
Lysosomal storage diseases and associated enzymatic defects

Disease	Enzymatic Defect
Pompe disease	acid α -glucosidase (e.g., Myozyme®, Lumizyme®)
MPSI* (Hurler disease)	α -L-iduronidase (e.g., Aldurazyme®)
MPSII (Hunter disease)	iduronate sulfatase
MPSIII (Sanfilippo)	heparan N-sulfatase
MPS IV (Morquio A)	galactose-6-sulfatase
MPS IV (Morquio B)	acid β -galactosidase
MPS VII (Sly disease)	β -glucuronidase
I-cell disease	N-acetylglucosamine-1-phosphotransferase
Schindler disease	α -N-acetylgalactosaminidase (α -galactosidase B)
Wolman disease	acid lipase
Cholestrol ester storage disease	acid lipase
Farber disease	lysosomal acid ceramidase
Niemann-Pick disease	acid sphingomyelinase
Gaucher disease	β -glucosidase (e.g. Cerezyme®, Ceredase®)
Krabbe disease	galactosylceramidase
Fabry disease	α -galactosidase A
GM1 gangliosidosis	acid β -galactosidase
Galactosialidosis	β -galactosidase and neuraminidase
Tay-Sach's disease	hexosaminidase A
Sandhoff disease	hexosaminidase A and B

*MPS = mucopolysaccharidosis

FIG. 1

Precursor Human Alpha-Galactosidase-A Protein and cDNA (SEQ ID NOs: 1 and 2)

ATG	CAG	CTG	AGG	AAC	CCA	GAA	CTA	CAT	CTG	GGC	TGC	GCG	CTT	GCG	CTT			
Met	Gln	Leu	Arg	Asn	Pro	Glu	Leu	His	Leu	Gly	Cys	Ala	Leu	Ala	Leu			
1				5					10					15				
CGC	TTC	CTG	GCC	CTC	GTT	TCC	TGG	GAC	ATC	CCT	GGG	GCT	AGA	GCA	CTG			
Arg	Phe	Leu	Ala	Leu	Val	Ser	Trp	Asp	Ile	Pro	Gly	Ala	Arg	Ala	Leu			
			20					25					30					
GAC	AAT	GGA	TTG	GCA	AGG	ACG	CCT	ACC	ATG	GGC	TGG	CTG	CAC	TGG	GAG			
Asp	Asn	Gly	Leu	Ala	Arg	Thr	Pro	Thr	Met	Gly	Trp	Leu	His	Trp	Glu			
		35					40					45						
CGC	TTC	ATG	TGC	AAC	CTT	GAC	TGC	CAG	GAA	GAG	CCA	GAT	TCC	TGC	ATC			
Arg	Phe	Met	Cys	Asn	Leu	Asp	Cys	Gln	Glu	Glu	Pro	Asp	Ser	Cys	Ile			
	50					55					60							
AGT	GAG	AAG	CTC	TTC	ATG	GAG	ATG	GCA	GAG	CTC	ATG	GTC	TCA	GAA	GGC			
Ser	Glu	Lys	Leu	Phe	Met	Glu	Met	Ala	Glu	Leu	Met	Val	Ser	Glu	Gly			
65					70					75					80			
TGG	AAG	GAT	GCA	GGT	TAT	GAG	TAC	CTC	TGC	ATT	GAT	GAC	TGT	TGG	ATG			
Trp	Lys	Asp	Ala	Gly	Tyr	Glu	Tyr	Leu	Cys	Ile	Asp	Asp	Cys	Trp	Met			
				85					90					95				
GCT	CCC	CAA	AGA	GAT	TCA	GAA	GGC	AGA	CTT	CAG	GCA	GAC	CCT	CAG	CGC			
Ala	Pro	Gln	Arg	Asp	Ser	Glu	Gly	Arg	Leu	Gln	Ala	Asp	Pro	Gln	Arg			
			100					105					110					
TTT	CCT	CAT	GGG	ATT	CGC	CAG	CTA	GCT	AAT	TAT	GTT	CAC	AGC	AAA	GGA			
Phe	Pro	His	Gly	Ile	Arg	Gln	Leu	Ala	Asn	Tyr	Val	His	Ser	Lys	Gly			
		115					120					125						
CTG	AAG	CTA	GGG	ATT	TAT	GCA	GAT	GTT	GGA	AAT	AAA	ACC	TGC	GCA	GGC			
Leu	Lys	Leu	Gly	Ile	Tyr	Ala	Asp	Val	Gly	Asn	Lys	Thr	Cys	Ala	Gly			
	130					135					140							
TTC	CCT	GGG	AGT	TTT	GGA	TAC	TAC	GAC	ATT	GAT	GCC	CAG	ACC	TTT	GCT			
Phe	Pro	Gly	Ser	Phe	Gly	Tyr	Tyr	Asp	Ile	Asp	Ala	Gln	Thr	Phe	Ala			
145					150				155						160			
GAC	TGG	GGA	GTA	GAT	CTG	CTA	AAA	TTT	GAT	GGT	TGT	TAC	TGT	GAC	AGT			
Asp	Trp	Gly	Val	Asp	Leu	Leu	Lys	Phe	Asp	Gly	Cys	Tyr	Cys	Asp	Ser			
				165					170					175				
TTG	GAA	AAT	TTG	GCA	GAT	GGT	TAT	AAG	CAC	ATG	TCC	TTG	GCC	CTG	AAT			
Leu	Glu	Asn	Leu	Ala	Asp	Gly	Tyr	Lys	His	Met	Ser	Leu	Ala	Leu	Asn			
			180					185					190					

FIG. 2 (PAGE 1 OF 3)

AGG	ACT	GGC	AGA	AGC	ATT	GTG	TAC	TCC	TGT	GAG	TGG	CCT	CTT	TAT	ATG
Arg	Thr	Gly	Arg	Ser	Ile	Val	Tyr	Ser	Cys	Glu	Trp	Pro	Leu	Tyr	Met
		195					200					205			
TGG	CCC	TTT	CAA	AAG	CCC	AAT	TAT	ACA	GAA	ATC	CGA	CAG	TAC	TGC	AAT
Trp	Pro	Phe	Gln	Lys	Pro	Asn	Tyr	Thr	Glu	Ile	Arg	Gln	Tyr	Cys	Asn
	210					215					220				
CAC	TGG	CGA	AAT	TTT	GCT	GAC	ATT	GAT	GAT	TCC	TGG	AAA	AGT	ATA	AAG
His	Trp	Arg	Asn	Phe	Ala	Asp	Ile	Asp	Asp	Ser	Trp	Lys	Ser	Ile	Lys
225					230					235					240
AGT	ATC	TTG	GAC	TGG	ACA	TCT	TTT	AAC	CAG	GAG	AGA	ATT	GTT	GAT	GTT
Ser	Ile	Leu	Asp	Trp	Thr	Ser	Phe	Asn	Gln	Glu	Arg	Ile	Val	Asp	Val
				245					250					255	
GCT	GGA	CCA	GGG	GGT	TGG	AAT	GAC	CCA	GAT	ATG	TTA	GTG	ATT	GGC	AAC
Ala	Gly	Pro	Gly	Gly	Trp	Asn	Asp	Pro	Asp	Met	Leu	Val	Ile	Gly	Asn
			260					265					270		
TTT	GGC	CTC	AGC	TGG	AAT	CAG	CAA	GTA	ACT	CAG	ATG	GCC	CTC	TGG	GCT
Phe	Gly	Leu	Ser	Trp	Asn	Gln	Gln	Val	Thr	Gln	Met	Ala	Leu	Trp	Ala
		275					280					285			
ATC	ATG	GCT	GCT	CCT	TTA	TTC	ATG	TCT	AAT	GAC	CTC	CGA	CAC	ATC	AGC
Ile	Met	Ala	Ala	Pro	Leu	Phe	Met	Ser	Asn	Asp	Leu	Arg	His	Ile	Ser
	290					295					300				
CCT	CAA	GCC	AAA	GCT	CTC	CTT	CAG	GAT	AAG	GAC	GTA	ATT	GCC	ATC	AAT
Pro	Gln	Ala	Lys	Ala	Leu	Leu	Gln	Asp	Lys	Asp	Val	Ile	Ala	Ile	Asn
305					310					315					320
CAG	GAC	CCC	TTG	GGC	AAG	CAA	GGG	TAC	CAG	CTT	AGA	CAG	GGA	GAC	AAC
Gln	Asp	Pro	Leu	Gly	Lys	Gln	Gly	Tyr	Gln	Leu	Arg	Gln	Gly	Asp	Asn
				325				330					335		
TTT	GAA	GTG	TGG	GAA	CGA	CCT	CTC	TCA	GGC	TTA	GCC	TGG	GCT	GTA	GCT
Phe	Glu	Val	Trp	Glu	Arg	Pro	Leu	Ser	Gly	Leu	Ala	Trp	Ala	Val	Ala
			340					345					350		
ATG	ATA	AAC	CGG	CAG	GAG	ATT	GGT	GGA	CCT	CGC	TCT	TAT	ACC	ATC	GCA
Met	Ile	Asn	Arg	Gln	Glu	Ile	Gly	Gly	Pro	Arg	Ser	Tyr	Thr	Ile	Ala
		355					360					365			
GTT	GCT	TCC	CTG	GGT	AAA	GGA	GTG	GCC	TGT	AAT	CCT	GCC	TGC	TTC	ATC
Val	Ala	Ser	Leu	Gly	Lys	Gly	Val	Ala	Cys	Asn	Pro	Ala	Cys	Phe	Ile
	370					375					380				

FIG. 2 (PAGE 2 OF 3)

ACA CAG CTC CTC CCT GTG AAA AGG AAG CTA GGG TTC TAT GAA TGG ACT
 Thr Gln Leu Leu Pro Val Lys Arg Lys Leu Gly Phe Tyr Glu Trp Thr
 385 390 395 400

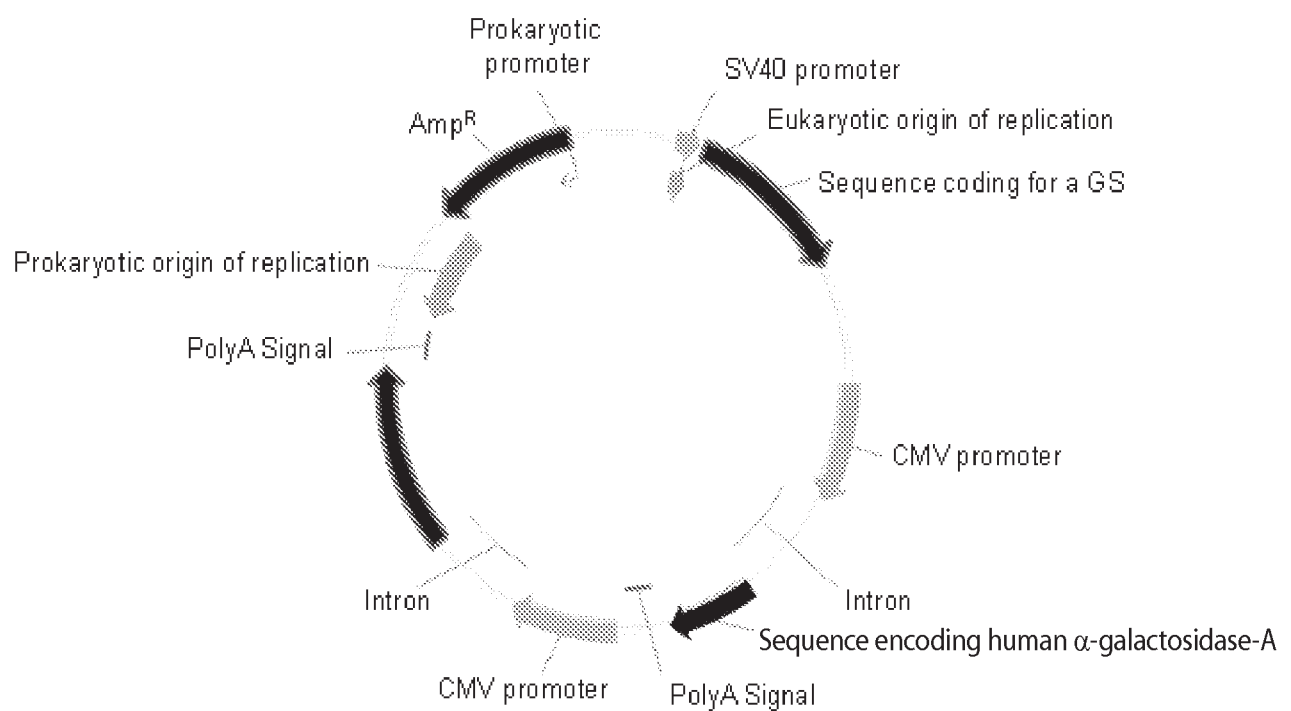
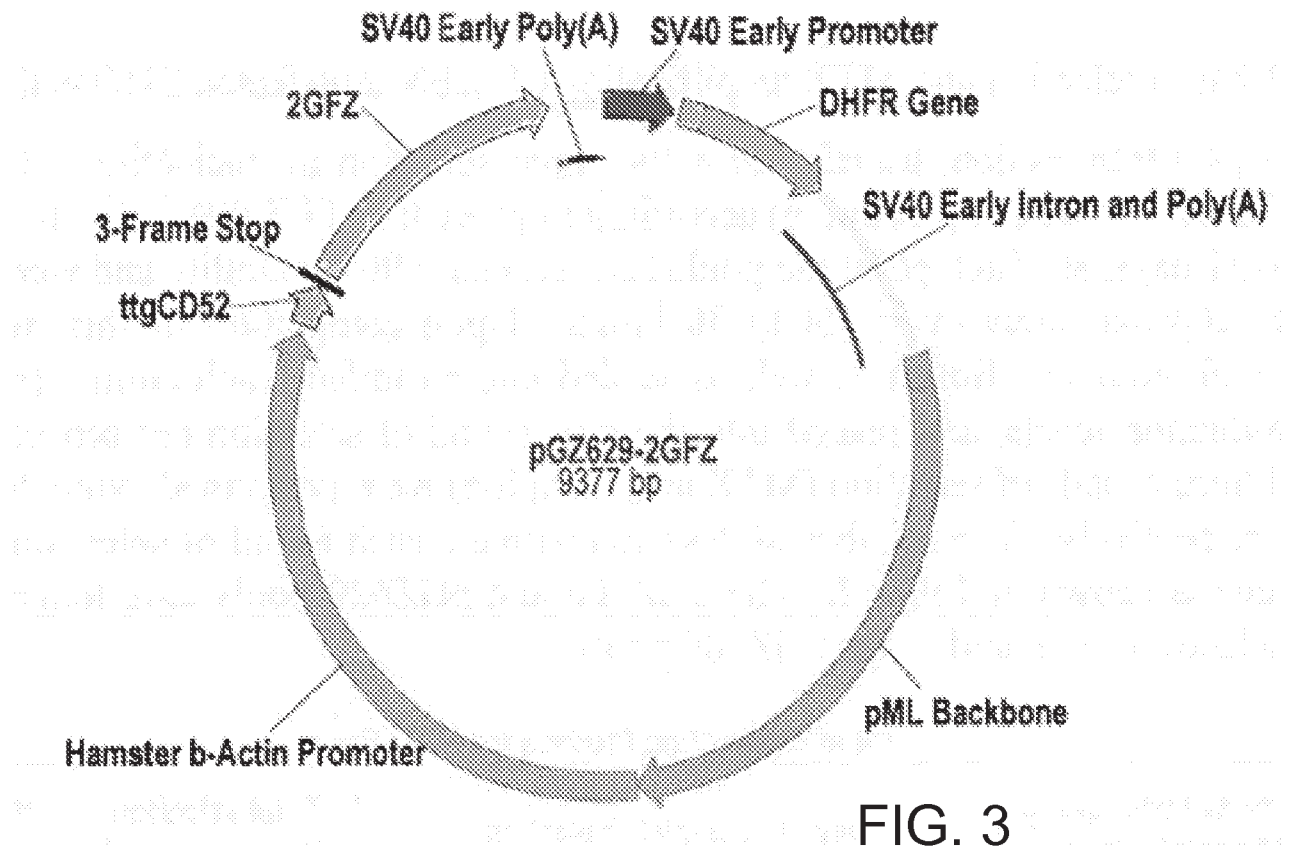
TCA AGG TTA AGA AGT CAC ATA AAT CCC ACA GGC ACT GTT TTG CTT CAG
 Ser Arg Leu Arg Ser His Ile Asn Pro Thr Gly Thr Val Leu Leu Gln
 405 410 415

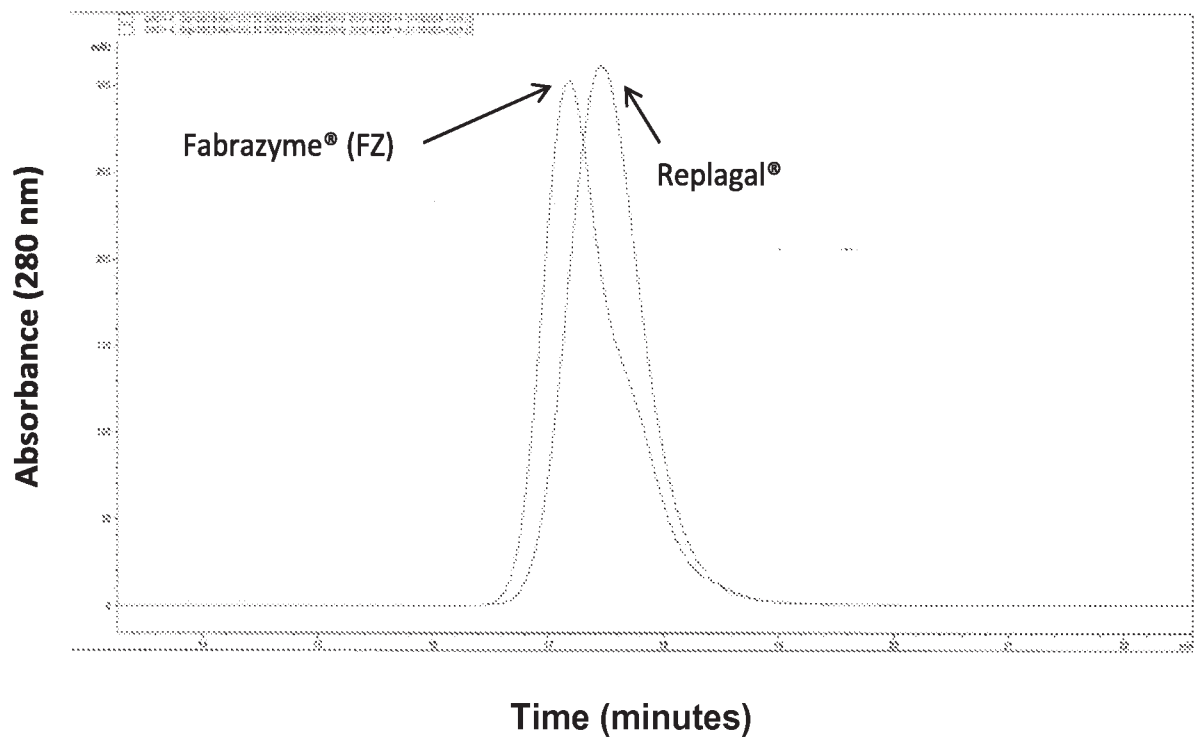
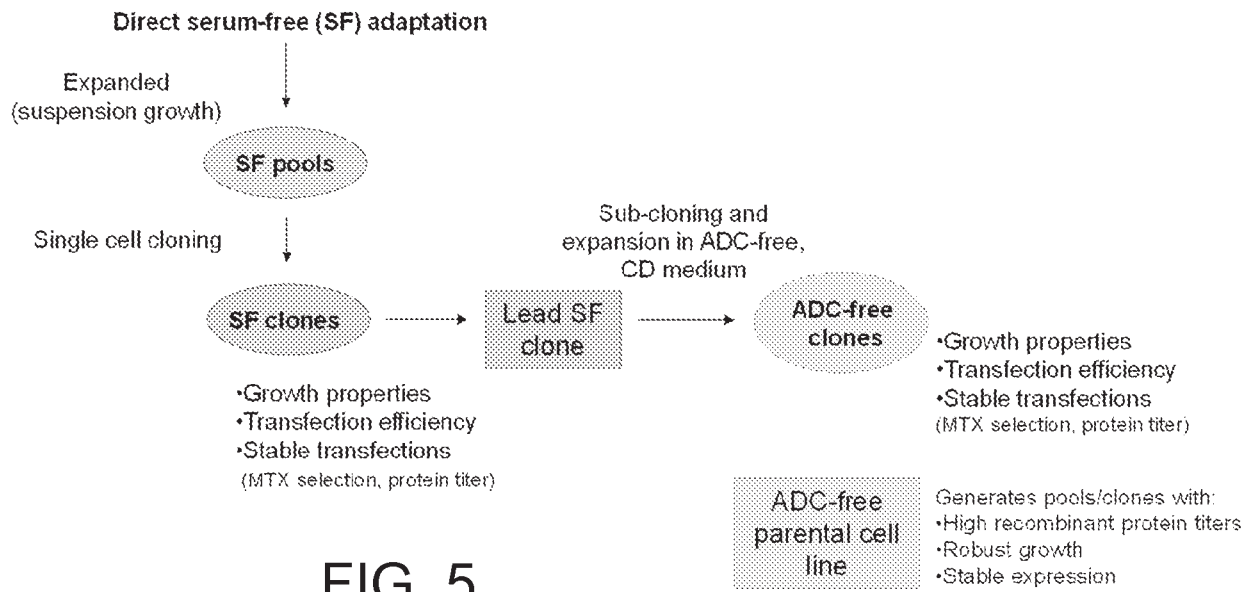
CTA GAA AAT ACA ATG CAG ATG TCA TTA AAA GAC TTA CTT TAAAAAAAAA
 Leu Glu Asn Thr Met Gln Met Ser Leu Lys Asp Leu Leu
 420 425 430

Mature Human Alpha-Galactosidase-A (SEQ ID NO: 3)

Leu	Asp	Asn	Gly	Leu	Ala	Arg	Thr	Pro	Thr	Met	Gly	Trp	Leu	His	Trp	16
Glu	Arg	Phe	Met	Cys	Asn	Leu	Asp	Cys	Gln	Glu	Glu	Pro	Asp	Ser	Cys	32
Ile	Ser	Glu	Lys	Leu	Phe	Met	Glu	Met	Ala	Glu	Leu	Met	Val	Ser	Glu	48
Gly	Trp	Lys	Asp	Ala	Gly	Tyr	Glu	Tyr	Leu	Cys	Ile	Asp	Asp	Cys	Trp	64
Met	Ala	Pro	Gln	Arg	Asp	Ser	Glu	Gly	Arg	Leu	Gln	Ala	Asp	Pro	Gln	80
Arg	Phe	Pro	His	Gly	Ile	Arg	Gln	Leu	Ala	Asn	Tyr	Val	His	Ser	Lys	96
Gly	Leu	Lys	Leu	Gly	Ile	Tyr	Ala	Asp	Val	Gly	Asn	Lys	Thr	Cys	Ala	112
Gly	Phe	Pro	Gly	Ser	Phe	Gly	Tyr	Tyr	Asp	Ile	Asp	Ala	Gln	Thr	Phe	128
Ala	Asp	Trp	Gly	Val	Asp	Leu	Leu	Lys	Phe	Asp	Gly	Cys	Tyr	Cys	Asp	144
Ser	Leu	Glu	Asn	Leu	Ala	Asp	Gly	Tyr	Lys	His	Met	Ser	Leu	Ala	Leu	160
Asn	Arg	Thr	Gly	Arg	Ser	Ile	Val	Tyr	Ser	Vys	Glu	Trp	Pro	Leu	Tyr	176
Met	Trp	Pro	Phe	Gln	Lys	Pro	Asn	Tyr	Thr	Glu	Ile	Arg	Gln	Tyr	Cys	192
Asn	His	Trp	Arg	Asn	Phe	Ala	Asp	Ile	Asp	Asp	Ser	Trp	Lys	Ser	Ile	208
Lys	Ser	Ile	Leu	Asp	Trp	Thr	Ser	Phe	Asn	Gln	Glu	Arg	Ile	Val	Asp	224
Val	Ala	Gly	Pro	Gly	Gly	trp	Asn	Asp	Pro	Asp	Met	Leu	Val	Ile	Gly	240
Asn	Phe	Gly	Leu	Ser	Trp	Asn	Gln	Gln	Val	Thr	Gln	Met	Ala	Leu	Trp	256
Ala	Ile	Met	Ala	Ala	Pro	Leu	Phe	Met	Ser	Asn	Asp	Leu	Arg	His	Ile	272
Ser	Pro	Gln	Ala	Lys	Ala	Leu	Leu	Gln	Asp	Lys	Asp	Val	Ile	Ala	Ile	288
Asn	Gln	Asp	Pro	Leu	Gly	Lys	Gln	Gly	Tyr	GLn	Leu	Arg	Gln	Gly	Asp	304
Asn	Phe	Glu	Val	Trp	Glu	Arg	Pro	Leu	Ser	Gly	Leu	Ala	Trp	Ala	Val	320
Ala	Met	Ile	Asn	Arg	Gln	Glu	Ile	Gly	Gly	Pro	Arg	Ser	Tyr	Thr	Ile	336
Ala	Val	Ala	Ser	Leu	Gly	Lys	Gly	Val	Ala	Cys	Asn	Pro	Ala	Cys	Phe	352
Ile	Thr	Gln	Leu	Leu	Pro	Val	Lys	Arg	Lys	Leu	Gly	Phe	Tyr	Glu	Trp	368
Thr	Ser	Arg	Leu	Arg	Ser	His	Ile	Asn	Pro	Thr	Gly	Thr	Val	Leu	Leu	384
Gln	Leu	Glu	Asn	Thr	Met	Gln	Met	Ser	Leu	Lys	Asp	Leu	Leu			398

FIG. 2 (PAGE 3 OF 3)



**FIG. 6**

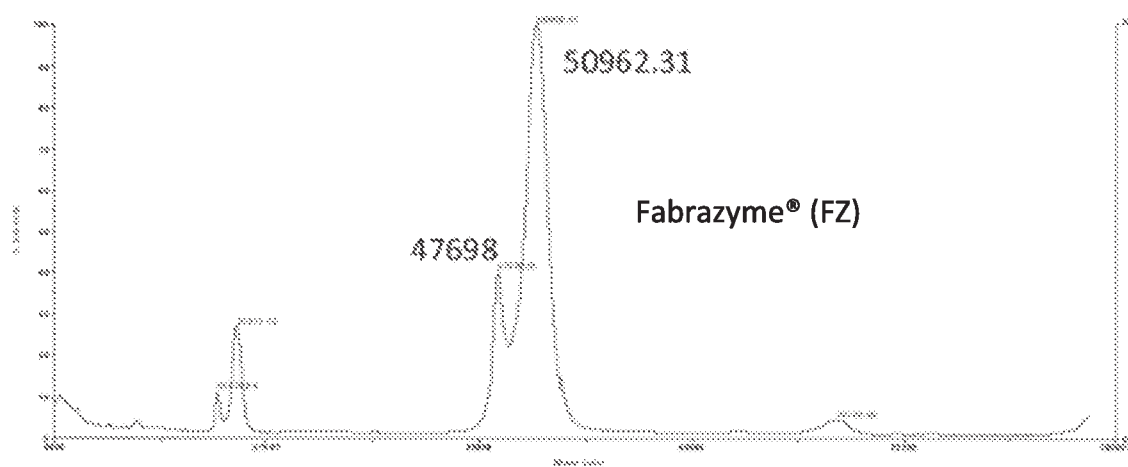


FIG. 7A

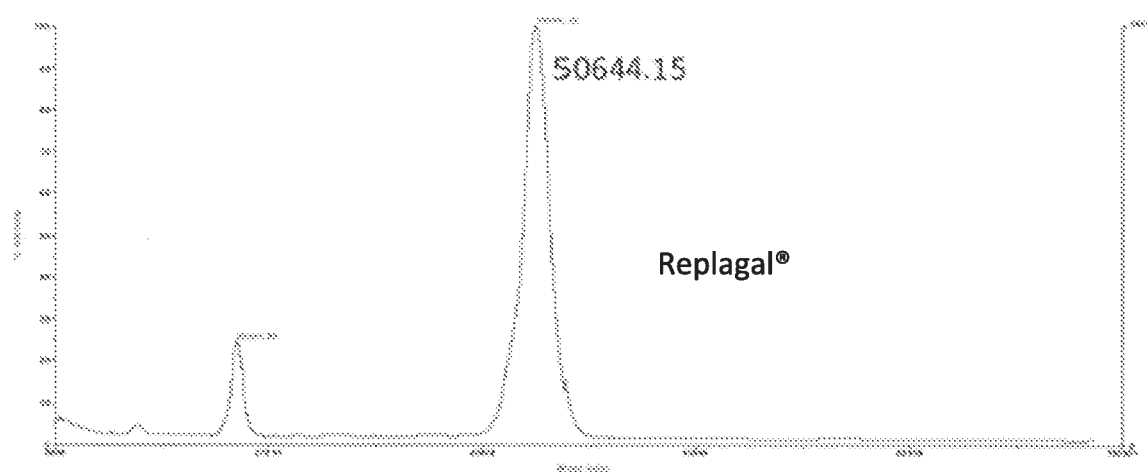


FIG. 7B

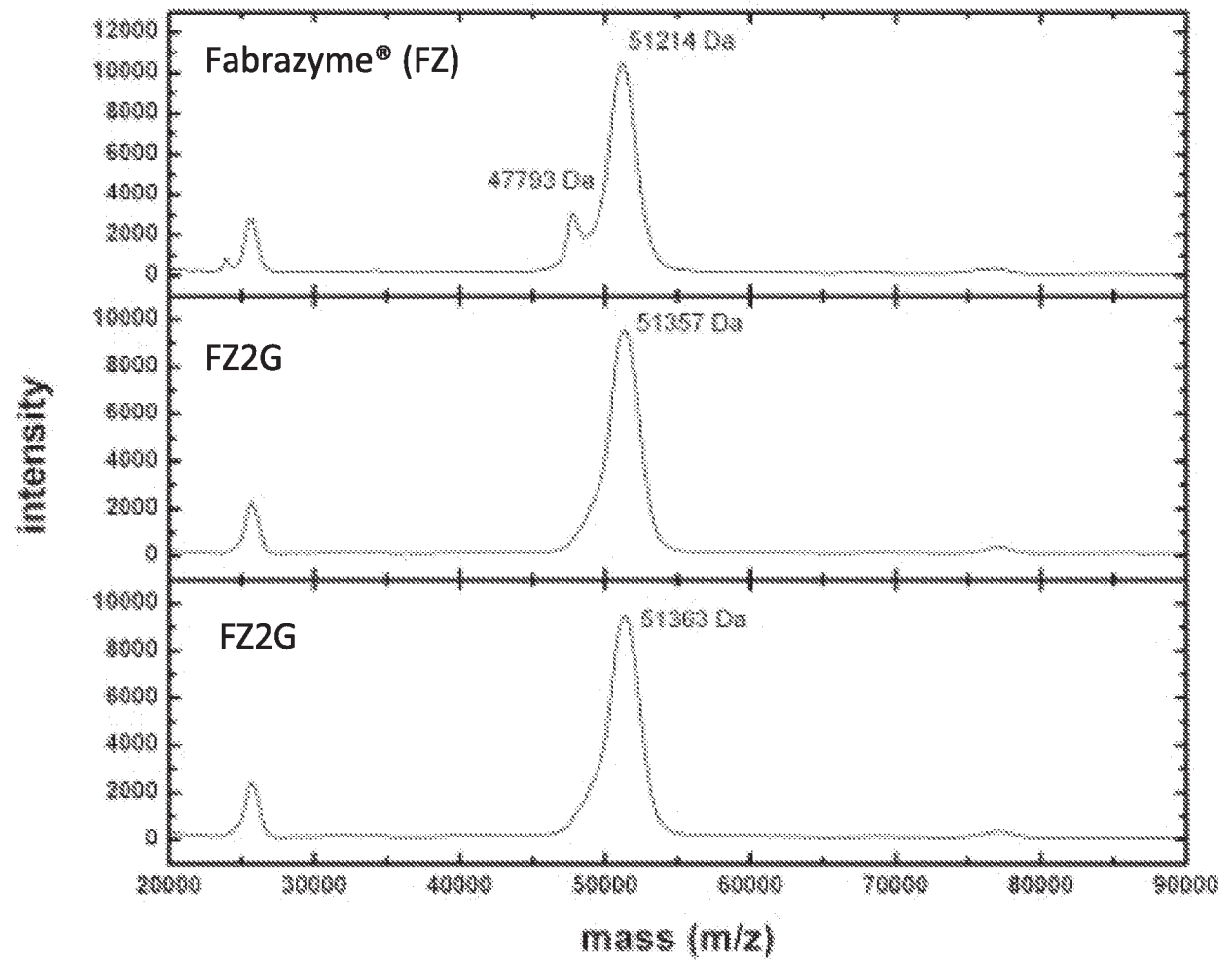


FIG. 8

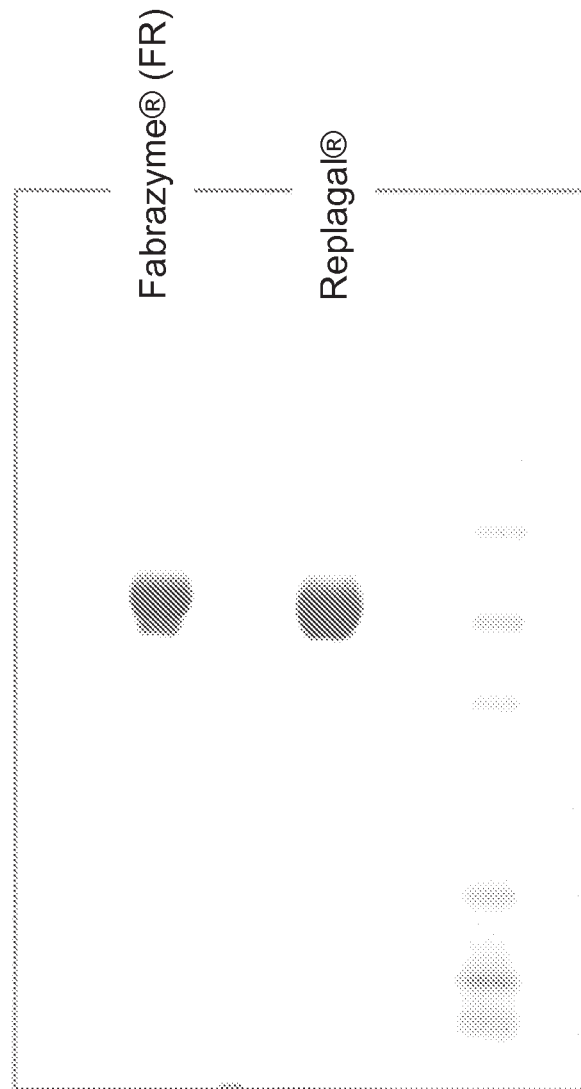
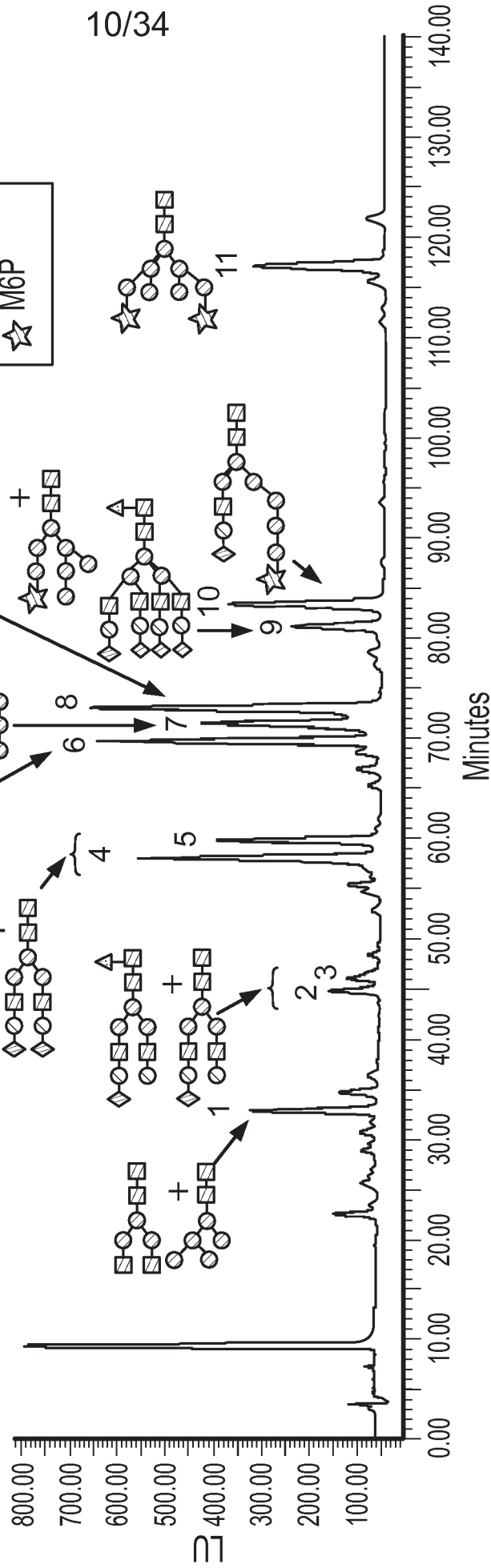
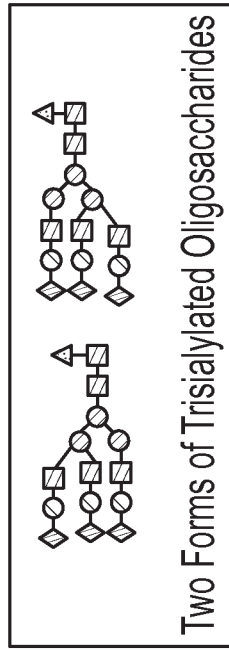


FIG. 9



Natural

Charged oligosaccharides

FIG. 10

FLD1 A, Ex=230, Em=425 (CT040312\CT040312 2012-04-0316-39-32\002-0201.D)
FLD1 A, Ex=230, Em=425 (CT040312\CT040312 2012-04-0316-39-32\003-0301.D)

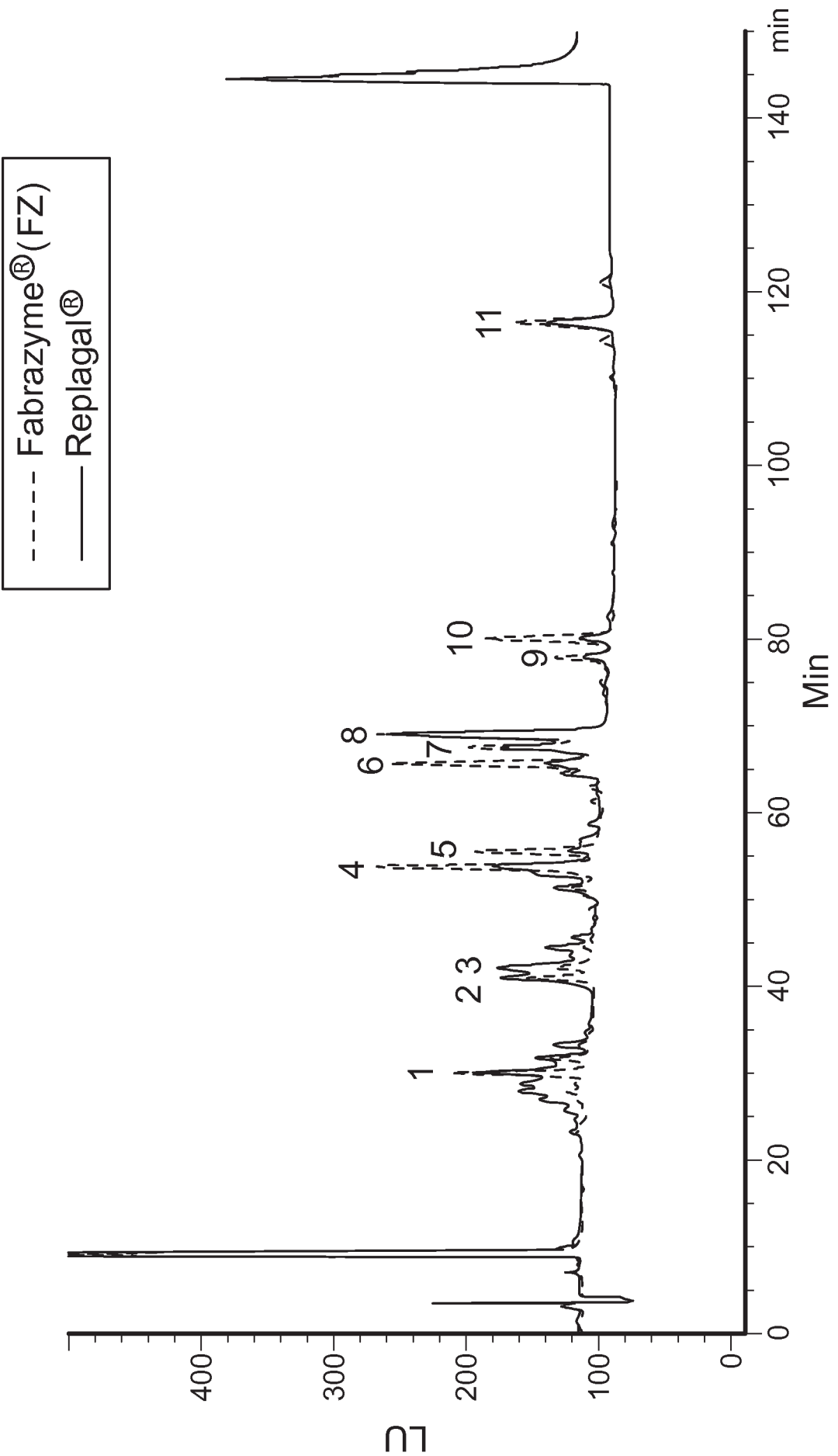


FIG. 11

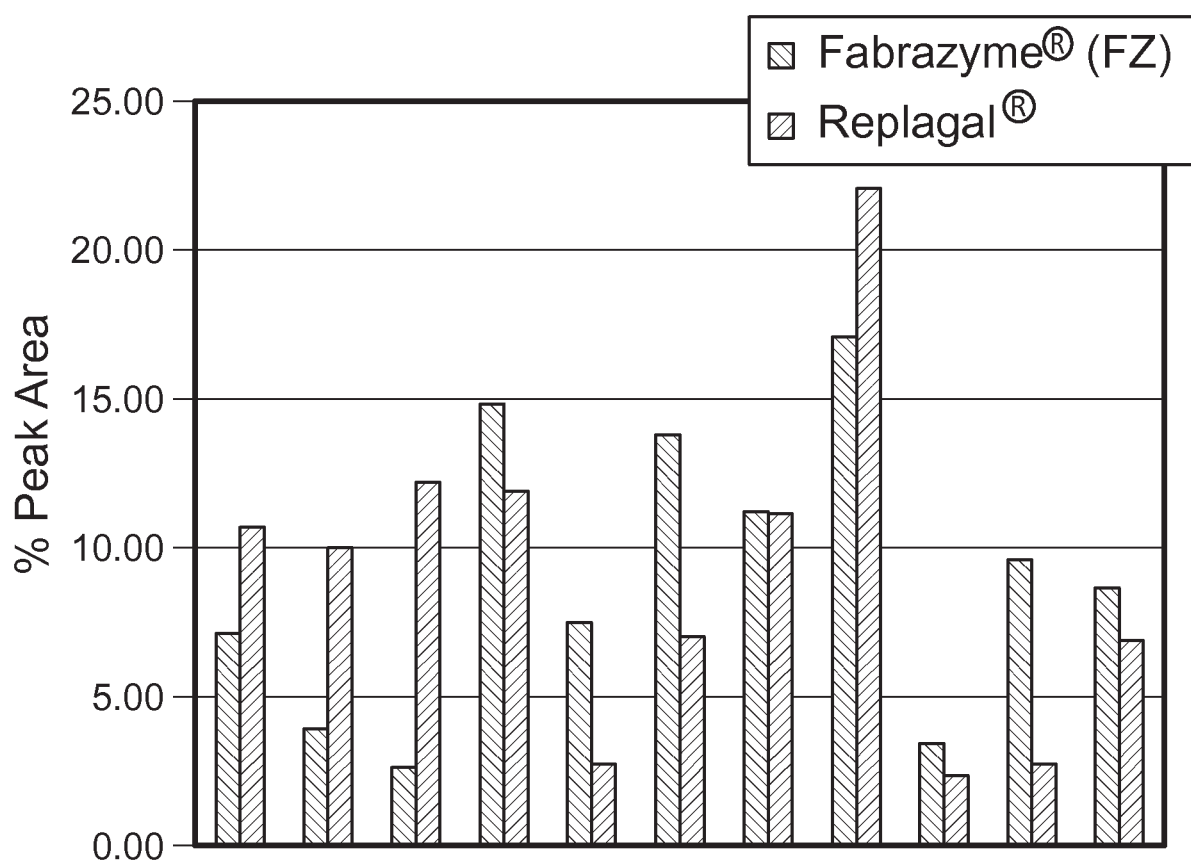


FIG. 12

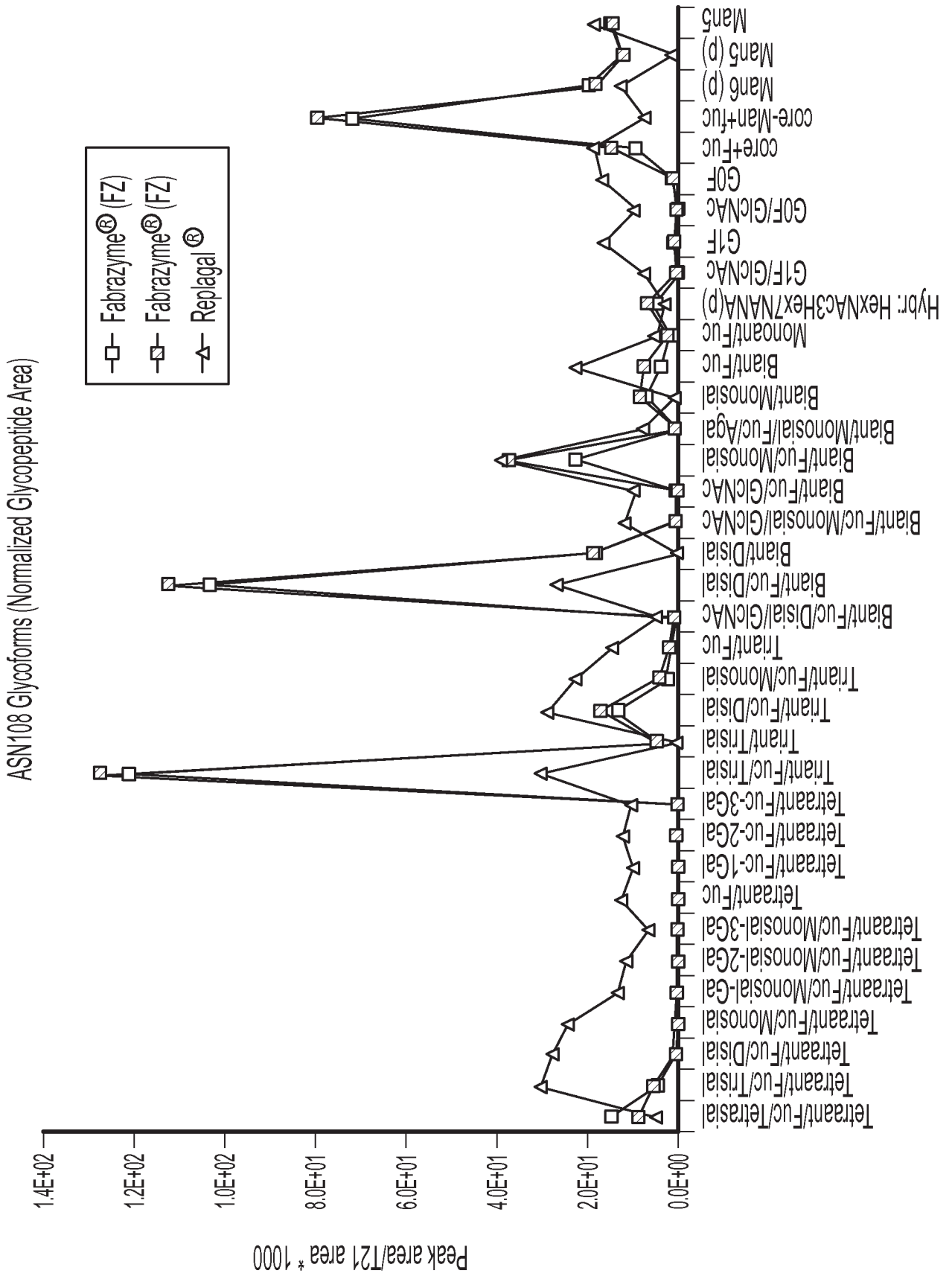


FIG. 13

ASN161 Glycoforms (Normalized Glycopeptide Peak Area)

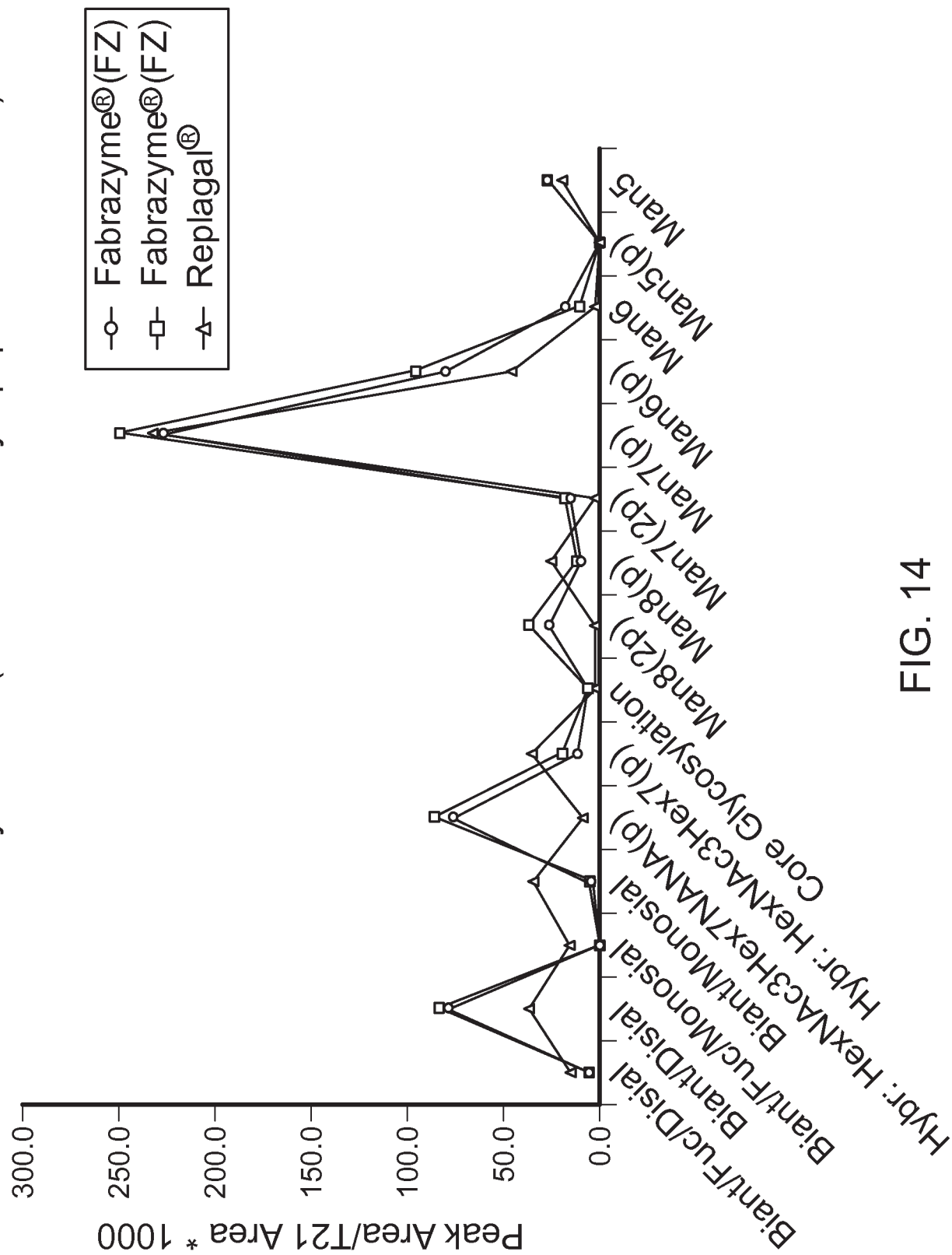


FIG. 14

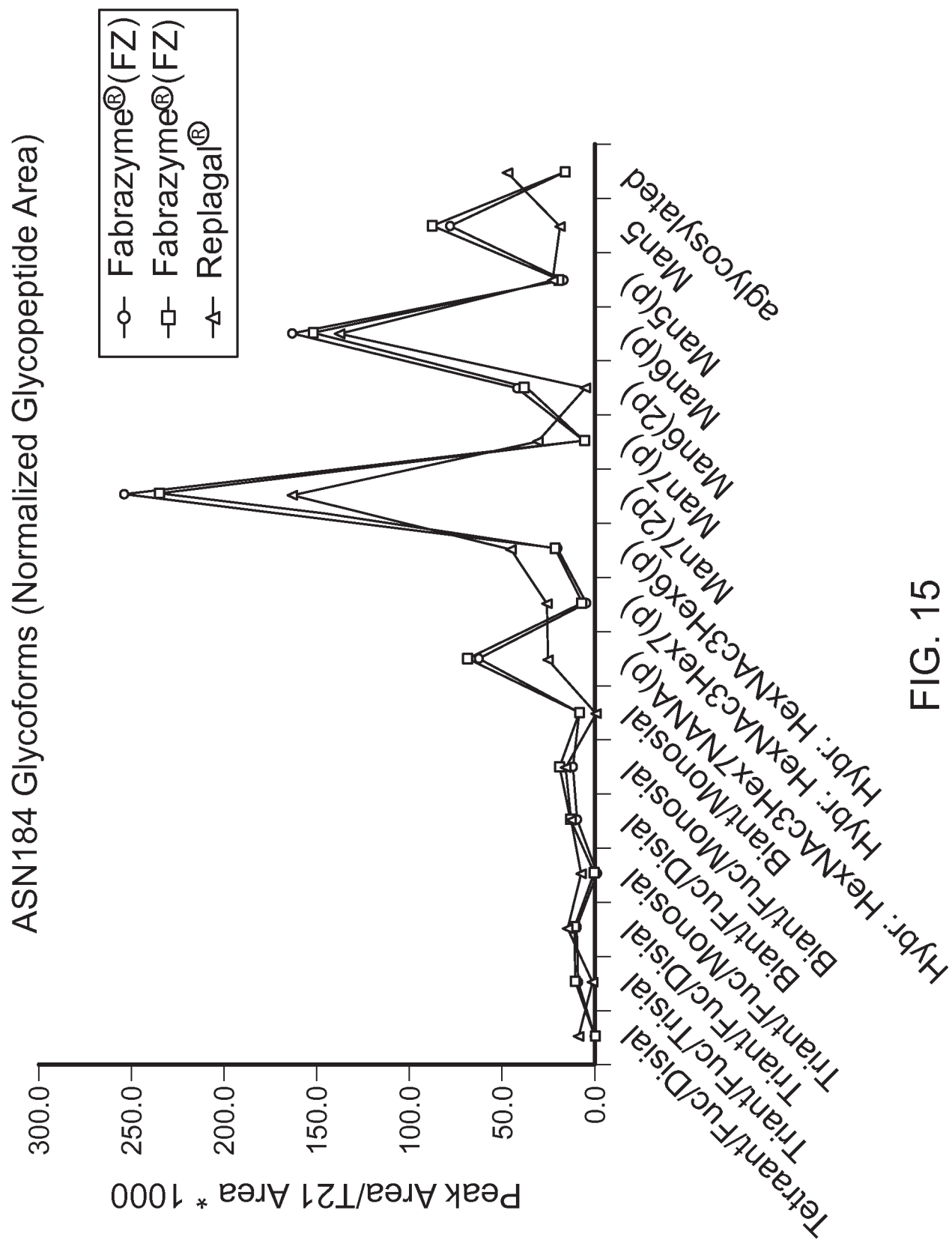
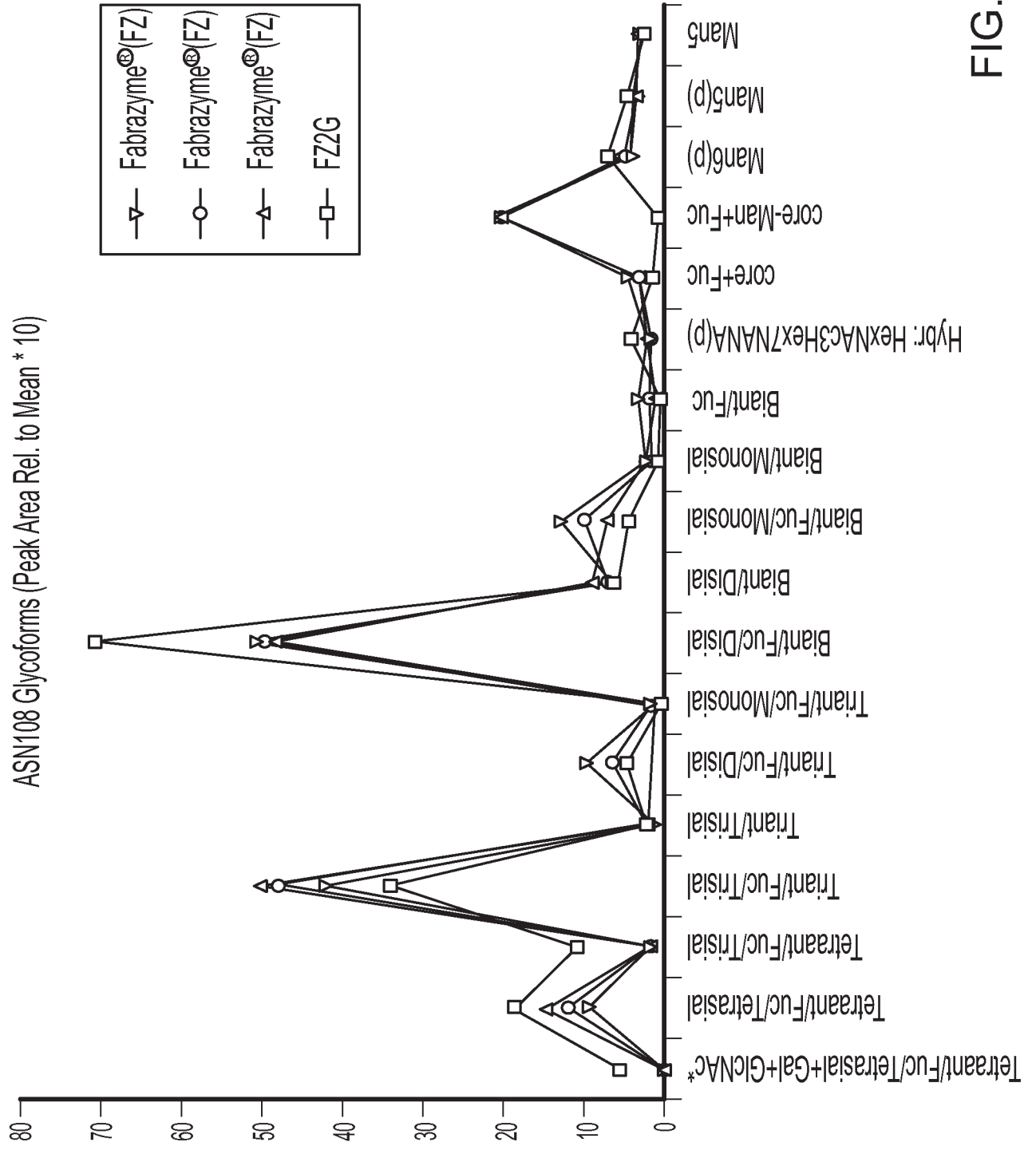


FIG. 15

FIG. 16



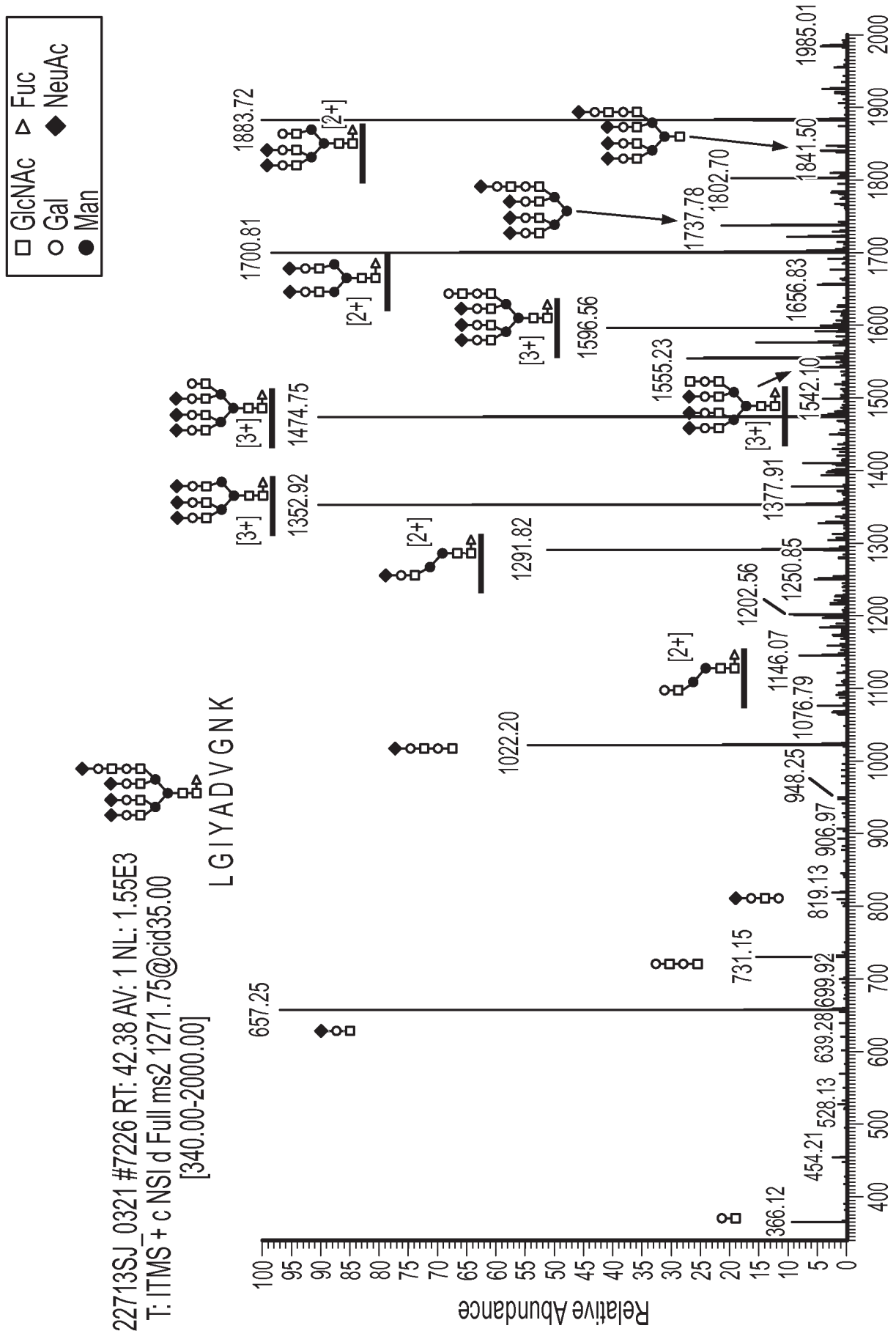


FIG. 17

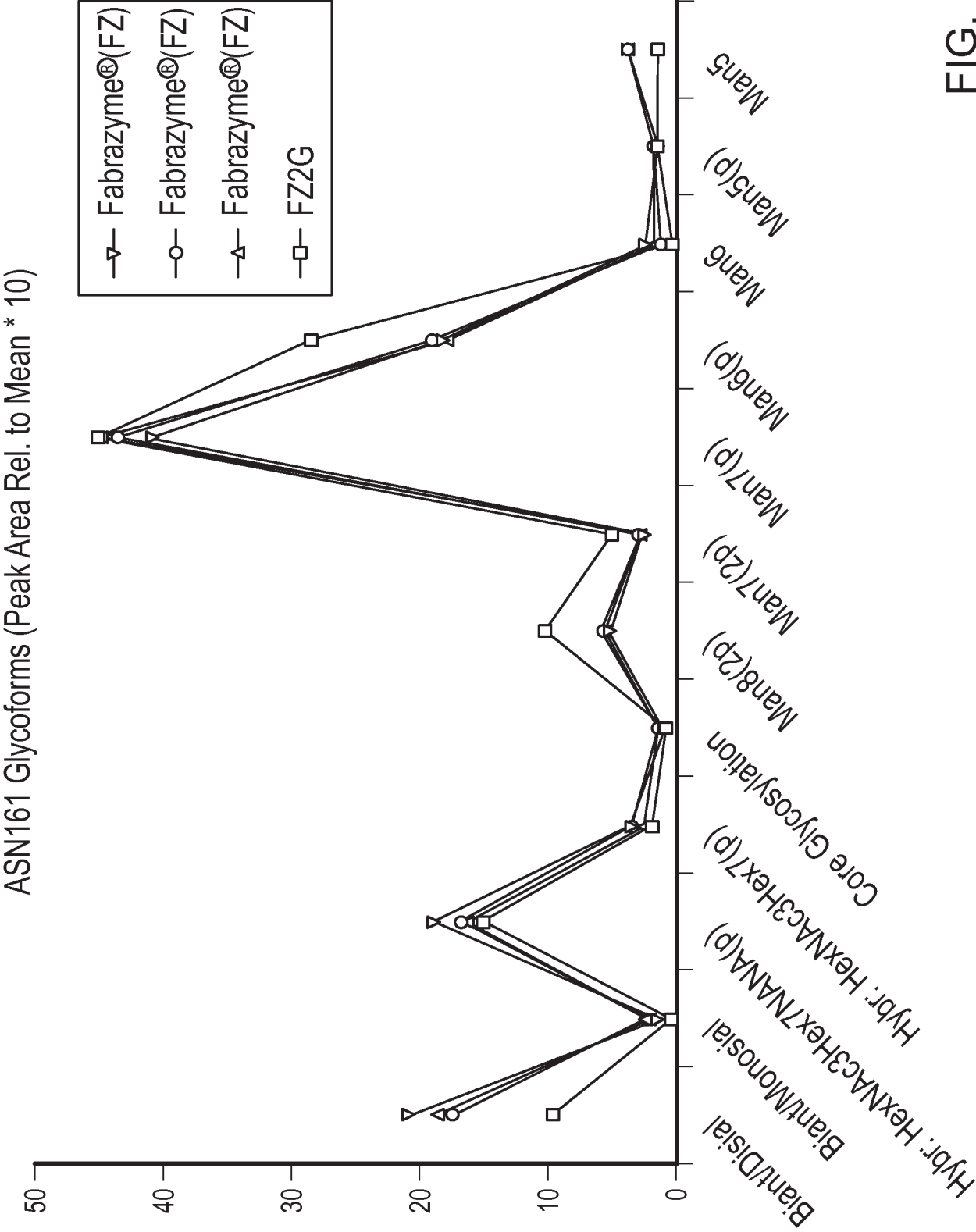


FIG. 18

ASN184 Glycoforms (Peak Area Rel. To Mean *10)

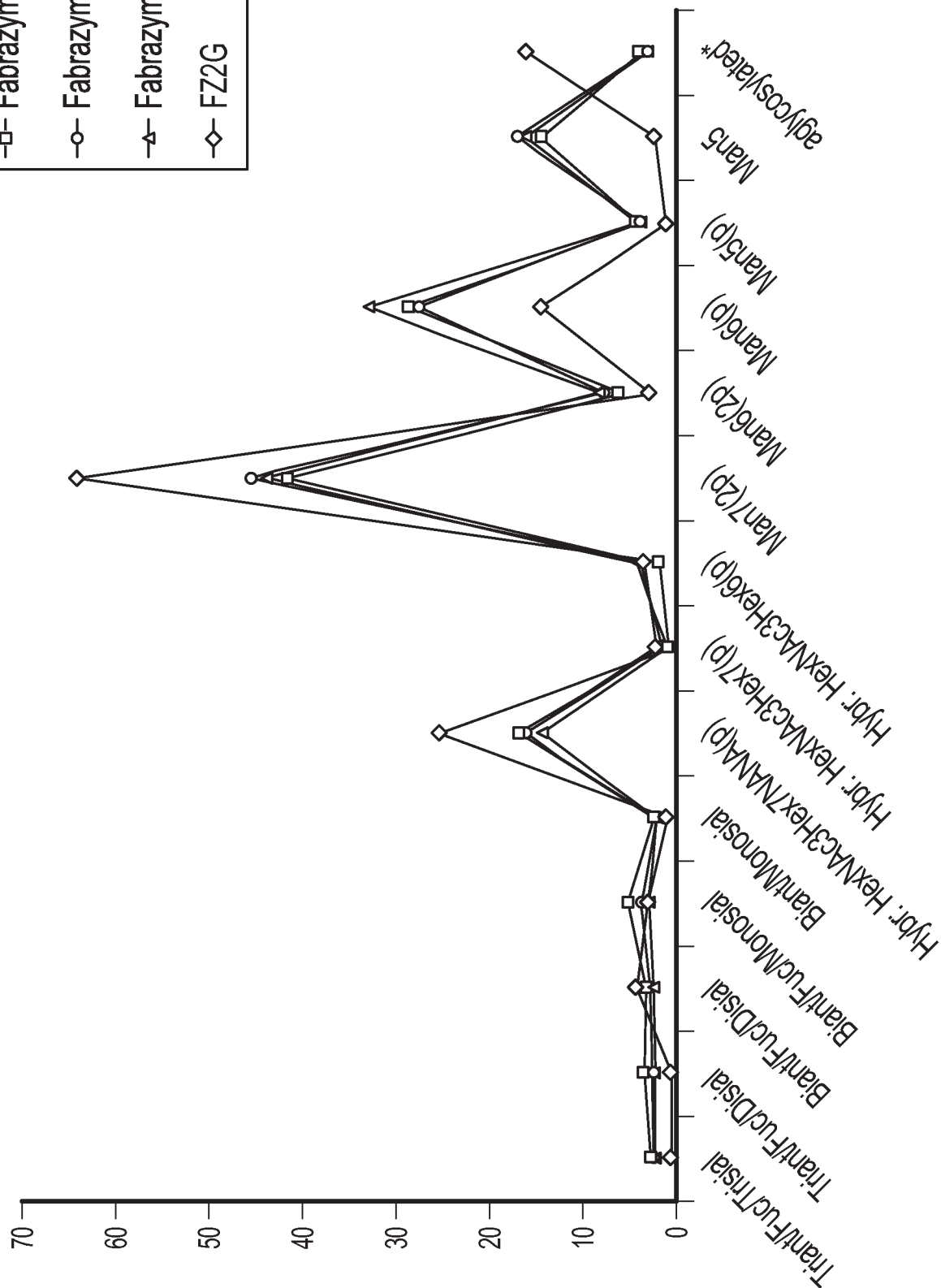
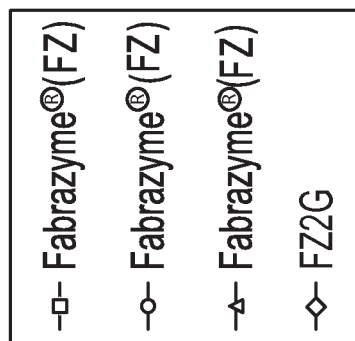


FIG. 19

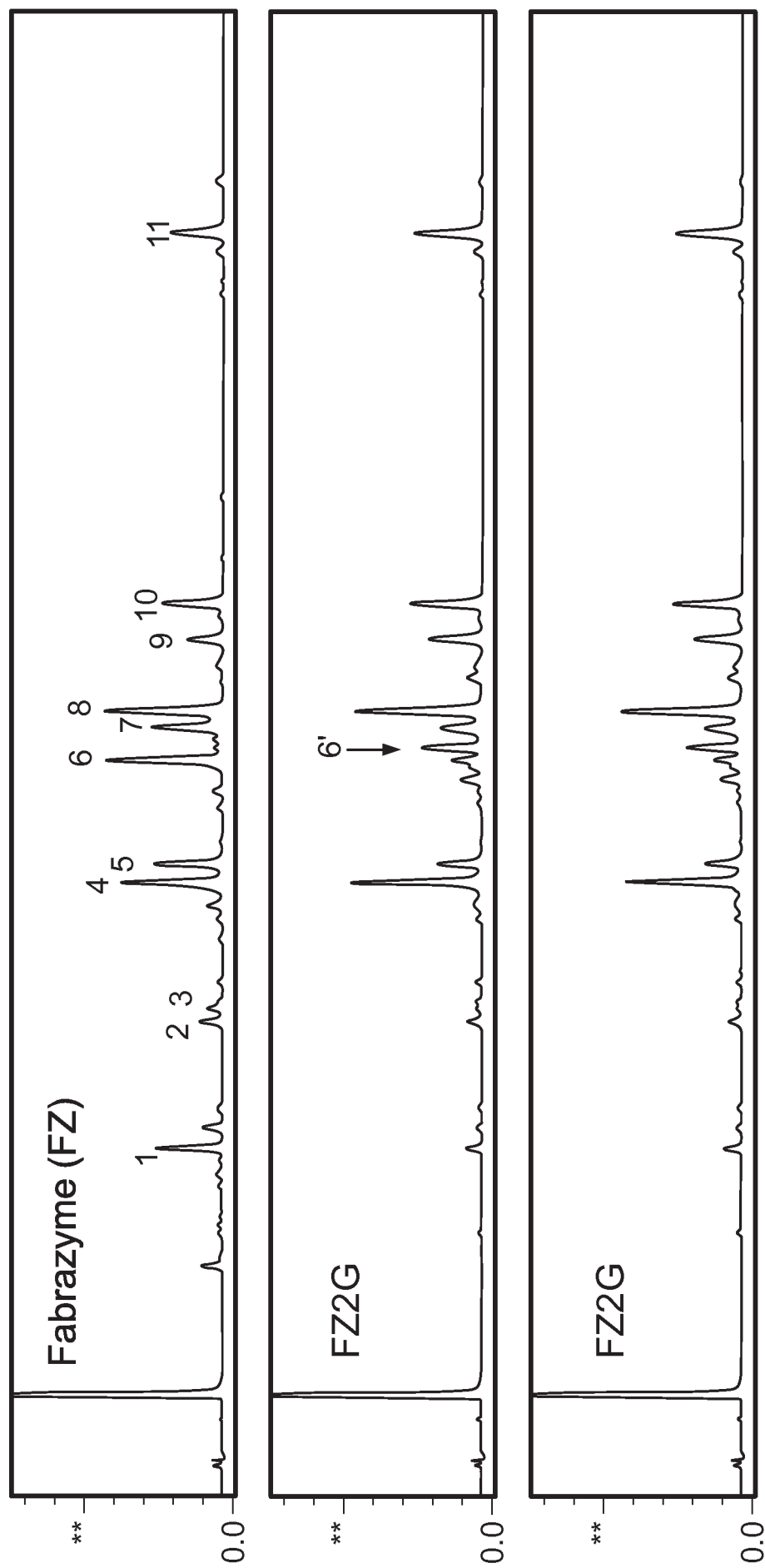
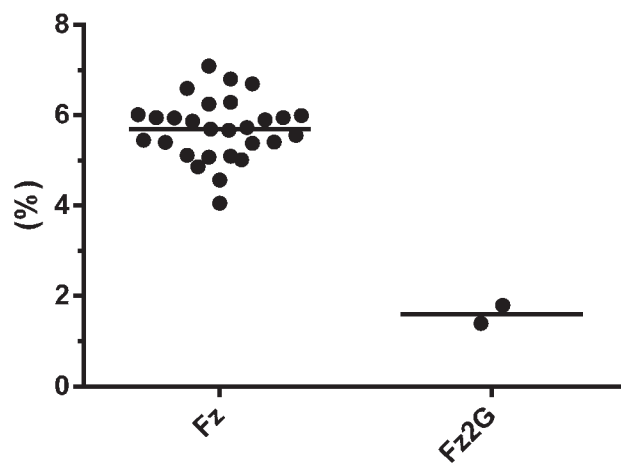


FIG. 20

21/34

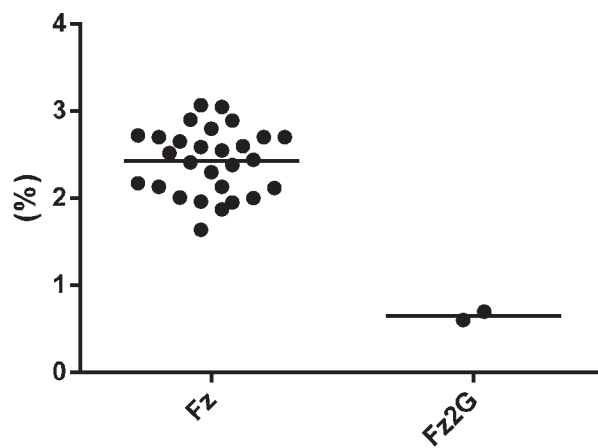
A

Peak 1



B

Peak 3



C

Peak 2

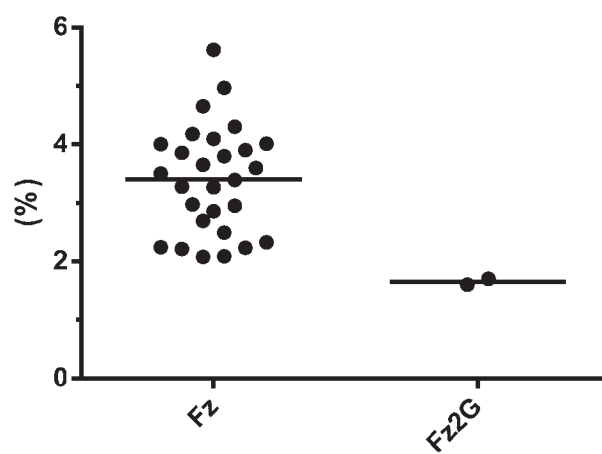
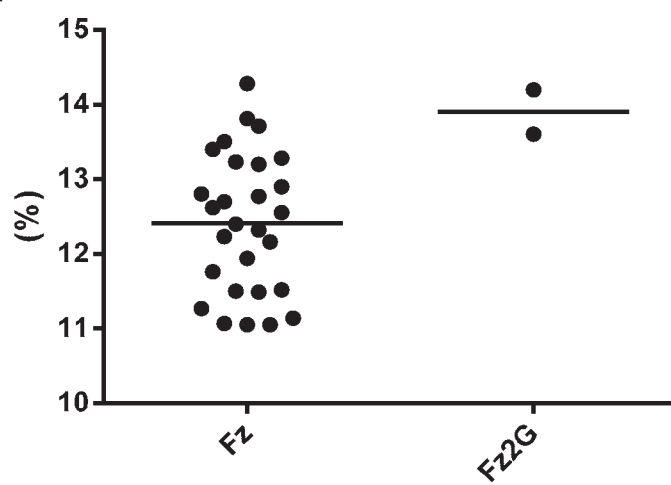


FIG. 21

22/34

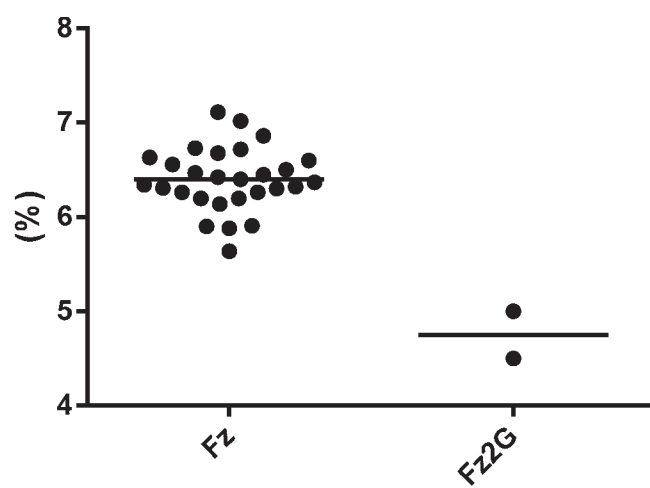
A

Peak 4



B

Peak 5



C

Peak 6

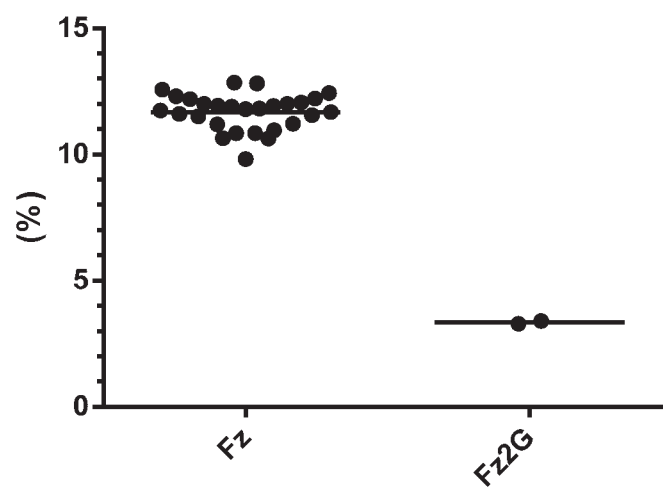
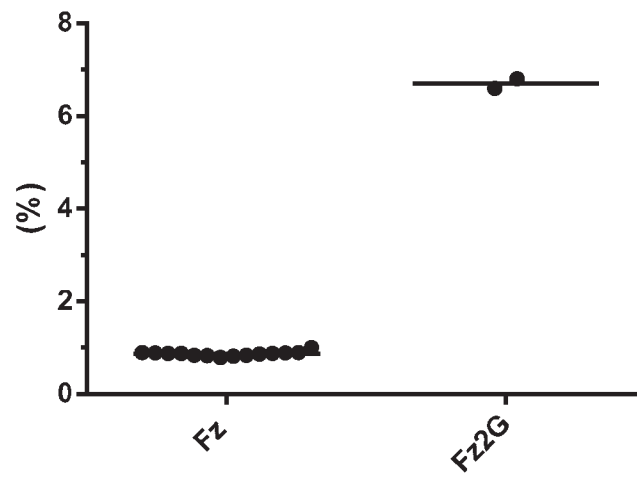


FIG. 22

23/34

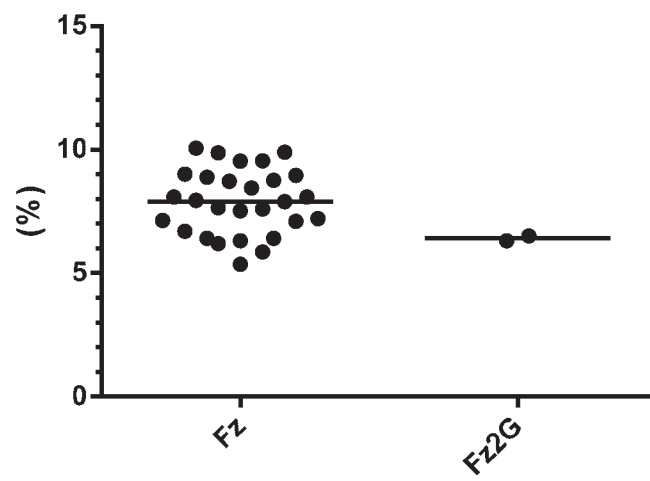
A

Peak 6'



B

Peak 7



C

Peak 8

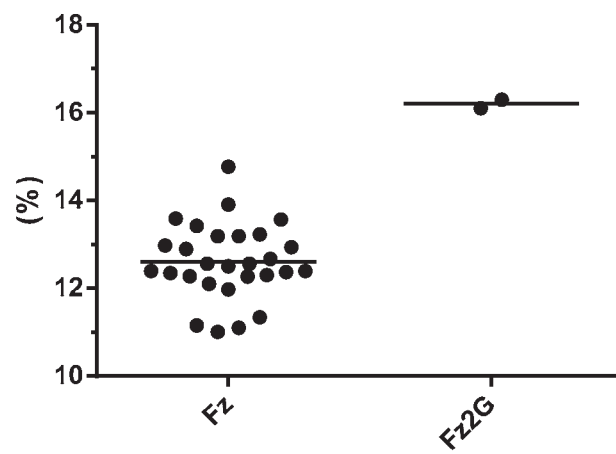
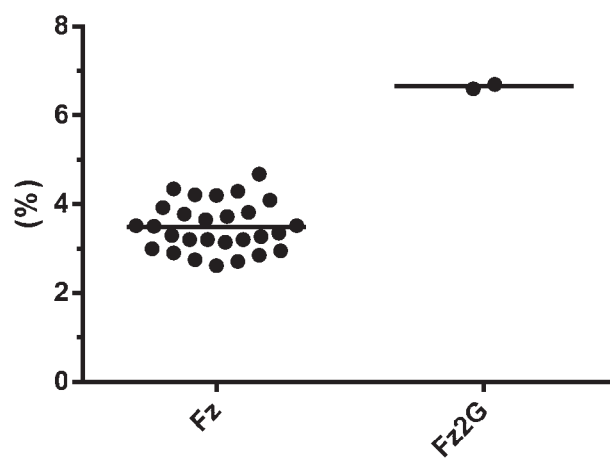


FIG. 23

24/34

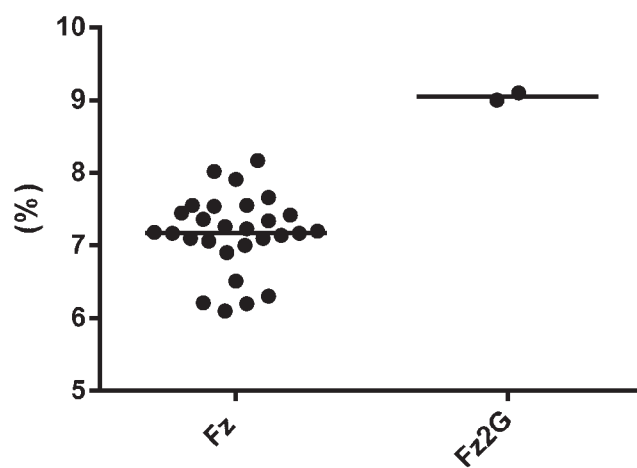
A

Oligo Peak 9



B

Peak 10



C

Peak 11

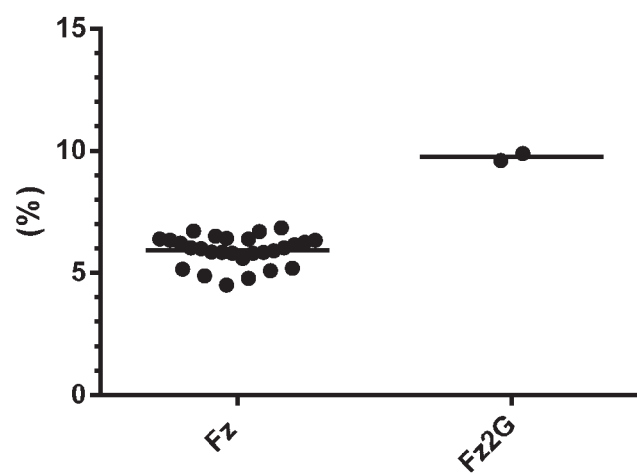
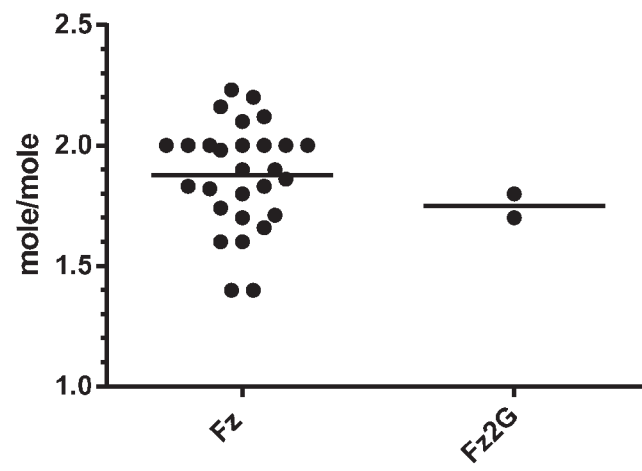


FIG. 24

25/34

A

M6P



B

NANA

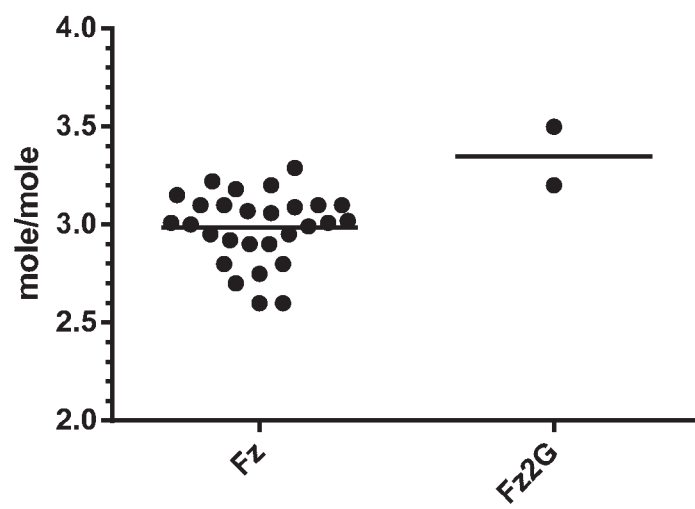


FIG. 25

26/34

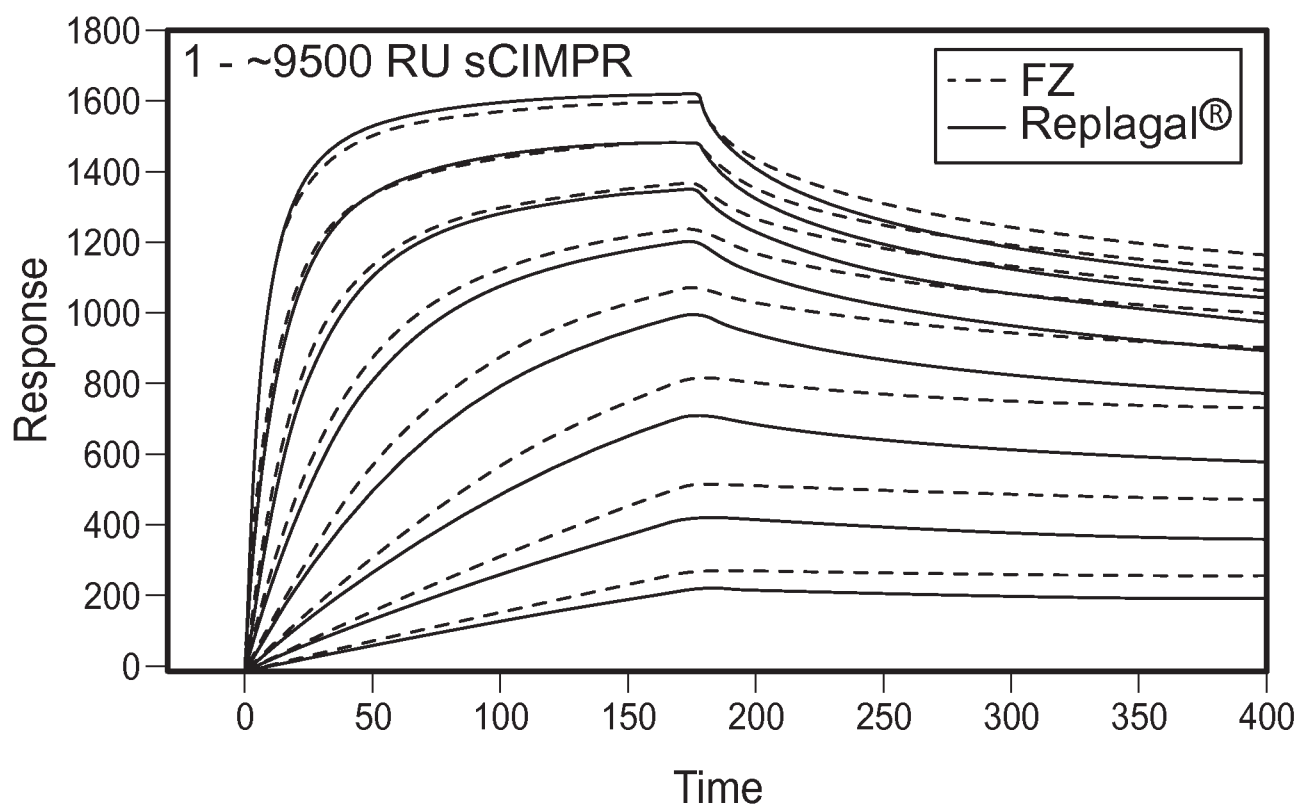


FIG. 26

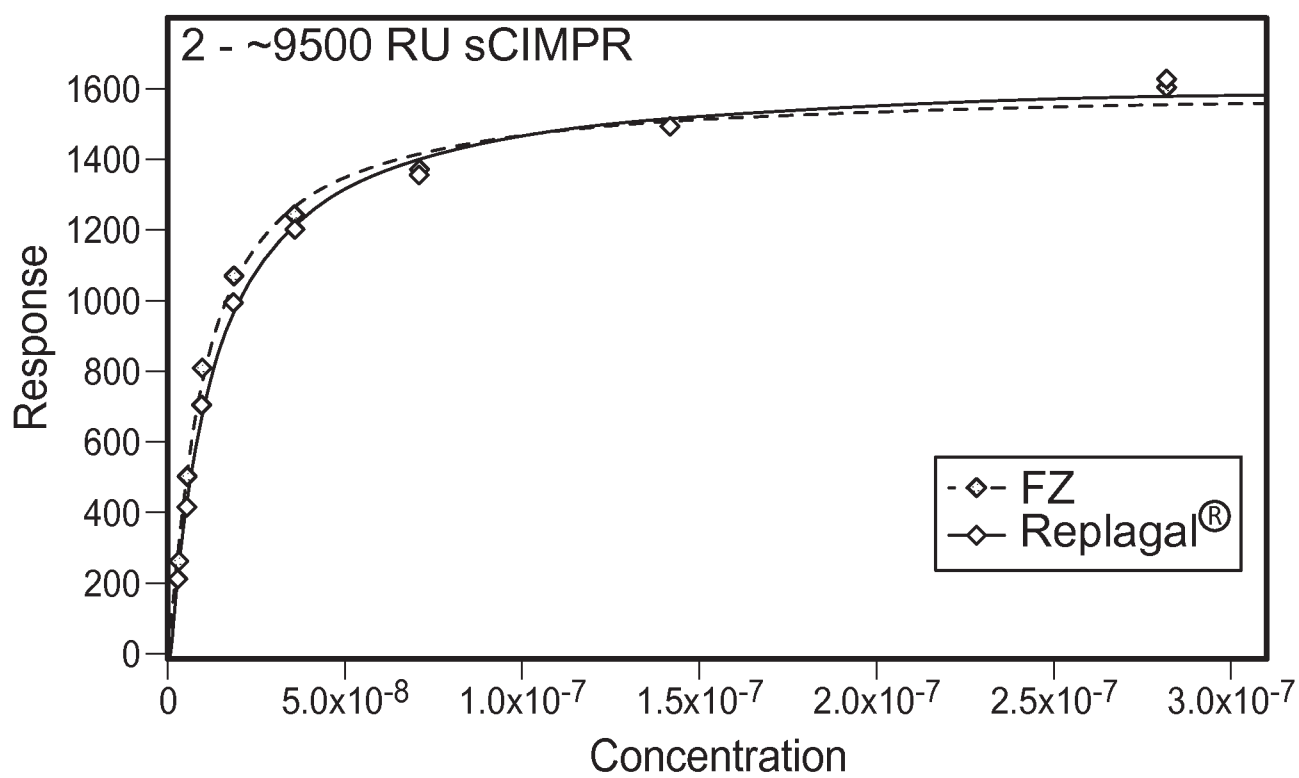


FIG. 27

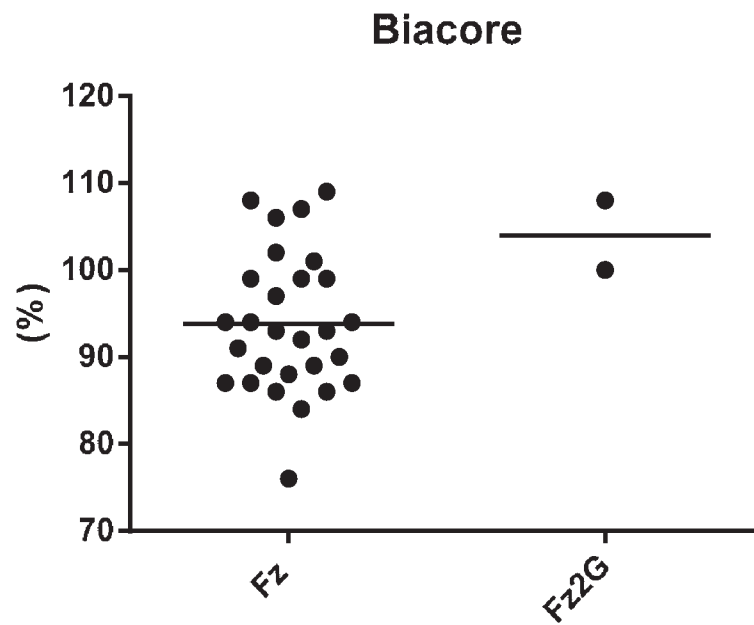


FIG. 28

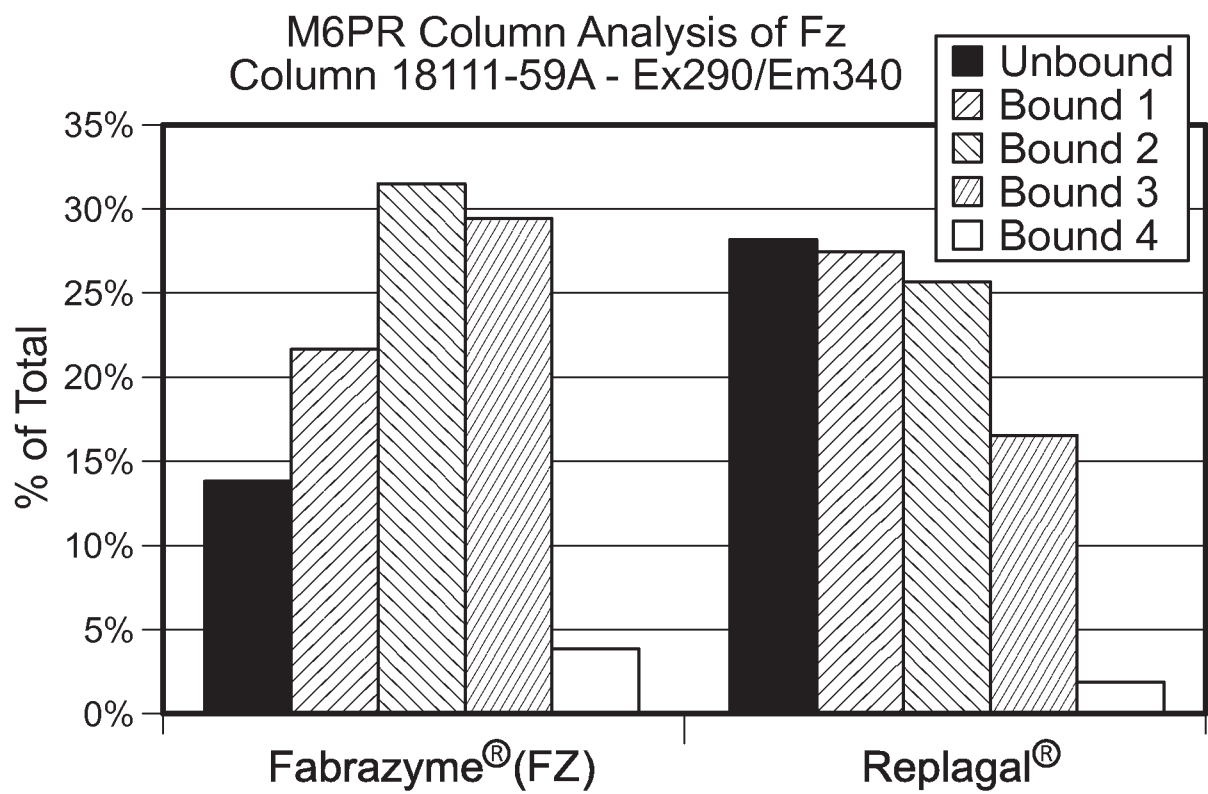


FIG. 29

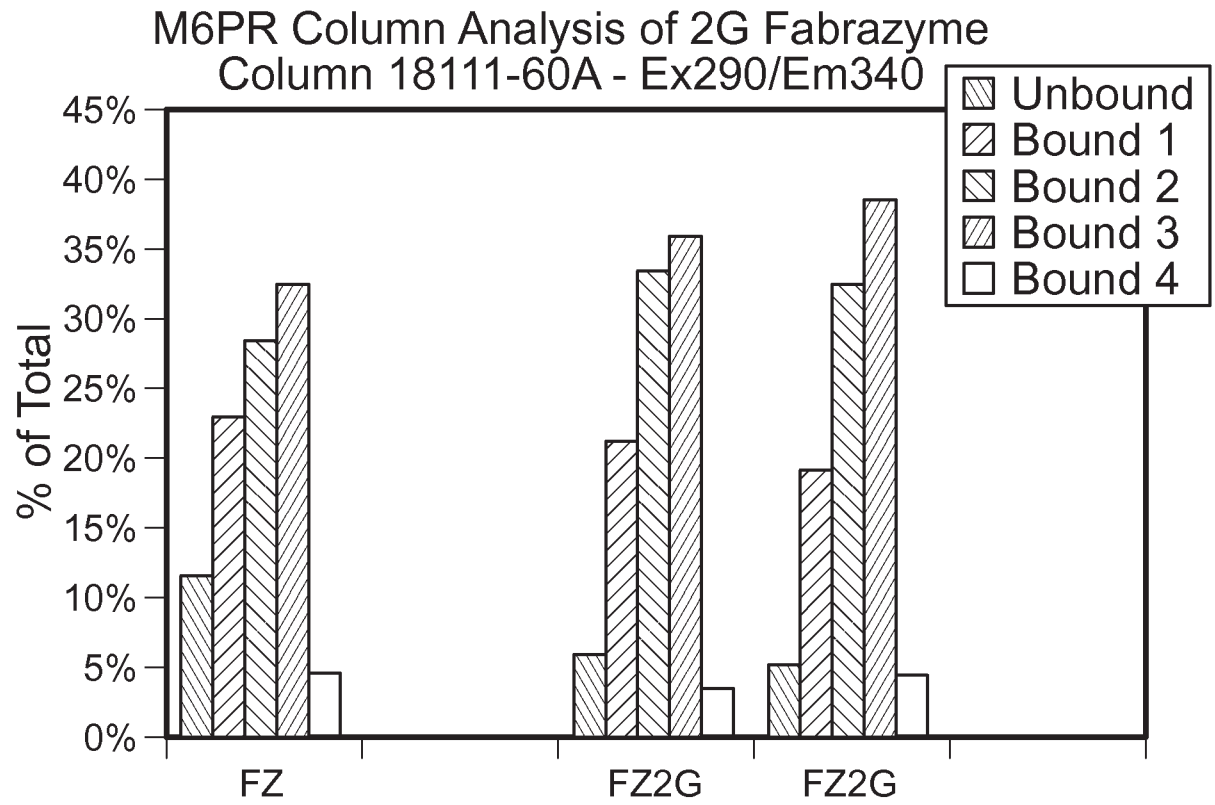


FIG. 30

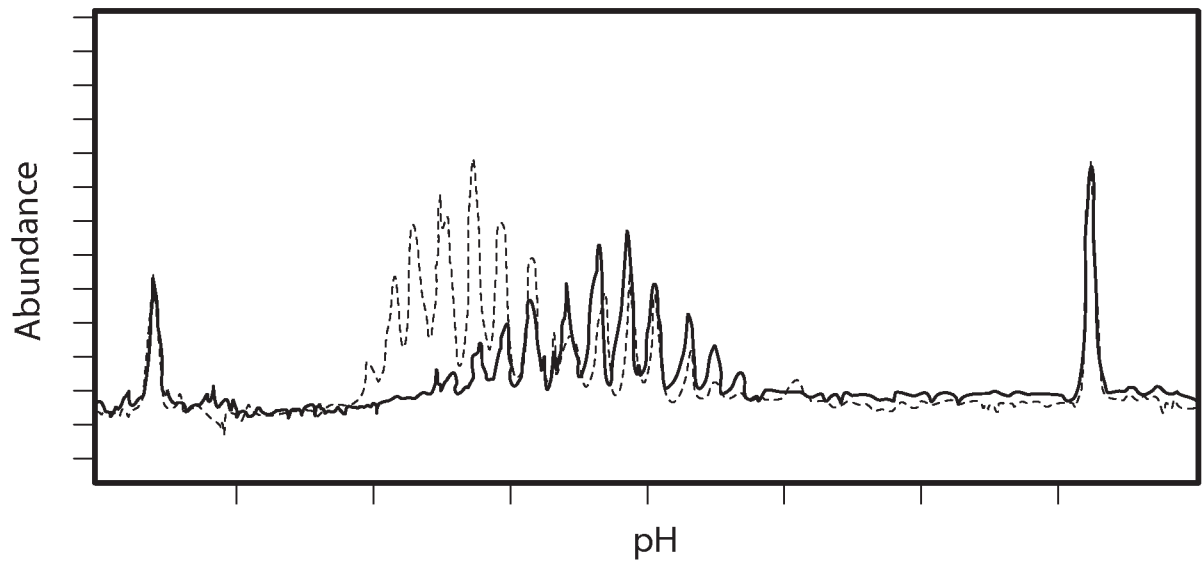
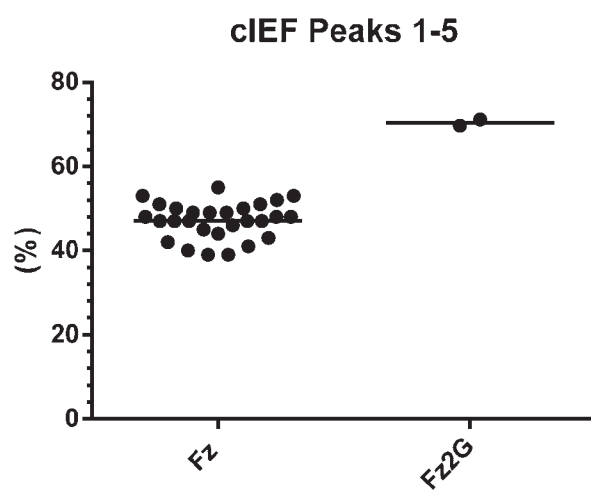


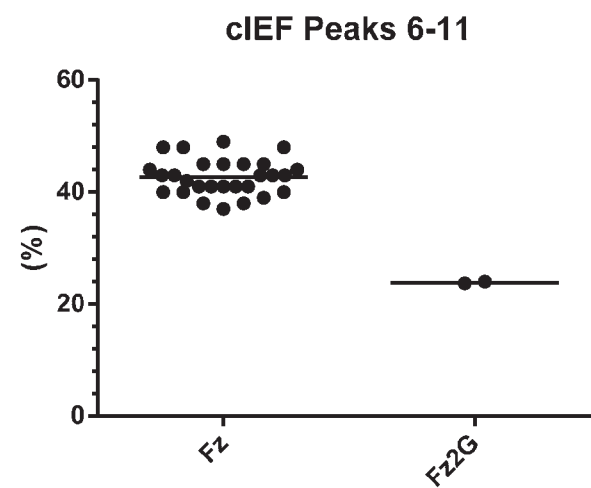
FIG. 31

29/34

A



B



C

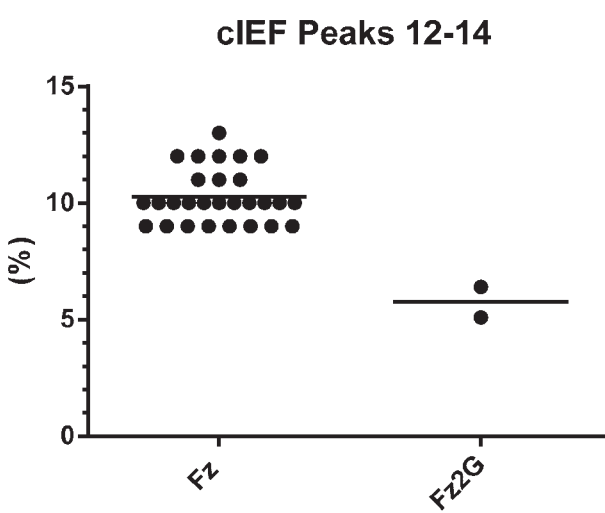


FIG. 32

30/34

Km

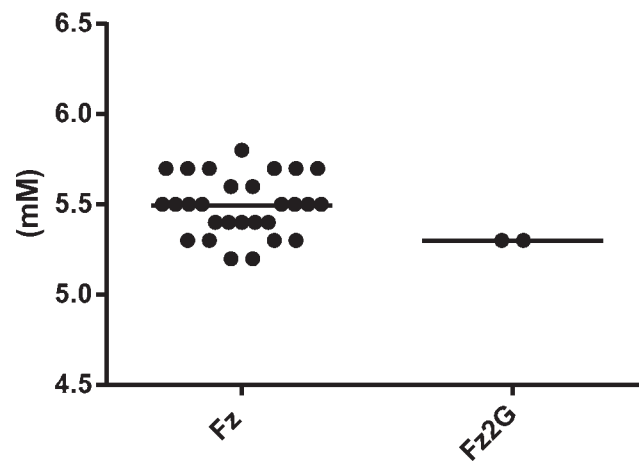


FIG. 33

Vmax

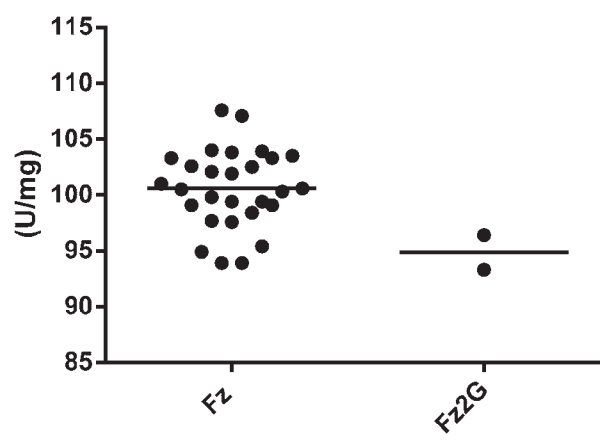
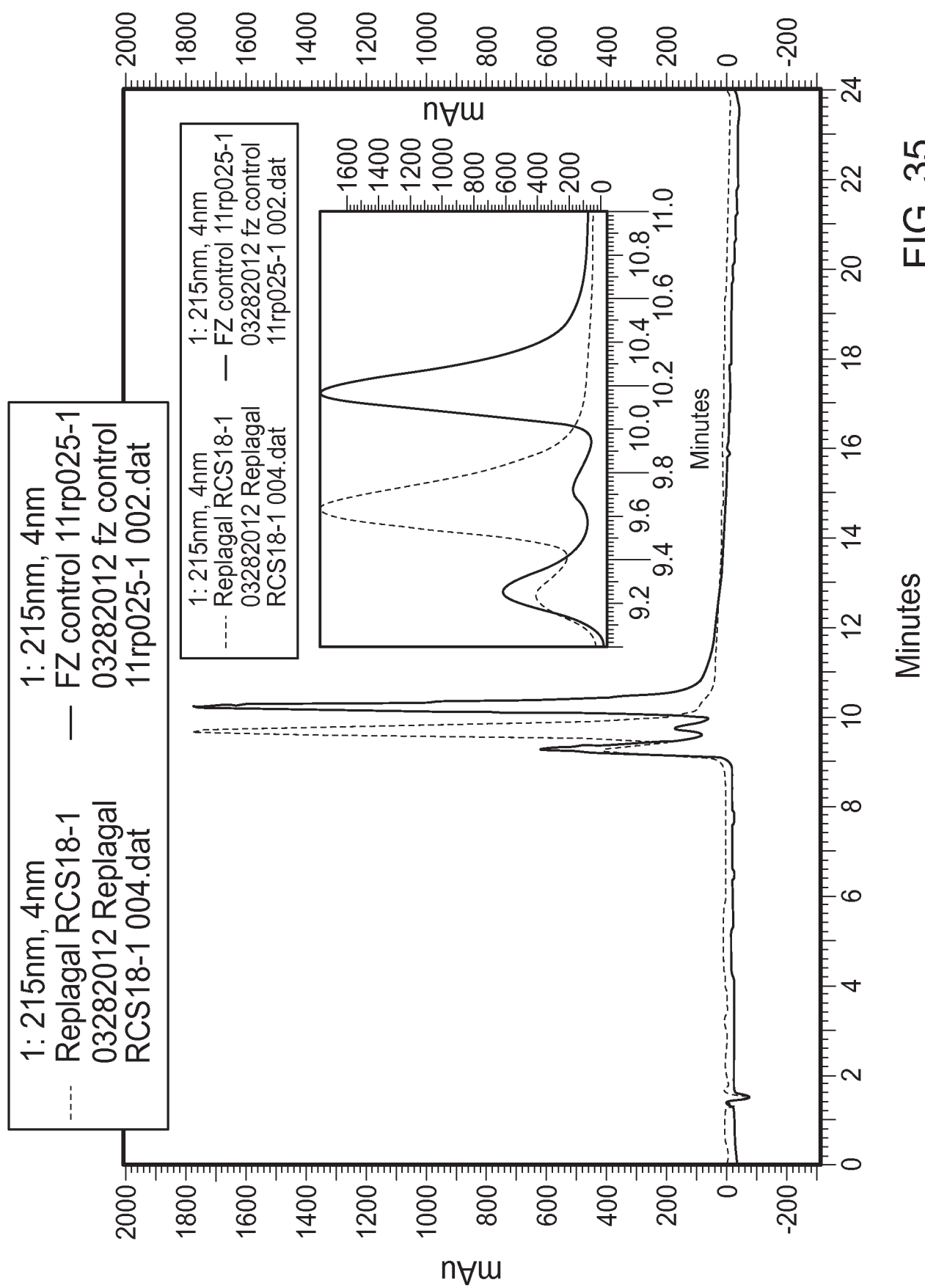


FIG. 34



32/34

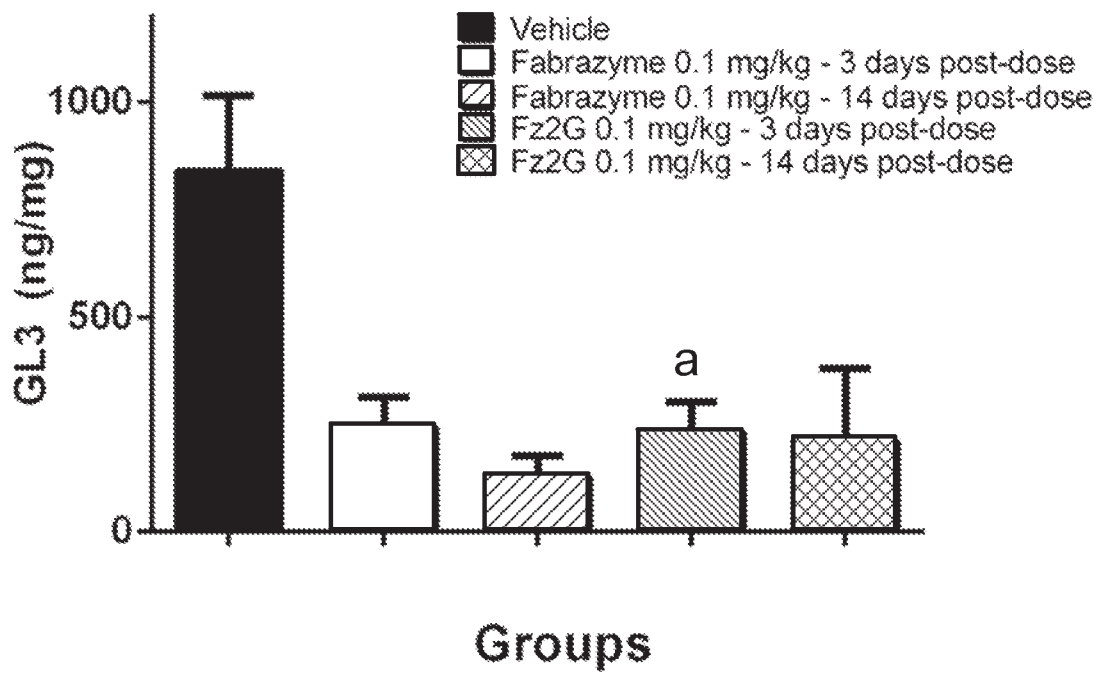


FIG. 36

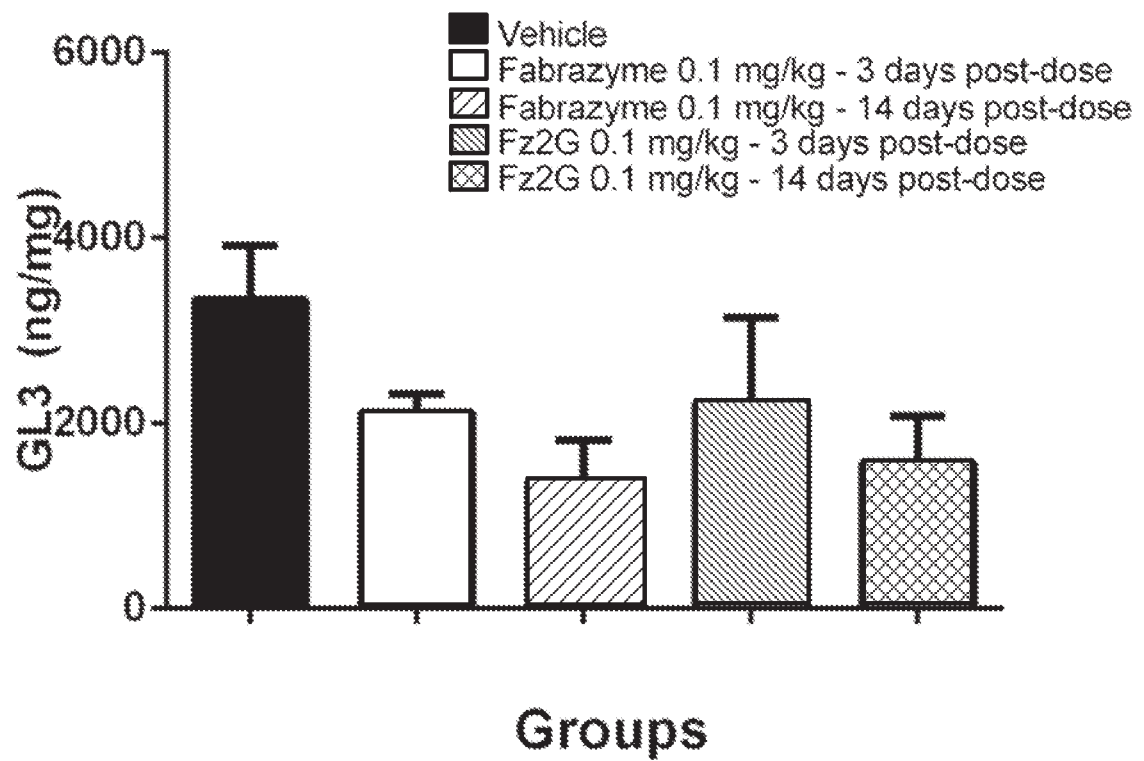


FIG. 37

33/34

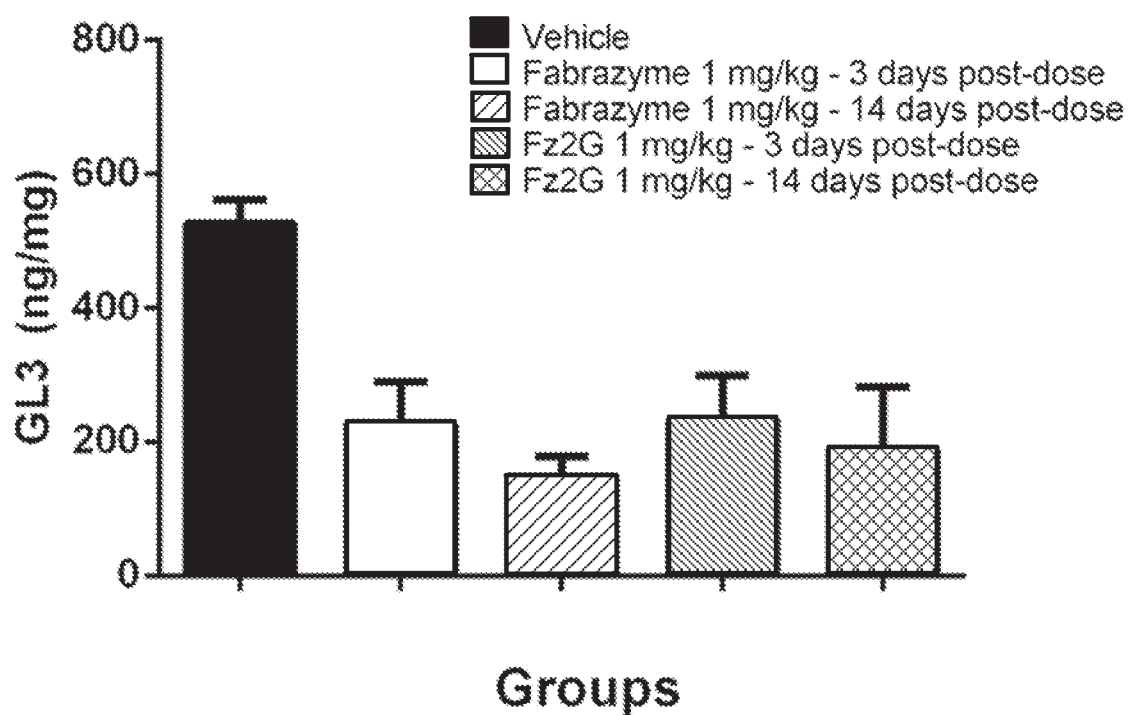


FIG. 38

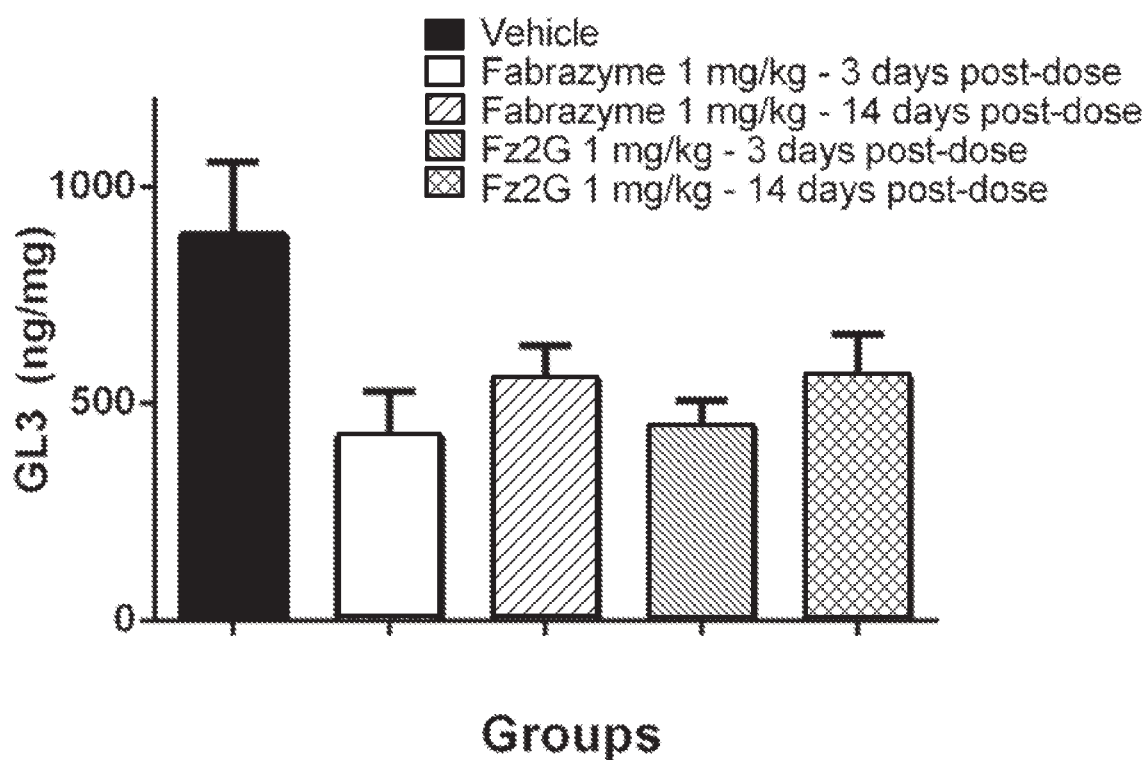


FIG. 39

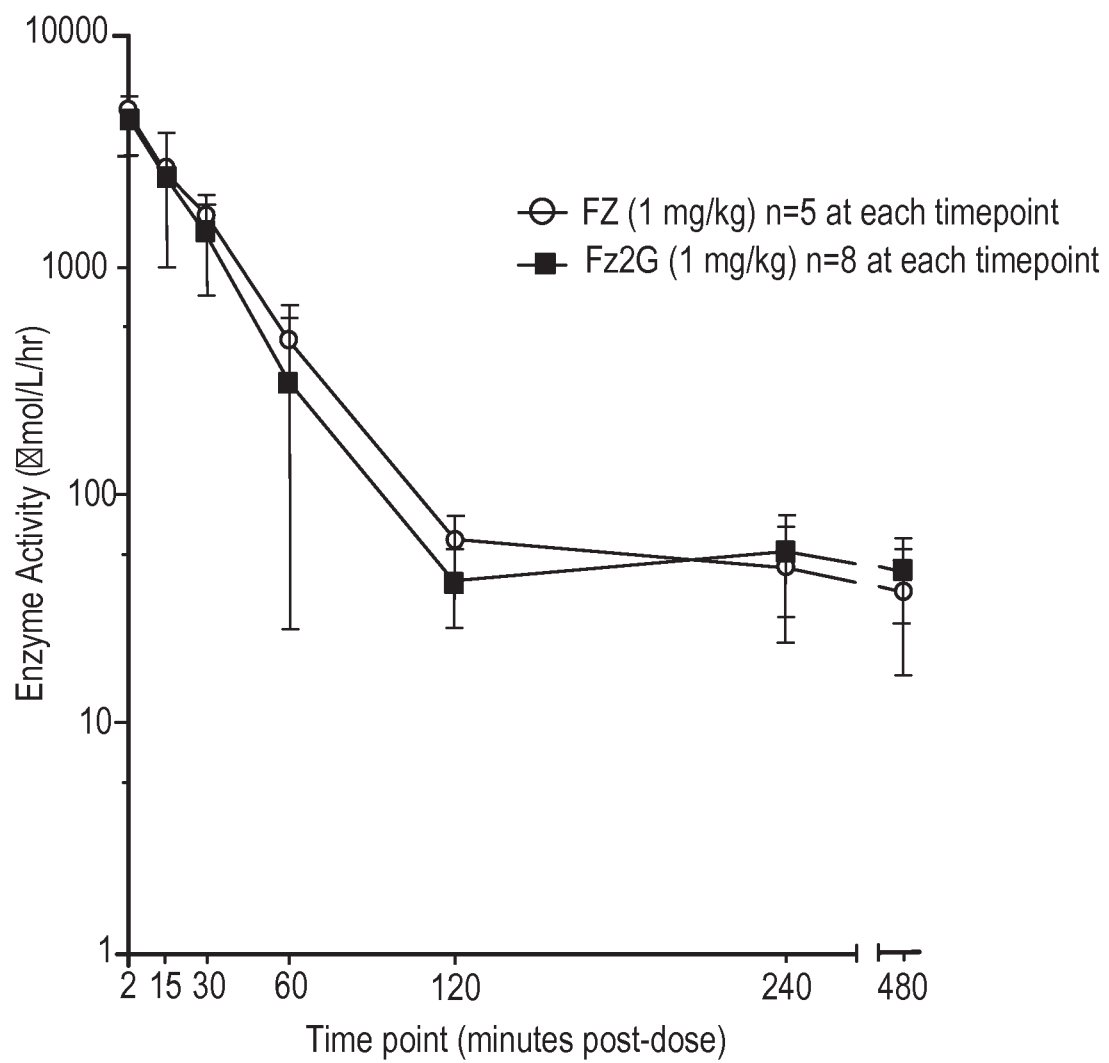


FIG. 40