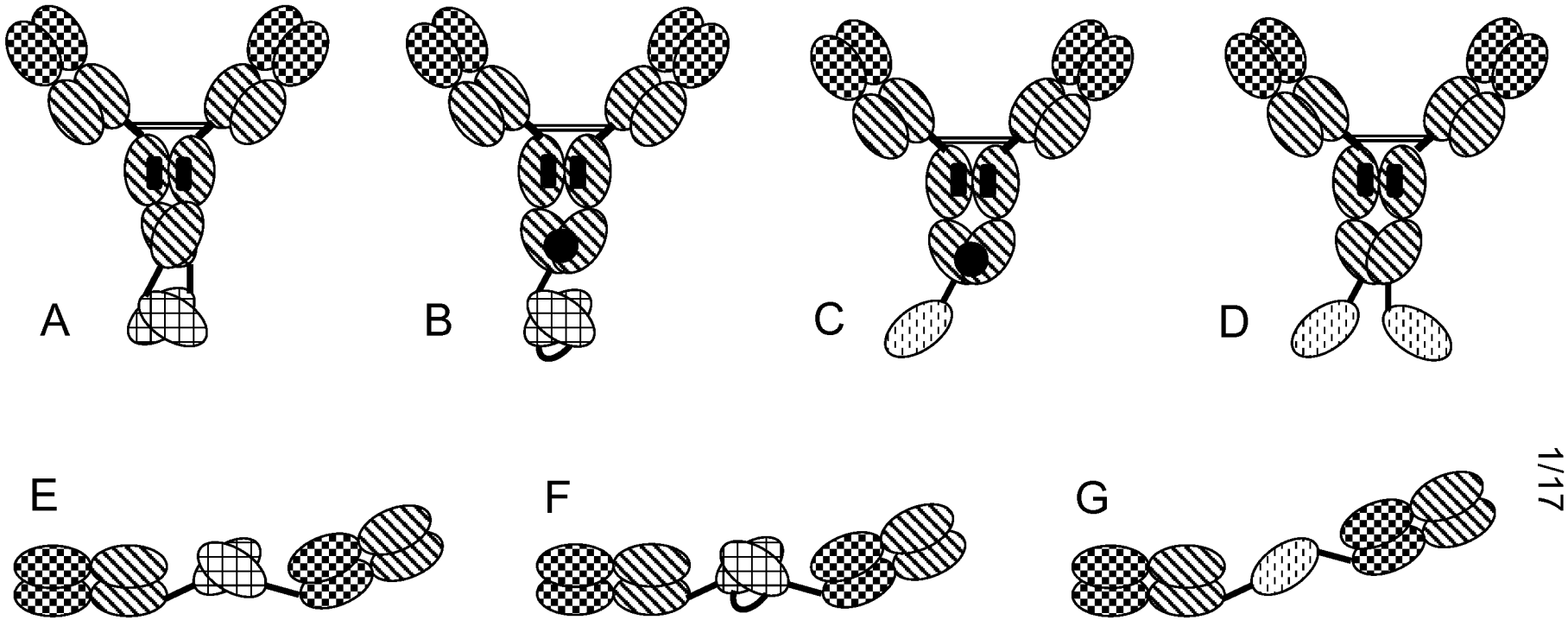

A
B
C
D
E
F
G
antibody V-domain
antibody C-domain
IL-10
LALA P329G mutation
knob-into-hole modification
monomeric IL-10

Figure 1

Figure 2

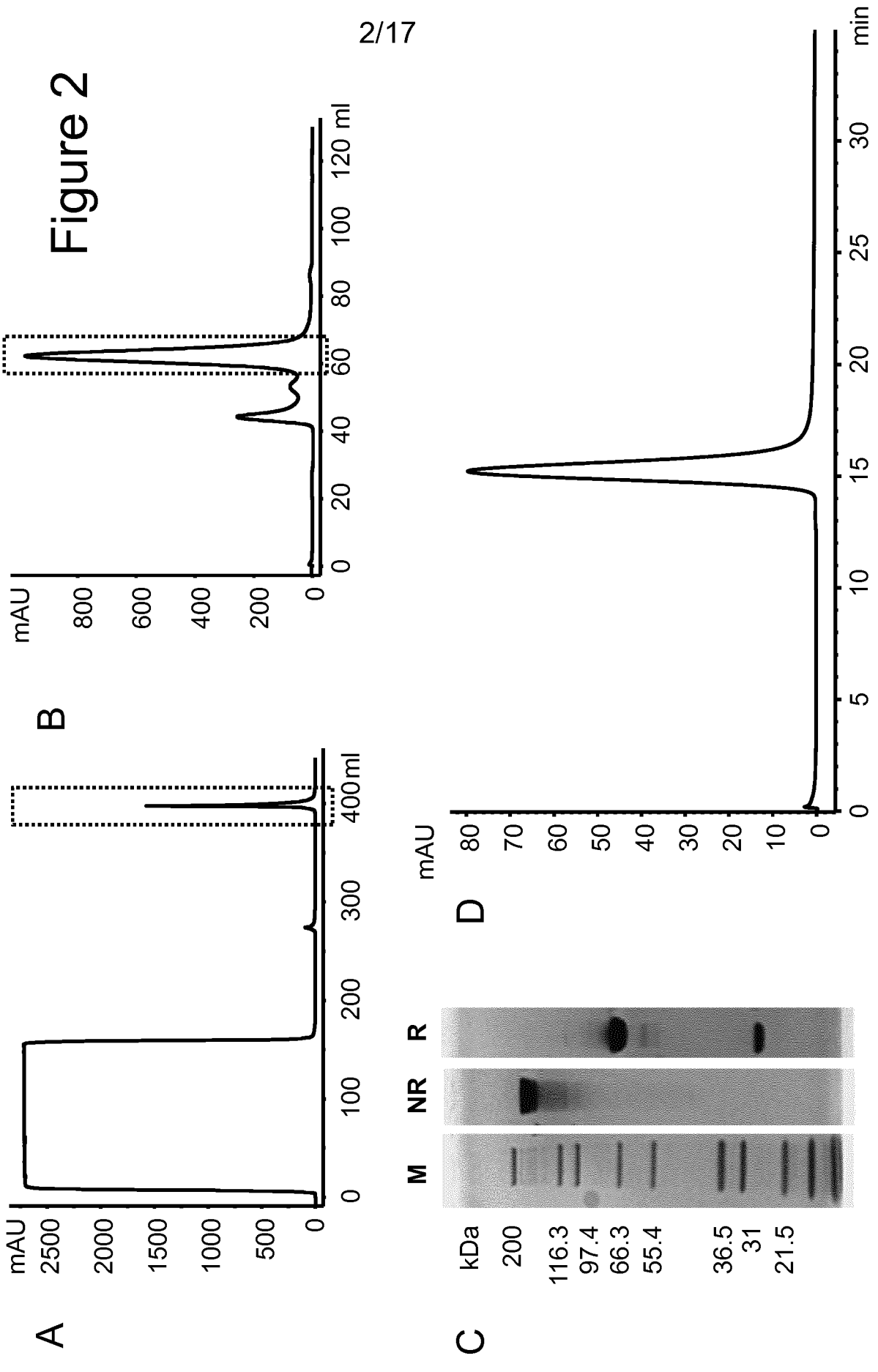

A
mAU
2500
2000
1500
1000
500
0
0
100
200
300
400ml
B
mAU
800
600
400
200
0
0
20
40
60
80
100
120 ml
C
kDa
200
116.3
97.4
66.3
55.4
36.5
31
21.5
M
NR
R
D
mAU
80
70
60
50
40
30
20
10
0
0
5
10
15
20
25
30
min

Figure 3

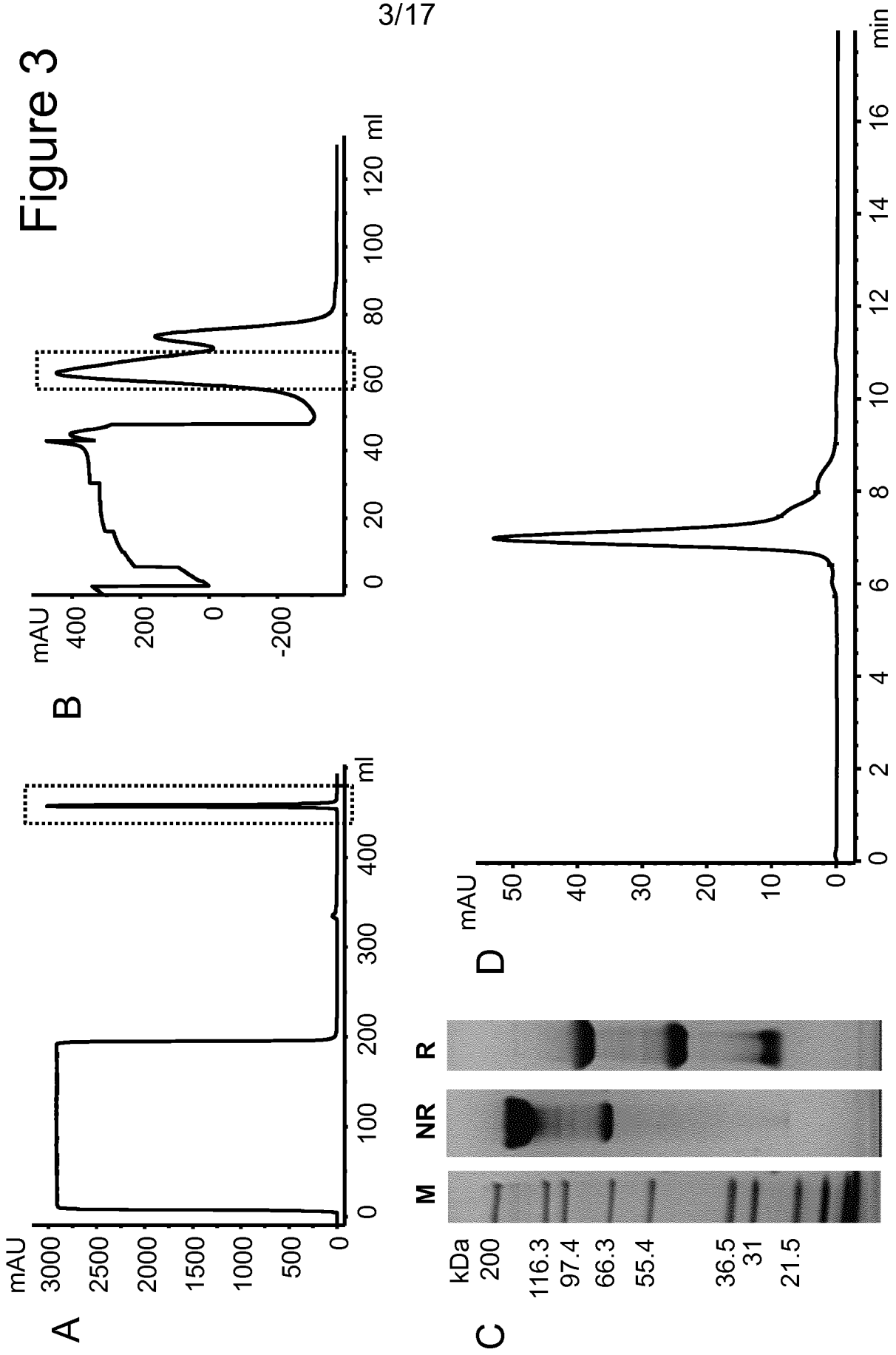
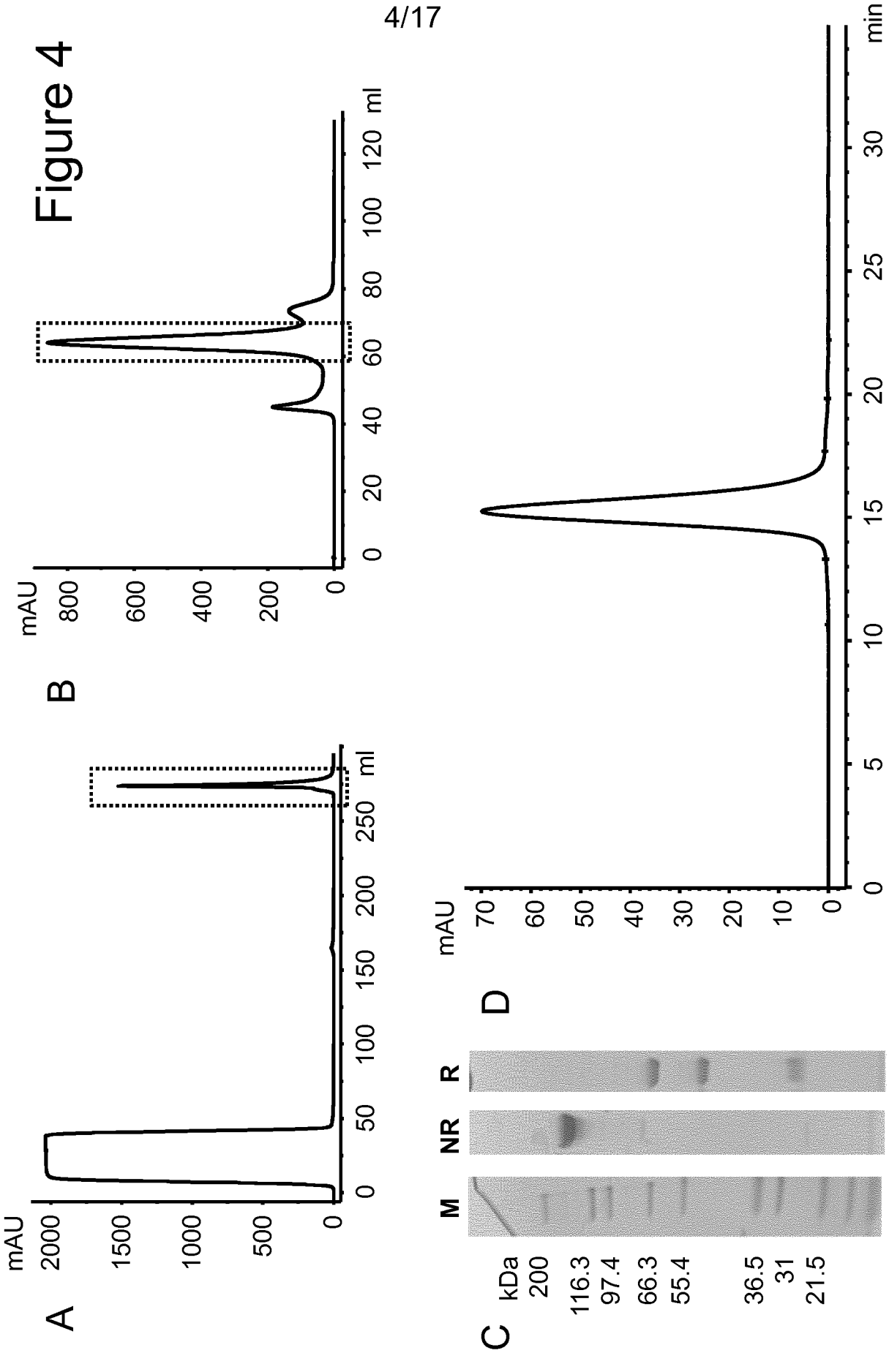

A
mAU
3000
2500
2000
1500
1000
500
0
0
100
200
300
400
ml
B
mAU
400
200
0
-200
0
20
40
60
80
100
120
ml
C
kDa
200
116.3
97.4
66.3
55.4
36.5
31
21.5
M
NR
R
D
mAU
50
40
30
20
10
0
0
2
4
6
8
10
12
14
16
min

Figure 4


A
mAU
2000
1500
1000
500
0
0
50
100
150
200
250
ml
B
mAU
800
600
400
200
0
0
20
40
60
80
100
120
ml
C
kDa
200
116.3
97.4
66.3
55.4
36.5
31
21.5
M
NR
R
D
mAU
70
60
50
40
30
20
10
0
0
5
10
15
20
25
30
min

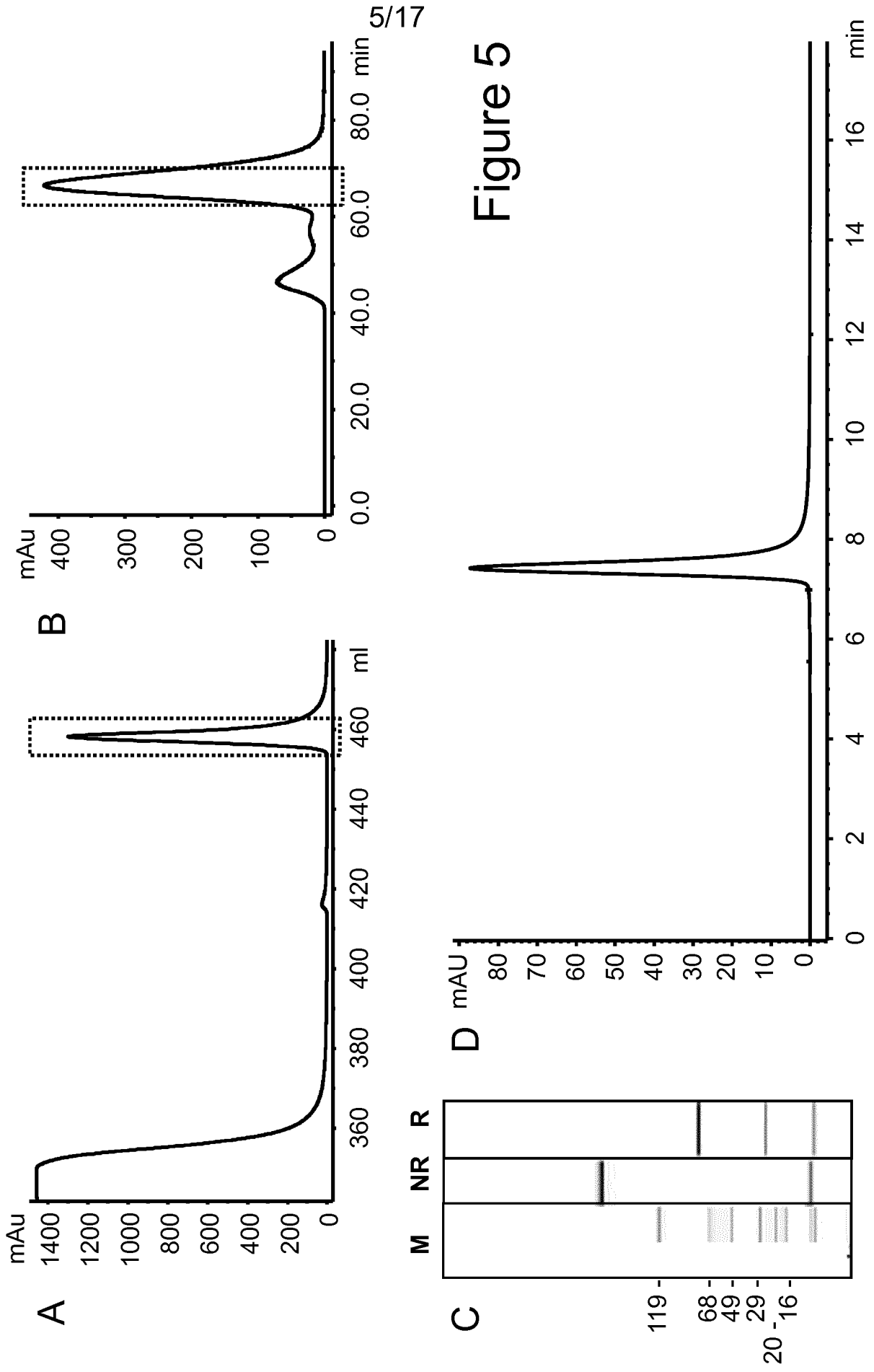
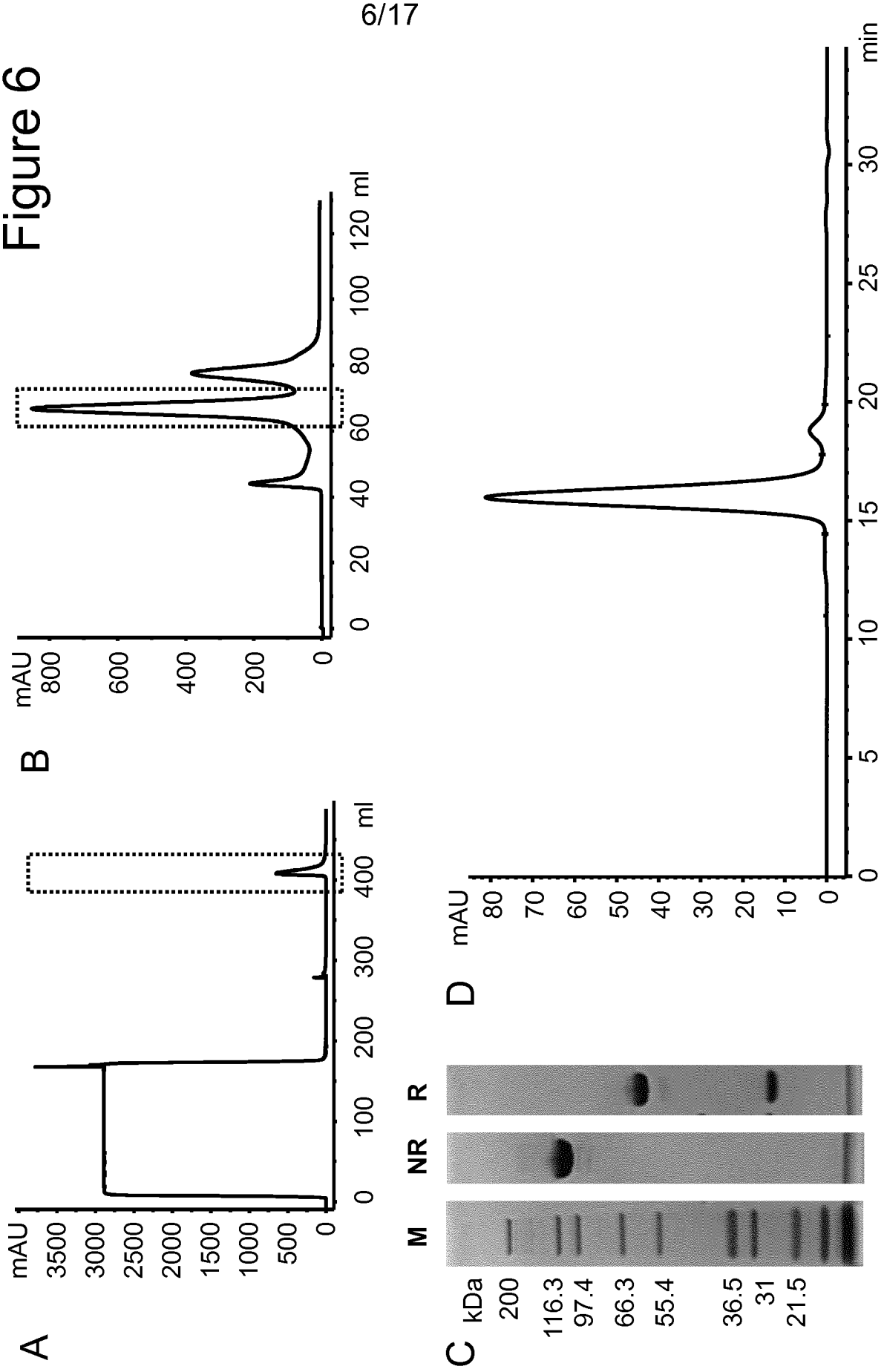

A
mAu
1400
1200
1000
800
600
400
200
0
360
380
400
420
440
460
ml
B
mAu
400
300
200
100
0
0.0
20.0
40.0
60.0
80.0
min
C
M
NR
R
119
68
49
29
20
16
D
mAU
80
70
60
50
40
30
20
10
0
0
2
4
6
8
10
12
14
16
min

Figure 5

Figure 6


A
mAU
3500
3000
2500
2000
1500
1000
500
0
0
100
200
300
400
ml
B
mAU
800
600
400
200
0
0
20
40
60
80
100
120
ml
C
kDa
200
116.3
97.4
66.3
55.4
36.5
31
21.5
M
NR
R
D
mAU
80
70
60
50
40
30
20
10
0
0
5
10
15
20
25
30
min

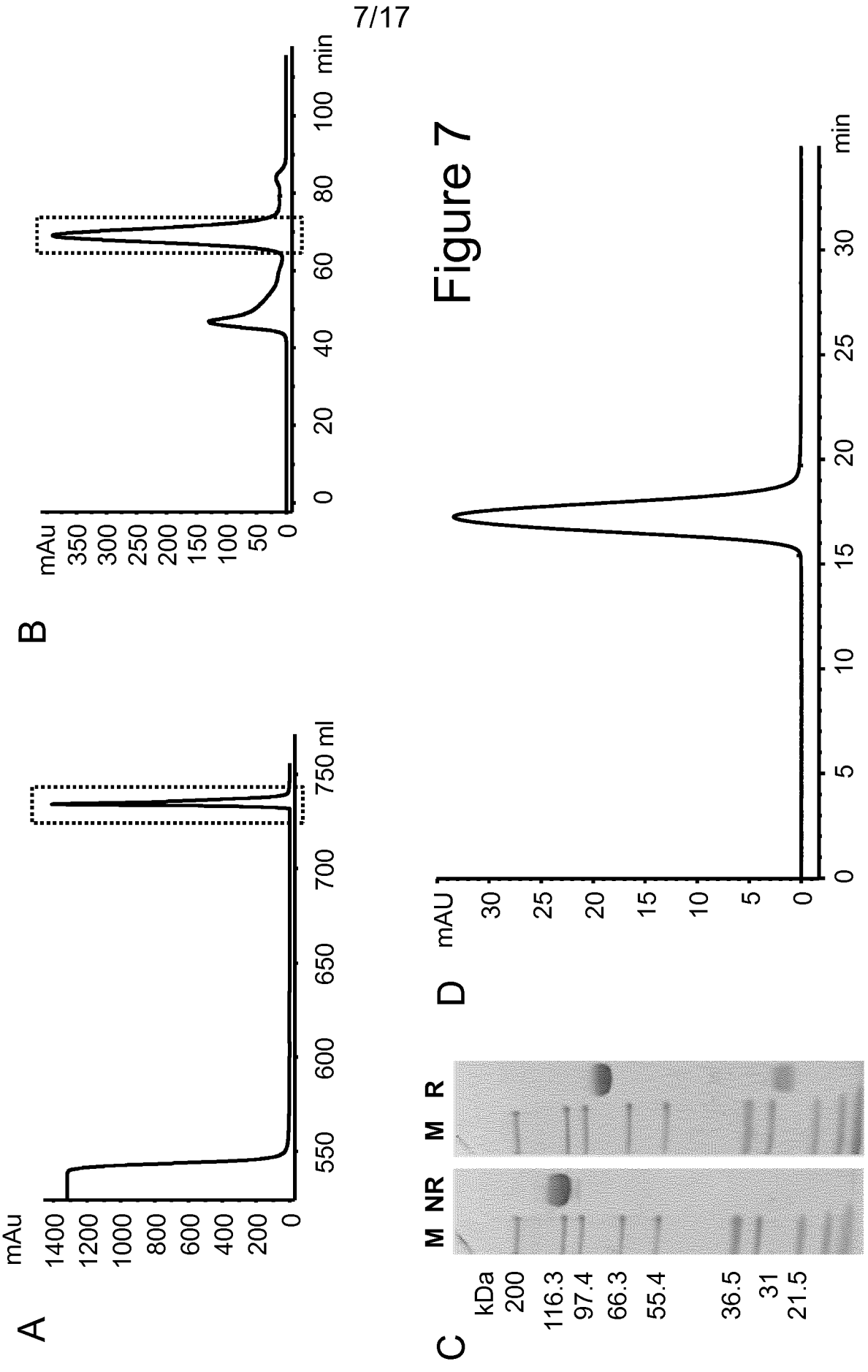

A
mAu
1400
1200
1000
800
600
400
200
0
550
600
650
700
750 ml
B
mAu
350
300
250
200
150
100
50
0
0
20
40
60
80
100 min
C
kDa
200
116.3
97.4
66.3
55.4
36.5
31
21.5
M NR
M R
D
mAU
30
25
20
15
10
5
0
0
5
10
15
20
25
30
min

Figure 7

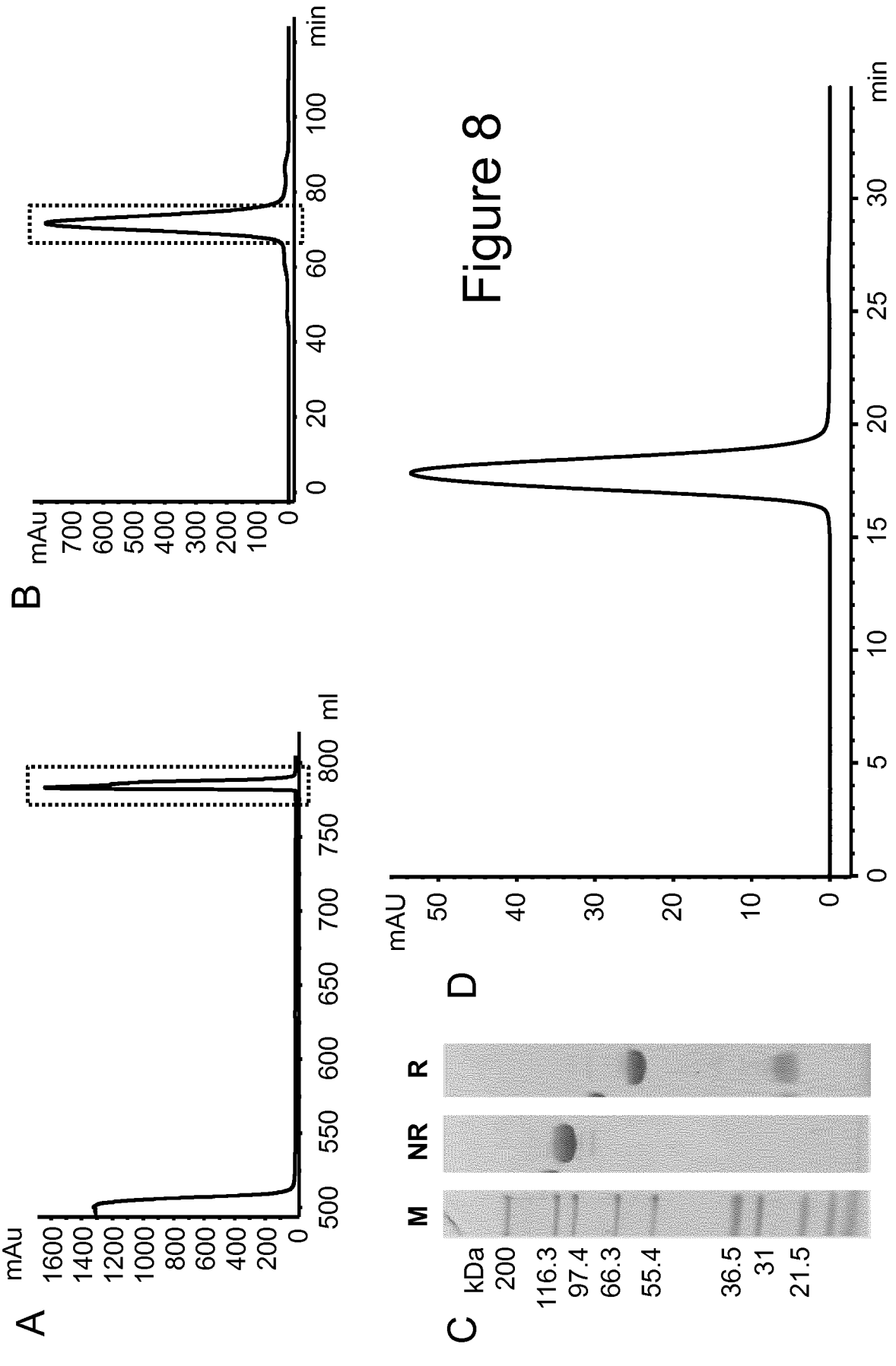

A
mAu
1600
1400
1200
1000
800
600
400
200
0
500
550
600
650
700
750
800
ml
B
mAu
700
600
500
400
300
200
100
0
0
20
40
60
80
100
min
C
kDa
200
116.3
97.4
66.3
55.4
36.5
31
21.5
M
NR
R
D
mAU
50
40
30
20
10
0
0
5
10
15
20
25
30
min

Figure 8

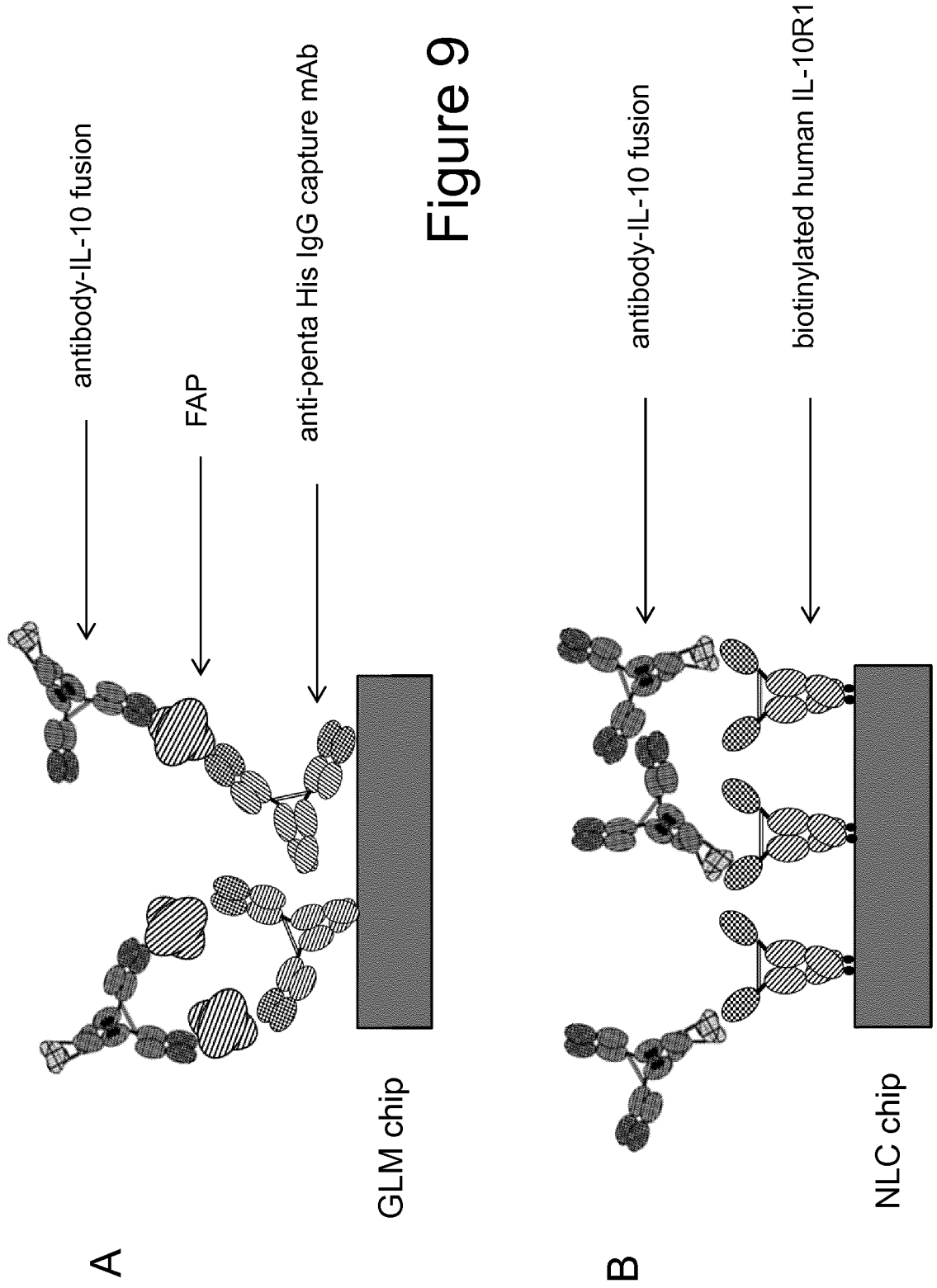

A
antibody-IL-10 fusion
FAP
anti-penta His IgG capture mAb
GLM chip
B
antibody-IL-10 fusion
biotinylated human IL-10R1
NLC chip

Figure 9

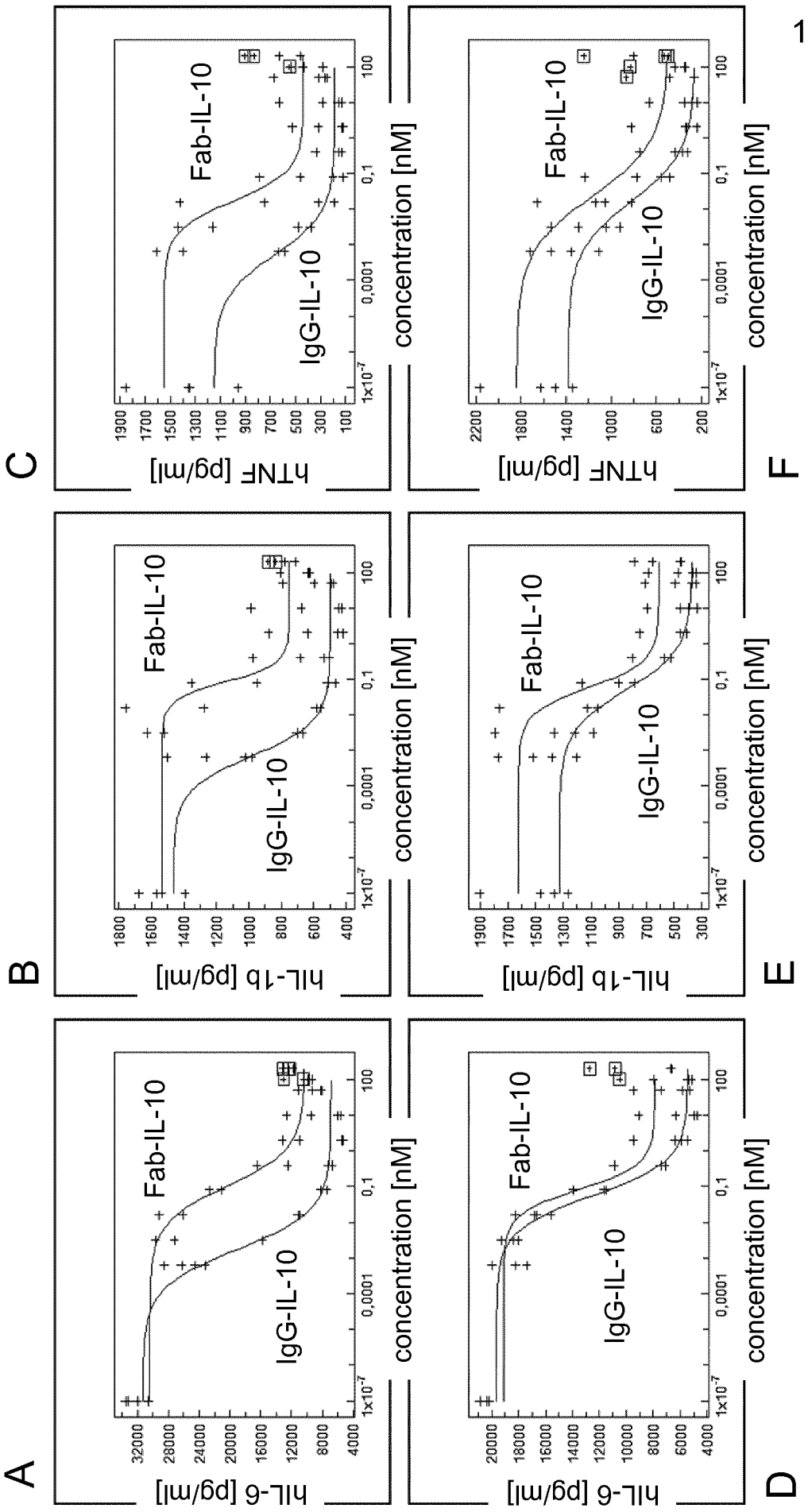

A
B
C
D
E
F
hIL-6 [pg/ml]
hIL-1b [pg/ml]
hTNF [pg/ml]
Fab-IL-10
IgG-IL-10
concentration [nM]

Figure 10

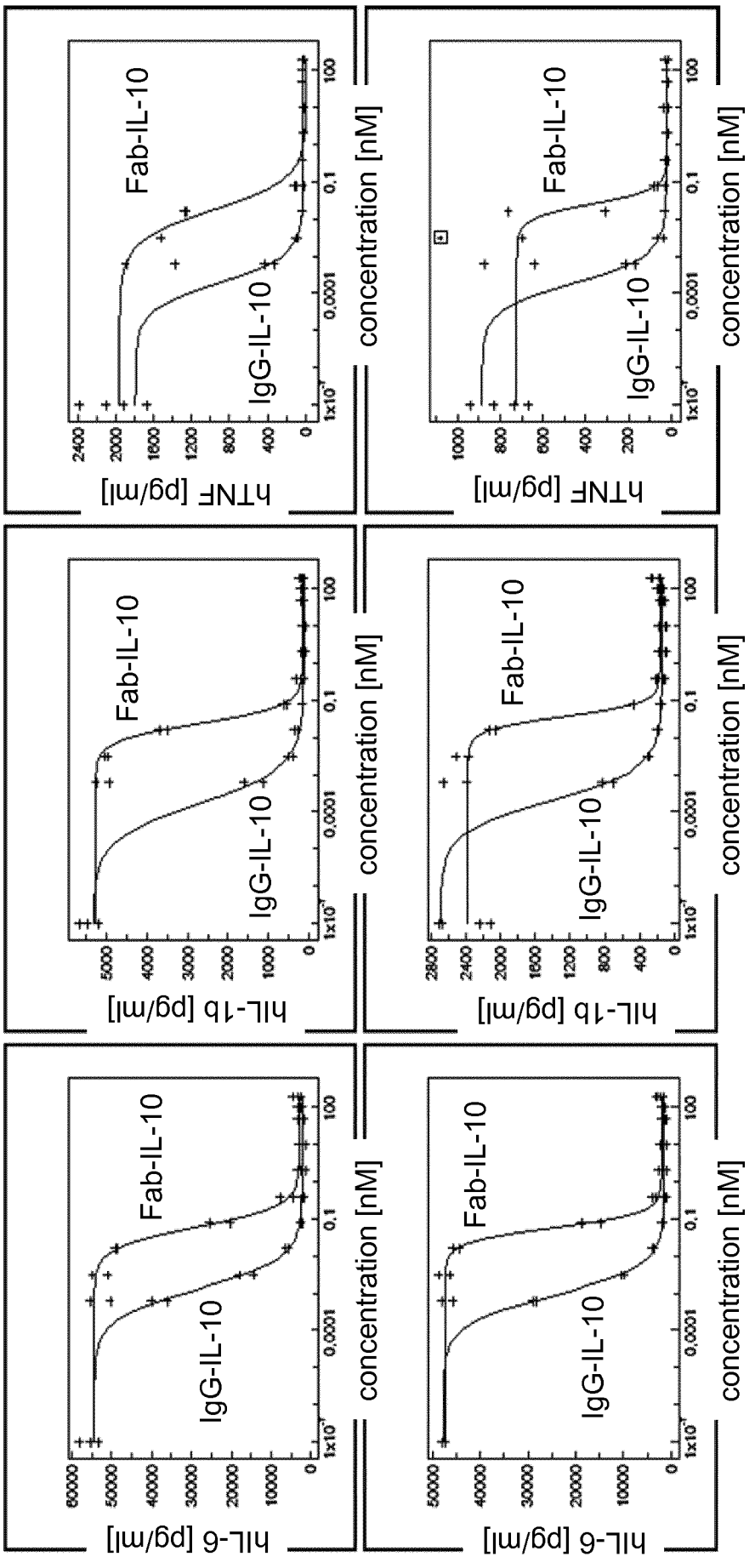

hIL-6 [pg/ml]
hIL-1b [pg/ml]
hTNF [pg/ml]
Fab-IL-10
IgG-IL-10
concentration [nM]

Figure 11A

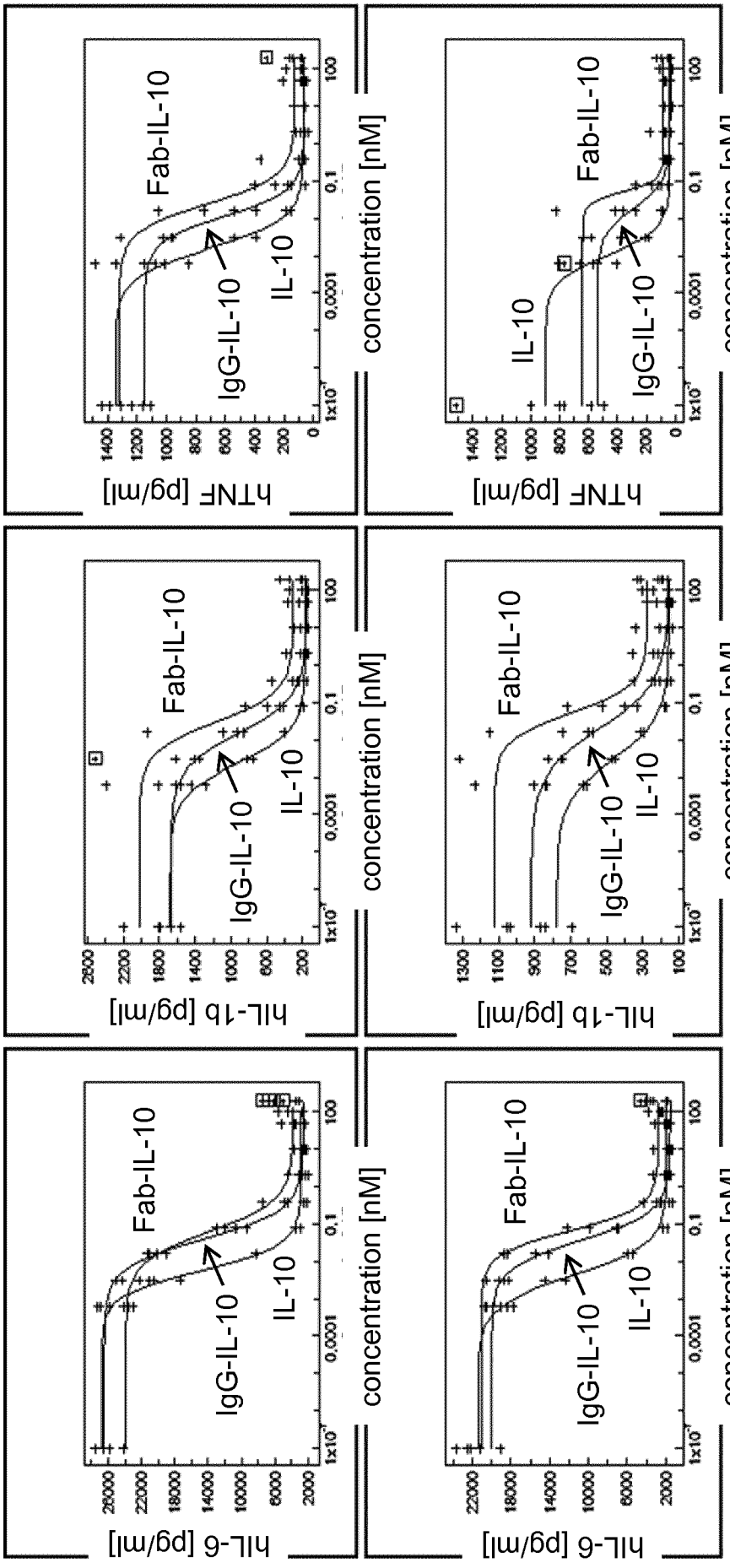

hIL-6 [pg/ml]
hIL-1b [pg/ml]
hTNF [pg/ml]
Fab-IL-10
IgG-IL-10
IL-10
concentration [nM]

Figure 11B

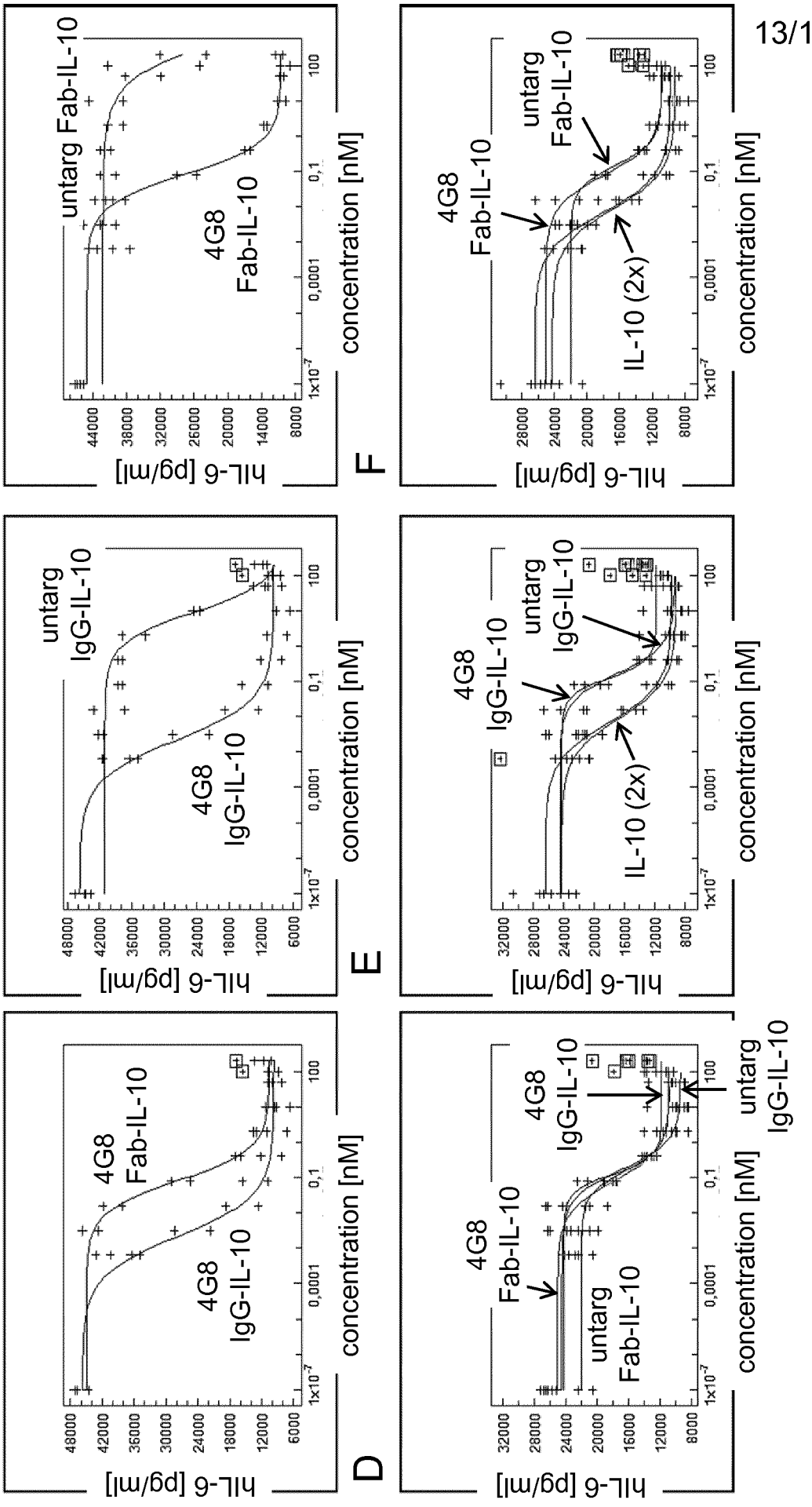

A
B
C
D
E
F
hIL-6 [pg/ml]
concentration [nM]
4G8
Fab-IL-10
4G8
IgG-IL-10
untarg
IgG-IL-10
untarg Fab-IL-10
IL-10 (2x)

Figure 12

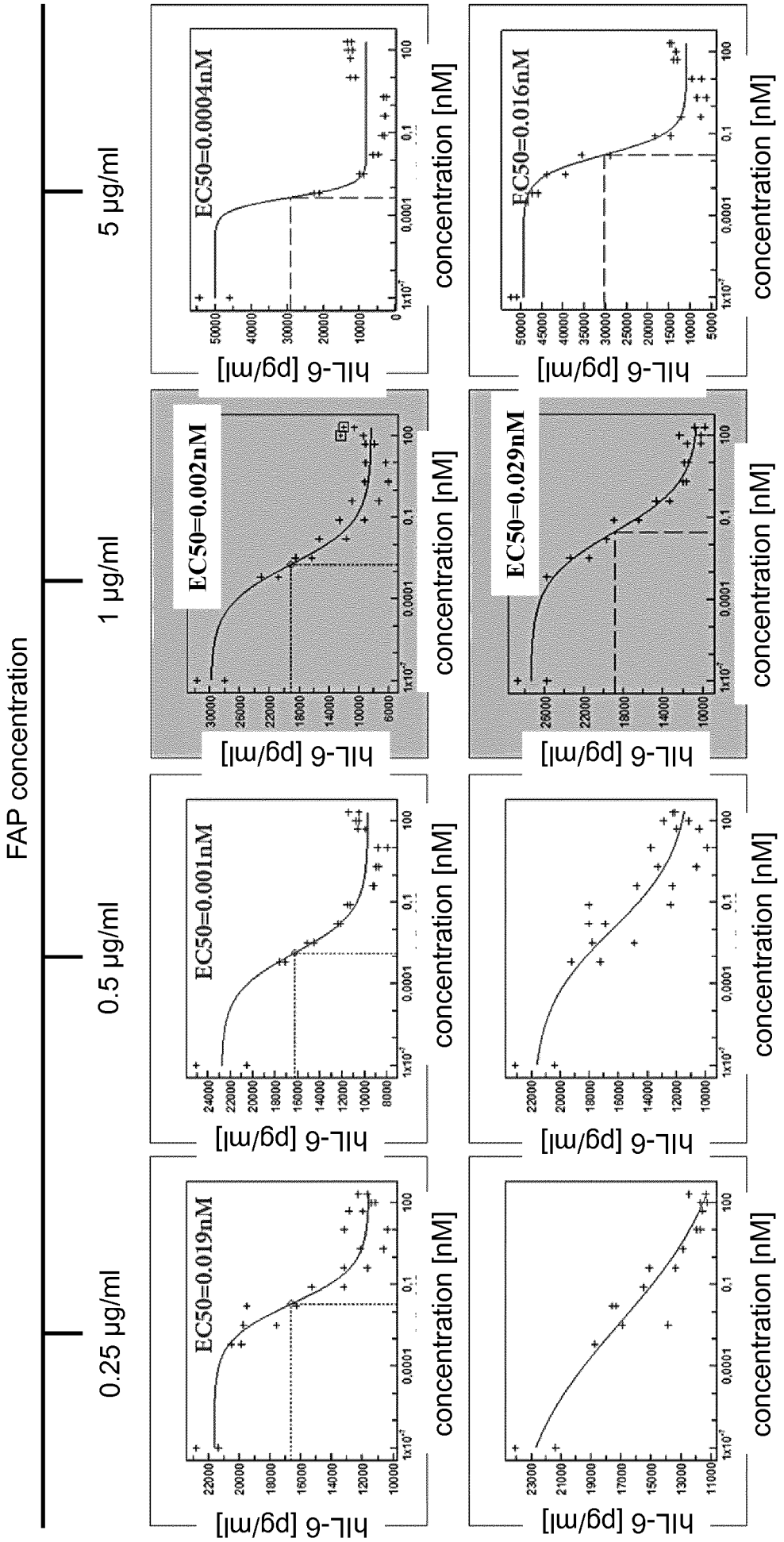

FAP concentration
0.25 µg/ml
0.5 µg/ml
1 µg/ml
5 µg/ml
EC50=0.019nM
EC50=0.001nM
EC50=0.002nM
EC50=0.029nM
EC50=0.0004nM
EC50=0.016nM
hIL-6 [pg/ml]
concentration [nM]
standard set-up

Figure 13

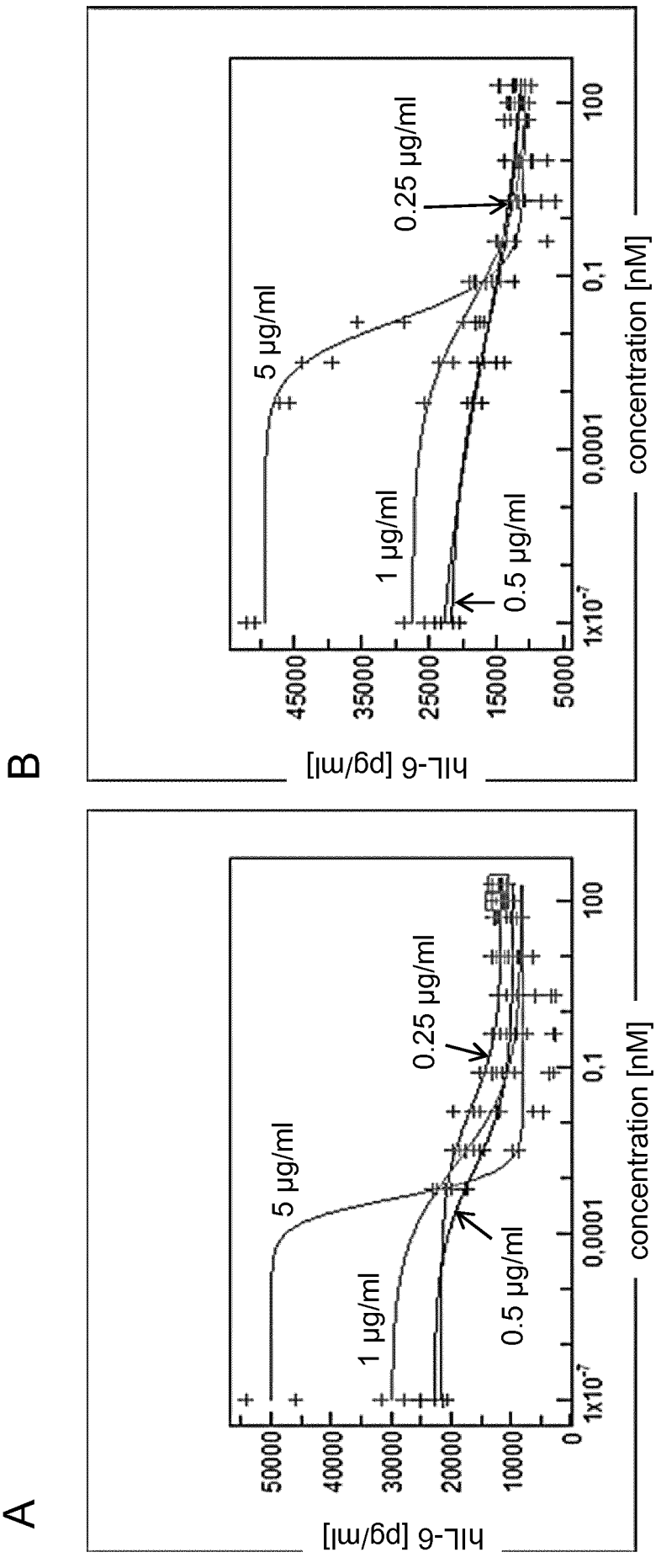

A
B
hIL-6 [pg/ml]
concentration [nM]
5 µg/ml
1 µg/ml
0.5 µg/ml
0.25 µg/ml
50000
40000
30000
20000
10000
0
45000
35000
25000
15000
5000
1x10⁻⁷
0,0001
0,1
100

Figure 14

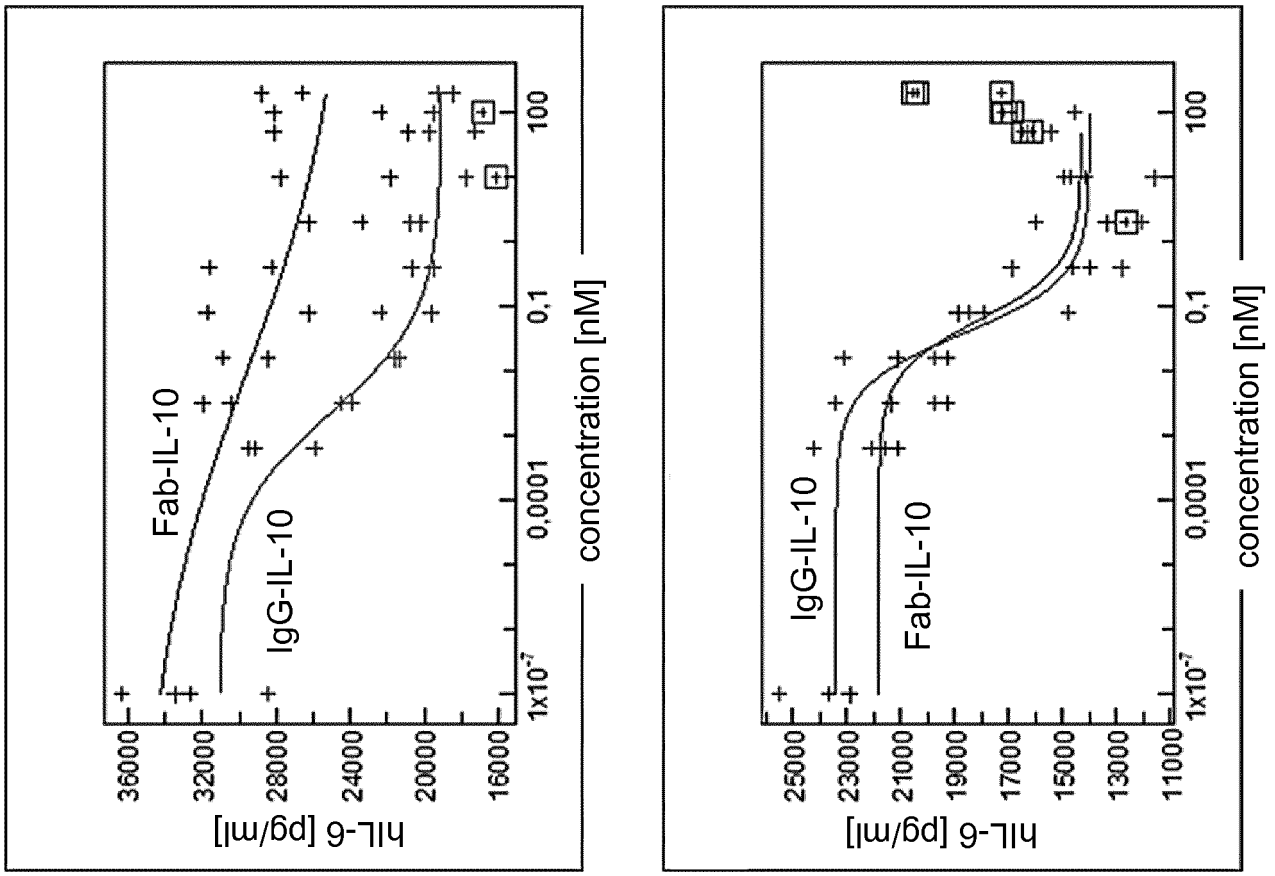

A
hIL-6 [pg/ml]
36000
32000
28000
24000
20000
16000
Fab-IL-10
IgG-IL-10
1x10-7
0,0001
0,1
100
concentration [nM]
B
hIL-6 [pg/ml]
25000
23000
21000
19000
17000
15000
13000
11000
IgG-IL-10
Fab-IL-10
1x10-7
0,0001
0,1
100
concentration [nM]

Figure 15

Figure 16

