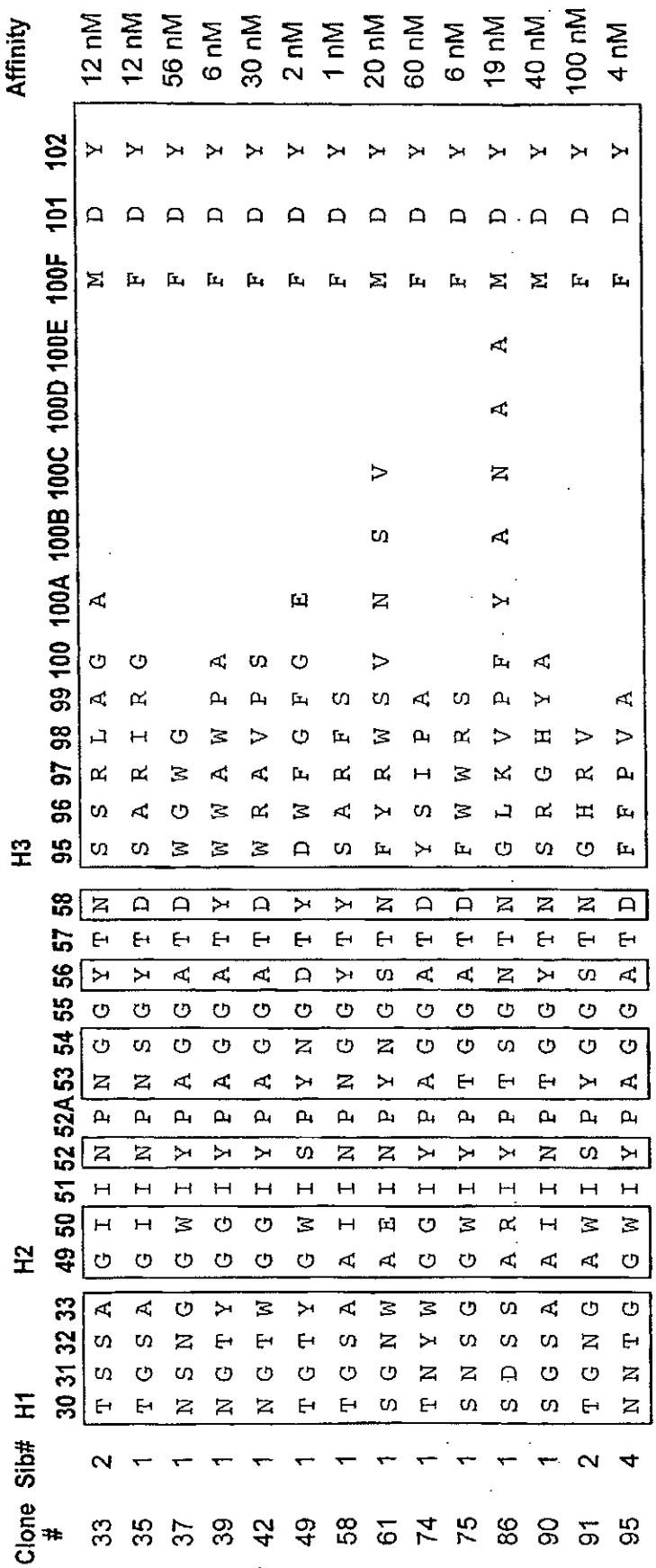

Clone #
Sib#
H1
30 31 32 33
H2
49 50 51 52 52A 53 54 55 56 57 58
H3
95 96 97 98 99 100 100A 100B 100C 100D 100E 100F 101 102
Affinity
33 2 T S S A G I I N P N G G Y T N S R L A G A M D Y 12 nM
35 1 T G S A G I I N P N S G Y T D S A R I R G F D Y 12 nM
37 1 N S N G G W I Y P A G G A T D W G W G F D Y 56 nM
39 1 N G T Y G G I Y P A G G A T Y W A W P A F D Y 6 nM
42 1 N G T W G G I Y P A G G A T D W R A V P S F D Y 30 nM
49 1 T G T Y G W I S P Y N G D T Y D W F G F G E F D Y 2 nM
58 1 T G S A A I I N P N G G Y T Y S A R F S F D Y 1 nM
61 1 S G N W A E I N P Y N G S T N F Y R W S V N S V M D Y 20 nM
74 1 T N Y W G G I Y P A G G A T D Y S I P P A F D Y 60 nM
75 1 S N S G G W I Y P T G G A T D F W W R R S F D Y 6 nM
86 1 S D S S A R I Y P P T S G N T N G L K V P F Y A A M D Y 19 nM
90 1 S G S A A I I N P T G G Y T N S R G H Y Y A M D Y 40 nM
91 2 T G N G A W I S P Y G G S T N G H R V F D Y 100 nM
95 4 N N T G G W I Y P A G G A T D F F P V A F D Y 4 nM

FIG. 1A

Clone #	H1												
	26	27	28	29	30	31	32	33	34	35	SEQ ID NO:
33	G	F	N	I	T	S	S	A	I	H	71
35	G	F	N	I	T	G	S	A	I	H	72
37	G	F	N	I	N	S	N	G	I	H	73
39	G	F	N	I	N	G	T	Y	I	H	74
42	G	F	N	I	N	G	T	W	I	H	75
49	G	F	N	I	T	G	T	Y	I	H	76
58	G	F	N	I	T	G	S	A	I	H	77
61	G	F	N	I	S	G	N	W	I	H	78
74	G	F	N	I	T	N	Y	W	I	H	79
75	G	F	N	I	S	N	S	G	I	H	80
86	G	F	N	I	S	D	S	S	I	H	81
90	G	F	N	I	S	G	S	A	I	H	82
91	G	F	N	I	T	G	N	G	I	H	83
95	G	F	N	I	N	N	T	G	I	H	84

FIG. 1B

Clone #	H2																				SEQ ID NO:
	49	50	51	52	52a	53	54	55	56	57	58	59	60	61	62	63	64	65			
33	G	I	I	N	P	N	G	G	Y	T	N	Y	A	D	S	V	K	G			85
35	G	I	I	N	P	N	S	G	Y	T	D	Y	A	D	S	V	K	G			86
37	G	W	I	Y	P	A	G	G	A	T	D	Y	A	D	S	V	K	G			87
39	G	G	I	Y	P	A	G	G	A	T	Y	Y	A	D	S	V	K	G			88
42	G	G	I	Y	P	A	G	G	A	T	D	Y	A	D	S	V	K	G			89
49	G	W	I	S	P	Y	N	G	D	T	Y	Y	A	D	S	V	K	G			90
58	A	I	I	N	P	N	G	G	Y	T	Y	Y	A	D	S	V	K	G			91
61	A	E	I	N	P	Y	N	G	S	T	N	Y	A	D	S	V	K	G			92
74	G	G	I	Y	P	A	G	G	A	T	D	Y	A	D	S	V	K	G			93
75	G	W	I	Y	P	T	G	G	A	T	D	Y	A	D	S	V	K	G			94
86	A	R	I	Y	P	T	S	G	N	T	N	Y	A	D	S	V	K	G			95
90	A	I	I	N	P	T	G	G	Y	T	N	Y	A	D	S	V	K	G			96
91	A	W	I	S	P	Y	G	G	S	T	N	Y	A	D	S	V	K	G			97
95	G	W	I	Y	P	A	G	G	A	T	D	Y	A	D	S	V	K	G			98

FIG. 1C

Clone #	H3																							SEQ ID NO:
	93	94	95	96	97	98	99	100	a	b	c	d	e	f	101	102								
33	A	R	S	S	R	L	A	G	A					M	D	Y								99
35	A	R	S	A	R	I	R	G						F	D	Y								100
37	A	R	W	G	W	G								F	D	Y								101
39	A	R	W	W	A	W	P	A						F	D	Y								102
42	A	R	W	R	A	V	P	S						F	D	Y								103
49	A	R	D	W	F	G	F	G	E					F	D	Y								104
58	A	R	S	A	R	F	S							F	D	Y								105
61	A	R	F	Y	R	W	S	V	N	S	V			M	D	Y								106
74	A	R	Y	S	I	P	A							F	D	Y								107
75	A	R	F	W	W	R	S							F	D	Y								108
86	A	R	G	L	K	V	P	F	Y	A	N	A	A	M	D	Y								109
90	A	R	S	R	G	H	Y	A						M	D	Y								110
91	A	R	G	H	R	V								F	D	Y								111
95	A	R	F	F	P	V	A							F	D	Y								112

FIG. 1D

Framework Sequences of Light Chain

LC-FR1 1 Asp Ile Gln Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val Gly Asp Arg Val Thr Ile Thr 23 Cys (SEQ ID NO: 55)

LC-FR2 35 Trp Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys Leu Leu Ile 49 Tyr (SEQ ID NO: 56)

LC-FR3 57 Gly Val Pro Ser Arg Phe Ser Gly Ser Arg Ser Gly Thr Asp Phe Thr Leu Thr Ile Ser Ser Leu Gln Pro Glu Asp Phe Ala Thr Tyr Tyr 88 Cys (SEQ ID NO: 57)

LC-FR4 98 Phe Gly Gln Gly Thr Lys Val Glu Ile 107 Lys (SEQ ID NO: 58)

Framework Sequences of Heavy Chain

HC-FR1 1 Glu Val Gln Leu Val Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly Ser Leu Arg Leu Ser Cys Ala Ala 25 Ser (SEQ ID NO: 59)

HC-FR2 36 Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp 48 Val (SEQ ID NO: 60)

HC-FR3 66 Arg Phe Thr Ile Ser Ala Asp Thr Ser Lys Asn Thr Ala Tyr Leu Gln Met 83 Asn Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr 92 Cys (SEQ ID NO: 61)
83a 83b 83c

HC-FR4 103 Trp Gly Gln Gly Thr Leu Val Thr Val Ser 113 Ser (SEQ ID NO: 62)

FIG. 1E

Framework Sequences of Light Chain

LC-FR1 1 Asp Ile Gln Met Thr Gln Ser Pro Ser Ser Leu Ser Ala Ser Val
Gly Asp Arg Val Thr Ile Thr Cys 23 (SEQ ID NO: 63)

LC-FR2 35 Trp Tyr Gln Gln Lys Pro Gly Lys Ala Pro Lys Leu Leu Ile Tyr 49 (SEQ ID NO: 64)

LC-FR3 57 Gly Val Pro Ser Arg Phe Ser Gly Ser Gly Ser Gly Thr Asp Phe Thr Leu Thr Ile
Ser Ser Leu Gln Pro Glu Asp Phe Ala Thr Tyr Tyr Cys 88 (SEQ ID NO: 65)

LC-FR4 98 Phe Gly Gln Gly Thr Lys Val Glu Ile Lys 107 (SEQ ID NO: 66)

Framework Sequences of Heavy Chain

HC-FR1 1 Glu Val Gln Leu Val Glu Ser Gly Gly Gly Leu Val Gln Pro Gly Gly Ser Leu
Arg Leu Ser Cys Ala Ala Ser 25 (SEQ ID NO: 67)

HC-FR2 36 Trp Val Arg Gln Ala Pro Gly Lys Gly Leu Glu Trp Val 48 (SEQ ID NO: 68)

HC-FR3 66 Arg Phe Thr Ile Ser Arg Asp Asn Ser Lys Asn Thr Leu Tyr Leu Gln Met Asn 83
83a 83b 83c
Ser Leu Arg Ala Glu Asp Thr Ala Val Tyr Tyr Cys 92 (SEQ ID NO: 69)

HC-FR4 103 Trp Gly Gln Gly Thr Leu Val Thr Val Ser Ser 113 (SEQ ID NO: 70)

FIG. 1F

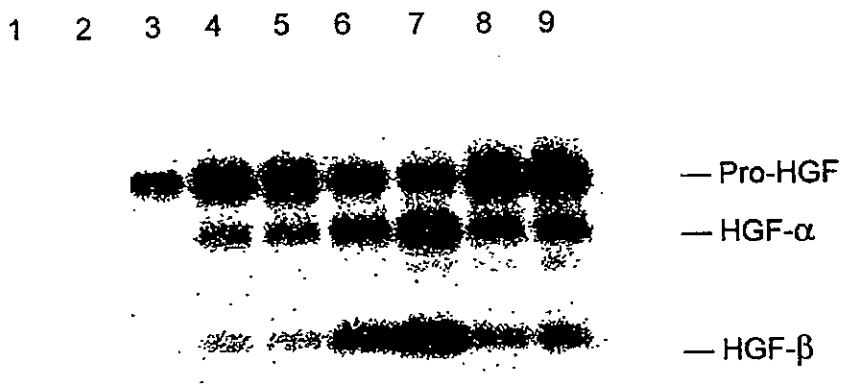


FIG. 2A

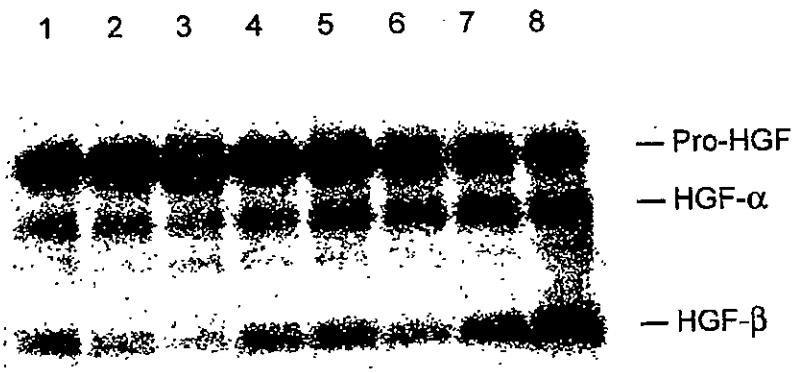


FIG. 2B

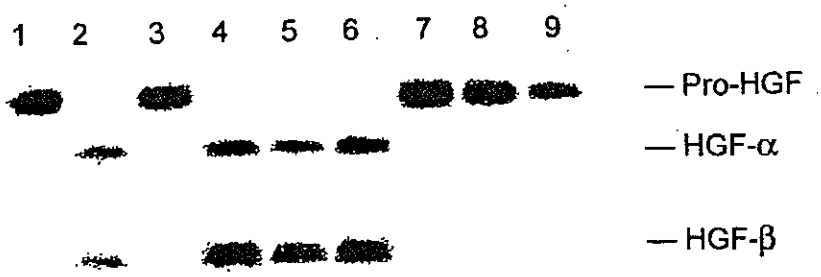


FIG. 3

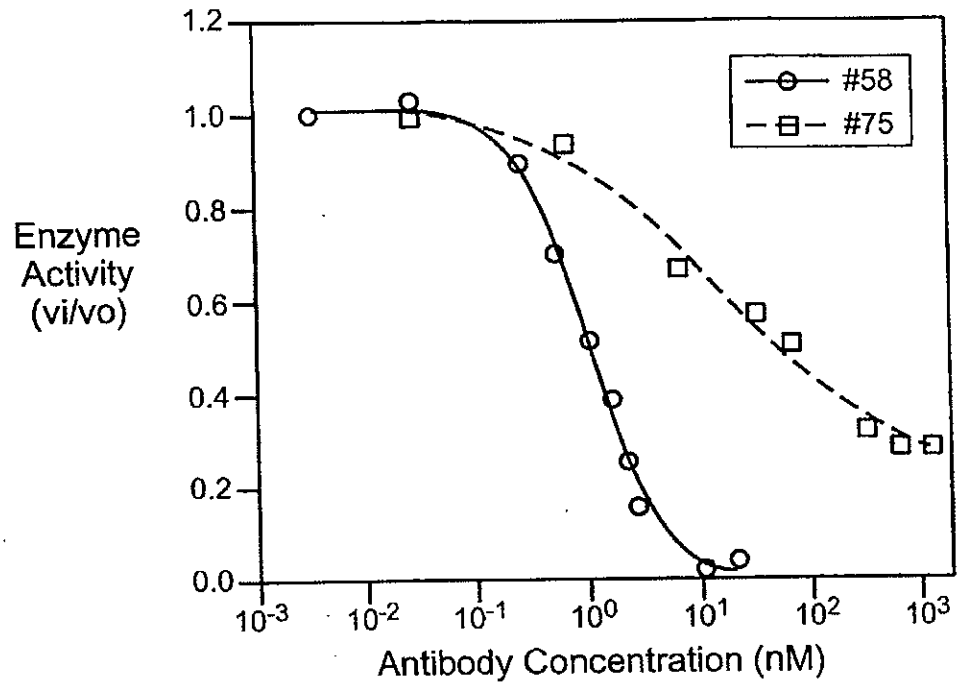

1.2
1.0
0.8
0.6
0.4
0.2
0.0
Enzyme Activity (vi/vo)
#58
#75
10^{-3}
10^{-2}
10^{-1}
10^{0}
10^{1}
10^{2}
10^{3}
Antibody Concentration (nM)

FIG. 4

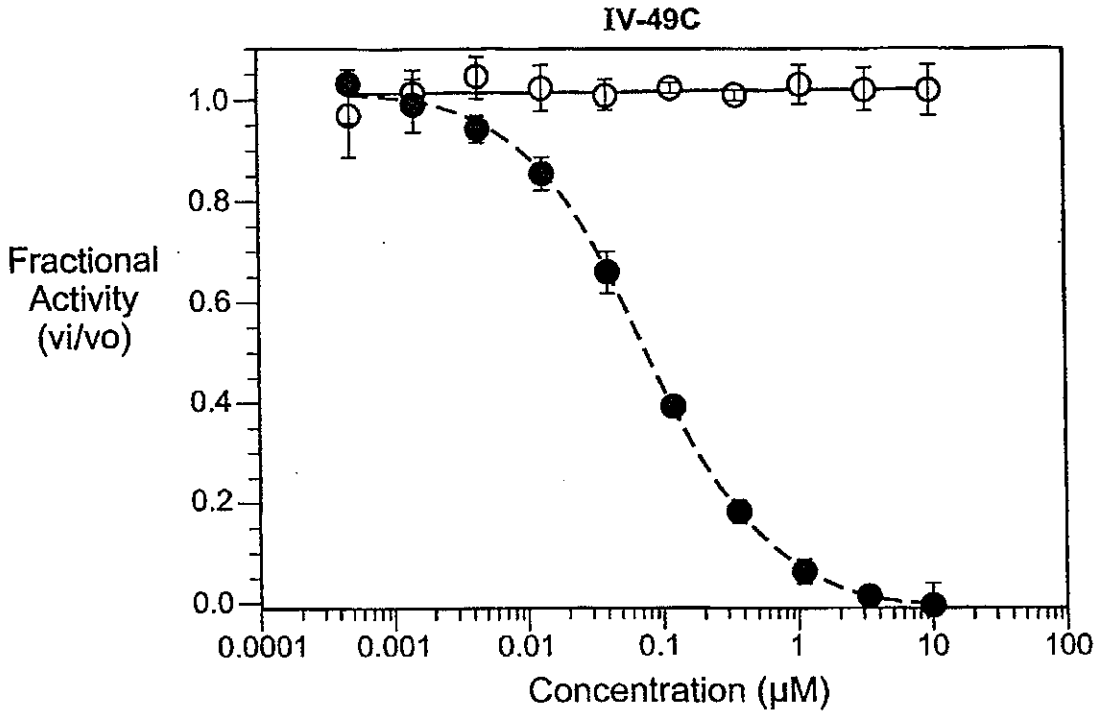

IV-49C
Fractional Activity (vi/vo)
1.0
0.8
0.6
0.4
0.2
0.0
0.0001
0.001
0.01
0.1
1
10
100
Concentration (μM)

FIG. 5A

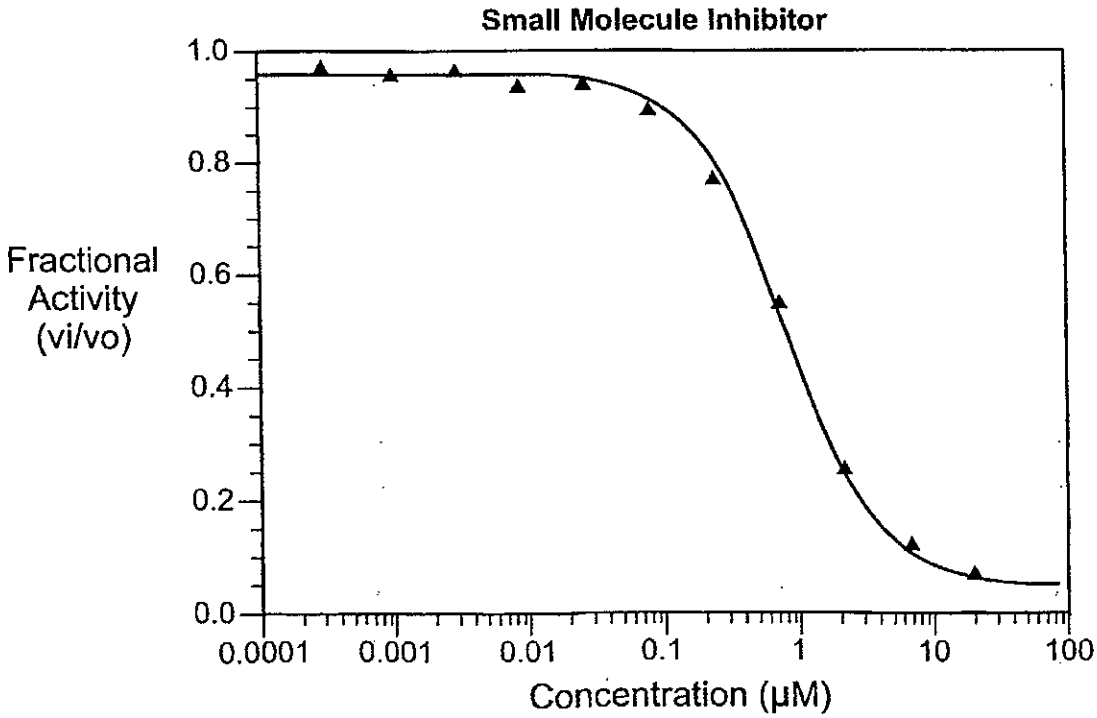

Small Molecule Inhibitor
Fractional Activity (vi/vo)
1.0
0.8
0.6
0.4
0.2
0.0
0.0001
0.001
0.01
0.1
1
10
100
Concentration (μM)

FIG. 5B

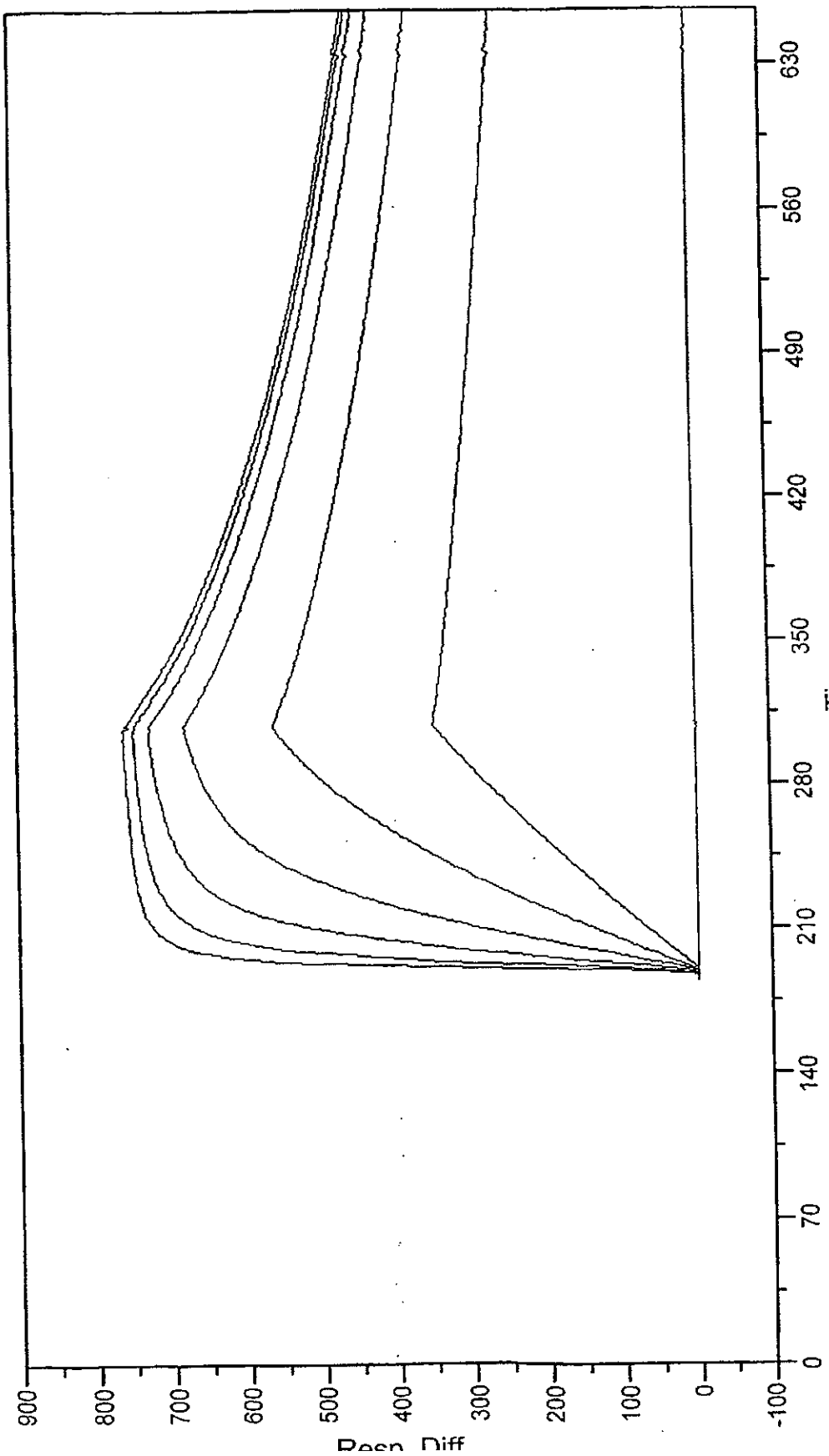

900
800
700
600
500
400
300
200
100
0
-100
Resp Diff
0
70
140
210
280
350
420
490
560
630
Time

FIG. 6A

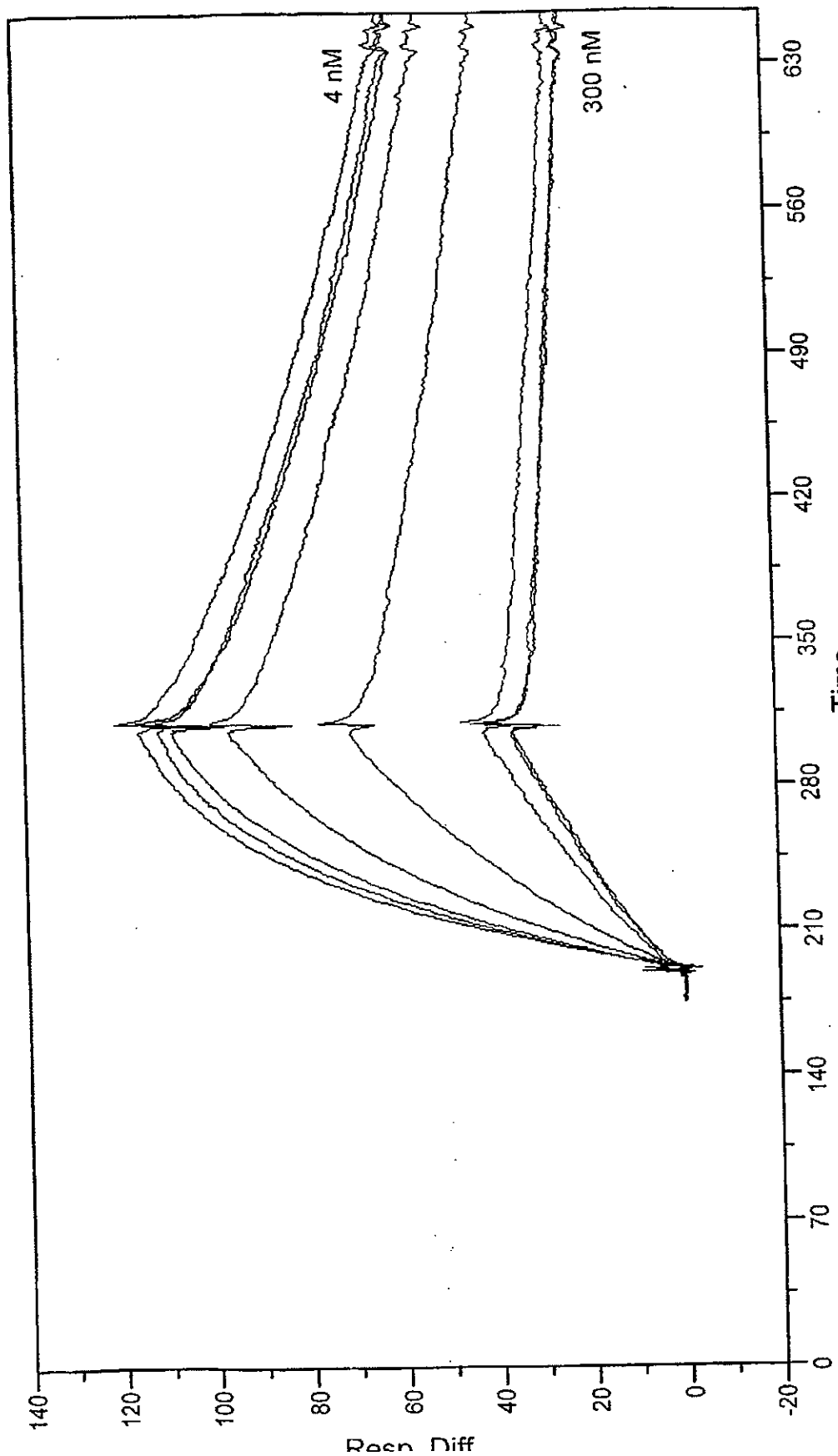

4 nM
300 nM
Time
Resp. Diff.
0
70
140
210
280
350
420
490
560
630
-20
0
20
40
60
80
100
120
140

FIG. 6B

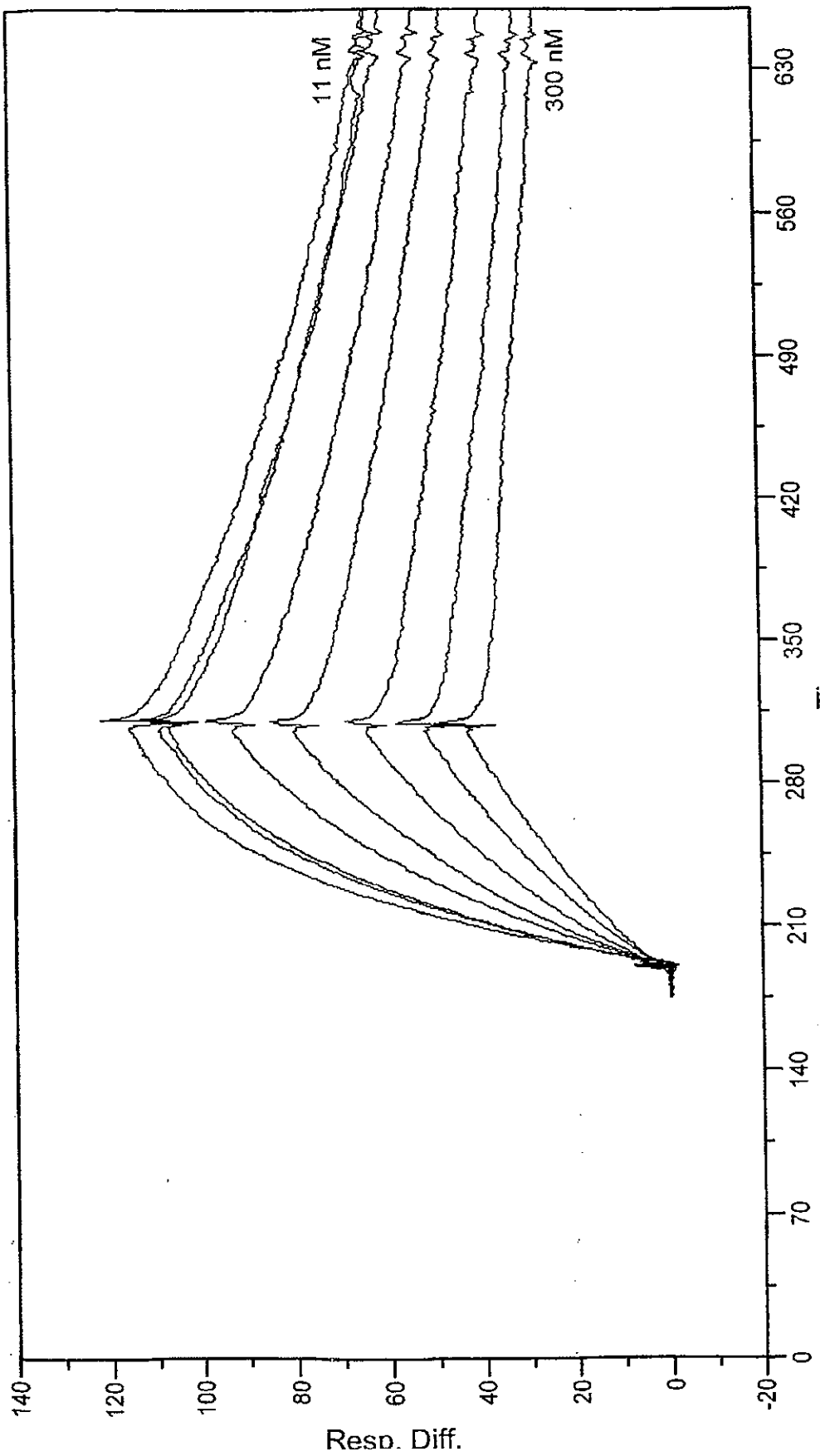

11 nM
300 nM
Resp. Diff.
Time
140
120
100
80
60
40
20
0
-20
0
70
140
210
280
350
420
490
560
630

FIG. 6C

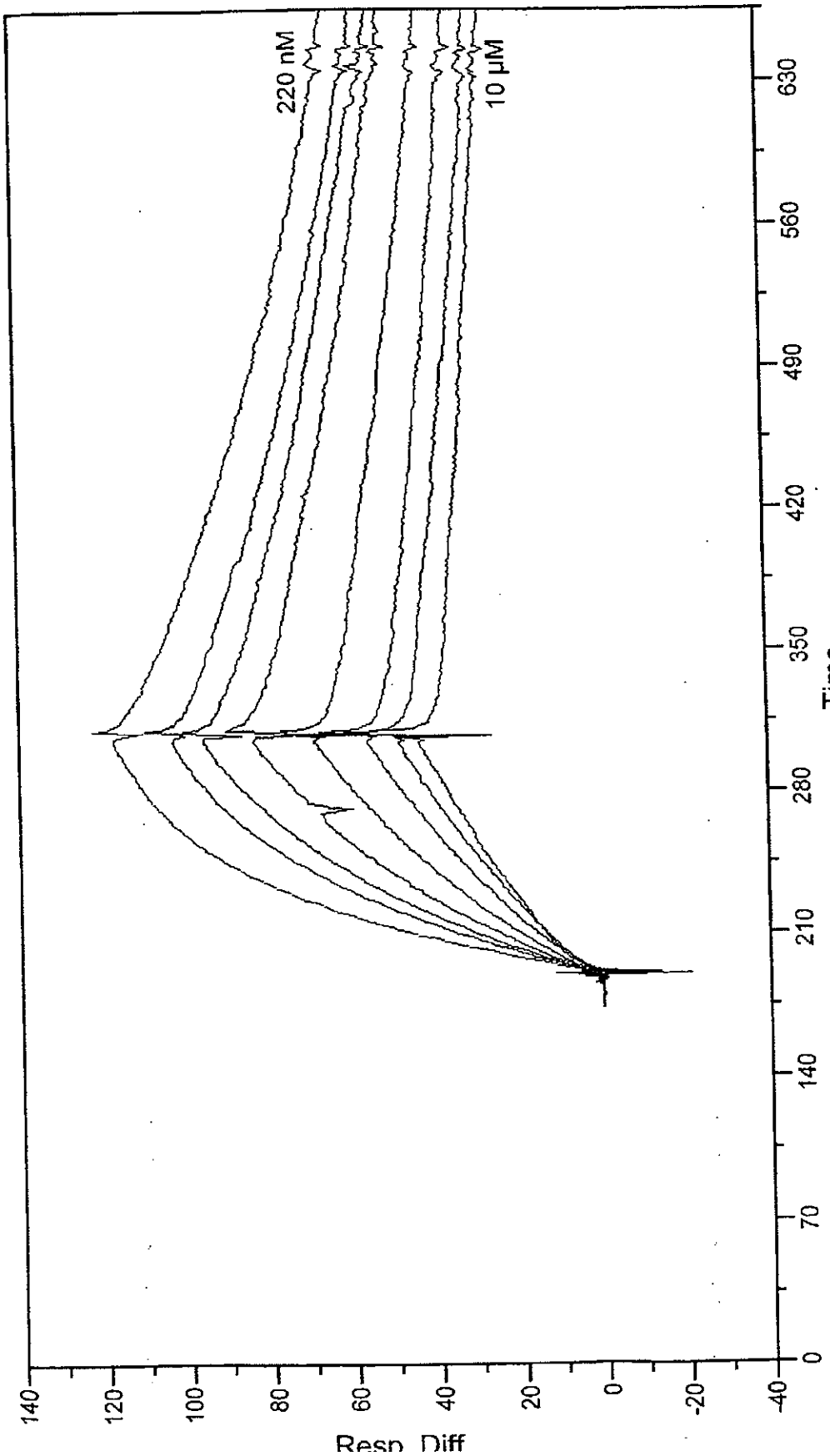

220 nM
10 µM
Resp. Diff
Time
140
120
100
80
60
40
20
0
-20
-40
0
70
140
210
280
350
420
490
560
630

FIG. 6D

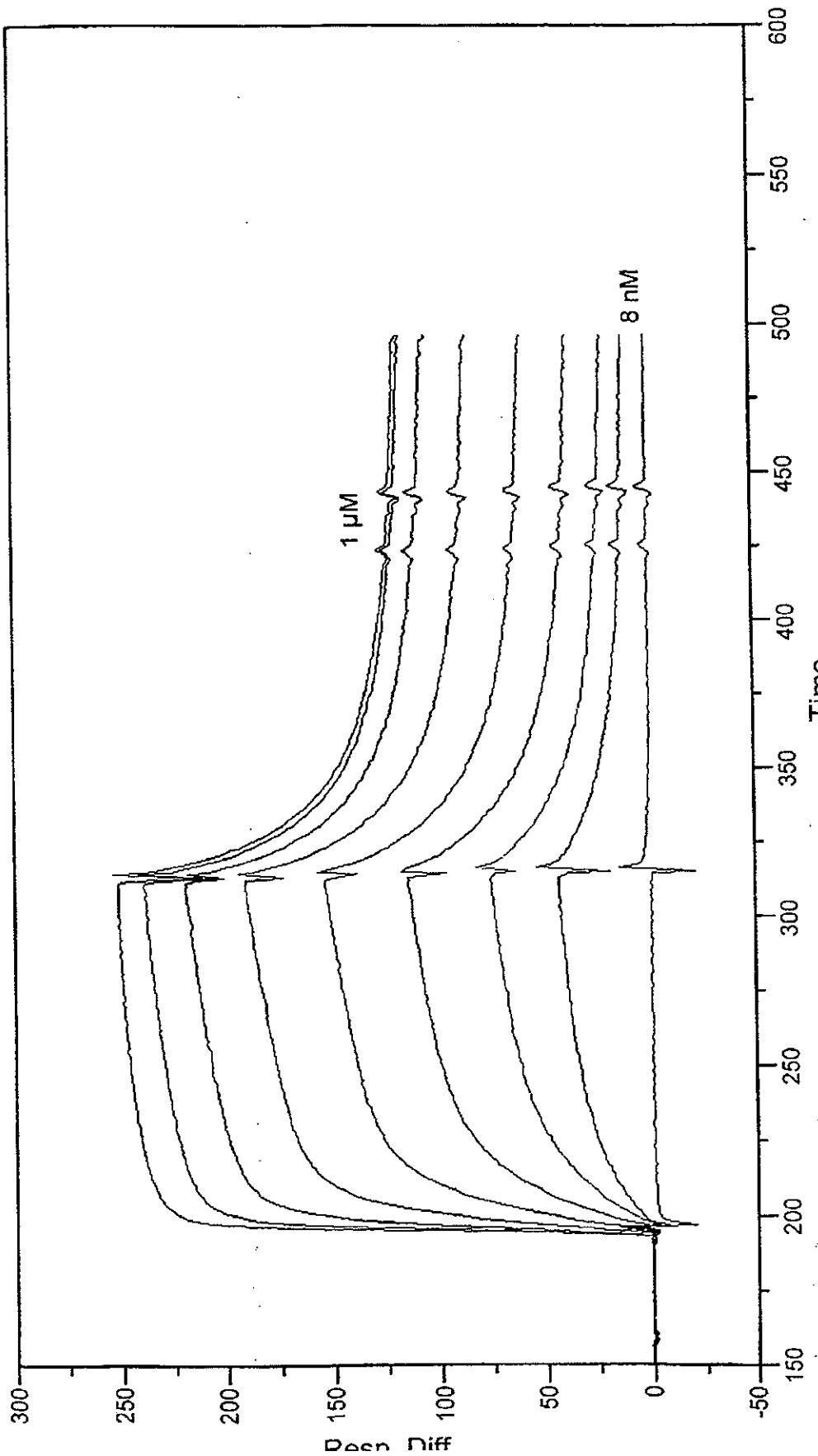

1 µM
8 nM
Time
Resp. Diff.
600
550
500
450
400
350
300
250
200
150
-50
0
50
100
150
200
250
300

FIG. 6E

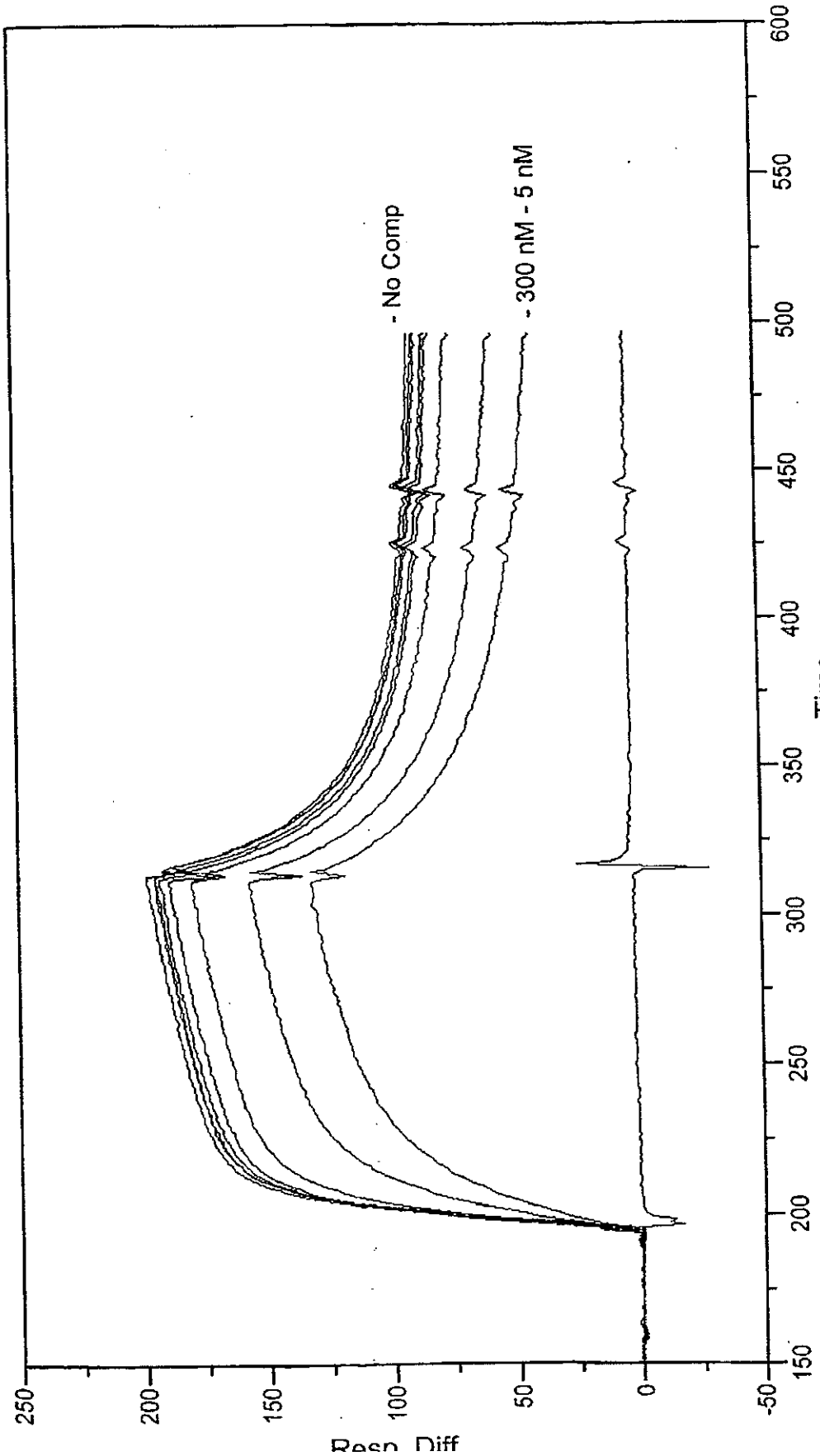

- No Comp
- 300 nM - 5 nM
Time
Resp. Diff
250
200
150
100
50
0
-50
150
200
250
300
350
400
450
500
550
600

FIG. 6F

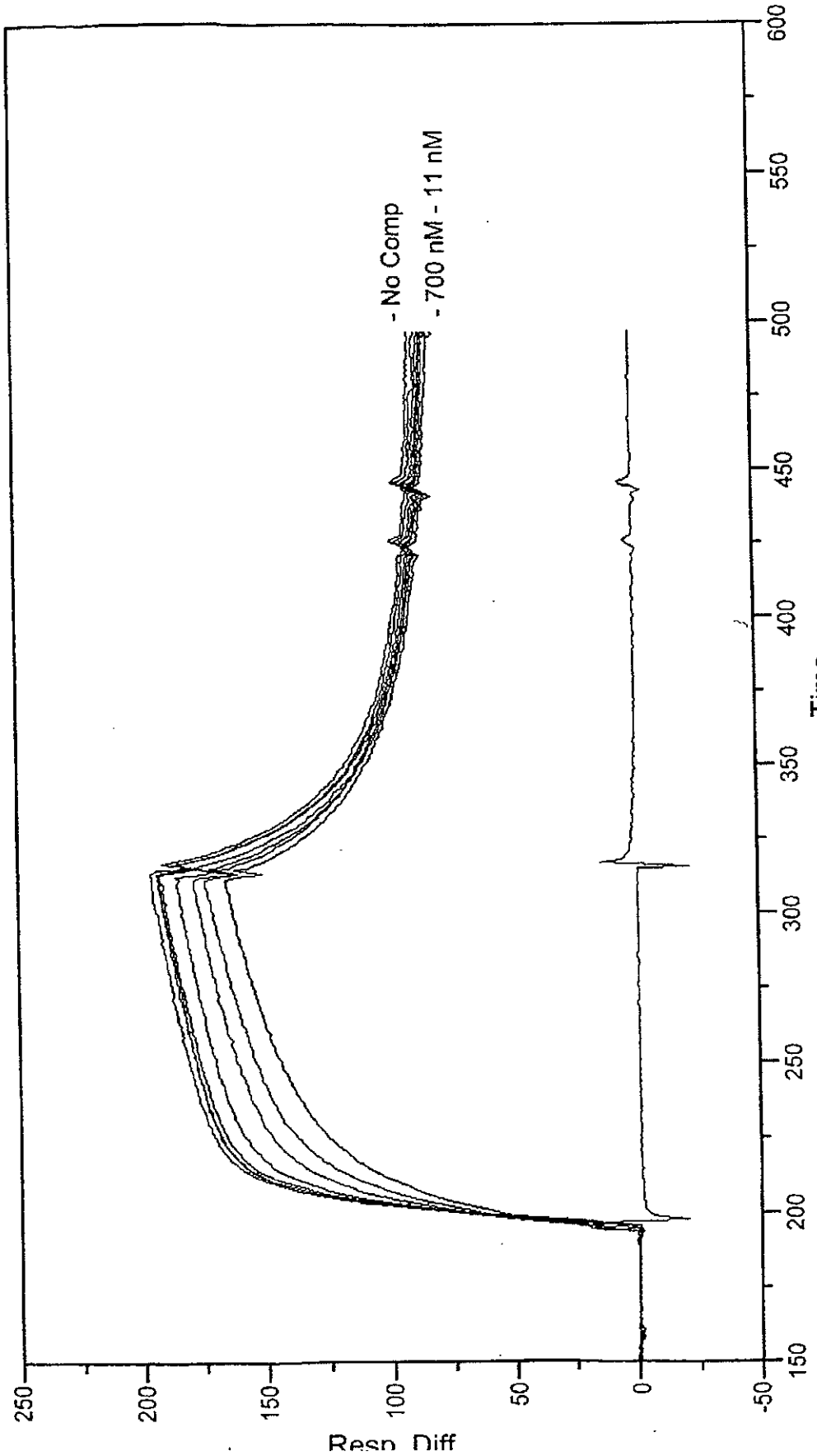

- No Comp
- 700 nM - 11 nM
Resp Diff
Time
250
200
150
100
50
0
-50
150
200
250
300
350
400
450
500
550
600

FIG. 6G

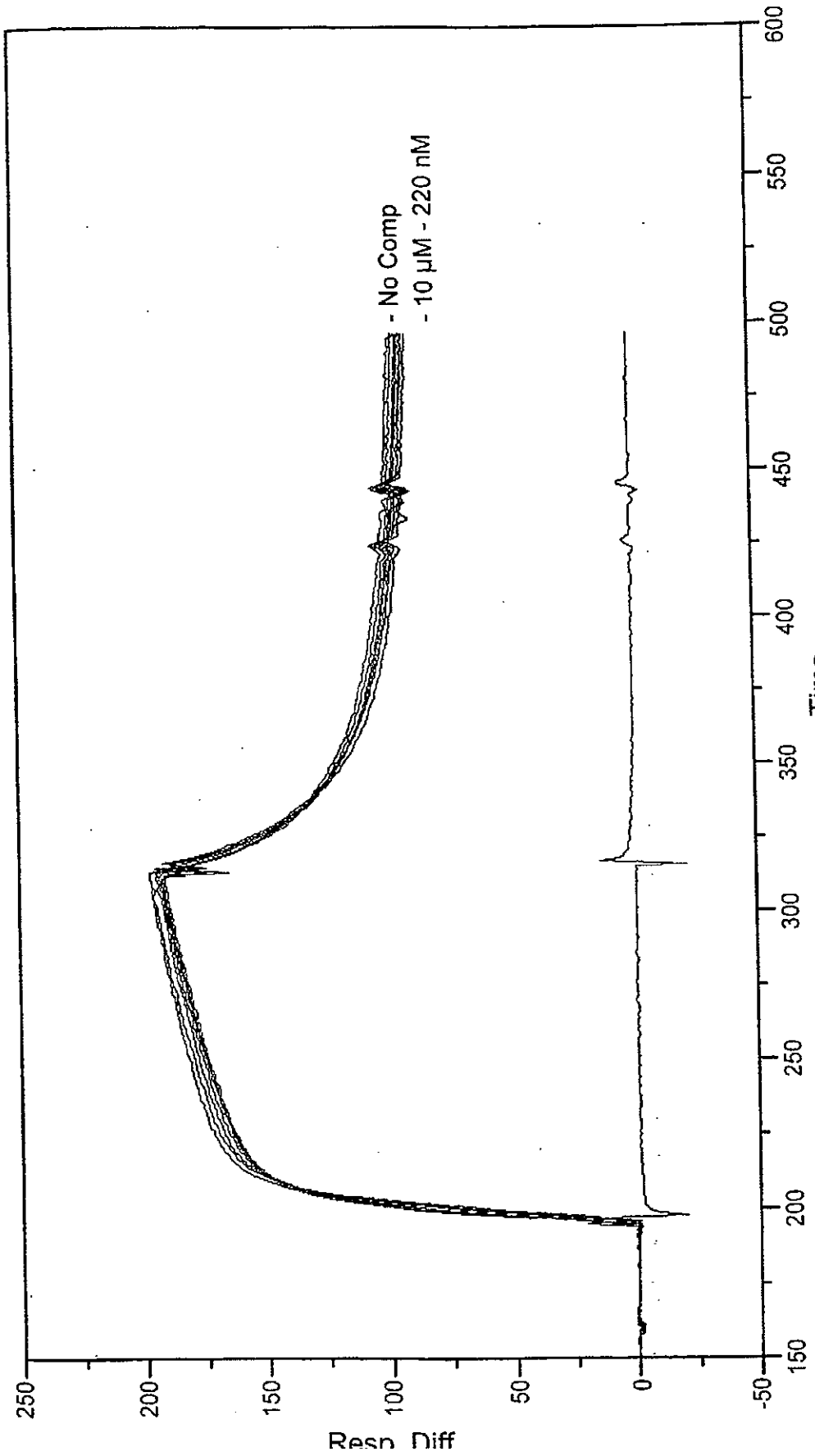

- No Comp
- 10 µM - 220 nM
Resp Diff
Time
250
200
150
100
50
0
-50
150
200
250
300
350
400
450
500
550
600

FIG. 6H

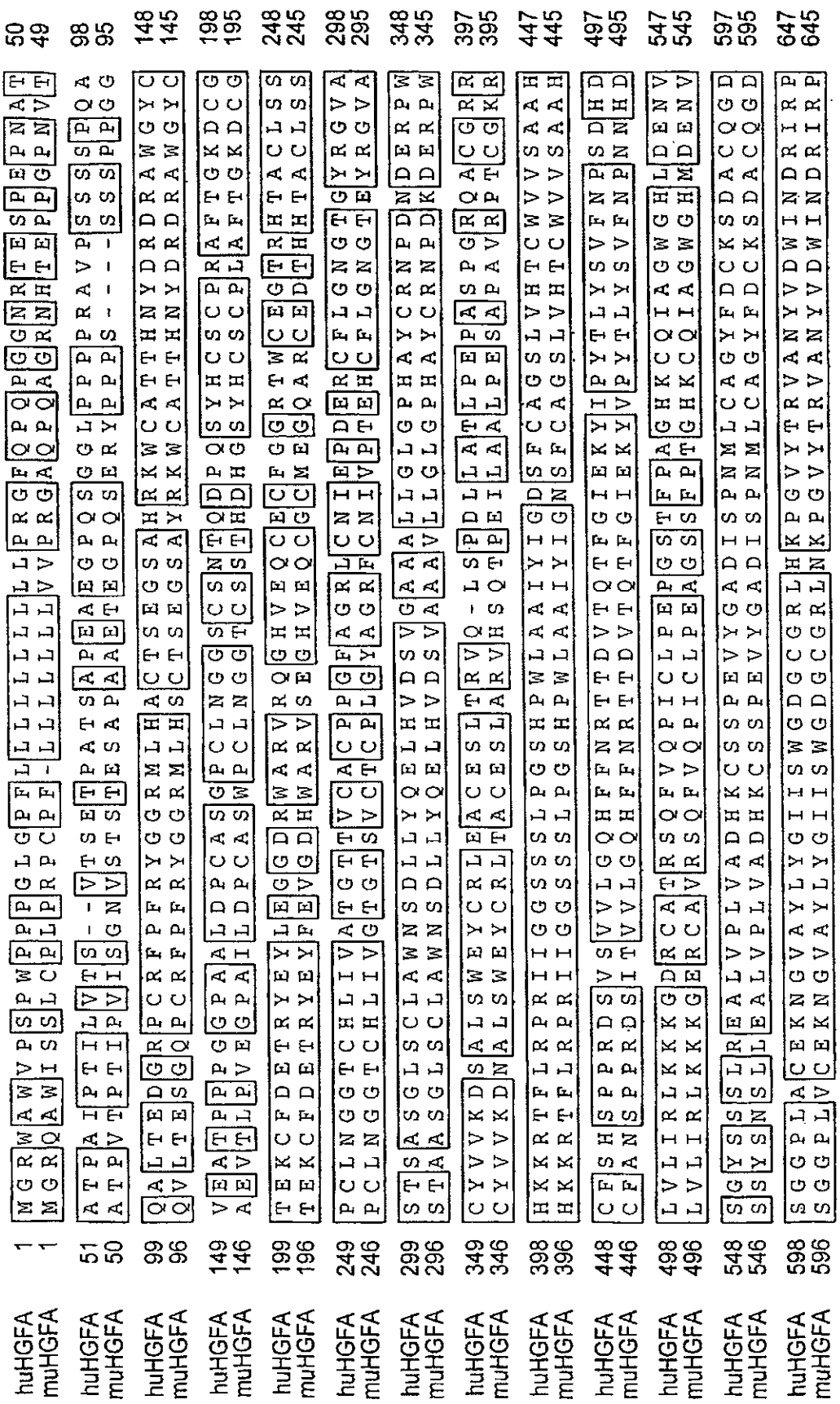


FIG. 7

Inhibition of HGFA Enzymatic Activity by Anti-HGFA Antibodies

Antibody #	Chromogenic Substrate Assay[a] IC_{50} (nM)	Macromolecular Substrate proHGF Inhibition at 670nM Antibody[b]
33	15.3	**
35	21.3	**
37	334	**
39	>500	*
42	>500	**
49	>500	N.I.
58	1.3	***
61	>500	**
74	>500	**
75	32.6	**
86	>500	*
90	203	**
91	169	**
95	>500	*

[a]Substrate was Spectrozyme® fVIIa at 0.2mM (~K_M)
[b]Qualitative assessment of inhibition based on disappearance of single chain HGF band: *, weak inhibition, ** strong inhibition, *** very strong inhibition; N.I., no inhibition.

FIG. 8

Binding of HGFA to Anti-HGFA Antibodies

Anti-HGFA Phage Antibody	Affinity by Phage ELISA IC_{50} (nM)	Affinity by BIAcore K_D (nM)	Inhibition of HGFA Binding by IV49	Inhibition of HGFA Binding by Small Molecule	Inhibition of HGFA Binding by sHAI-1B
58	2	1.3	++	++	++
75	6	100	+/-	-	+/-
37	56	ND[a]	+/-	+/-	ND[a]
74	60	ND[a]	+/-	-	+/-
42	30	ND[a]	+	+/-	+
61	20	ND[a]	+	-	+

++, +: >50% Inhibition
+/-: 20-50% Inhibition
-: <20% Inhibition
[HGFA] : 70 nM
[Competitor]: start at 10 fold above reported KD, and decrease by 2-fold dilution
[a]ND, Not Determined

FIG. 9