FIG. 1

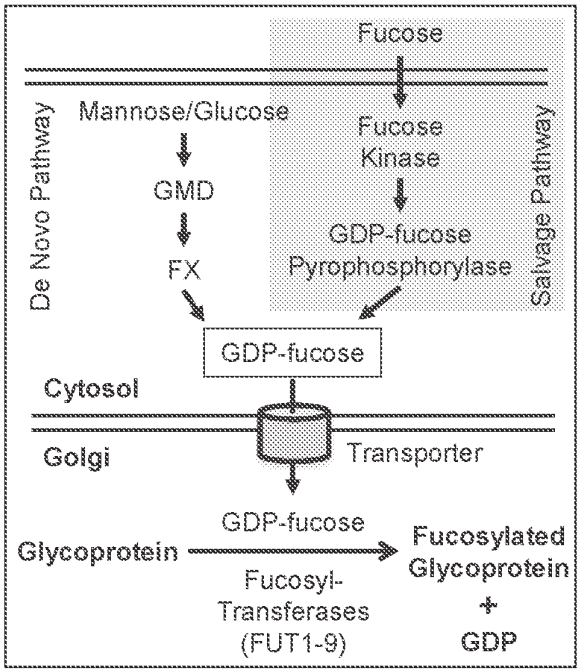

Fucose
De Novo Pathway
Mannose/Glucose
GMD
FX
Fucose Kinase
GDP-fucose Pyrophosphorylase
Salvage Pathway
GDP-fucose
Cytosol
Golgi
Transporter
GDP-fucose
Glycoprotein
Fucosyl-Transferases (FUT1-9)
Fucosylated Glycoprotein
+
GDP

FIG. 1

FIG. 2A

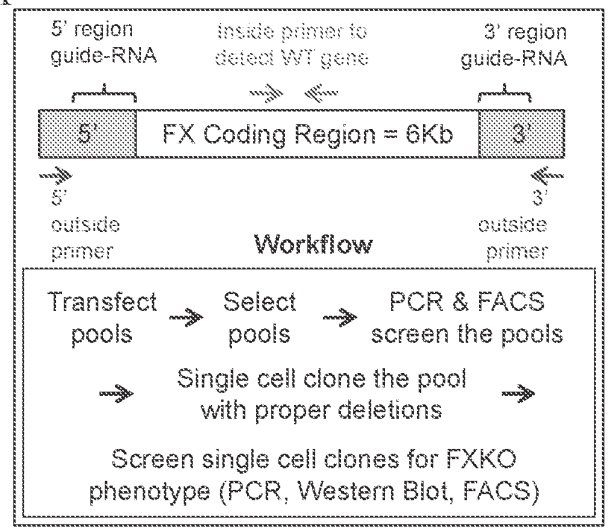
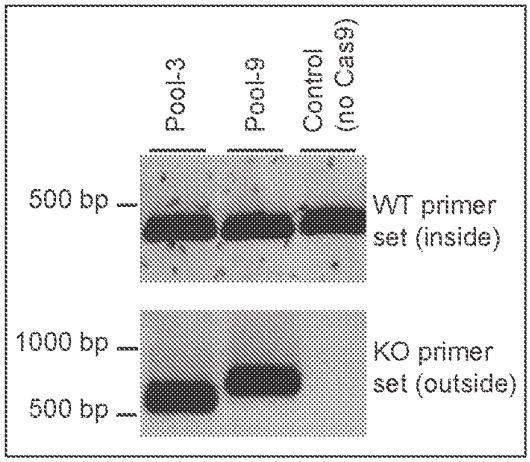


FIG. 2B



FIGS. 2A-2B

FIG. 3

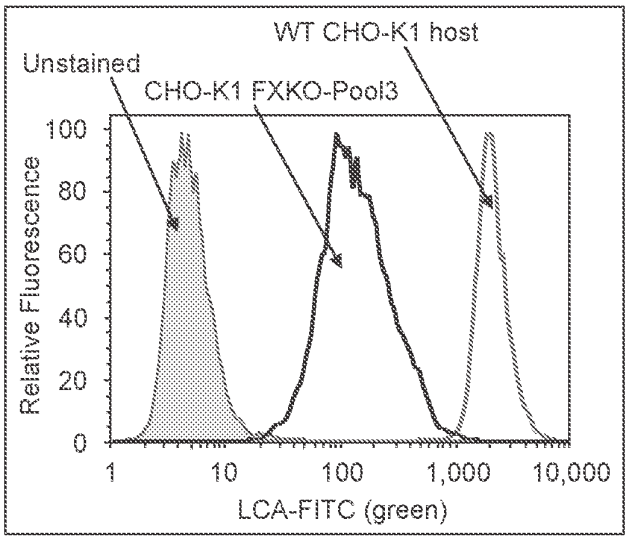

WT CHO-K1 host
Unstained
CHO-K1 FXKO-Pool3
Relative Fluorescence
100
80
60
40
20
0
1
10
100
1,000
10,000
LCA-FITC (green)

FIG. 3

FIG. 4A

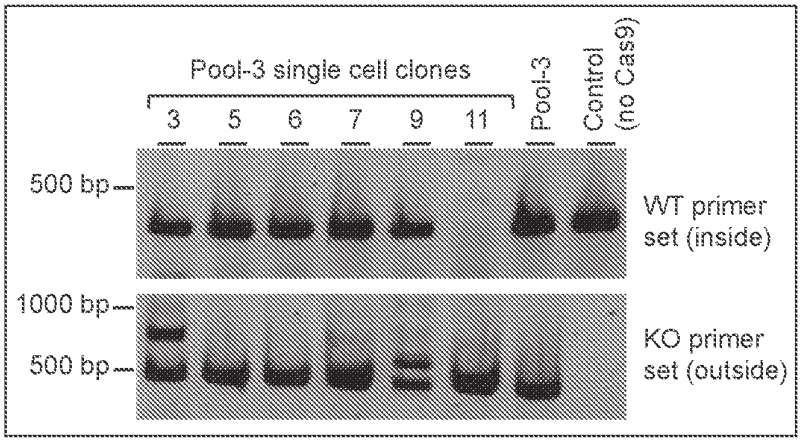
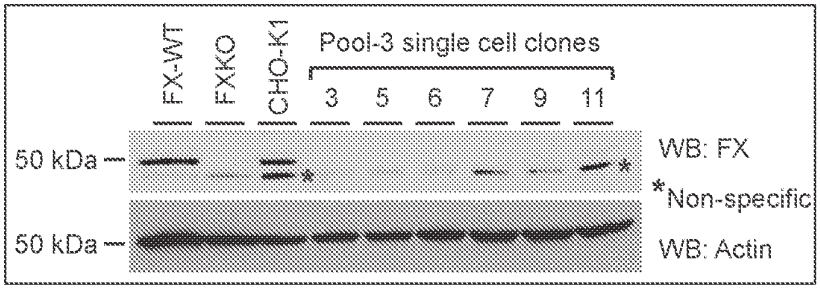
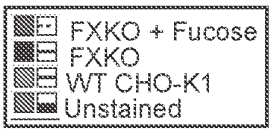


FIG. 4B



FIGS. 4A-4B

FIG. 5A



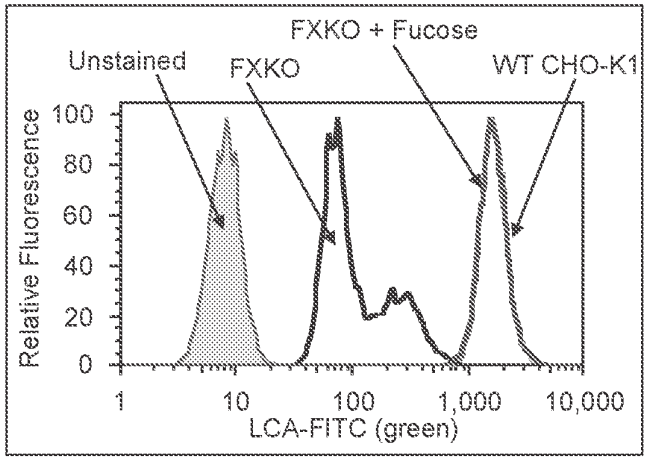
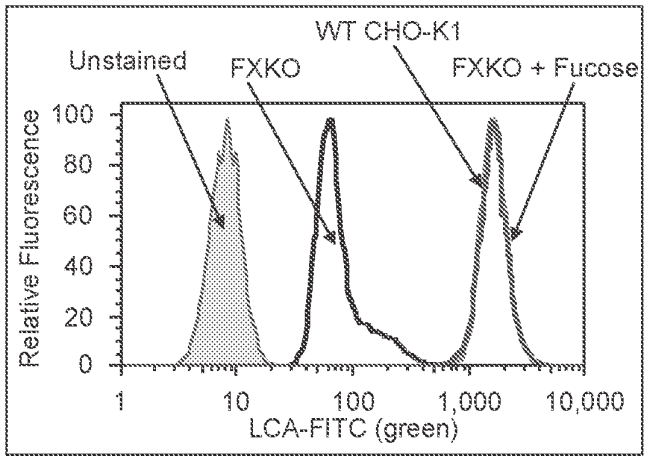


FIG. 5B



FIGS. 5A-5B

FIG. 5C

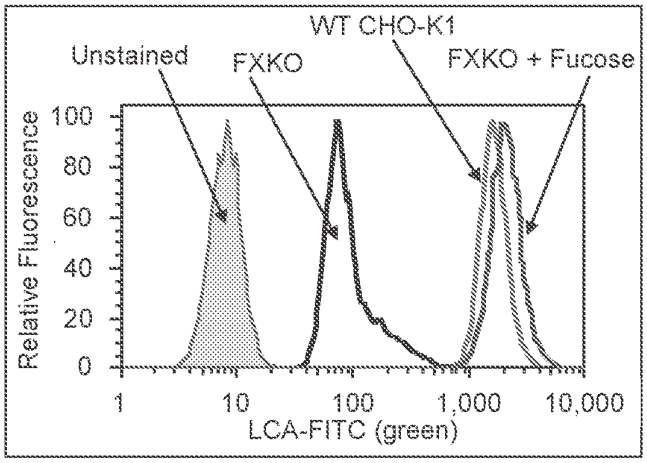
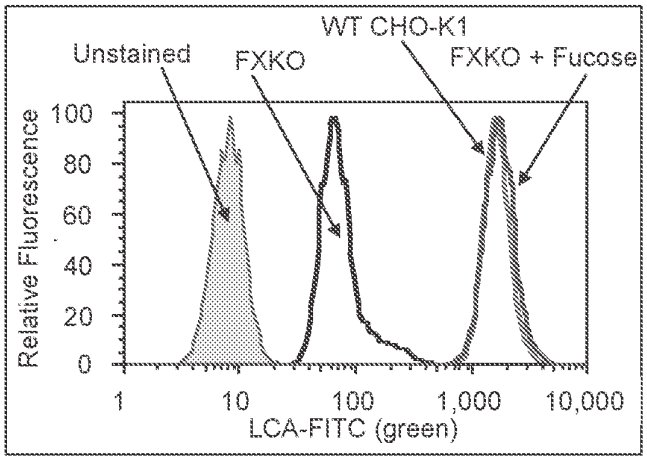


FIG. 5D



FIGS. 5C-5D

FIG. 5E

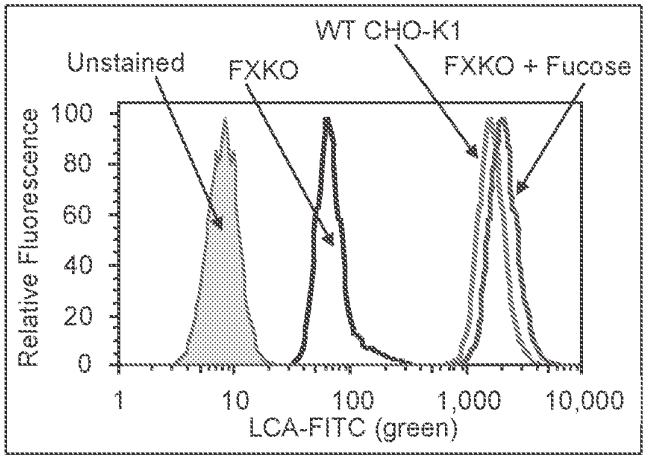
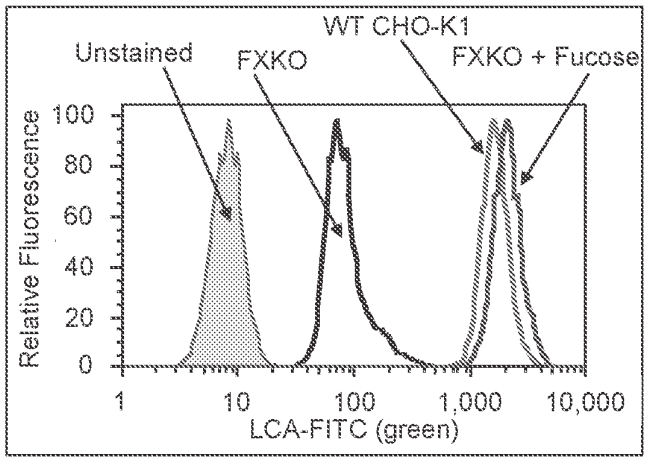


FIG. 5F



FIGS. 5E-5F

FIG. 6A

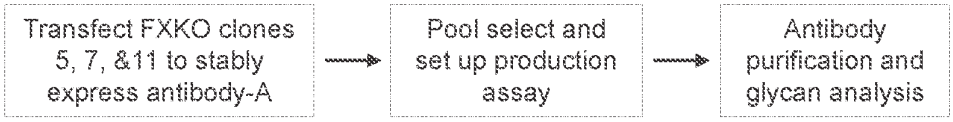
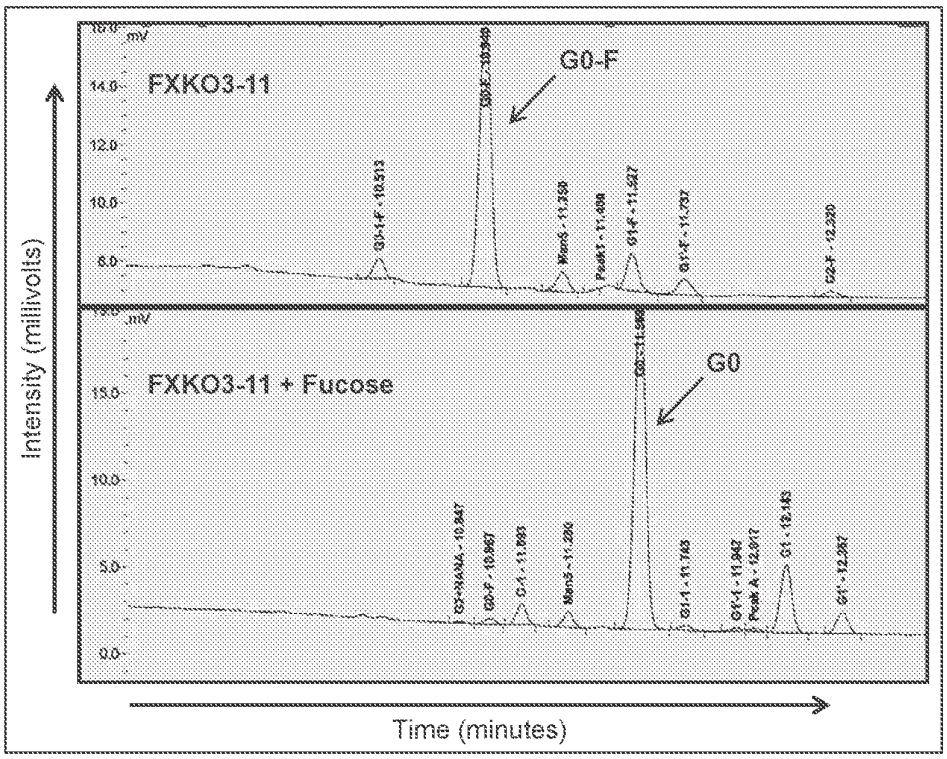


FIG. 6B



FIGS. 6A-6B

FIG. 6C

Glycan type	G-1	G0	G1	G1'	G2	Man5	G0-1-F	G0-F	G1-F	G1'-F	G2-F
FXKO3-5	n/a	n/a	n/a	n/a	n/a	5.1	6.2	70.2	11.0	5.1	1.9
FXKO3-7	n/a	n/a	n/a	n/a	n/a	4.7	5.0	68.2	13.3	6.1	2.3
FXKO3-11	n/a	n/a	n/a	n/a	n/a	5.2	5.4	73.1	9.6	4.9	1.6
CHO-K1	9.5	64.6	11.8	4.0	1.5	3.7	n/a	1.4	n/a	n/a	n/a
FXKO3-5 +Fucose	5.2	64.8	14.8	4.8	2.3	3.2	n/a	1.8	n/a	n/a	n/a
FXKO3-7 +Fucose	3.7	63.2	18.2	5.6	2.9	2.3	n/a	1.4	n/a	n/a	n/a
FXKO3-11 +Fucose	4.1	69.0	13.8	4.3	2.3	3.1	n/a	1.2	n/a	n/a	n/a

FIG. 6C

FIG. 7A

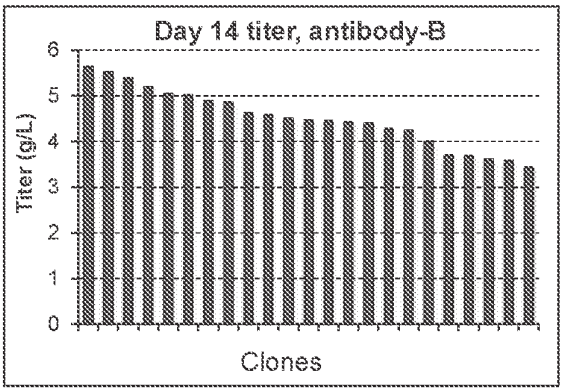
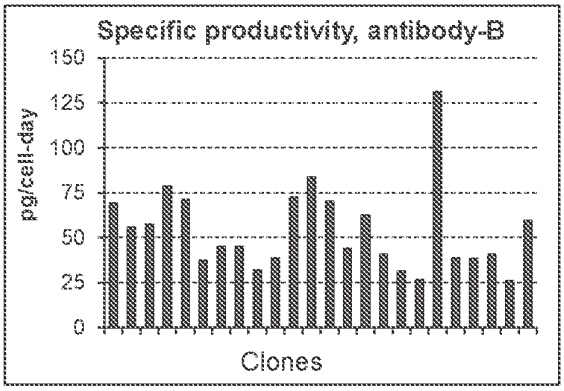


FIG. 7B



FIGS. 7A-7B

FIG. 7C

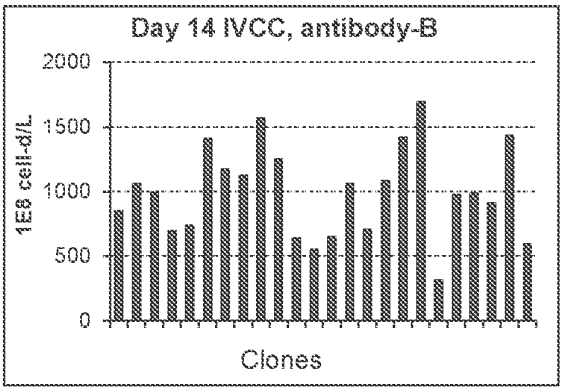
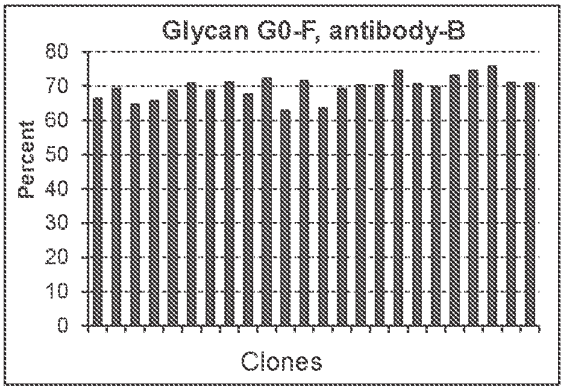


FIG. 7D



FIGS. 7C-7D

FIG. 7E

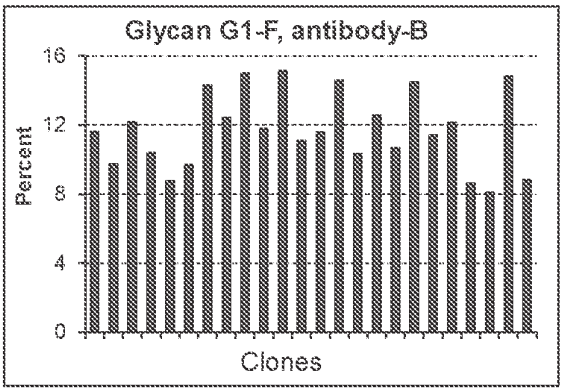
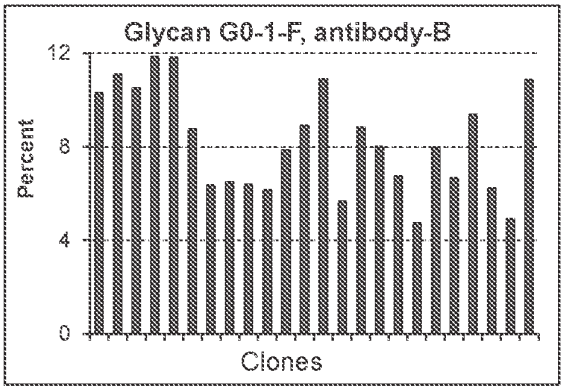


FIG. 7F



FIGS. 7E-7F

FIG. 8A

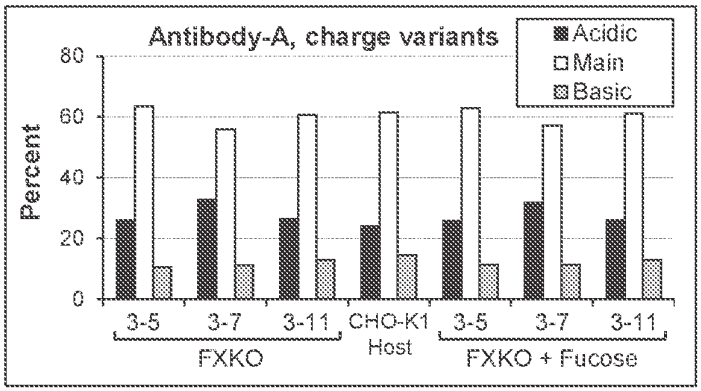
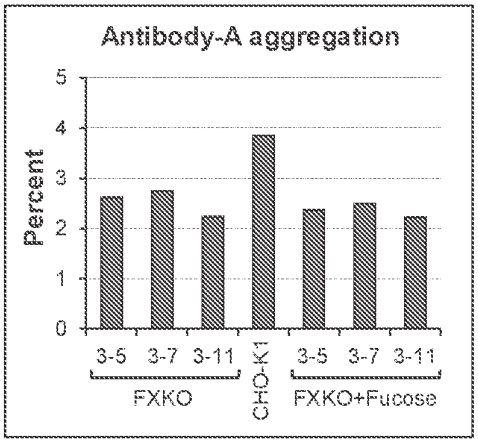


FIG. 8B



FIGS. 8A-8B

FIG. 8C

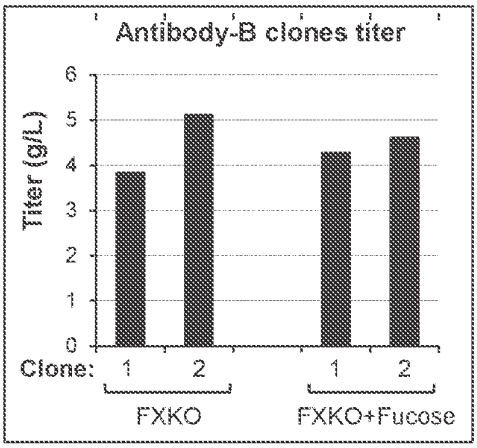
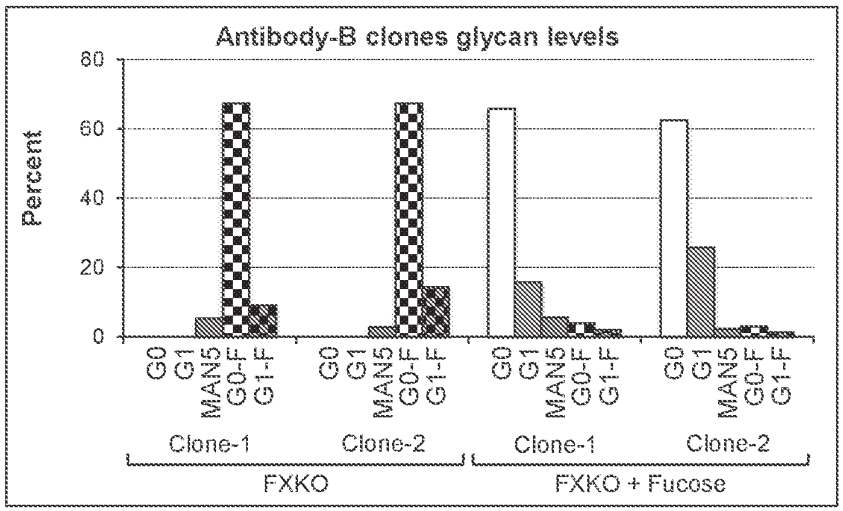


FIG. 8D



FIGS. 8C-8D

FIG. 8E

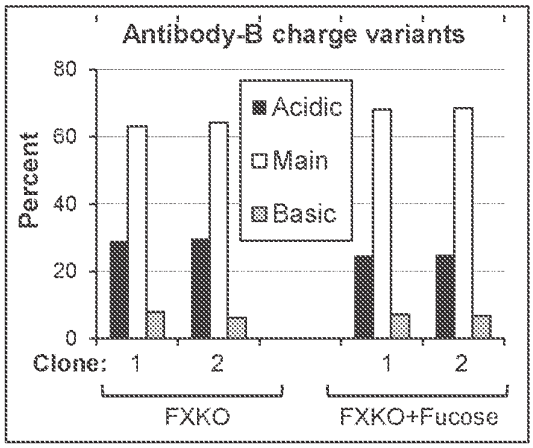
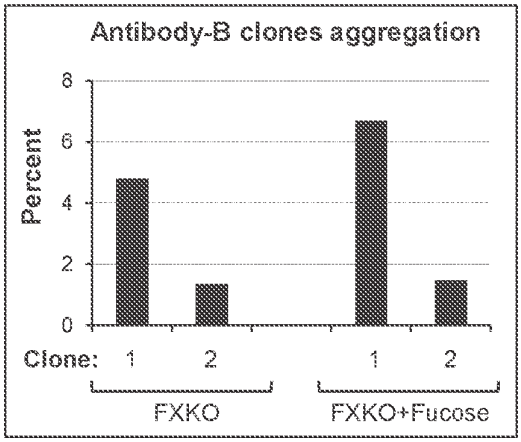


FIG. 8F



FIGS. 8E-8F

FIG. 9A

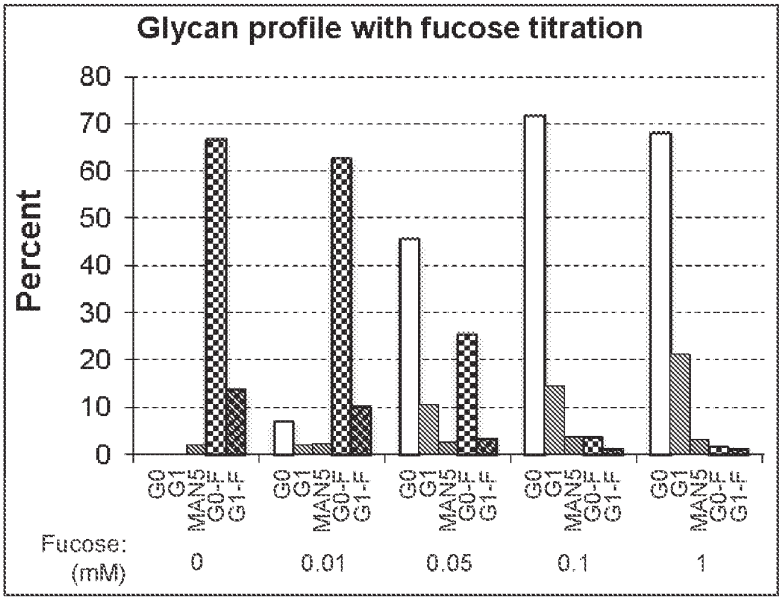
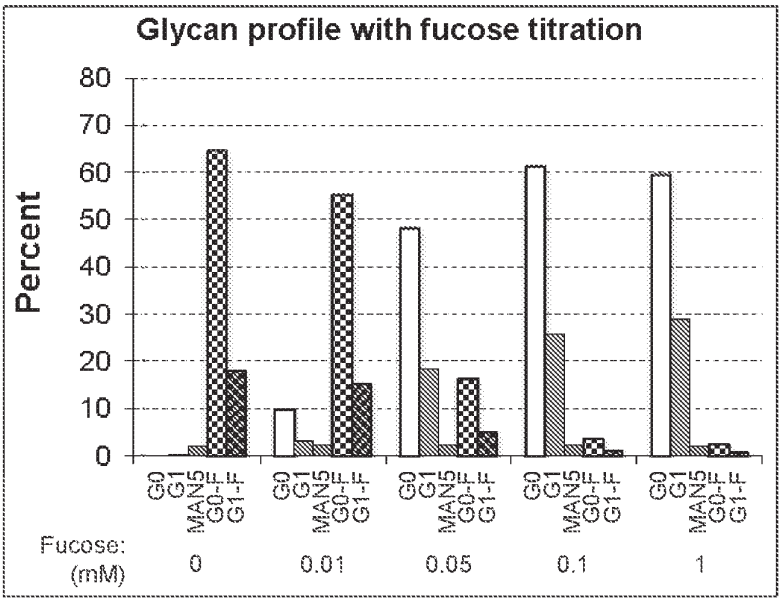


FIG. 9B



FIGS. 9A-9B

FIG. 9C

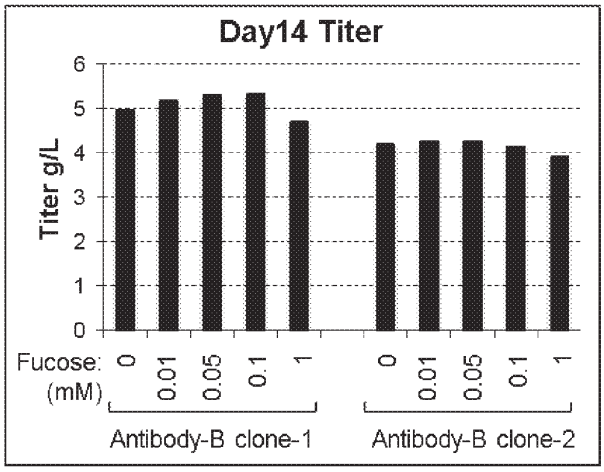
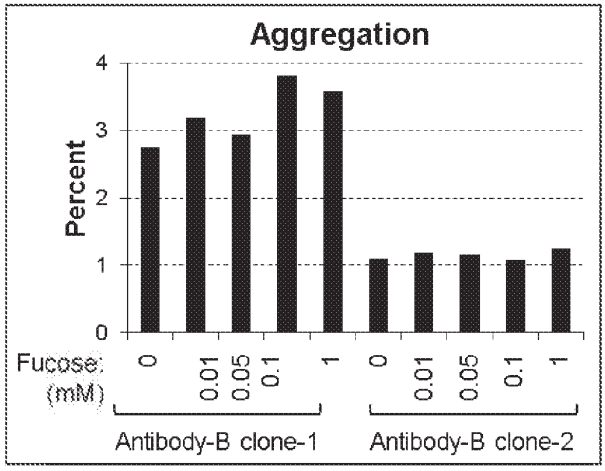


FIG. 9D



FIGS. 9C-9D

FIG. 9E

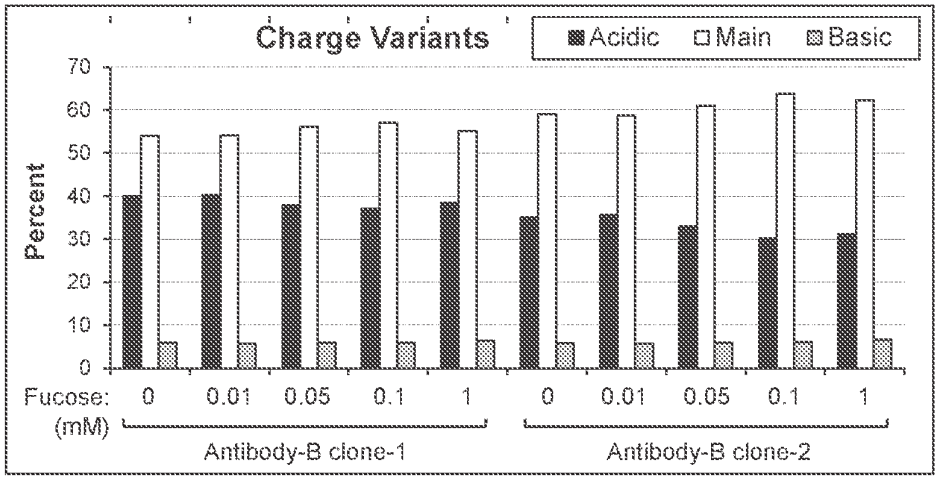
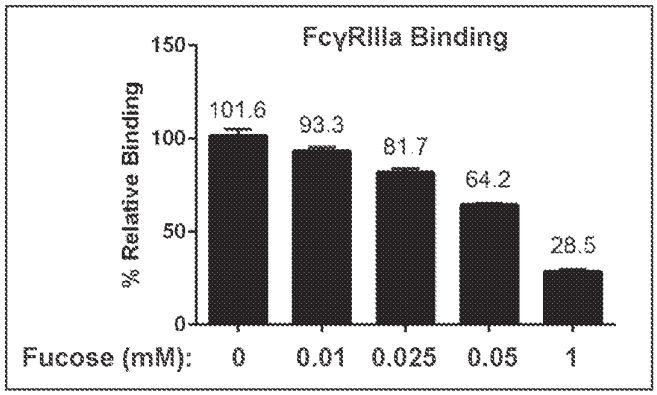

Charge Variants
Acidic
Main
Basic
Percent
70
60
50
40
30
20
10
0
Fucose: (mM)
0
0.01
0.05
0.1
1
0
0.01
0.05
0.1
1
Antibody-B clone-1
Antibody-B clone-2

FIG. 9E

FIG. 10A

Fucose concentration (mM)	G0-F	G1-F	G2-F	Man5	G0	G1	G2
0	67.3	14.3	1	2.8	0	0	0
0.01	54.5	11.6	0.8	2.9	13.4	4	0.4
0.025	32	7.1	0.5	2.9	34.4	11	1.3
0.05	16.5	5.2	0.4	2.2	48.4	18.3	2.3
1	3	1.3	0	2.4	62.5	25.8	2.5

FIG. 10B

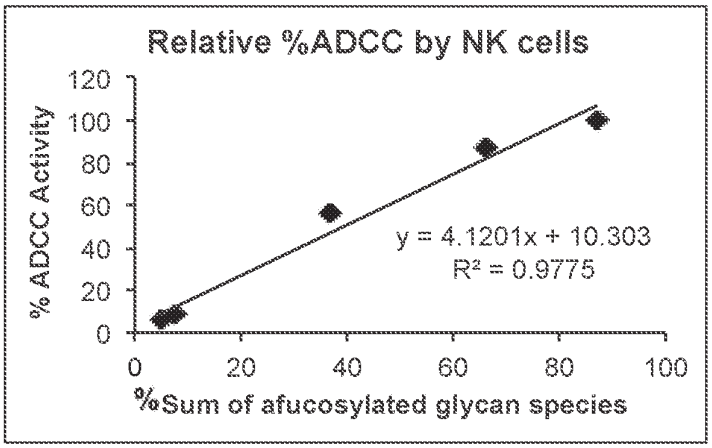


FIGS. 10A-10B

FIG. 10C

Fucose (mM)	G0-F	G1-F	G2-F	Man5	G0	G1	G2
0	73.6	9.5	0.9	2.9	0.2	0.5	0
0.01	54.9	7.6	0.8	3	17.8	3	0.5
0.02	29.4	4.7	0.4	2.3	46.7	7.4	1
0.03	9.6	2.1	0.2	3.3	67.5	10.5	1.4
0.05	3.2	0.9	0.1	3.2	74.4	12.5	1.6
1	2.1	0.7	0	2.1	76.5	14.4	1.9

FIG. 10D

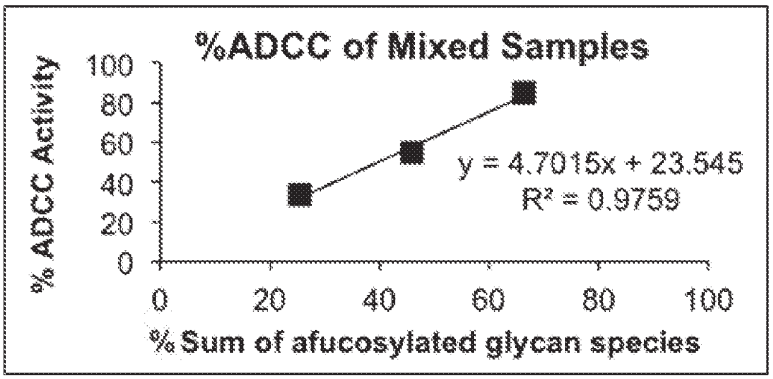


FIGS. 10C-10D

FIG. 10E

Fucose (mM)	% sum of afucosylated glycans	% relative ADCC Activity
1:3 mix (0&1 mM)	25.4	34
1:1 mix (0&1 mM)	45.9	54
3:1 mix (0&1 mM)	66.4	85

FIG. 10F



FIGS. 10E-10F

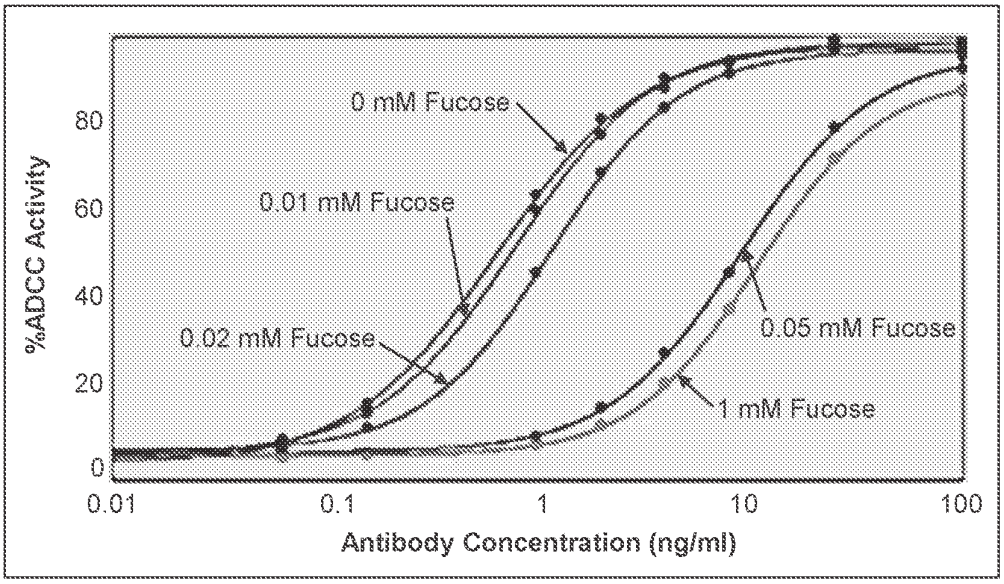

%ADCC Activity
80
60
40
20
0
0.01
0.1
1
10
100
Antibody Concentration (ng/ml)
0 mM Fucose
0.01 mM Fucose
0.02 mM Fucose
0.05 mM Fucose
1 mM Fucose

FIG. 10G