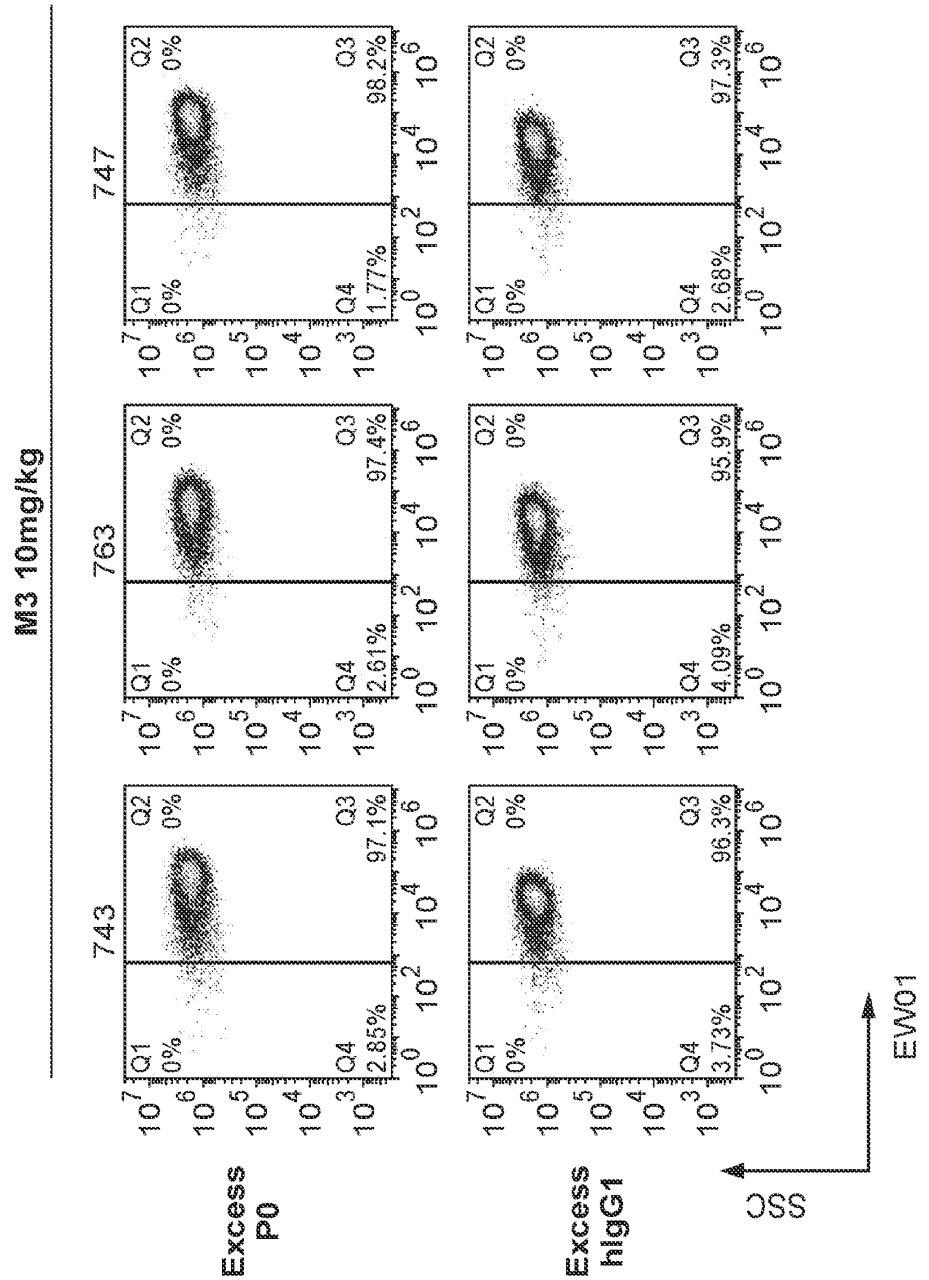


FIG. 16B Continued

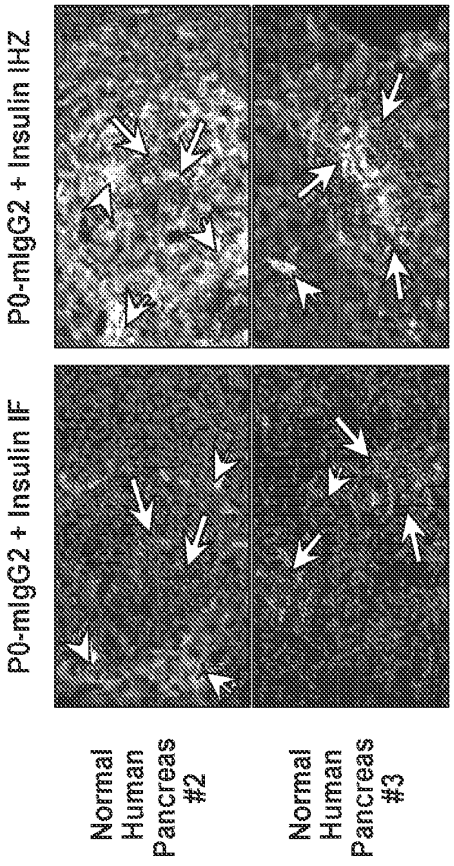

P0-mIgG2 + Insulin IHZ
P0-mIgG2 + Insulin IF
Normal Human Pancreas #2
Normal Human Pancreas #3

FIG. 17

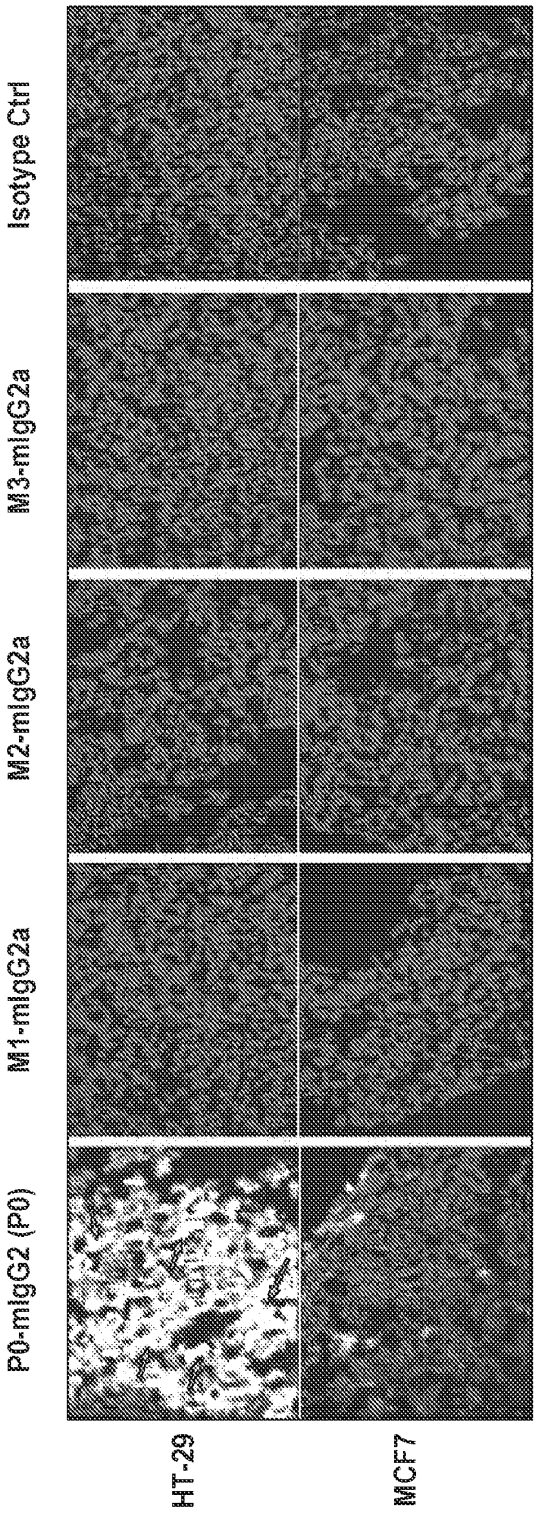

Isotype Ctrl
M3-mIgG2a
M2-mIgG2a
M1-mIgG2a
P0-mIgG2 (P0)
HT-29
MCF7

FIG. 18A

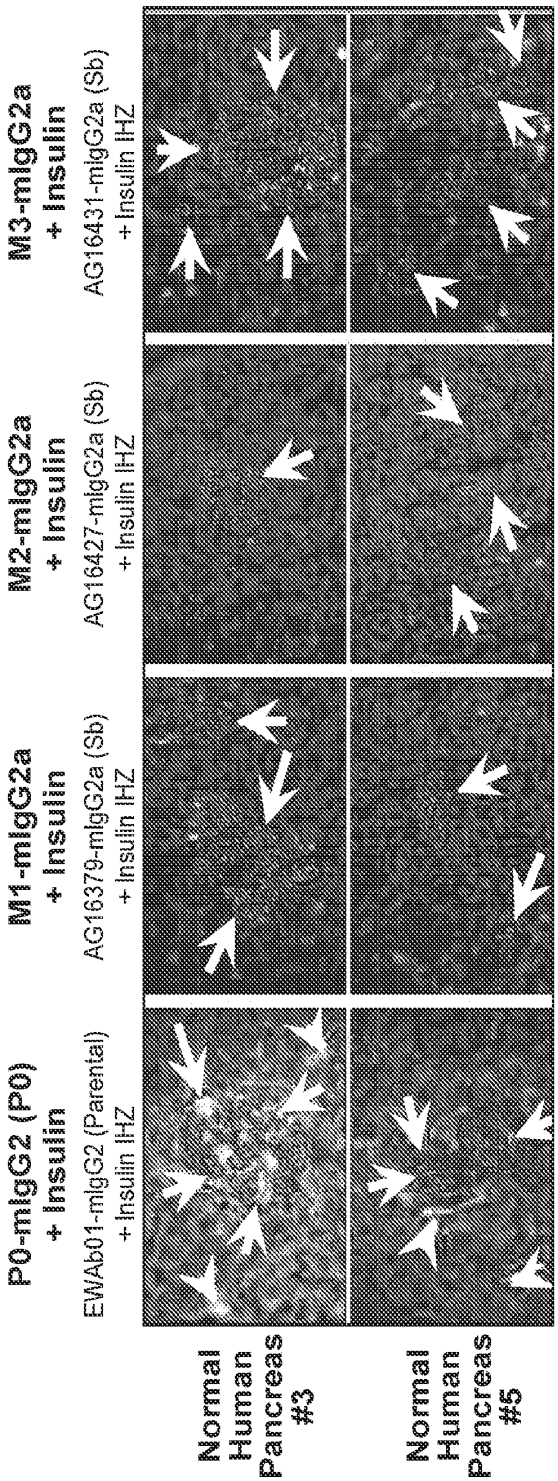

P0-mIgG2 (P0)
+ Insulin
EWAb01-mIgG2 (Parental)
+ Insulin IHZ
M1-mIgG2a
+ Insulin
AG16379-mIgG2a (Sb)
+ Insulin IHZ
M2-mIgG2a
+ Insulin
AG16427-mIgG2a (Sb)
+ Insulin IHZ
M3-mIgG2a
+ Insulin
AG16431-mIgG2a (Sb)
+ Insulin IHZ
Normal Human Pancreas #3
Normal Human Pancreas #5

FIG. 18B

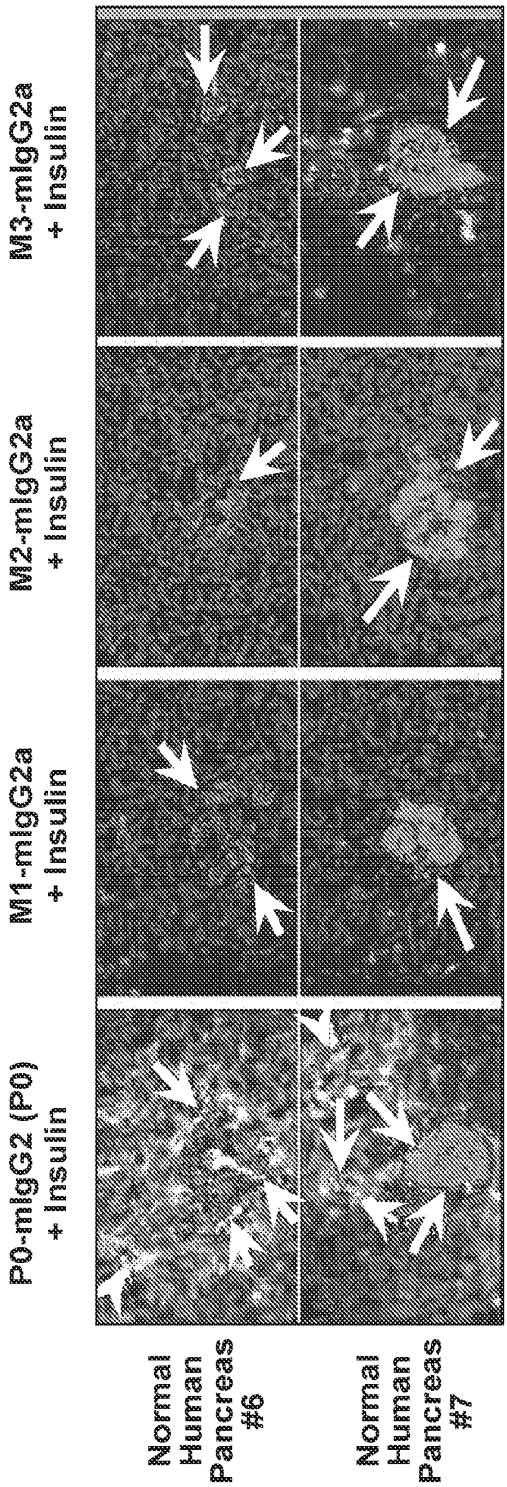

P0-mIgG2 (P0)
+ Insulin
M1-mIgG2a
+ Insulin
M2-mIgG2a
+ Insulin
M3-mIgG2a
+ Insulin
Normal Human Pancreas #6
Normal Human Pancreas #7

FIG. 18C

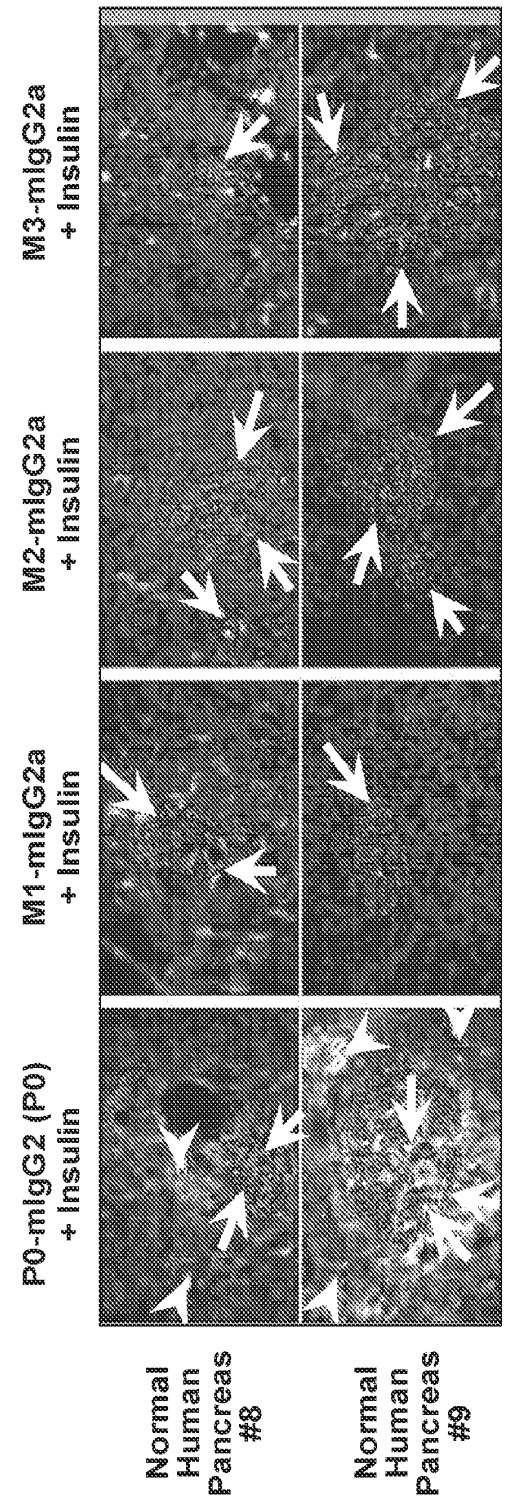

P0-mIgG2 (P0)
+ Insulin
M1-mIgG2a
+ Insulin
M2-mIgG2a
+ Insulin
M3-mIgG2a
+ Insulin
Normal Human Pancreas #8
Normal Human Pancreas #9

FIG. 18D

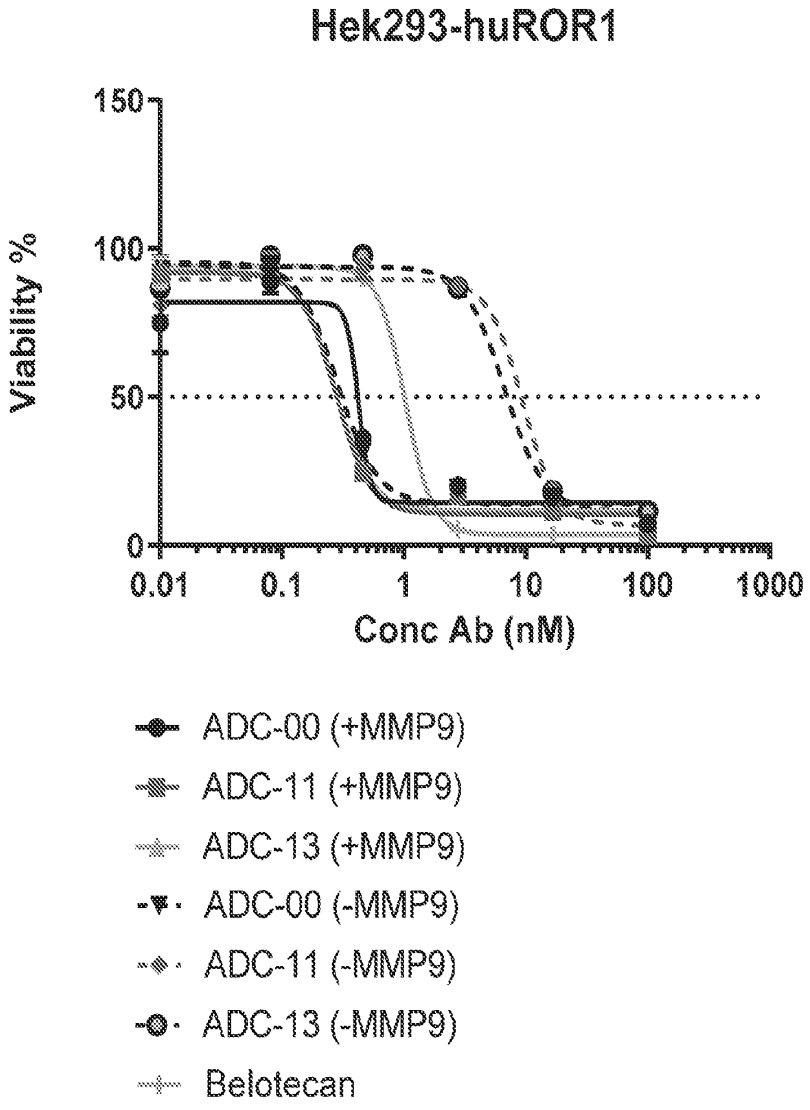

Hek293-huROR1
Viability %
150
100
50
0
0.01
0.1
1
10
100
1000
Conc Ab (nM)
ADC-00 (+MMP9)
ADC-11 (+MMP9)
ADC-13 (+MMP9)
ADC-00 (-MMP9)
ADC-11 (-MMP9)
ADC-13 (-MMP9)
Belotecan

FIG. 19A

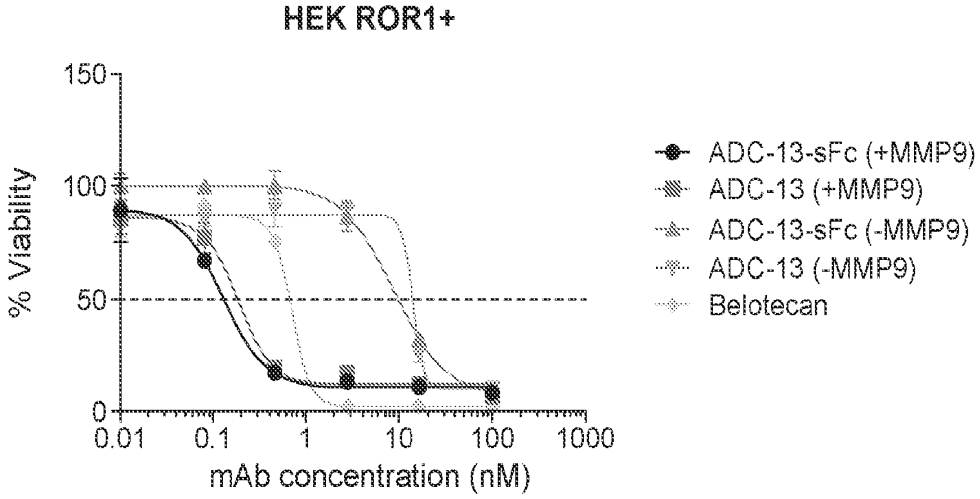


FIG. 19B

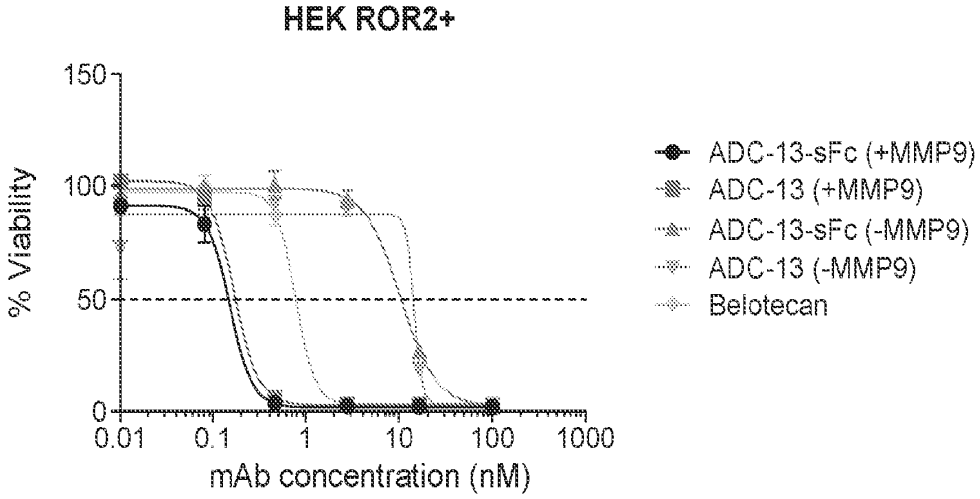
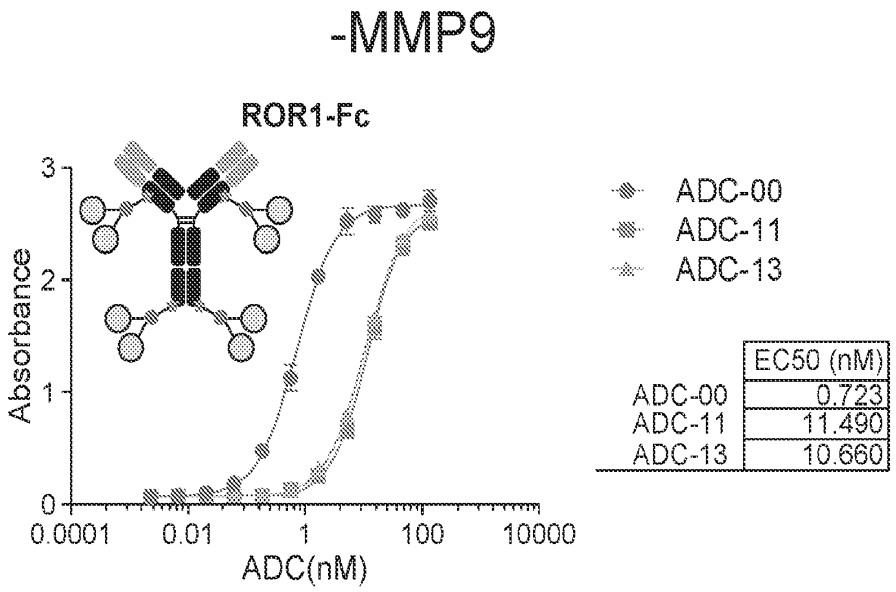
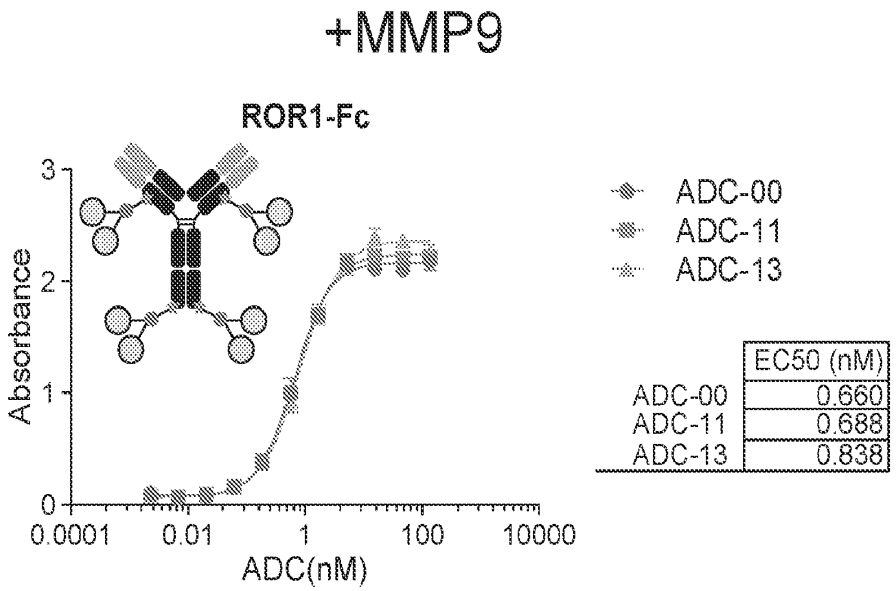


FIG. 19C



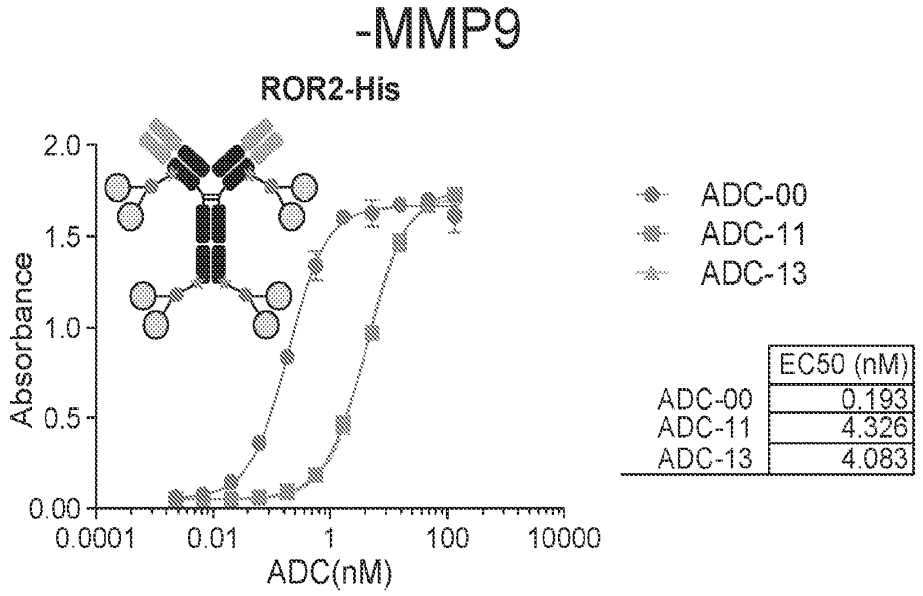
	EC50 (nM)
ADC-00	0.723
ADC-11	11.490
ADC-13	10.660

FIG. 20A



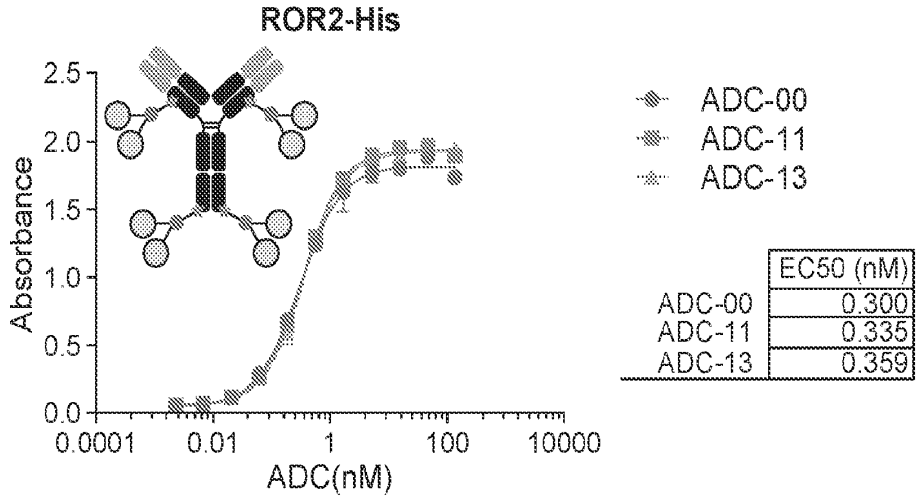
	EC50 (nM)
ADC-00	0.660
ADC-11	0.688
ADC-13	0.838

FIG. 20B



	EC50 (nM)
ADC-00	0.193
ADC-11	4.326
ADC-13	4.083

FIG. 20C



	EC50 (nM)
ADC-00	0.300
ADC-11	0.335
ADC-13	0.359

FIG. 20D

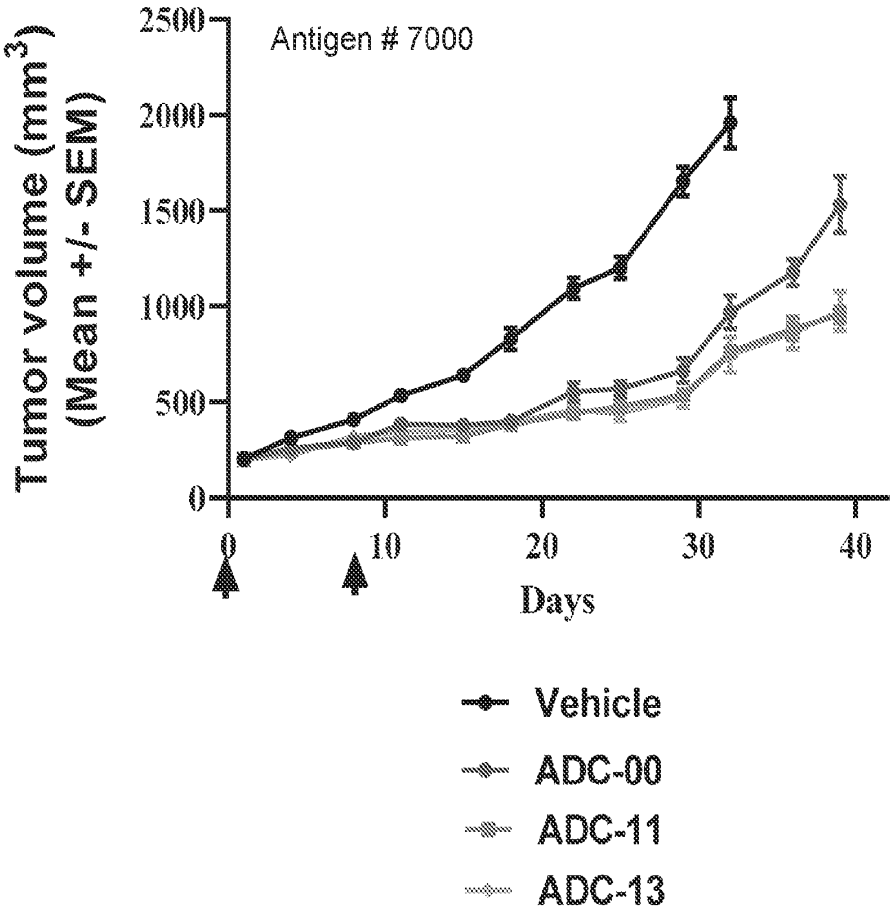

Antigen # 7000
Tumor volume (mm3)
(Mean +/- SEM)
2500
2000
1500
1000
500
0
0
10
20
30
40
Days
Vehicle
ADC-00
ADC-11
ADC-13

FIG. 21A

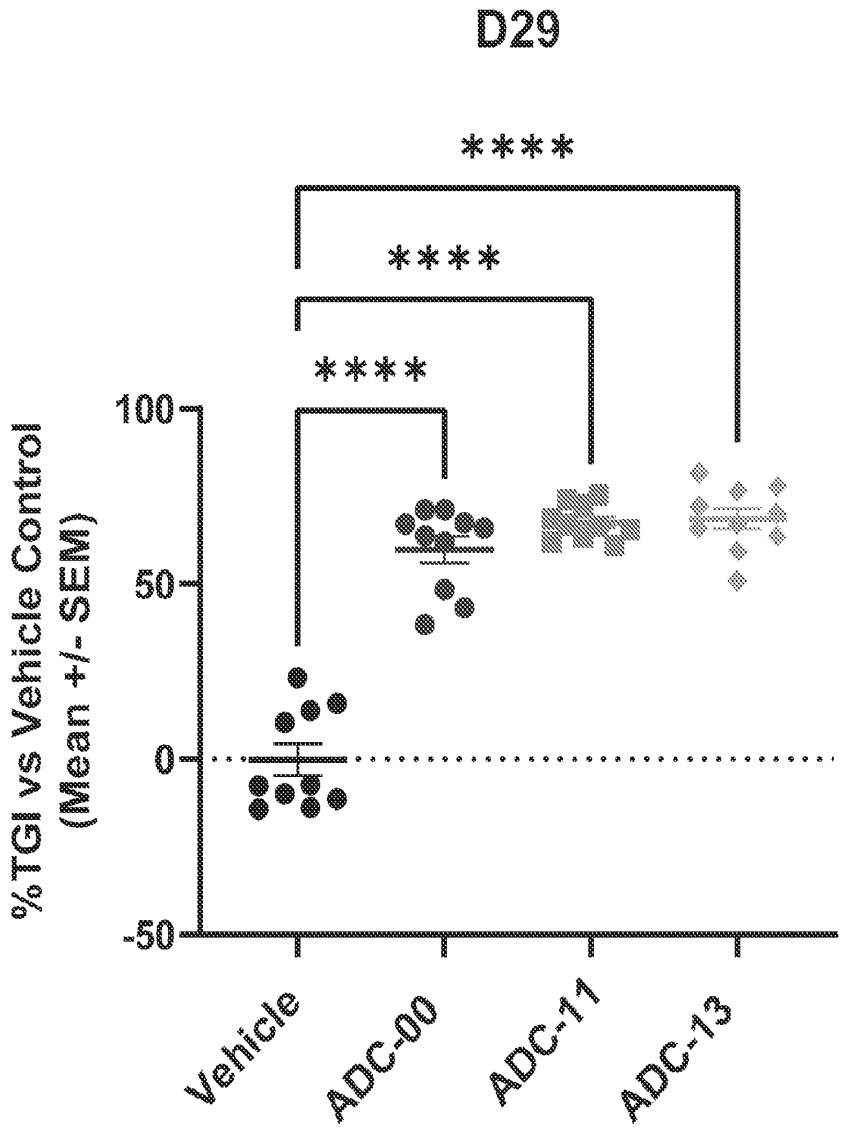

D29
****
****
****
100
50
0
-50
%TGI vs Vehicle Control
(Mean +/- SEM)
Vehicle
ADC-00
ADC-11
ADC-13

FIG. 21B

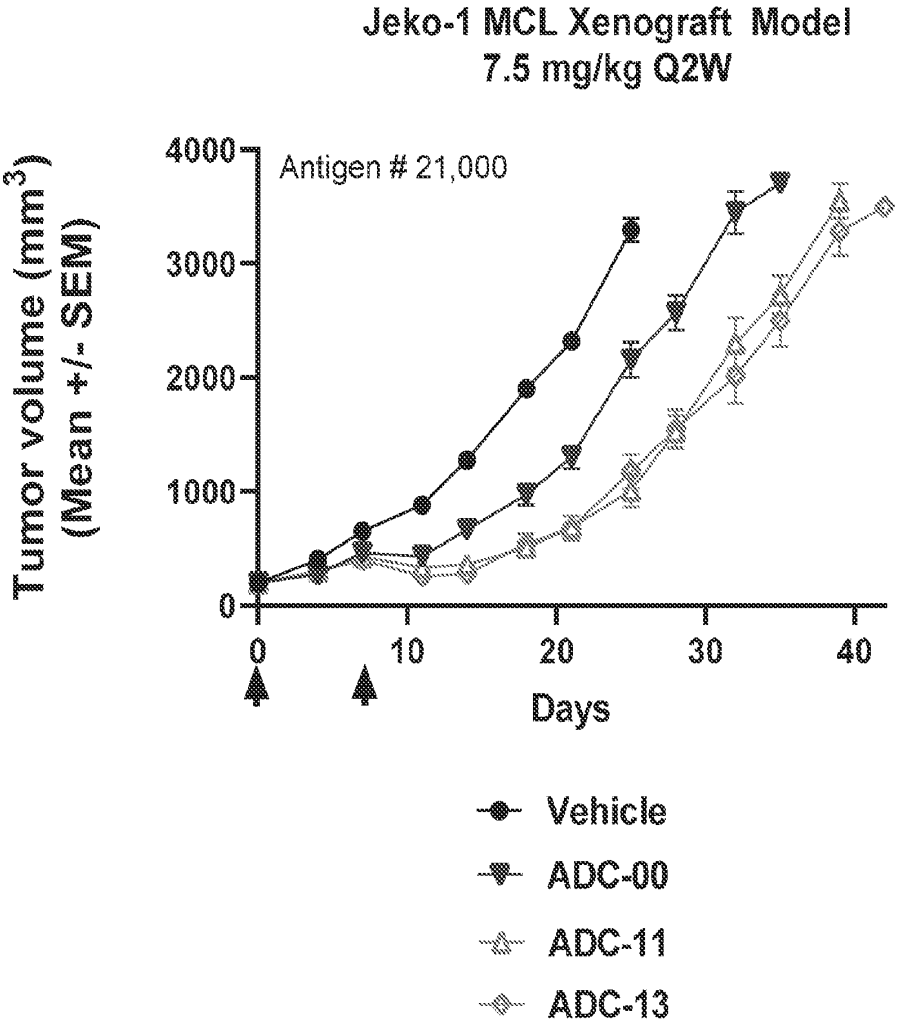

Jeko-1 MCL Xenograft Model
7.5 mg/kg Q2W
Antigen # 21,000
Tumor volume (mm3)
(Mean +/- SEM)
4000
3000
2000
1000
0
0
10
20
30
40
Days
Vehicle
ADC-00
ADC-11
ADC-13

FIG. 22A

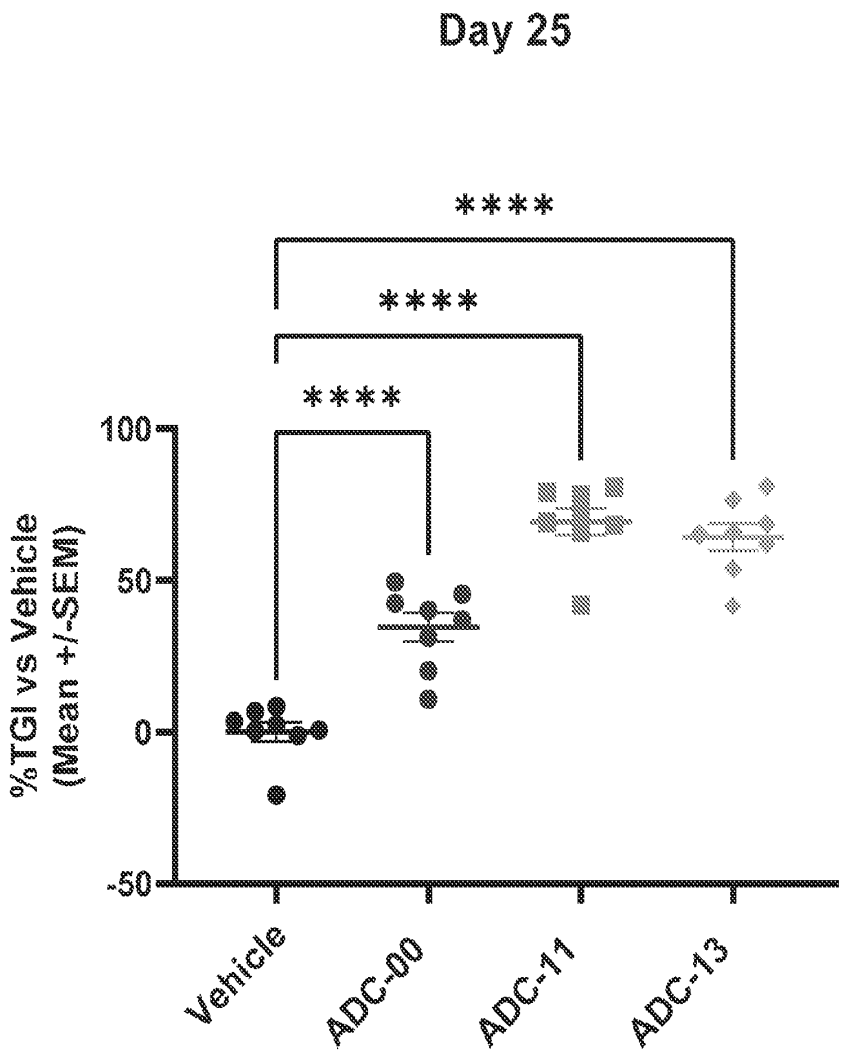

Day 25
****
****
****
100
50
0
-50
%TGI vs Vehicle
(Mean +/-SEM)
Vehicle
ADC-00
ADC-11
ADC-13

FIG. 22B

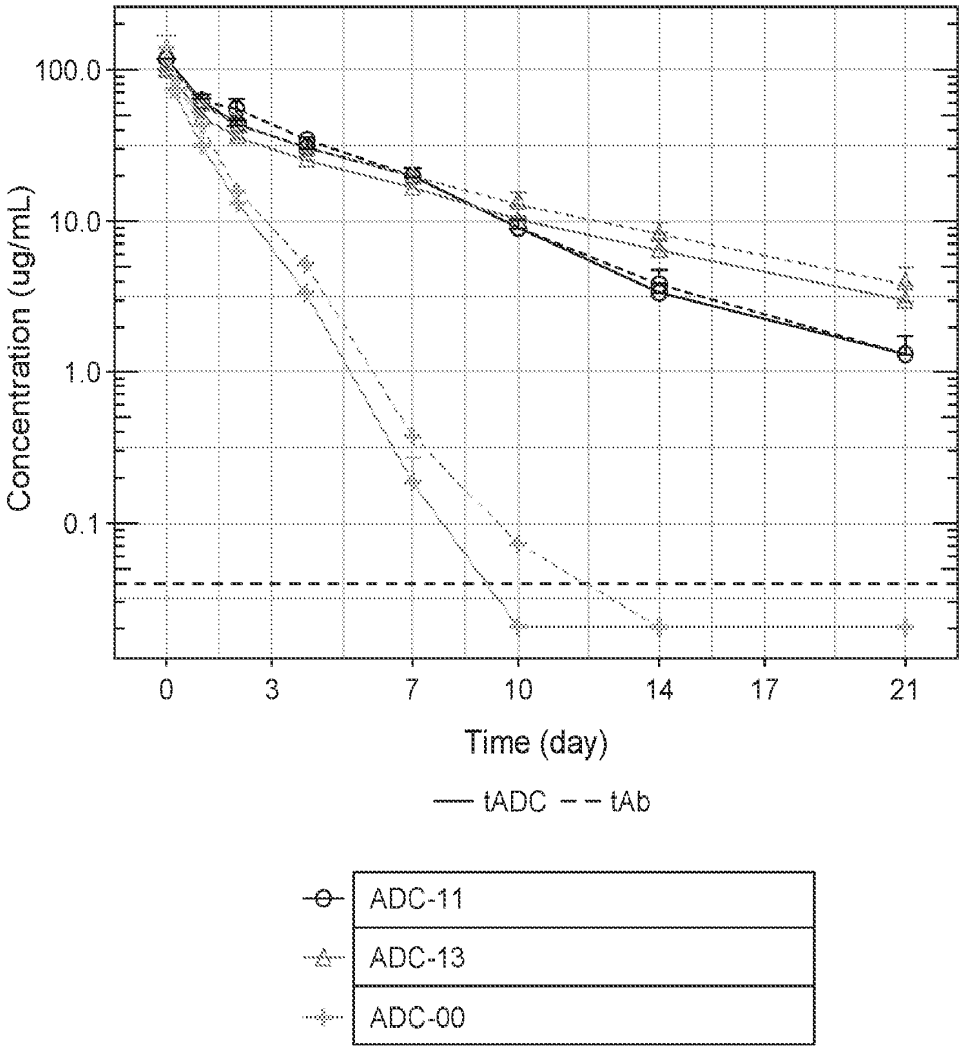

100.0
10.0
1.0
0.1
Concentration (ug/mL)
0
3
7
10
14
17
21
Time (day)
tADC
tAb
ADC-11
ADC-13
ADC-00

FIG. 23

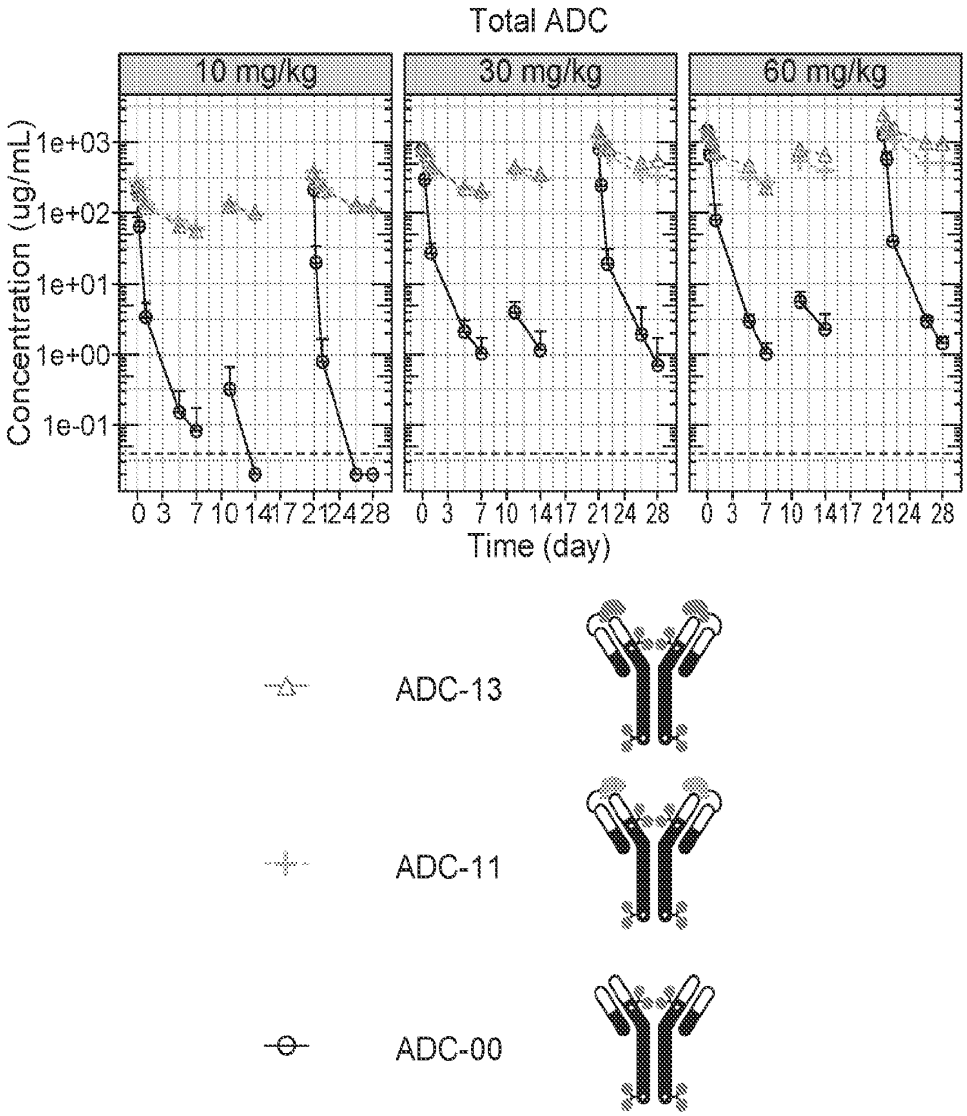
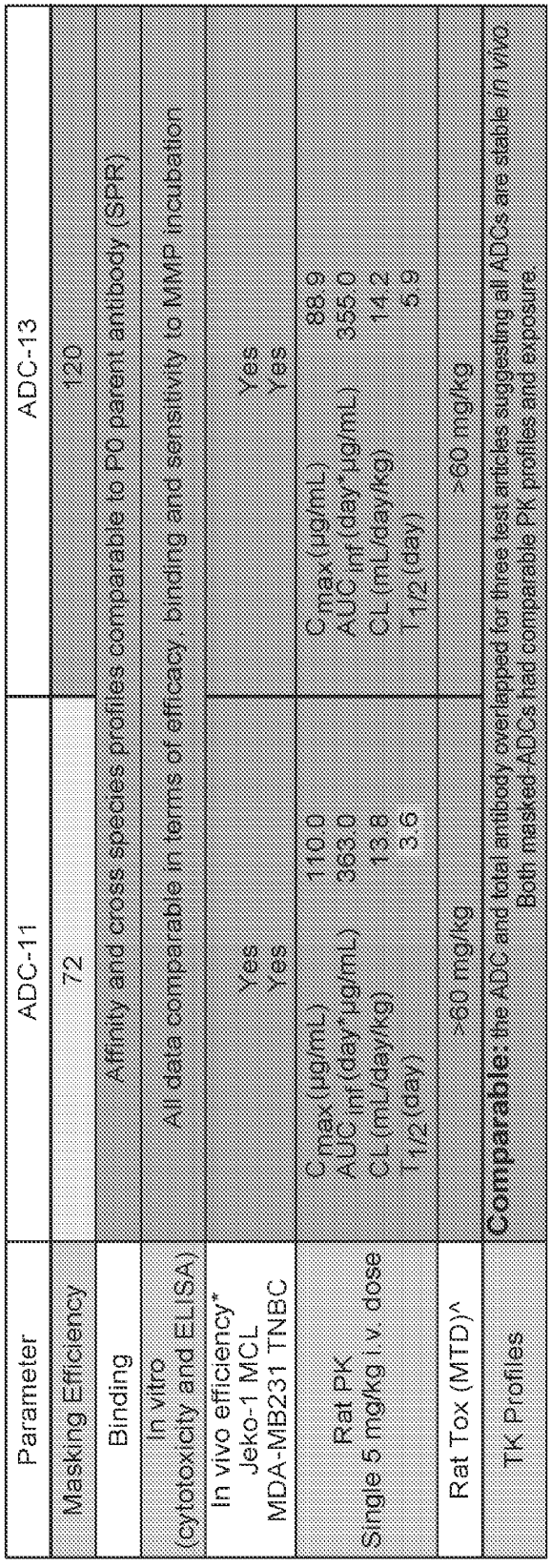

Total ADC
10 mg/kg
30 mg/kg
60 mg/kg
Concentration (ug/mL)
1e+03
1e+02
1e+01
1e+00
1e-01
0 3 7 10 14 17 21 24 28
Time (day)
ADC-13
ADC-11
ADC-00

FIG. 24



Parameter	ADC-11	ADC-13
Masking Efficiency	72	120
Binding	Affinity and cross species profiles comparable to P0 parent antibody (SPR)	
In vitro (cytotoxicity and ELISA)	All data comparable in terms of efficacy, binding and sensitivity to MMP incubation	
In vivo efficiency* Jeko-1 MCL	Yes	Yes
MDA-MB231 TNBC	Yes	Yes
Rat PK Single 5 mg/kg i.v. dose	C_{max} (µg/mL) 110.0 AUC_{inf} (day*µg/mL) 363.0 CL (mL/day/kg) 13.8 T1/2 (day) 3.6	C_{max} (µg/mL) 88.9 AUC_{inf} (day*µg/mL) 355.0 CL (mL/day/kg) 14.2 T1/2 (day) 5.9
Rat Tox (MTD)^	>80 mg/kg	>80 mg/kg
TK Profiles	**Comparable:** the ADC and total antibody overlapped for three test articles suggesting all ADCs are stable *in vivo* Both masked ADCs had comparable PK profiles and exposure	

*p < 0.05 %TGI vs isotype control
^projected

FIG. 25

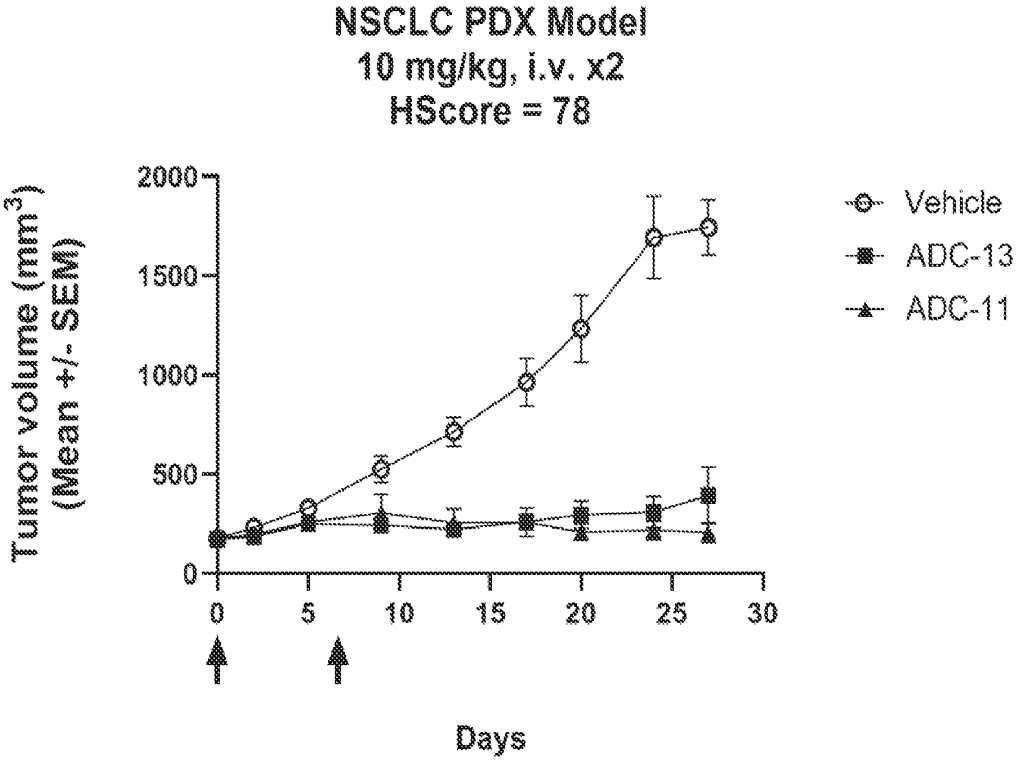

NSCLC PDX Model
10 mg/kg, i.v. x2
HScore = 78
Tumor volume (mm3)
(Mean +/- SEM)
2000
1500
1000
500
0
0
5
10
15
20
25
30
Days
Vehicle
ADC-13
ADC-11

FIG. 26