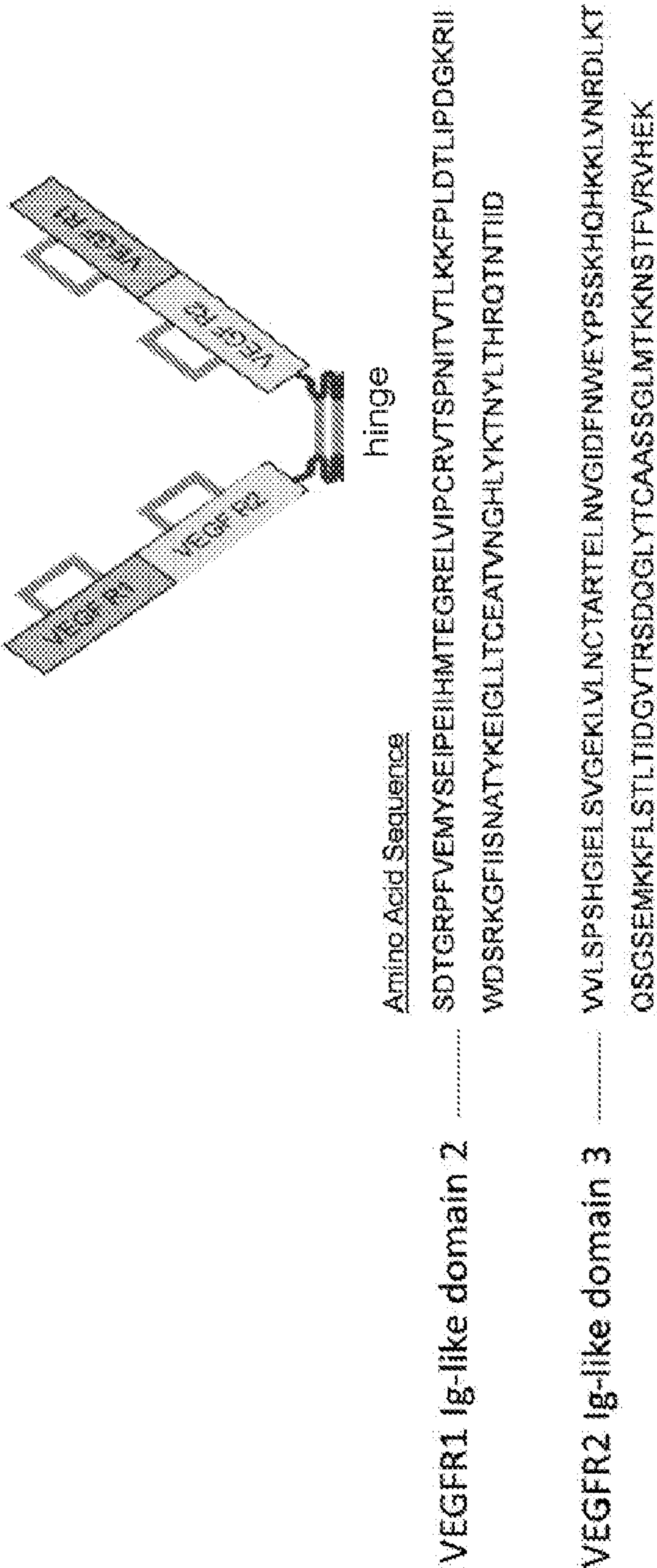


FIG. 1

Fabricator cleaved mini Trap: Fabricator enzyme cleaved product from aflibercept REGN3



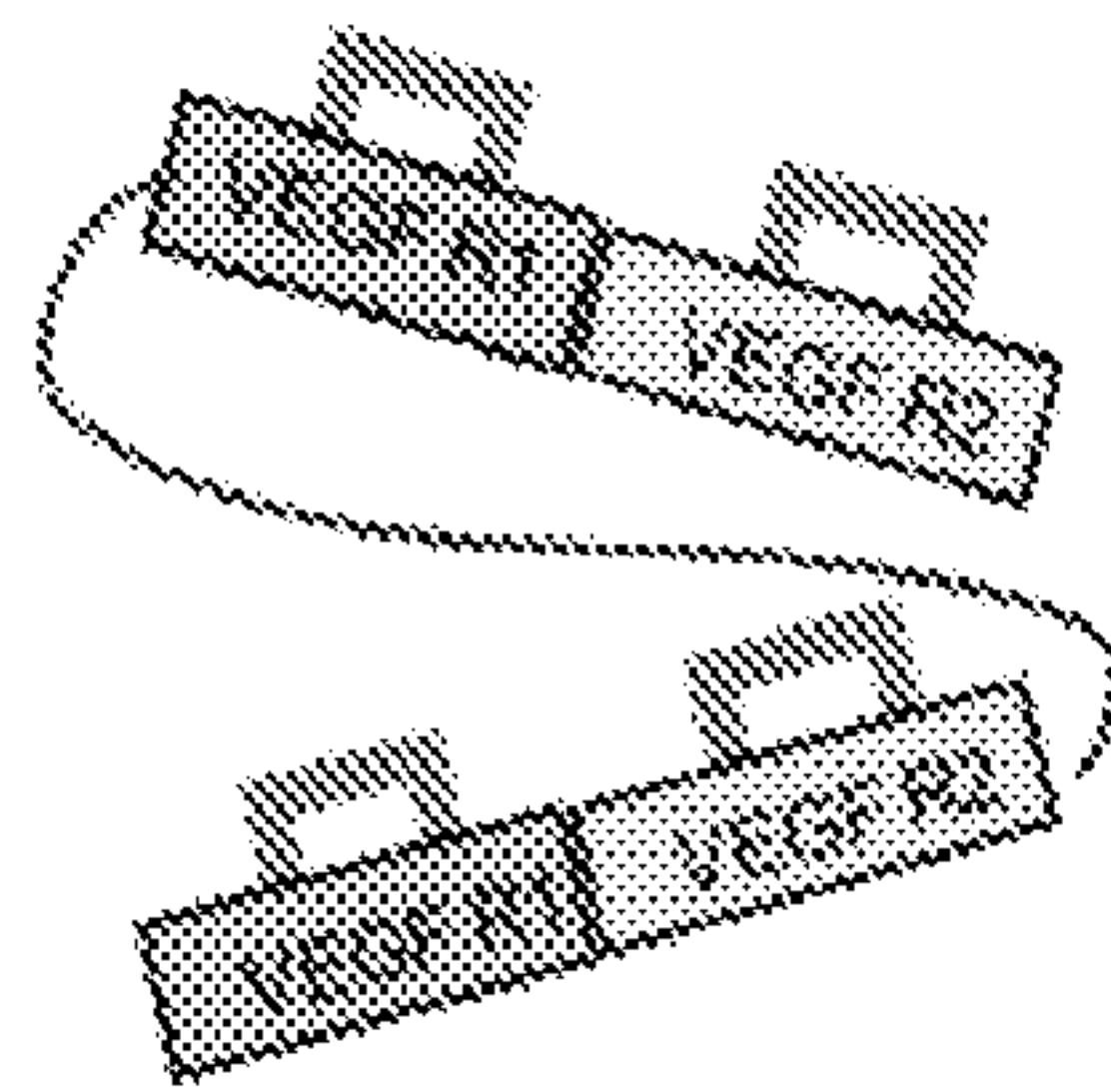
Fc hinge region to cleavage site at PAPELLG DKTHYCPAPPELLG //

cleaved off Fc fragment GPSVFLFPPKPKDTLMISRTPEVTCWVDVSHEDPEVKFNWYVDGVEVH
NAKTPREEQYNSTYRVWSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISK
AKGQPREPQVYTLPPSRDELTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNY
KTTTPPVLDSDGSGFFLYSKLTVDKSRWQQGNVFCSSVMHEALHNHYTQKSLSLS
PGK

// = FABRICATOR cleavage site

FIG. 2

Single chain mini Trap: VEGR1-VEGR2-(G4S)6-VEGFR1-VEGFR2



Amino Acid Sequence

VEGFR1 Ig-like domain 2 -----
SDTGRPFVEMYSEIPEIIHMTGRELVIPCRVTSPNITVTLKKFPLDLPDGKRIWDSRKGFIISNATYKEIGLLTCEAT
VNGHLYKTNLYLTHRQTNTIID

VEGFR2 Ig-like domain 3 _____
W L S P S H G I E L S V G E K L V L N C T A R T E L N V G I D F N W E Y P S S K H Q H K K L V N R D L K T Q S G S E M K K F L S T L T I D G V T R S D Q
GLYTCAASSGLMTKKNSTFVRVHEK

(G₄S)₆ linker ----- GGGSGGGSGGGSGGGSGGGSS

VEGFR1 Ig-like domain 2 _____
SDTGRPFVEMYSEIPEIIHMTGRELVIPCRVTSPNITVTLKKFPLDLPDGKRIWDSRKGFIIISNATYKEIGLLTCEAT
VNGHLYKTNLYLTHRQTNTIID

VEGFR2 Ig-like domain 3 _____
WLSPSHGIELSVGEKLVNCTARTELVGIDFNWEYPSSKHQHKLVNRDLKTQSGSEMKKFLSTLTIDGVTRSDQ
GLYTCAASSGLMTKKKNSTFVRVHEK

FIG. 3A

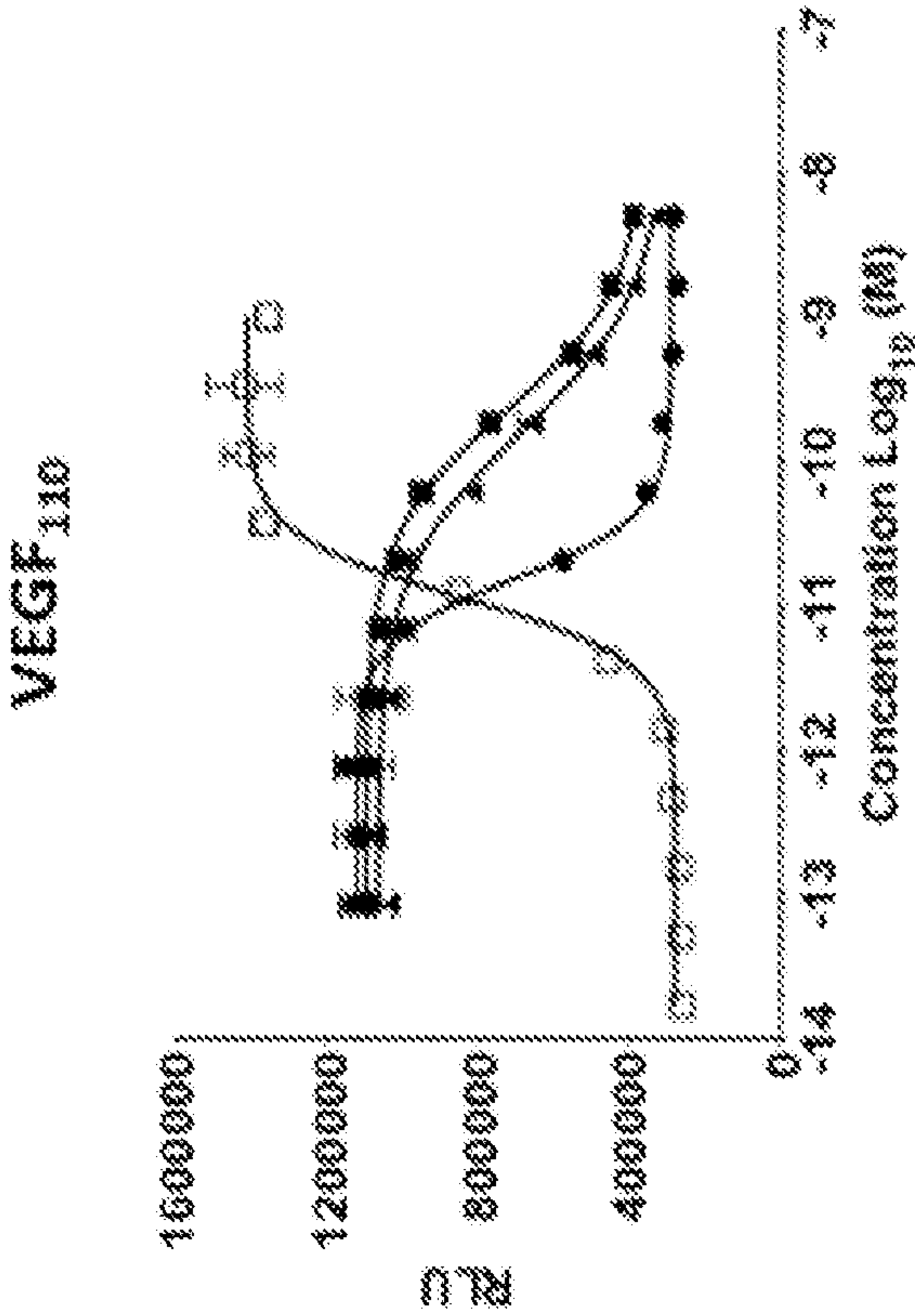


FIG. 3B

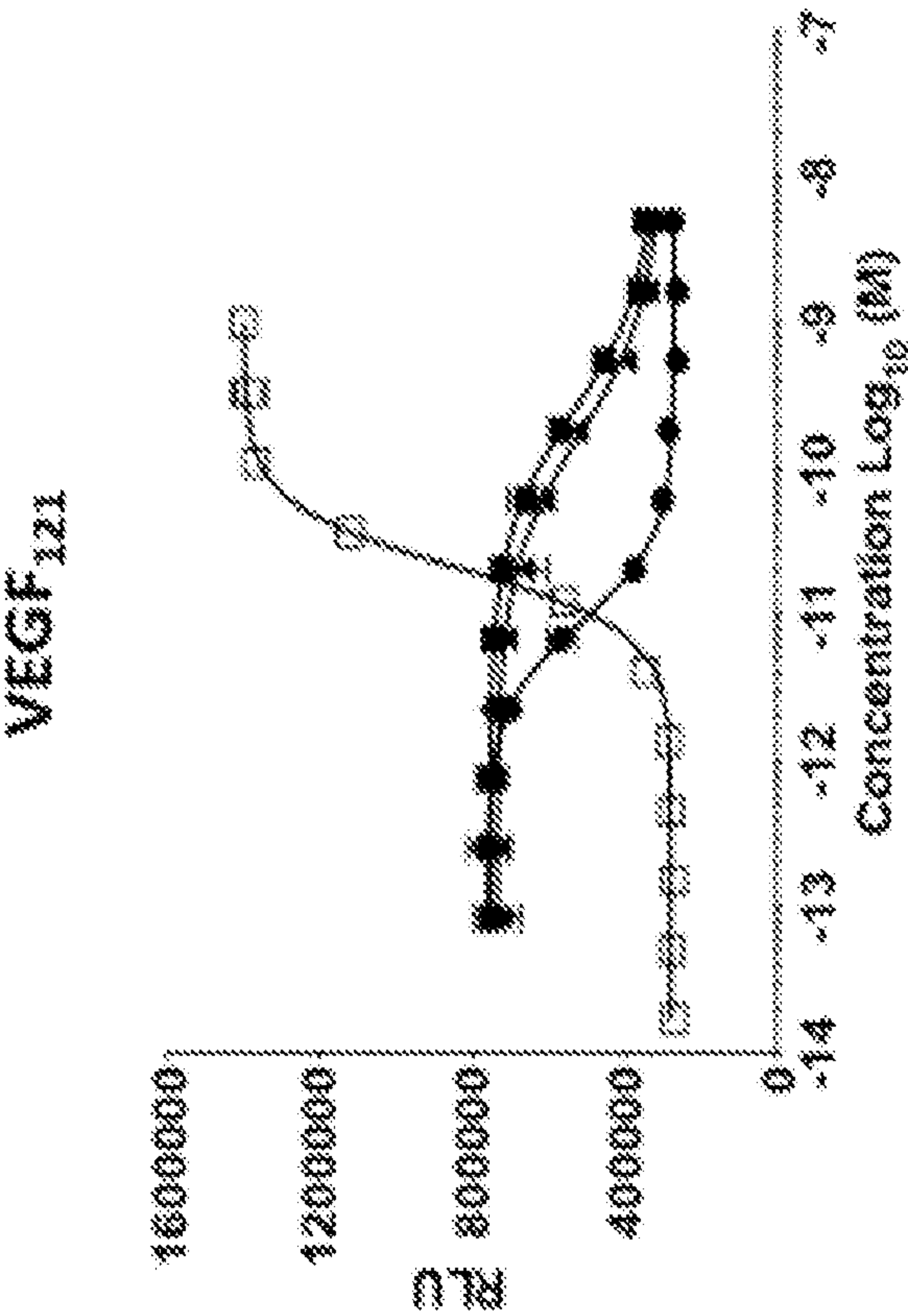


FIG. 3C

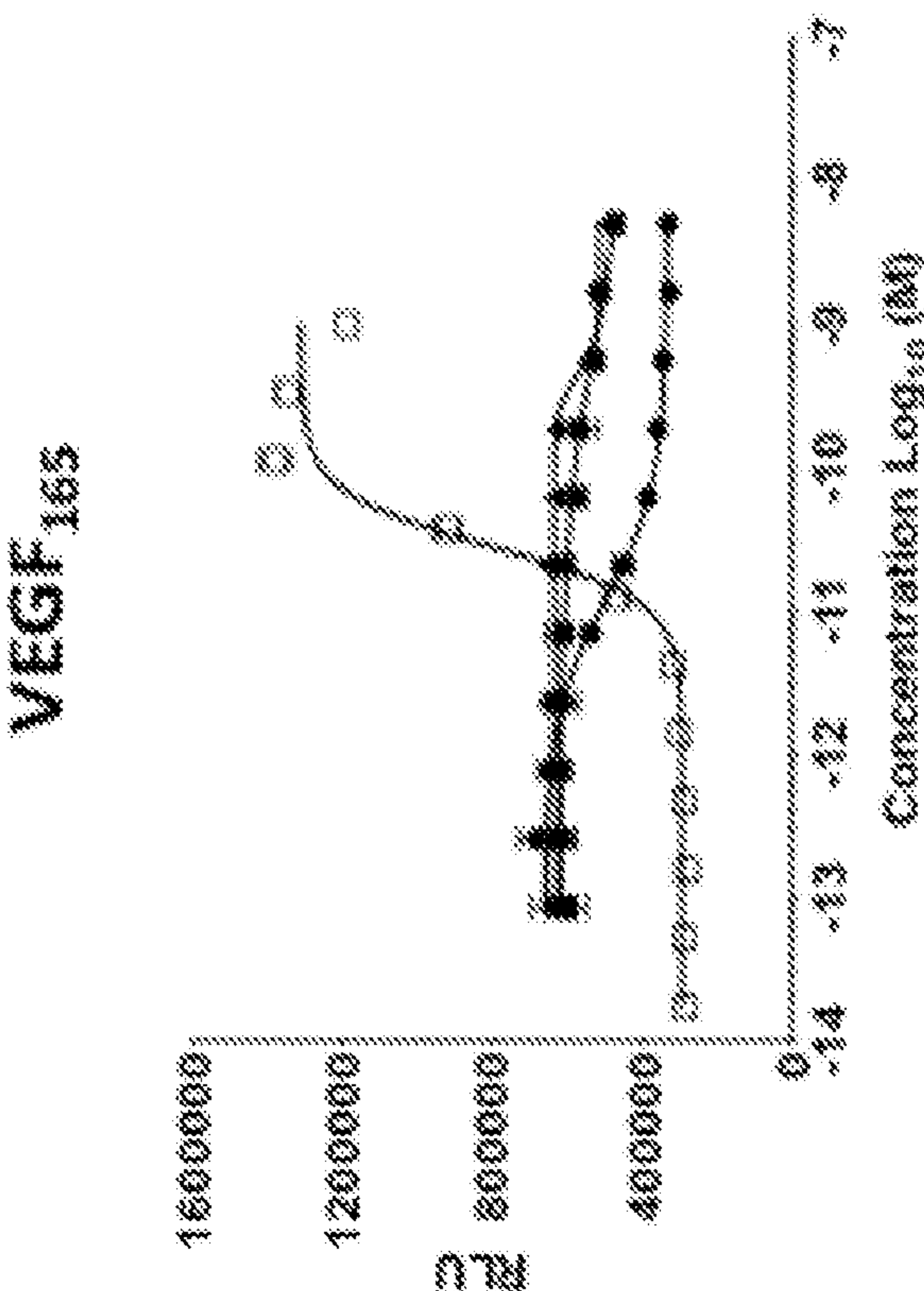


FIG. 4A

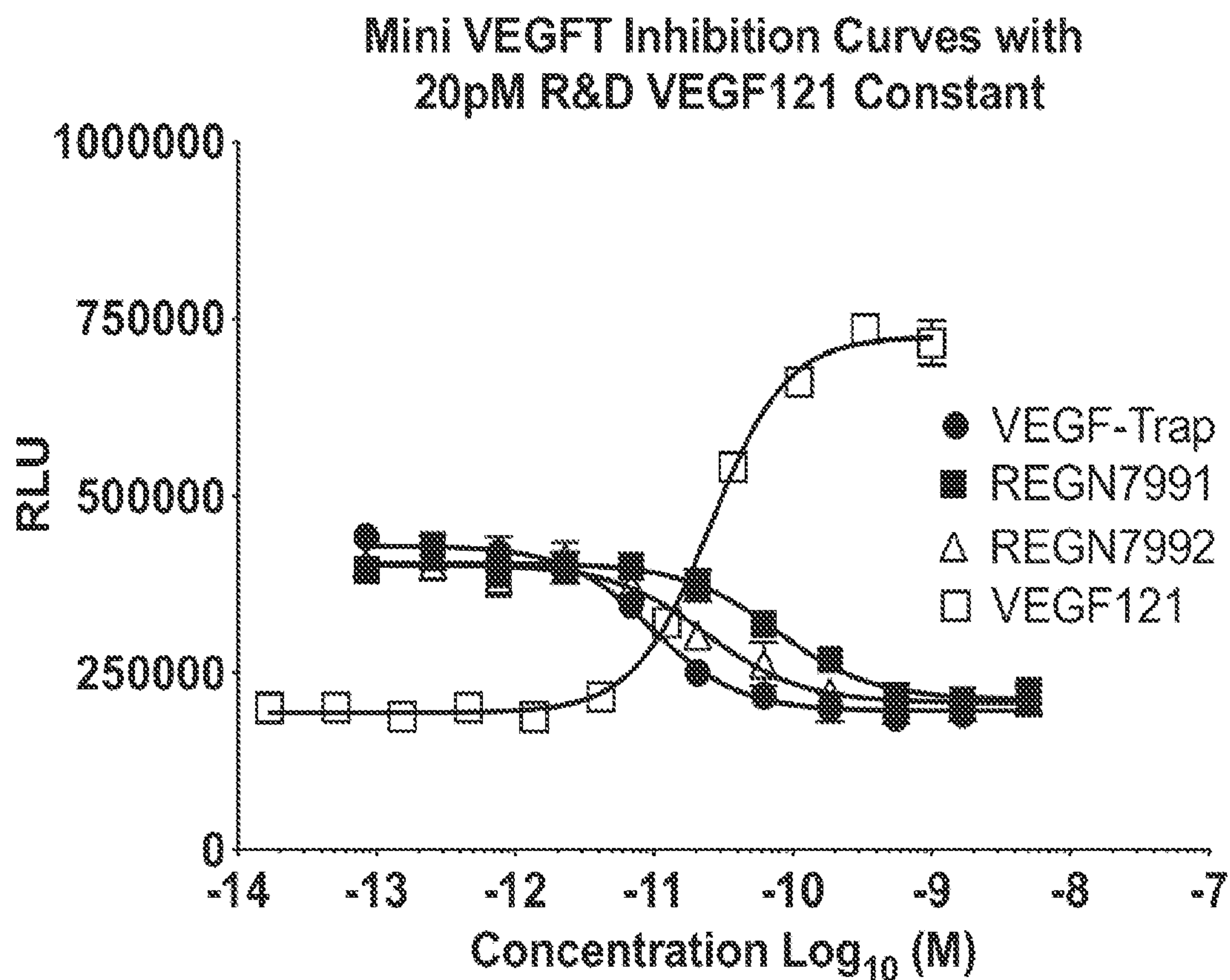


FIG. 4B

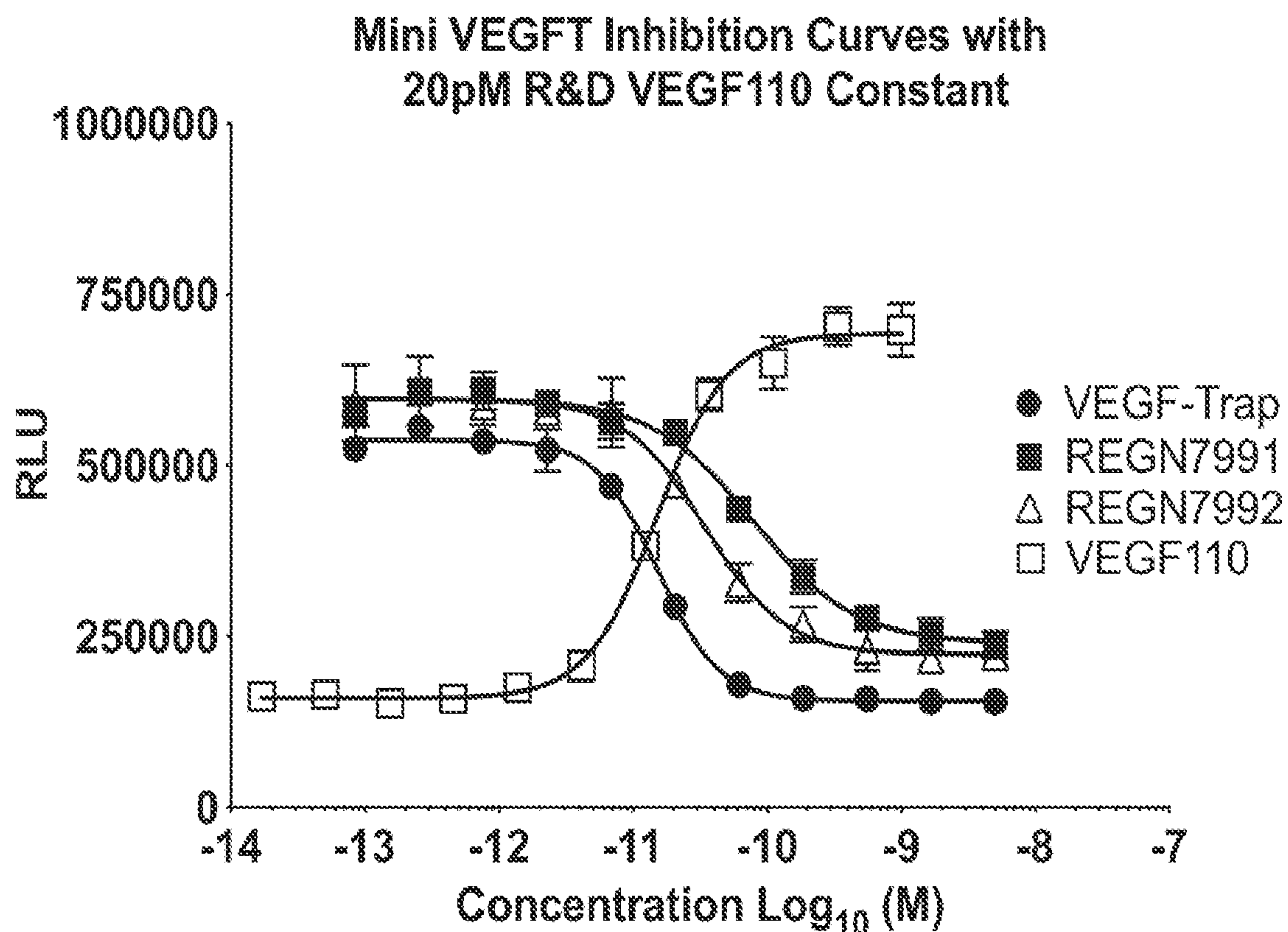


FIG. 5A

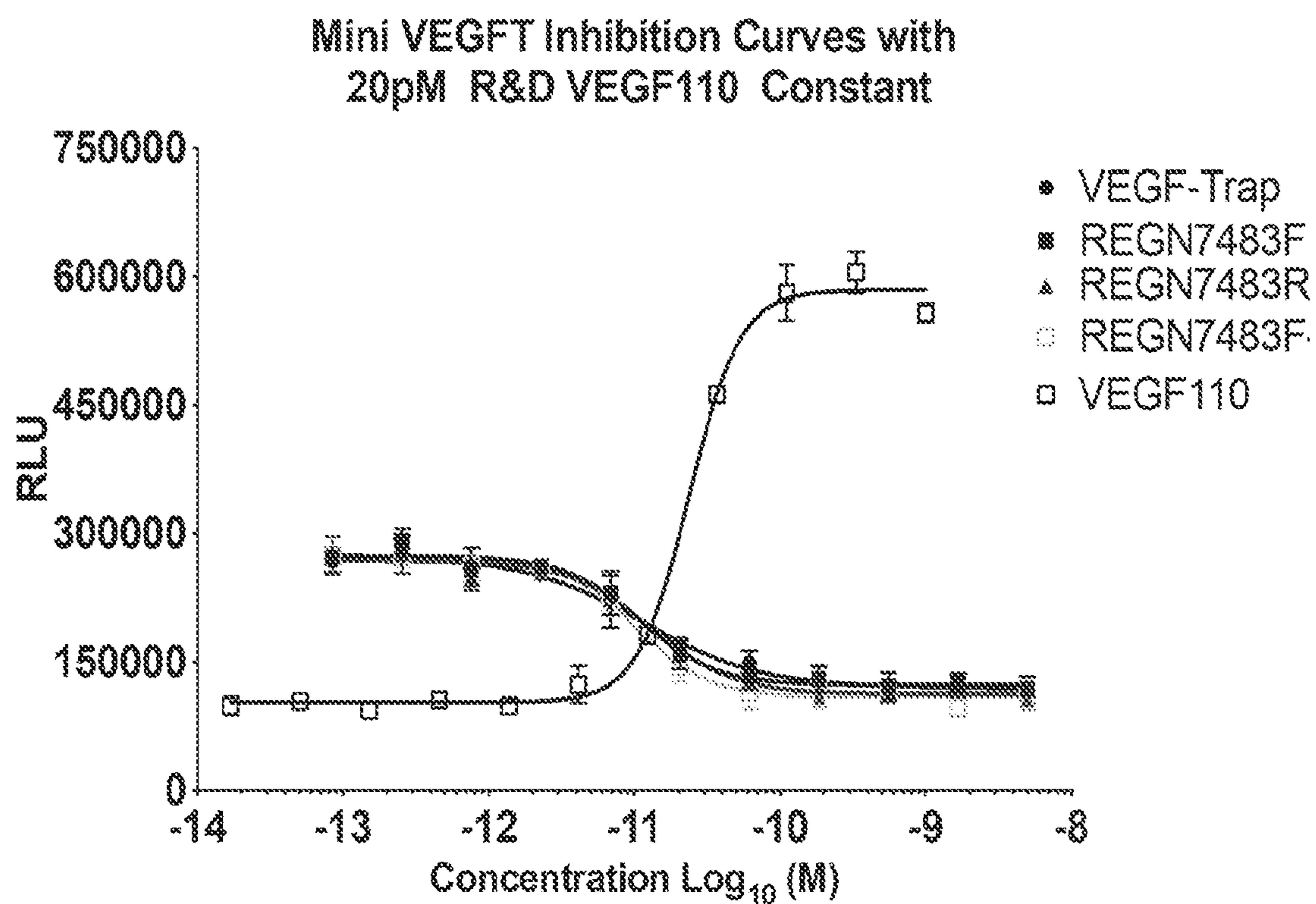


FIG. 5B

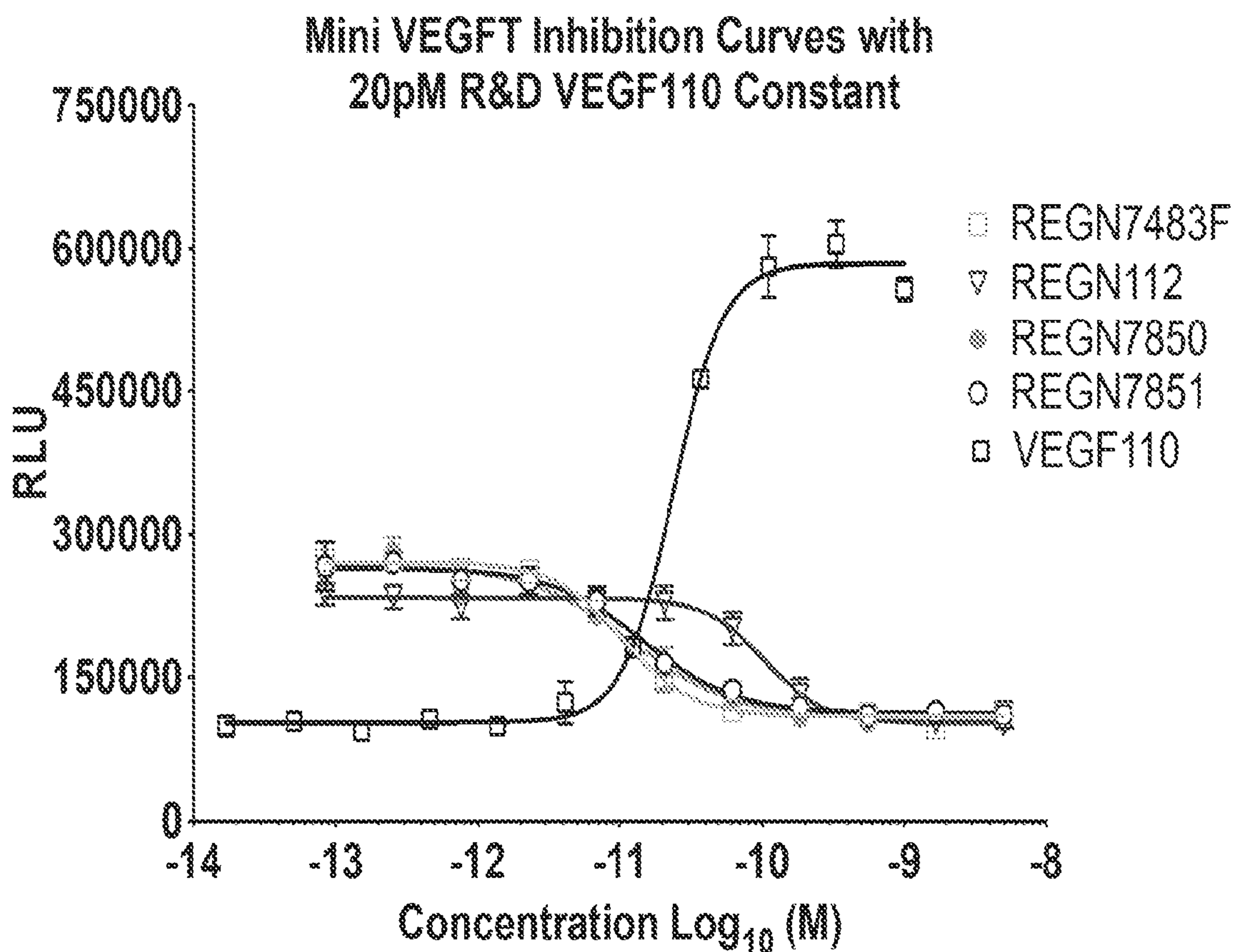


FIG. 5C

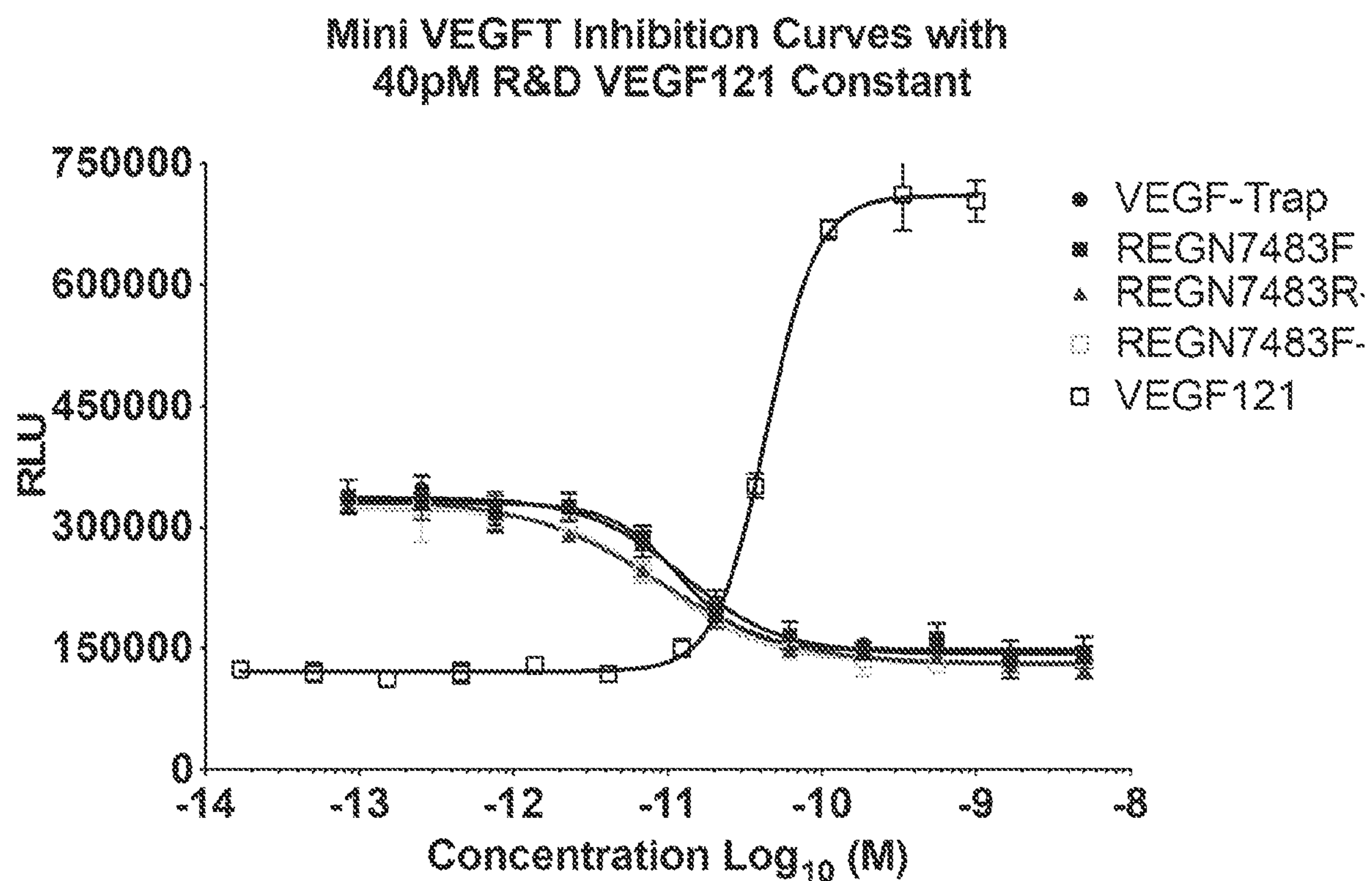


FIG. 5D

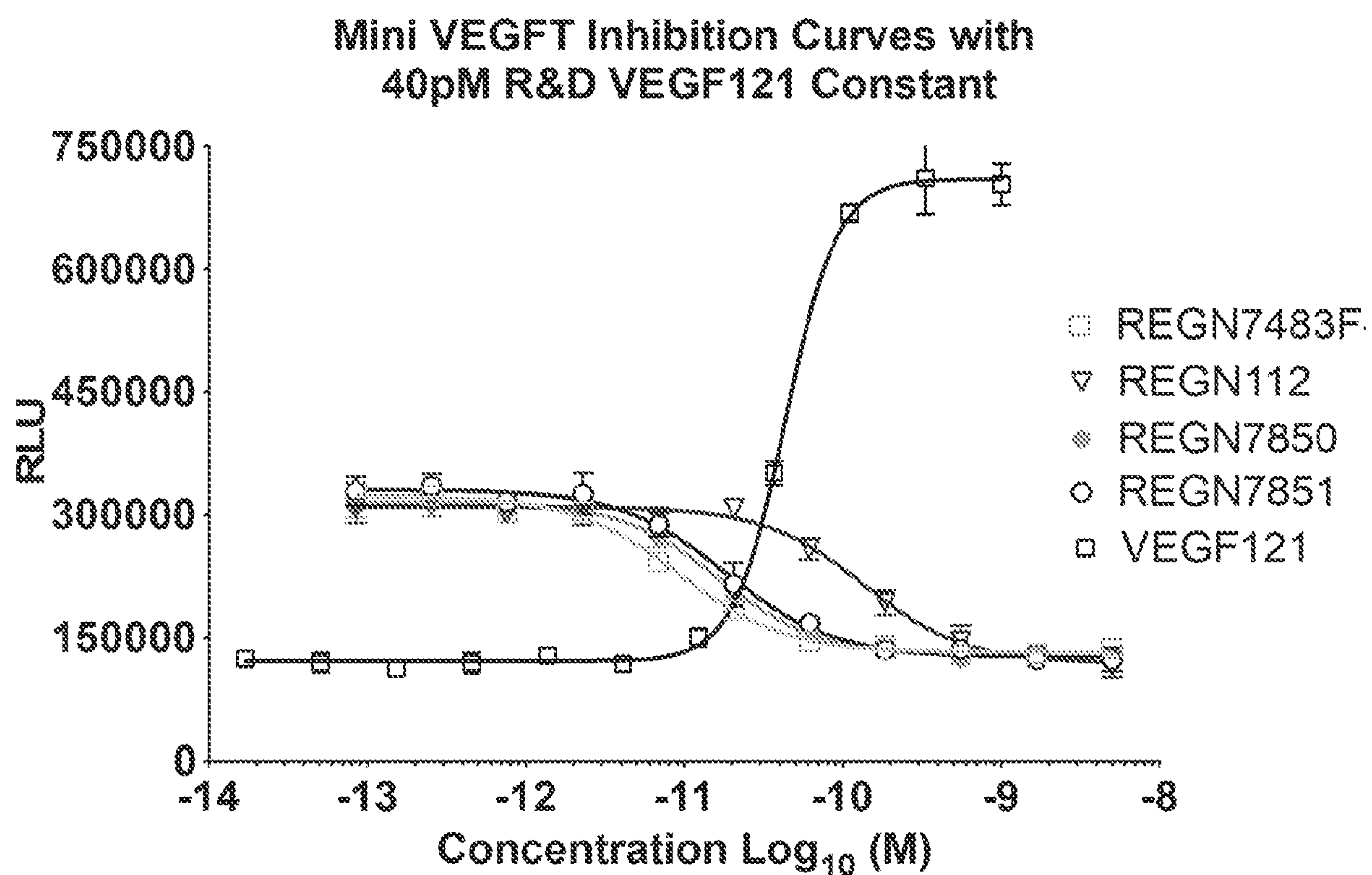


FIG. 5E

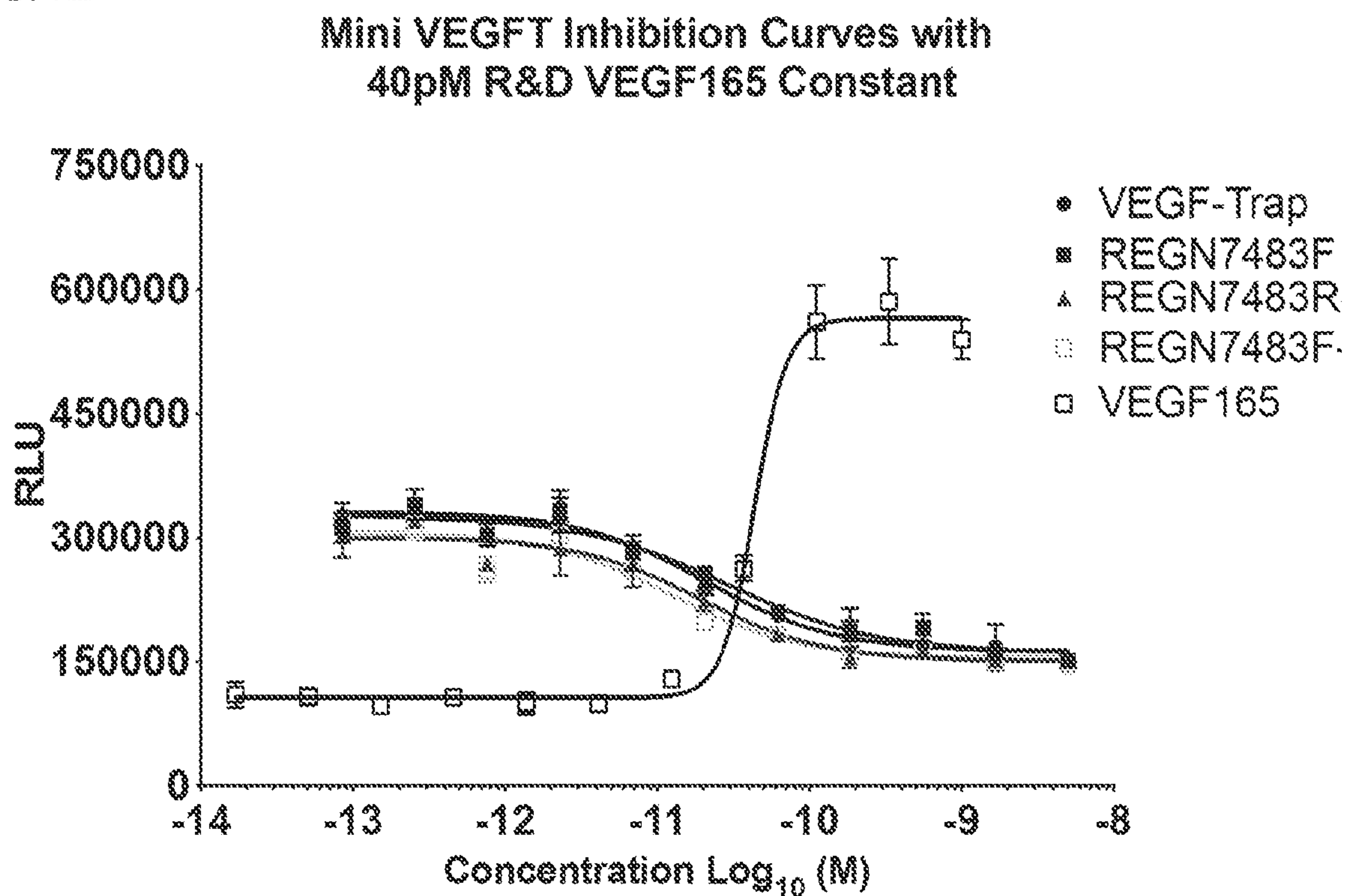


FIG. 5F

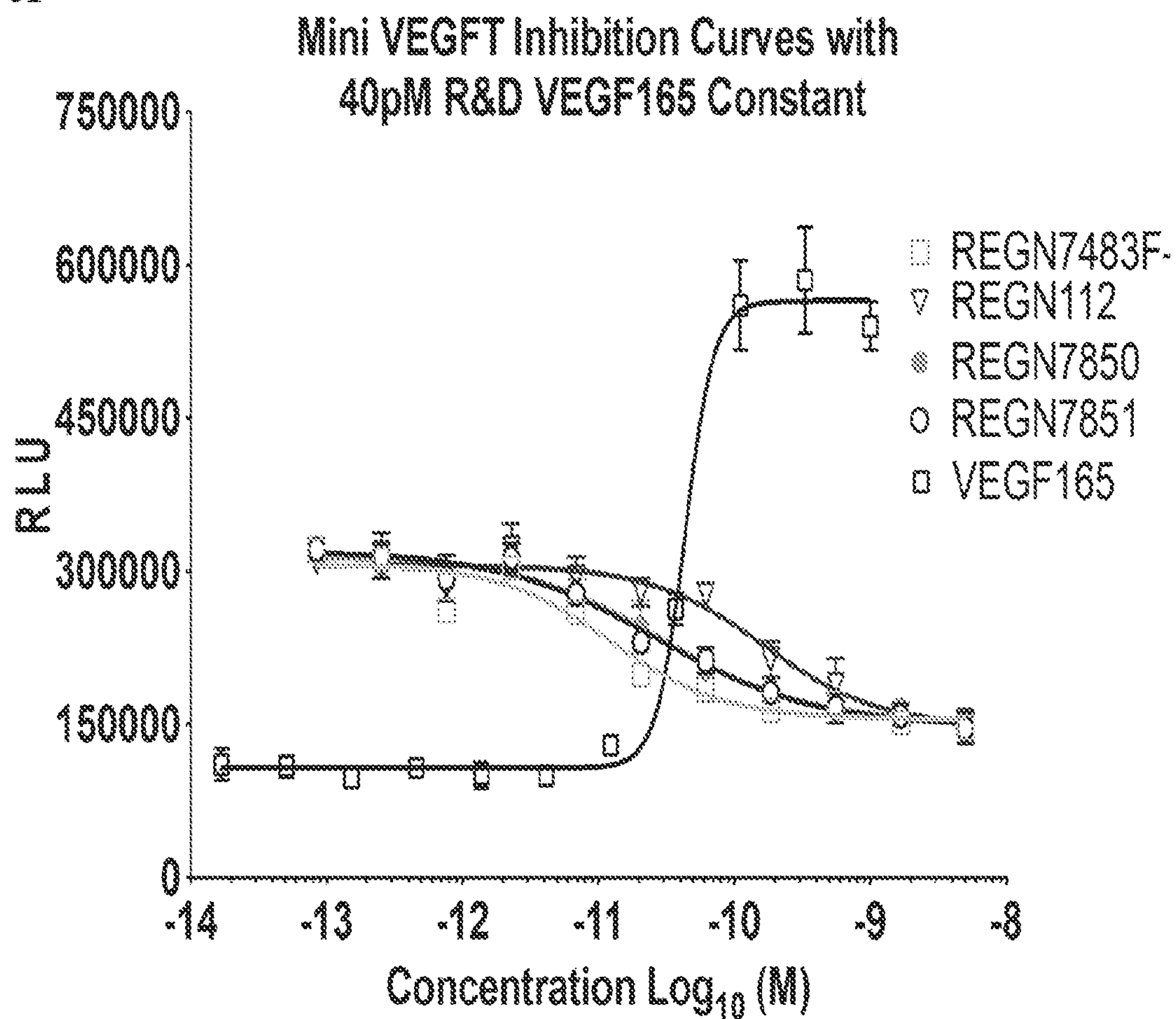


FIG. 6

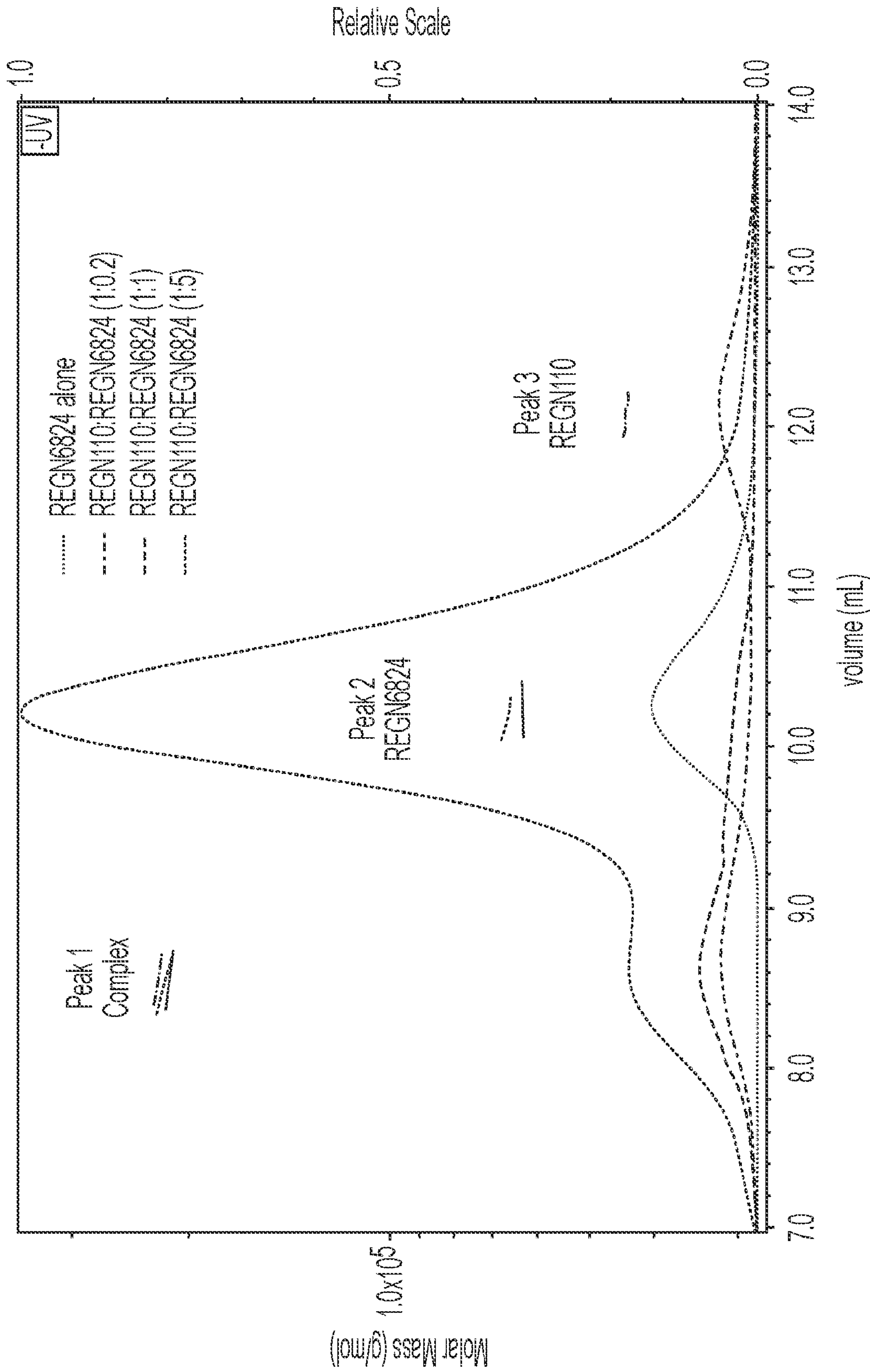


FIG. 7

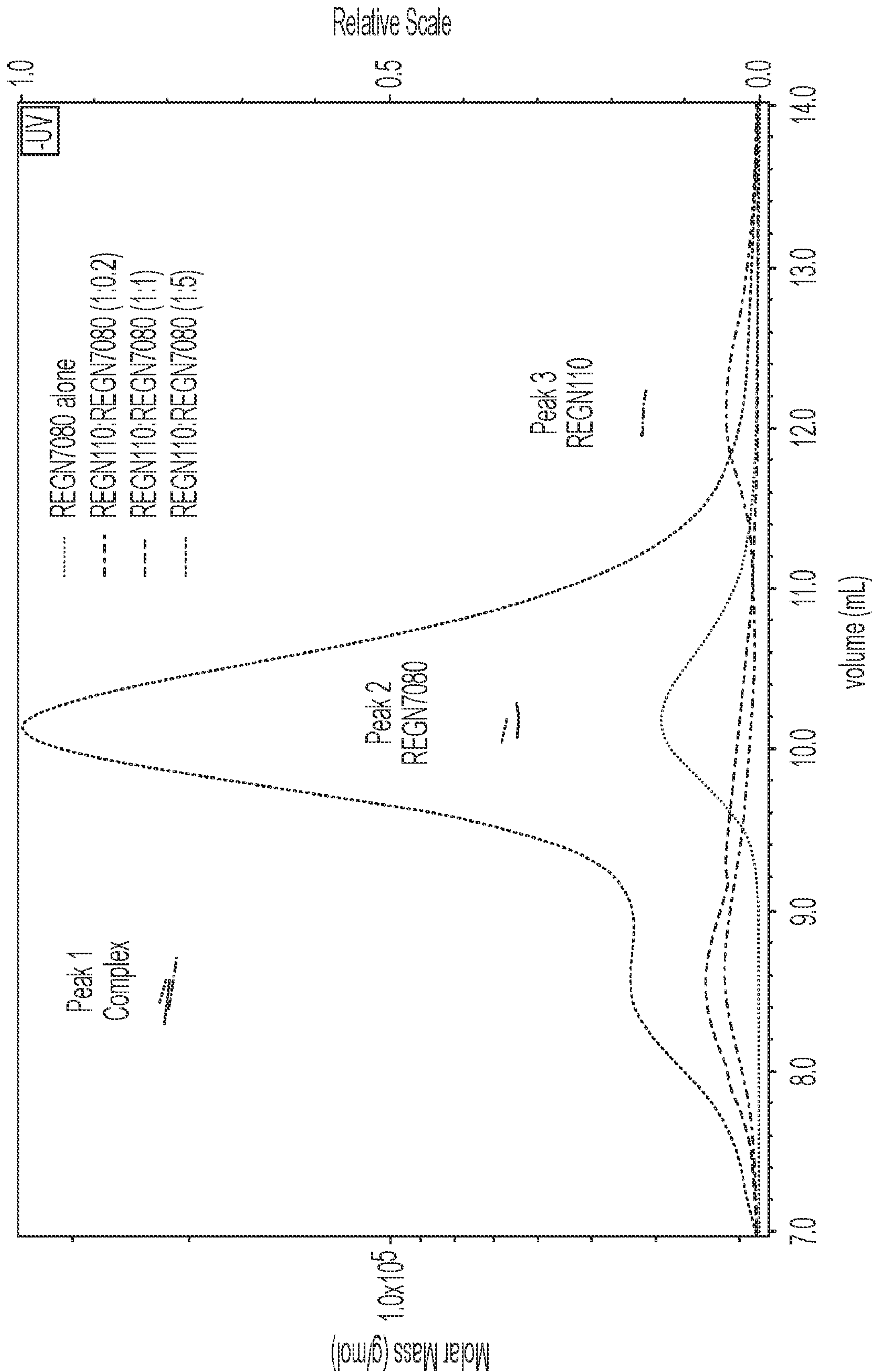
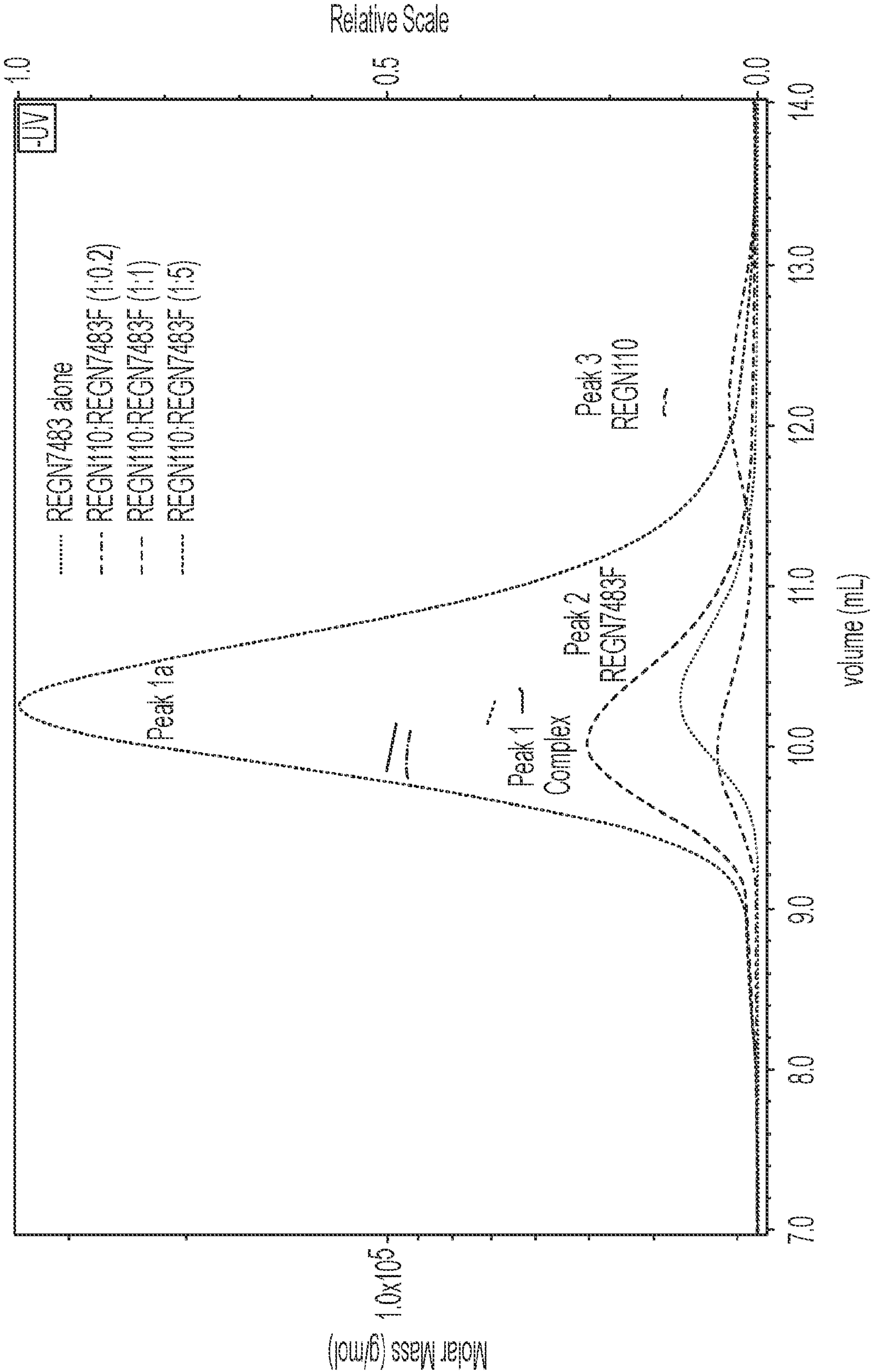
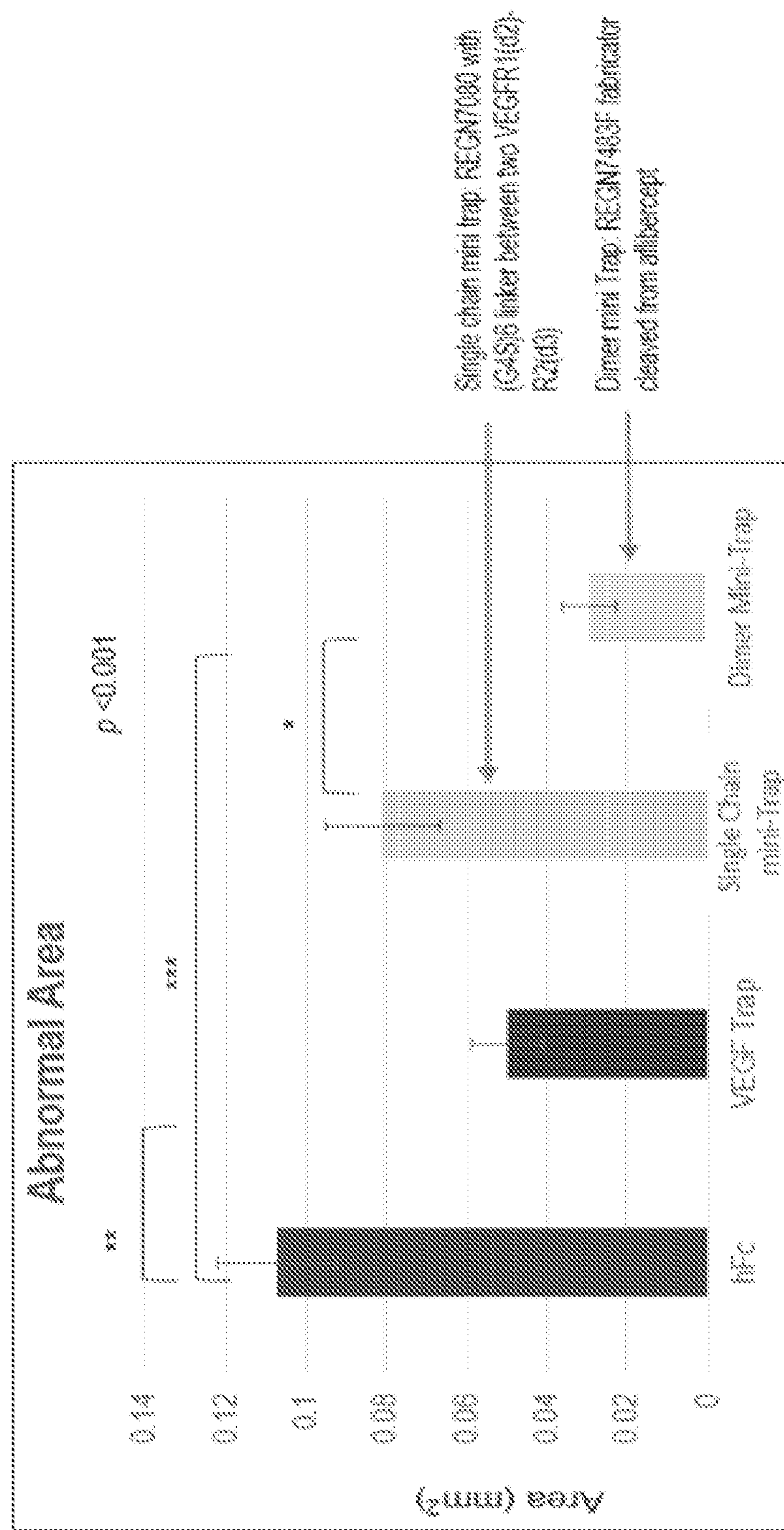


FIG. 8



CHILD



Oxygen induced retinopathy model; equimolar doses (0.25 μ g VEGF Trapeye, 0.125 μ g MNV-Trap/eye)

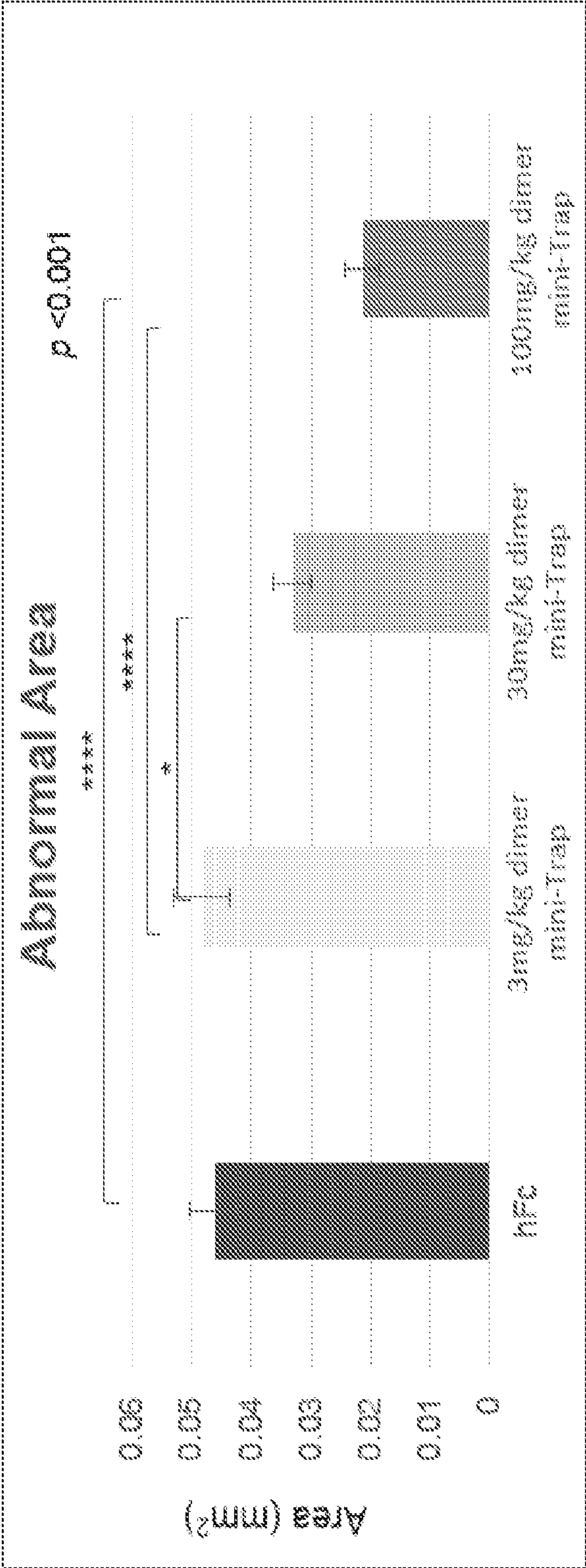


FIG. 10A

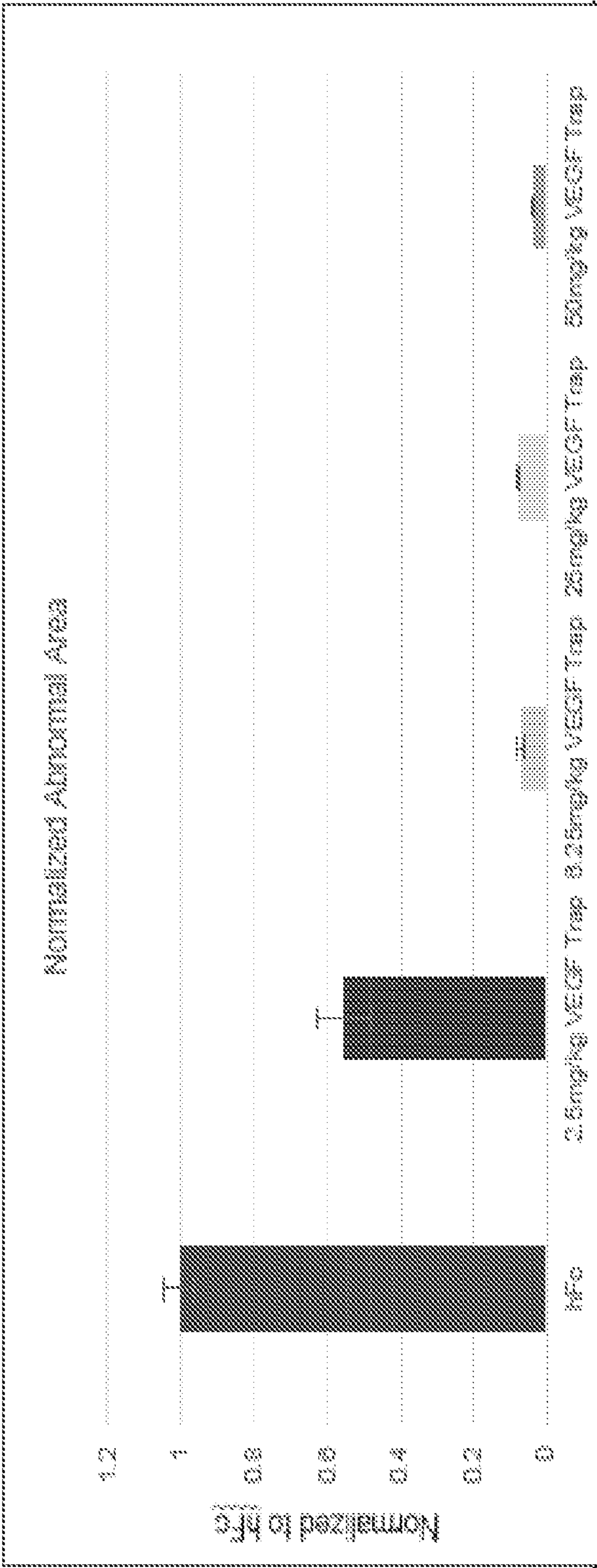


FIG. 10B

FIG. 11



REGN112

SDTGRPFVEMYSEIPEIIHMTEGRELVIPCRVTSPNITVTLKKFPLDTLIPDGKRIIWDSRKGFIISNATYKEIGLL
TCEATVNGHLYKTNLYLTHRQTNTIIDVVLSPSHGIELSVGEKLVNCTARTELVGIDFNWEYPSSKHQHKLVNRD
LKTQSGSEMKKFLSTLTIDGVTRSDQGLYTCAASSGLMTKKNSTFVRVHEKDKTHTCPPC
(SEQ ID NO: 26)

REGN7850

SDTGRPFVEMYSEIPEIIHMTEGRELVIPCRVTSPNITVTLKKFPLDTLIPDGKRIIWDSRKGFIISNATYKEIGLL
TCEATVNGHLYKTNLYLTHRQTNTIIDVVLSPSHGIELSVGEKLVNCTARTELVGIDFNWEYPSSKHQHKLVNRD
LKTQSGSEMKKFLSTLTIDGVTRSDQGLYTCAASSGLMTKKNSTFVRVHEKDKTHTCPPCPPC
(SEQ ID NO: 27)

REGN7851

SDTGRPFVEMYSEIPEIIHMTEGRELVIPCRVTSPNITVTLKKFPLDTLIPDGKRIIWDSRKGFIISNATYKEIGLL
TCEATVNGHLYKTNLYLTHRQTNTIIDVVLSPSHGIELSVGEKLVNCTARTELVGIDFNWEYPSSKHQHKLVNRD
LKTQSGSEMKKFLSTLTIDGVTRSDQGLYTCAASSGLMTKKNSTFVRVHEKDKTHTCPPCPPCPPC
(SEQ ID NO: 28)

REGN7483

SDTGRPFVEMYSEIPEIIHMTEGRELVIPCRVTSPNITVTLKKFPLDTLIPDGKRIIWDSRKGFIISNATYKEIGLL
TCEATVNGHLYKTNLYLTHRQTNTIIDVVLSPSHGIELSVGEKLVNCTARTELVGIDFNWEYPSSKHQHKLVNRD
LKTQSGSEMKKFLSTLTIDGVTRSDQGLYTCAASSGLMTKKNSTFVRVHEKDKTHTCPPCPAPELLG
(SEQ ID NO: 12)

FIG. 12

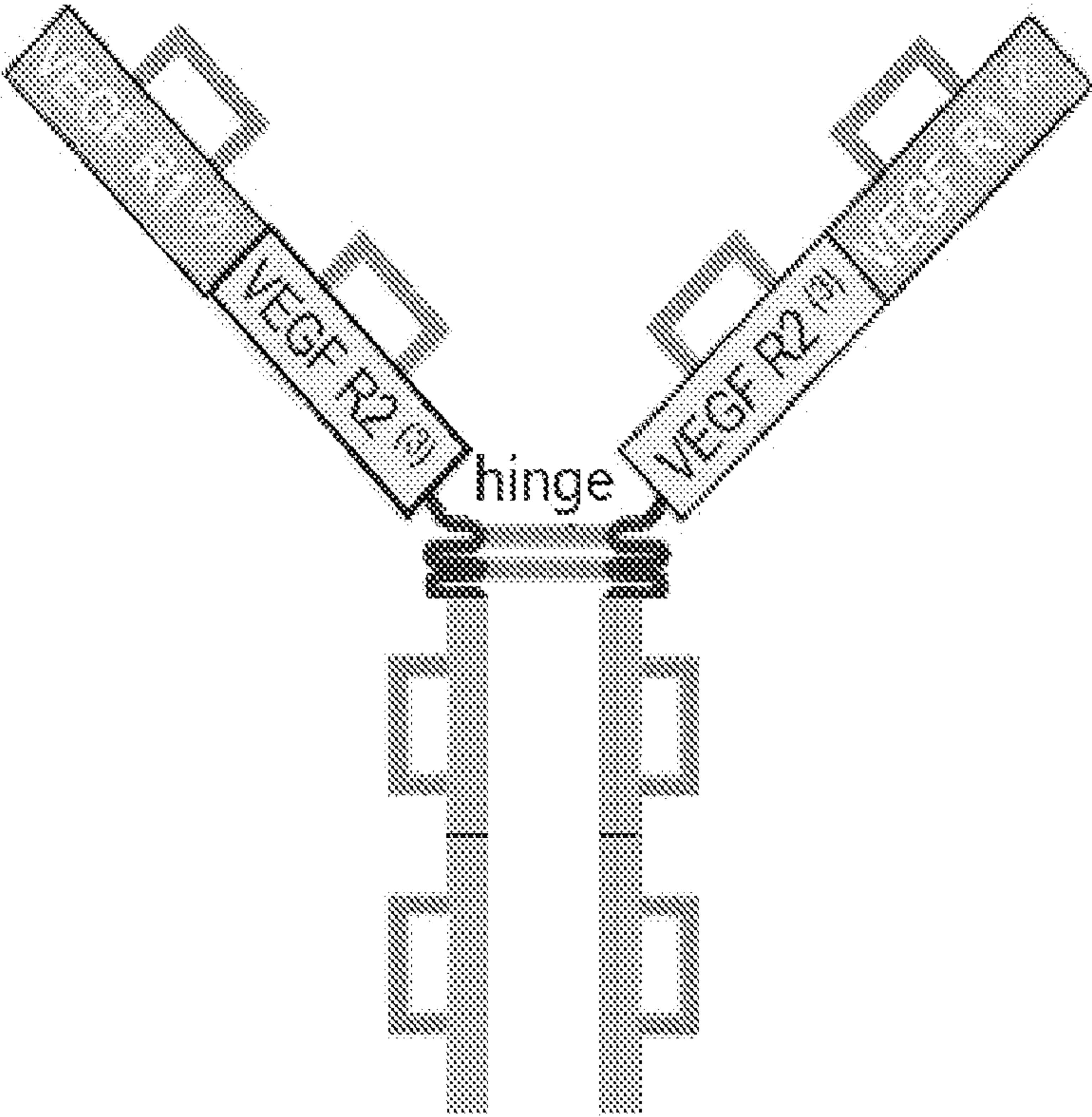
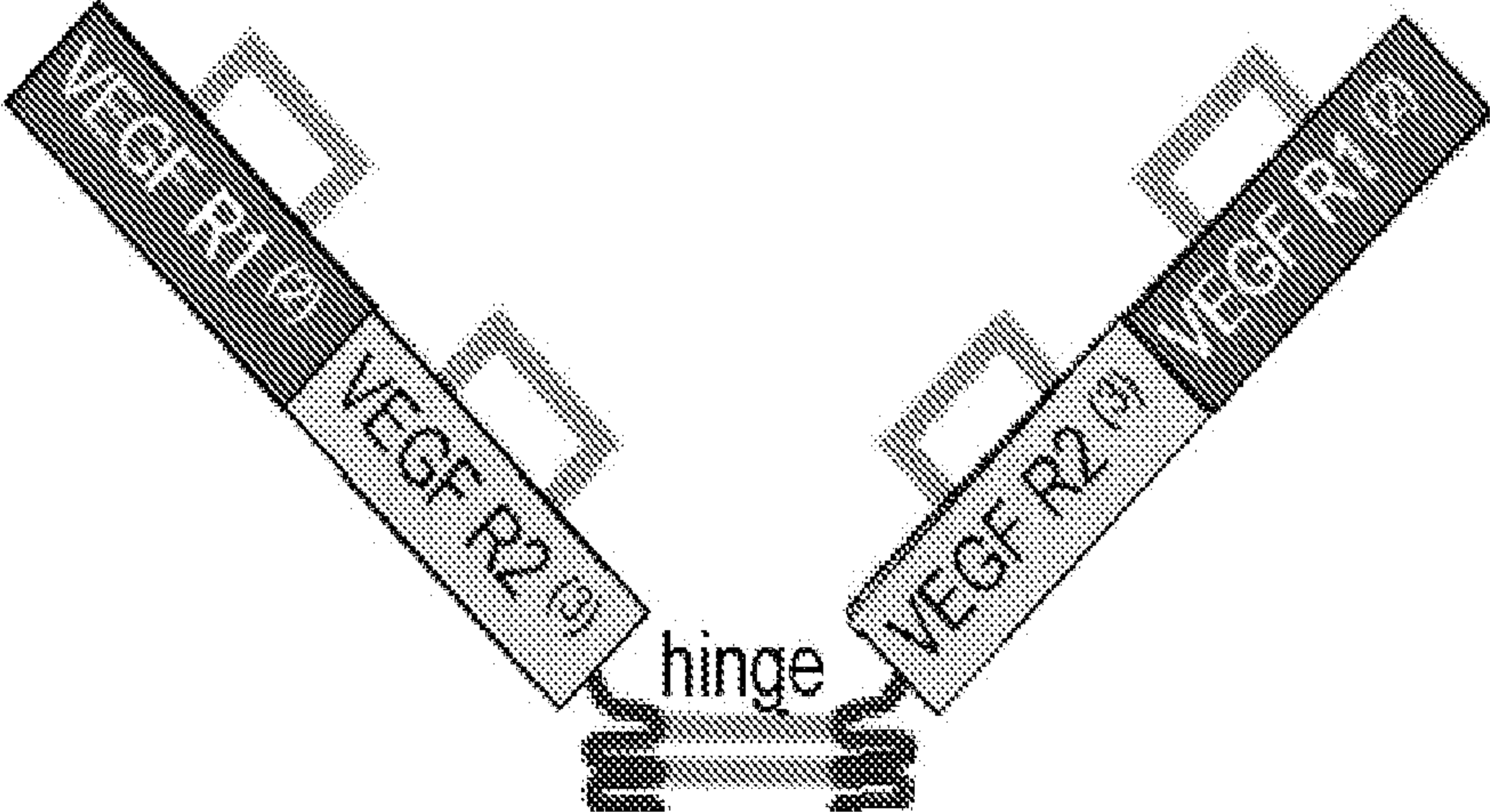
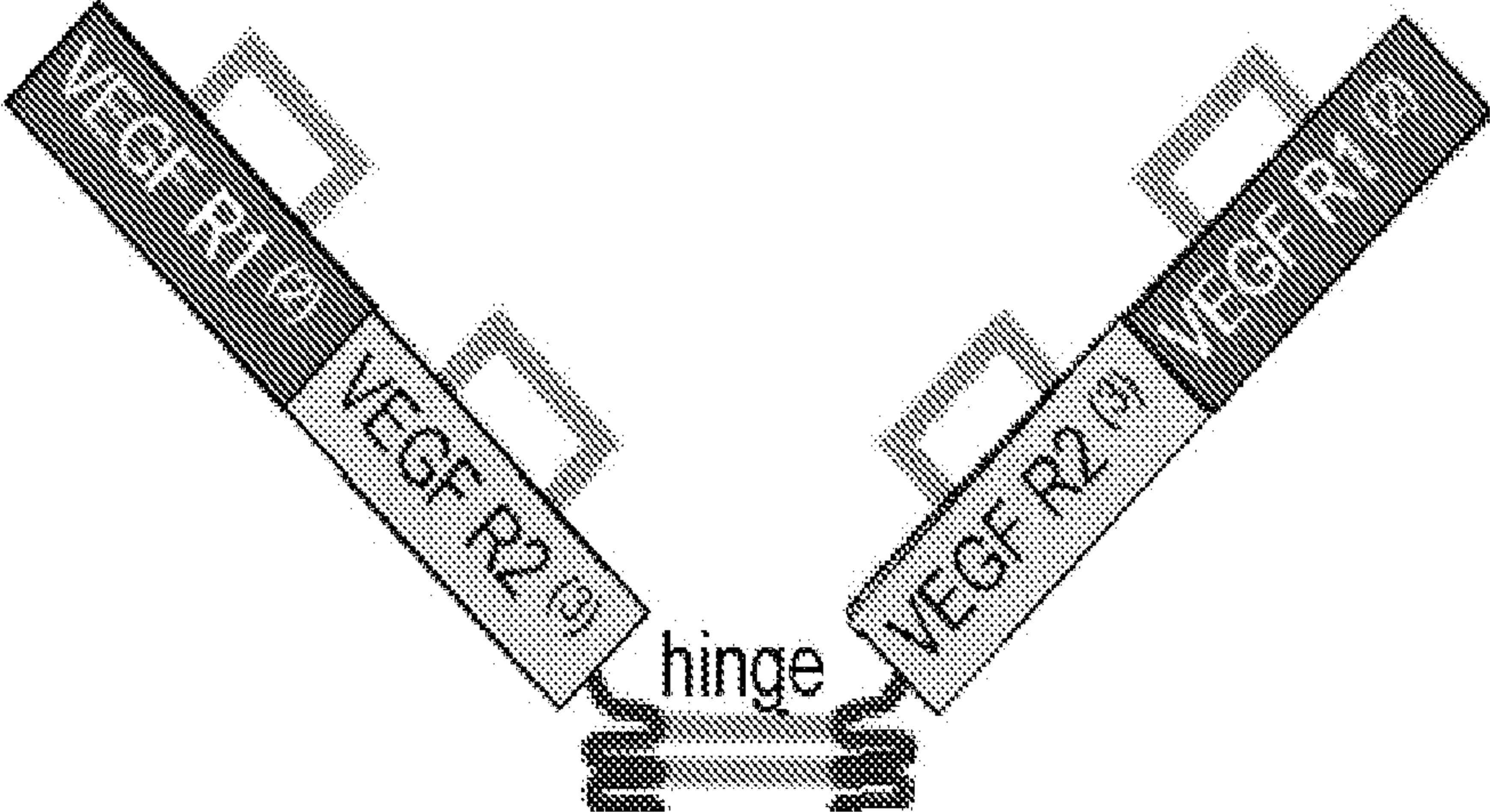
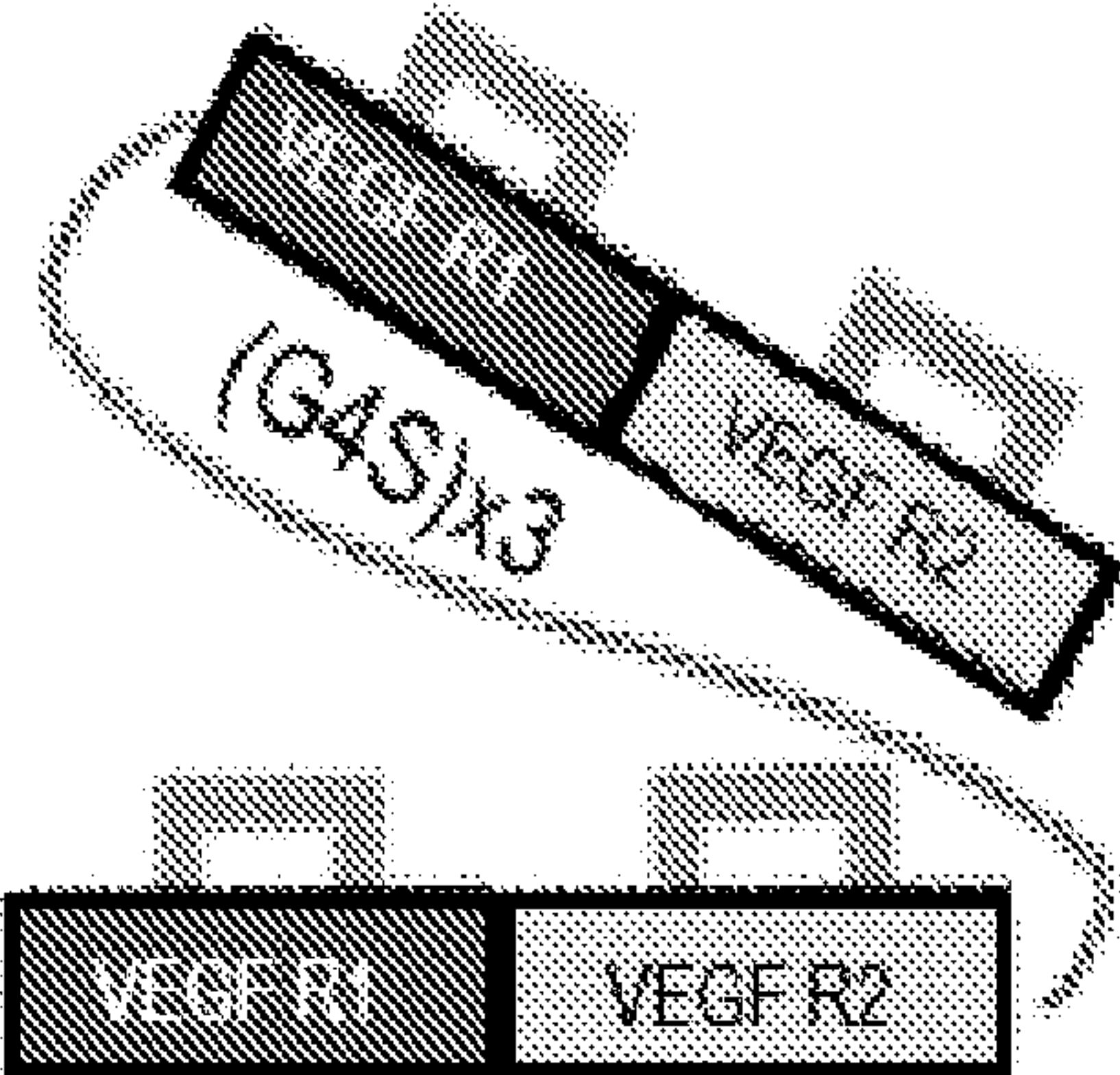
REGN#	Construct Details
REGN3	
REGN7483 ^F	
REGN7483 ^R	
REGN6824	 VEGF R1 is VEGF Receptor 1 Ig-like domain 2 VEGF R2 is VEGF Receptor 2 Ig-like domain 3

FIG. 12 cont.

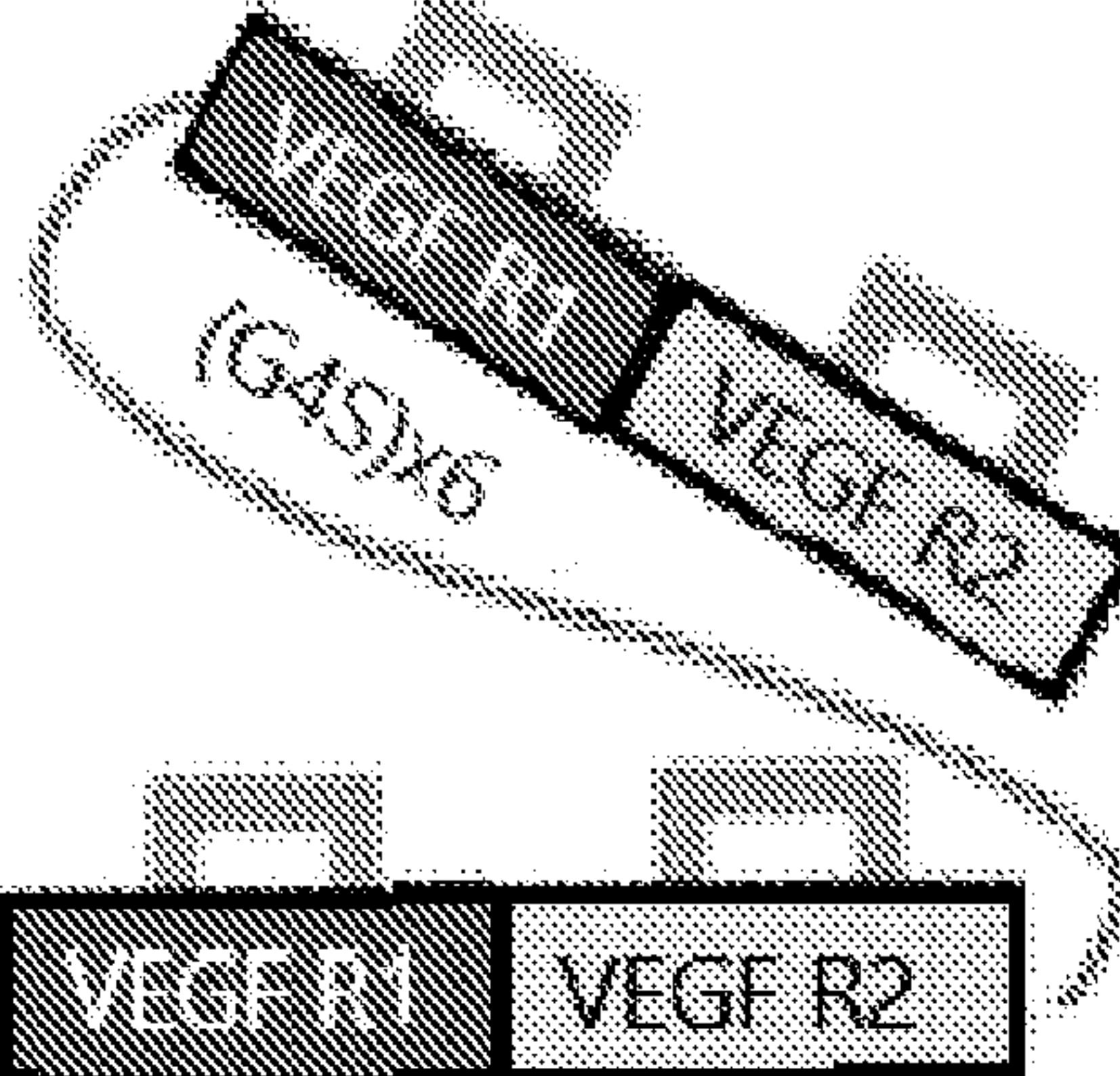
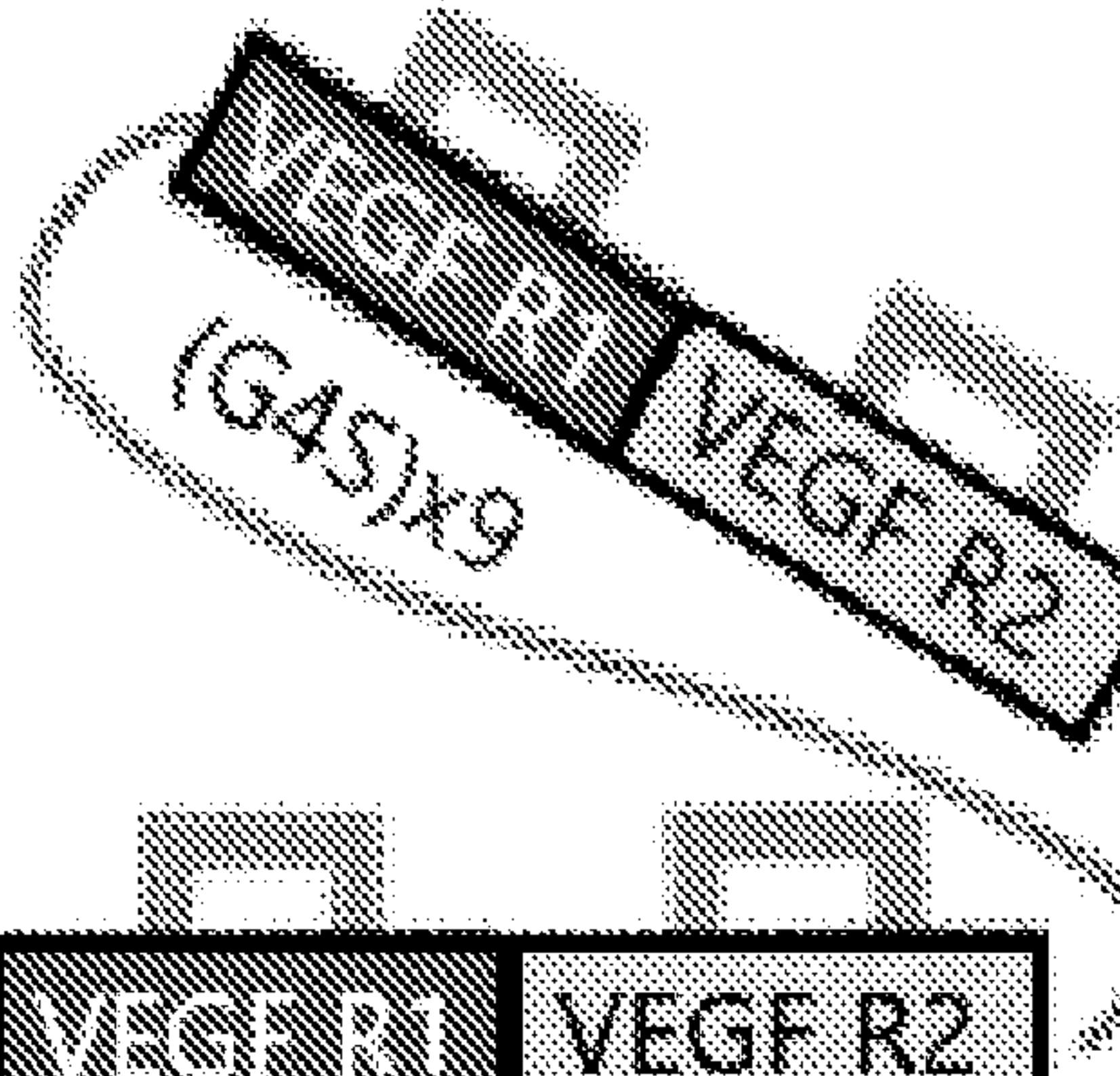
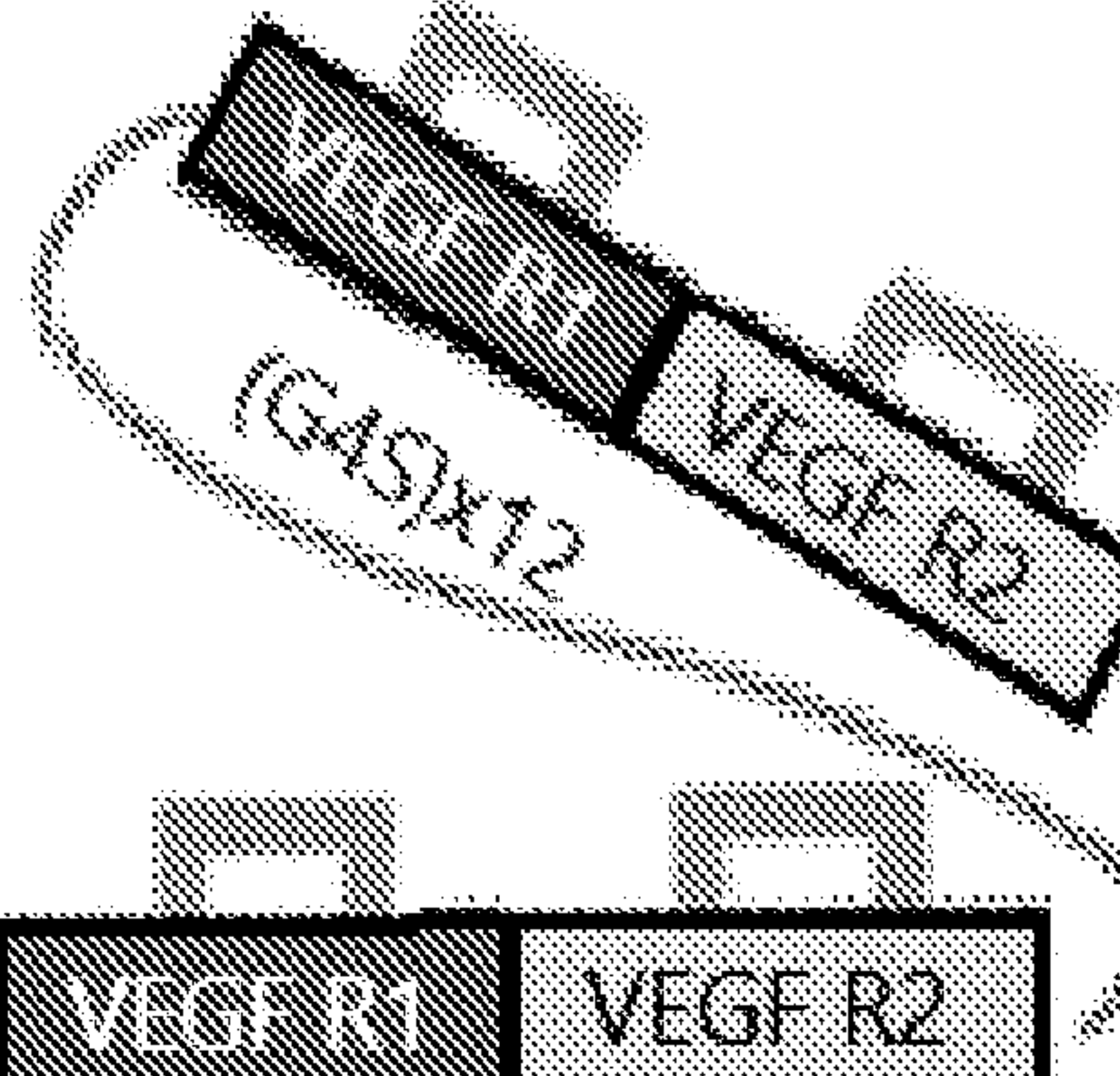
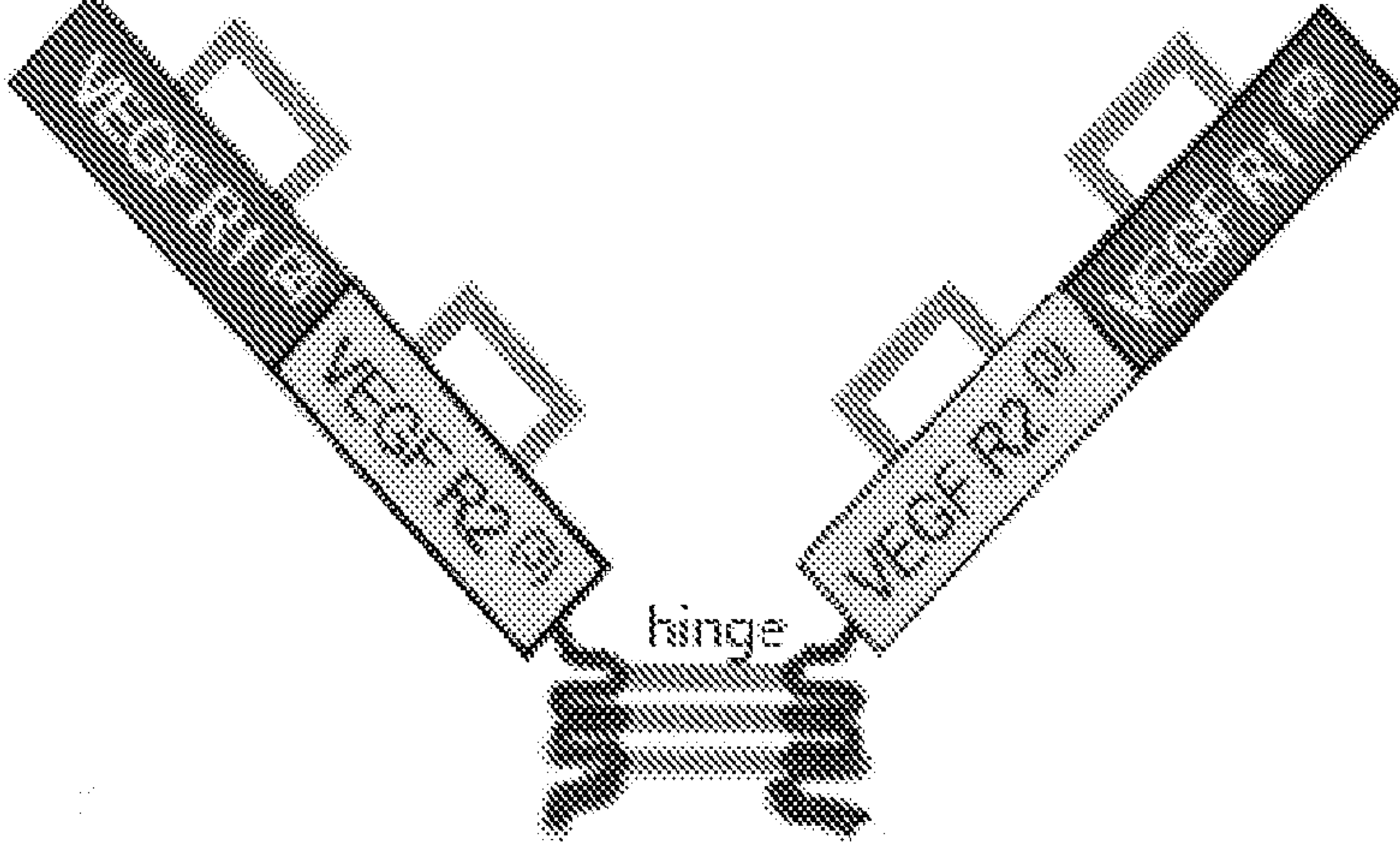
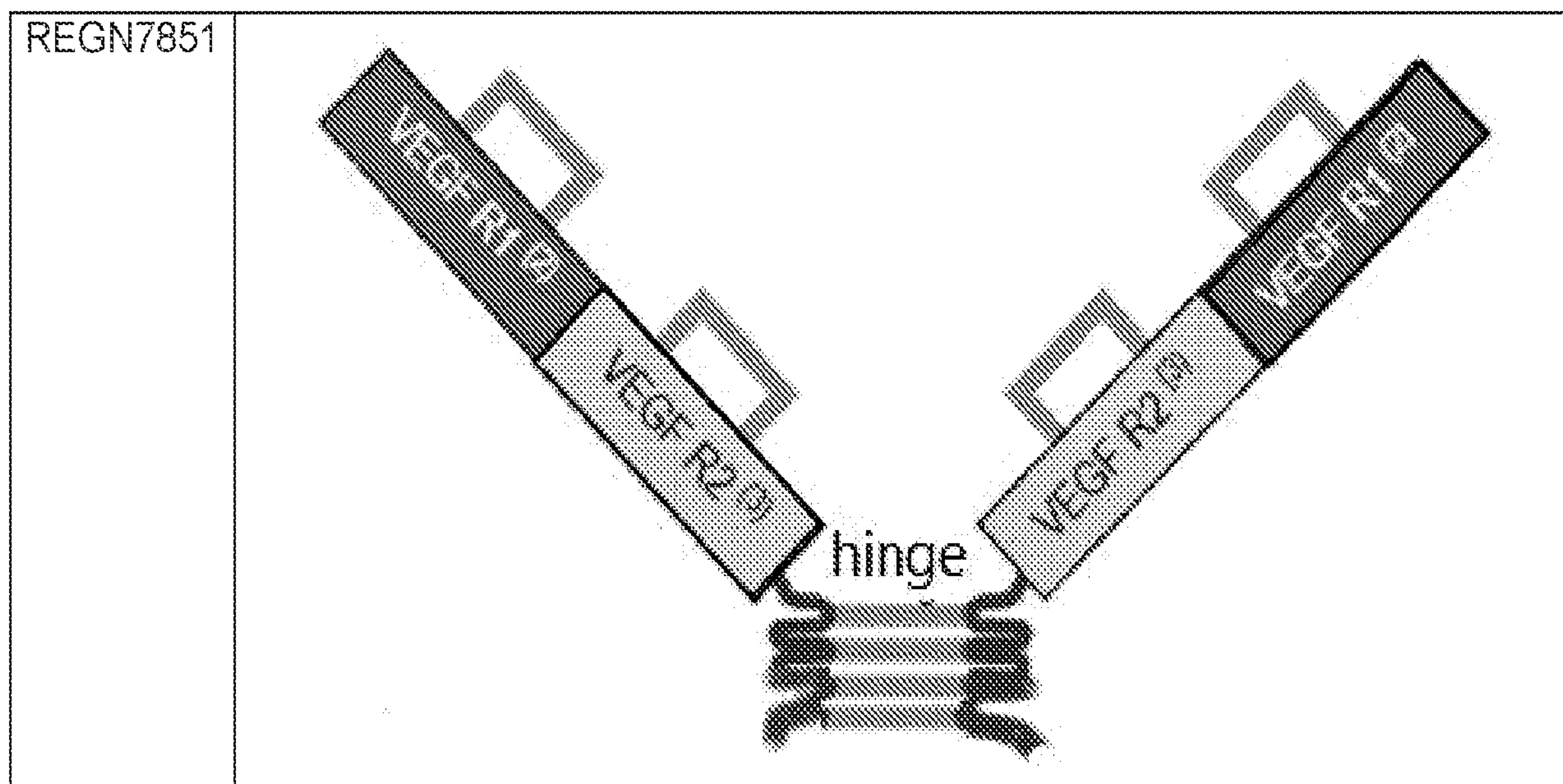
REGN7080	 VEGF R1 is VEGF Receptor 1 Ig-like domain 2 VEGF R2 is VEGF Receptor 2 Ig-like domain 3
REGN7991	 VEGF R1 is VEGF Receptor 1 Ig-like domain 2 VEGF R2 is VEGF Receptor 2 Ig-like domain 3
REGN7992	 VEGF R1 is VEGF Receptor 1 Ig-like domain 2 VEGF R2 is VEGF Receptor 2 Ig-like domain 3
REGN7850	 hinge

FIG. 12 cont.



VEGF R1 (2) is the VEGF Receptor 1 Ig-like domain 2

VEGF R2 (3) is the VEGF Receptor 2 Ig-like domain 3

 and , in the hinge region, represents a disulfide bridge.

FIG. 13

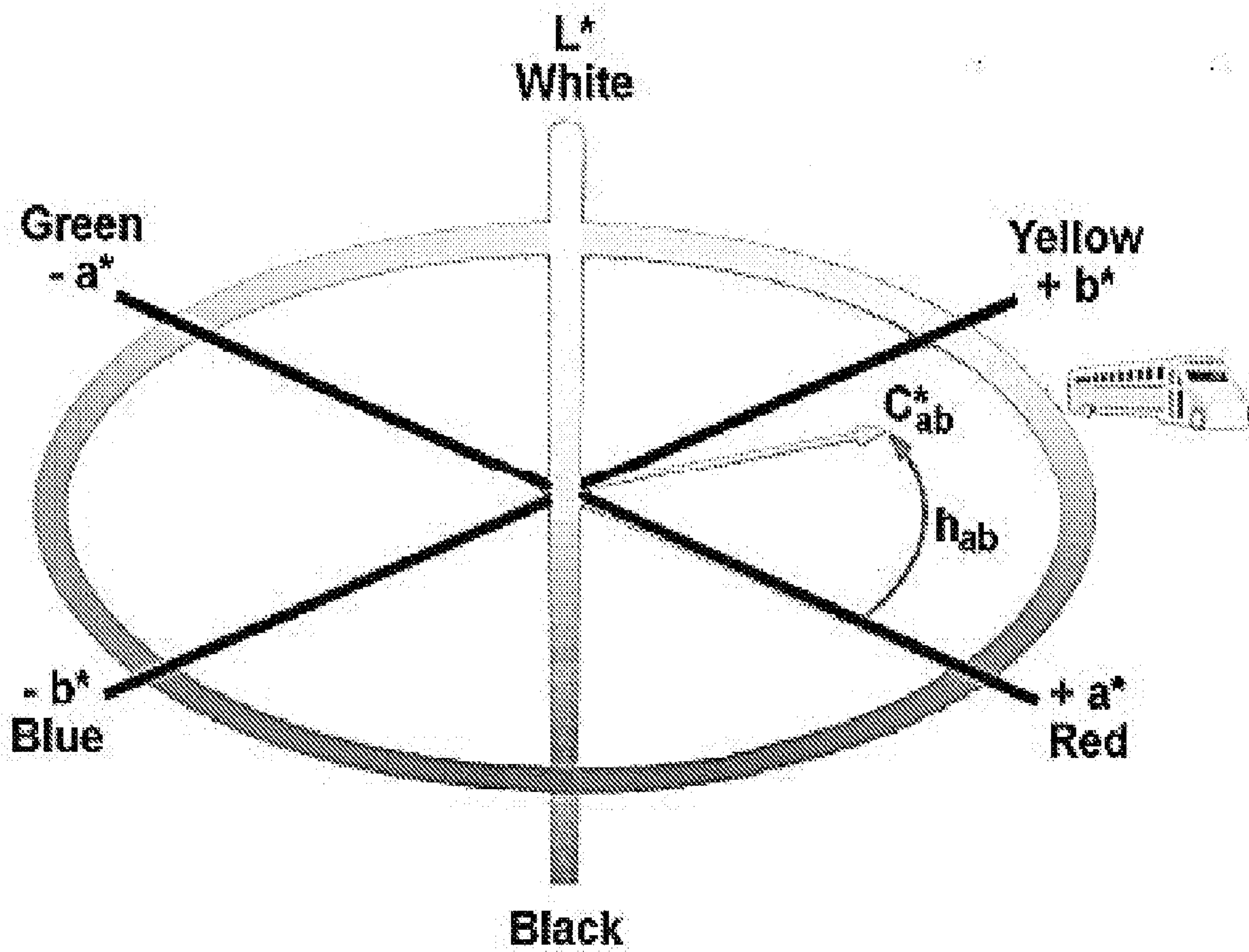


FIG. 14A

Glycan Annotation	Possible Glycan Structure	Glycosylation site								
		REGN7483 ^F				Eylea				
		N36	N68	N123	N195	N36	N68	N123	N196	N282
G0-GlcNAc		x	x	1.0%	1.4%	x	x	0.1%	0.2%	x
G1-GlcNAc		x	x	4.8%	2.7%	x	x	1.0%	0.5%	x
G1S-GlcNAc		x	x	4.1%	2.2%	x	x	2.9%	1.4%	x
G0		x	x	x	x	x	x	x	x	12.0%
G1		x	x	x	6.1%	x	x	x	3.1%	8.4%
G1S		x	x	x	1.9%	x	x	x	3.4%	x
G2		x	x	11.5%	18.1%	x	x	10.1%	16.5%	x
G2S		x	x	14.5%	18.4%	x	x	32.7%	36.9%	x
G2S2		x	x	1.5%	3.7%	x	x	7.1%	16.9%	x
G0F		x	x	x	x	x	x	x	x	28.7%
G2F2S		2.0%	2.0%	x	x	0.8%	1.3%	x	x	x
G2F2S2		1.6%	0.5%	x	x	1.9%	0.9%	x	x	x
G1F		5.6%	6.1%	x	x	0.3%	0.5%	x	x	40.1%
G1FS		x	3.8%	x	x	x	0.8%	x	x	x
G2F		20.2%	28.0%	1.8%	2.1%	4.9%	11.0%	1.7%	1.8%	10.8%
G2FS		35.2%	48.9%	2.8%	2.2%	29.2%	56.5%	5.6%	4.0%	x
G2FS2		22.4%	9.1%	0.3%	0.6%	41.6%	26.8%	1.0%	2.0%	x
G3FS		3.4%	1.6%	x	x	2.1%	2.2%	x	x	x
G3FS3		1.7%	x	x	x	5.2%	x	x	x	x
G0-2GlcNAc		x	x	3.4%	2.6%	x	x	0.2%	0.1%	x
Man4		x	x	0.5%	1.6%	x	x	0.2%	0.2%	x
Man4_G1		x	x	3.6%	2.1%	x	x	2.7%	0.8%	x
Man4_G1S1		x	x	4.6%	3.0%	x	x	9.9%	3.2%	x
Man5		x	x	32.4%	27.1%	x	x	13.1%	5.5%	x
Man5_G1		x	x	4.8%	2.8%	x	x	3.9%	1.1%	x
Man5_G1S1		x	x	3.3%	1.5%	x	x	7.5%	2.2%	x
Man6		x	x	1.3%	x	x	x	x	x	x
Man6_G0+Phosphate		1.7%	x	x	x	1.4%	x	x	x	x
Man6+Phosphate		6.2%	x	x	x	12.5%	x	x	x	x
Man7		x	x	3.6%	x	x	x	0.1%	x	x

Min

Max

- Fucose (Fuc, F)
 N-acetylglucosamine (GlcNAc)
 Mannose (Man)
 Galactose (Gal, G)
 N-acetylneuraminic acid (Sialic Acid, S)
 P = Phosphate

FIG. 14B

PTM	Site	Peptide sequence	Eylea	REGN7483 ^F
Asn Deamidation	Asn84	EIGLLTCEATVNGHLYK	35.1%	26.9%
	Asn99	QTNTIIDVVLSPSHGIELSVGEK	0.7%	0.7%
	Asn152	LVNR	2.7%	2.7%
Asp Succinimide+isomerization	Asp173	FLSTLTIDGVTR	2.2%	1.9%
Met Oxidation	Met10	SDTGRPFVEMYSEIPEIIHMTEGR	9.5%	5.5%
	Met20	SDTGRPFVEMYSEIPEIIHMTEGR	8.4%	1.9%
	Met163	TQSGSEMK	4.7%	7.3%
	Met192	SDQGLYTCAASSGLMTK	8.2%	6.6%
Trp DiOxidation	Trp58	IIWDSR	0.2%	0.3%
Arg 3-deoxyglucosone	Arg5	SDTGRPFVEMYSEIPEIIHMTEGR	6.7%	8.1%
C-term loss	Gly221	THTCPPCPAPELLG	NA	7.2%
Non-glycosylated glycosite	Asn36	VTSPNITVTLK	0.3%	1.7%
	Asn68	GFIISNATYK	34.5%	47.3%
	Asn123	LVLNCTAR	0.0%	0.2%
	Asn196	NSTFVR	0.2%	0.8%
	Asn282	EEQYNSTYR	0.3%	N/A

FIG. 14C

³²VTSPN³⁶ITVTLK⁴²

Glycans at N36	VEGF trap_ RefStd	REGN7483 ^F	REGN7483 ^R	REGN7711
G0F-GlcNAc	0.3%	2.0%	1.8%	1.8%
G1F	0.3%	3.2%	1.0%	1.4%
G1F-GlcNAc	0.6%	4.8%	4.6%	4.9%
G1FS-GlcNAc	1.3%	3.1%	3.8%	3.1%
G2F	2.8%	17.4%	15.1%	19.8%
G2F2S	1.4%	1.7%	2.0%	2.2%
G2FS	24.8%	34.2%	31.5%	31.9%
G2FS2	42.4%	20.4%	25.8%	19.0%
G3FS	2.1%	2.3%	4.0%	5.5%
G3FS2	6.6%	2.6%	4.7%	5.0%
G3FS3	8.8%	1.1%	2.4%	1.9%
G1_Man5+Phos	0.0%	1.2%	0.3%	0.2%
Man6+Phos	8.3%	5.7%	2.5%	2.8%

FIG. 14C cont.

⁶²KGFIISN⁶⁸ATYK⁷²

⁶³GFIISN⁶⁸ATYK⁷²

Glycans at N68	VEGF trap_ RefStd	REGN7483 ^F	REGN7483 ^R	REGN7711
G0F-GlcNAc	0.2%	1.2%	1.1%	1.1%
G1F	0.8%	5.1%	1.4%	1.7%
G1F-GlcNAc	0.9%	3.9%	3.9%	4.0%
G1FS	0.5%	1.2%	0.4%	0.4%
G1FS1-GlcNAc	0.7%	1.2%	1.6%	1.4%
G2F	9.3%	27.4%	23.6%	28.6%
G2F2S	2.2%	2.2%	3.0%	3.4%
G2FS	67.1%	52.4%	55.2%	50.2%
G2FS2	14.8%	3.9%	6.9%	5.8%
G3FS	1.4%	0.5%	1.2%	1.6%
G3FS2	1.7%	0.4%	1.1%	1.2%

FIG. 14C cont.

¹²⁰LVLN¹²³CTAR¹²⁷

Glycans at N123	VEGF trap_ RefStd	REGN7483 ^F	REGN7483 ^R	REGN7711
G0-GlcNAc	1.0%	3.5%	3.7%	3.5%
G1-GlcNAc	2.2%	6.2%	6.8%	6.4%
G1S-GlcNAc	3.7%	4.1%	3.5%	2.8%
G2	8.4%	10.6%	16.7%	17.1%
G2F	1.3%	1.5%	7.2%	7.0%
G2FS	5.7%	2.1%	13.6%	14.2%
G2S	34.8%	12.7%	26.1%	25.5%
G2S2	6.9%	1.3%	5.0%	6.6%
G1_Man4	2.2%	3.8%	1.3%	1.4%
G1S_Man4	10.6%	3.9%	2.1%	1.8%
G1_Man5	3.0%	4.0%	1.2%	1.1%
G1S_Man5	7.1%	3.2%	1.4%	1.4%
Man4	0.5%	2.6%	1.9%	1.8%
Man5	10.9%	35.5%	4.3%	3.1%
Man6	0.0%	1.1%	0.1%	0.1%
Man7	0.1%	2.8%	0.1%	0.1%

FIG. 14C cont.

¹⁹⁵K¹⁹⁶NSTFVR²⁰¹¹⁹⁶NSTFVR²⁰¹

Glycans at N196	VEGF trap_ RefStd	REGN7483 ^F	REGN7483 ^R	REGN7711
G0-GlcNAc	0.7%	1.9%	1.8%	1.9%
G1	4.4%	4.1%	3.6%	4.2%
G1-GlcNAc	0.8%	1.9%	2.5%	2.4%
G1S-GlcNAc	1.8%	2.9%	2.6%	1.8%
G2	14.0%	20.7%	28.2%	32.1%
G2F	1.9%	2.0%	5.1%	6.0%
G2FS	4.7%	2.0%	6.1%	6.2%
G2FS2	1.8%	0.5%	1.6%	1.3%
G2S	39.3%	17.7%	31.2%	29.9%
G2S2	17.9%	4.4%	9.7%	6.7%
G3S	0.4%	0.1%	0.7%	1.0%
G1S_Man4	1.5%	1.0%	0.3%	0.3%
G1_Man5	1.0%	2.3%	0.5%	0.5%
Man3	0.2%	3.1%	0.7%	0.6%
Man4	0.2%	2.7%	0.8%	0.6%
Man5	5.6%	30.4%	3.6%	3.4%

FIG. 14D

Additional glycan structures:

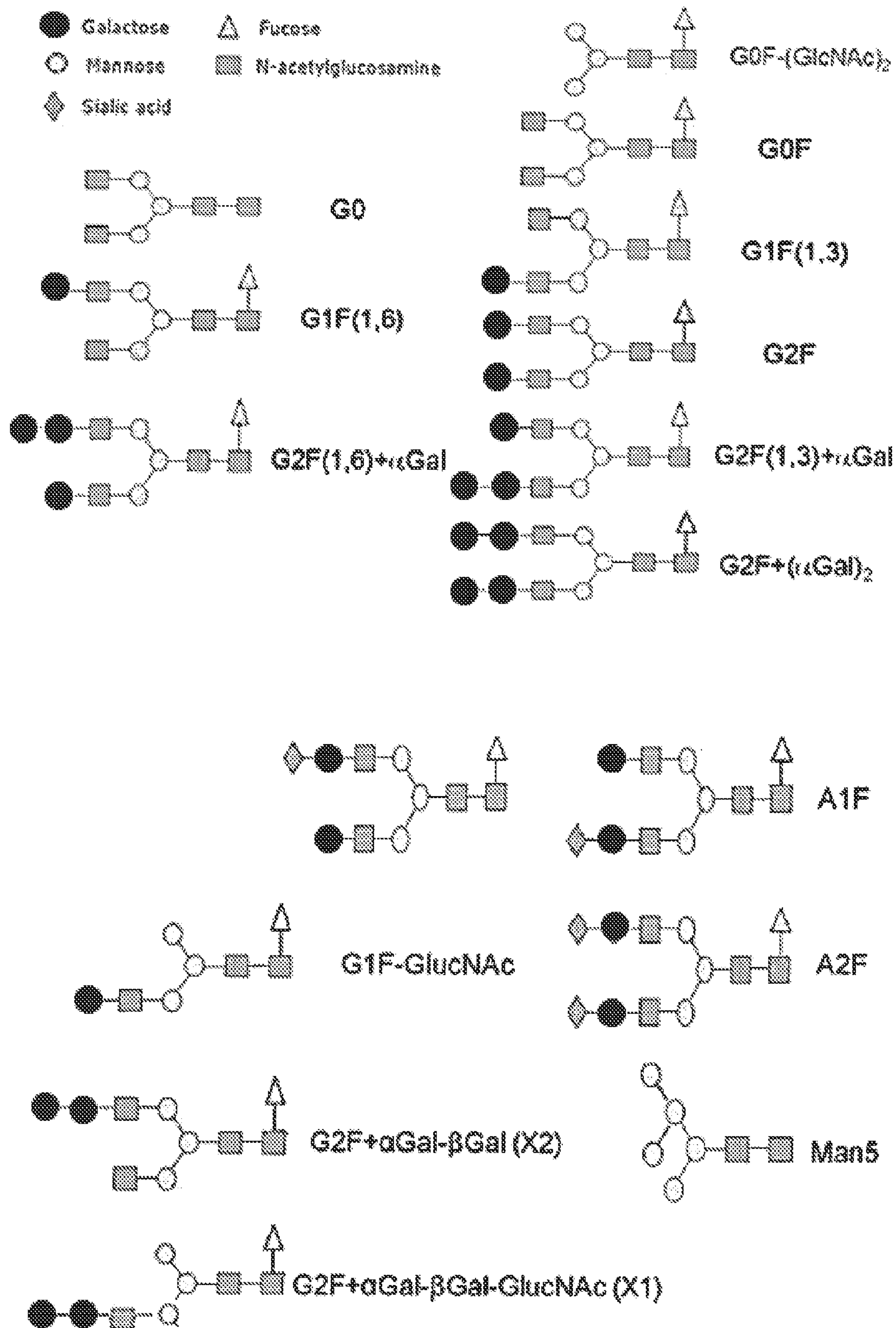


FIG. 15

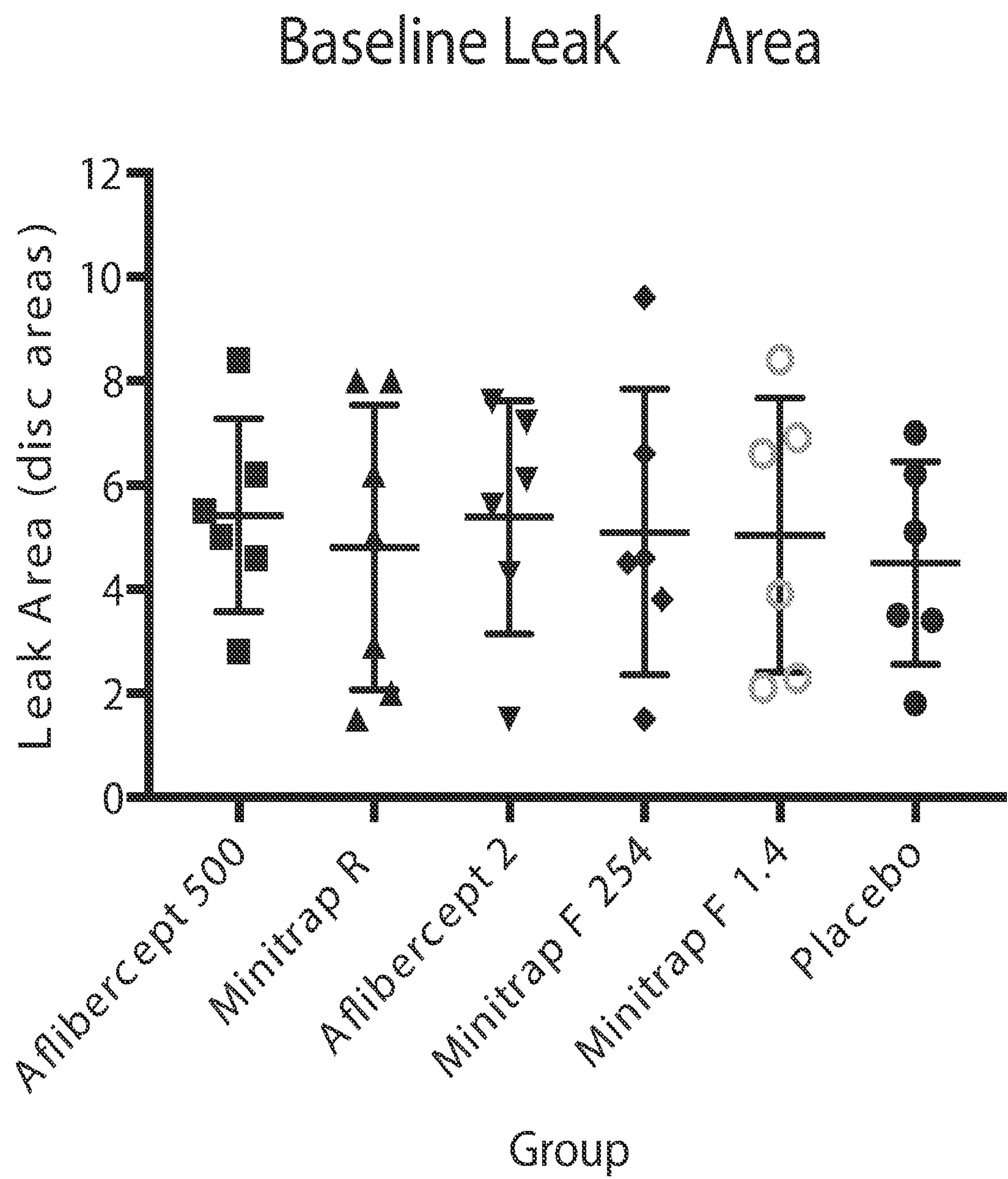


FIG. 16

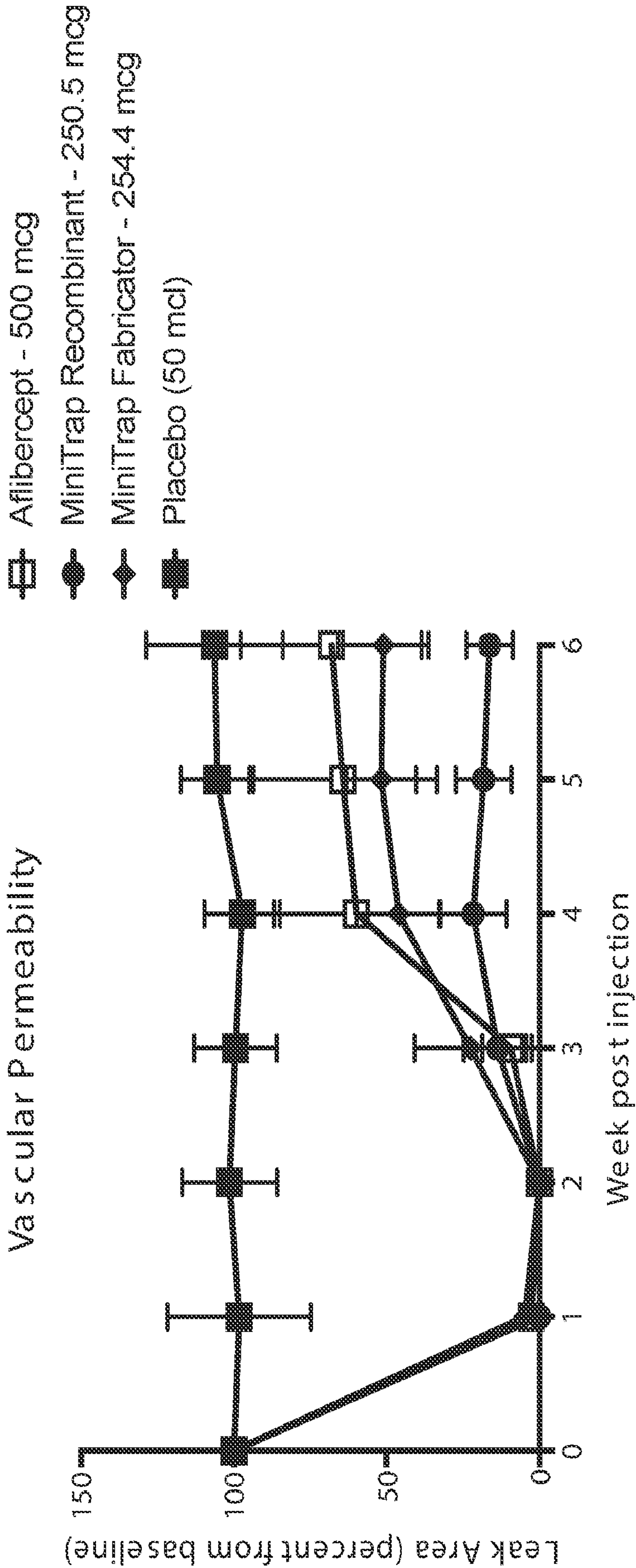


FIG. 17

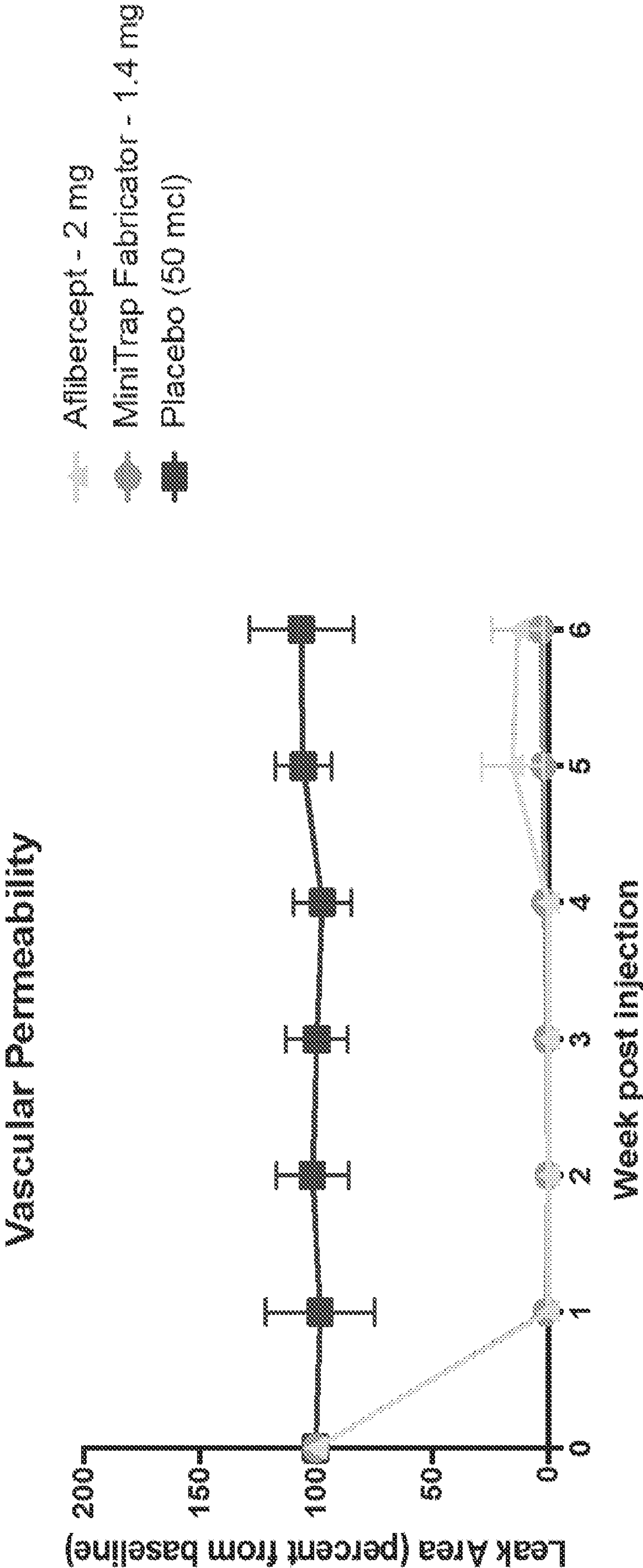


FIG. 18

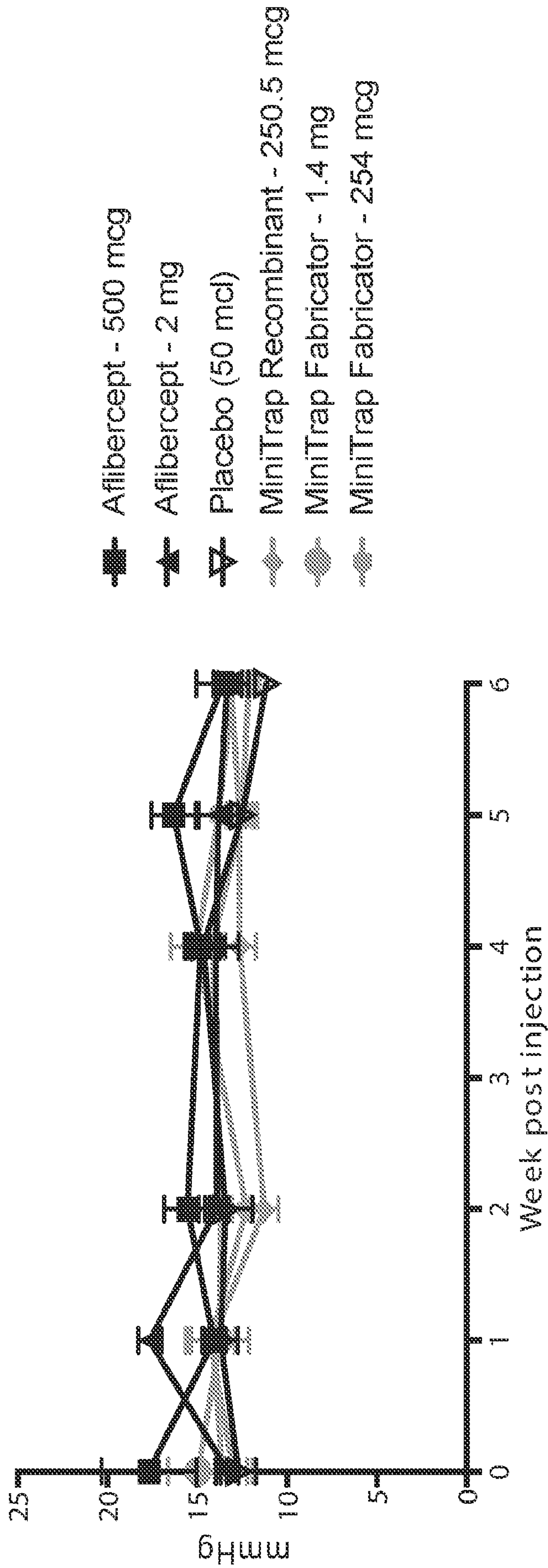
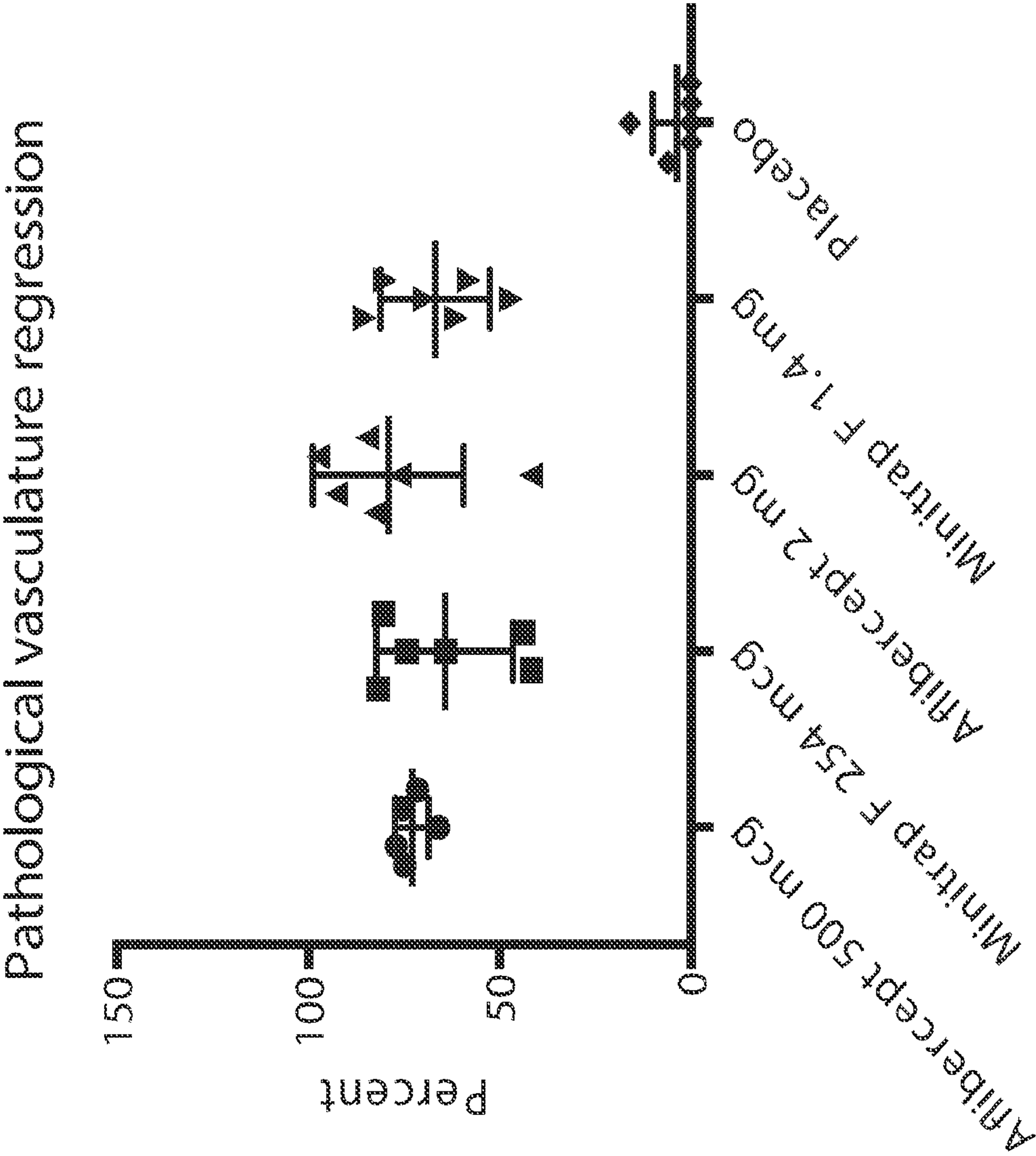


FIG. 19



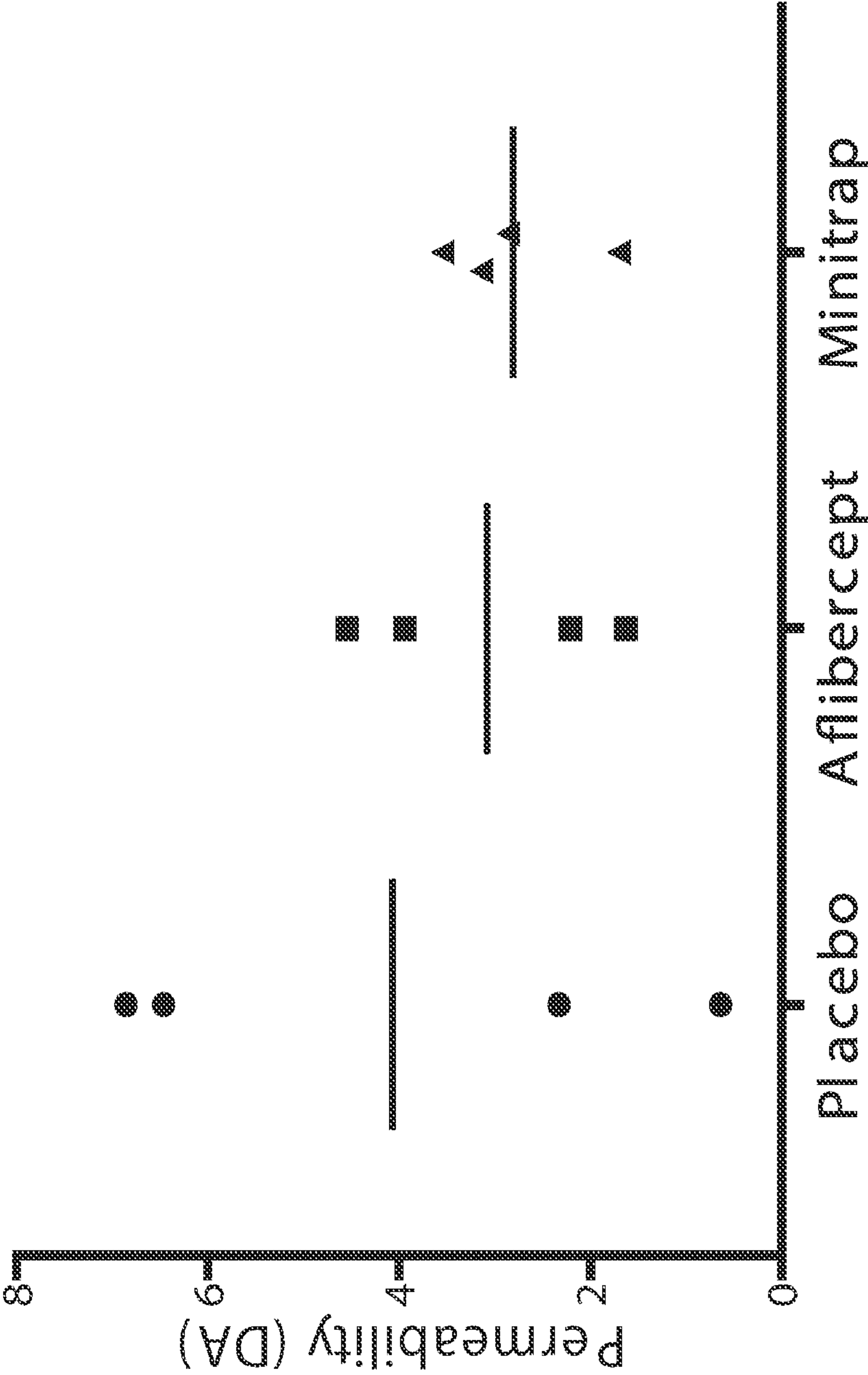


FIG. 20

FIG. 21

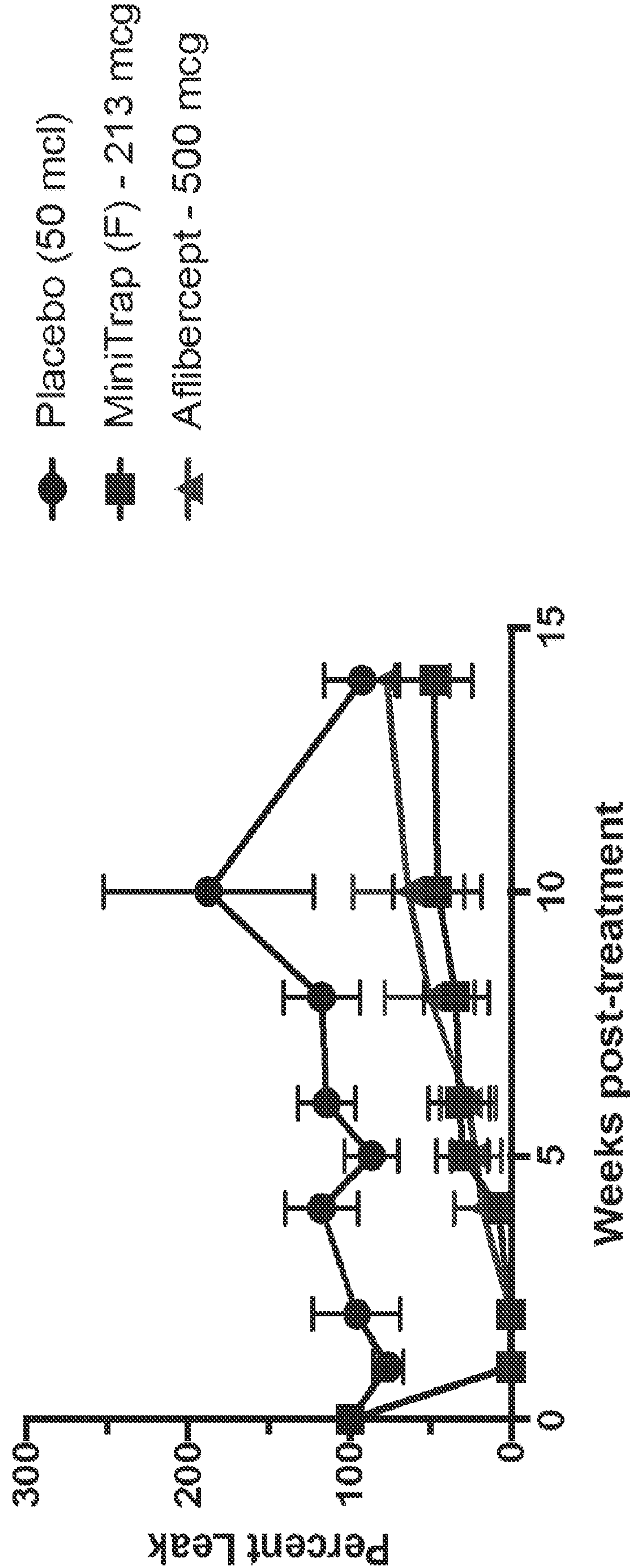


FIG. 22

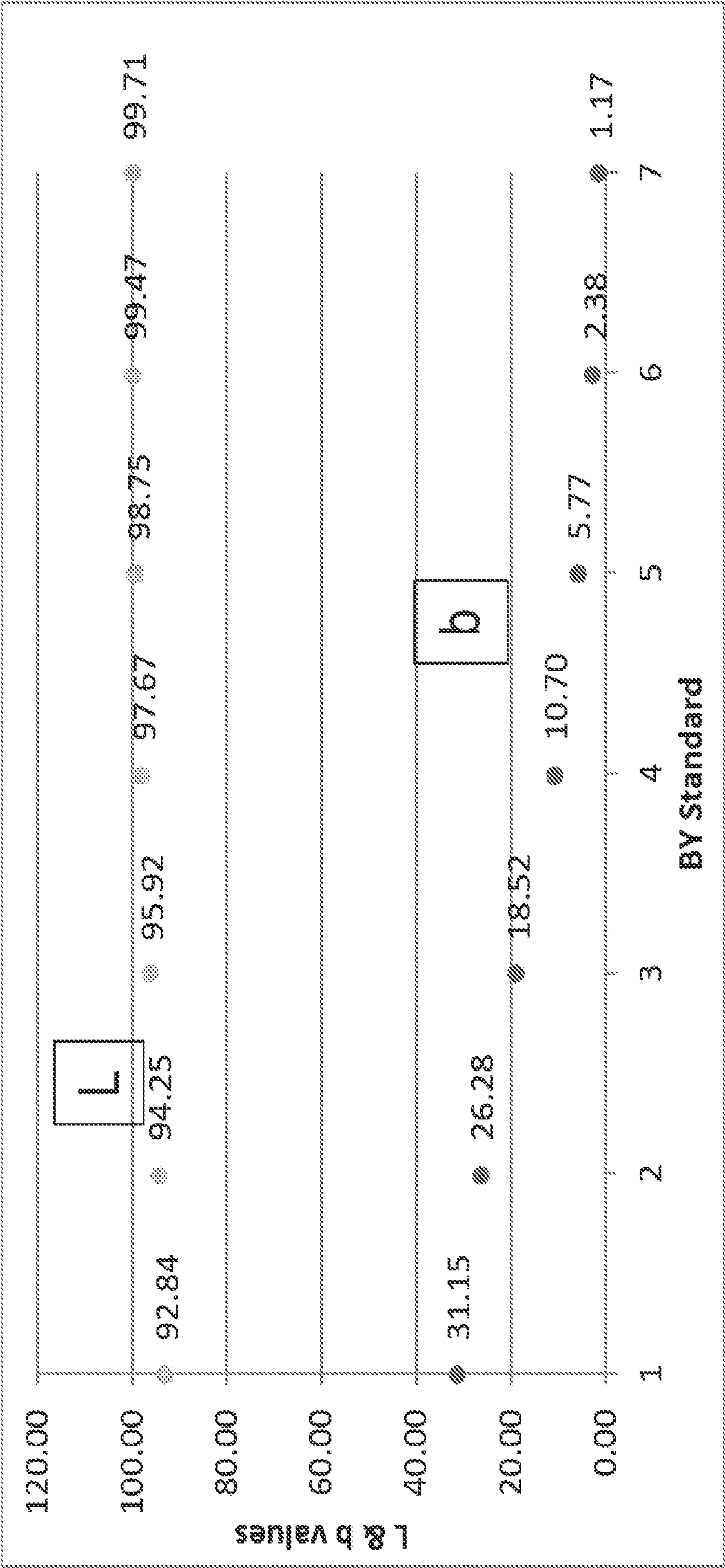


FIG. 23

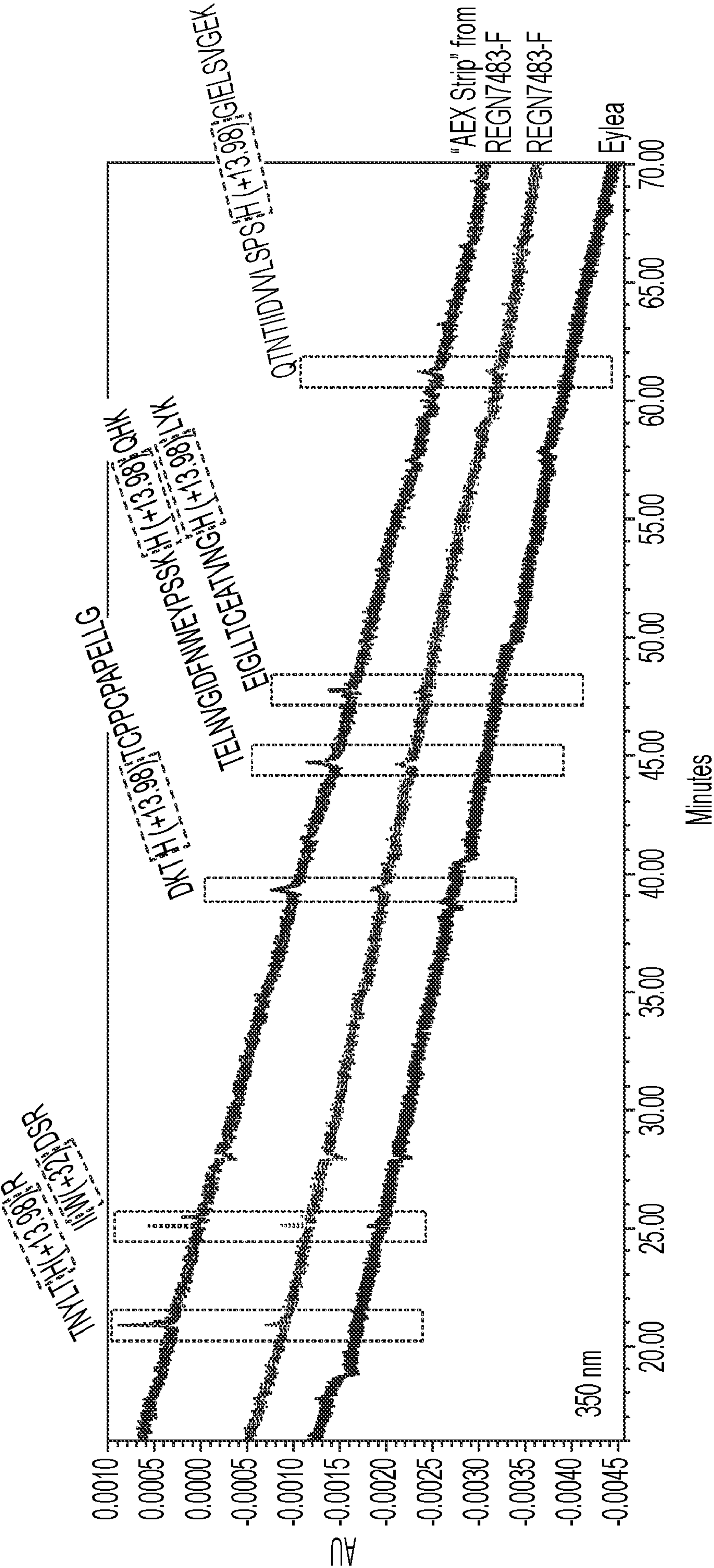


FIG. 24A

Parameter Estimates					
Term	Estimate	Std Error	t Ratio	Prob> t	VIF
Intercept	2.1696718	0.045304	47.89	<.0001*	.
Cysteine(0,16.6)	1.6083682	0.04544	35.40	<.0001*	1.0224715
B-Vits Group(0,2)	0.1731314	0.045861	3.78	0.0008*	1.0441432
Iron(0,230)	0.3235496	0.046683	6.93	<.0001*	1.0791787
Zinc(0,0.536)	0.2495801	0.047522	5.25	<.0001*	1.1211453
Folic Acid(0,173)	0.175624	0.047137	3.73	0.0009*	1.1002915
Cysteine*B-Vits Group	0.1175578	0.045586	2.58	0.0155*	1.0290749
Cysteine*Iron	0.335687	0.046851	7.17	<.0001*	1.0896913
Cysteine*Zinc	0.1572243	0.046217	3.40	0.0020*	1.0577611
Cysteine*Folic Acid	0.1382794	0.047348	2.92	0.0068*	1.1129431
B-Vits Group*Iron	0.1062442	0.046085	2.31	0.0288*	1.0517205
B-Vits Group*Folic Acid	-0.104795	0.047468	-2.21	0.0356*	1.1157801

mM: Cysteine, Zinc

uM: Iron, Copper, Folic Acid

x: B-vitamins Group

FIG. 24B

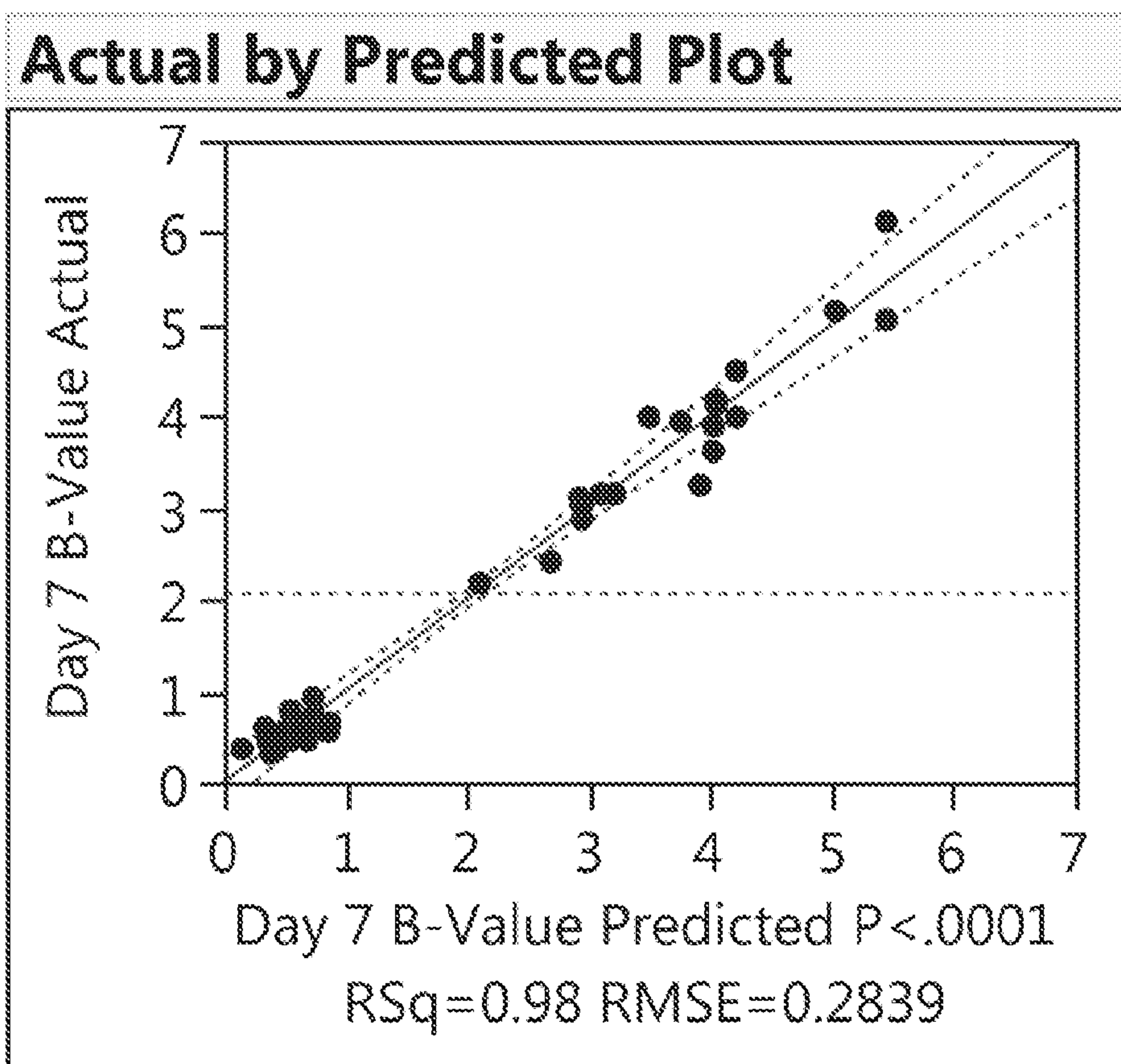
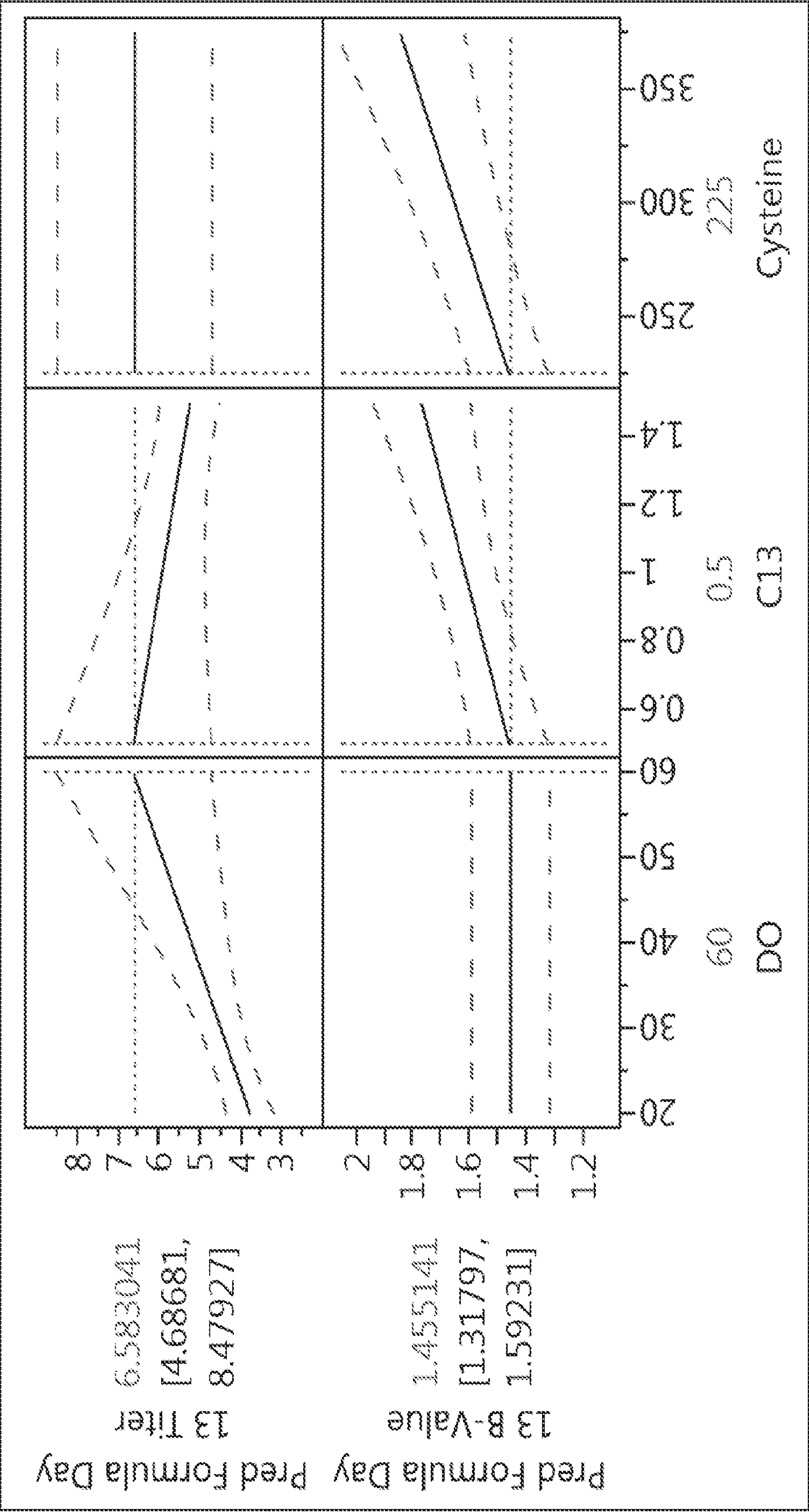
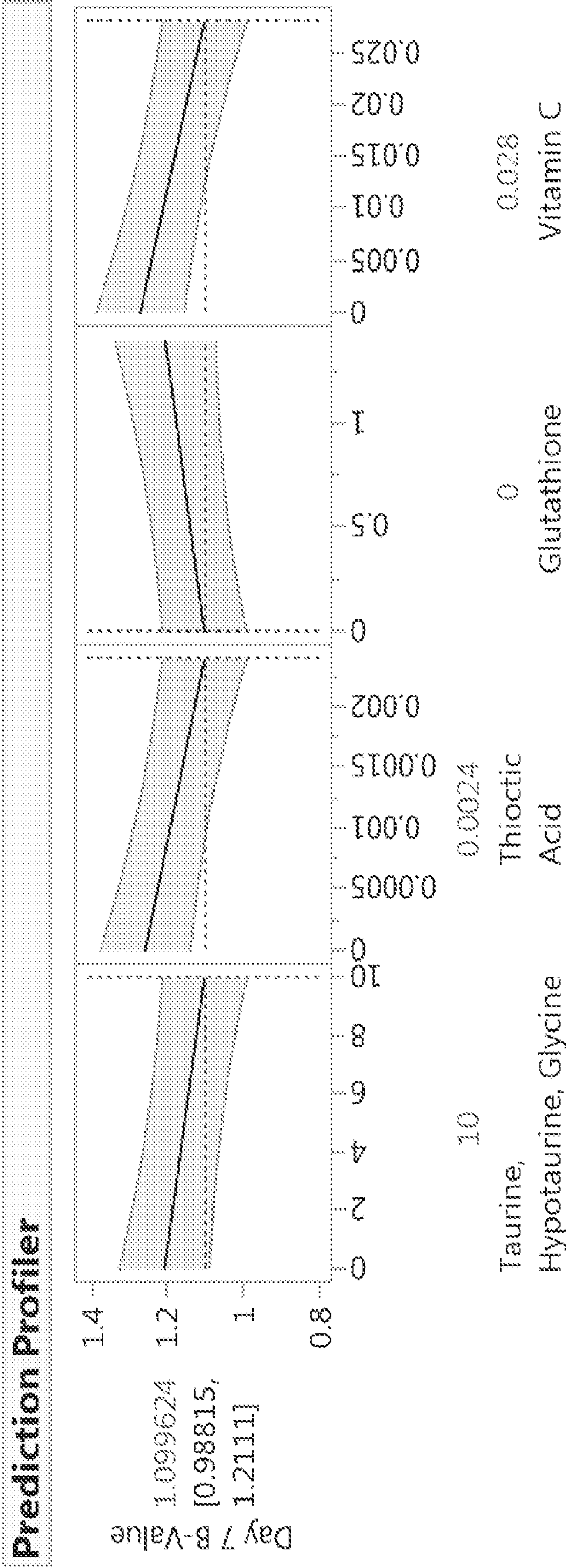


FIG. 25



Control b-value = 1.67

FIG. 26A

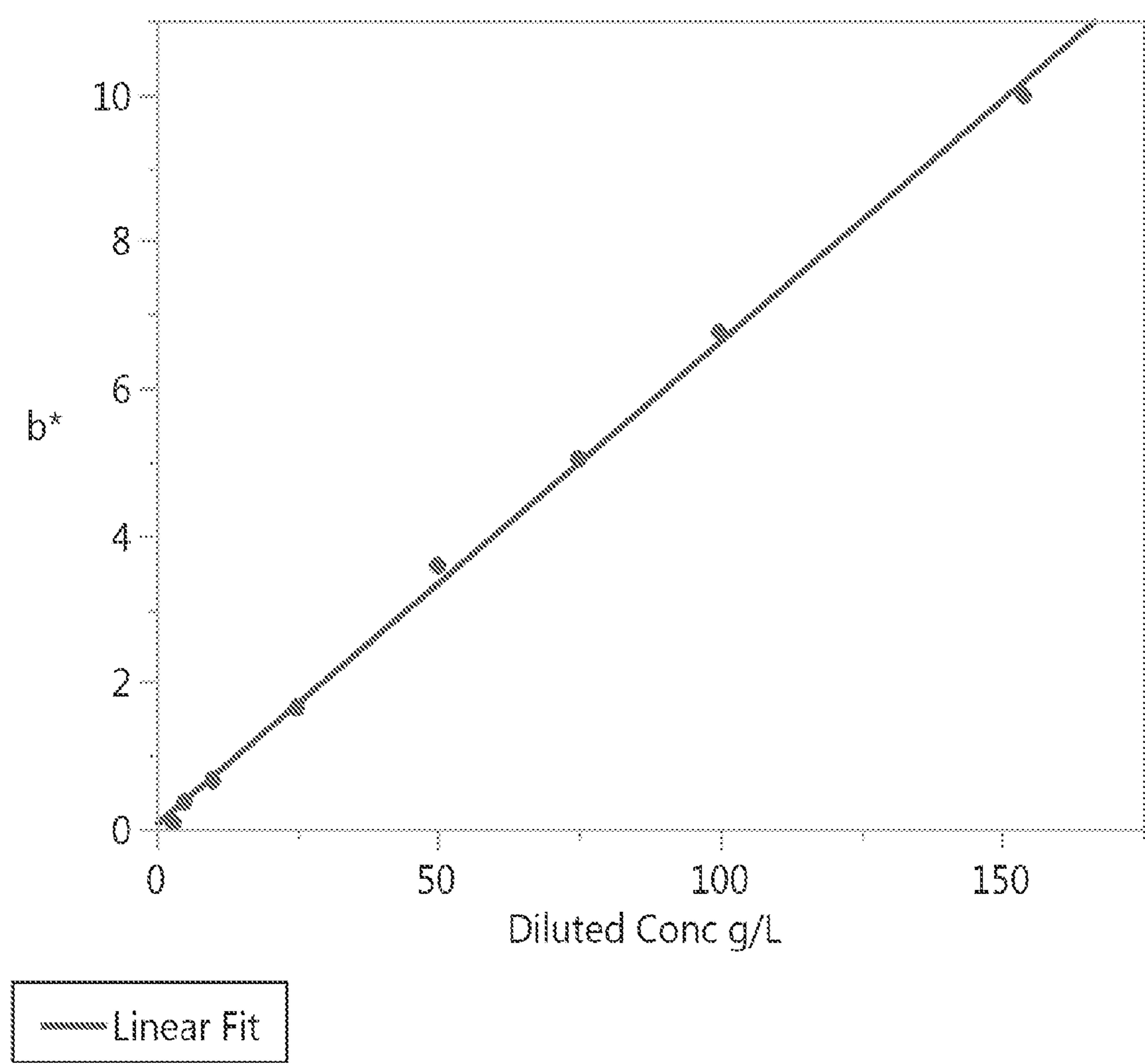


All components in mM. Taurine/hypotaaurine/glycine each at 10 mM

FIG. 26B

Scaled Estimates		Bar Chart			
Term	Scaled Estimate	Std Error	t Ratio	Prob> t	
Intercept	1.3997181	0.022019	63.57	<.0001*	
Taurine, Hypotaaurine, Glycine(0,10)	-0.153727	0.02201	-6.98	<.0001*	
Thiocctic Acid(0,0.0024)	-0.010756	0.022019	-0.49	0.6293	
Glutathione(0,1.4)	0.152492	0.022112	6.90	<.0001*	
Vitamin C(0,0.028)	0.017603	0.02201	0.80	0.4311	
Taurine, Hypotaaurine, Glycine*Glutathione	-0.101323	0.022186	-4.57	0.0001*	
Thiocctic Acid*Vitamin C	-0.06684	0.02201	-3.04	0.0054*	

FIG. 27



Linear Fit

$b^* = 0.0469318 + 0.0656506 \cdot \text{Diluted Conc g/L}$

Summary of Fit

RSquare	0.998283
RSquare Adj	0.997997

Fig. 28

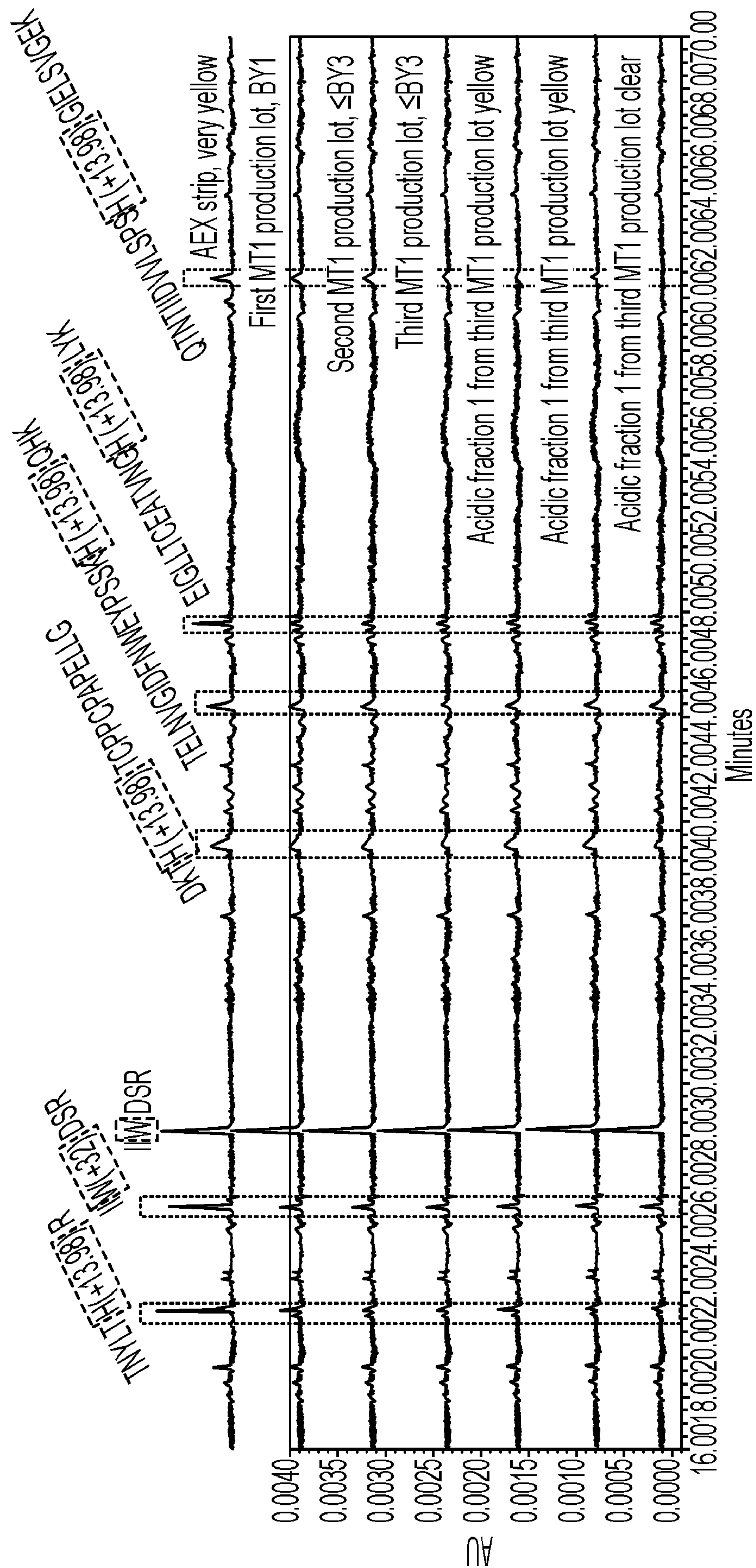


FIG. 29

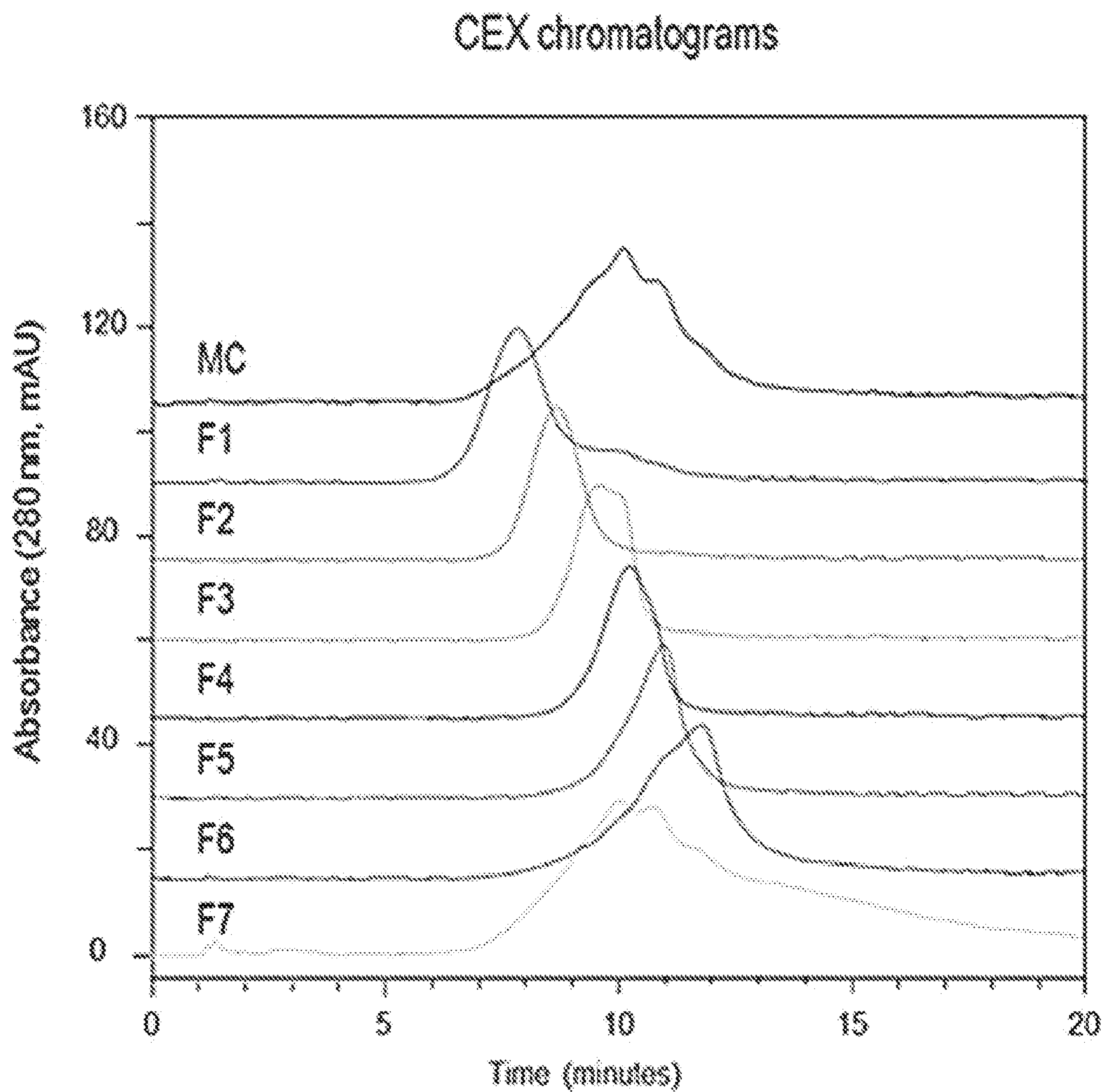


FIG. 30

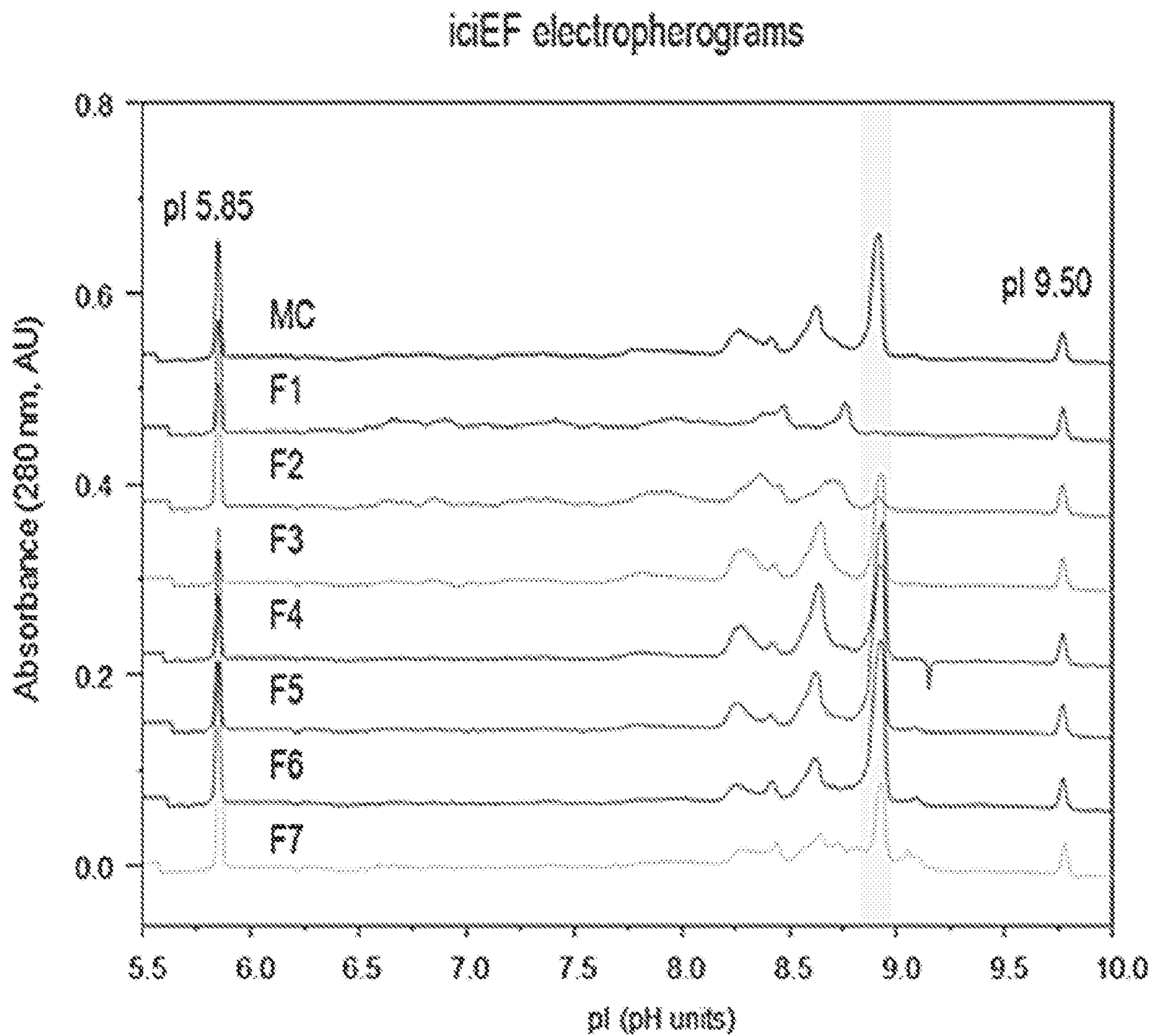
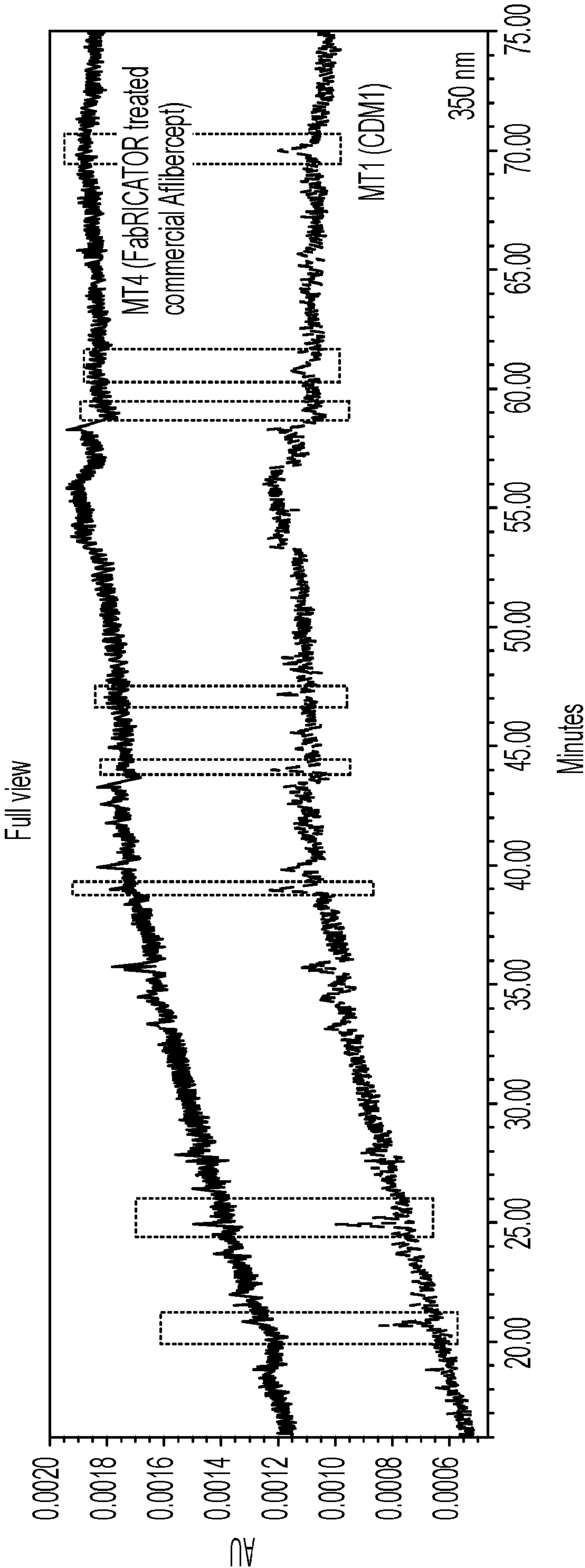


FIG. 31A



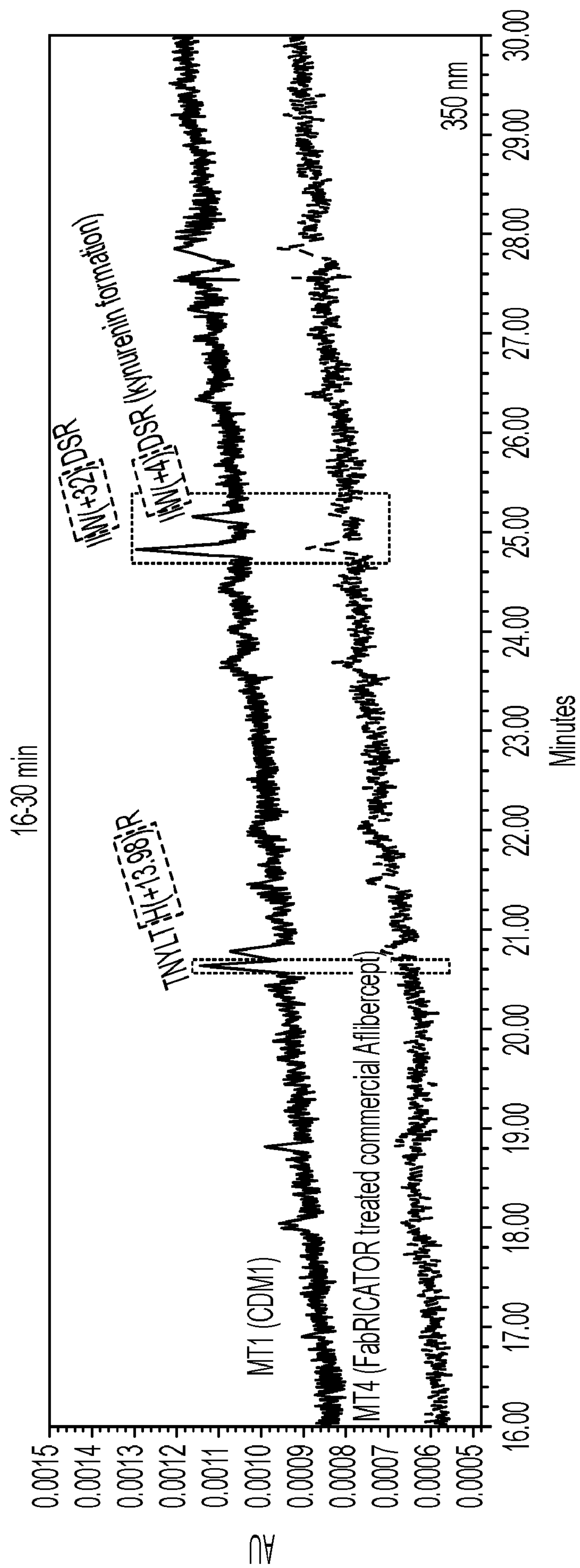


FIG. 31C

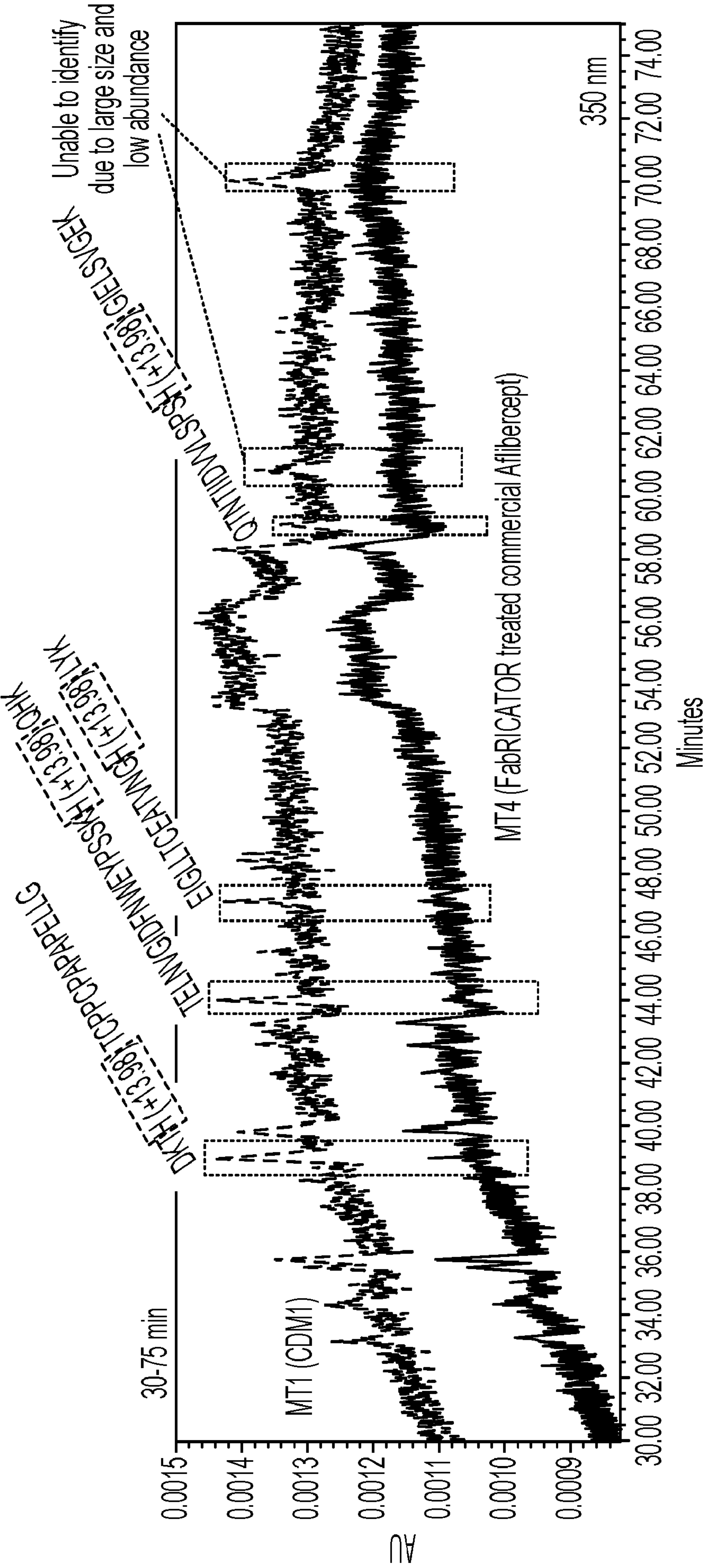


FIG. 32A

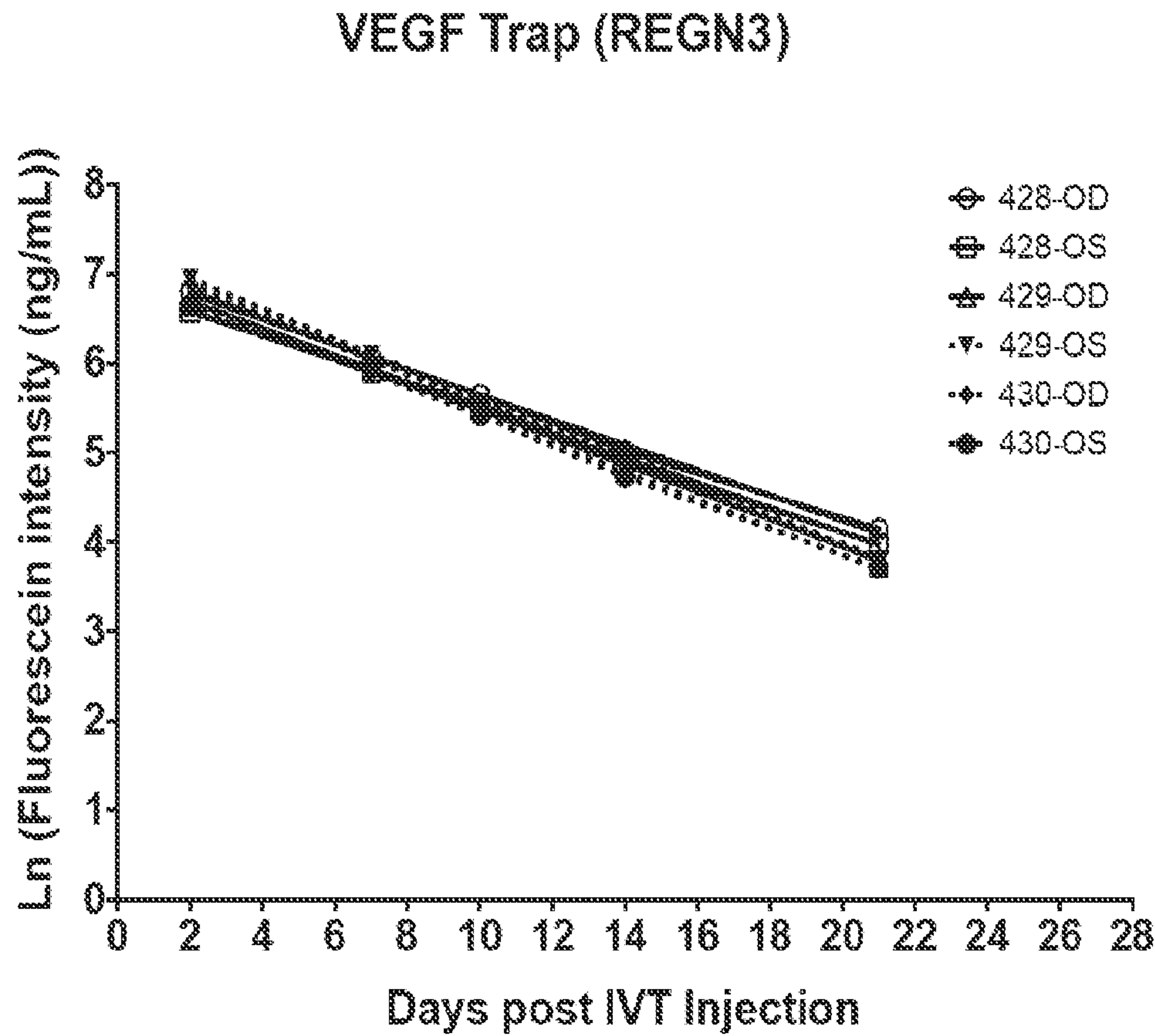


FIG. 32B

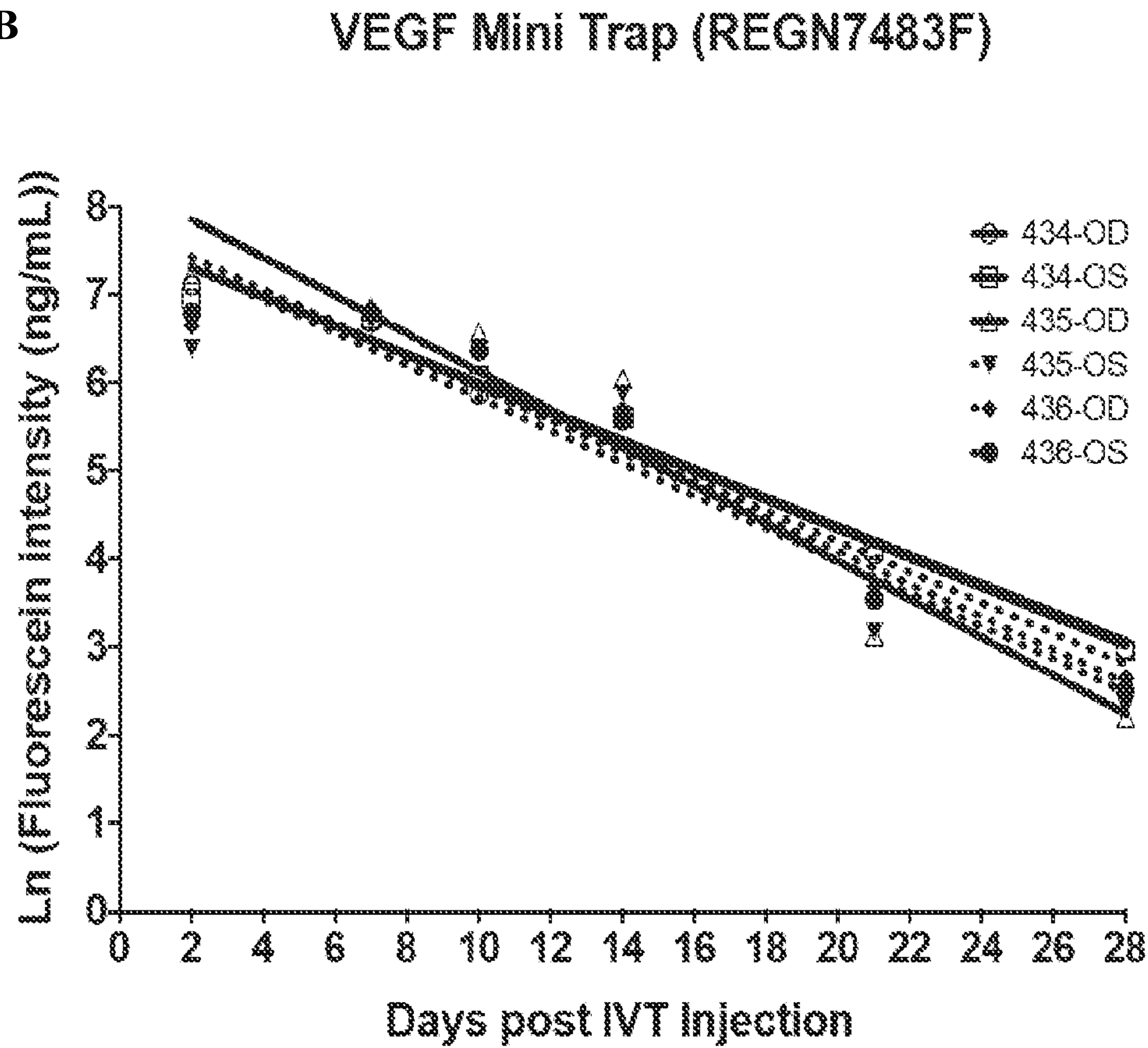


FIG. 33A

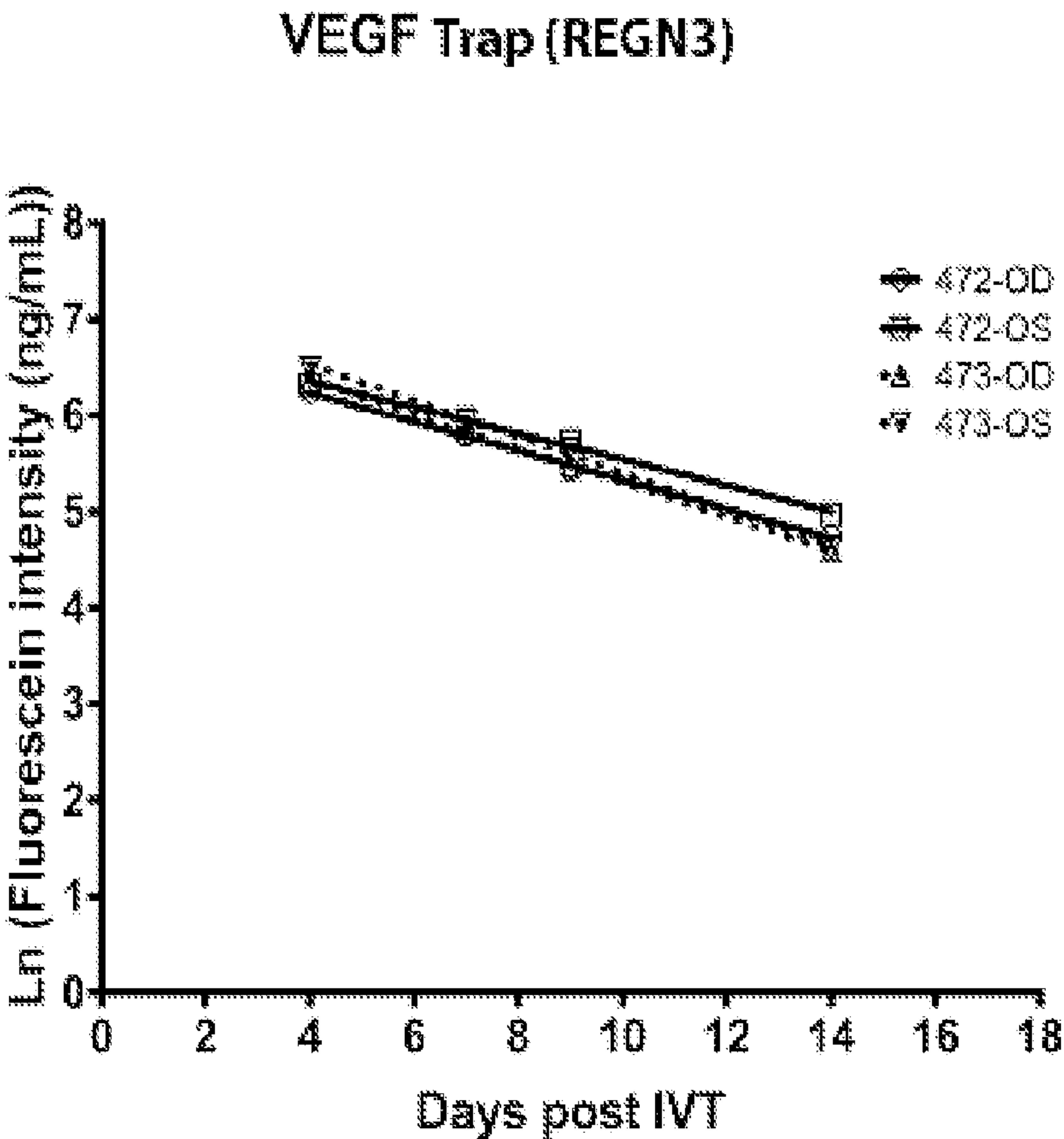


FIG. 33B

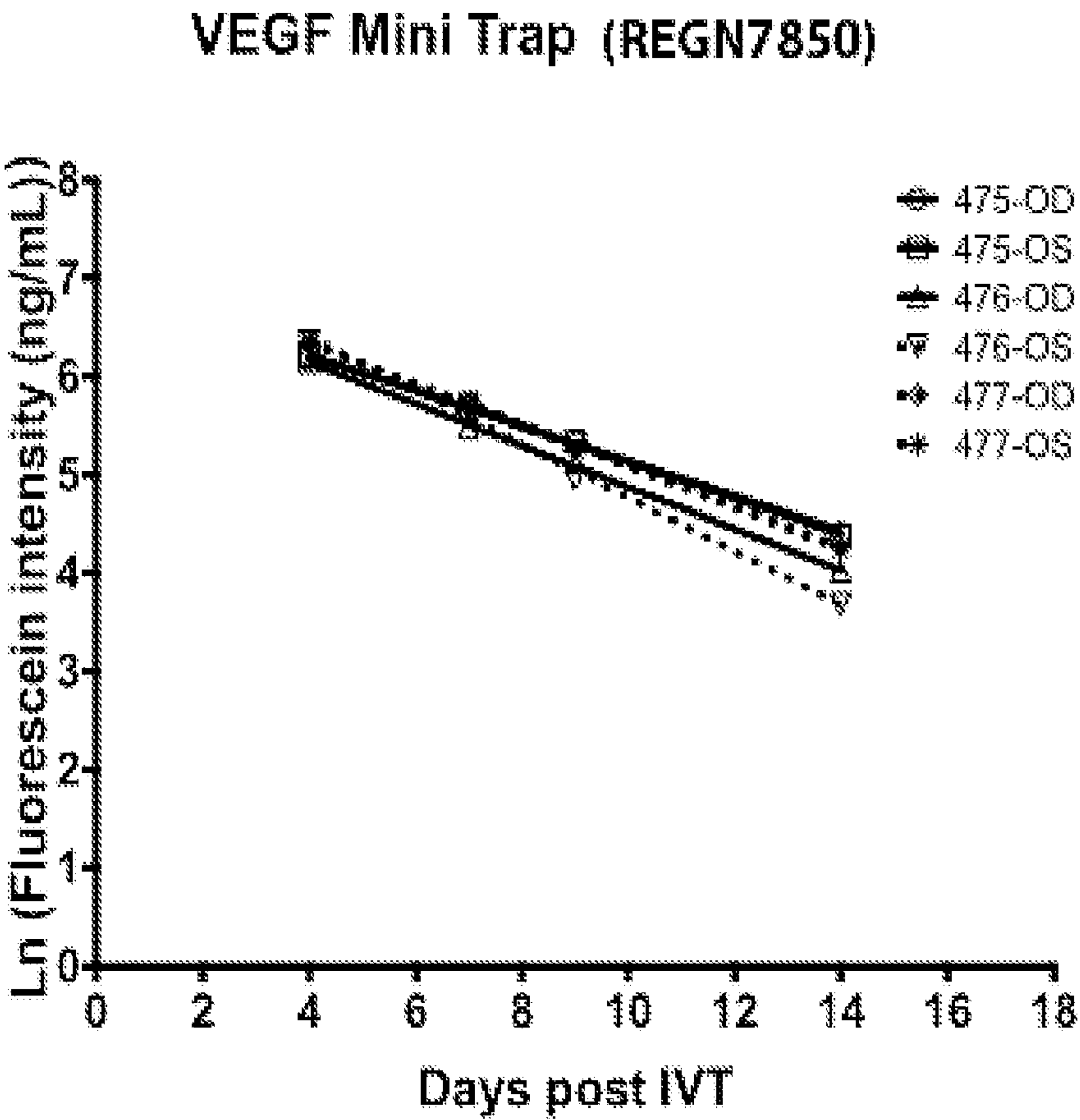


FIG. 33C

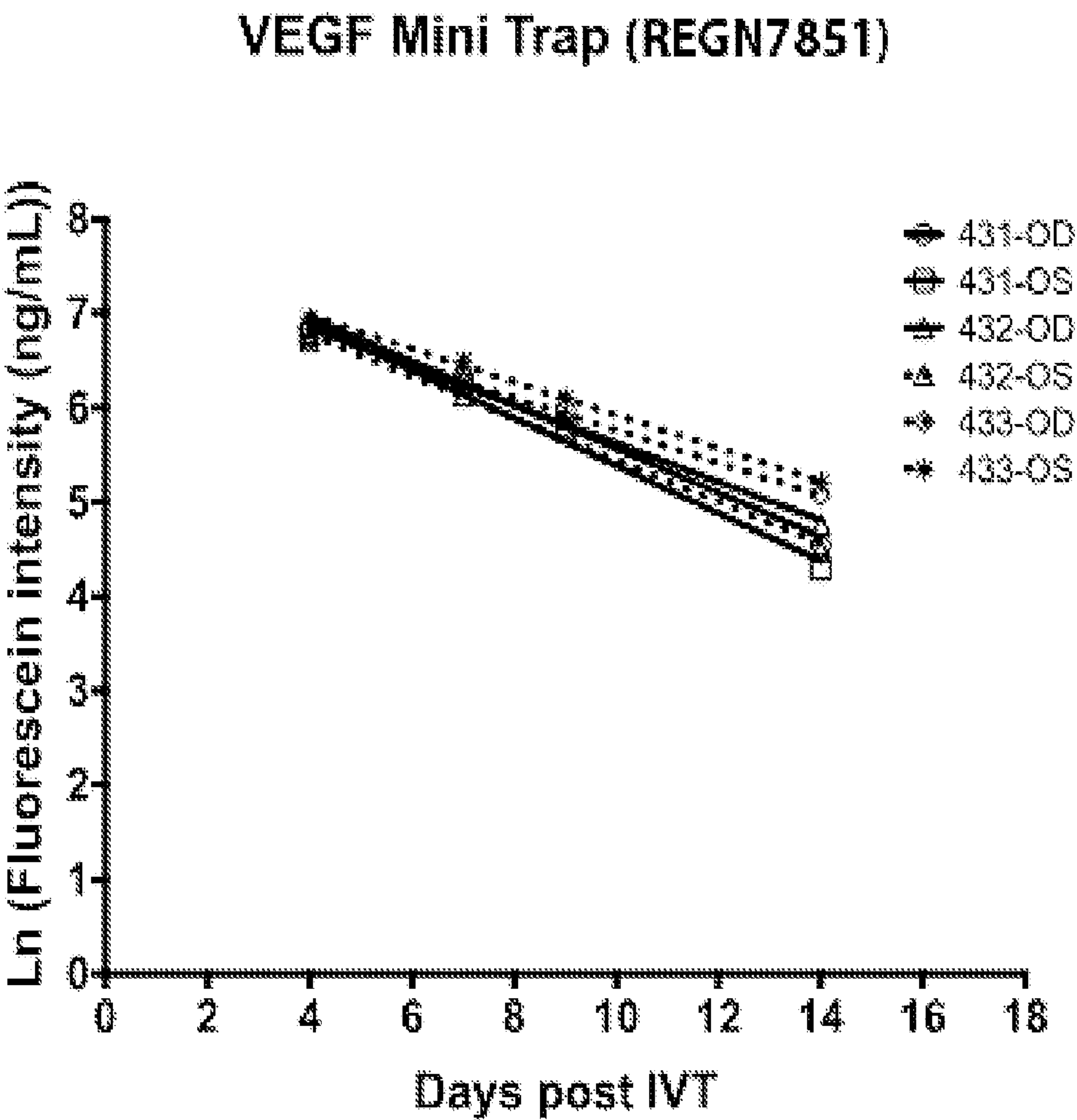


FIG. 34

