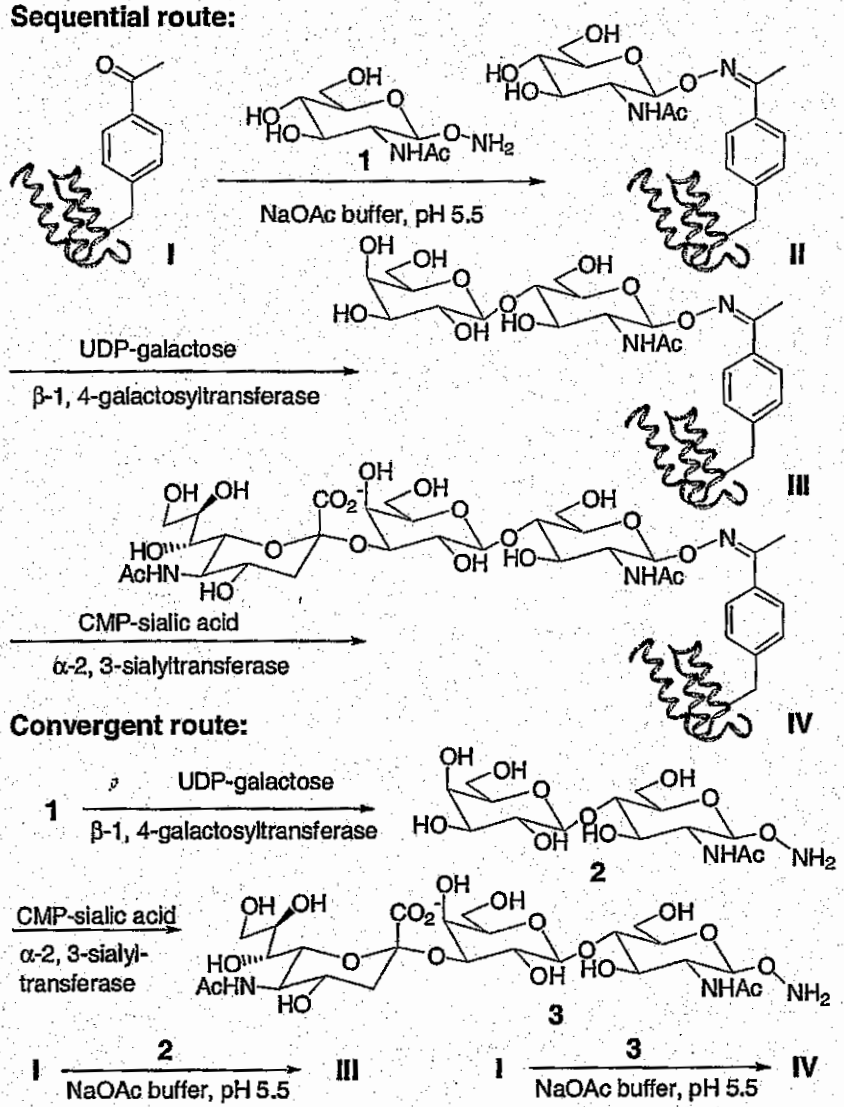

Sequential route:
1
NaOAc buffer, pH 5.5
I
II
UDP-galactose
β-1, 4-galactosyltransferase
III
CMP-sialic acid
α-2, 3-sialyltransferase
IV
Convergent route:
1
UDP-galactose
β-1, 4-galactosyltransferase
2
CMP-sialic acid
α-2, 3-sialyl-
transferase
3
I
2
NaOAc buffer, pH 5.5
III
I
3
NaOAc buffer, pH 5.5
IV

Fig. 1

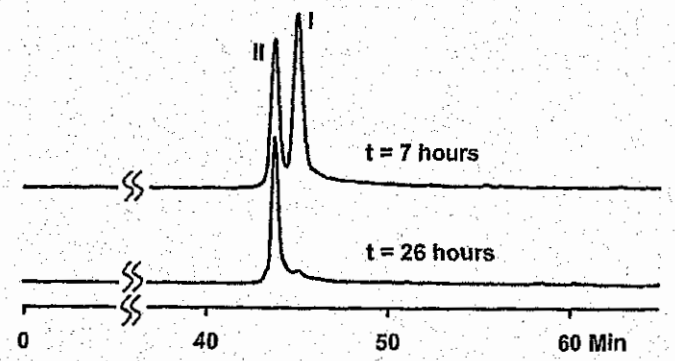

II
I
t = 7 hours
t = 26 hours
0
40
50
60 Min

Fig. 2

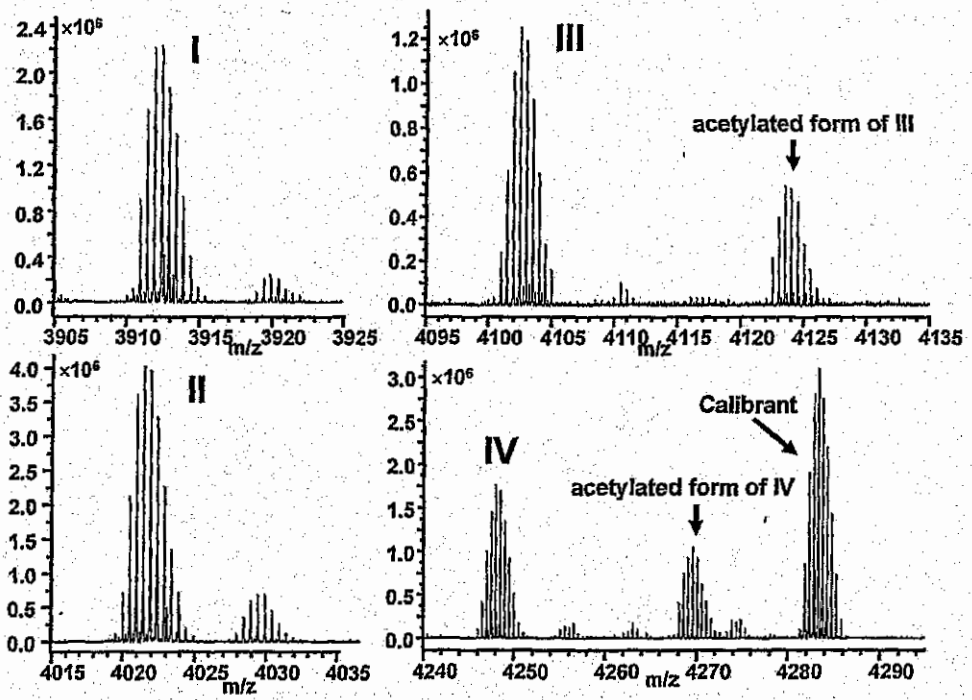

I
III
acetylated form of III
II
IV
Calibrant
acetylated form of IV
m/z

Fig. 3

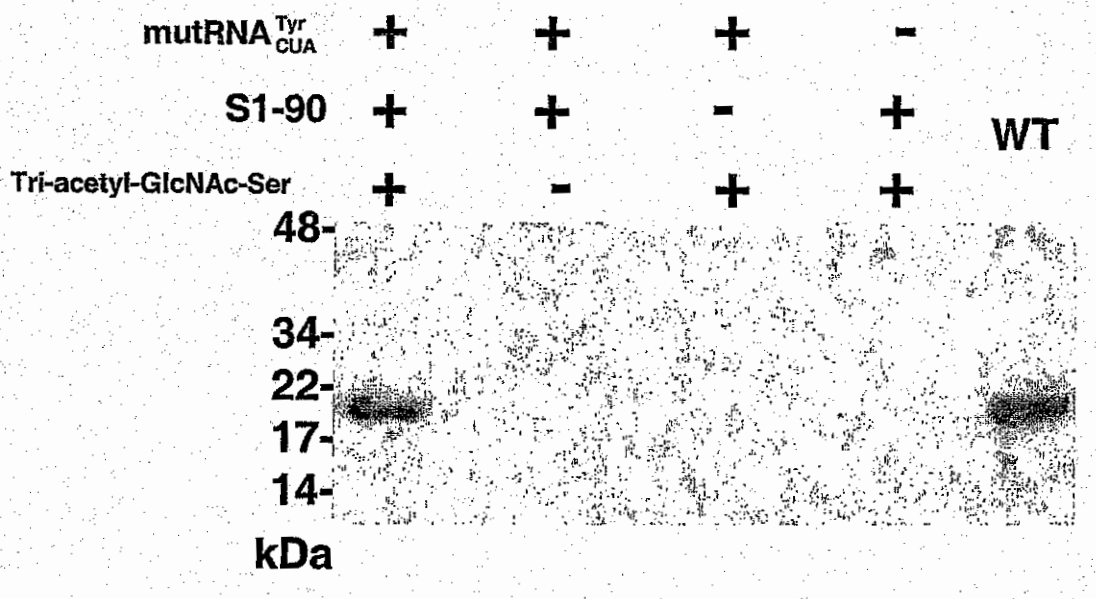

mutRNA Tyr CUA + + + -
S1-90 + + - + WT
Tri-acetyl-GlcNAc-Ser + - + +
48-
34-
22-
17-
14-
kDa

Fig. 4

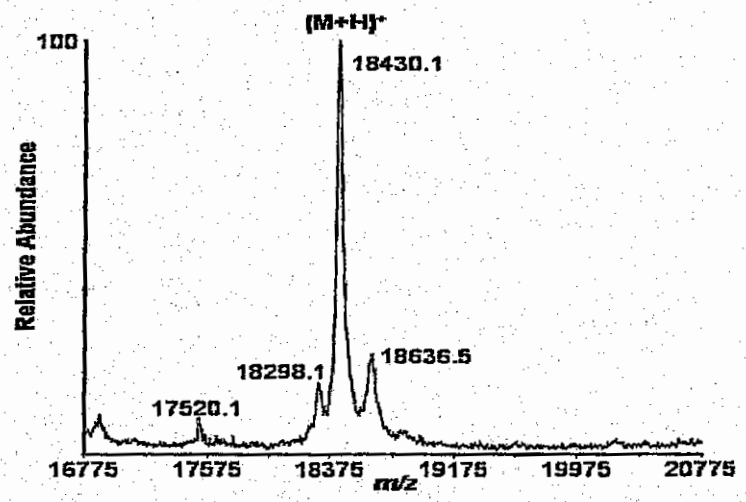

(M+H)+
100
18430.1
Relative Abundance
18298.1
18636.5
17520.1
16775
17575
18375
19175
19975
20775
m/z

Fig. 5

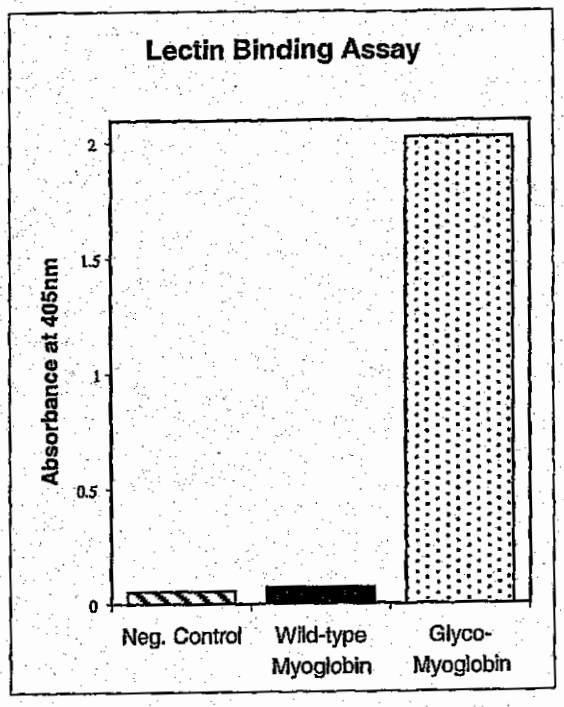

Lectin Binding Assay
Absorbance at 405nm
2
1.5
1
0.5
0
Neg. Control
Wild-type Myoglobin
Glyco-Myoglobin

Fig. 6A

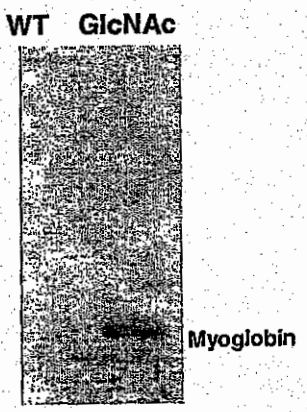

WT
GlcNAc
Myoglobin

Fig. 6B

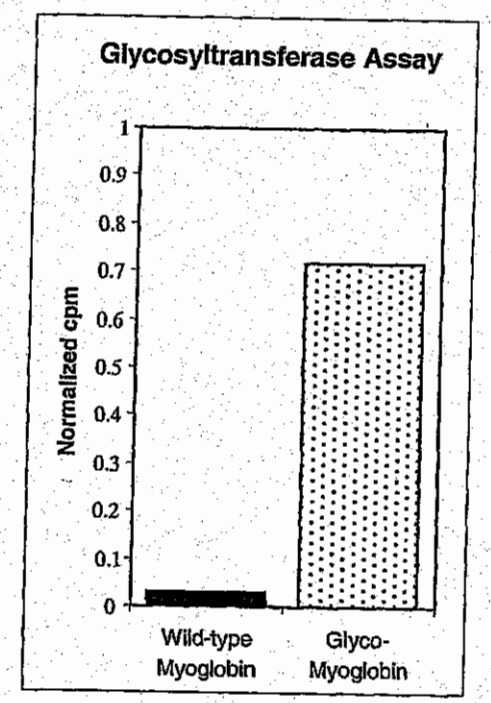

Glycosyltransferase Assay
Normalized cpm
1
0.9
0.8
0.7
0.6
0.5
0.4
0.3
0.2
0.1
0
Wild-type Myoglobin
Glyco-Myoglobin

Fig. 6C