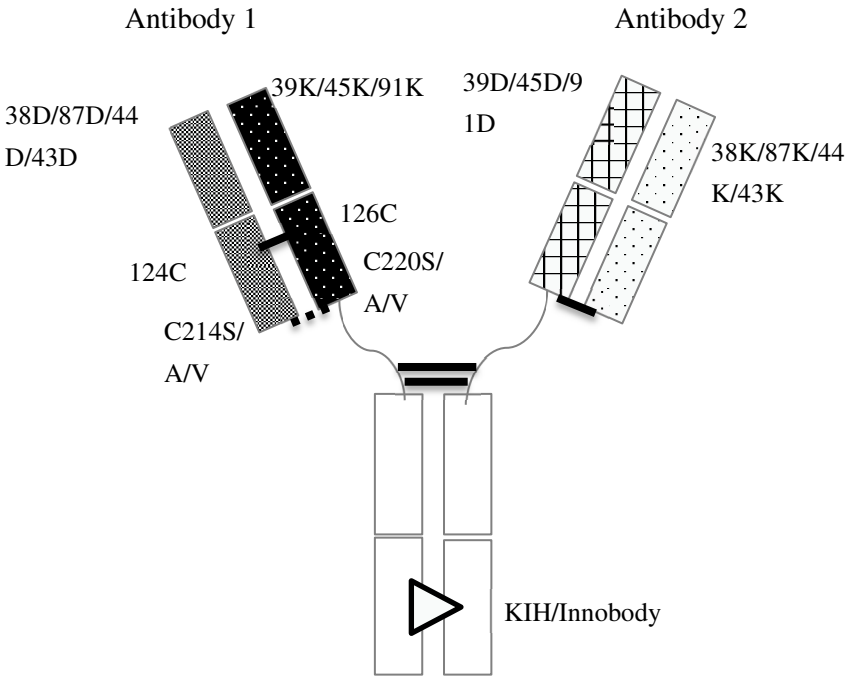

Antibody 1
Antibody 2
38D/87D/44
D/43D
39K/45K/91K
39D/45D/9
1D
38K/87K/44
K/43K
126C
124C
C220S/
A/V
C214S/
A/V
KIH/Innobody

Figure 1

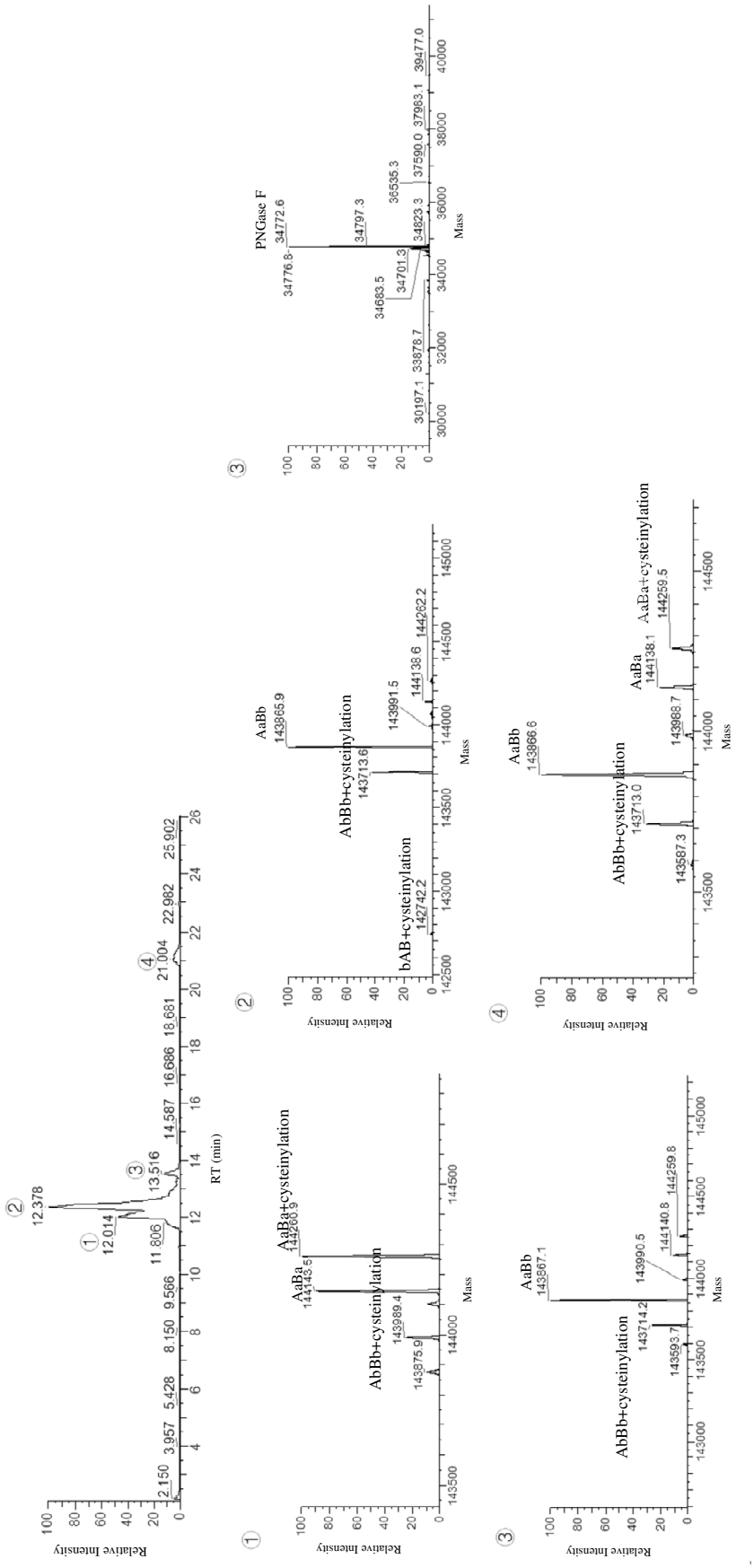

PNGase F
34772.6
34776.8
34797.3
34683.5
34701.3
34823.3
36535.3
37590.0
37983.1
39477.0
30197.1
33878.7
Mass
AaBb
143865.9
AbBb+cysteinylation
143713.6
143991.5
144138.6
144262.2
bAB+cysteinylation
142742.2
AaBa+cysteinylation
144260.9
AaBa
144143.5
AbBb+cysteinylation
143989.4
143875.9
AaBb
143866.6
AbBb+cysteinylation
143713.0
143587.3
AaBa
144138.1
AaBa+cysteinylation
144259.5
143988.7
AaBb
143867.1
AbBb+cysteinylation
143714.2
143990.5
144140.8
144259.8
143593.7
RT (min)
Relative Intensity

Figure 2A

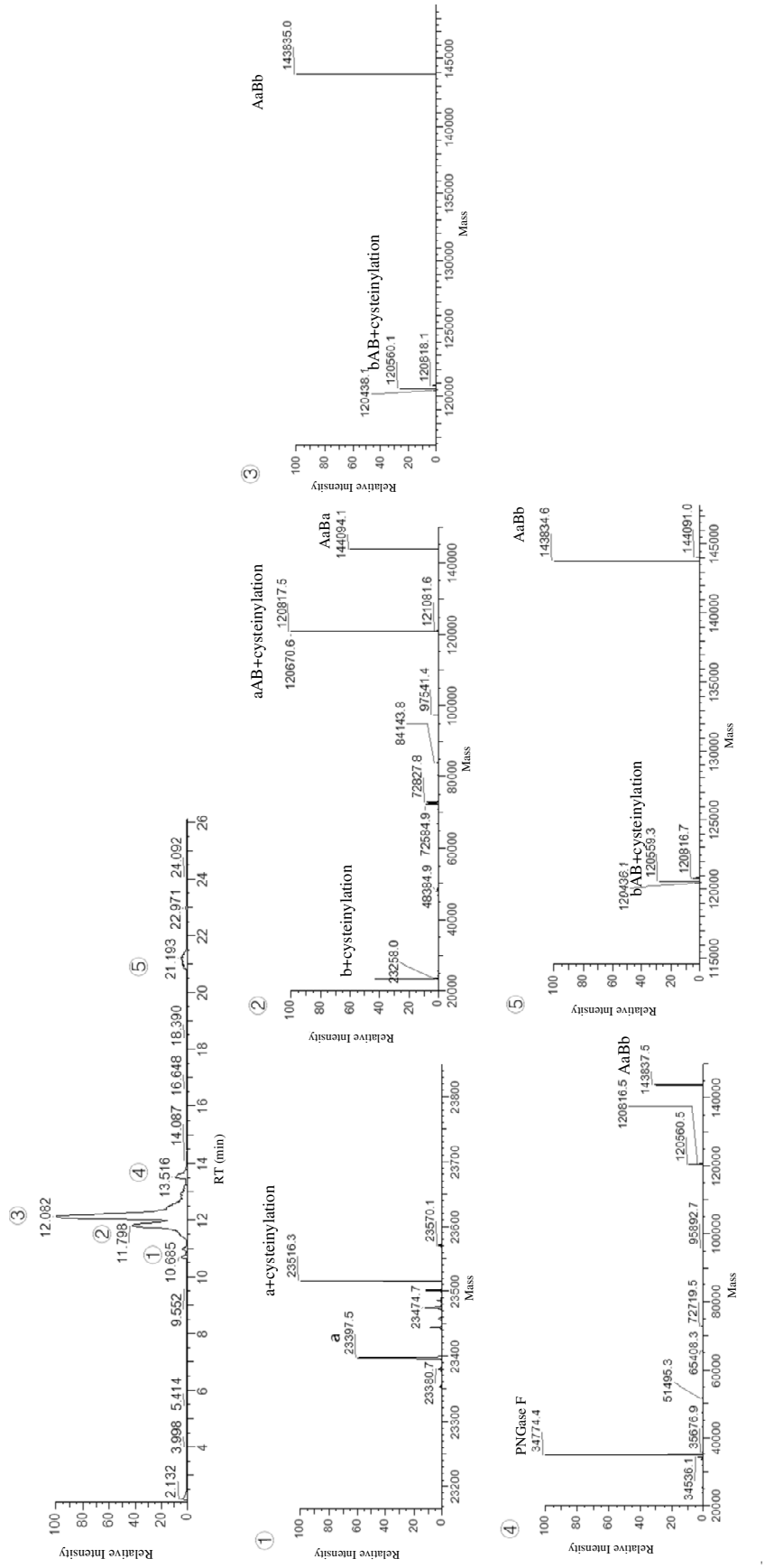

Relative Intensity
RT (min)
2.132
3.998
5.414
9.552
10.685
11.798
12.082
13.516
14.087
16.648
18.390
21.193
22.971
24.092
①
②
③
④
⑤
a+cysteinylation
23516.3
a
23397.5
23474.7
23570.1
23380.7
b+cysteinylation
23258.0
48384.9
72584.9
72827.8
84143.8
97541.4
aAB+cysteinylation
120670.6
120817.5
AaBa
144094.1
121081.6
bAB+cysteinylation
120438.1
120560.1
120618.1
AaBb
143835.0
PNGase F
34774.4
34536.1
35676.9
51496.3
65408.3
72719.5
95892.7
120616.5
AaBb
143837.5
120560.5
120436.1
bAB+cysteinylation
120559.3
120816.7
AaBb
143834.6
144091.0
Mass

Figure 2B

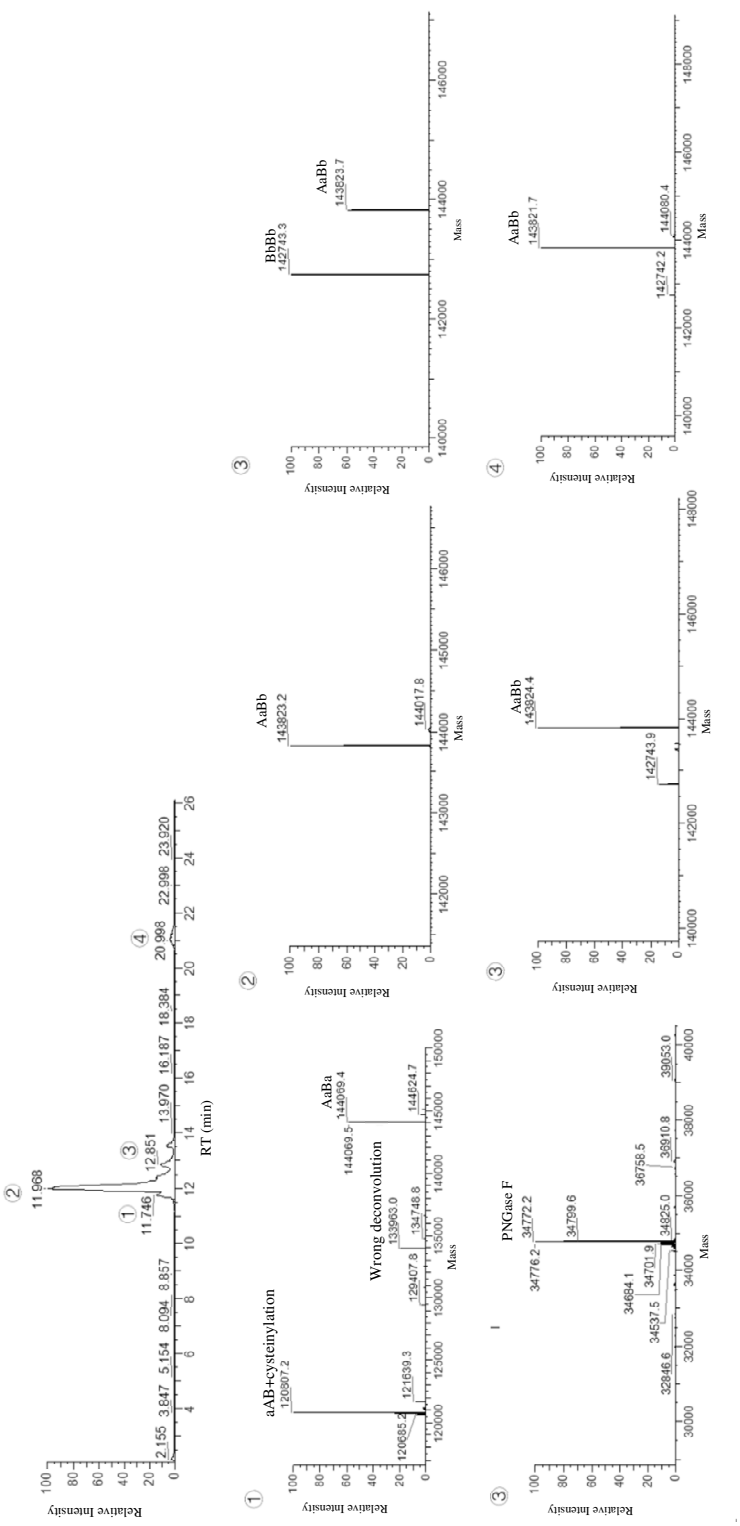

RT (min)
Relative Intensity
Mass
aAB+cysteinylation
120907.2
120685.2
121639.3
129407.8
133963.0
134748.8
144524.7
Wrong deconvolution
AaBa
144069.4
144069.5
AaBb
143823.2
144017.8
BbBb
142743.3
AaBb
143823.7
PNGase F
34772.2
34776.2
34759.6
34684.1
34701.9
34537.5
34825.0
36758.5
38910.8
39053.0
32846.6
AaBb
143824.4
142743.9
AaBb
143821.7
142742.2
144080.4

Figure 2C

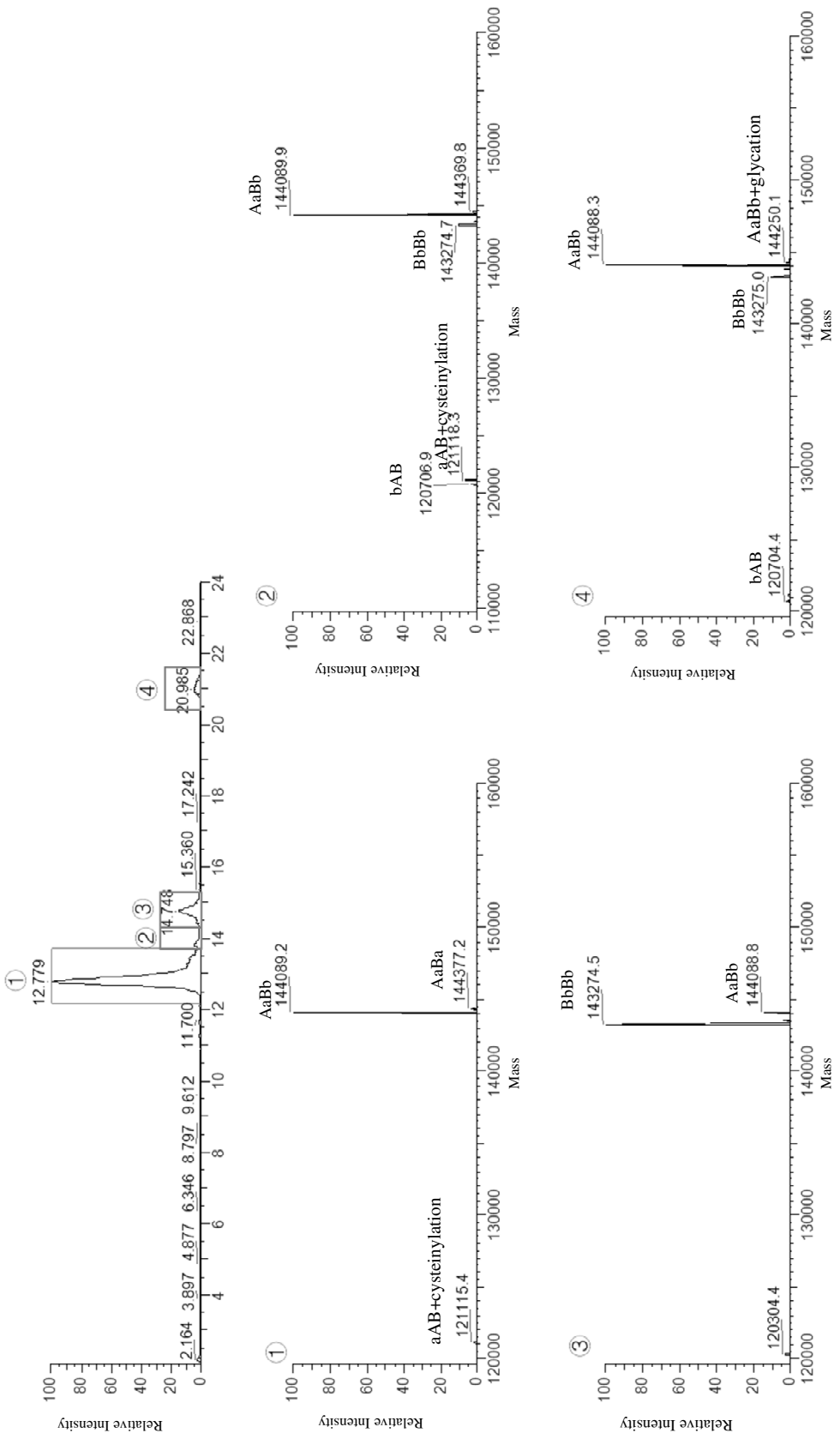

①
12.779
②
③
14.748
④
20.985
22.868
15.360
17.242
11.700
9.612
8.797
6.346
4.877
3.897
2.164
Relative Intensity
aAB+cysteinylation
121115.4
AaBb
144089.2
AaBa
144377.2
Mass
bAB
120706.9
aAB+cysteinylation
121118.3
AaBb
144089.9
BbBb
143274.7
144369.8
BbBb
143274.5
AaBb
144088.8
120304.4
bAB
120704.4
AaBb
144088.3
BbBb
143275.0
AaBb+glycation
144250.1

Figure 3A

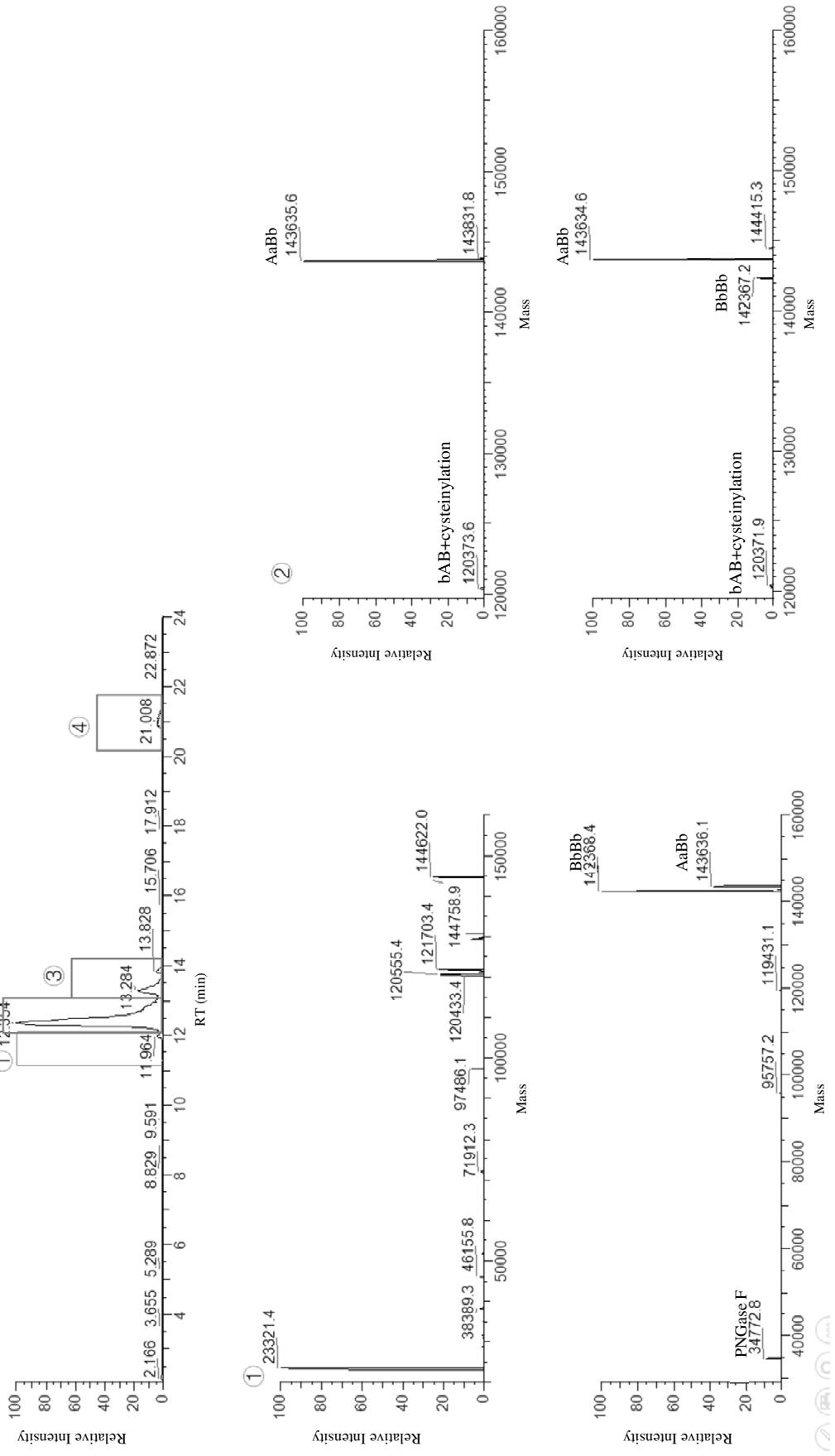

Relative Intensity
RT (min)
① 12.354
② 11.964
③ 13.284
④ 21.008
2.166
3.655
5.289
8.829
9.591
13.828
15.706
17.912
22.872
Mass
AaBb
143635.6
143831.8
bAB+cysteinylation
120373.6
BbBb
142367.2
144415.3
120371.9
23321.4
38389.3
46155.8
71912.3
97486.1
120433.4
120555.4
121703.4
144758.9
144622.0
BbBb
142368.4
AaBb
143636.1
PNGase F
34772.8
95757.2
119431.1

Figure 3B

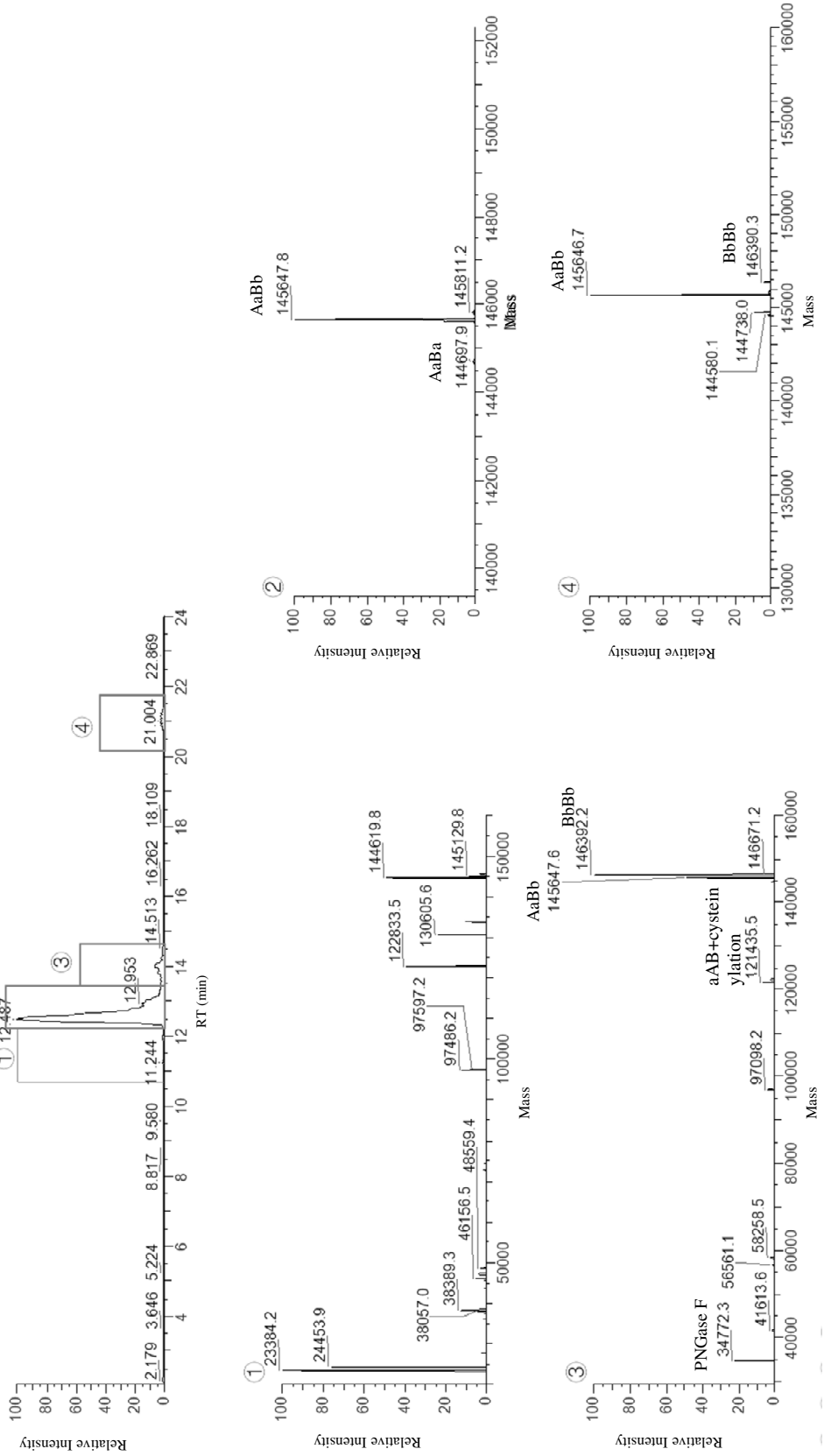

RT (min)
Relative Intensity
Mass
AaBb
145647.8
AaBa
144697.9
145811.2
BbBb
146390.3
144580.1
144738.0
145646.7
23384.2
24453.9
38057.0
38389.3
46156.5
48559.4
97597.2
97486.2
122833.5
130605.6
144619.8
145129.8
PNGase F
34772.3
56561.1
41613.6
58258.5
97098.2
aAB+cystein
ylation
121435.5
146392.2
146671.2

Figure 3C

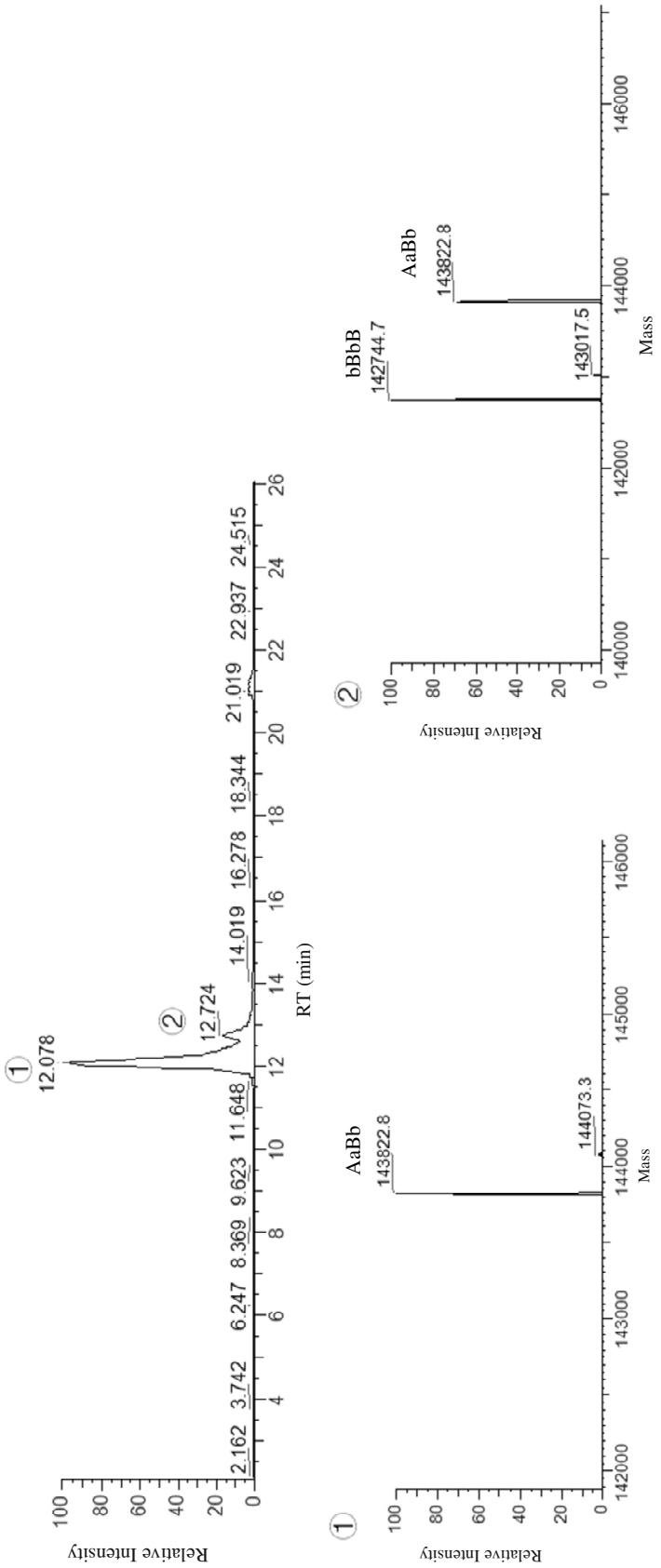

①
12.078
②
12.724
14.019
2.162
3.742
6.247
8.369
9.623
11.648
16.278
18.344
21.019
22.937
24.515
RT (min)
Relative Intensity
AaBb
143822.8
144073.3
Mass
bBbB
142744.7
143017.5
AaBb
143822.8

Figure 4A

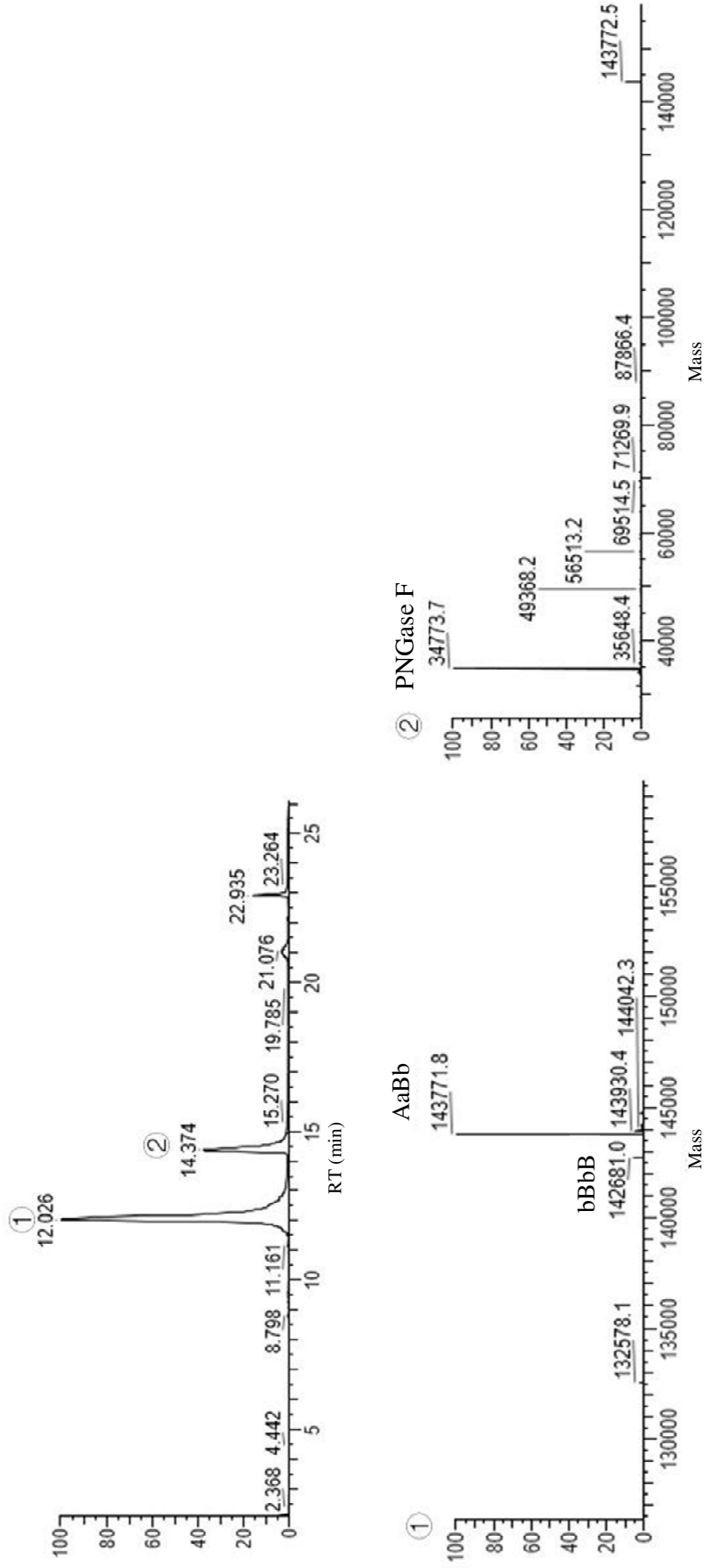

①
12.026
②
14.374
2.368
4.442
8.798
11.161
15.270
19.785
21.076
22.935
23.264
RT (min)
PNGase F
34773.7
49368.2
56513.2
35648.4
69514.5
71269.9
87866.4
143772.5
Mass
AaBb
143771.8
bBbB
142681.0
143930.4
144042.3
132578.1
Mass

Figure 4B

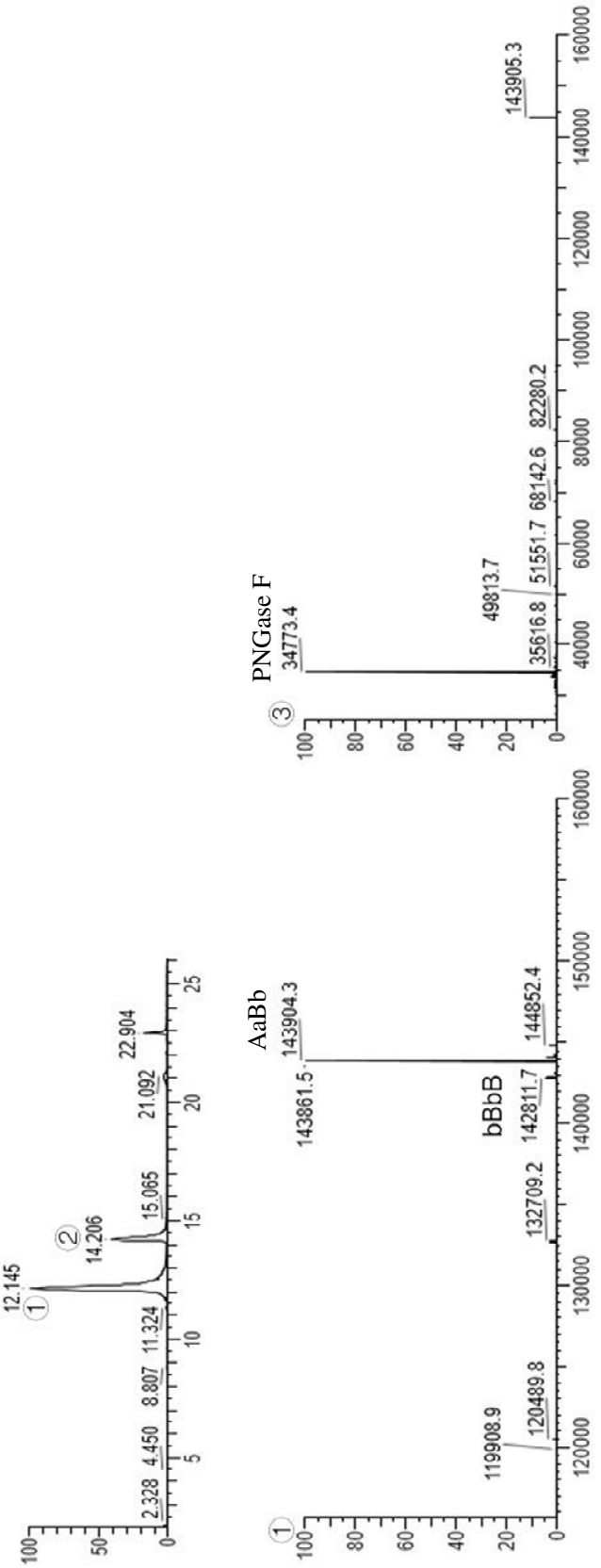

12.145
14.206
15.065
21.092
22.904
2.328
4.450
8.807
11.324
AaBb
143904.3
143861.5
144852.4
bBbB
142811.7
132709.2
119908.9
120489.8
PNGase F
34773.4
143905.3
49813.7
35616.8
51551.7
68142.6
82280.2

Figure 4C

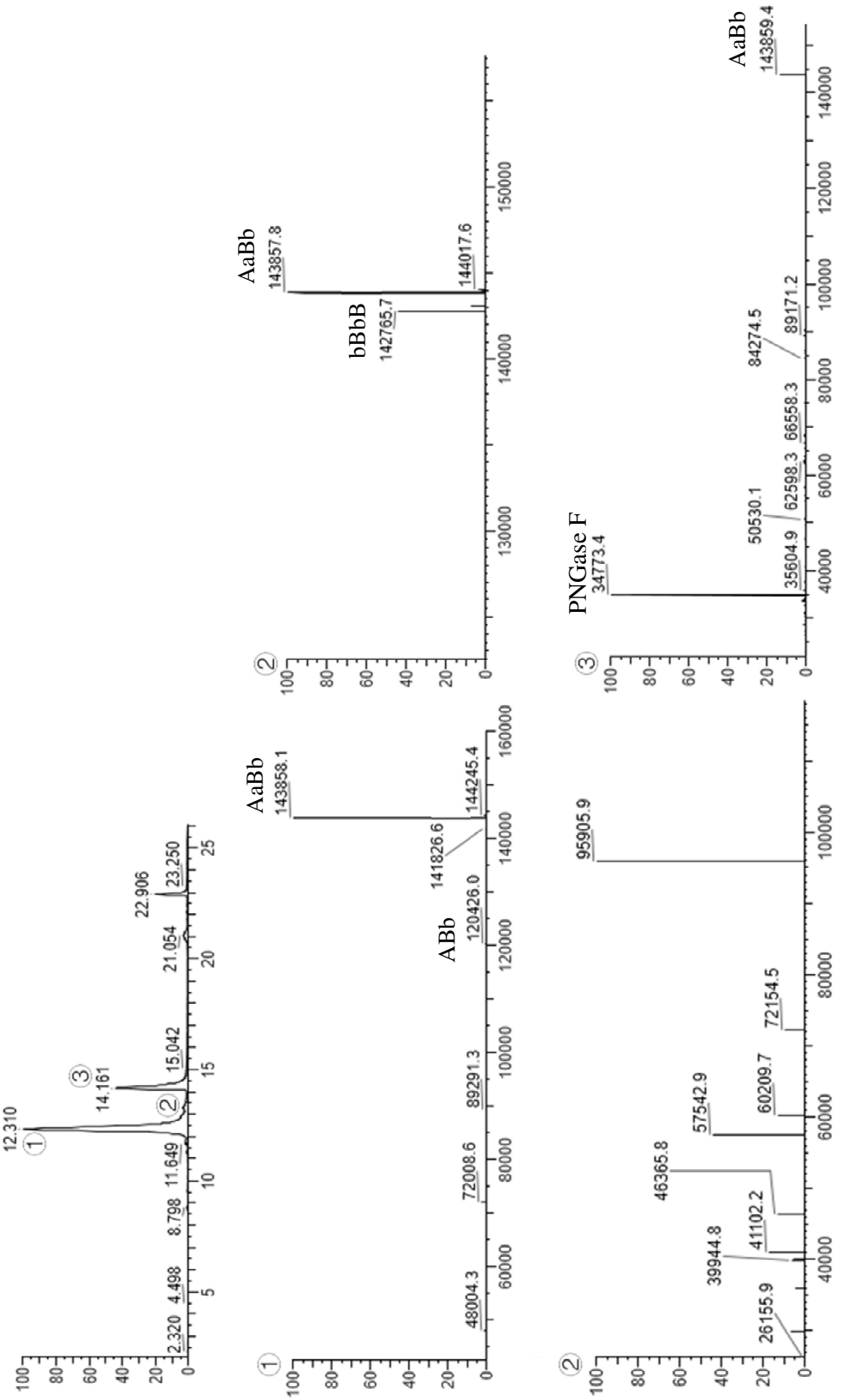

12.310
14.161
15.042
22.906
23.250
21.054
11.649
8.798
4.498
2.320
AaBb
143858.1
144245.4
141826.6
ABb
120426.0
89291.3
72008.6
48004.3
AaBb
143857.8
bBbB
142765.7
144017.6
PNGase F
34773.4
95905.9
46365.8
57542.9
60209.7
72154.5
39944.8
41102.2
26155.9
50530.1
62598.3
66558.3
84274.5
89171.2
35604.9
AaBb
143859.4

Figure 4D

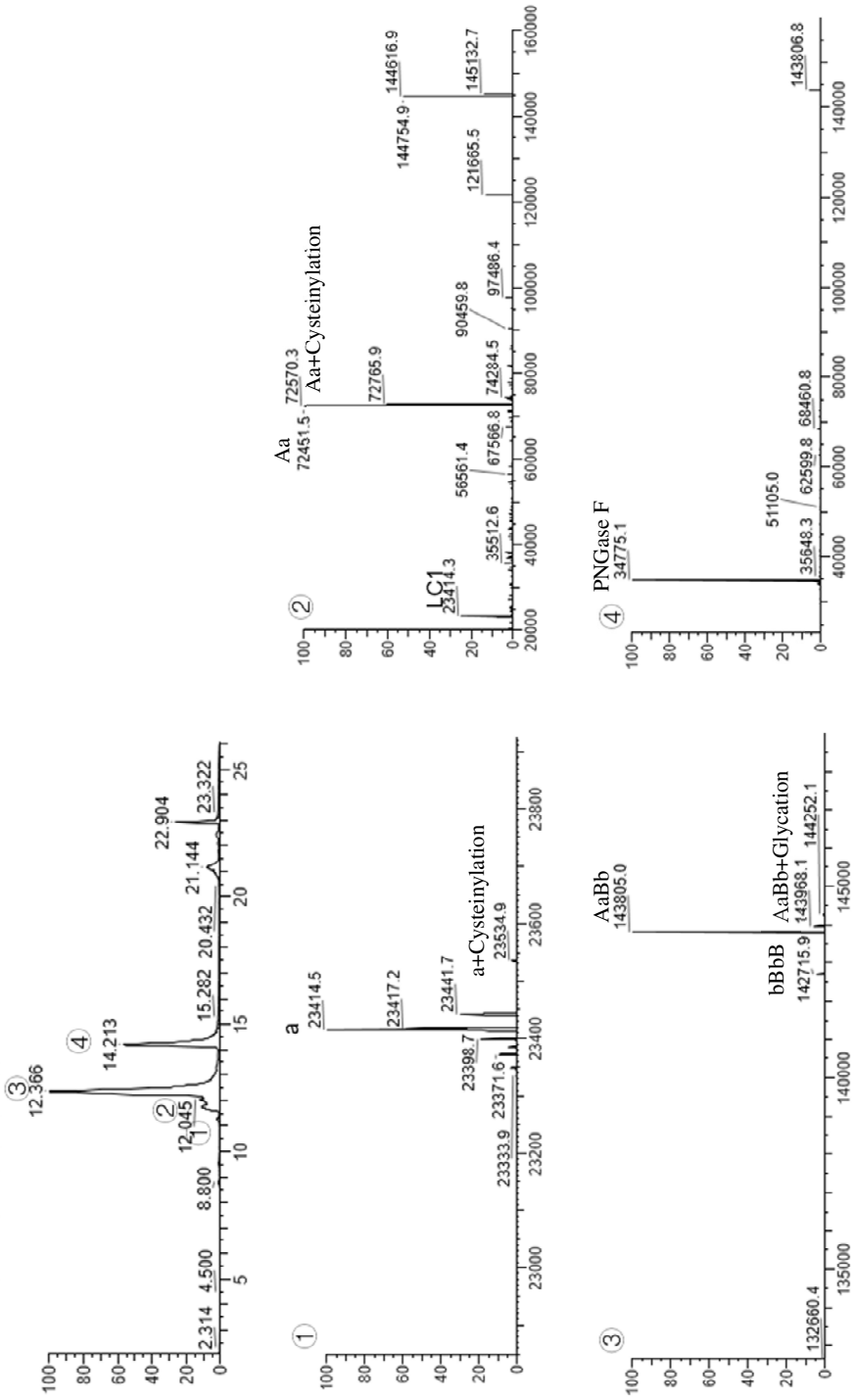

2.314
4.500
8.800
12.045
12.366
14.213
15.282
20.432
21.144
22.904
23.322
a
23414.5
23417.2
23441.7
a+Cysteinylation
23534.9
23398.7
23371.6
23333.9
AaBb
143805.0
bBbB
142715.9
AaBb+Glycation
143968.1
144252.1
132660.4
Aa
72451.5
72570.3
Aa+Cysteinylation
72765.9
LC1
23414.3
35512.6
56561.4
67566.8
74284.5
90459.8
97486.4
121665.5
144754.9
144616.9
145132.7
PNGase F
34775.1
35648.3
51105.0
62599.8
68460.8
143806.8

Figure 4E

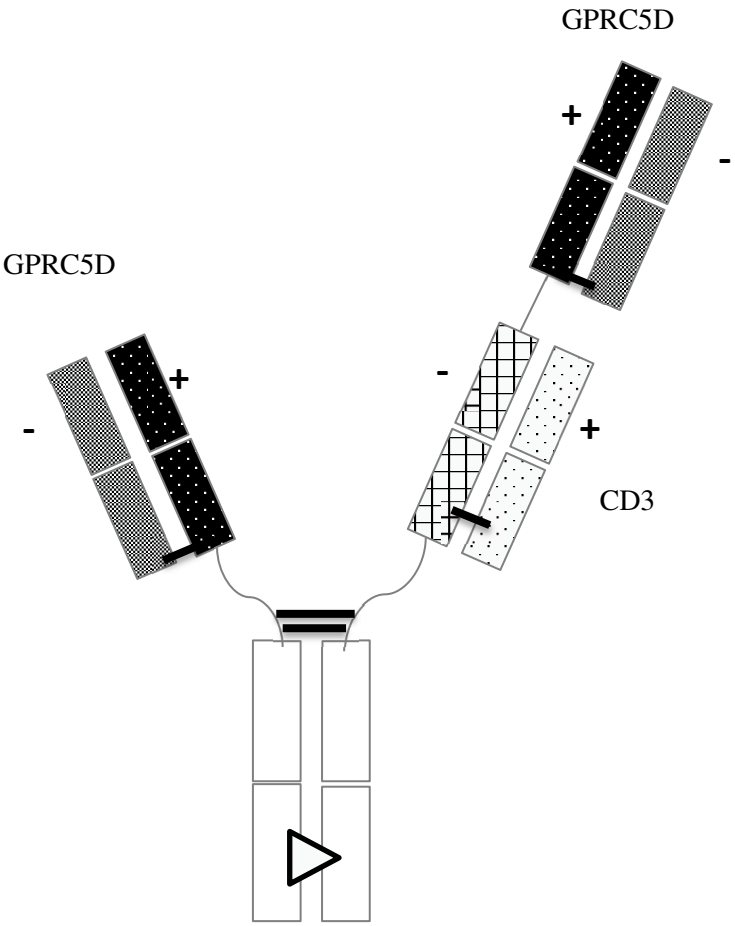


Figure 5A

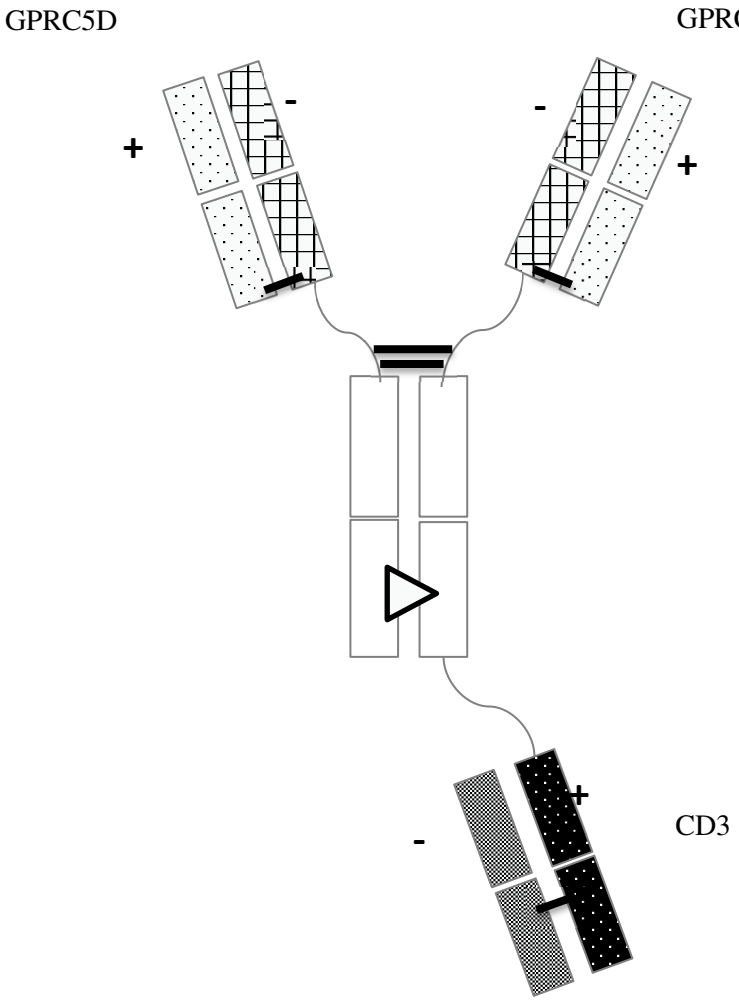


Figure 5B

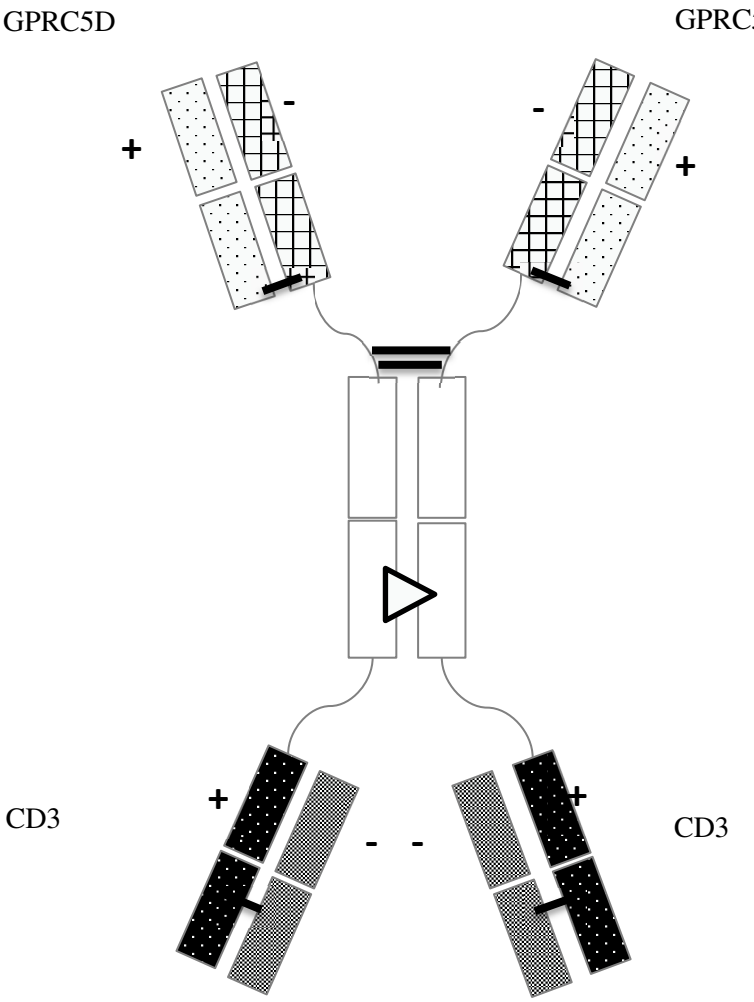


Figure 5C

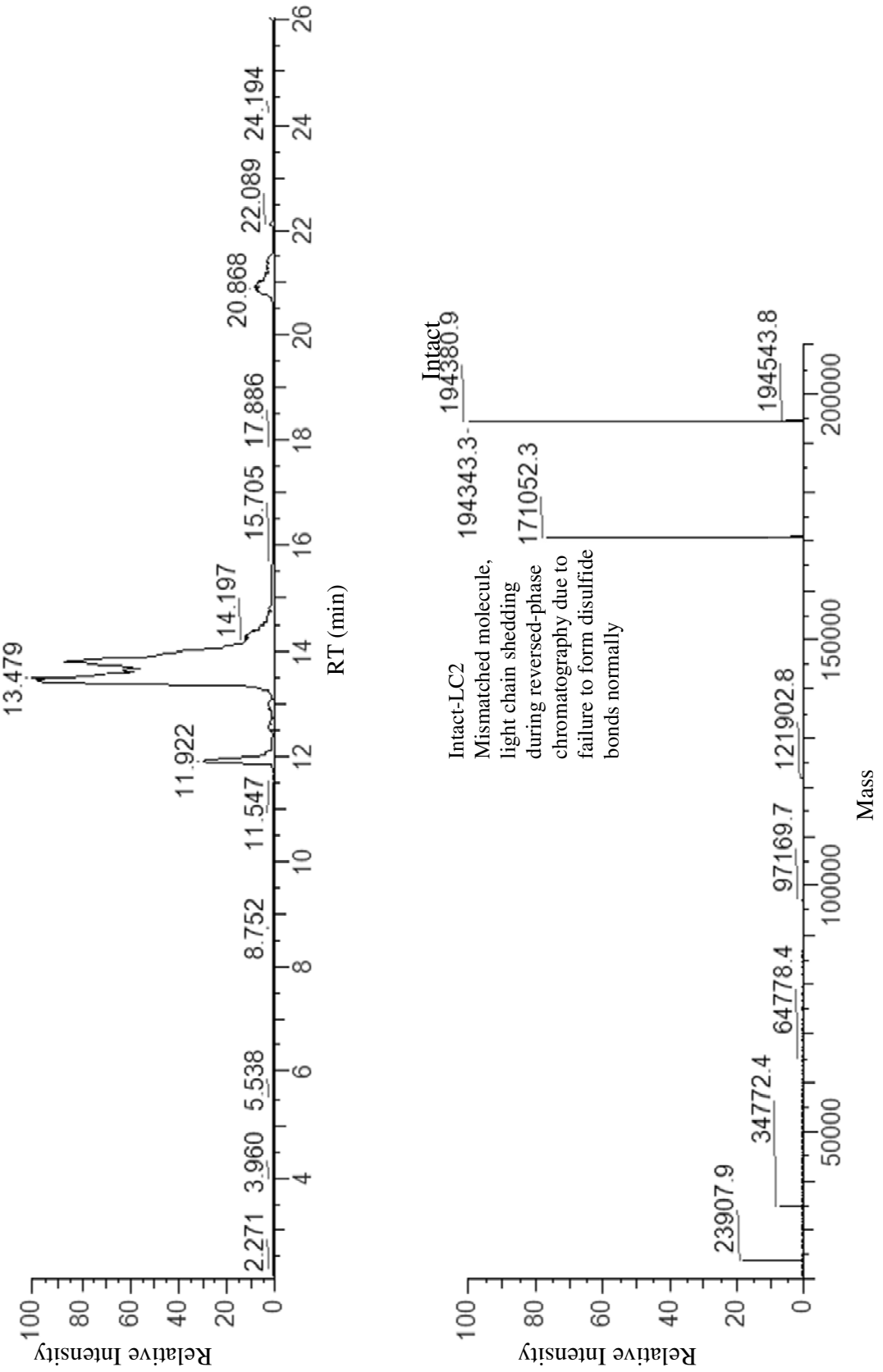

Relative Intensity
RT (min)
2.271
3.960
5.538
8.752
11.547
11.922
13.479
14.197
15.705
17.886
20.868
22.089
24.194
Intact
194380.9
194343.3
171052.3
194543.8
Intact-LC2
Mismatched molecule, light chain shedding during reversed-phase chromatography due to failure to form disulfide bonds normally
23907.9
34772.4
64778.4
97169.7
121902.8
Mass

Figure 6A

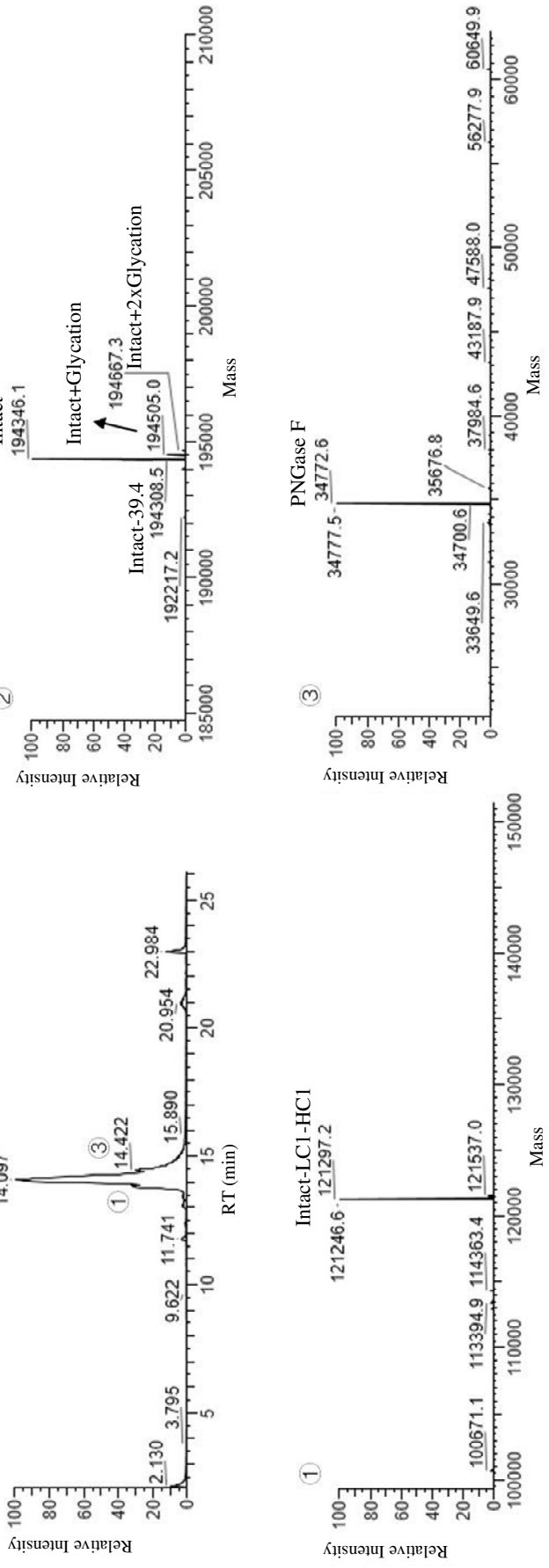

Intact
194346.1
Intact+Glycation
194667.3
Intact+2xGlycation
194505.0
Intact-39.4
194308.5
192217.2
Relative Intensity
Mass
PNGase F
34772.6
34777.5
35676.8
37984.6
43187.9
47588.0
56277.9
60649.9
34700.6
33649.6
Intact-LC1-HC1
121297.2
121246.6
121537.0
114363.4
113394.9
100671.1
2.130
3.795
9.622
11.741
14.097
14.422
15.890
20.954
22.984
RT (min)

Figure 6B

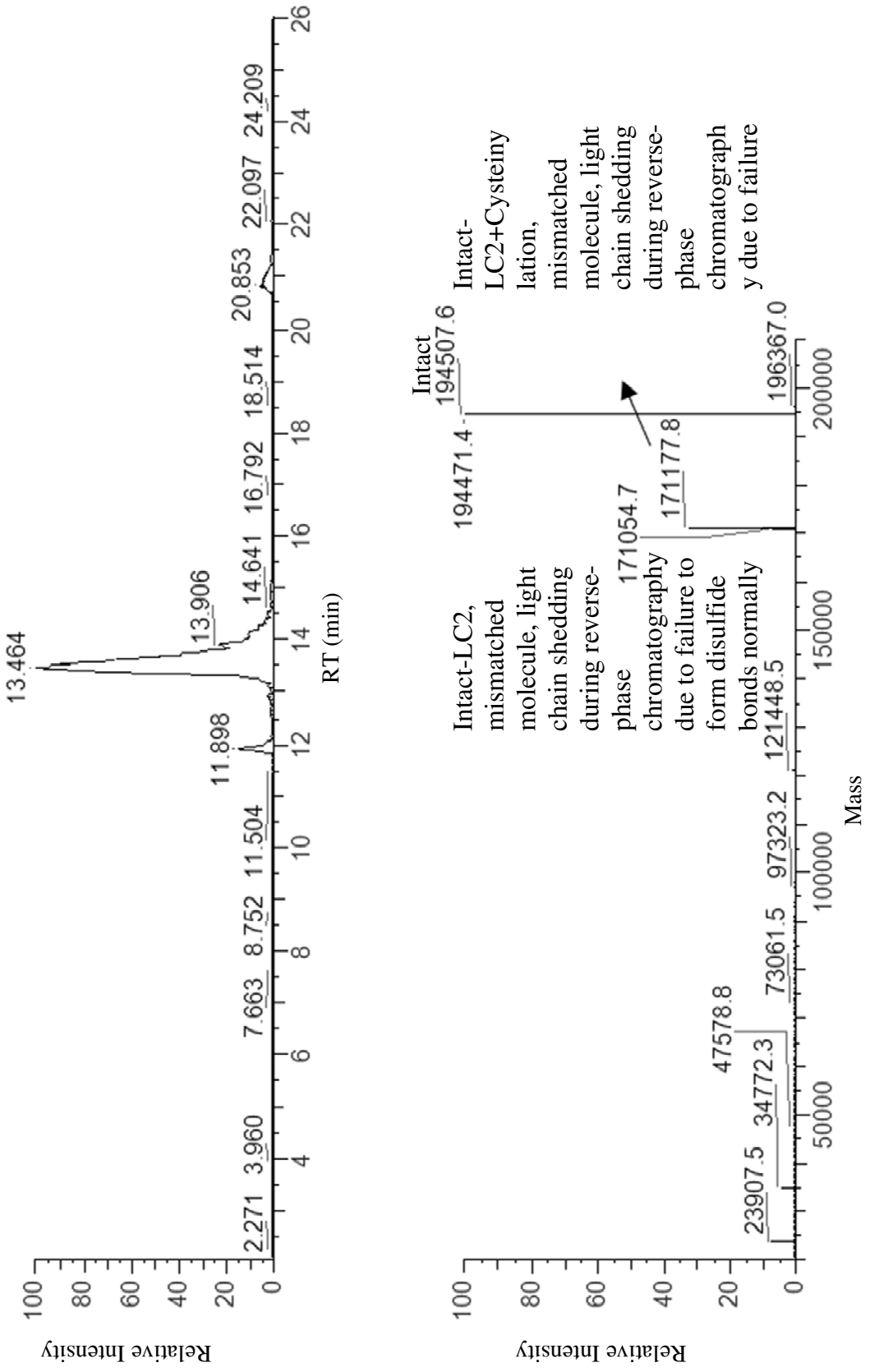

Relative Intensity
RT (min)
13.464
13.906
11.898
14.641
11.504
16.792
8.752
18.514
7.663
20.853
3.960
22.097
2.271
24.209
Intact
194507.6
Intact-LC2+Cysteinylation, mismatched molecule, light chain shedding during reverse-phase chromatography y due to failure
Intact-LC2, mismatched molecule, light chain shedding during reverse-phase chromatography due to failure to form disulfide bonds normally
194471.4
171054.7
171177.8
196367.0
121448.5
97323.2
73061.5
47578.8
34772.3
23907.5
Mass

Figure 6C

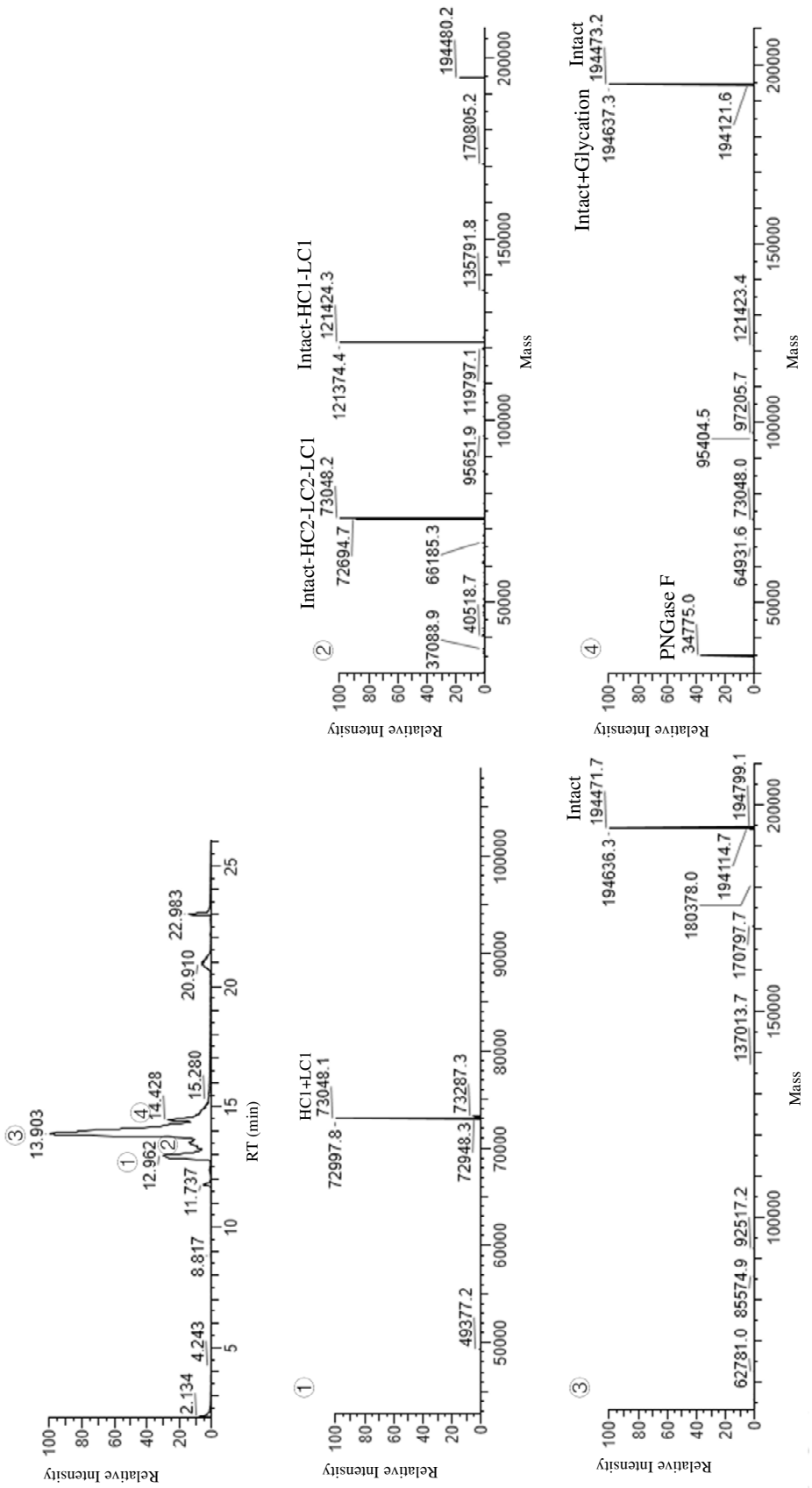

Relative Intensity
RT (min)
Mass
HC1+LC1
Intact-HC2-LC2-LC1
Intact-HC1-LC1
PNGase F
Intact+Glycation
Intact

Figure 6D

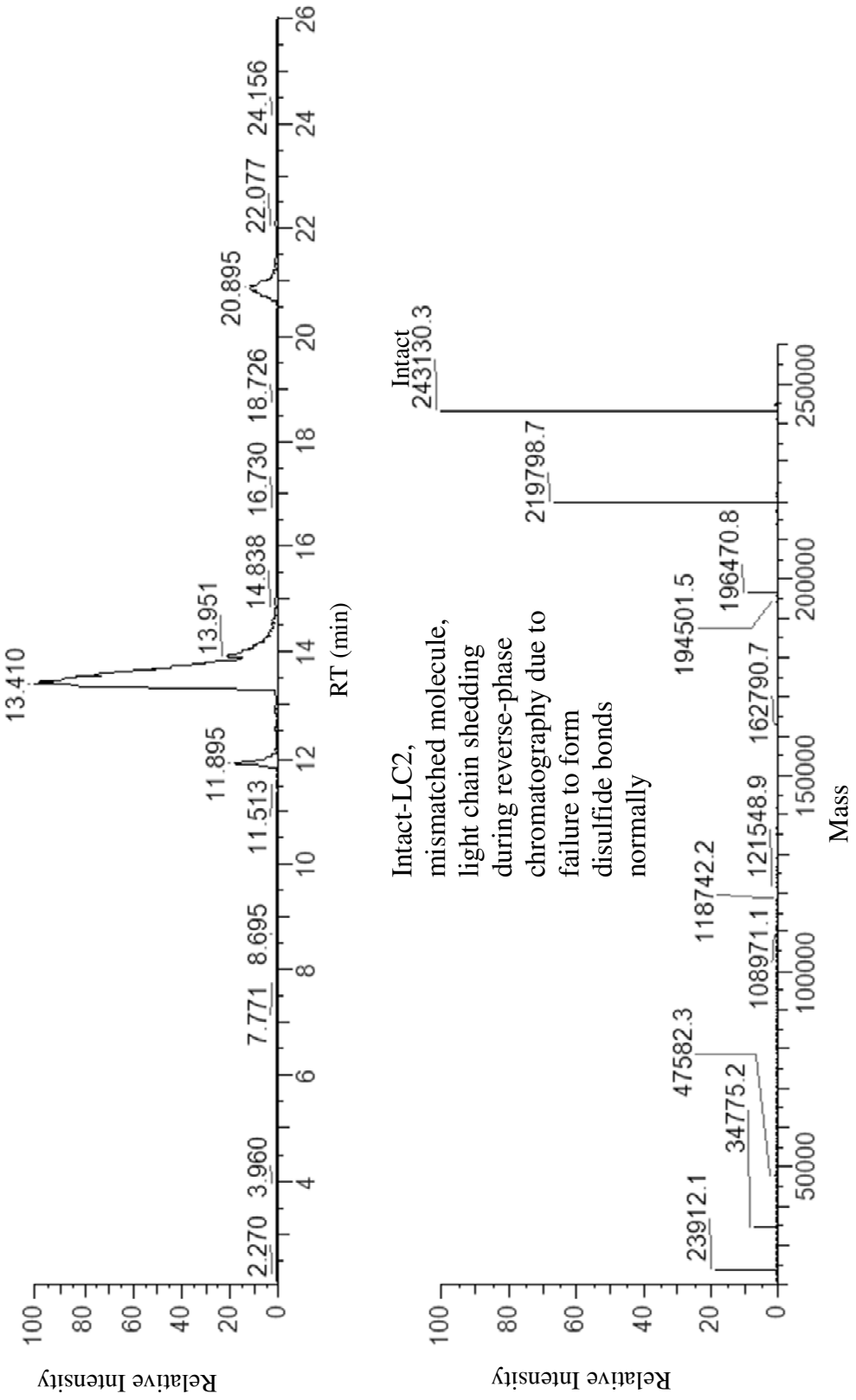

Relative Intensity
RT (min)
13.410
13.951
11.895
11.513
14.838
16.730
18.726
20.895
22.077
24.156
2.270
3.960
7.771
8.695
Intact-LC2,
mismatched molecule,
light chain shedding
during reverse-phase
chromatography due to
failure to form
disulfide bonds
normally
Intact
243130.3
219798.7
194501.5
196470.8
118742.2
121548.9
108971.1
162790.7
47582.3
34775.2
23912.1
Mass

Figure 6E

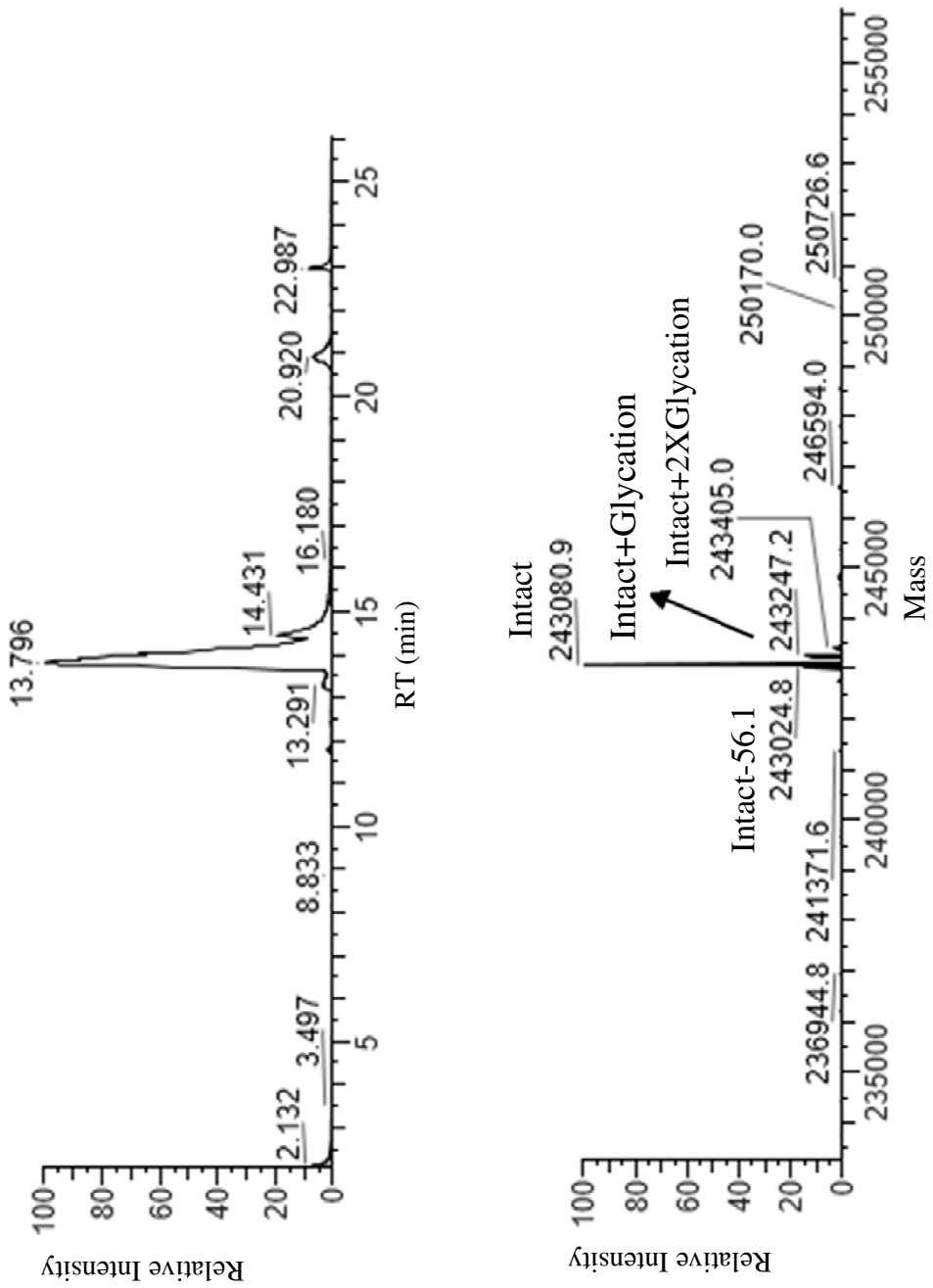

Relative Intensity
RT (min)
13.796
14.431
16.180
20.920
22.987
13.291
8.833
3.497
2.132
Intact
243080.9
Intact+Glycation
Intact+2XGlycation
243405.0
250170.0
250726.6
246594.0
243247.2
243024.8
Intact-56.1
241371.6
236944.8
Mass

Figure 6F

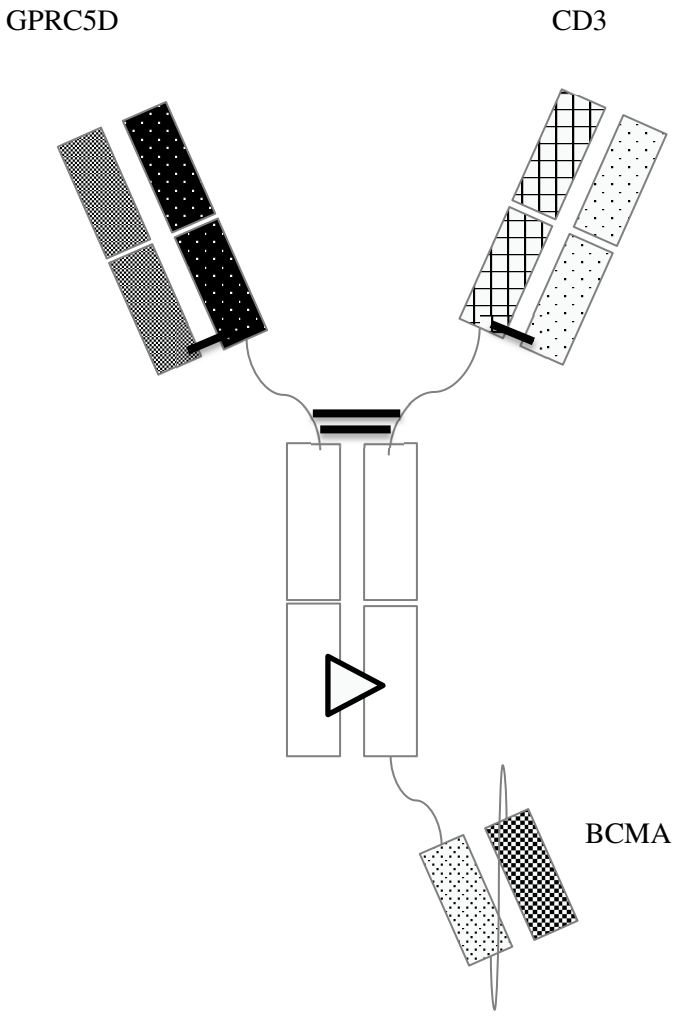


Figure 7A BCMA/GPRC5D/CD3

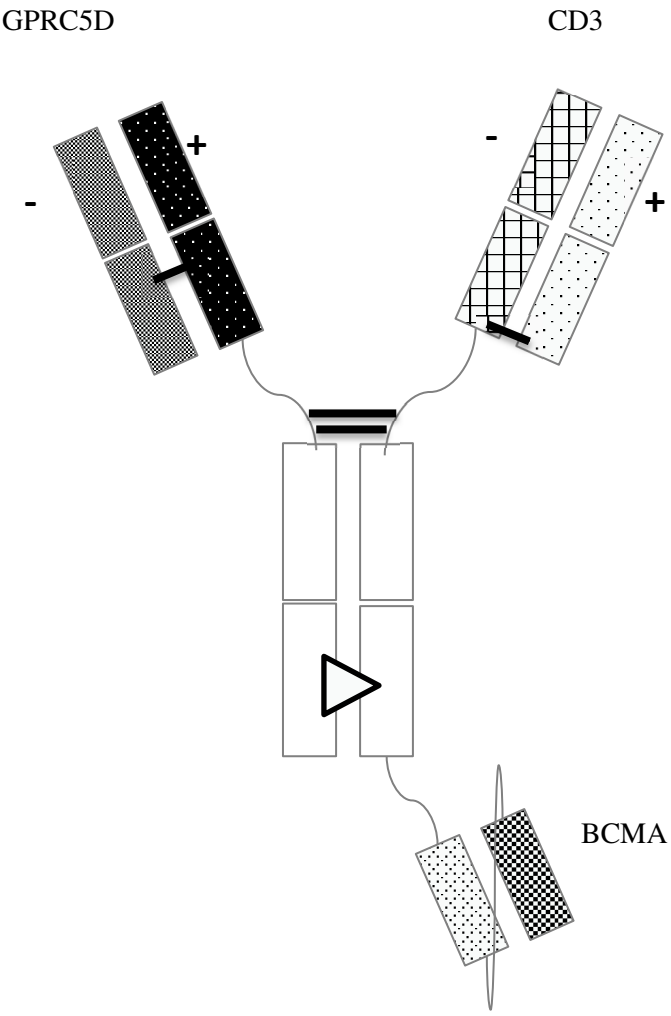


Figure 7B BCMA/GPRC5D/CD3 with Charge and cysteine mutation

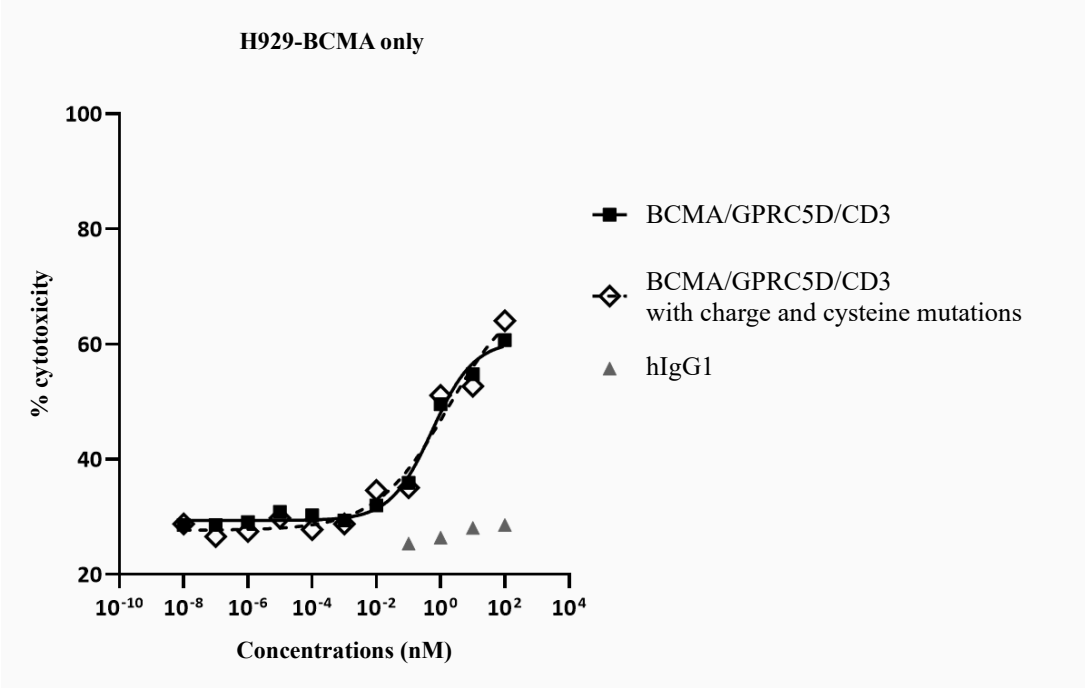


Figure 8A

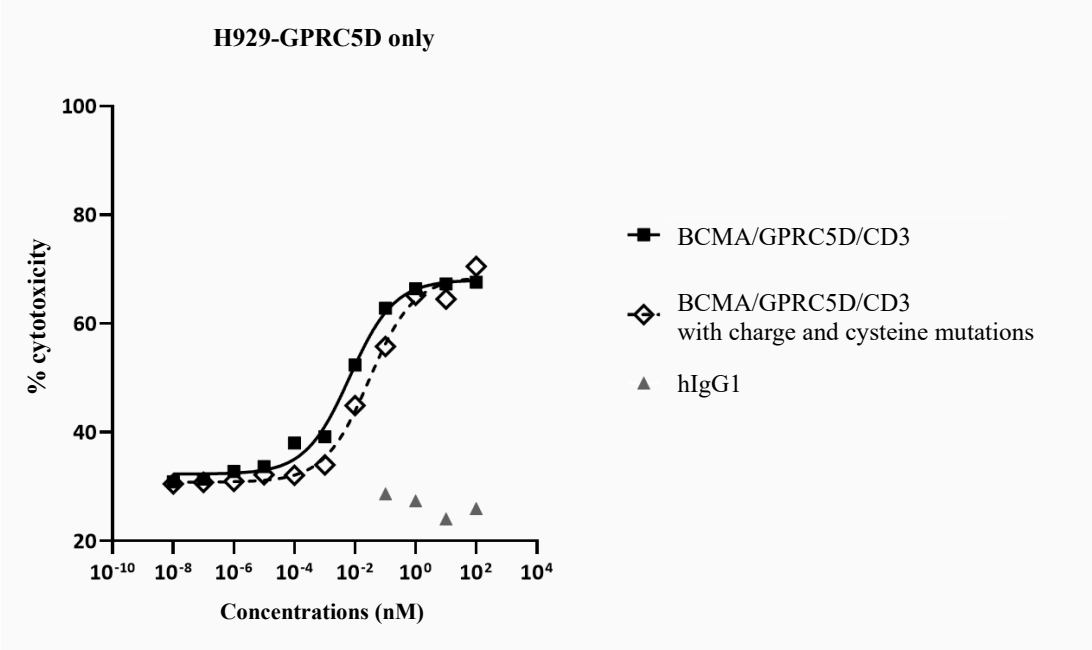
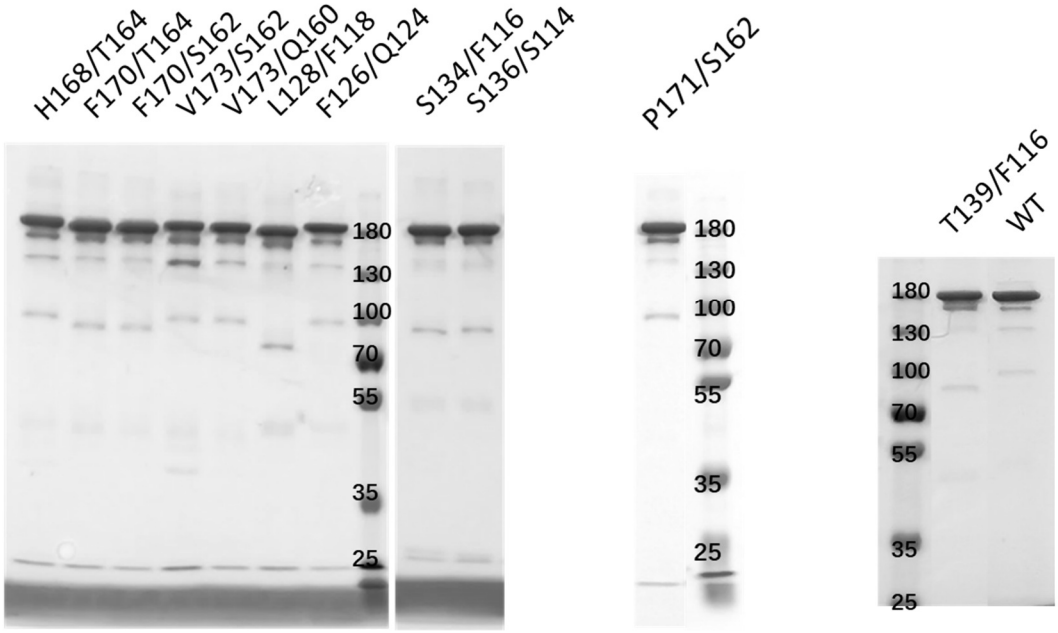


Figure 8B



Figures 9A 9B 9C

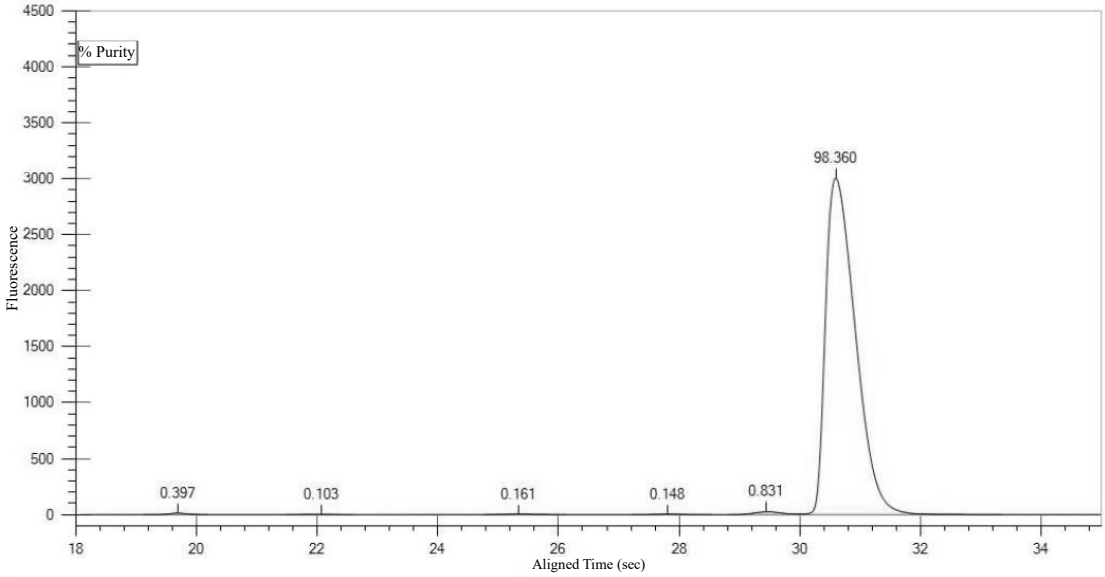


Figure 9D F126C/Q124C Disulfide Bond Reshaped Antibody

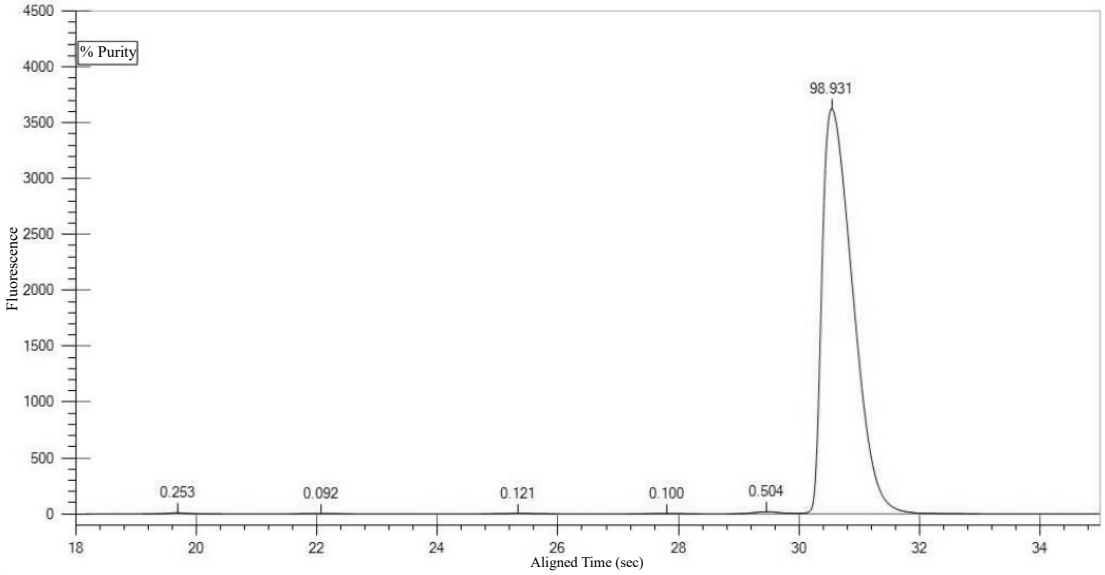


Figure 9E H168C/T164C Disulfide Bond Reshaped Antibody

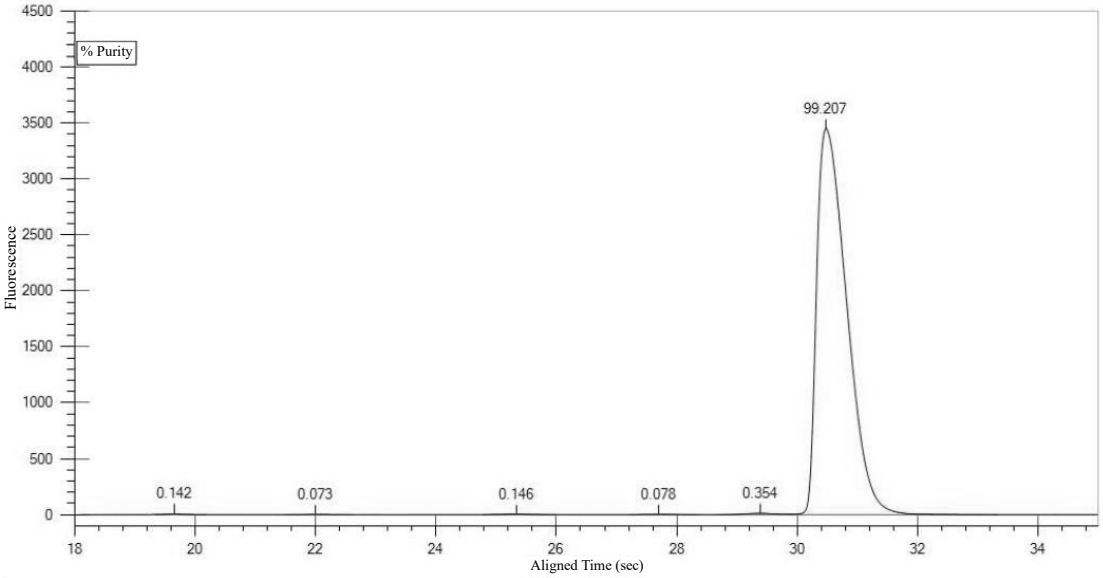


Figure 9F F170C/T164C Disulfide Bond Reshaped Antibody

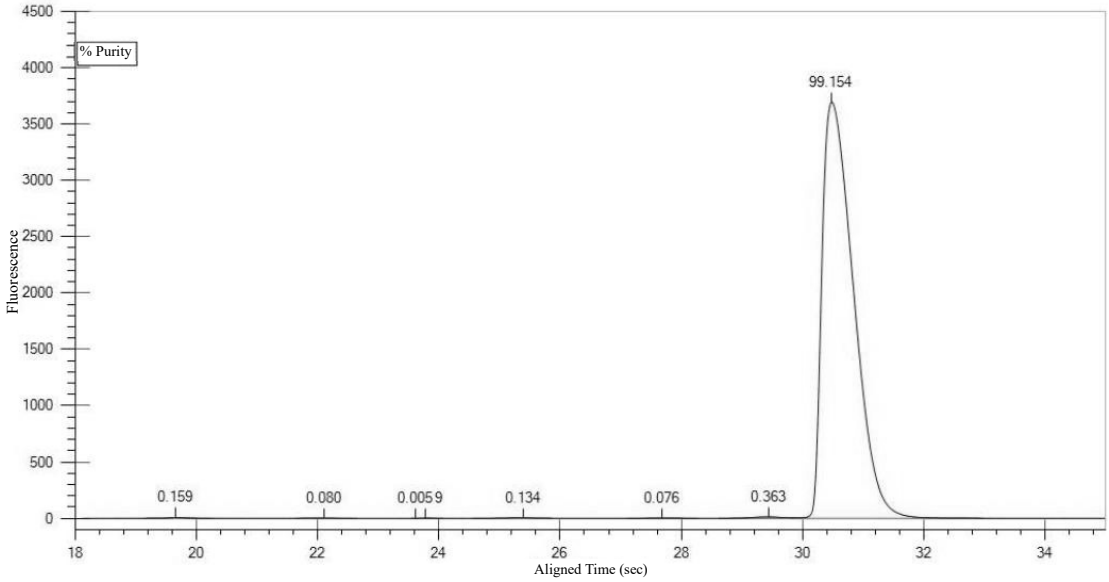


Figure 9G F170C/S162C Disulfide Bond Reshaped Antibody

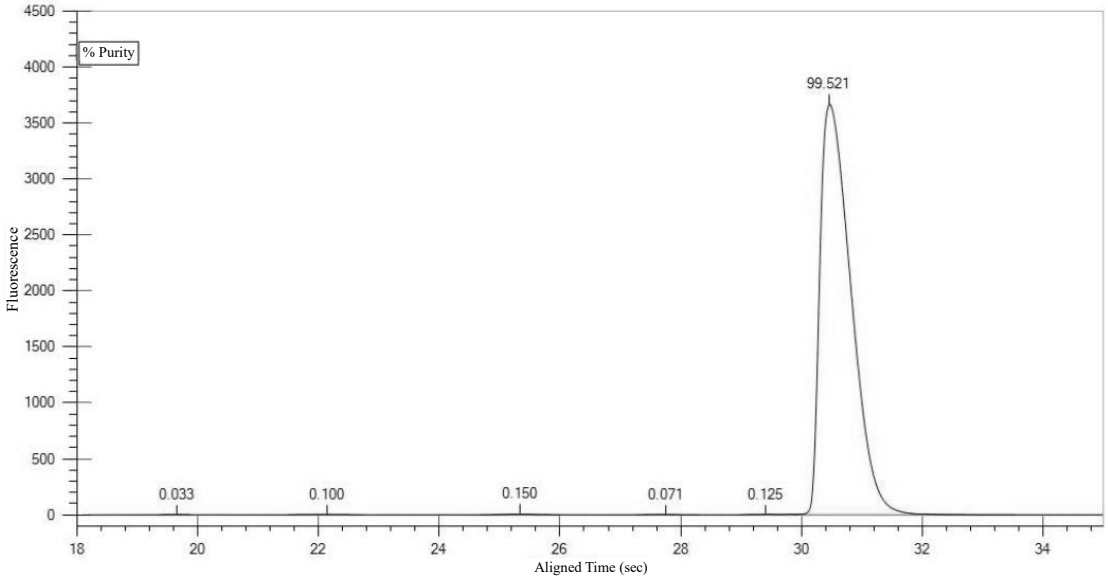


Figure 9H V173C/S162C Disulfide Bond Reshaped Antibody

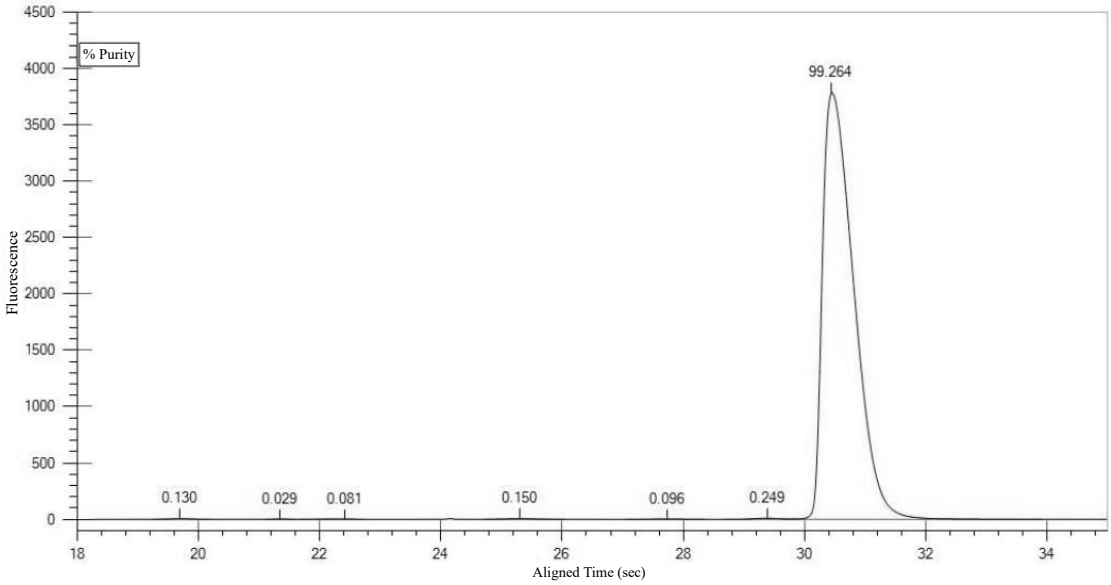


Figure 9I V173C/Q160C Disulfide Bond Reshaped Antibody

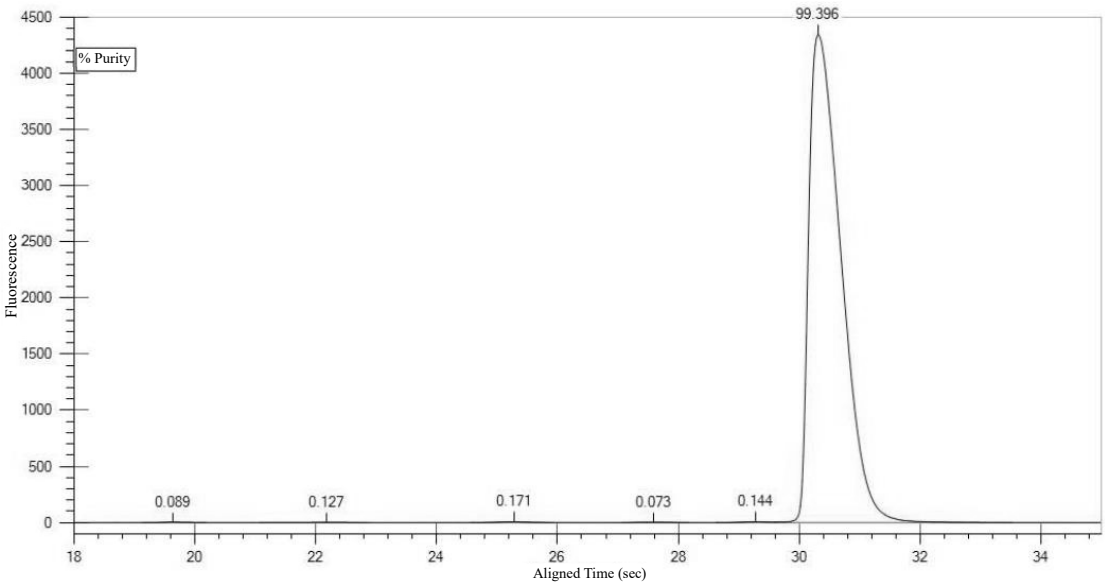


Figure 9J S134C/F116C Disulfide Bond Reshaped Antibody

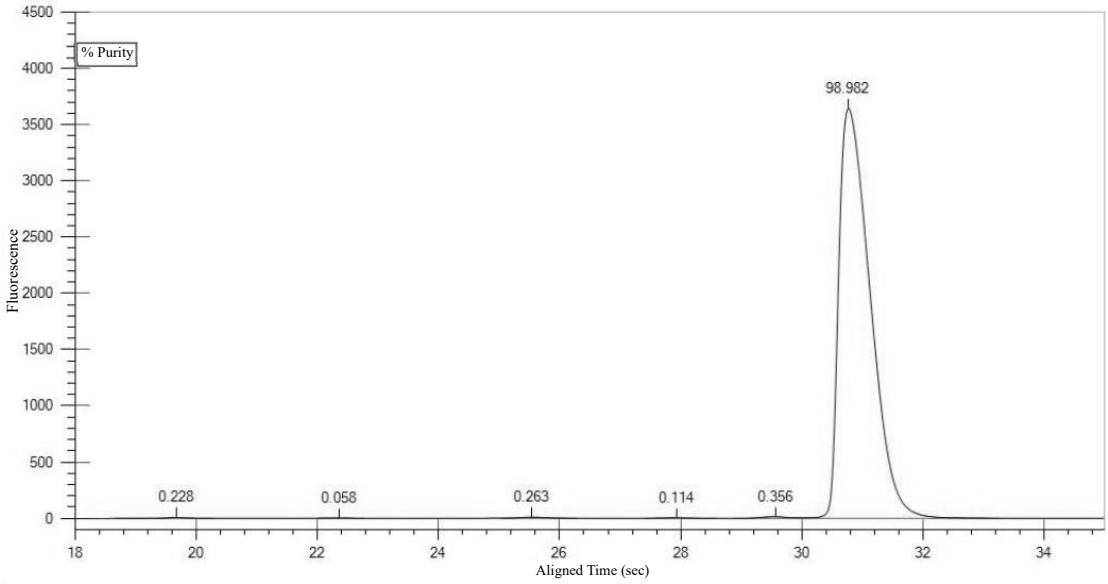


Figure 9K S136C/S114C Disulfide Bond Reshaped Antibody

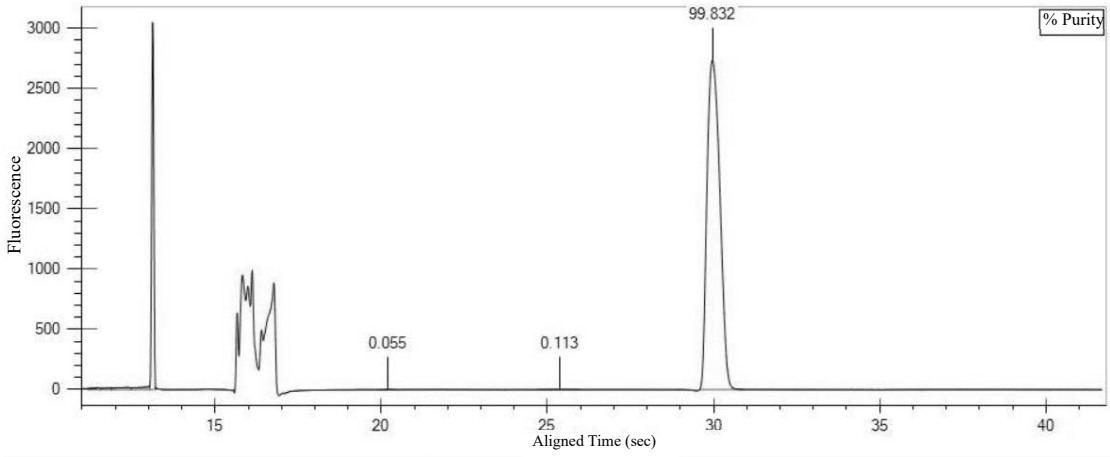


Figure 9L P171/S162 Disulfide Bond Reshaped Antibody

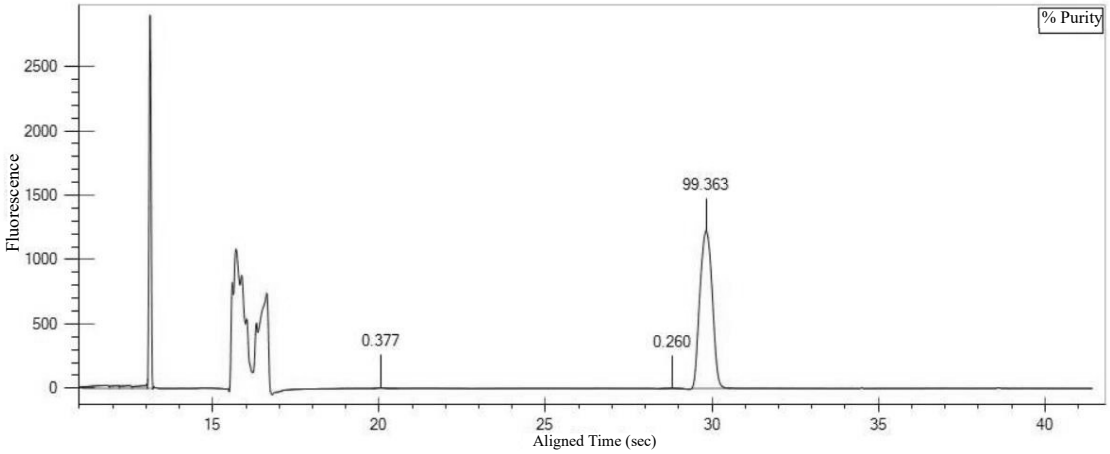


Figure 9M T139/F116 Disulfide Bond Reshaped Antibody