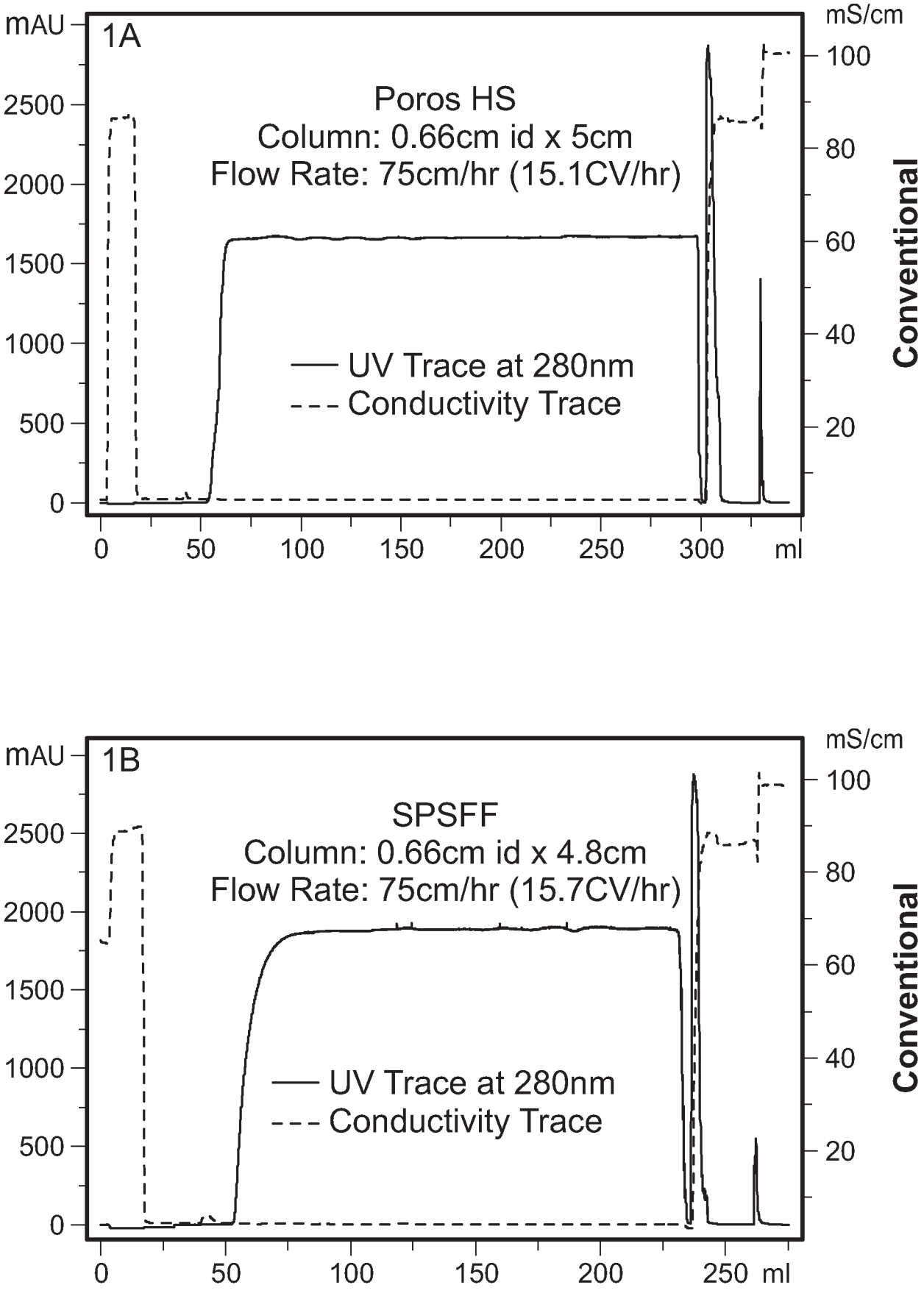
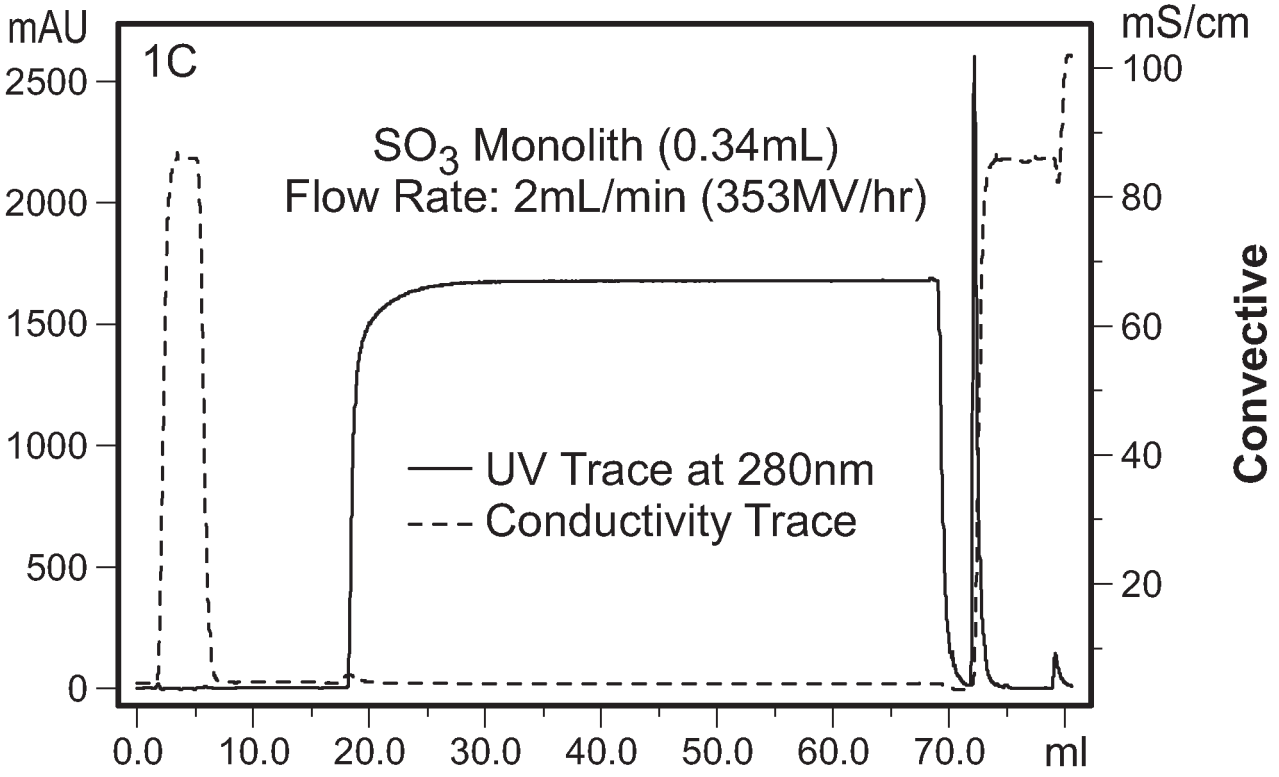

1A
Poros HS
Column: 0.66cm id x 5cm
Flow Rate: 75cm/hr (15.1CV/hr)
UV Trace at 280nm
Conductivity Trace
mAU
mS/cm
Conventional
ml
1B
SPSFF
Column: 0.66cm id x 4.8cm
Flow Rate: 75cm/hr (15.7CV/hr)
UV Trace at 280nm
Conductivity Trace
mAU
mS/cm
Conventional
ml

FIG. 1


mAU
mS/cm
1C
SO_3 Monolith (0.34mL)
Flow Rate: 2mL/min (353MV/hr)
UV Trace at 280nm
Conductivity Trace
Convective
2500
2000
1500
1000
500
0
100
80
60
40
20
0.0
10.0
20.0
30.0
40.0
50.0
60.0
70.0
ml

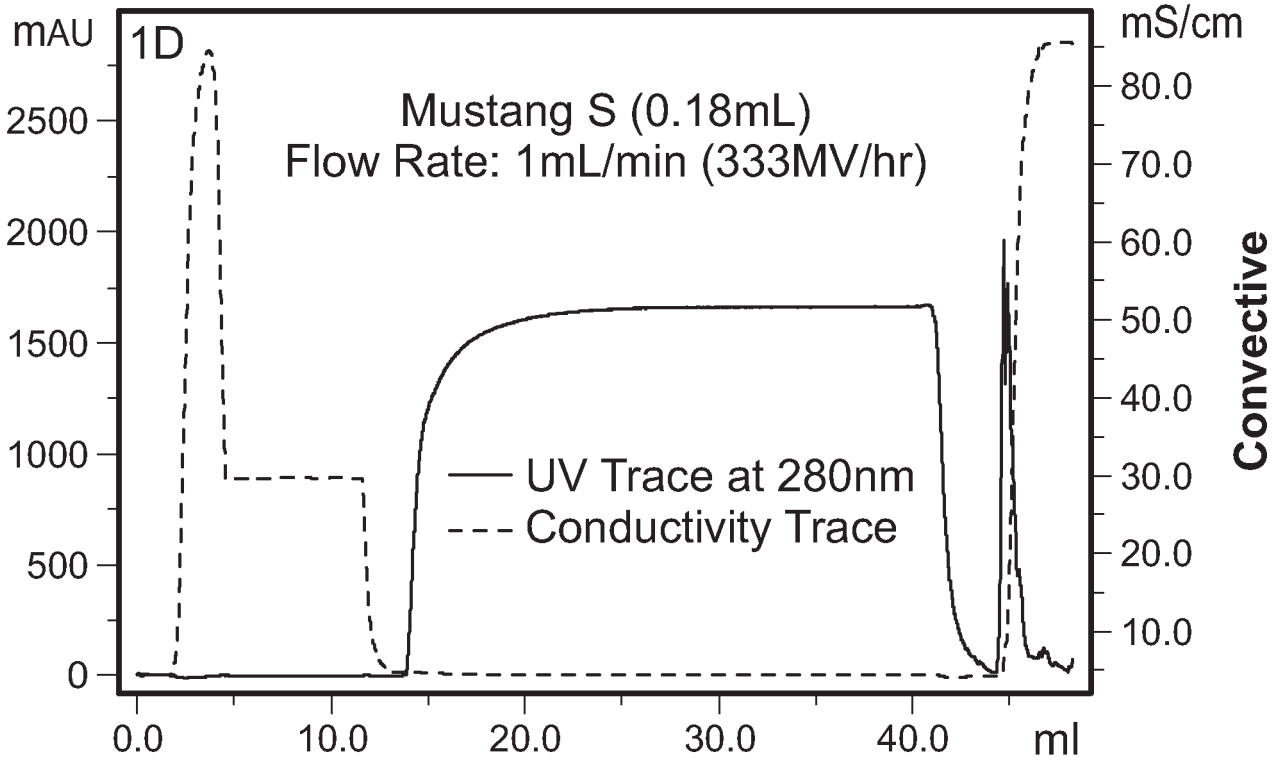

mAU
mS/cm
1D
Mustang S (0.18mL)
Flow Rate: 1mL/min (333MV/hr)
UV Trace at 280nm
Conductivity Trace
Convective
2500
2000
1500
1000
500
0
80.0
70.0
60.0
50.0
40.0
30.0
20.0
10.0
0.0
10.0
20.0
30.0
40.0
ml

FIG. 1 (Cont.)

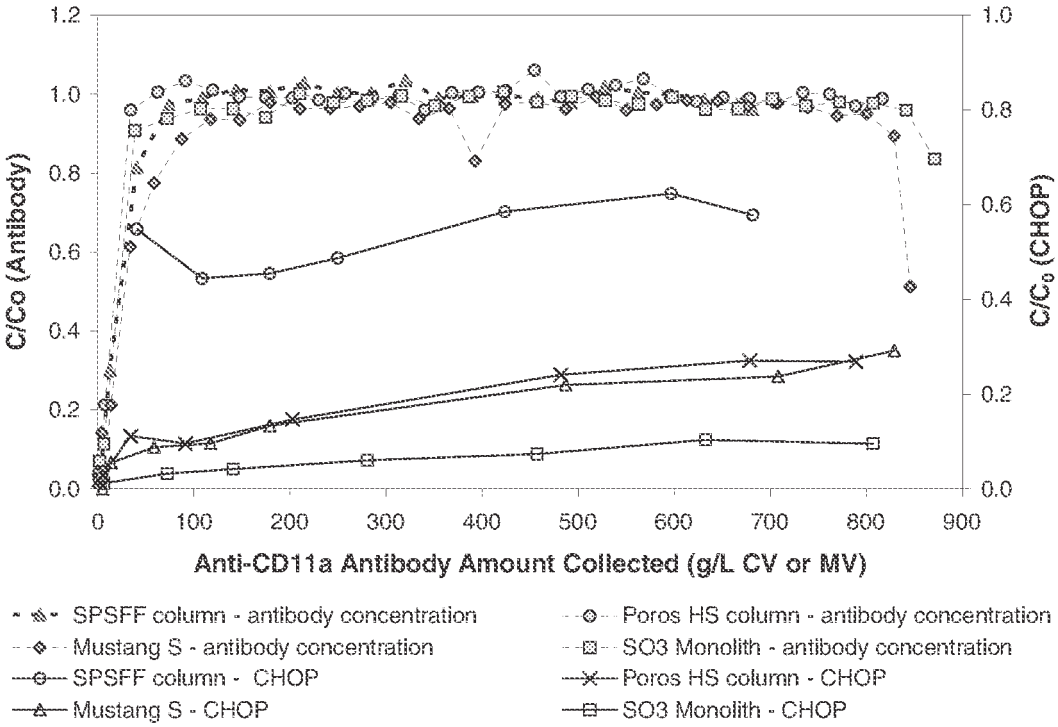

C/Co (Antibody)
C/C0 (CHOP)
Anti-CD11a Antibody Amount Collected (g/L CV or MV)
SPSFF column - antibody concentration
Mustang S - antibody concentration
SPSFF column - CHOP
Mustang S - CHOP
Poros HS column - antibody concentration
SO3 Monolith - antibody concentration
Poros HS column - CHOP
SO3 Monolith - CHOP

FIG. 2

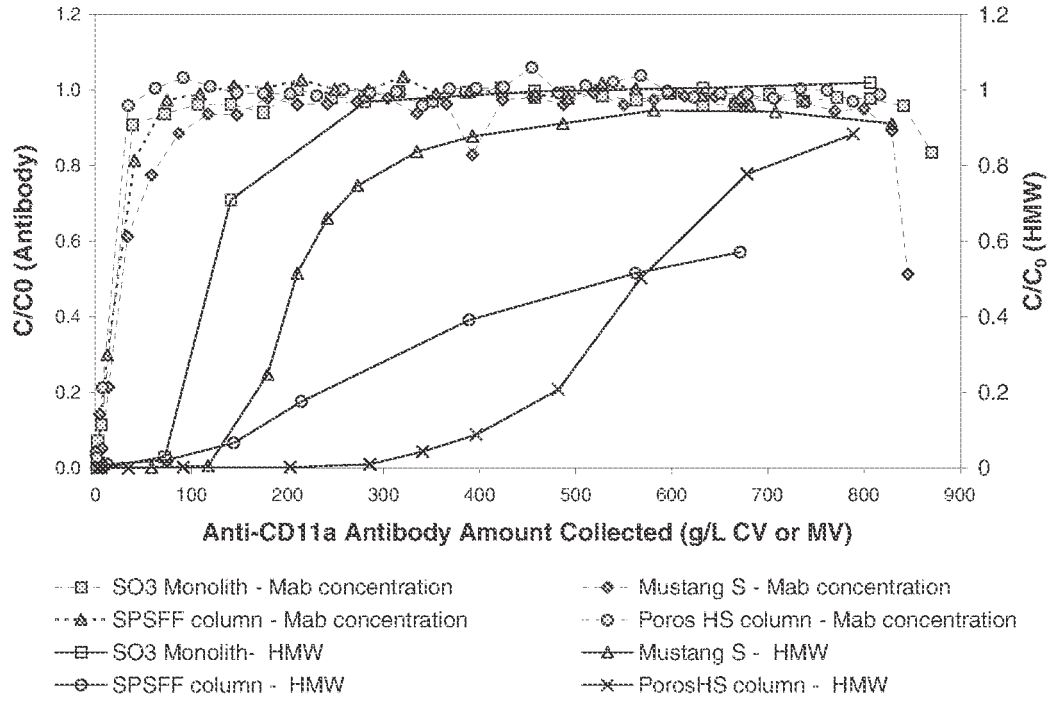
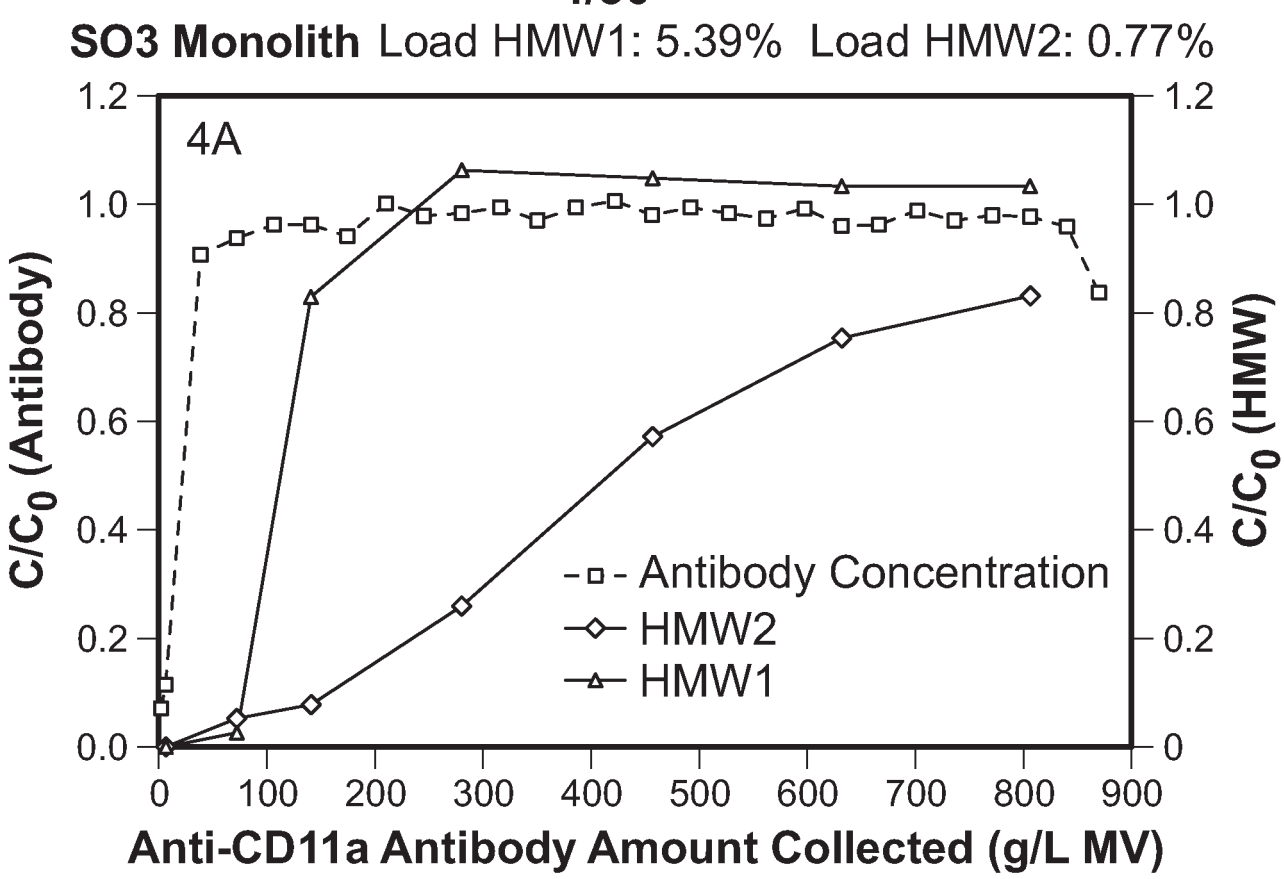

C/C0 (Antibody)
C/C0 (HMW)
Anti-CD11a Antibody Amount Collected (g/L CV or MV)
SO3 Monolith - Mab concentration
SPSFF column - Mab concentration
SO3 Monolith- HMW
SPSFF column - HMW
Mustang S - Mab concentration
Poros HS column - Mab concentration
Mustang S - HMW
PorosHS column - HMW

FIG.3


SO3 Monolith Load HMW1: 5.39% Load HMW2: 0.77%
4A
C/C0 (Antibody)
C/C0 (HMW)
Antibody Concentration
HMW2
HMW1
Anti-CD11a Antibody Amount Collected (g/L MV)

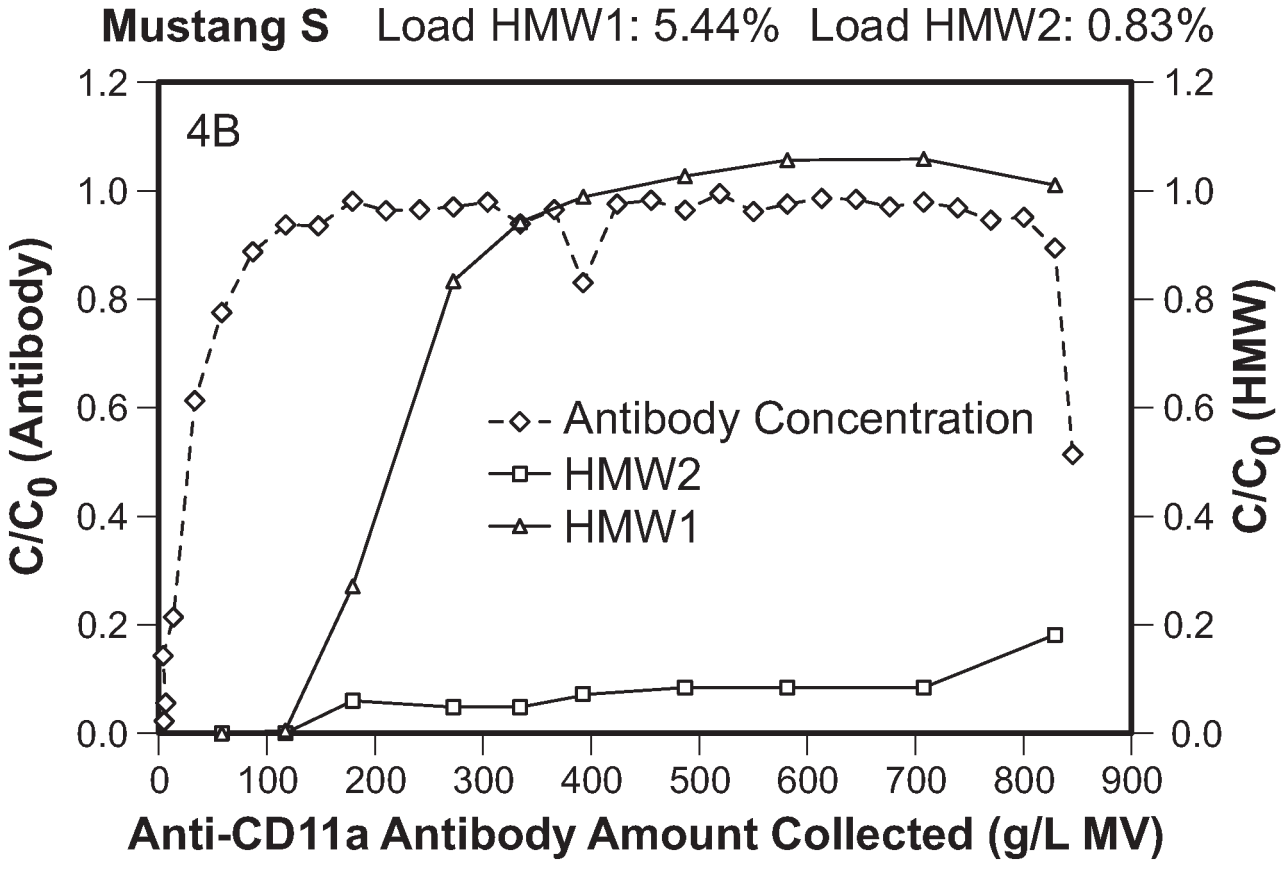
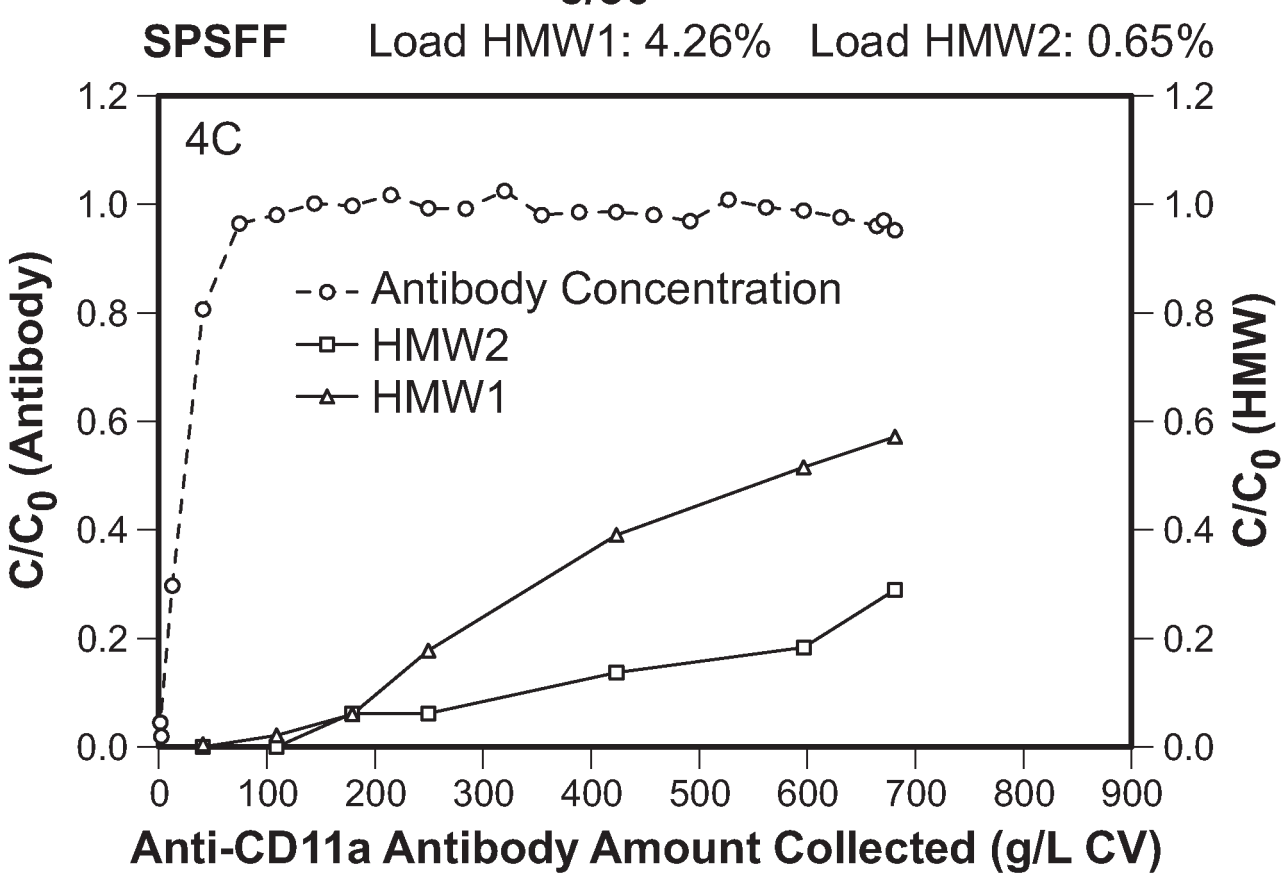

Mustang S Load HMW1: 5.44% Load HMW2: 0.83%
4B
C/C0 (Antibody)
C/C0 (HMW)
Antibody Concentration
HMW2
HMW1
Anti-CD11a Antibody Amount Collected (g/L MV)

FIG. 4


SPSFF Load HMW1: 4.26% Load HMW2: 0.65%
4C
Antibody Concentration
HMW2
HMW1
C/C0 (Antibody)
C/C0 (HMW)
Anti-CD11a Antibody Amount Collected (g/L CV)

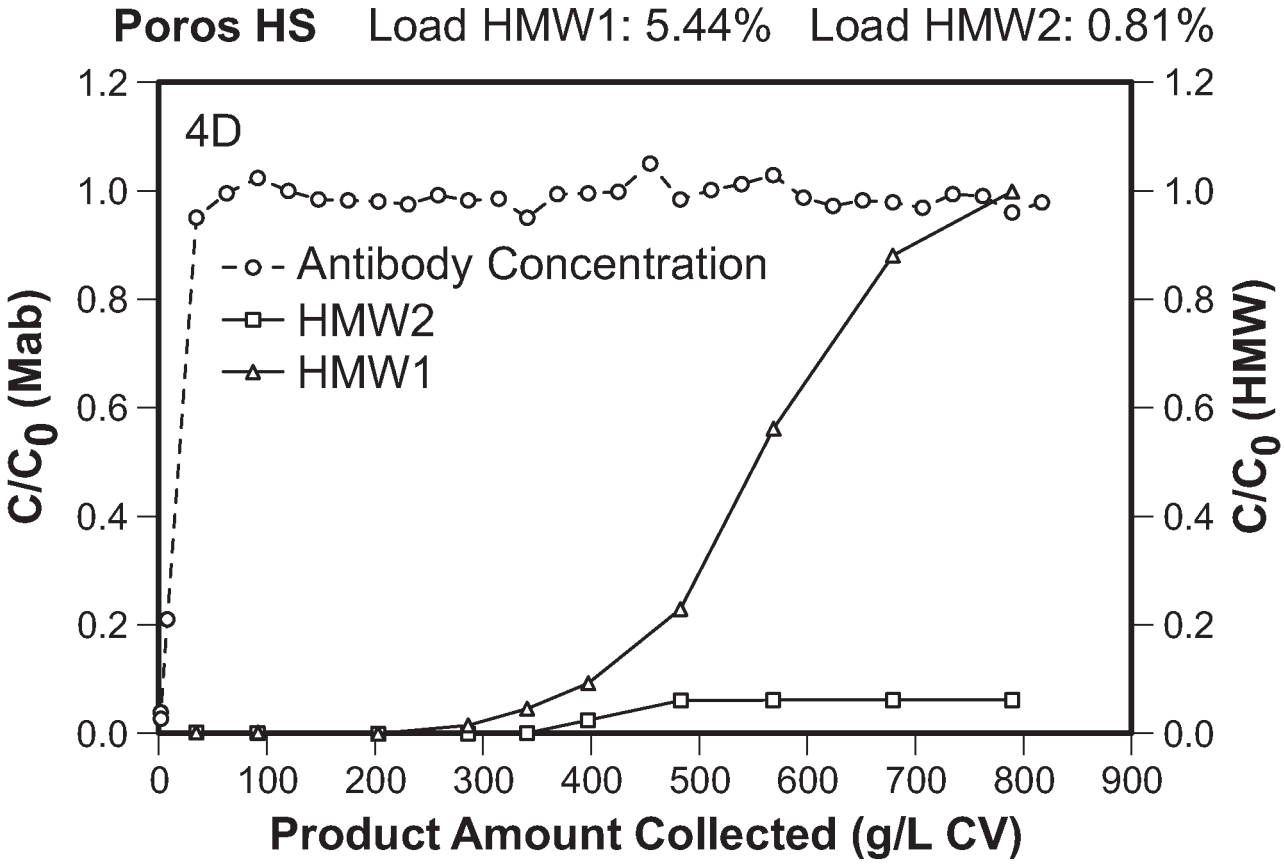
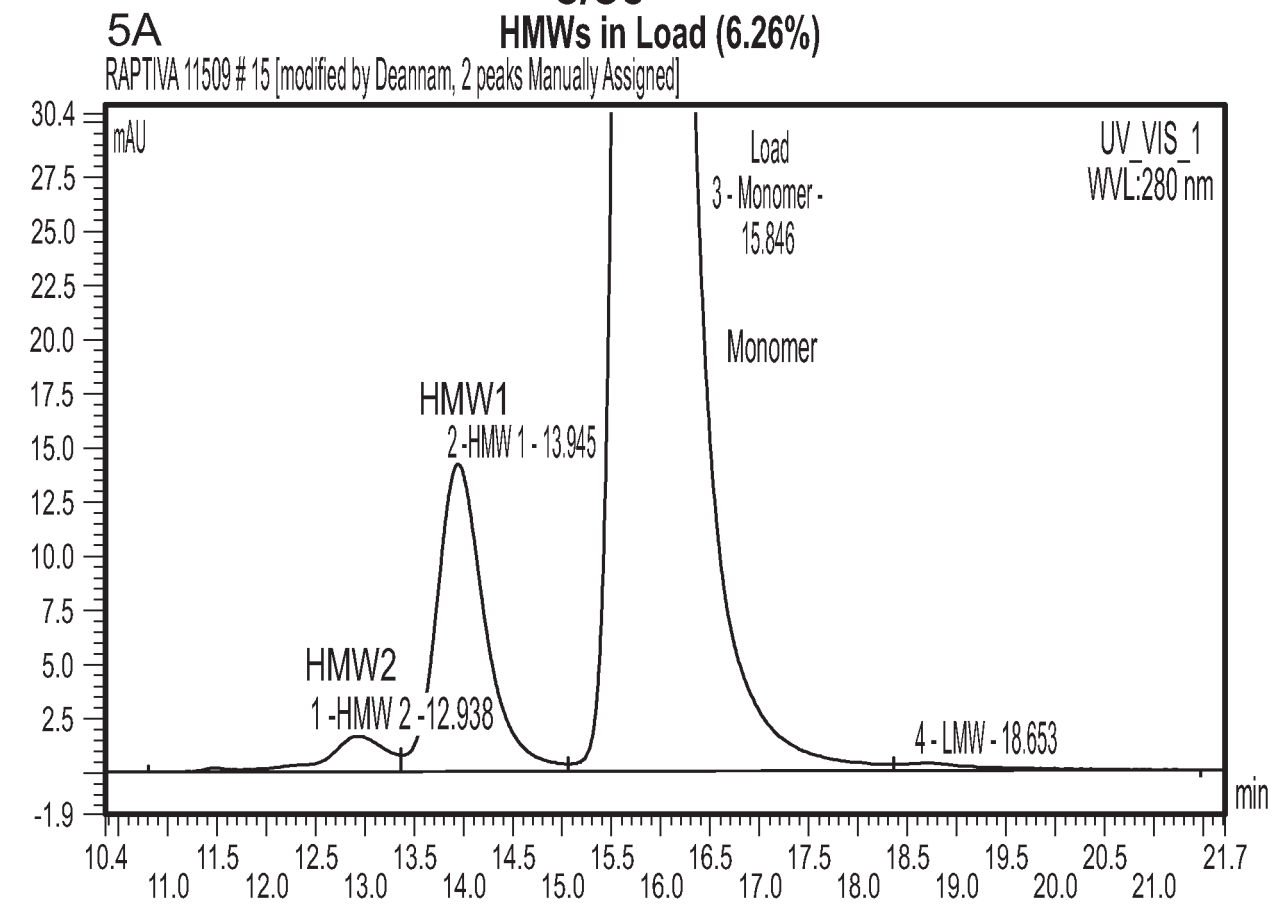

Poros HS Load HMW1: 5.44% Load HMW2: 0.81%
4D
Antibody Concentration
HMW2
HMW1
C/C0 (Mab)
C/C0 (HMW)
Product Amount Collected (g/L CV)

FIG. 4 (Cont.)


5A
HMWs in Load (6.26%)
RAPTIVA 11509 # 15 [modified by Deannam, 2 peaks Manually Assigned]
mAU
UV_VIS_1
WVL:280 nm
Load
3 - Monomer - 15.846
Monomer
HMW1
2 - HMW 1 - 13.945
HMW2
1 - HMW 2 - 12.938
4 - LMW - 18.653
min
30.4
27.5
25.0
22.5
20.0
17.5
15.0
12.5
10.0
7.5
5.0
2.5
-1.9
10.4
11.0
11.5
12.0
12.5
13.0
13.5
14.0
14.5
15.0
15.5
16.0
16.5
17.0
17.5
18.0
18.5
19.0
19.5
20.0
20.5
21.0
21.7

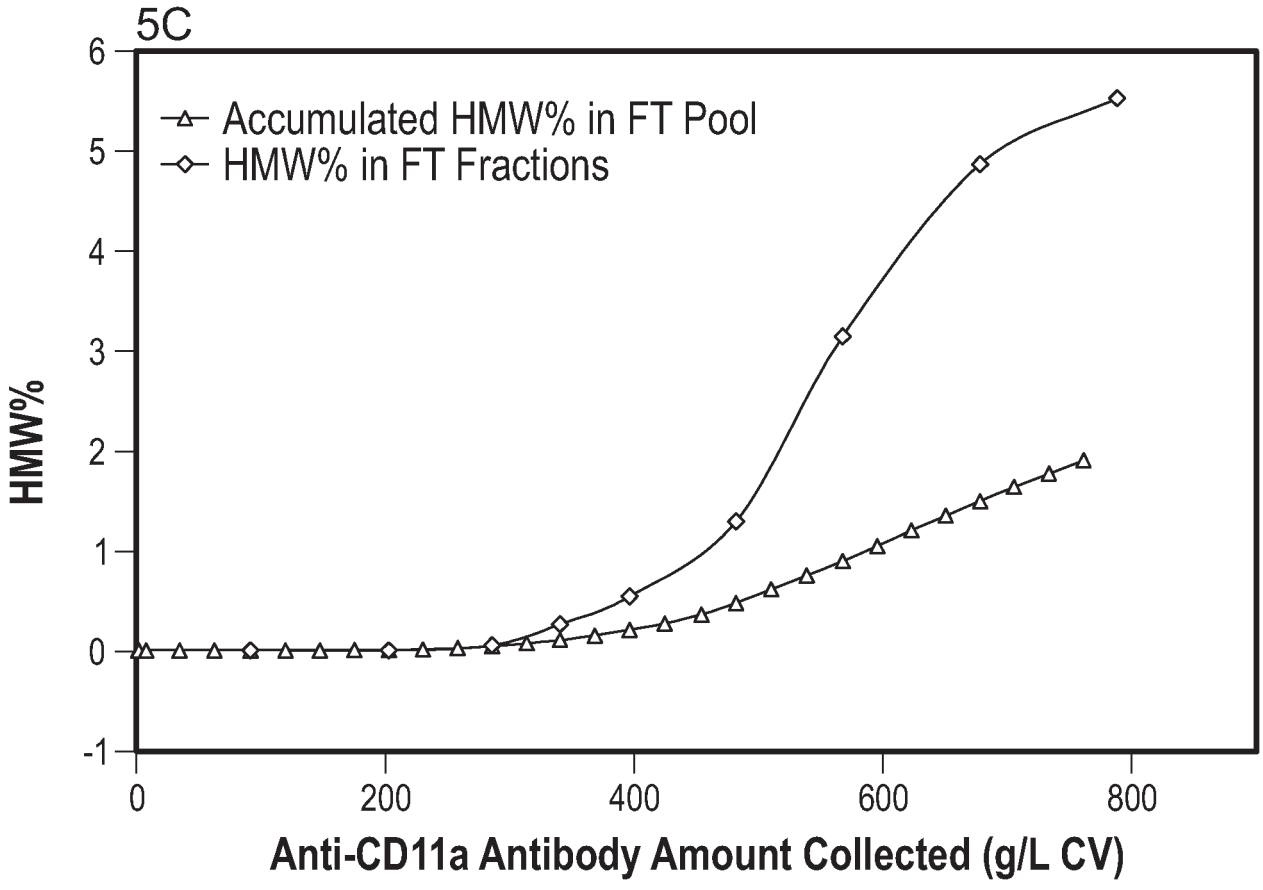

5C
Accumulated HMW% in FT Pool
HMW% in FT Fractions
HMW%
6
5
4
3
2
1
0
-1
0
200
400
600
800
Anti-CD11a Antibody Amount Collected (g/L CV)

FIG. 5

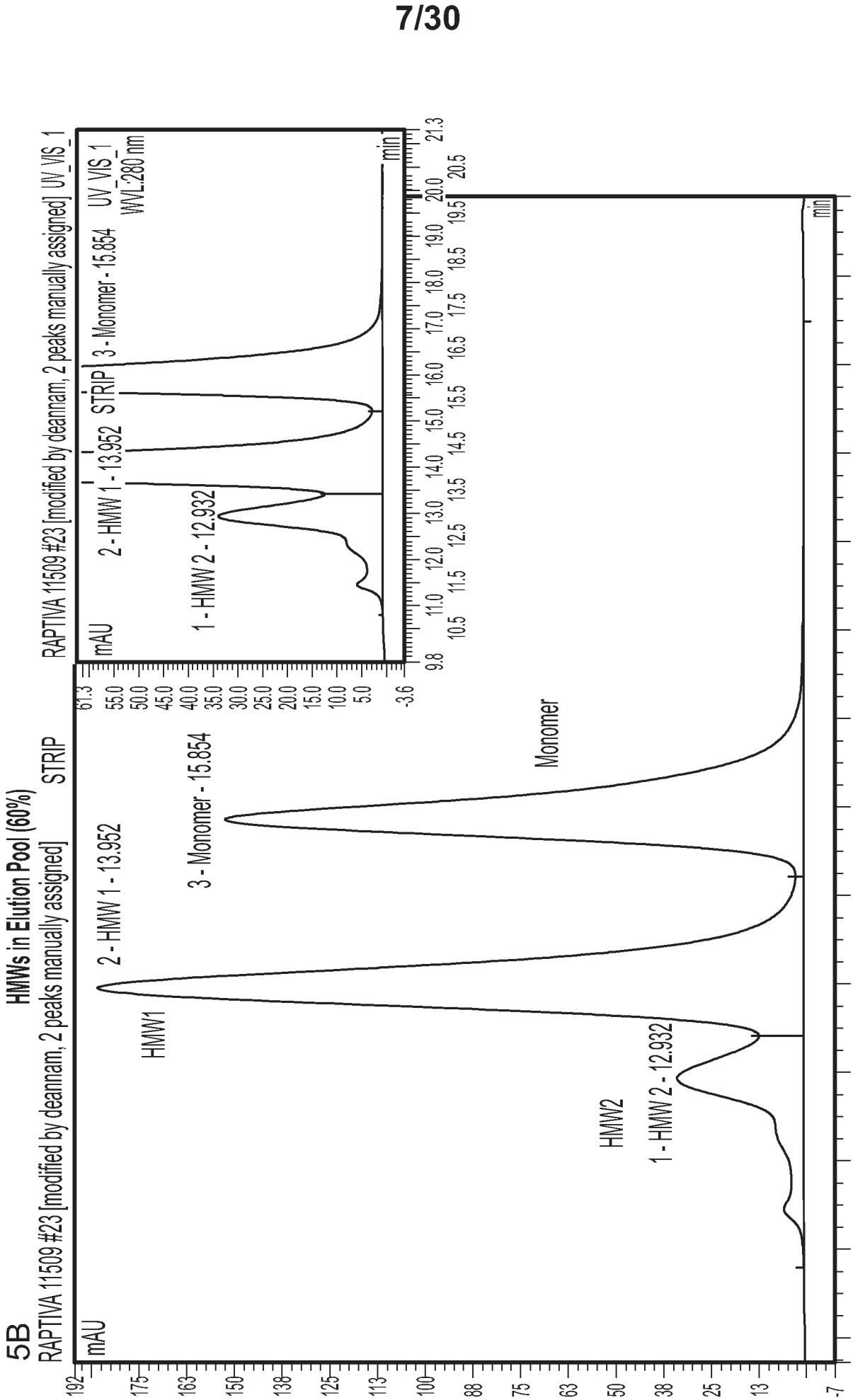

5B
HMWs in Elution Pool (60%)
RAPTIVA 11509 #23 [modified by deannam, 2 peaks manually assigned] STRIP
mAU
2 - HMW 1 - 13.952
3 - Monomer - 15.854
1 - HMW 2 - 12.932
HMW1
HMW2
Monomer
min
RAPTIVA 11509 #23 [modified by deannam, 2 peaks manually assigned] UV_VIS_1
mAU
2 - HMW 1 - 13.952
STRIP
3 - Monomer - 15.854
UV_VIS_1
WVL:280 nm
1 - HMW 2 - 12.932
min

FIG. 5 (Cont.)

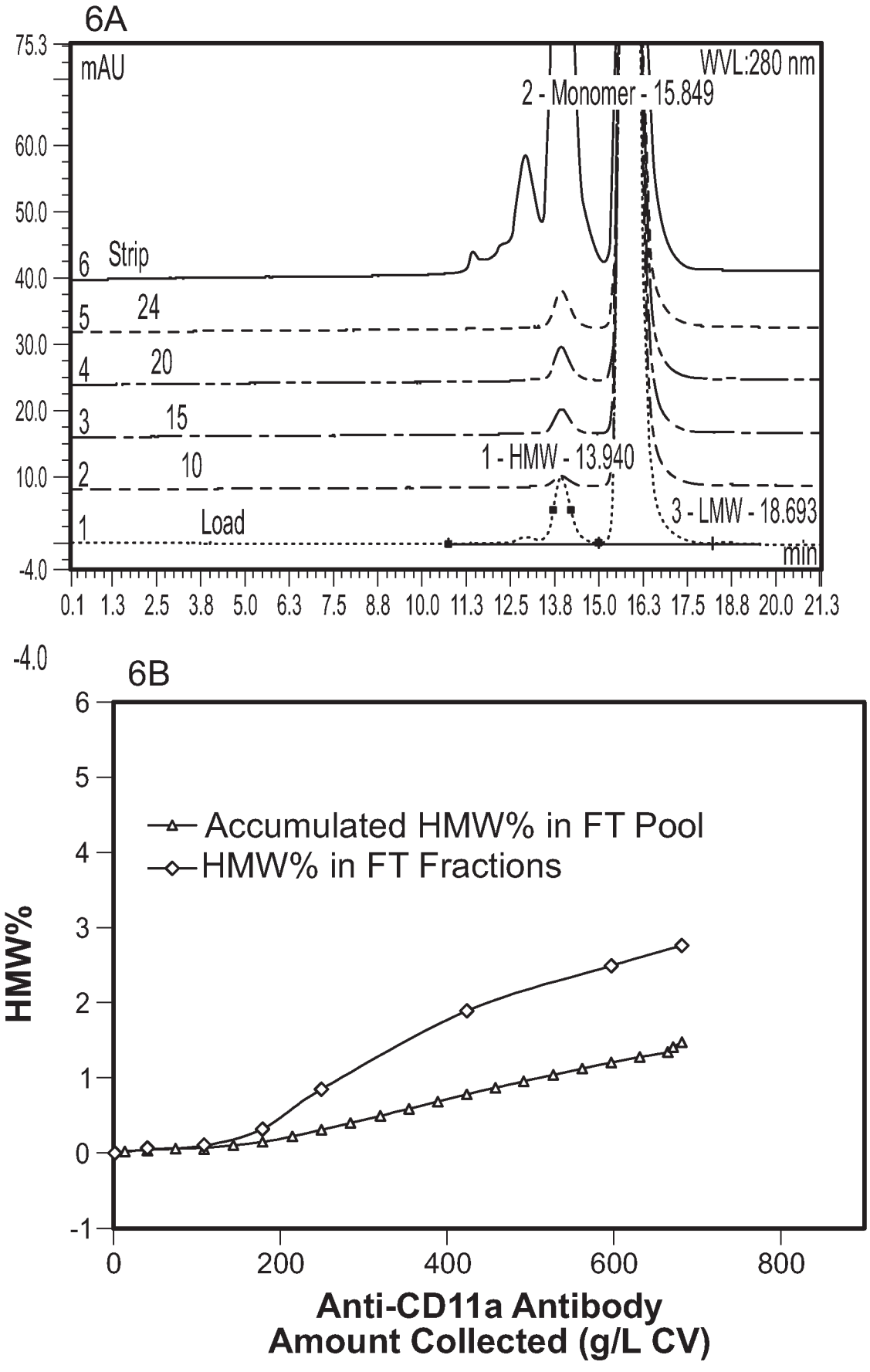

6A
mAU
WVL:280 nm
2 - Monomer - 15.849
1 - HMW - 13.940
3 - LMW - 18.693
Strip
24
20
15
10
Load
min
75.3
60.0
50.0
40.0
30.0
20.0
10.0
-4.0
0.1 1.3 2.5 3.8 5.0 6.3 7.5 8.8 10.0 11.3 12.5 13.8 15.0 16.3 17.5 18.8 20.0 21.3
6B
Accumulated HMW% in FT Pool
HMW% in FT Fractions
HMW%
Anti-CD11a Antibody
Amount Collected (g/L CV)
0
200
400
600
800

FIG. 6

6C

Sample ID	Anti-CD11a Collected (g/L CV)	HMW%
Load	-	4.83
2	2.20	n.a.
4	40.9	0.05
6	109	0.10
8	179	0.32
10	250	0.85
15	424	1.89
20	597	2.49
24	681	2.76
Strip	-	35.3

FIG. 6 (Cont.)

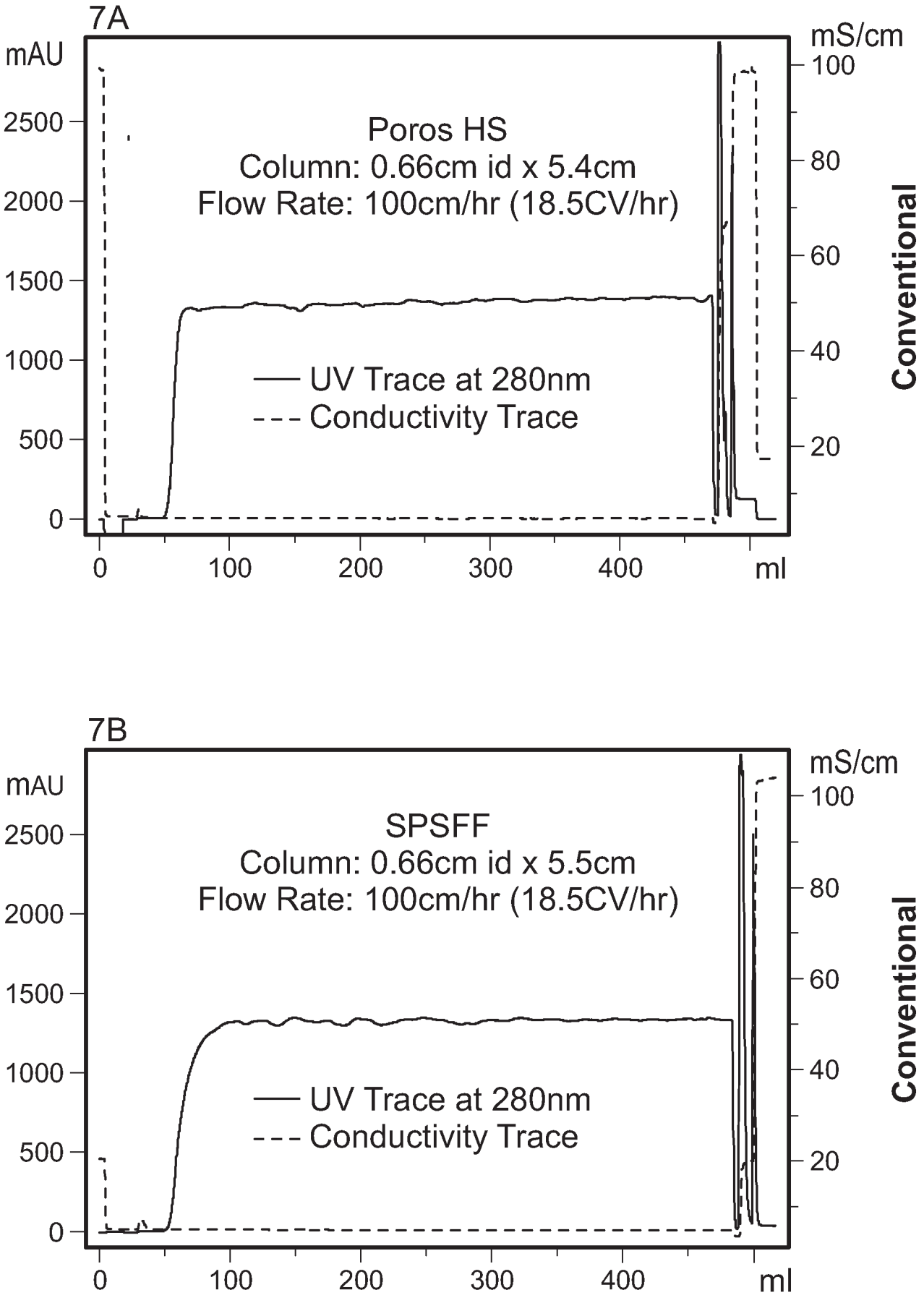
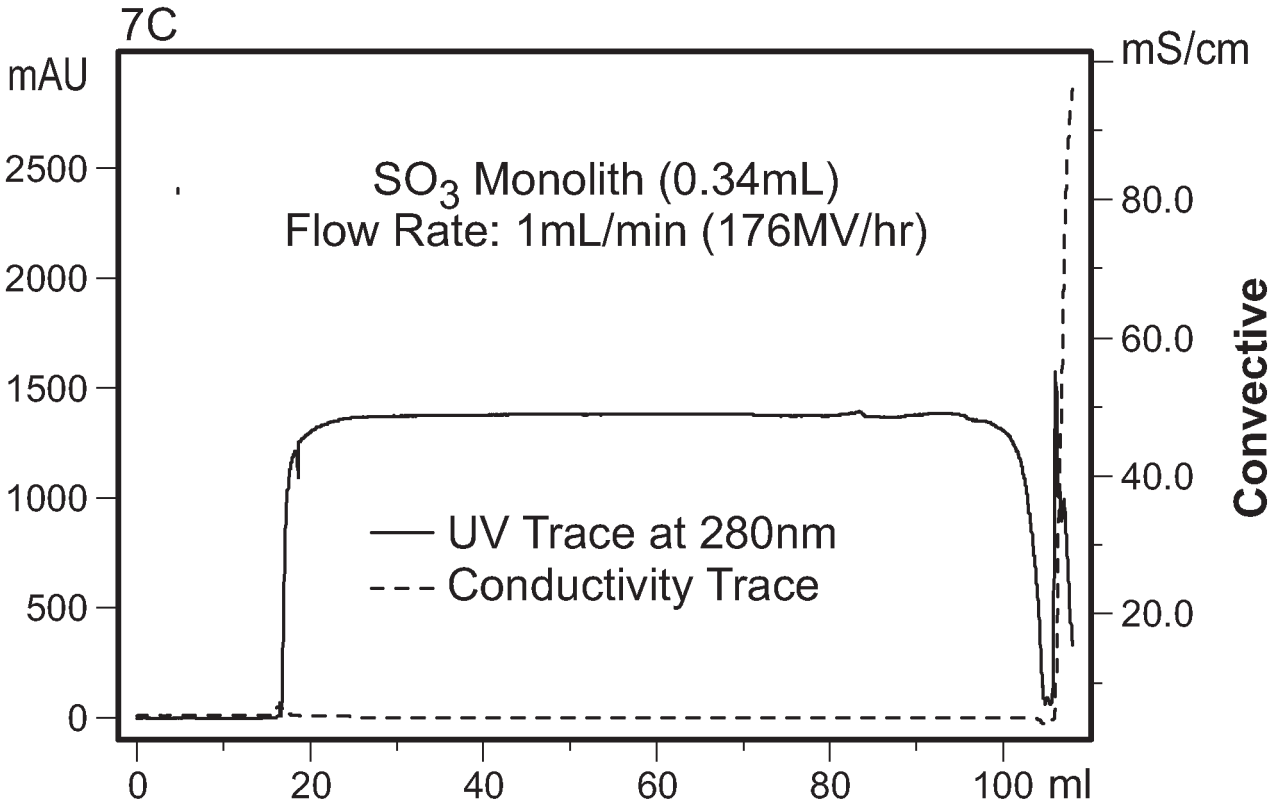

7A
mAU
2500
2000
1500
1000
500
0
mS/cm
100
80
60
40
20
Conventional
Poros HS
Column: 0.66cm id x 5.4cm
Flow Rate: 100cm/hr (18.5CV/hr)
UV Trace at 280nm
Conductivity Trace
0
100
200
300
400
ml
7B
mAU
2500
2000
1500
1000
500
0
mS/cm
100
80
60
40
20
Conventional
SPSFF
Column: 0.66cm id x 5.5cm
Flow Rate: 100cm/hr (18.5CV/hr)
UV Trace at 280nm
Conductivity Trace
0
100
200
300
400
ml

FIG. 7


7C
mAU
2500
2000
1500
1000
500
0
SO_3 Monolith (0.34mL)
Flow Rate: 1mL/min (176MV/hr)
UV Trace at 280nm
Conductivity Trace
0
20
40
60
80
100 ml
mS/cm
80.0
60.0
40.0
20.0
Convective

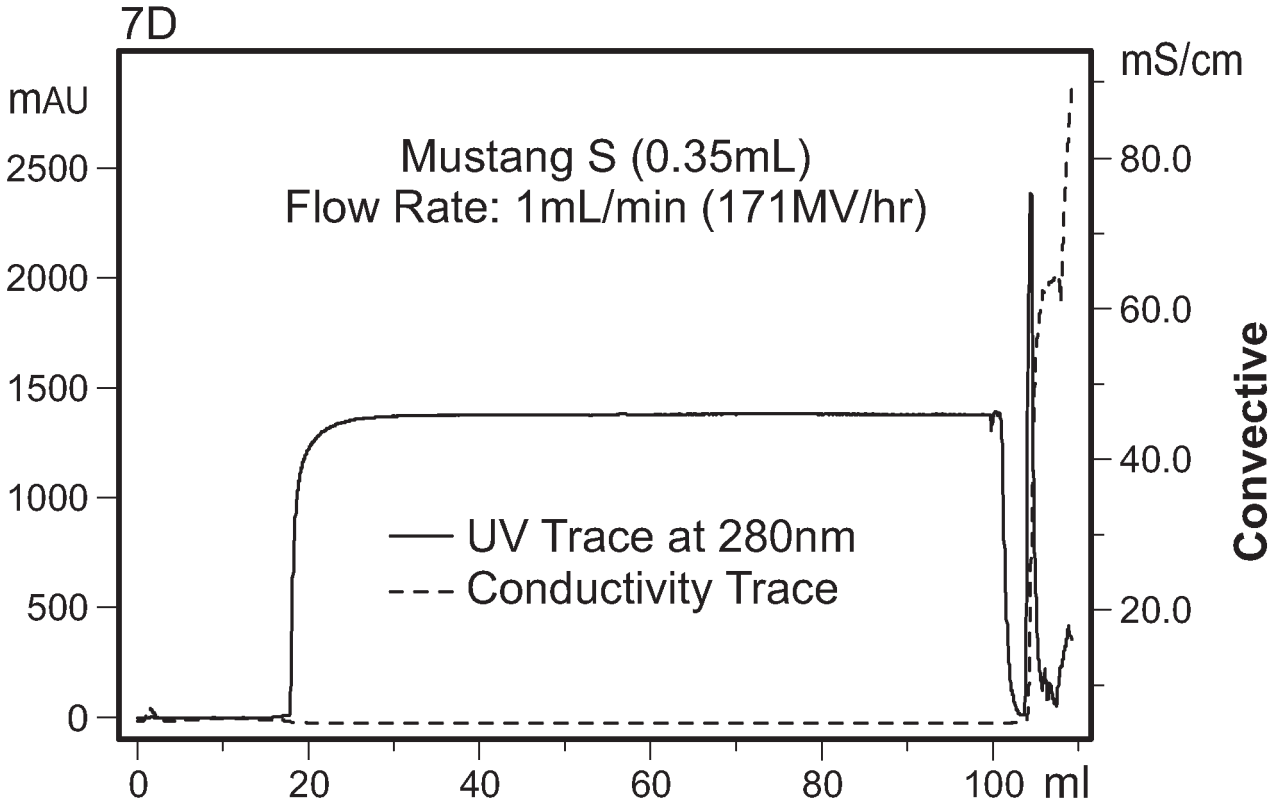

7D
mAU
2500
2000
1500
1000
500
0
Mustang S (0.35mL)
Flow Rate: 1mL/min (171MV/hr)
UV Trace at 280nm
Conductivity Trace
0
20
40
60
80
100 ml
mS/cm
80.0
60.0
40.0
20.0
Convective

FIG. 7 (Cont.)

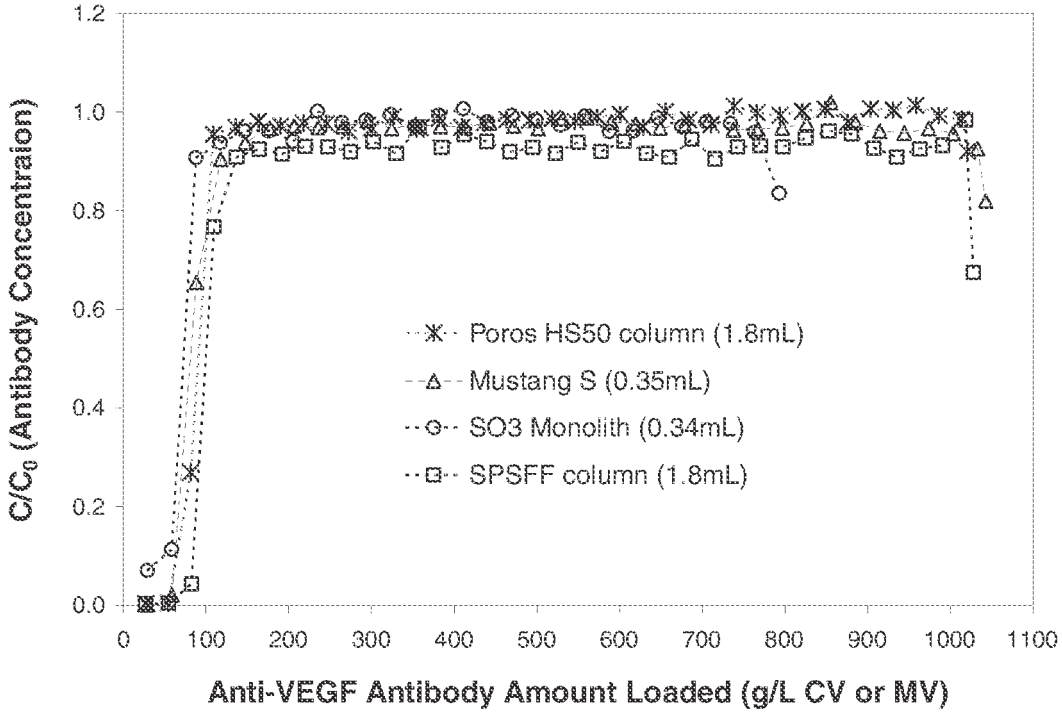

1.2
1.0
0.8
0.6
0.4
0.2
0.0
C/C0 (Antibody Concentraion)
0
100
200
300
400
500
600
700
800
900
1000
1100
Anti-VEGF Antibody Amount Loaded (g/L CV or MV)
Poros HS50 column (1.8mL)
Mustang S (0.35mL)
SO3 Monolith (0.34mL)
SPSFF column (1.8mL)

FIG. 8

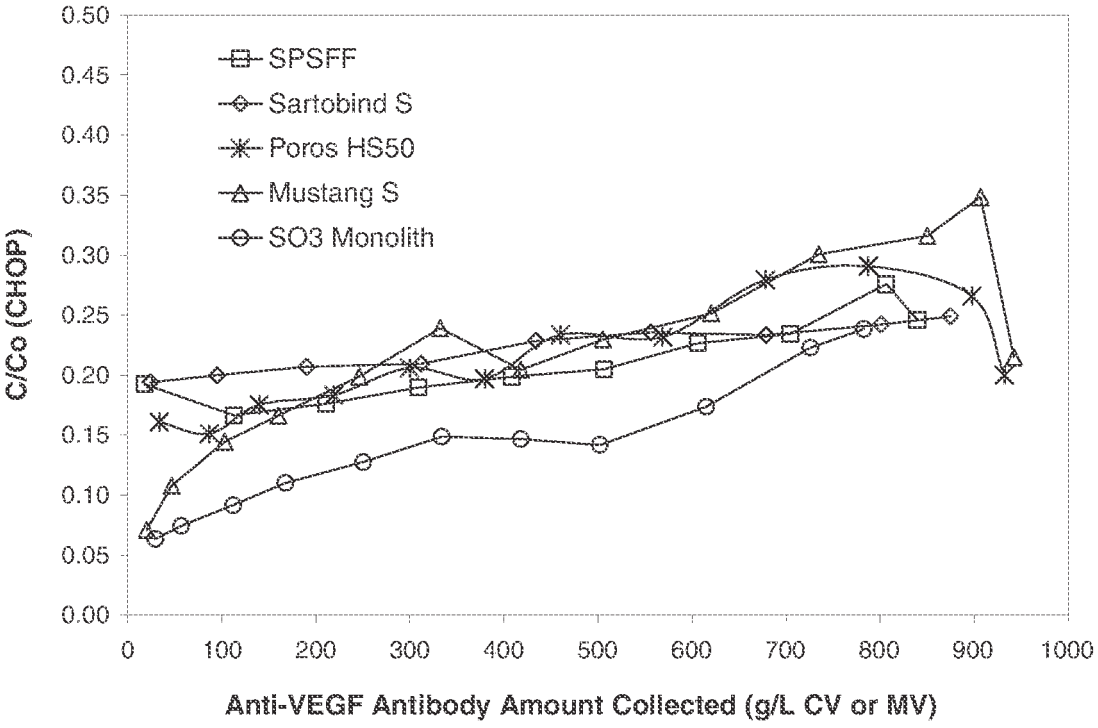

0.50
0.45
0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
0.00
C/Co (CHOP)
0
100
200
300
400
500
600
700
800
900
1000
Anti-VEGF Antibody Amount Collected (g/L CV or MV)
SPSFF
Sartobind S
Poros HS50
Mustang S
SO3 Monolith

FIG. 9

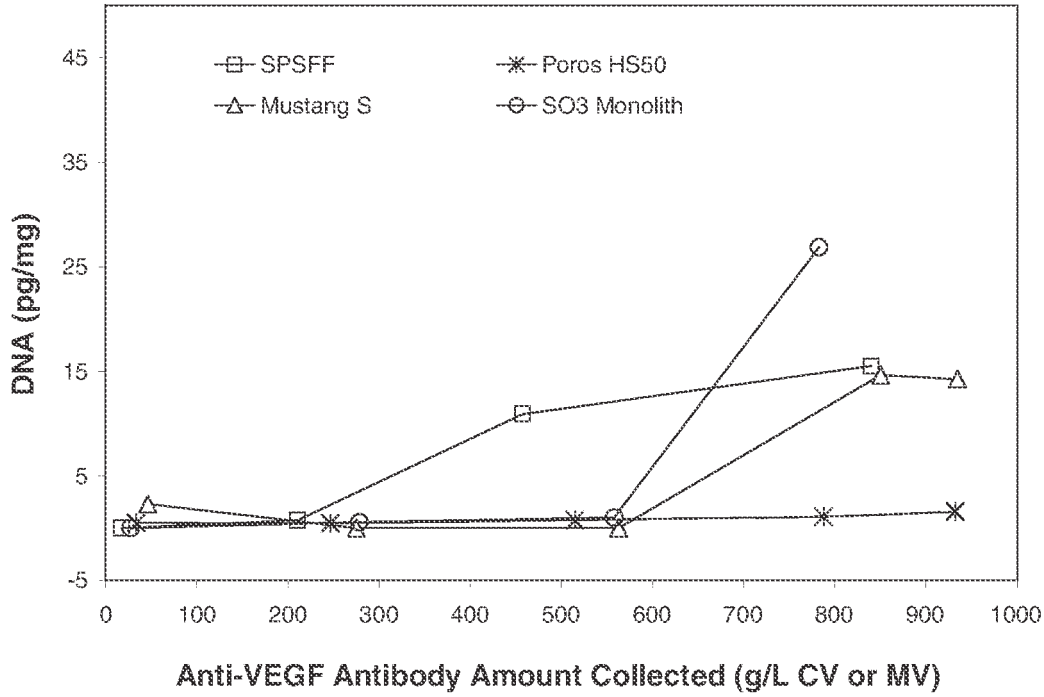

SPSFF
Poros HS50
Mustang S
SO3 Monolith
DNA (pg/mg)
45
35
25
15
5
-5
0
100
200
300
400
500
600
700
800
900
1000
Anti-VEGF Antibody Amount Collected (g/L CV or MV)

FIG. 10

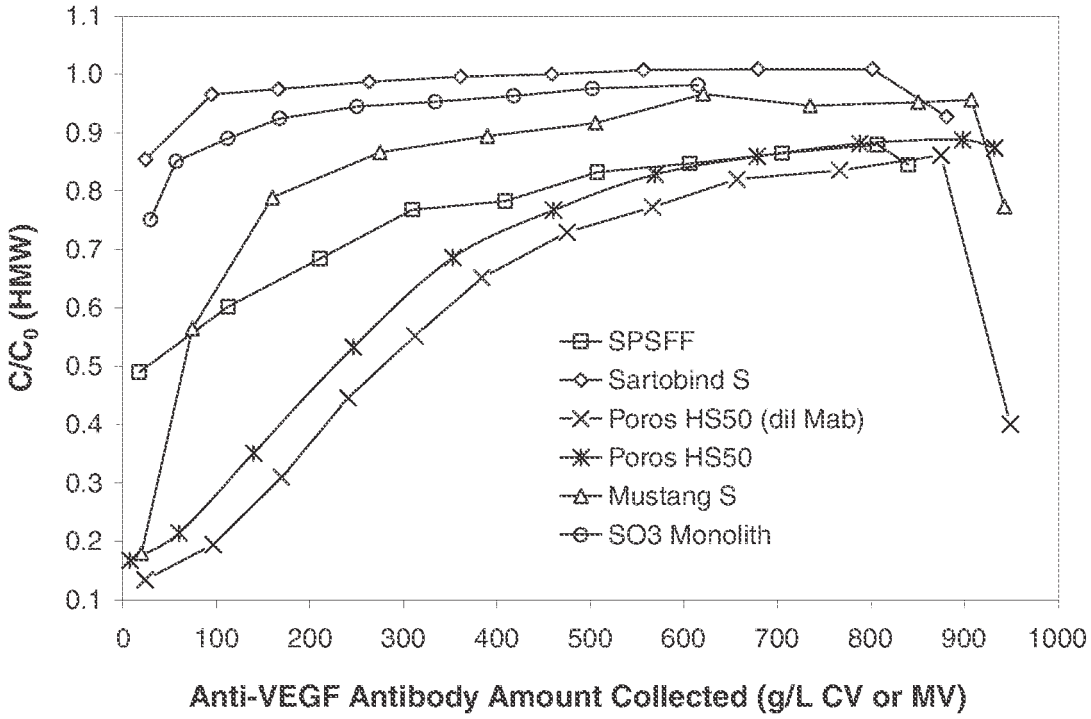

C/C0 (HMW)
1.1
1.0
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0
100
200
300
400
500
600
700
800
900
1000
SPSFF
Sartobind S
Poros HS50 (dil Mab)
Poros HS50
Mustang S
SO3 Monolith
Anti-VEGF Antibody Amount Collected (g/L CV or MV)

FIG. 11

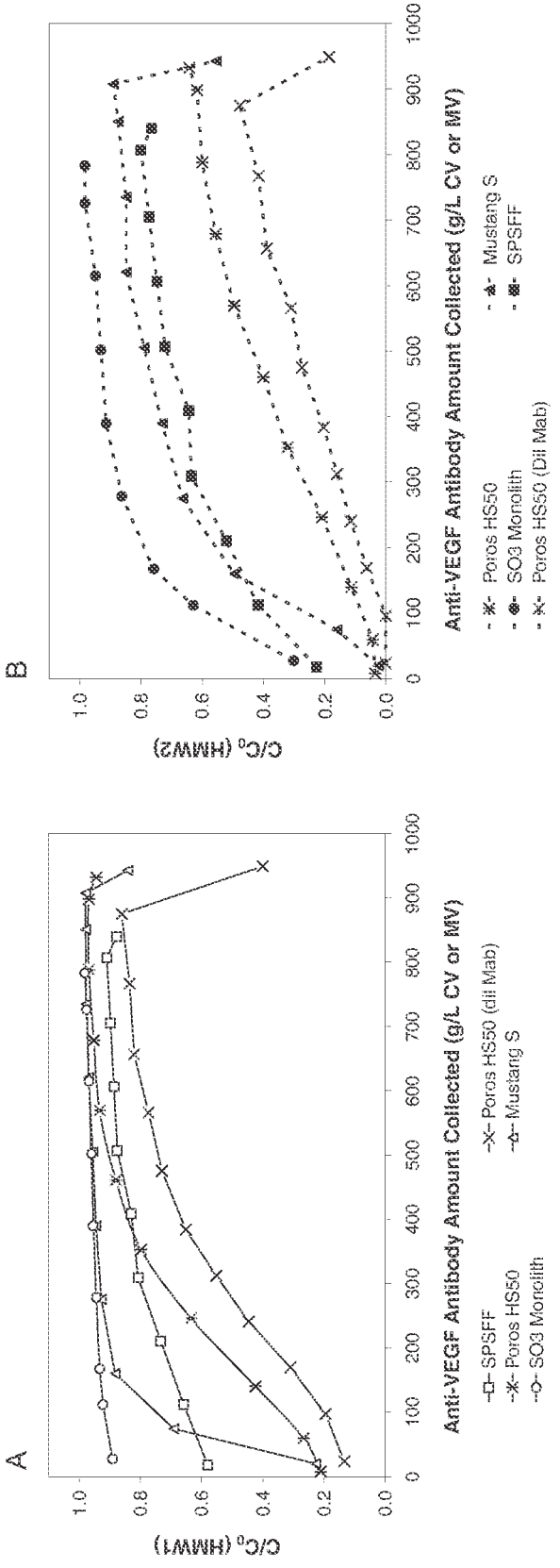

A
B
C/C0 (HMWS1)
C/C0 (HMWS2)
0.0
0.2
0.4
0.6
0.8
1.0
0
100
200
300
400
500
600
700
800
900
1000
Anti-VEGF Antibody Amount Collected (g/L CV or MV)
SPSFF
Poros HS50
SO3 Monolith
Poros HS50 (dil Mab)
Mustang S

FIG. 12

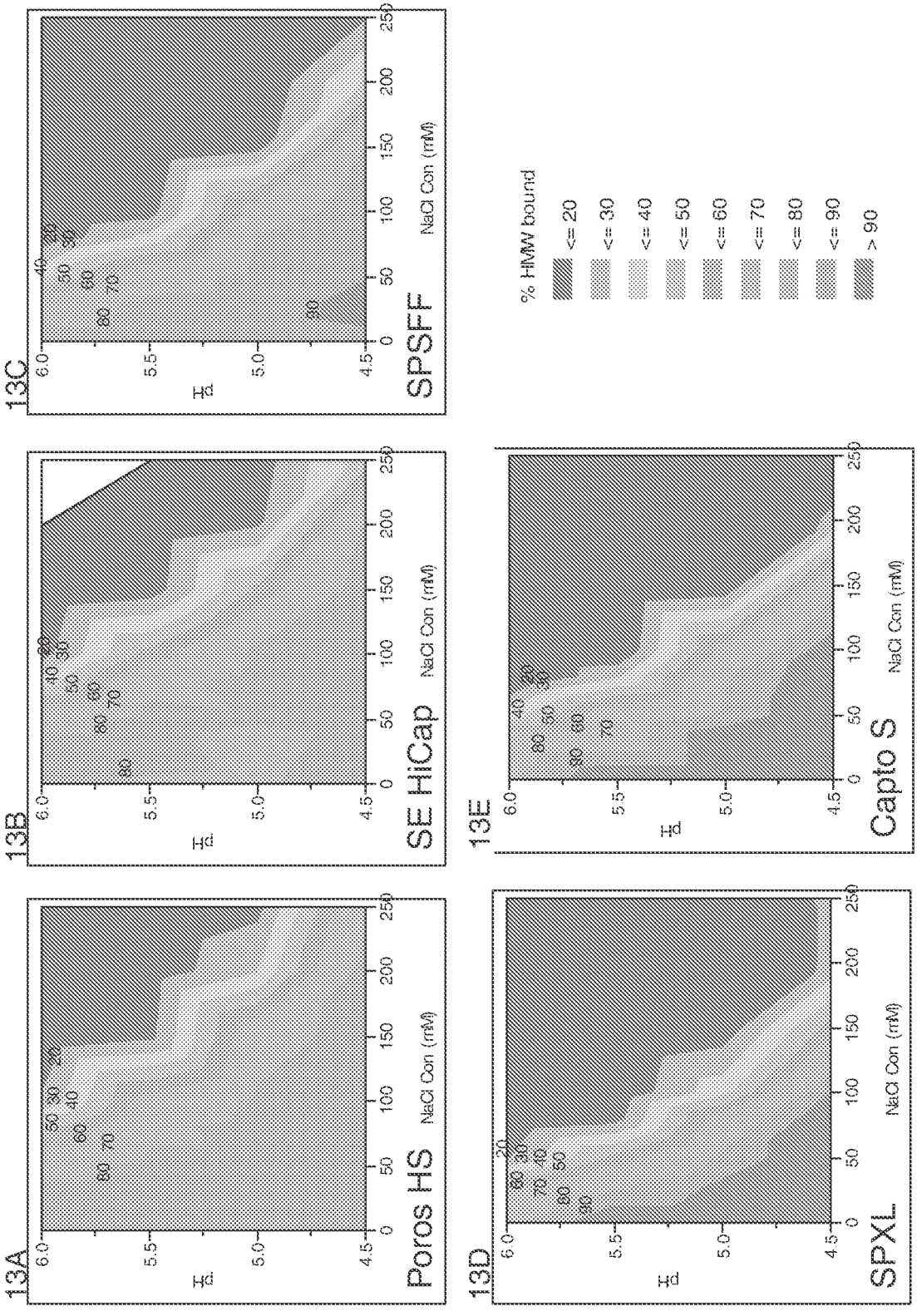

13A
Poros HS
13B
SE HiCap
13C
SPSFF
13D
SPXL
13E
Capto S
pH
NaCl Con (mM)
% HMW bound
<= 20
<= 30
<= 40
<= 50
<= 60
<= 70
<= 80
<= 90
> 90

FIG. 13

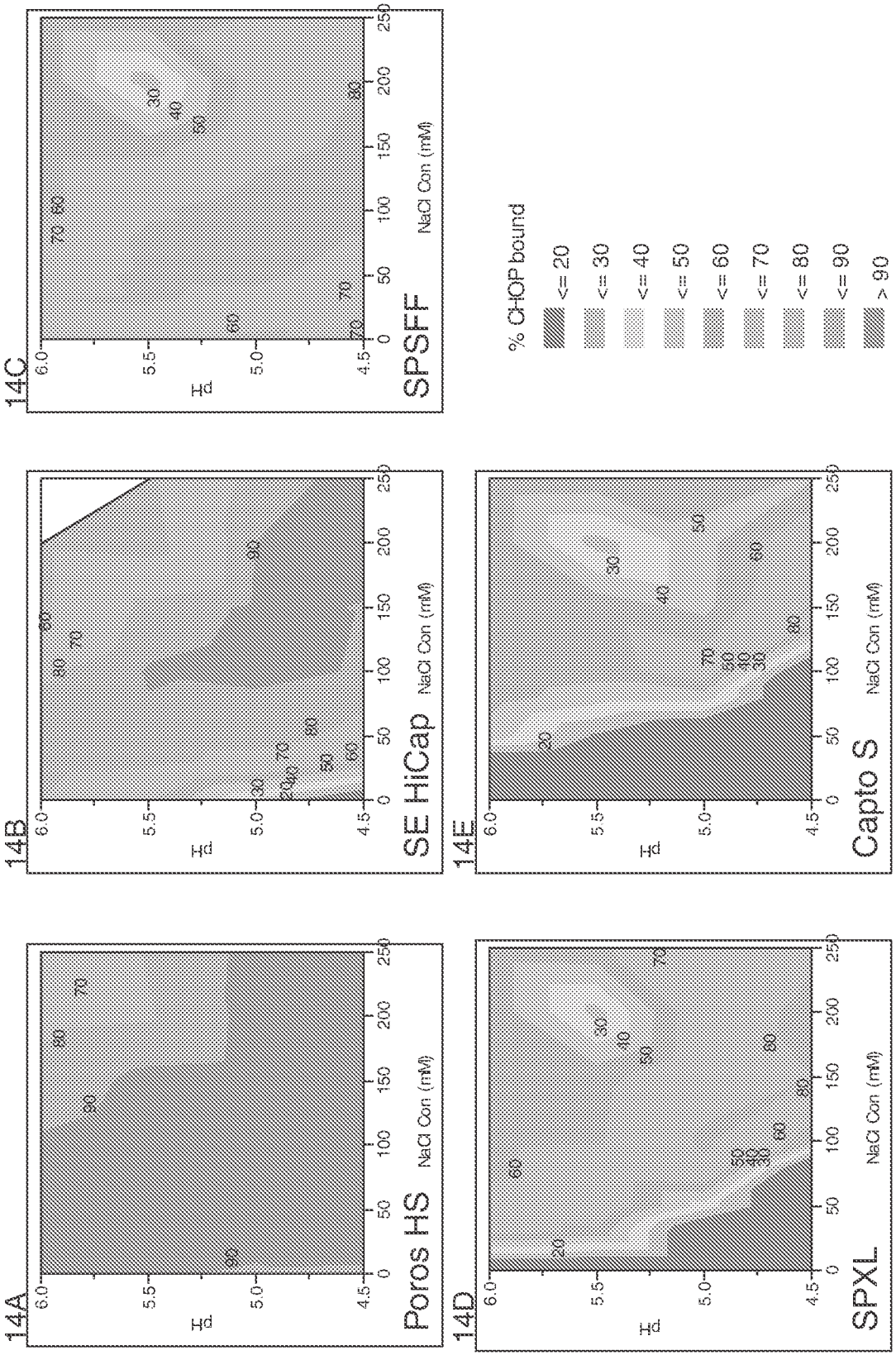

14A
Poros HS
14B
SE HiCap
14C
SPSFF
14D
SPXL
14E
Capto S
pH
NaCl Con (mM)
% CHOP bound
<= 20
<= 30
<= 40
<= 50
<= 60
<= 70
<= 80
<= 90
> 90

FIG. 14

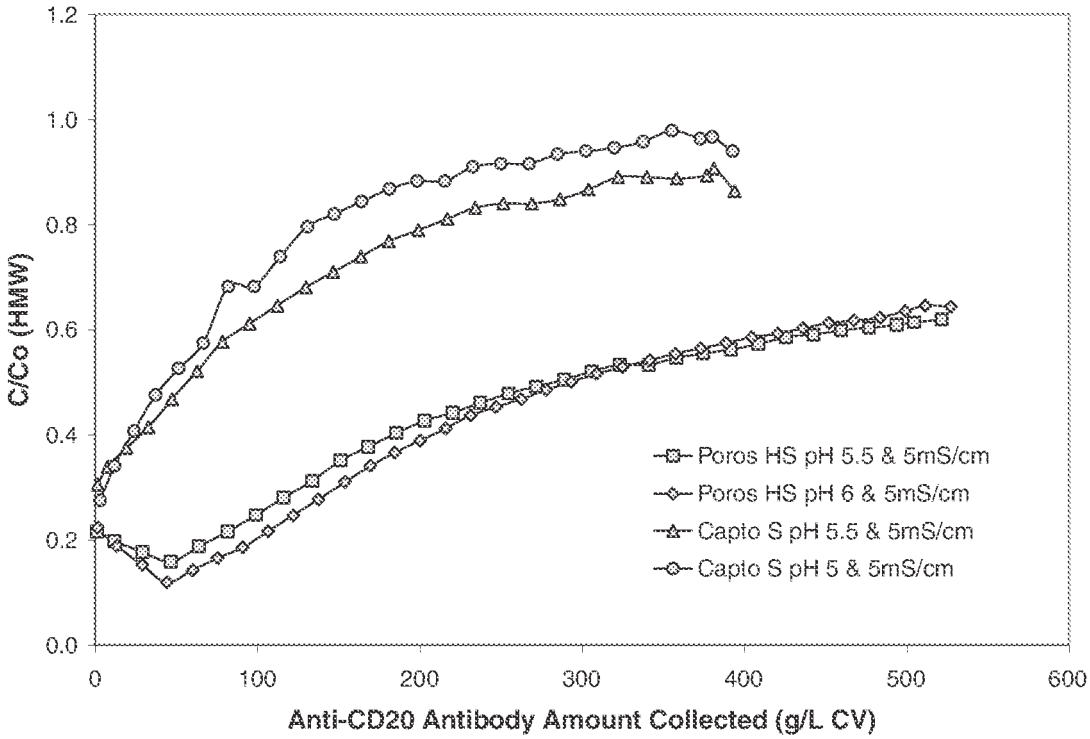

1.2
1.0
0.8
0.6
0.4
0.2
0.0
C/Co (HMW)
0
100
200
300
400
500
600
Anti-CD20 Antibody Amount Collected (g/L CV)
Poros HS pH 5.5 & 5mS/cm
Poros HS pH 6 & 5mS/cm
Capto S pH 5.5 & 5mS/cm
Capto S pH 5 & 5mS/cm

FIG. 15

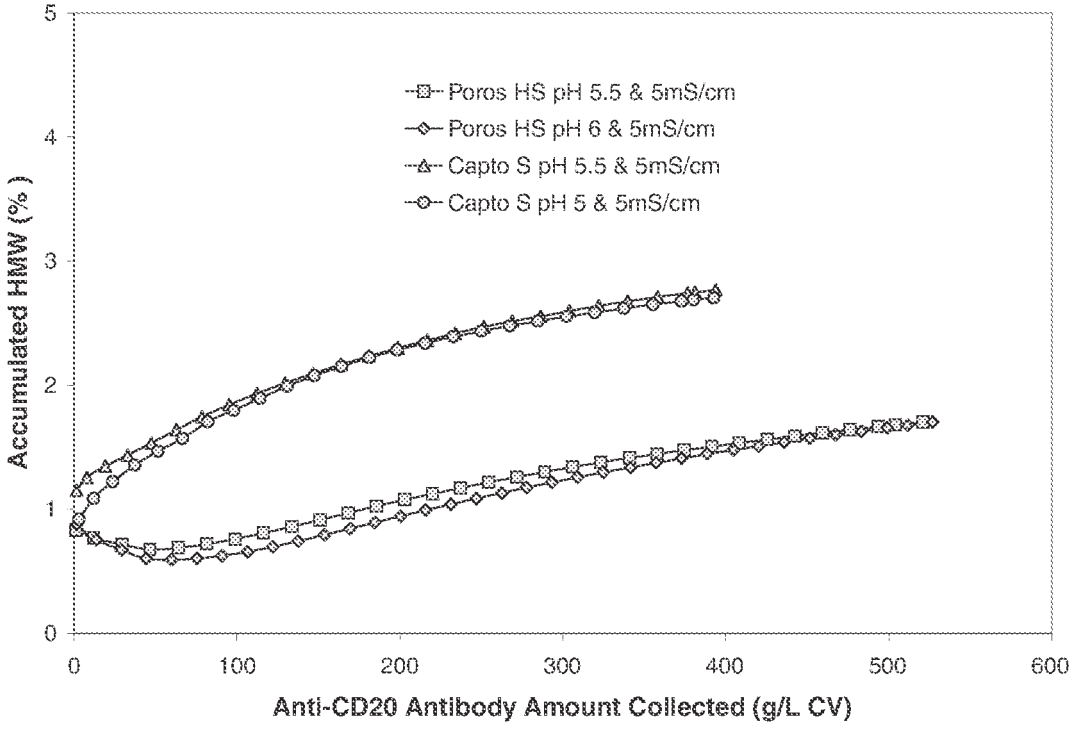

5
4
3
2
1
0
Accumulated HMW (%)
0
100
200
300
400
500
600
Anti-CD20 Antibody Amount Collected (g/L CV)
Poros HS pH 5.5 & 5mS/cm
Poros HS pH 6 & 5mS/cm
Capto S pH 5.5 & 5mS/cm
Capto S pH 5 & 5mS/cm

FIG. 16

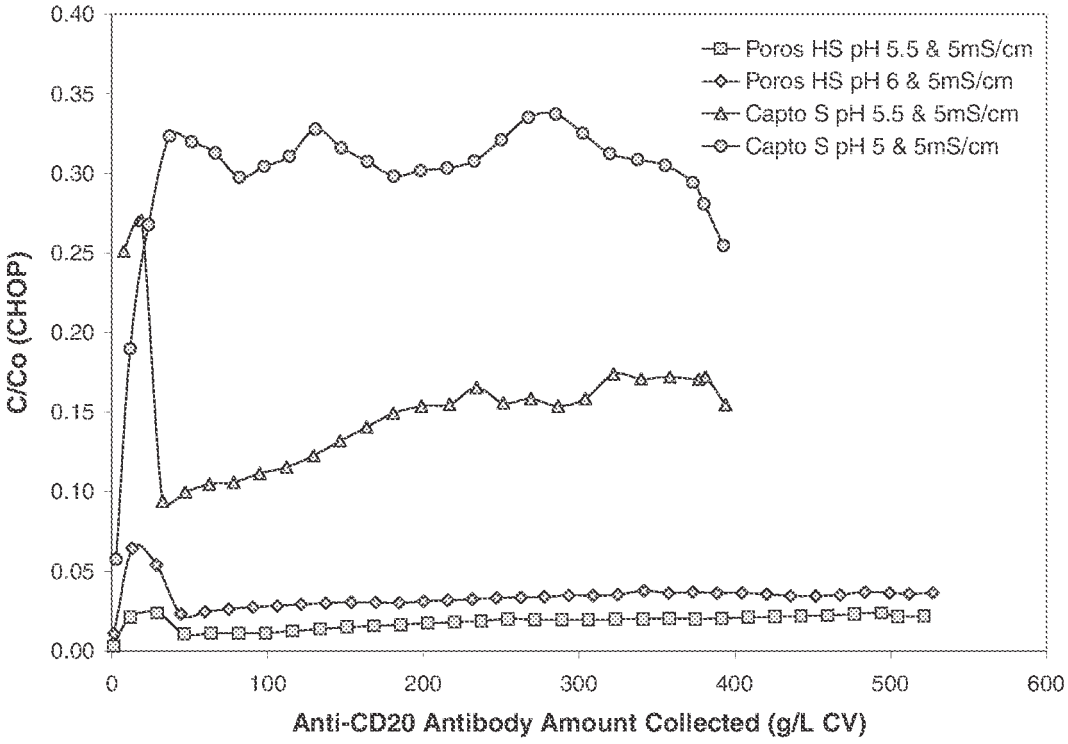

0.40
0.35
0.30
0.25
0.20
0.15
0.10
0.05
0.00
C/Co (CHOP)
0
100
200
300
400
500
600
Anti-CD20 Antibody Amount Collected (g/L CV)
Poros HS pH 5.5 & 5mS/cm
Poros HS pH 6 & 5mS/cm
Capto S pH 5.5 & 5mS/cm
Capto S pH 5 & 5mS/cm

FIG. 17

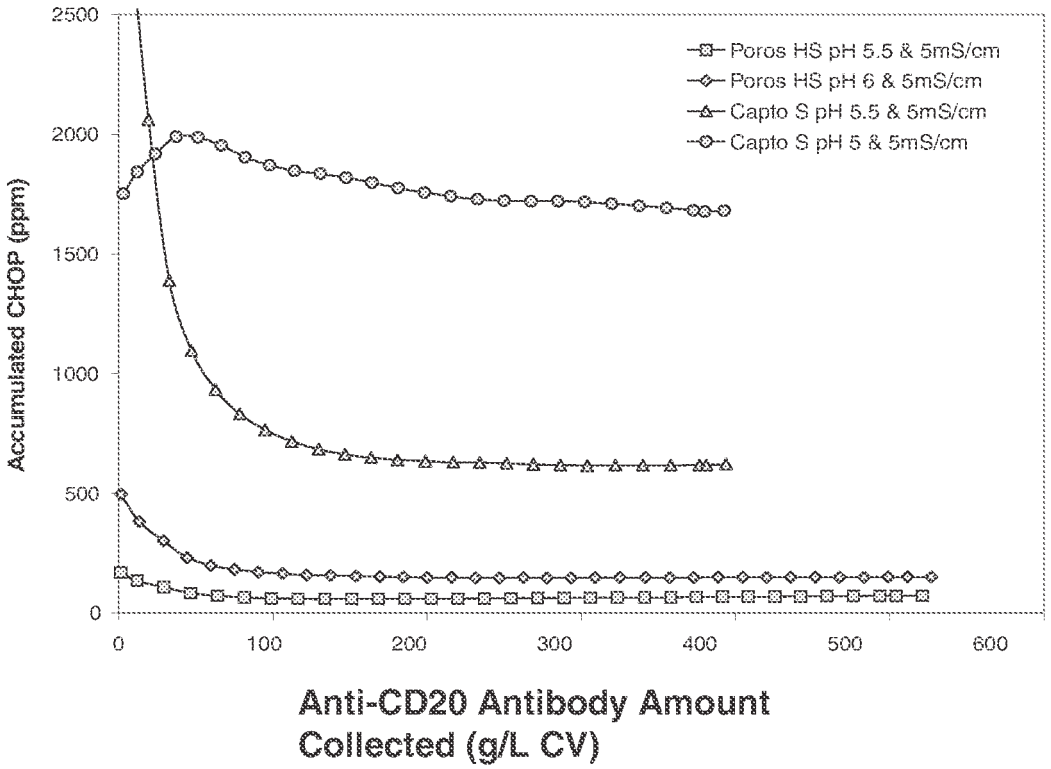
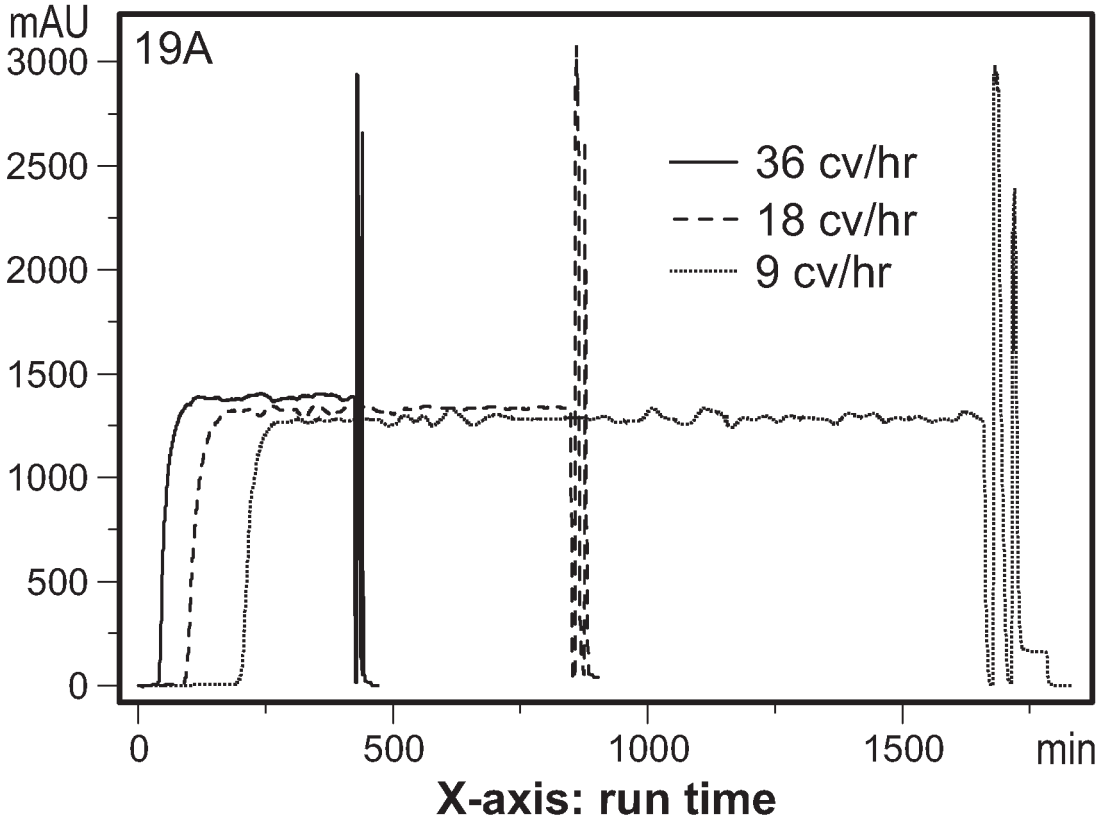

2500
2000
1500
1000
500
0
Accumulated CHOP (ppm)
0
100
200
300
400
500
600
Anti-CD20 Antibody Amount Collected (g/L CV)
Poros HS pH 5.5 & 5mS/cm
Poros HS pH 6 & 5mS/cm
Capto S pH 5.5 & 5mS/cm
Capto S pH 5 & 5mS/cm

FIG. 18


mAU
3000
2500
2000
1500
1000
500
0
19A
36 cv/hr
18 cv/hr
9 cv/hr
0
500
1000
1500
min
X-axis: run time

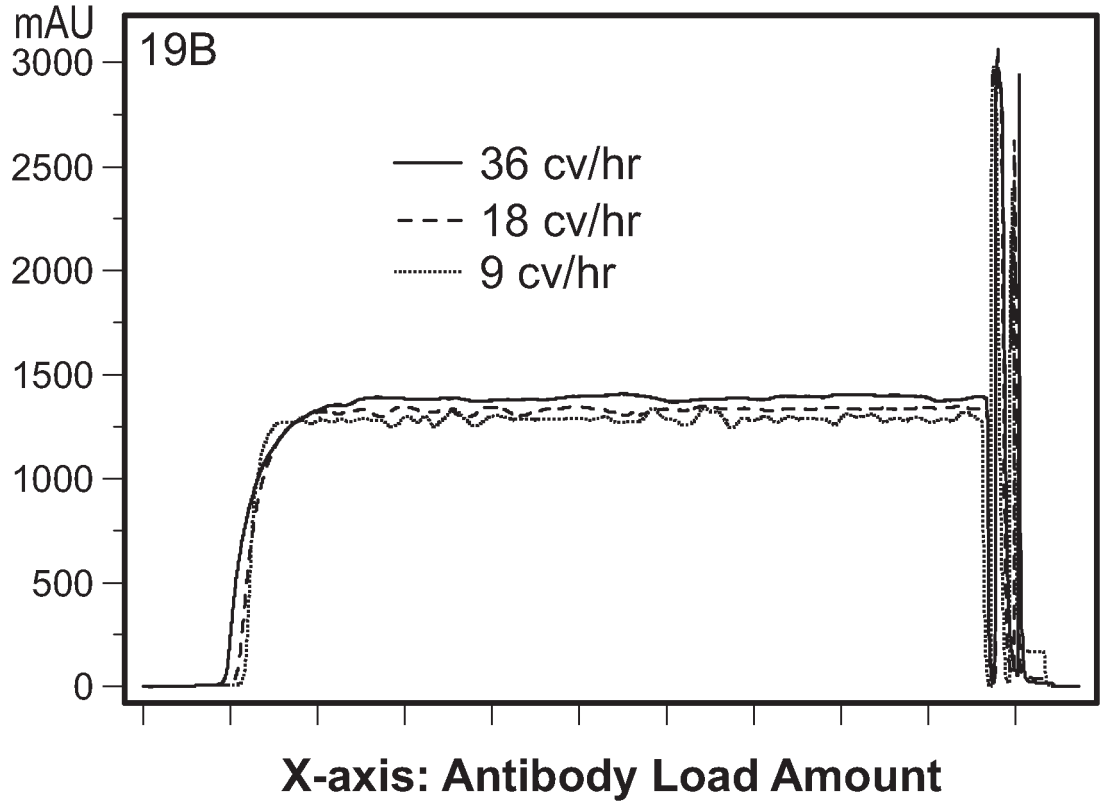

mAU
3000
2500
2000
1500
1000
500
0
19B
36 cv/hr
18 cv/hr
9 cv/hr
X-axis: Antibody Load Amount

FIG. 19

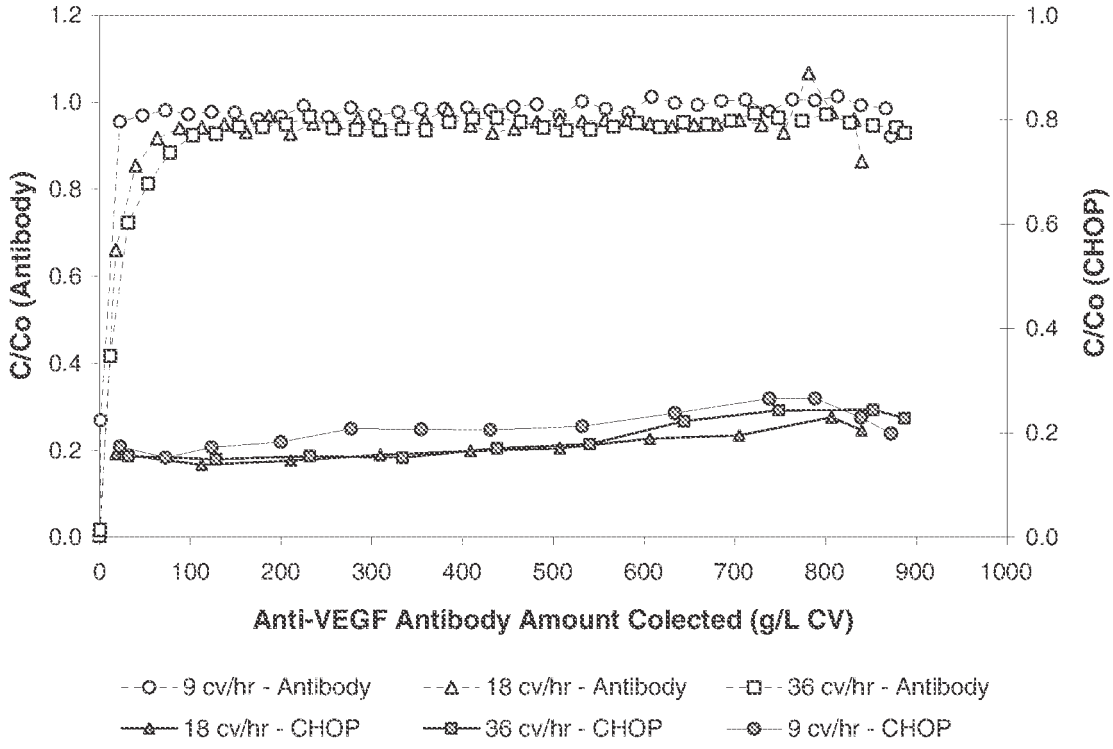

C/Co (Antibody)
C/Co (CHOP)
Anti-VEGF Antibody Amount Colected (g/L CV)
9 cv/hr - Antibody
18 cv/hr - Antibody
36 cv/hr - Antibody
18 cv/hr - CHOP
36 cv/hr - CHOP
9 cv/hr - CHOP

FIG. 20

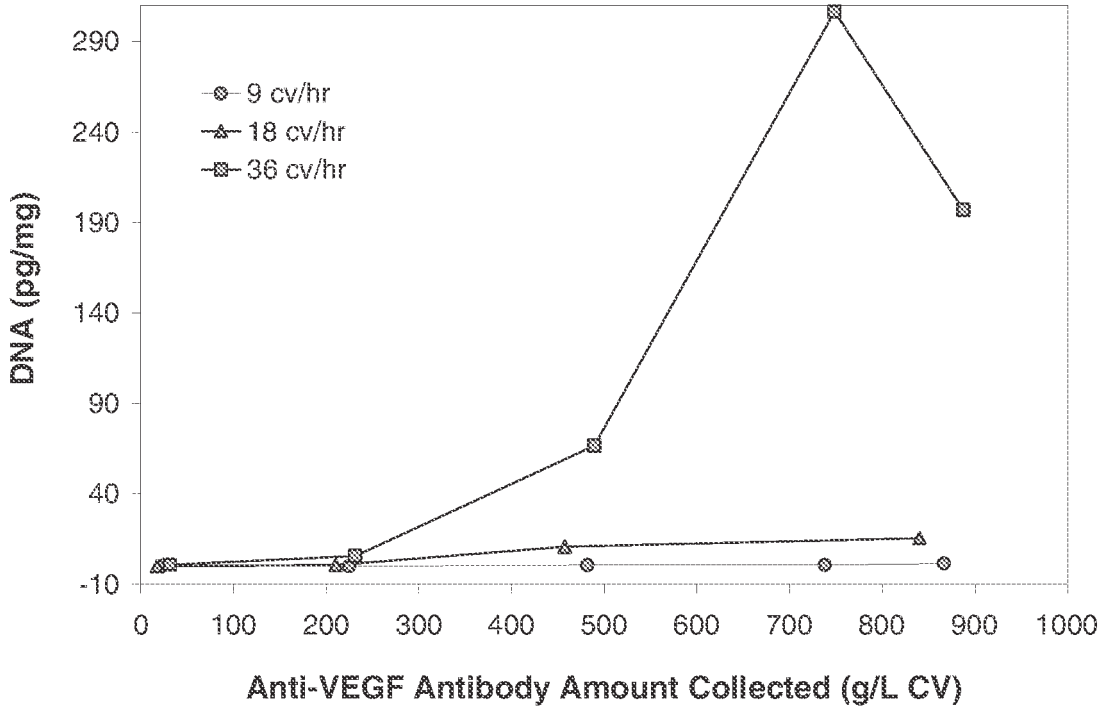

DNA (pg/mg)
9 cv/hr
18 cv/hr
36 cv/hr
Anti-VEGF Antibody Amount Collected (g/L CV)

FIG. 21

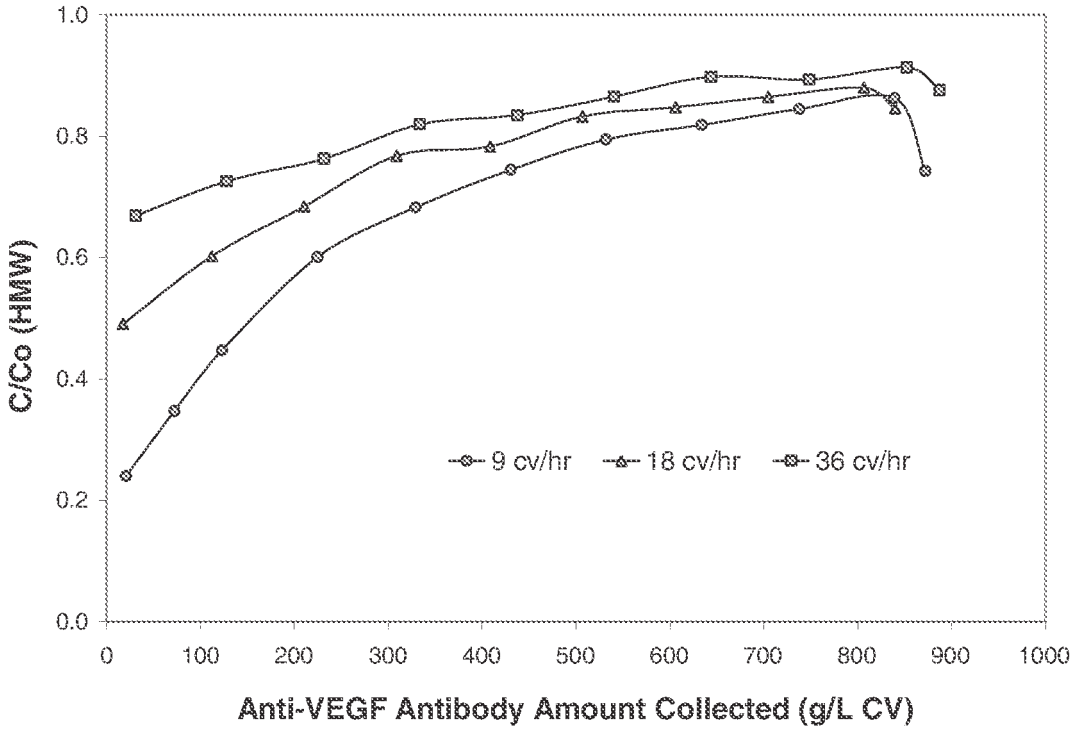

1.0
0.8
0.6
0.4
0.2
0.0
C/Co (HMW)
9 cv/hr
18 cv/hr
36 cv/hr
0
100
200
300
400
500
600
700
800
900
1000
Anti-VEGF Antibody Amount Collected (g/L CV)

FIG. 22

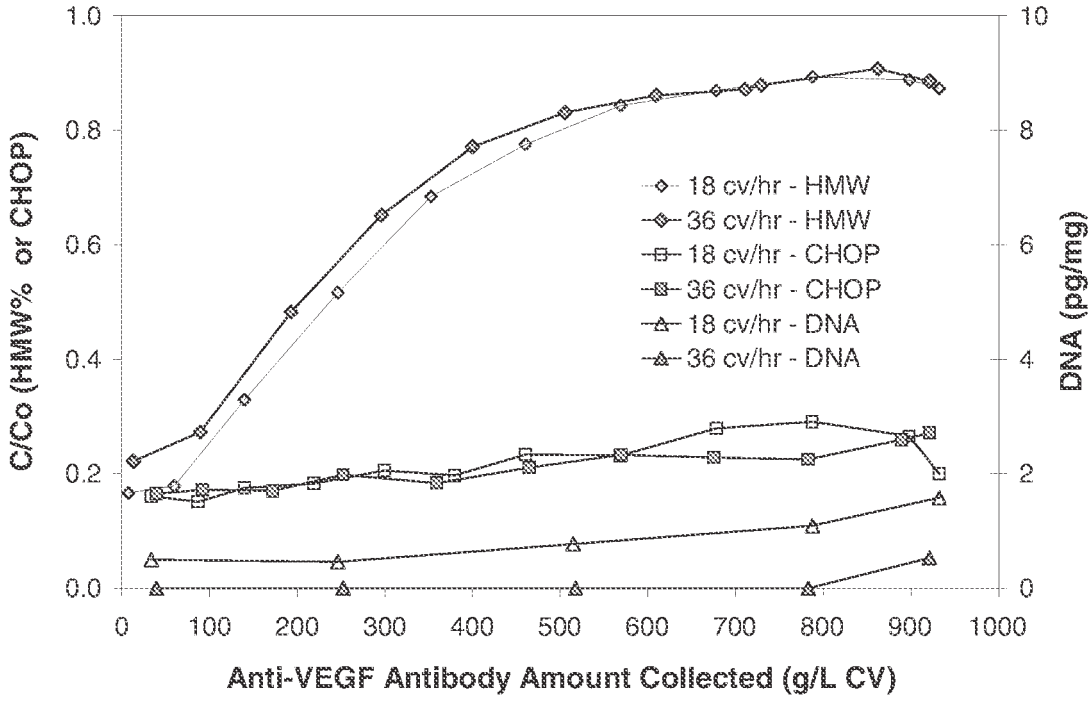

1.0
0.8
0.6
0.4
0.2
0.0
C/Co (HMW% or CHOP)
10
8
6
4
2
0
DNA (pg/mg)
18 cv/hr - HMW
36 cv/hr - HMW
18 cv/hr - CHOP
36 cv/hr - CHOP
18 cv/hr - DNA
36 cv/hr - DNA
0
100
200
300
400
500
600
700
800
900
1000
Anti-VEGF Antibody Amount Collected (g/L CV)

FIG. 23

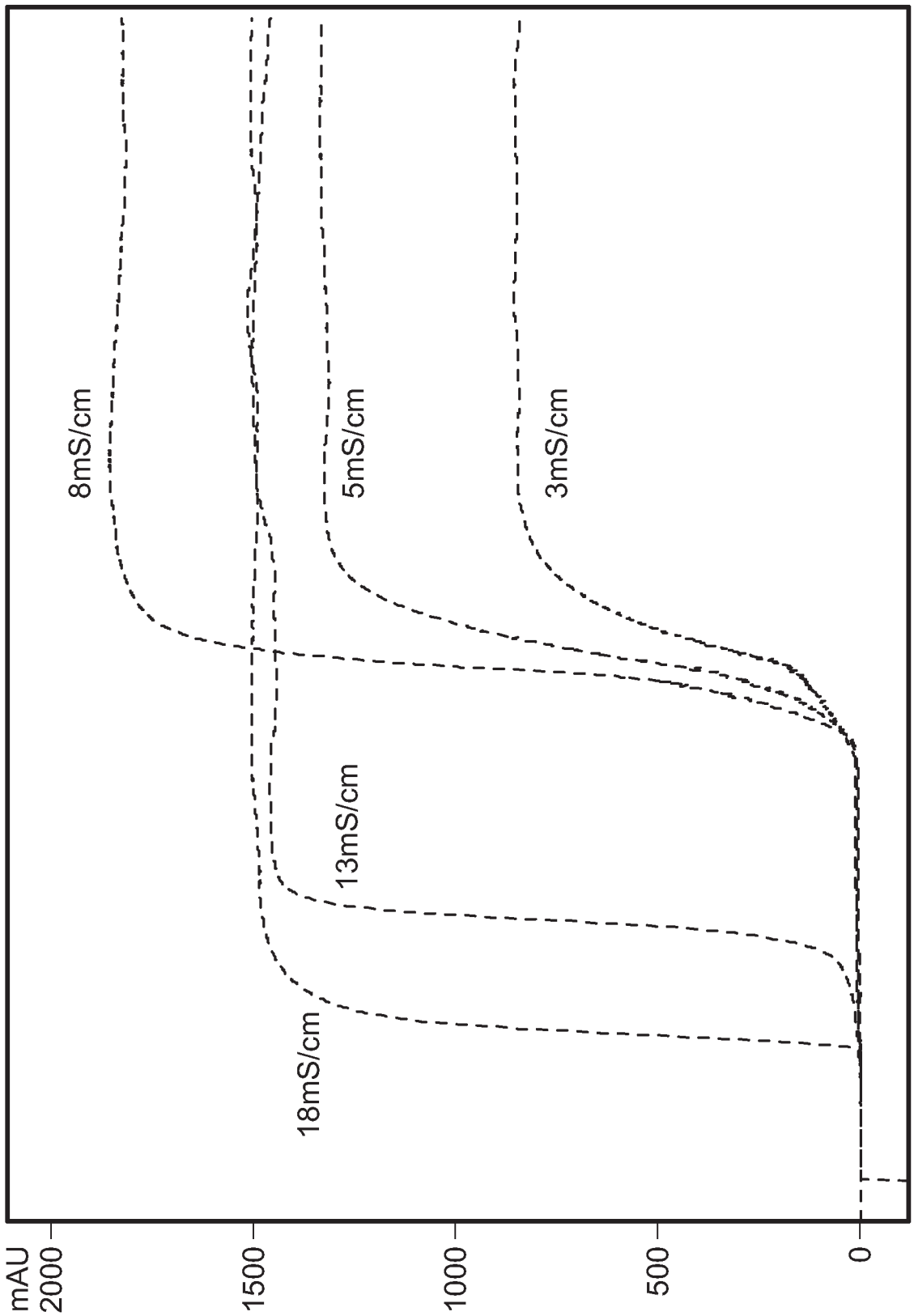

mAU
2000
1500
1000
500
0
8mS/cm
5mS/cm
3mS/cm
13mS/cm
18mS/cm
Normalized to anti-VEGF antibody load amount

FIG. 24

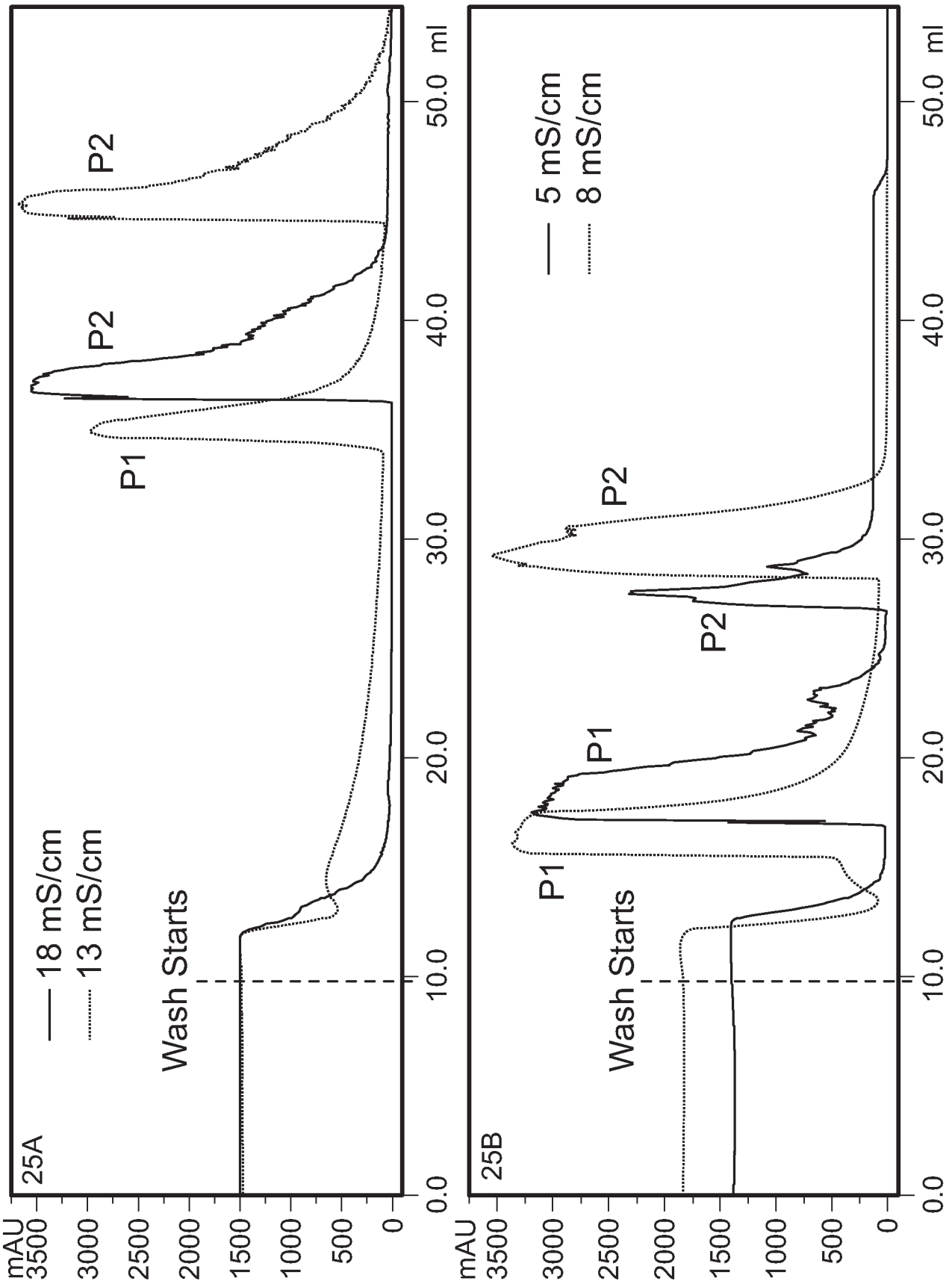

25A
18 mS/cm
13 mS/cm
Wash Starts
P1
P2
P2
mAU
3500
3000
2500
2000
1500
1000
500
0
0.0
10.0
20.0
30.0
40.0
50.0 ml
25B
5 mS/cm
8 mS/cm
Wash Starts
P1
P1
P2
P2
mAU
3500
3000
2500
2000
1500
1000
500
0
0.0
10.0
20.0
30.0
40.0
50.0 ml

FIG. 25

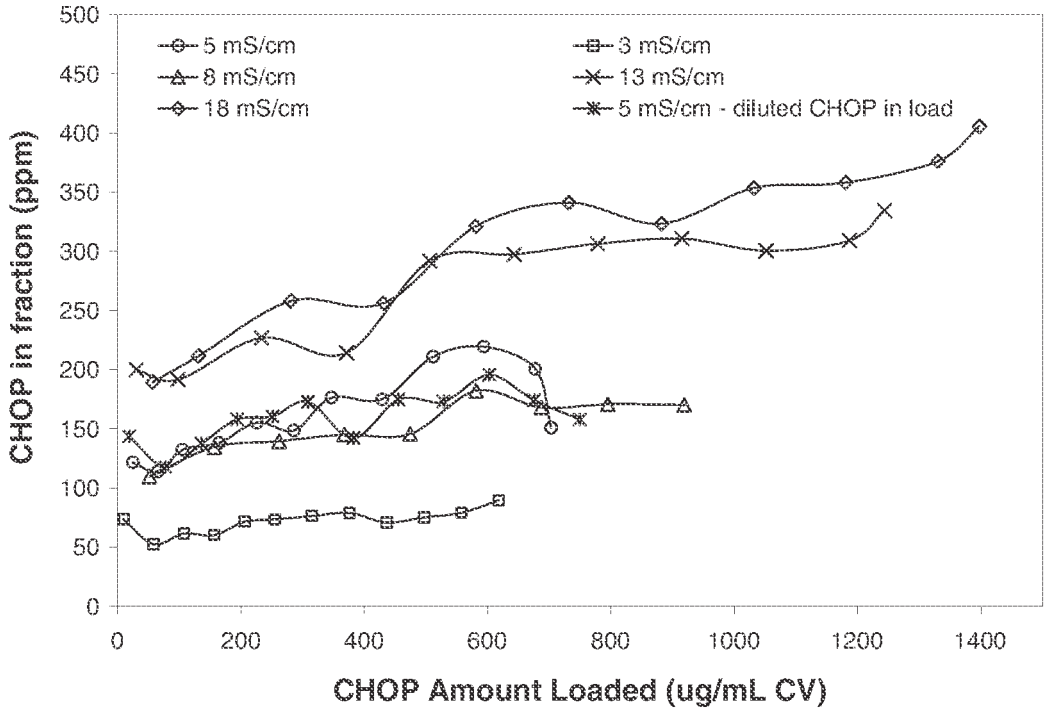

5 mS/cm
8 mS/cm
18 mS/cm
3 mS/cm
13 mS/cm
5 mS/cm - diluted CHOP in load
CHOP in fraction (ppm)
CHOP Amount Loaded (ug/mL CV)
0
50
100
150
200
250
300
350
400
450
500
0
200
400
600
800
1000
1200
1400

FIG. 26

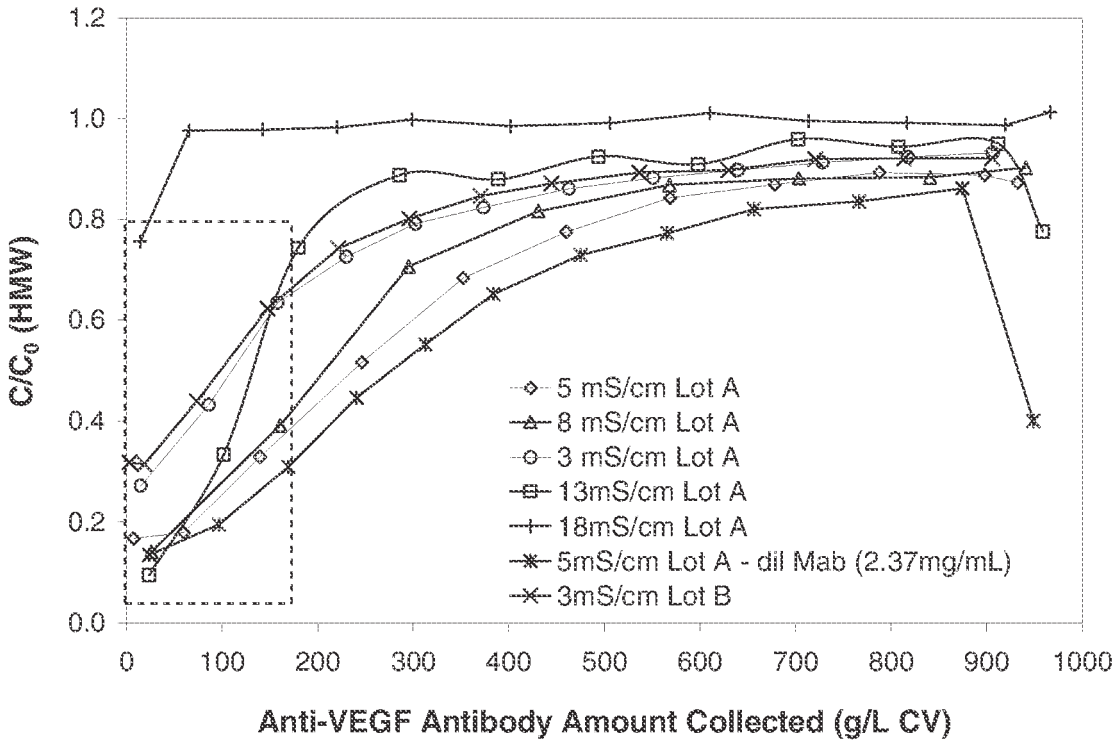

C/C0 (HMW)
Anti-VEGF Antibody Amount Collected (g/L CV)
5 mS/cm Lot A
8 mS/cm Lot A
3 mS/cm Lot A
13mS/cm Lot A
18mS/cm Lot A
5mS/cm Lot A - dil Mab (2.37mg/mL)
3mS/cm Lot B
0.0
0.2
0.4
0.6
0.8
1.0
1.2
0
100
200
300
400
500
600
700
800
900
1000

FIG. 27

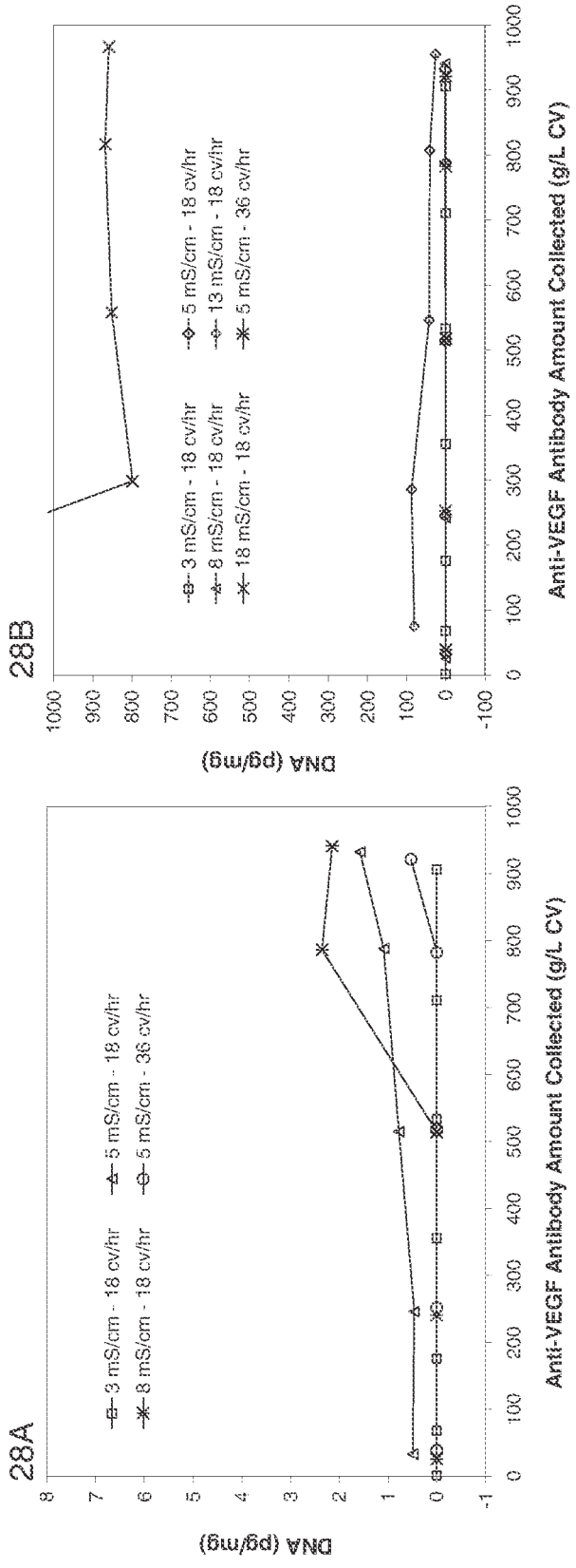

28A
DNA (pg/mg)
Anti-VEGF Antibody Amount Collected (g/L CV)
3 mS/cm - 18 cv/hr
5 mS/cm - 18 cv/hr
8 mS/cm - 18 cv/hr
5 mS/cm - 36 cv/hr
28B
DNA (pg/mg)
Anti-VEGF Antibody Amount Collected (g/L CV)
3 mS/cm - 18 cv/hr
5 mS/cm - 18 cv/hr
8 mS/cm - 18 cv/hr
13 mS/cm - 18 cv/hr
18 mS/cm - 18 cv/hr
5 mS/cm - 36 cv/hr

FIG. 28

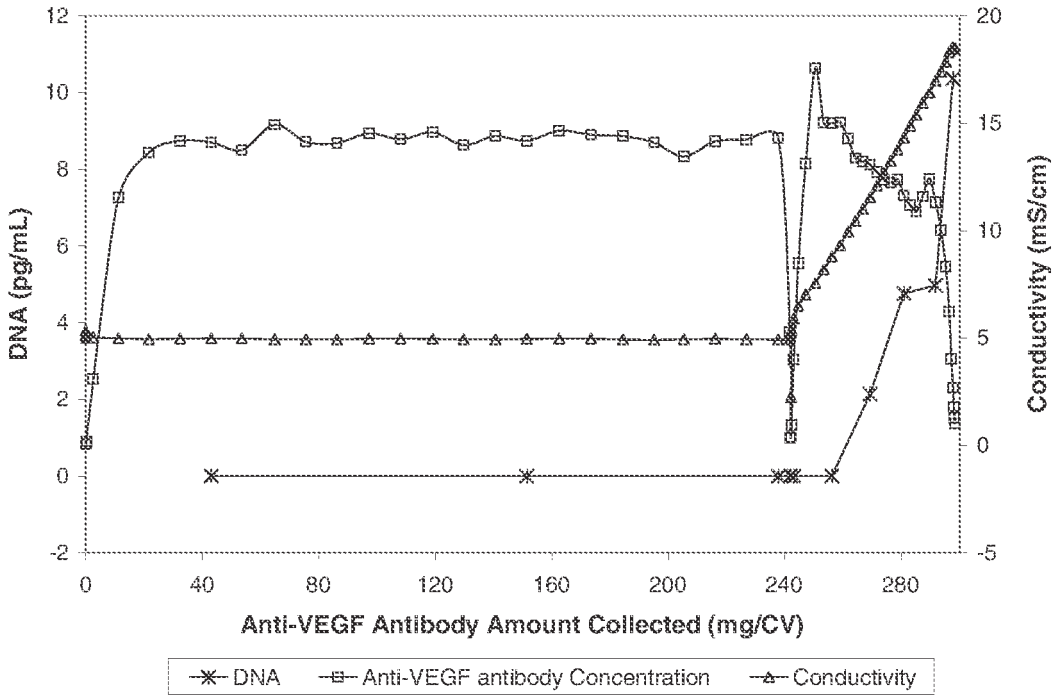

DNA (pg/mL)
Conductivity (mS/cm)
Anti-VEGF Antibody Amount Collected (mg/CV)
DNA
Anti-VEGF antibody Concentration
Conductivity

FIG. 29

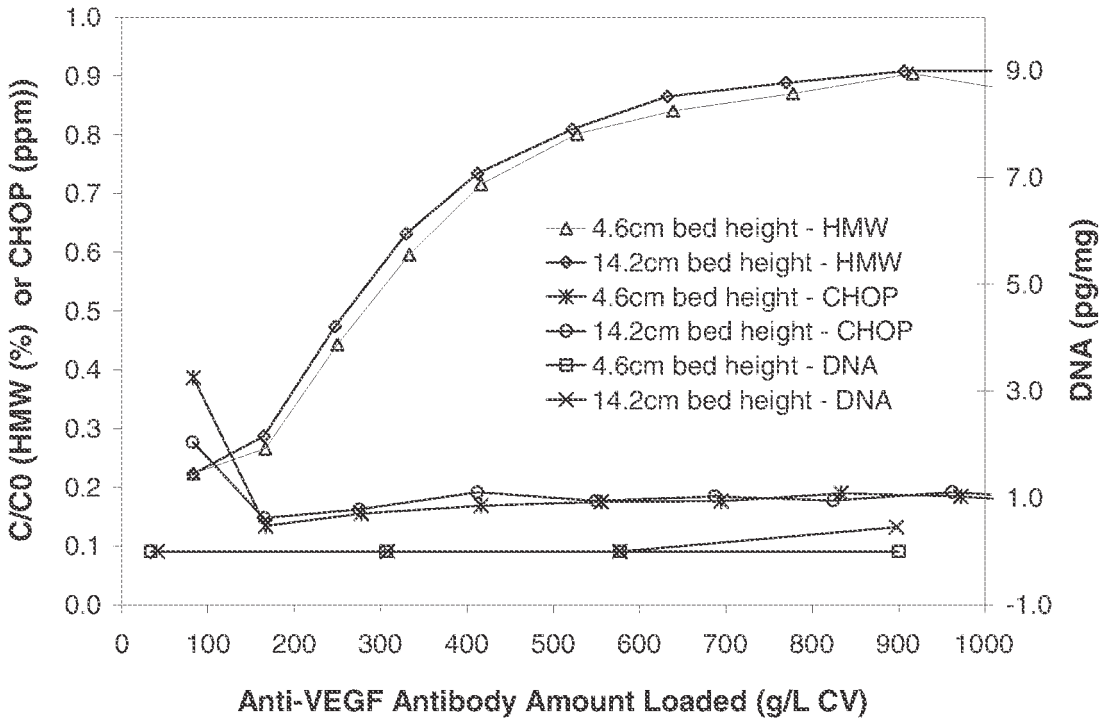

C/C0 (HMW (%) or CHOP (ppm))
DNA (pg/mg)
Anti-VEGF Antibody Amount Loaded (g/L CV)
4.6cm bed height - HMW
14.2cm bed height - HMW
4.6cm bed height - CHOP
14.2cm bed height - CHOP
4.6cm bed height - DNA
14.2cm bed height - DNA

FIG. 30

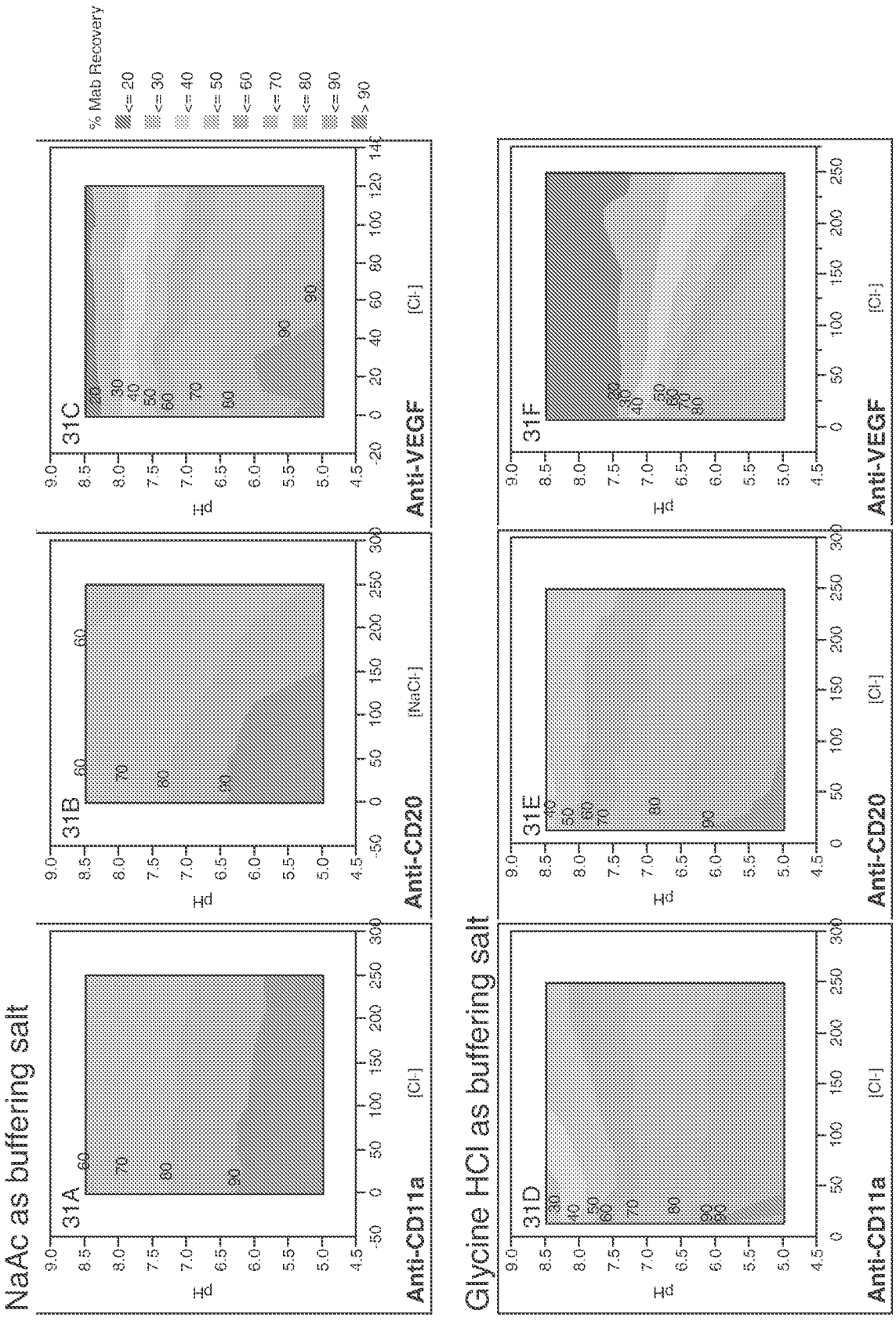

NaAc as buffering salt
Glycine HCl as buffering salt
31A
31B
31C
31D
31E
31F
Anti-CD11a
Anti-CD20
Anti-VEGF
[Cl-]
[NaCl-]
pH
% Mab Recovery
<= 20
<= 30
<= 40
<= 50
<= 60
<= 70
<= 80
<= 90
> 90

FIG. 31

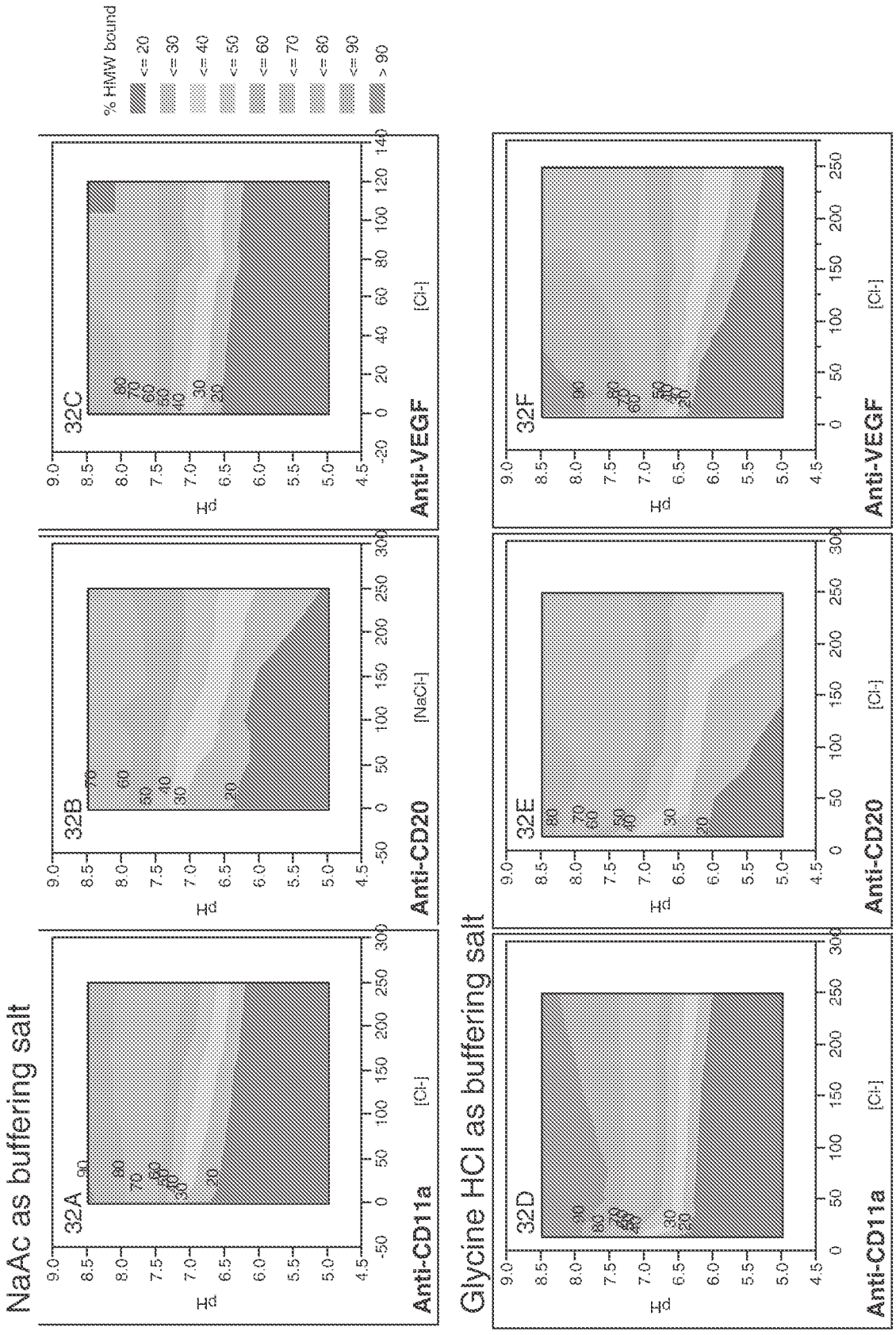

NaAc as buffering salt
Glycine HCl as buffering salt
% HMW bound
<= 20
<= 30
<= 40
<= 50
<= 60
<= 70
<= 80
<= 90
> 90
32A
32B
32C
32D
32E
32F
Anti-CD11a
Anti-CD20
Anti-VEGF
[Cl-]
[NaCl]
pH

FIG. 32

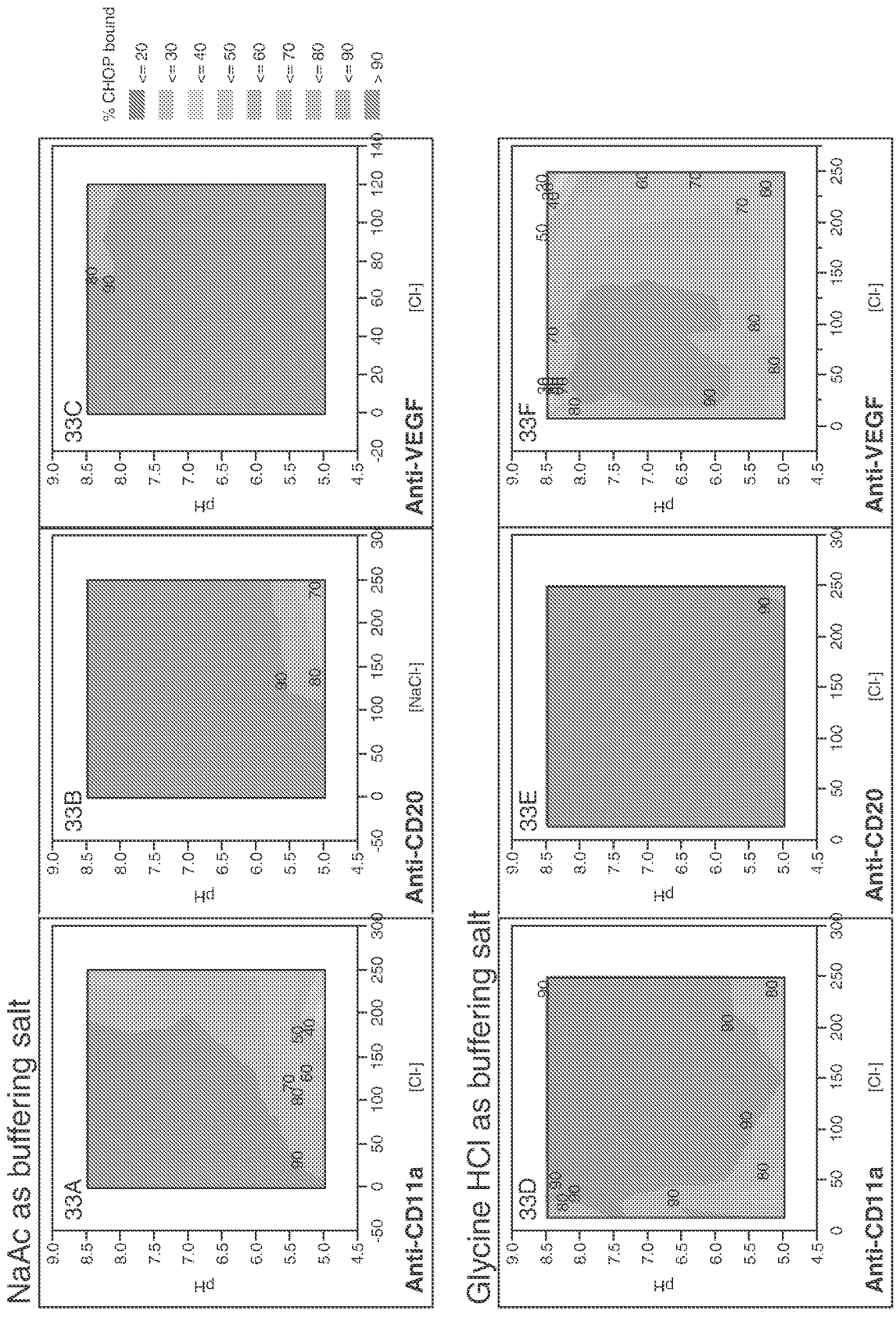

NaAc as buffering salt
Glycine HCl as buffering salt
33A
33B
33C
33D
33E
33F
pH
[Cl-]
[NaCl-]
Anti-CD11a
Anti-CD20
Anti-VEGF
% CHOP bound
<= 20
<= 30
<= 40
<= 50
<= 60
<= 70
<= 80
<= 90
> 90

FIG. 33

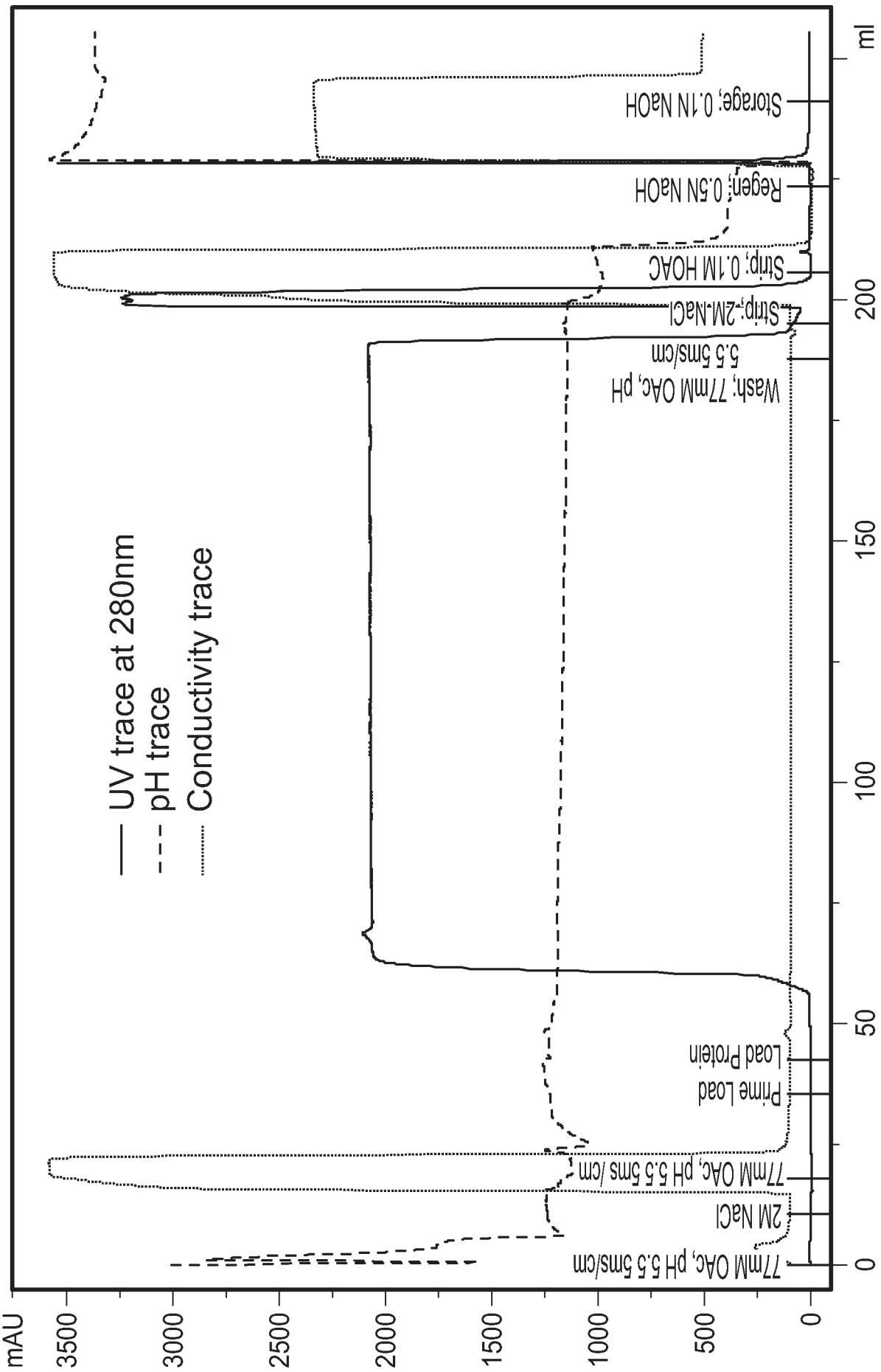

mAU
3500
3000
2500
2000
1500
1000
500
0
UV trace at 280nm
pH trace
Conductivity trace
77mM OAc, pH 5.5 5ms/cm
2M NaCl
77mM OAc, pH 5.5 5ms /cm
Prime Load
Load Protein
Wash; 77mM OAc, pH
5.5 5ms/cm
Strip; 2M NaCl
Strip; 0.1M HOAC
Regen; 0.5N NaOH
Storage; 0.1N NaOH
0
50
100
150
200
ml

FIG. 34