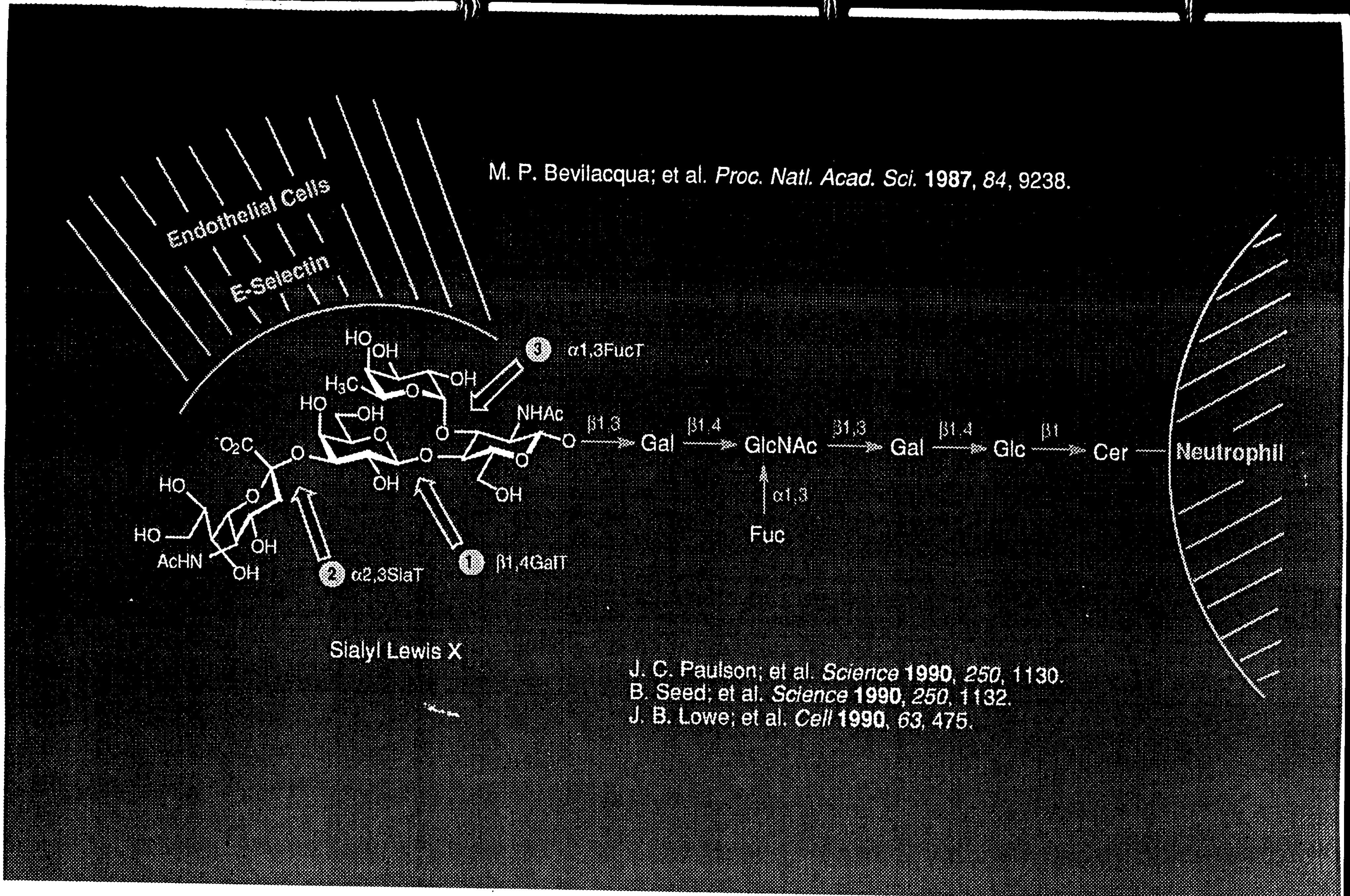
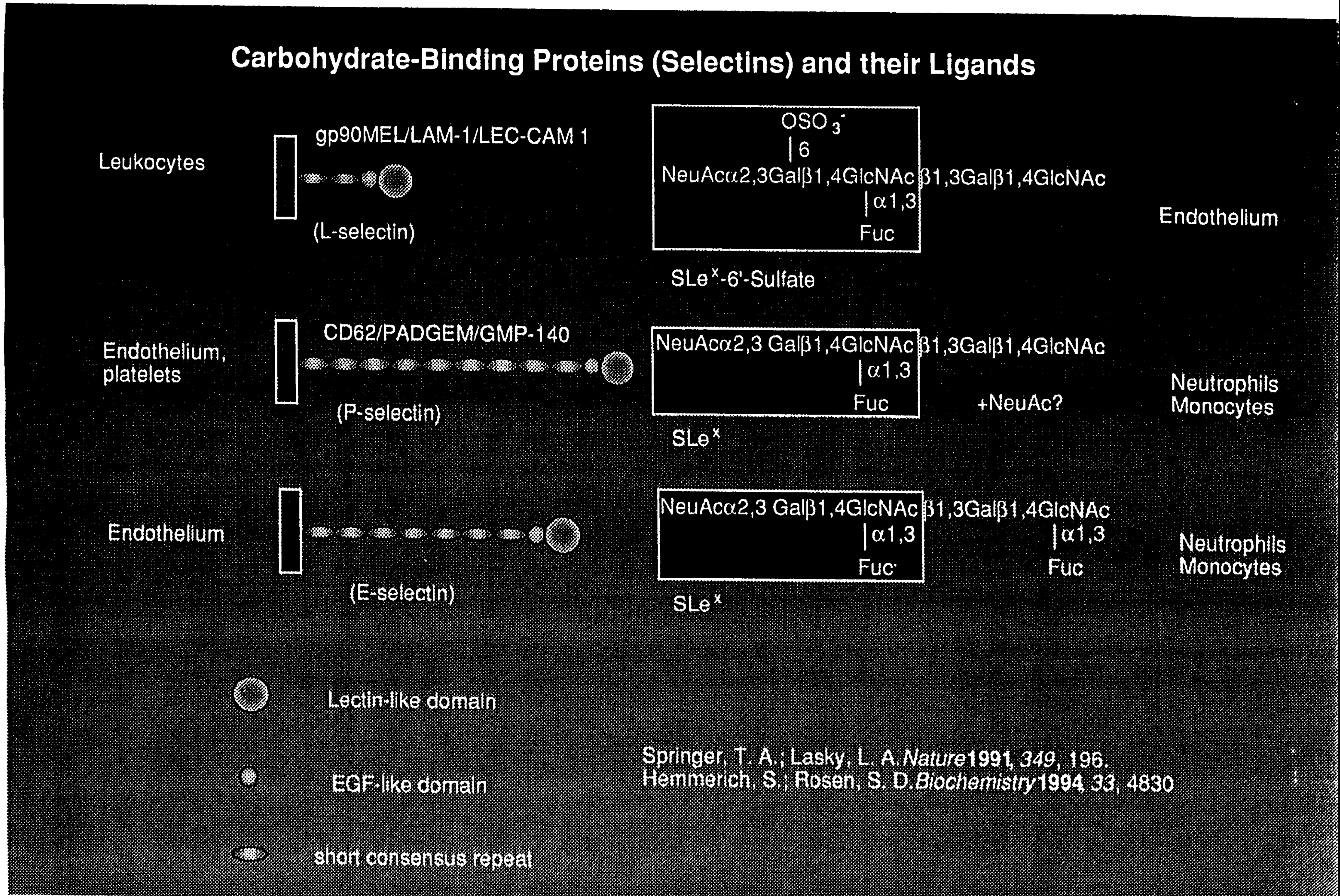
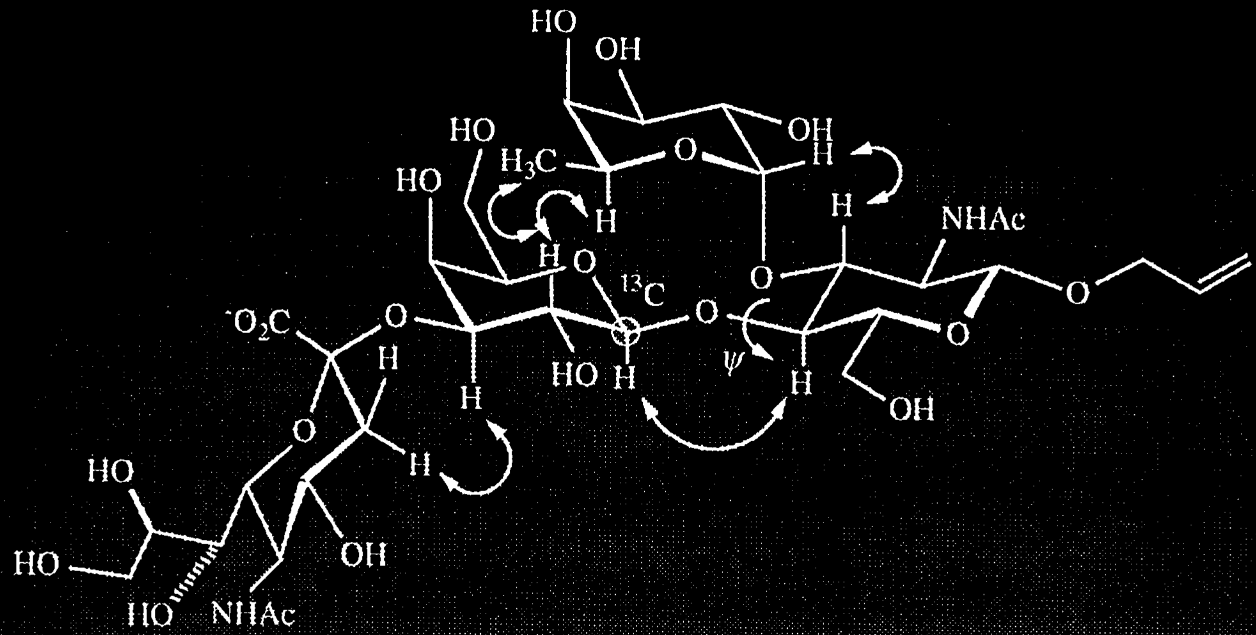

M. P. Bevilacqua; et al. Proc. Natl. Acad. Sci. 1987, 84, 9238.
Endothelial Cells
E-Selectin
HO
OH
H3C
OH
HO
OH
NHAc
-O2C
HO
HO
AcHN
OH
OH
OH
OH
3 α1,3FucT
1 β1,4GalT
2 α2,3SiaT
β1,3 Gal β1,4 GlcNAc β1,3 Gal β1,4 Glc β1 Cer — Neutrophil
α1,3
Fuc
Sialyl Lewis X
J. C. Paulson; et al. Science 1990, 250, 1130.
B. Seed; et al. Science 1990, 250, 1132.
J. B. Lowe; et al. Cell 1990, 63, 475.



WongC/CHEM/dia2c/062?94

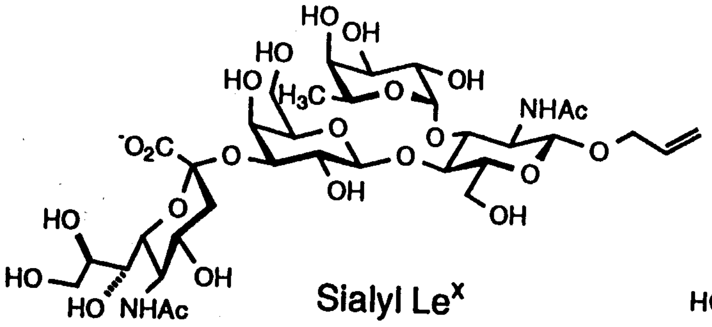


Calculated $\psi = 15^\circ$

$$^3J_{C\text{-}H} = 0.5 - 0.6\cos\psi + 5.7\cos^2\psi$$

$^3J_{C\text{-}H} = 5$ Hz ⟶ $\psi = 18^\circ$ for Gal-GlcNAc

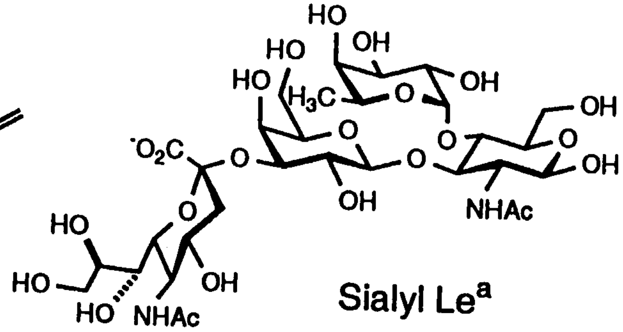
WongC/CHEM/dla5c/062894pm



Sialyl Le[x]

IC_{50} = 2.1 mM
1.0 μM (*in vivo*)

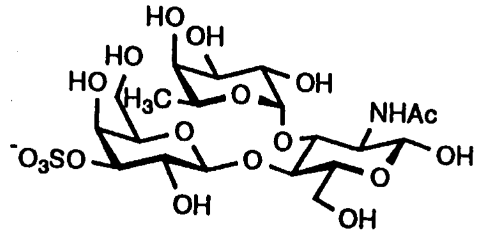
JACS **1992**, *114*, 9283.



Sialyl Le[a]

IC_{50} = 2.1 mM

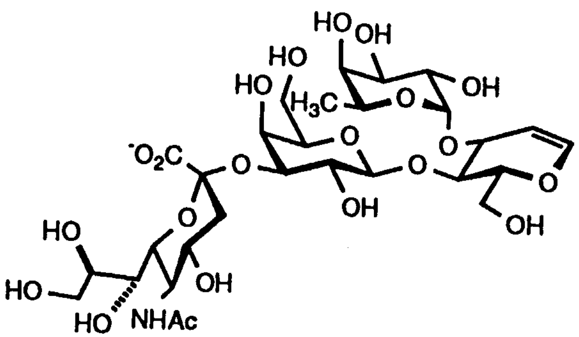
JACS **1992**, *114*, 9283.
PNAS **1991**, *88*, 10372.



Le[x] 3'-O-sulfate

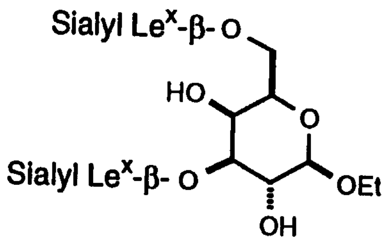
IC_{50} = 2.1 mM

Biochemistry **1992**, *31*, 9126



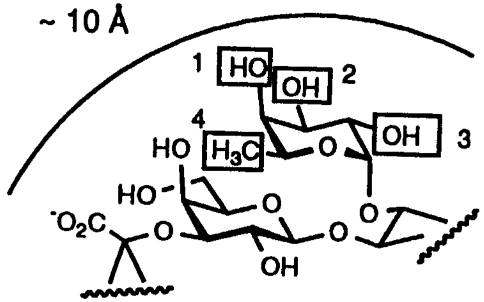
IC_{50} = 2.1 mM

JACS **1992**, *114*, 9283.



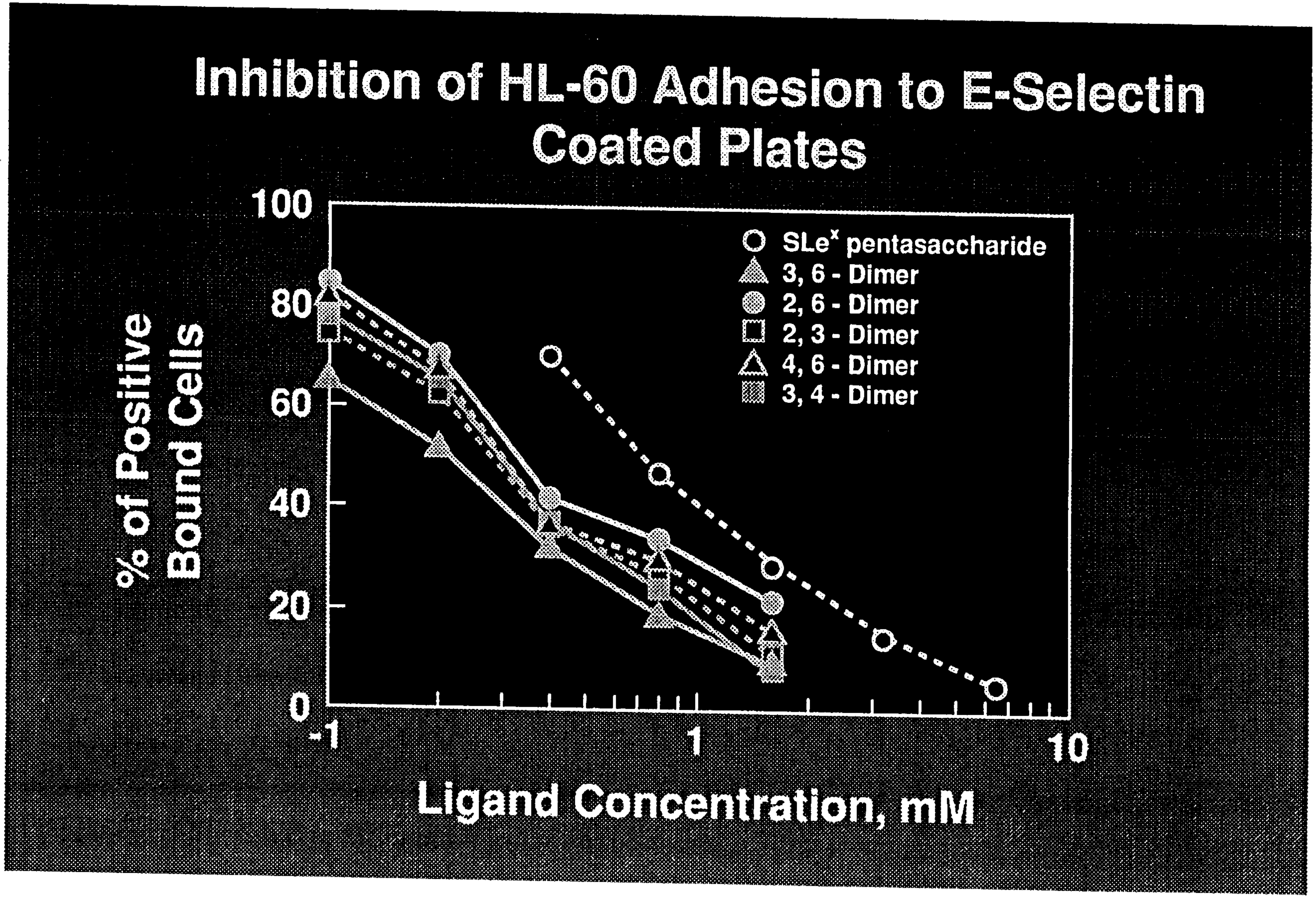
IC_{50} = 0.4 mM

JACS **1993**, *115*, 7549.



1-3: essential
4: nonessential

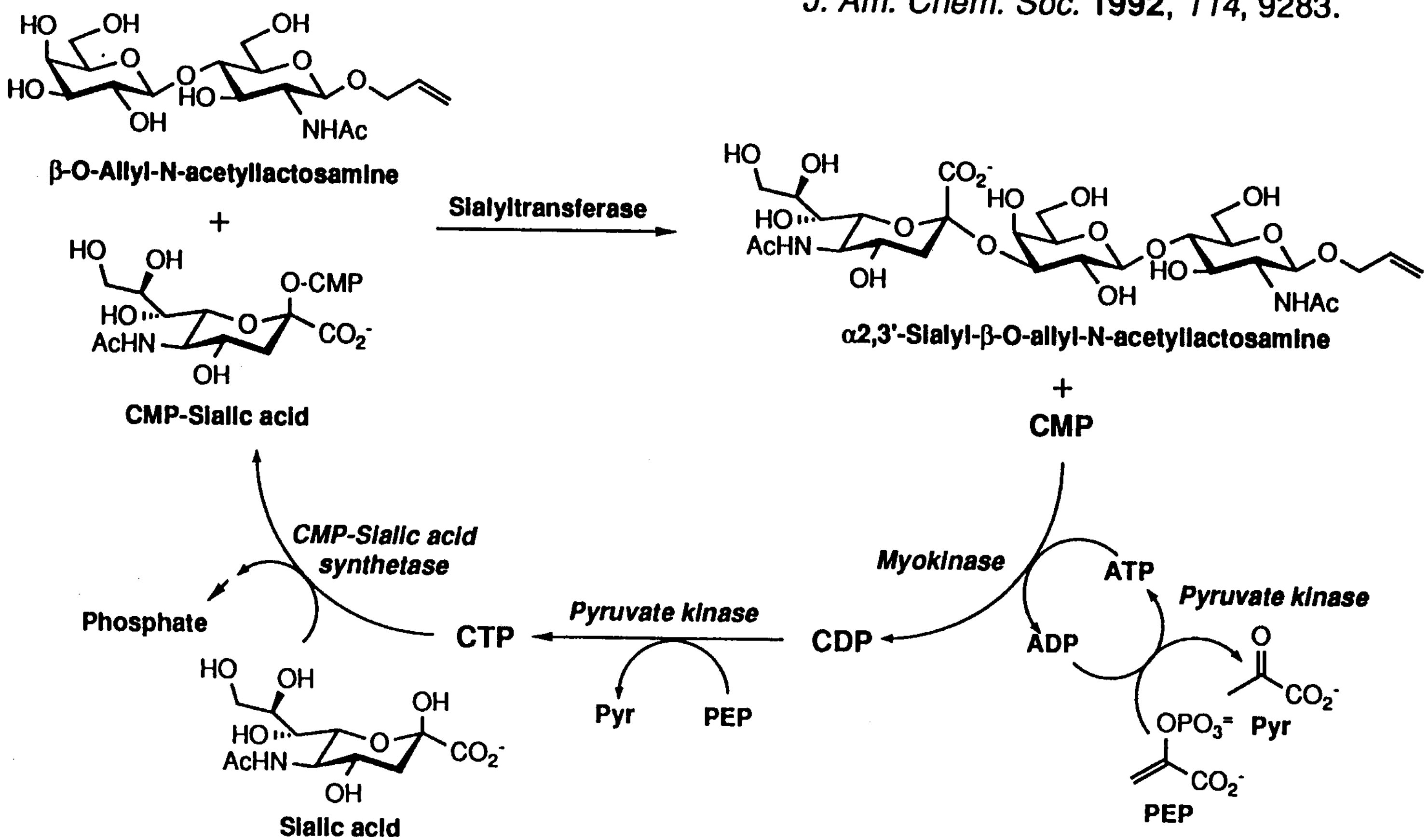
JACS **1992**, *114*, 9283.



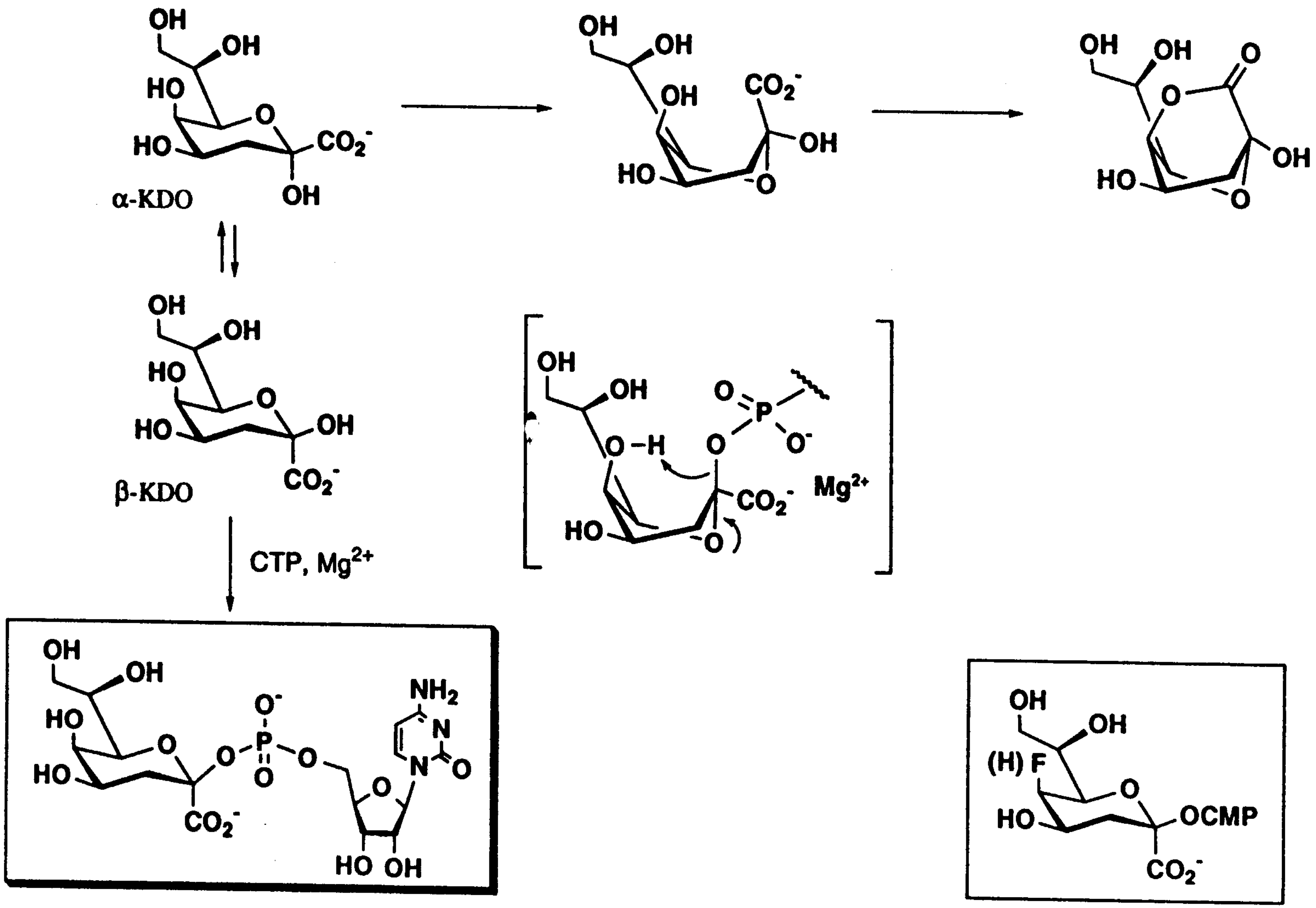
WongC/CHEM/gra1c/051294sa

Regeneration of Nucleotides Is a Key to Scale-Up: CMP-Sialic Acid

J. Am. Chem. Soc. **1992**, *114*, 9283.

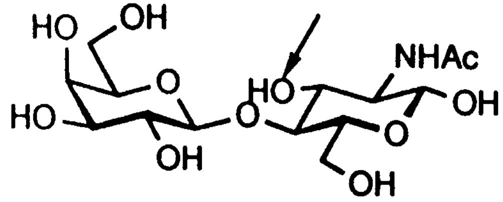


Stabilities of KDO and CMP-KDO

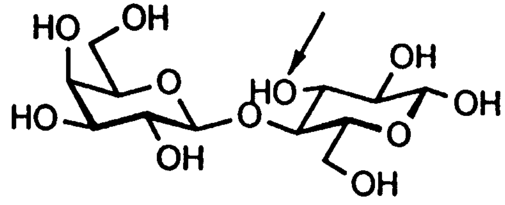


Unstable : $t_{1/2}$ = 20 min

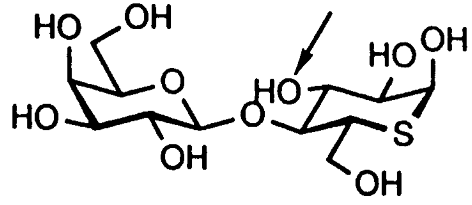
Stable : $t_{1/2}$ > 3 days



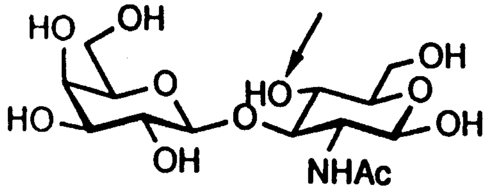
Galβ1,4GlcNAc
K_m= 35 mM, V= 100



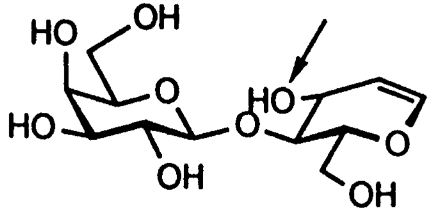
Galβ1,4Glc
K_m= 500 mM, V= 160



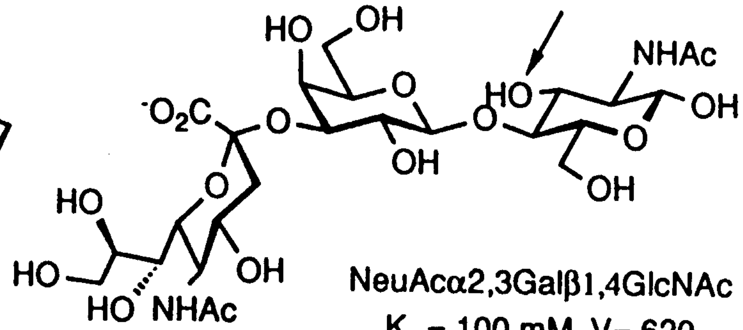
Galβ1,4(5-S-Glc)
K_m= 12 mM, V= 51



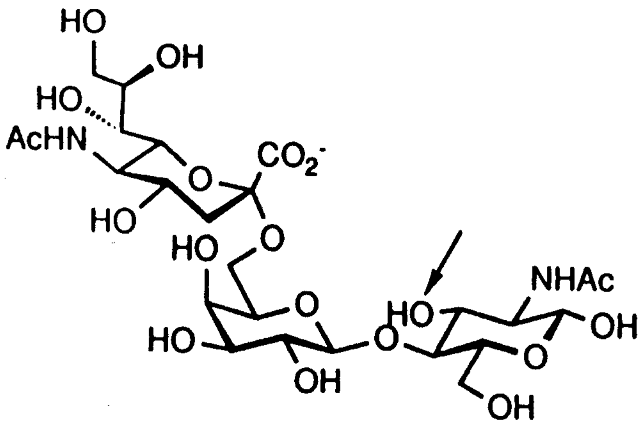
Galβ1,3GlcNAc
K_m= 600 mM, V= 130



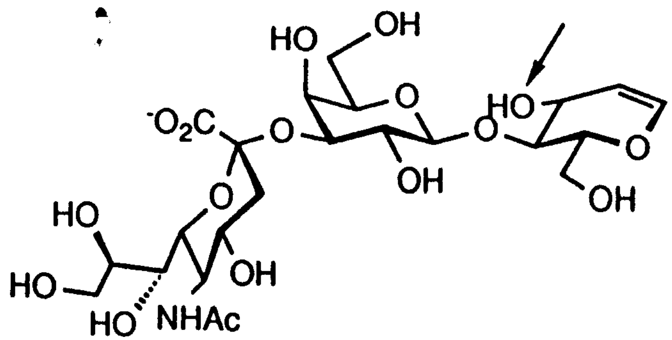
Galβ1,4Glucal
K_m= 34 mM, V= 10



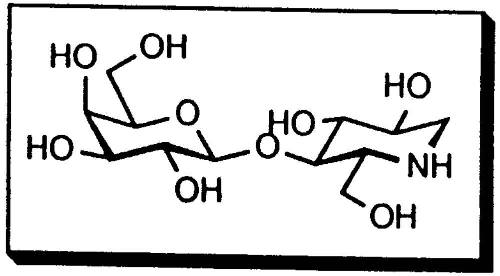
NeuAcα2,3Galβ1,4GlcNAc
K_m= 100 mM, V= 620



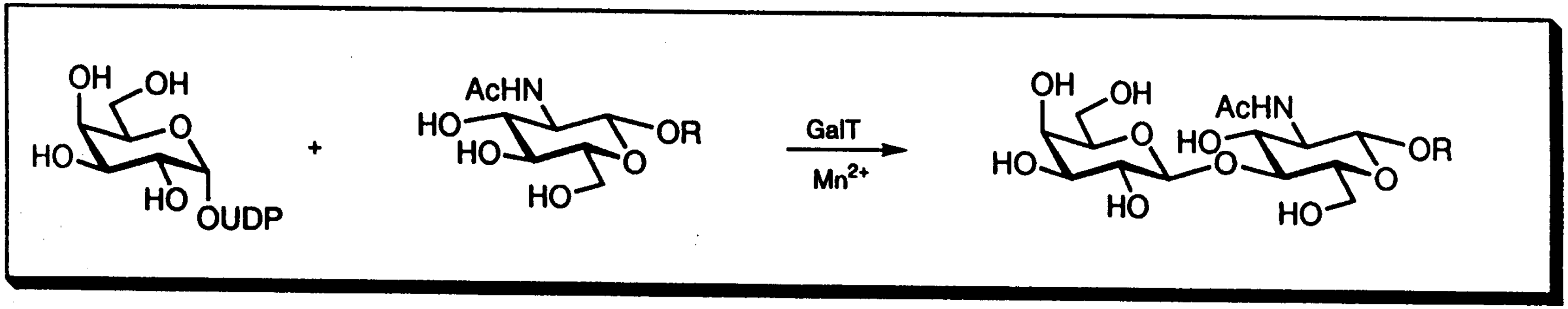
NeuAcα2,6Galβ1,4Glc
K_m= 70 mM, V= 13

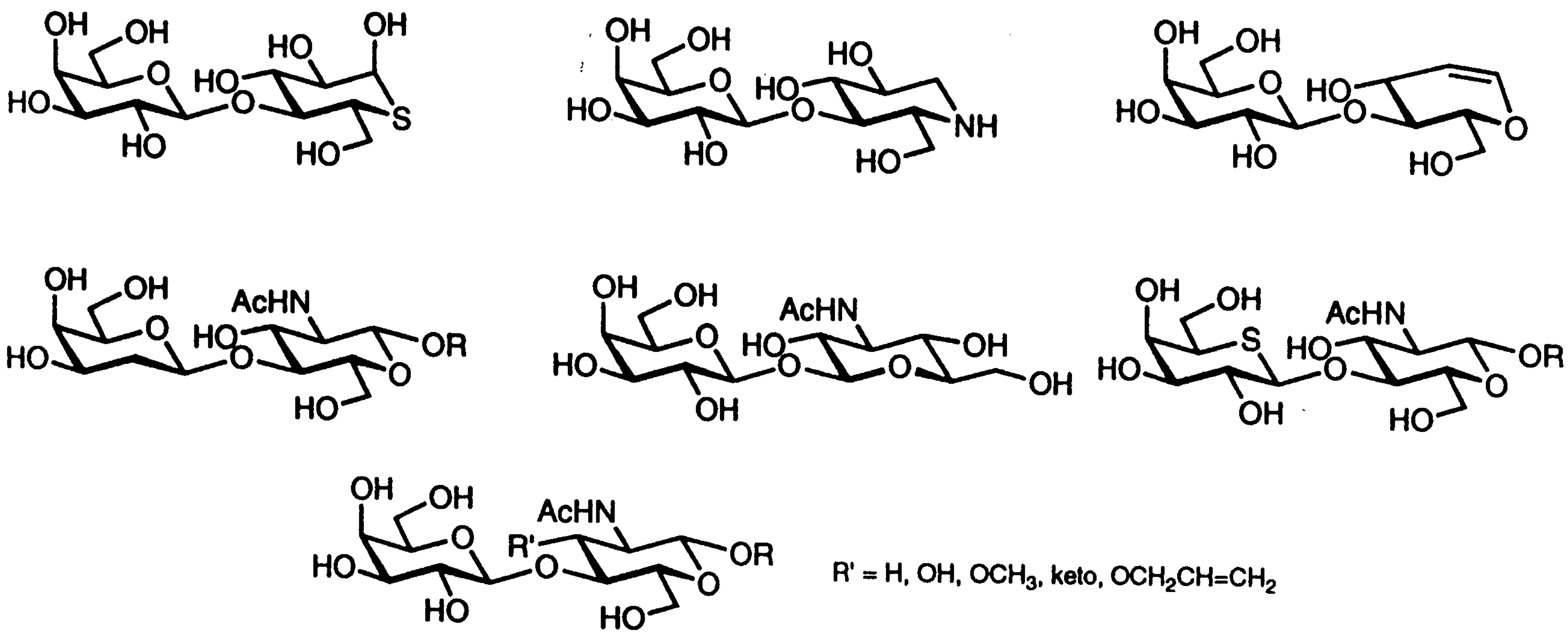


NeuAcα2,3Galβ1,4Glucal
K_m= 64 mM, V= 330



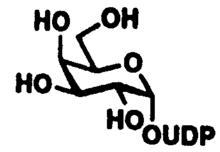
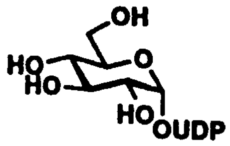
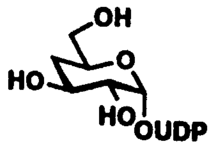
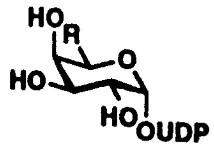
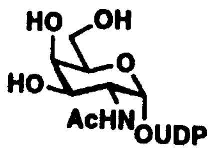
Galβ1,4deoxynojirimycin
IC_{50}= 8 mM

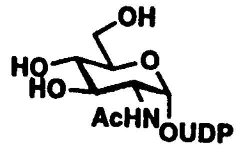
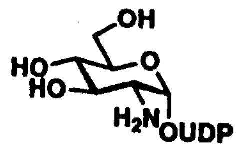
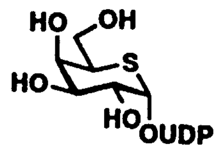
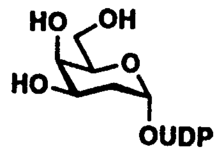




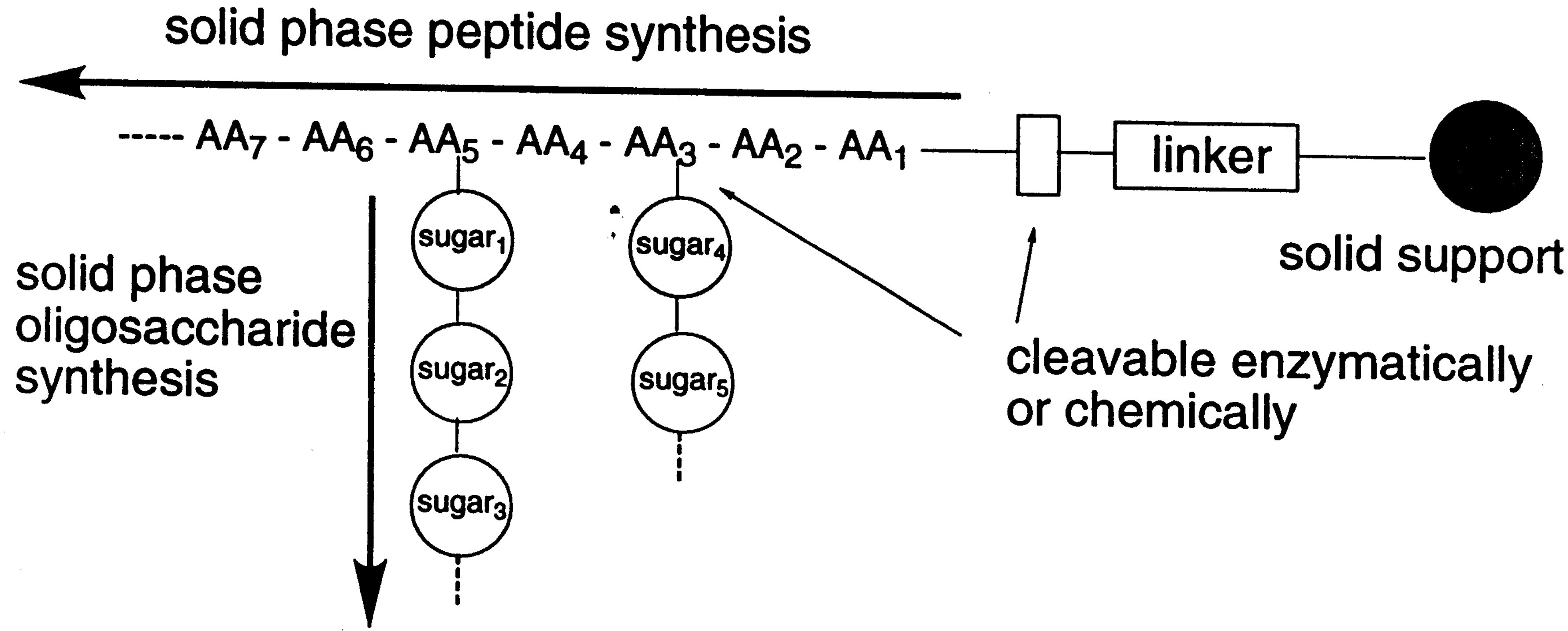
R' = H, OH, OCH_3, keto, $OCH_2CH=CH_2$

Donor Specificity of β-1,4-Galactosyltransferase

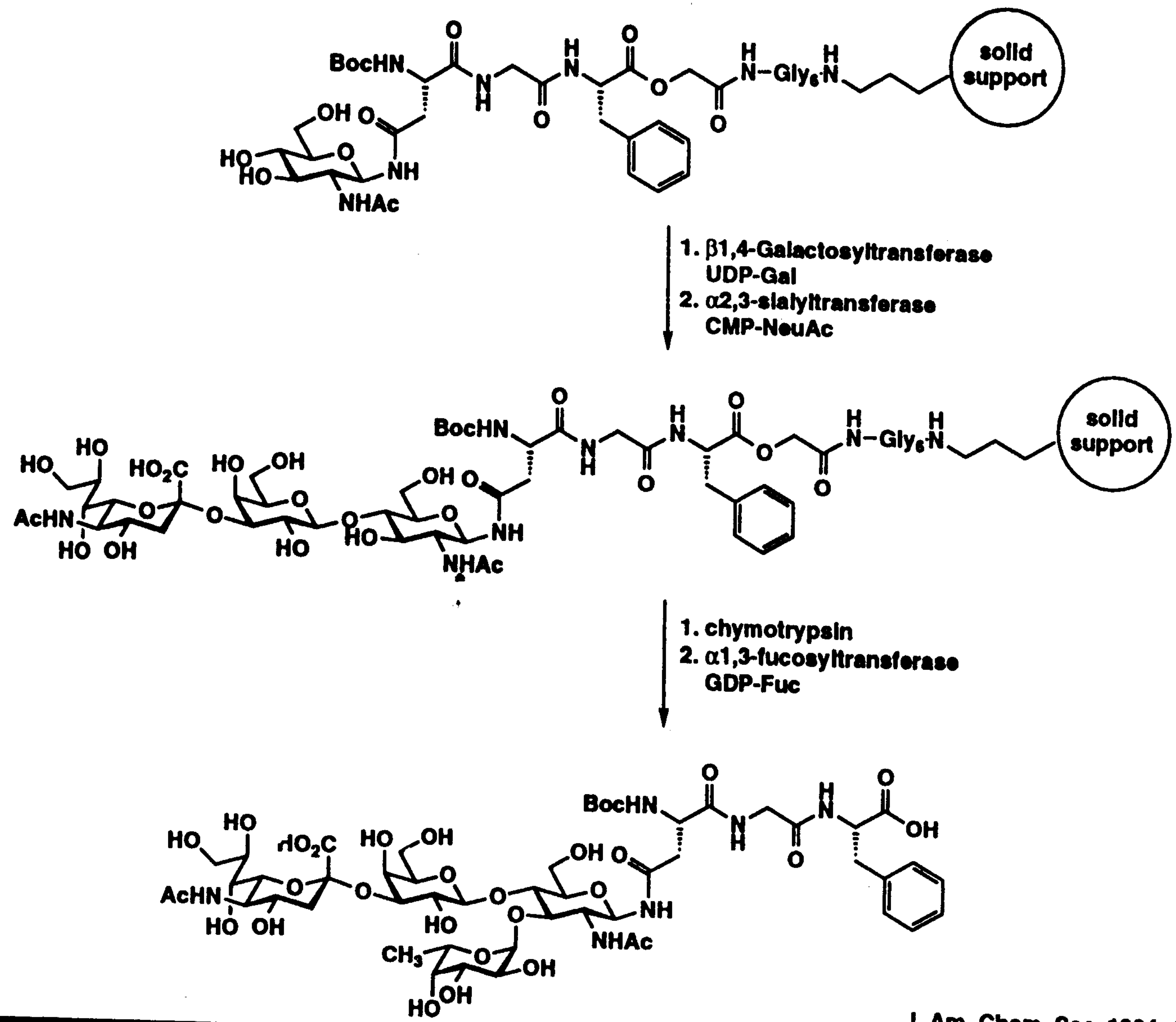
Donor	Rel. rate	Ref.
	100	Berliner & Robinson 1982
	0.3	Palcic & Hindsgaul 1991
	5.5	Berliner & Robinson 1982
	4.0 R = H 1.3 R = CH_3 0.2 R = CH_2F	Berliner & Robinson 1982
	4.0	Palcic & Hindsgaul 1991

Donor	Rel. rate	Ref.
	0.00	Palcic & Hindsgaul 1991
	0.09	Palcic & Hindsgaul 1991
	5.0	Yuasa 1992
	90	Srivastava & Hindsgaul 1993

Solid phase synthesis of glycopeptides and oligosaccharides

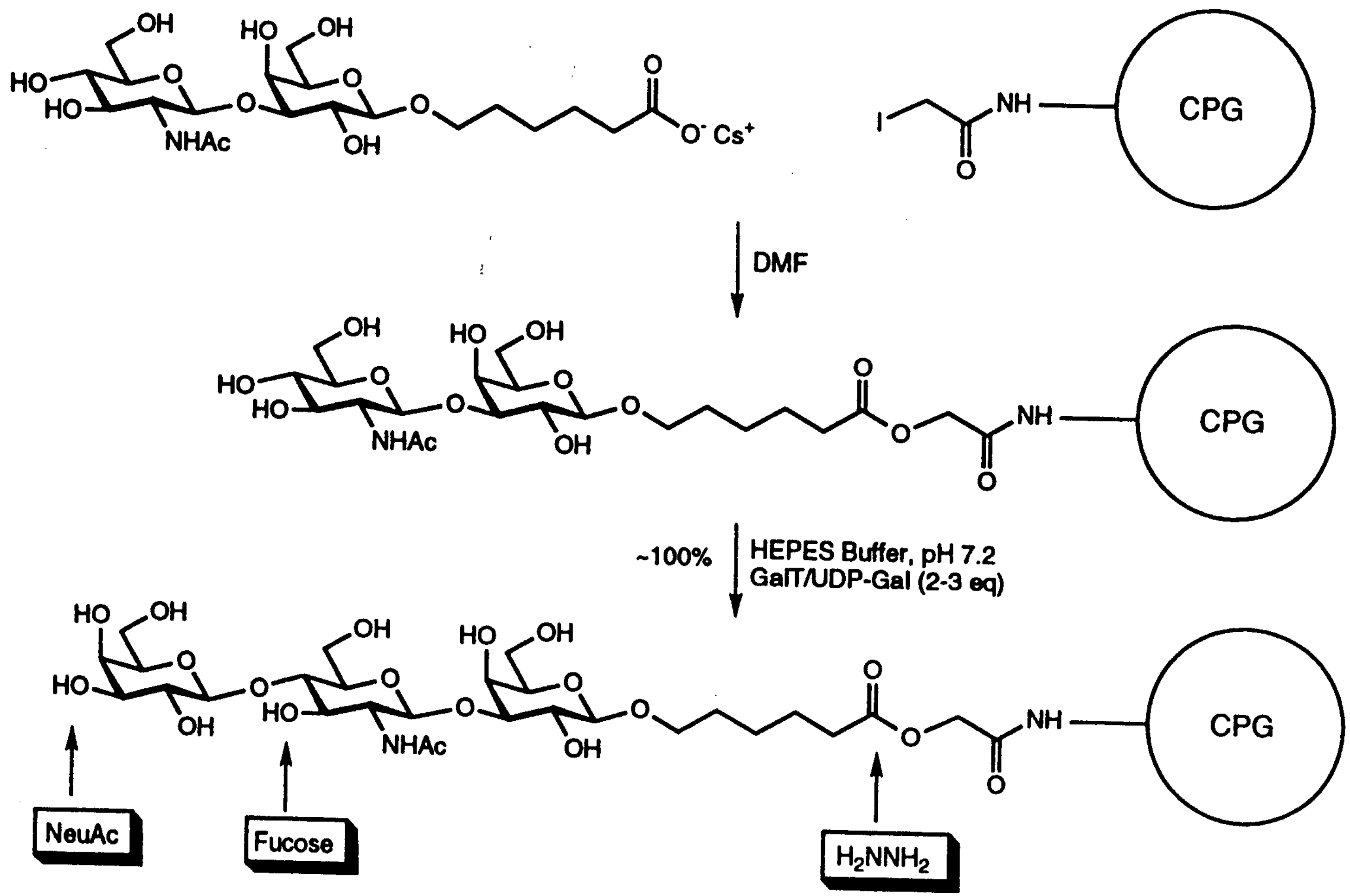


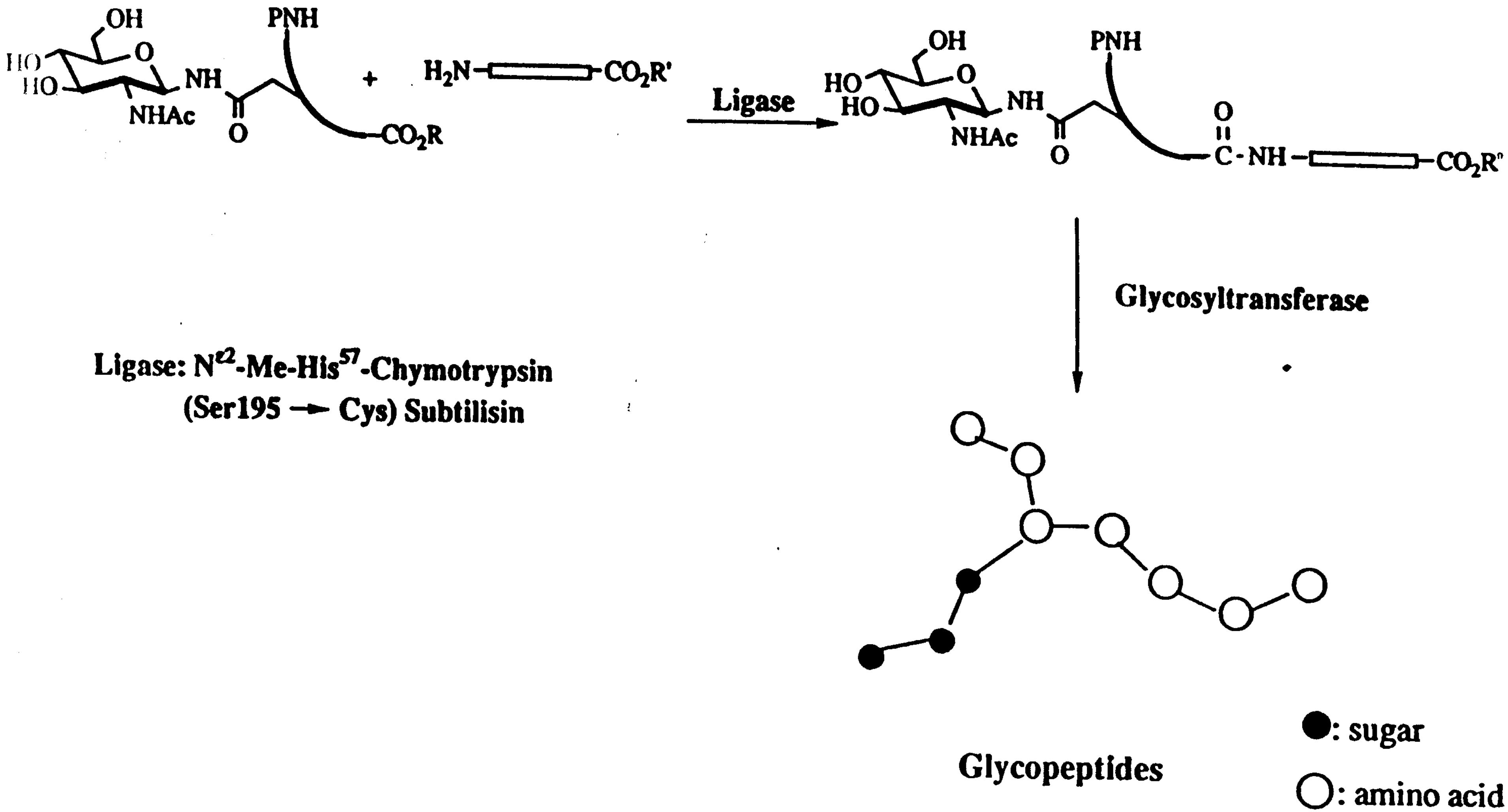
Solid-Phase Synthesis of Glycopeptides



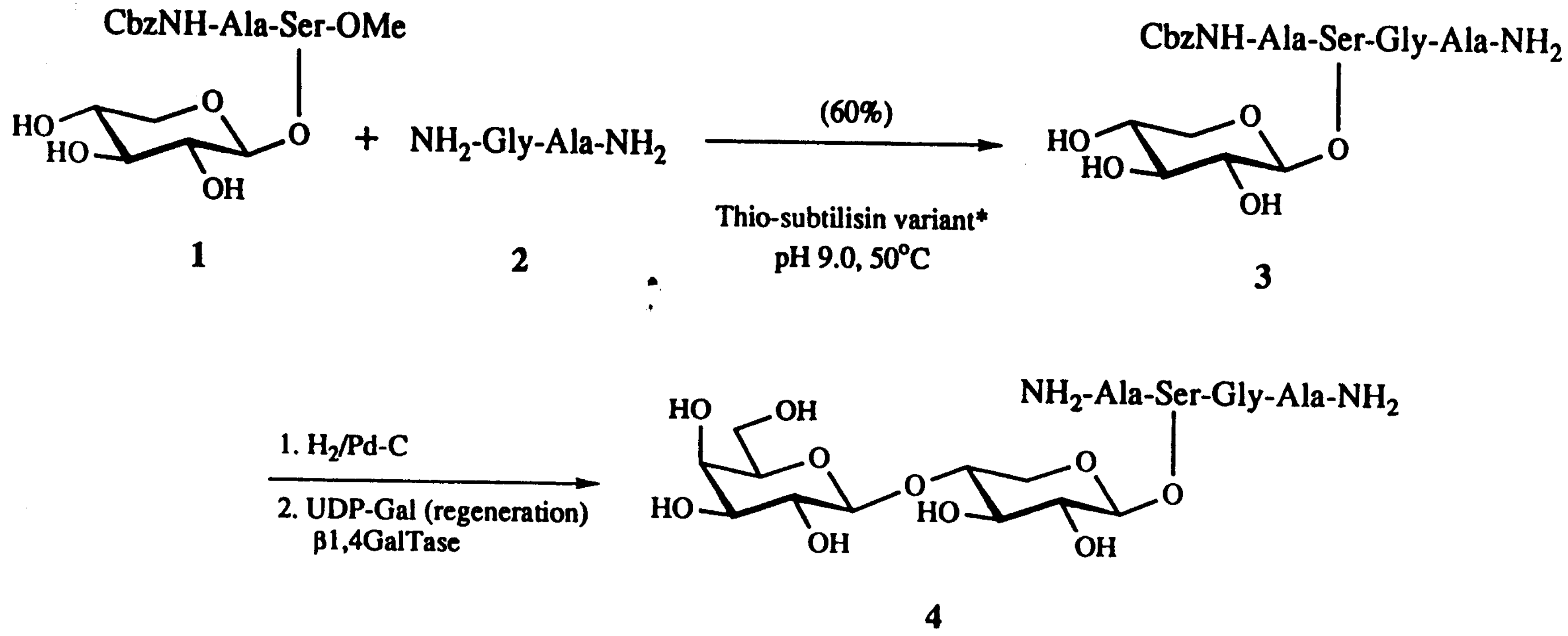
J. Am. Chem. Soc. 1994, 116, 1135

Solid-Phase Synthesis of Oligosaccharide

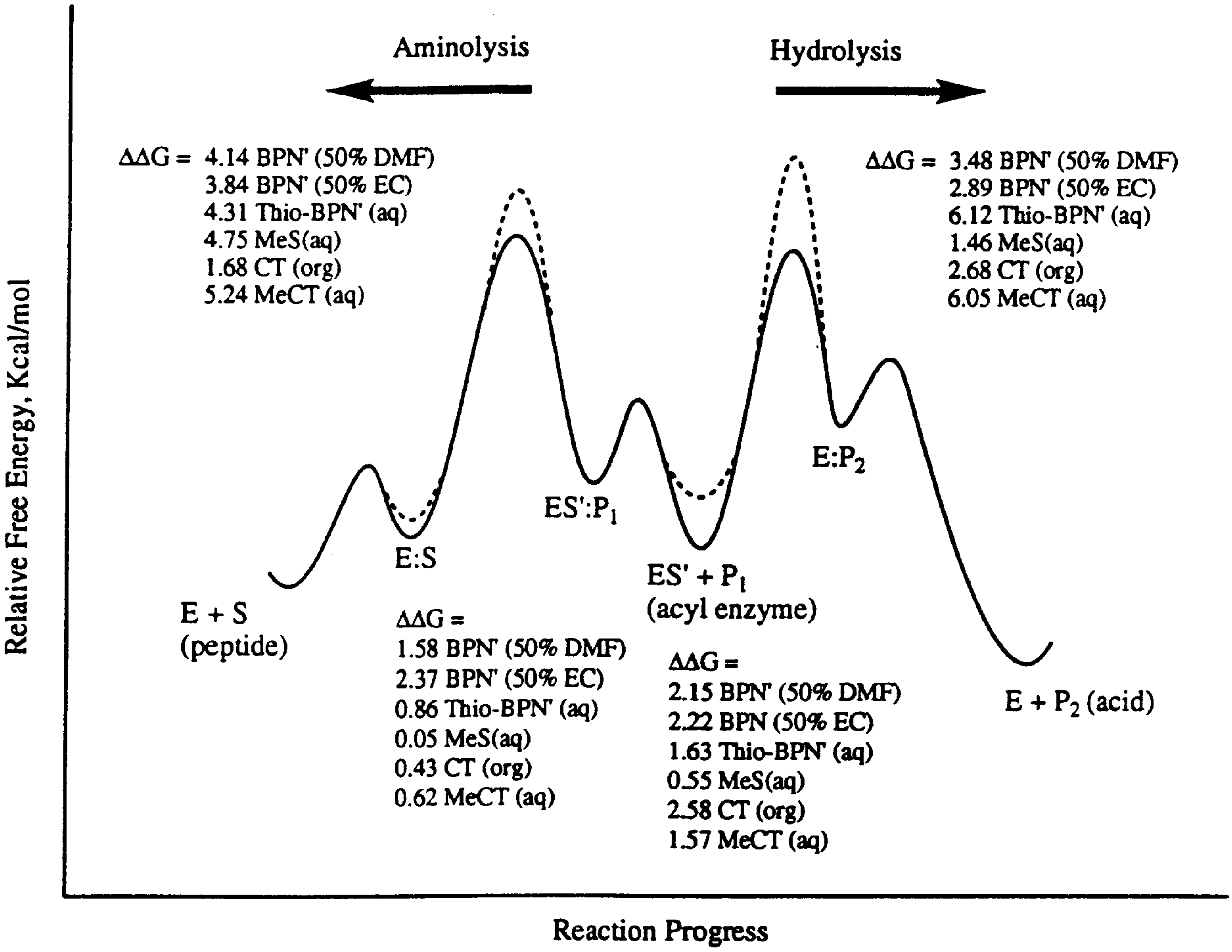



OH
PNH
HO
HO
NH
NHAc
O
CO2R
+
H2N
CO2R'
Ligase
OH
PNH
HO
HO
NH
NHAc
O
O
C-NH
CO2R'
Glycosyltransferase
Ligase: Nε2-Me-His57-Chymotrypsin
(Ser195 → Cys) Subtilisin
Glycopeptides
: sugar
: amino acid

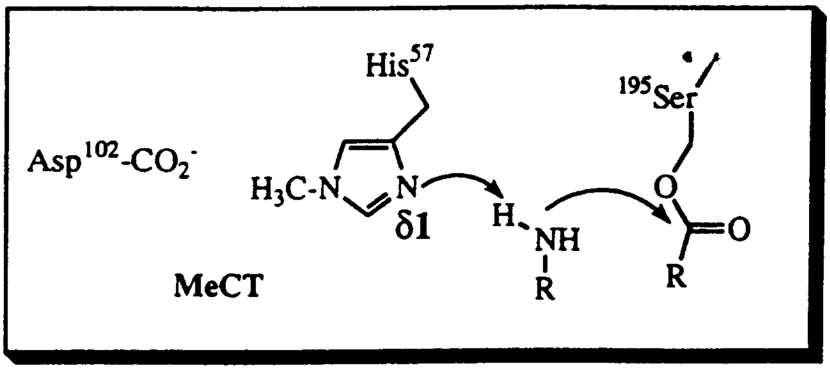
Enzymatic Synthesis of a Glycopeptide.



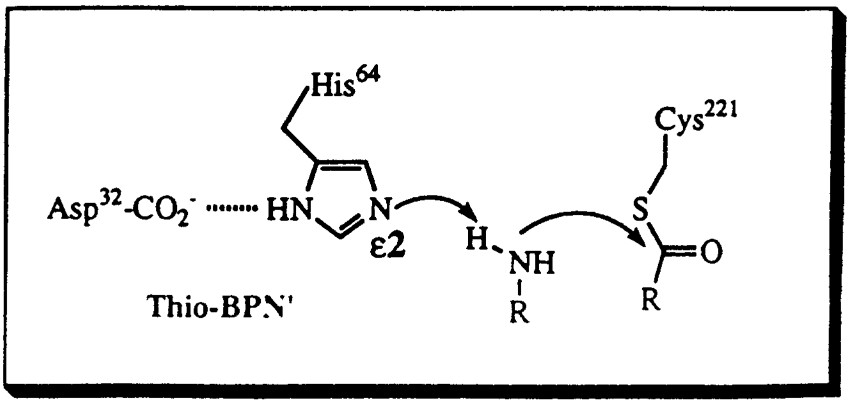
*Thio-subtilisin variant contains the following changes:
Met50Phe, Asn76Asp, Gly169Ala, Asn218Ser, and Ser221Cys.



Aminolysis
Hydrolysis
ΔΔG = 4.14 BPN' (50% DMF)
3.84 BPN' (50% EC)
4.31 Thio-BPN' (aq)
4.75 MeS(aq)
1.68 CT (org)
5.24 MeCT (aq)
ΔΔG = 3.48 BPN' (50% DMF)
2.89 BPN' (50% EC)
6.12 Thio-BPN' (aq)
1.46 MeS(aq)
2.68 CT (org)
6.05 MeCT (aq)
Relative Free Energy, Kcal/mol
E:P2
ES':P1
E:S
ES' + P1
(acyl enzyme)
E + S
(peptide)
ΔΔG =
1.58 BPN' (50% DMF)
2.37 BPN' (50% EC)
0.86 Thio-BPN' (aq)
0.05 MeS(aq)
0.43 CT (org)
0.62 MeCT (aq)
ΔΔG =
2.15 BPN' (50% DMF)
2.22 BPN (50% EC)
1.63 Thio-BPN' (aq)
0.55 MeS(aq)
2.58 CT (org)
1.57 MeCT (aq)
E + P2 (acid)
Reaction Progress

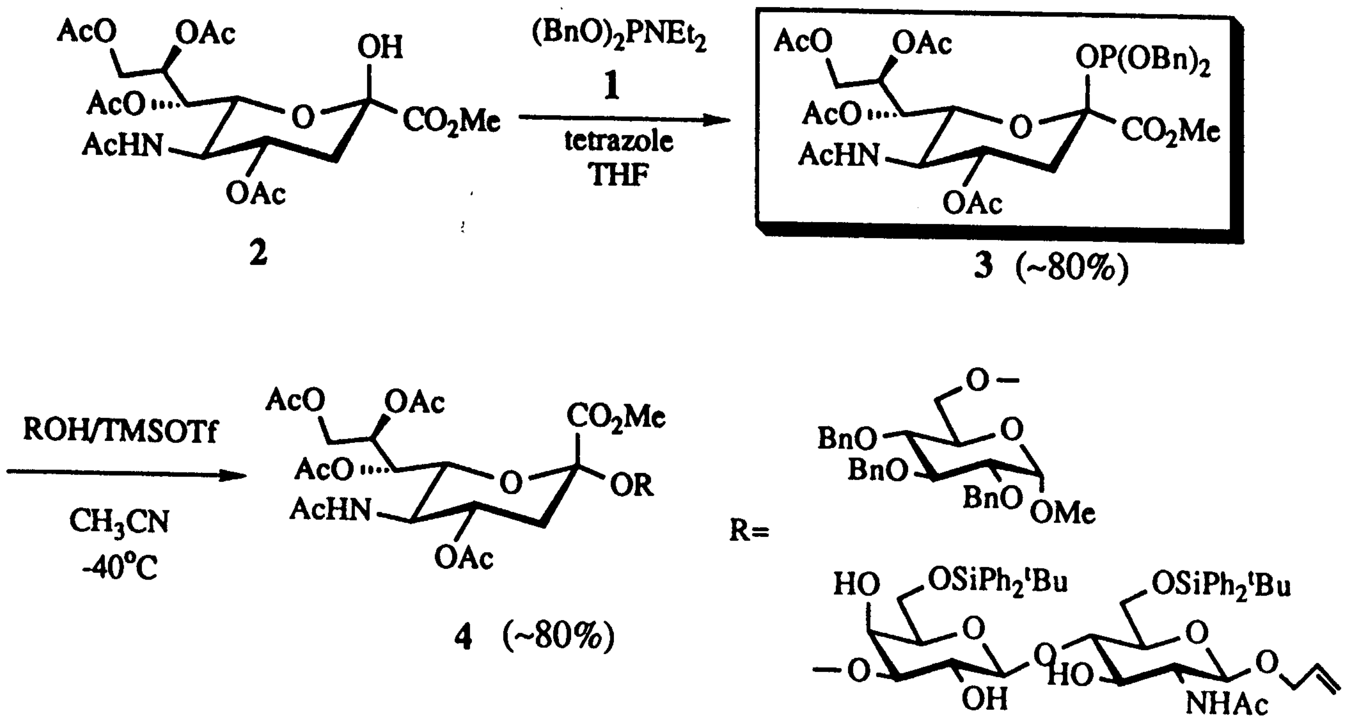


His57
195Ser
Asp102-CO2-
H3C-N
N
δ1
O
H
NH
R
O
R
MeCT



His64
Cys221
Asp32-CO2- .... HN
N
ε2
S
H
NH
R
O
R
Thio-BPN'

Chemical Sialylation Employing Sialyl Phosphite.

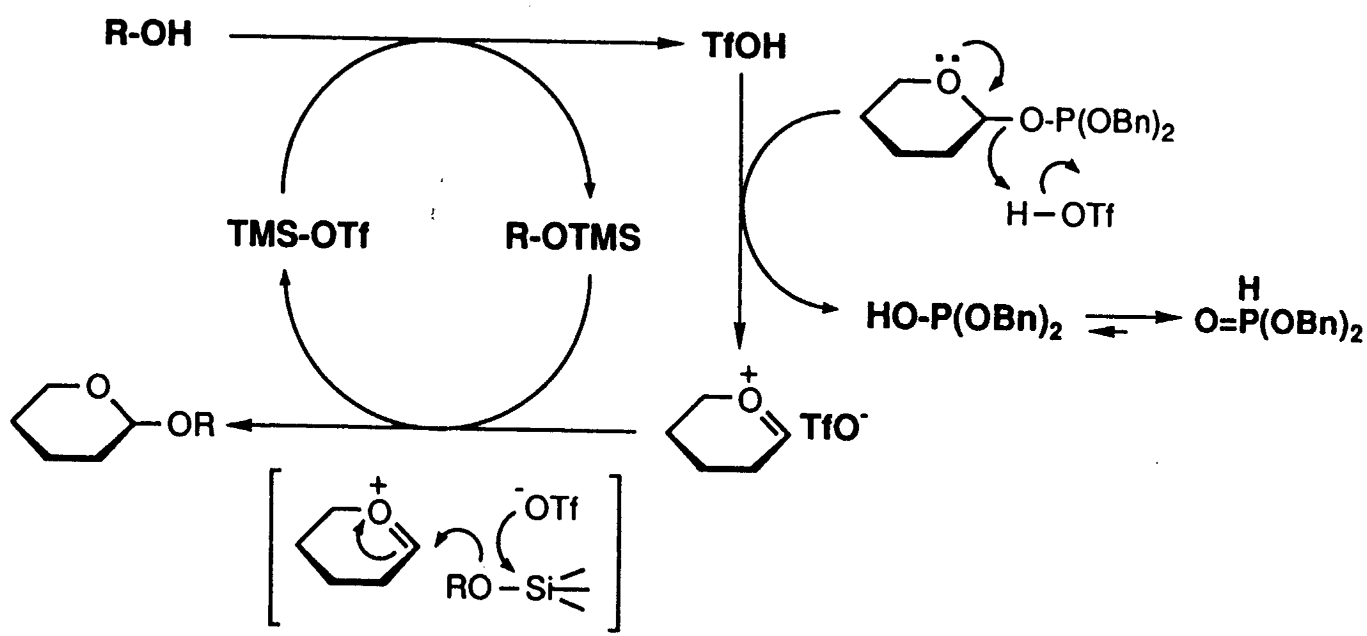


2

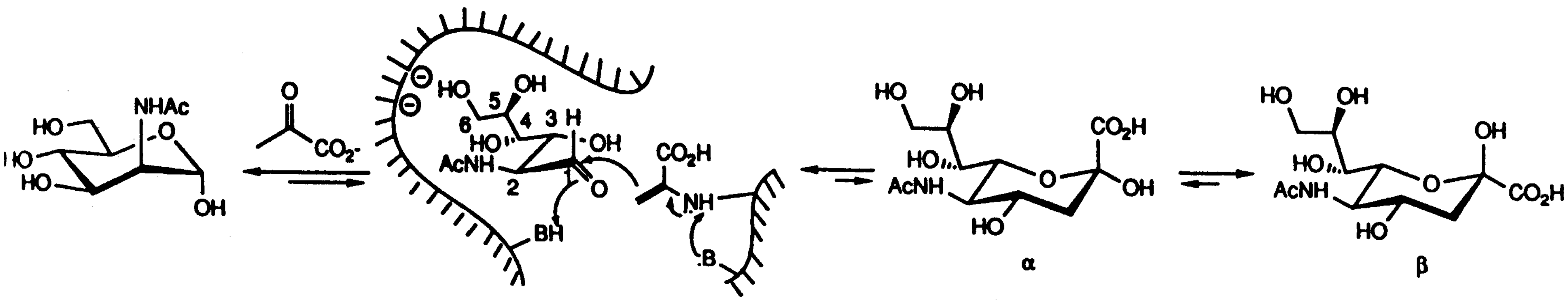
3 (~80%)

4 (~80%)

Proposed Mechanism of Glycosylphosphite-Mediated Glycosylation
TMSOTf Reacts with Acceptor First



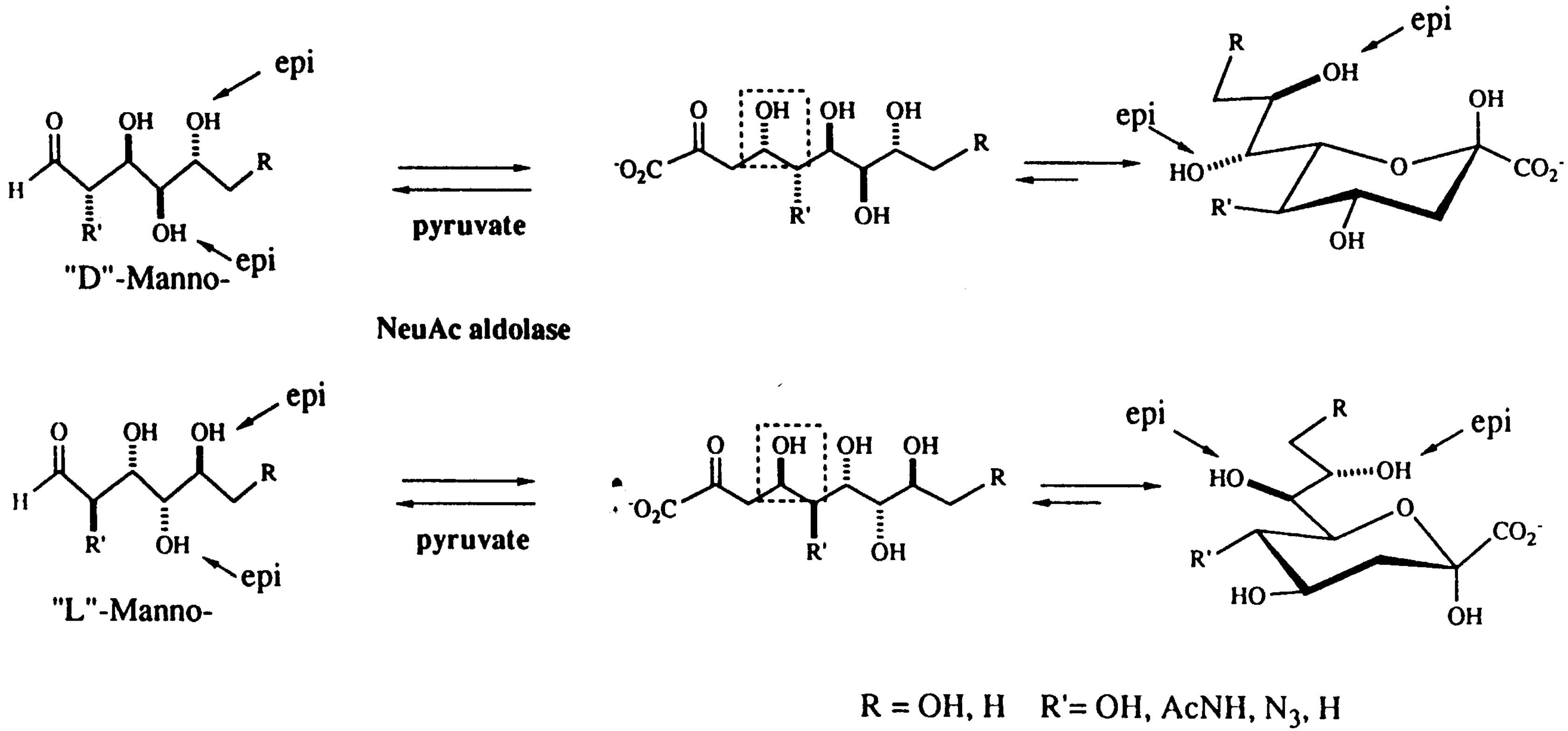
Sialic Acid Aldolase : Substrate Specificity



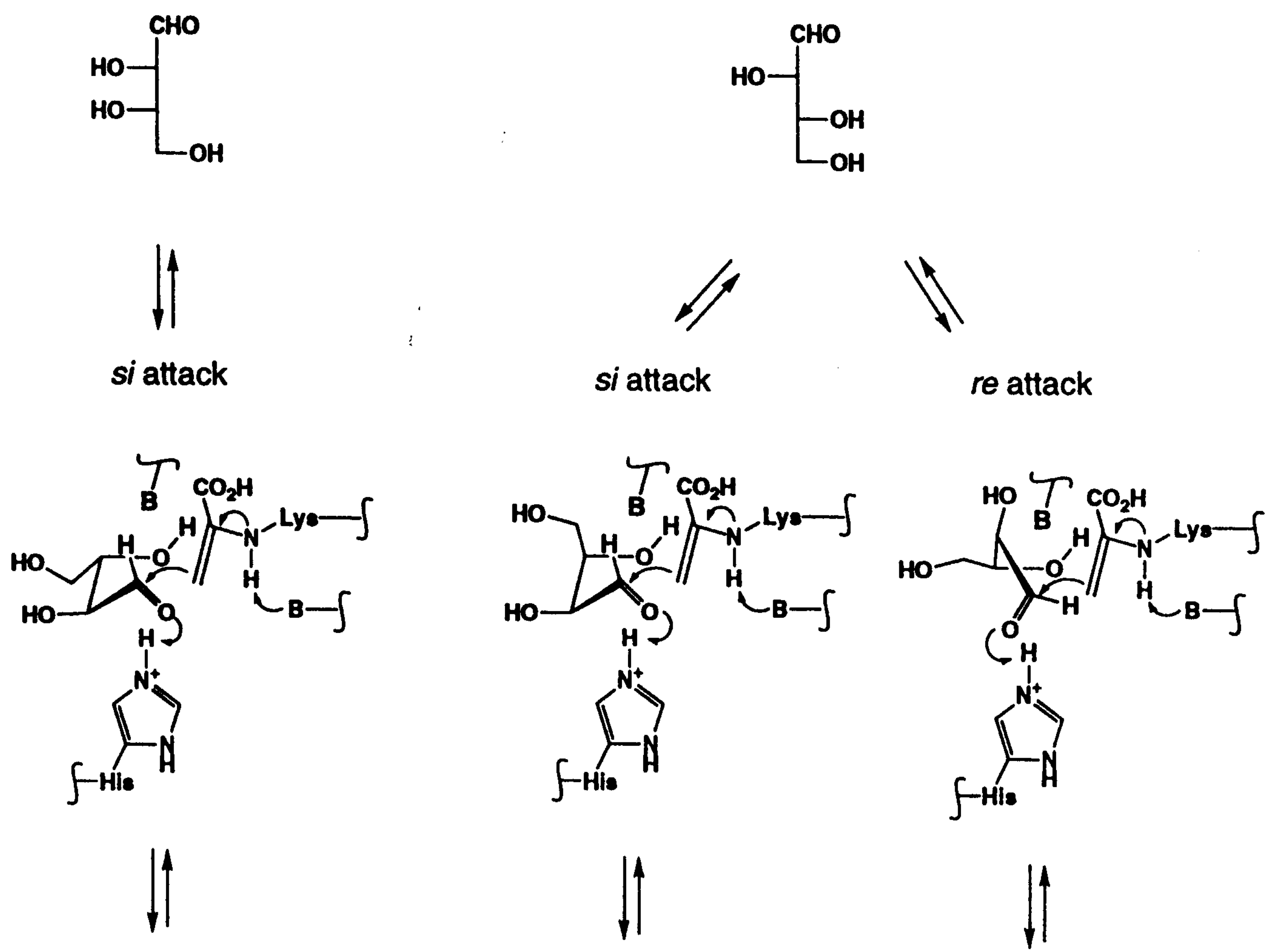
Acceptor Position	2	3	4	5	6
Very good	Ph		CH_3O	AcO	F
	HO		epi-F (6-F)		AcO
	H		AcO		$CH_3CH(OH)CO_2$
	$HOCH_2CONH$		epi-OH (2-OH)		CH_3O
	N_3		H (2-OH)		N_3
	epi-F	F (2 epi-OH)	F (2 epi-OH)		$(CH_3)_2P(O)O$-
Weak	epi-OH	epi-OH	epi-OH (2 epi-OH)	CH_3O	deleted to C-5
No	epi-NHAc				$OPO_3^{=}$

S David, *Tetrahedron*, **46**, 201 (1990)

Kim and Wong, *JACS*, **110**, 6481 (1988)
Liu, Ichikawa, Wong, *JACS*, **1992**

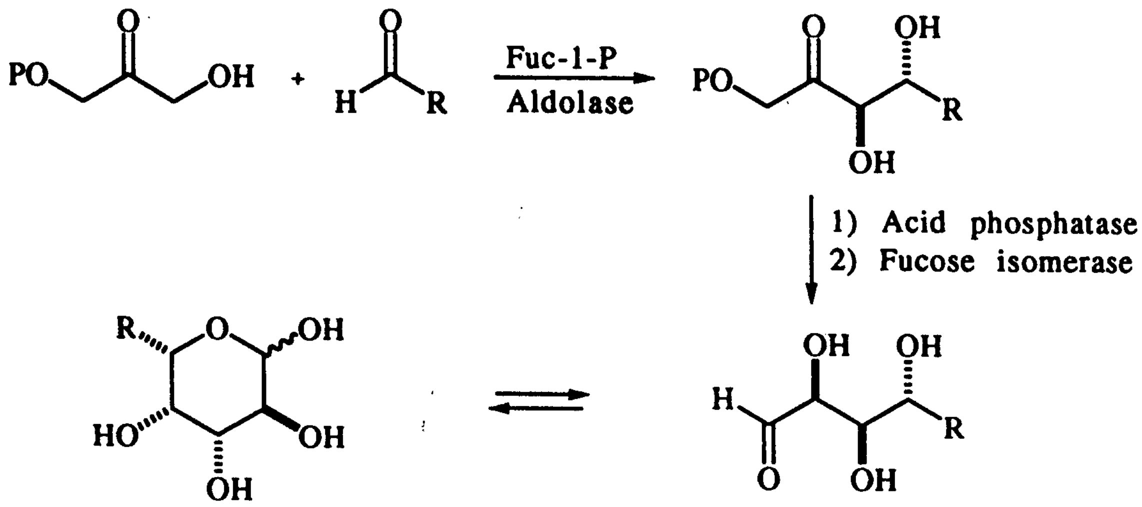


epi
"D"-Manno-
pyruvate
NeuAc aldolase
"L"-Manno-
pyruvate
R = OH, H R'= OH, AcNH, N3, H

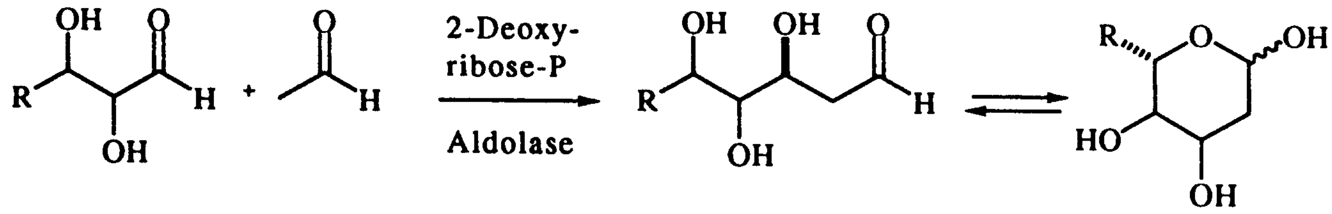


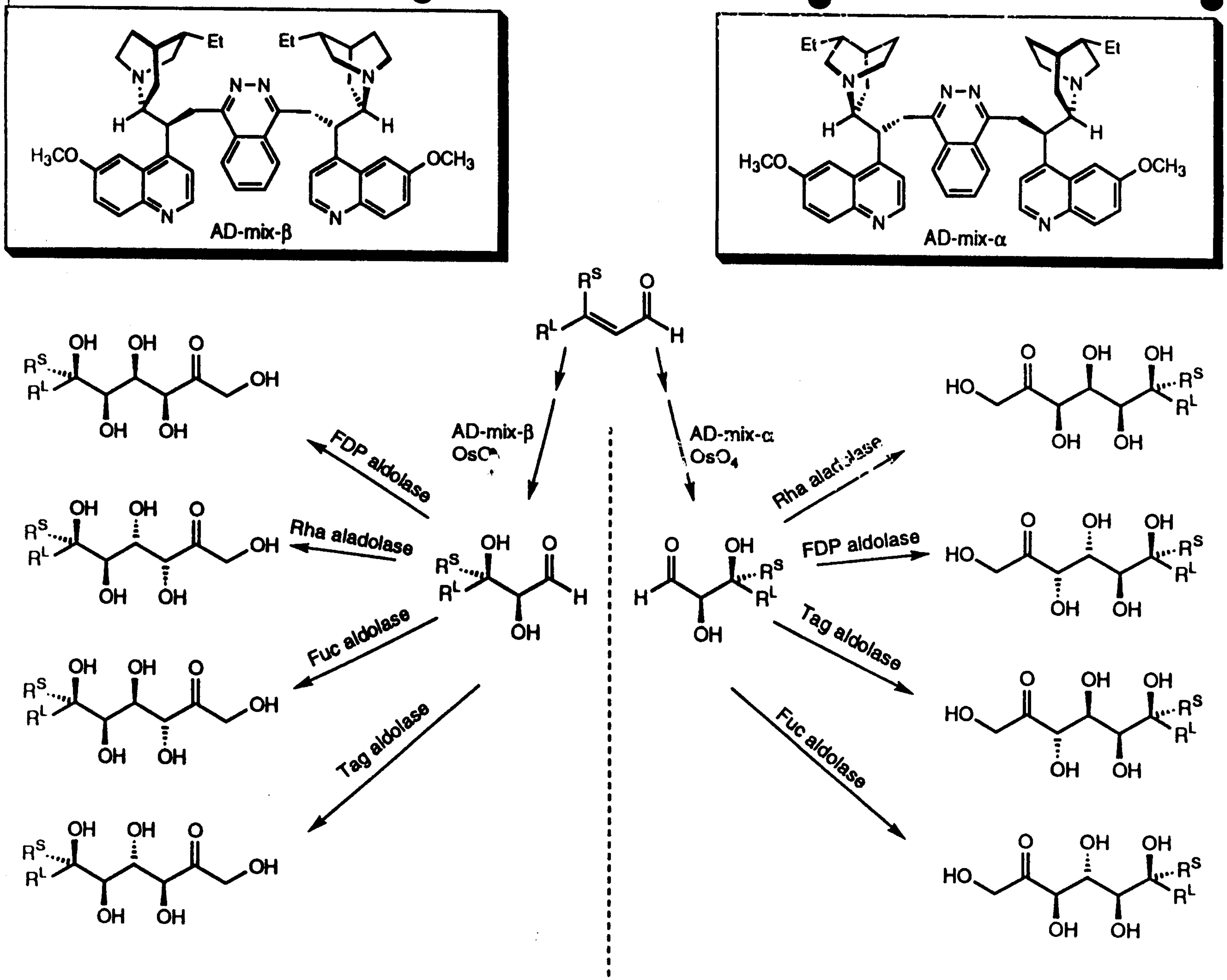
CHO
HO
HO
OH
CHO
HO
OH
OH
si attack
si attack
re attack
B
CO2H
Lys
His

Synthesis of Fucose Analogs



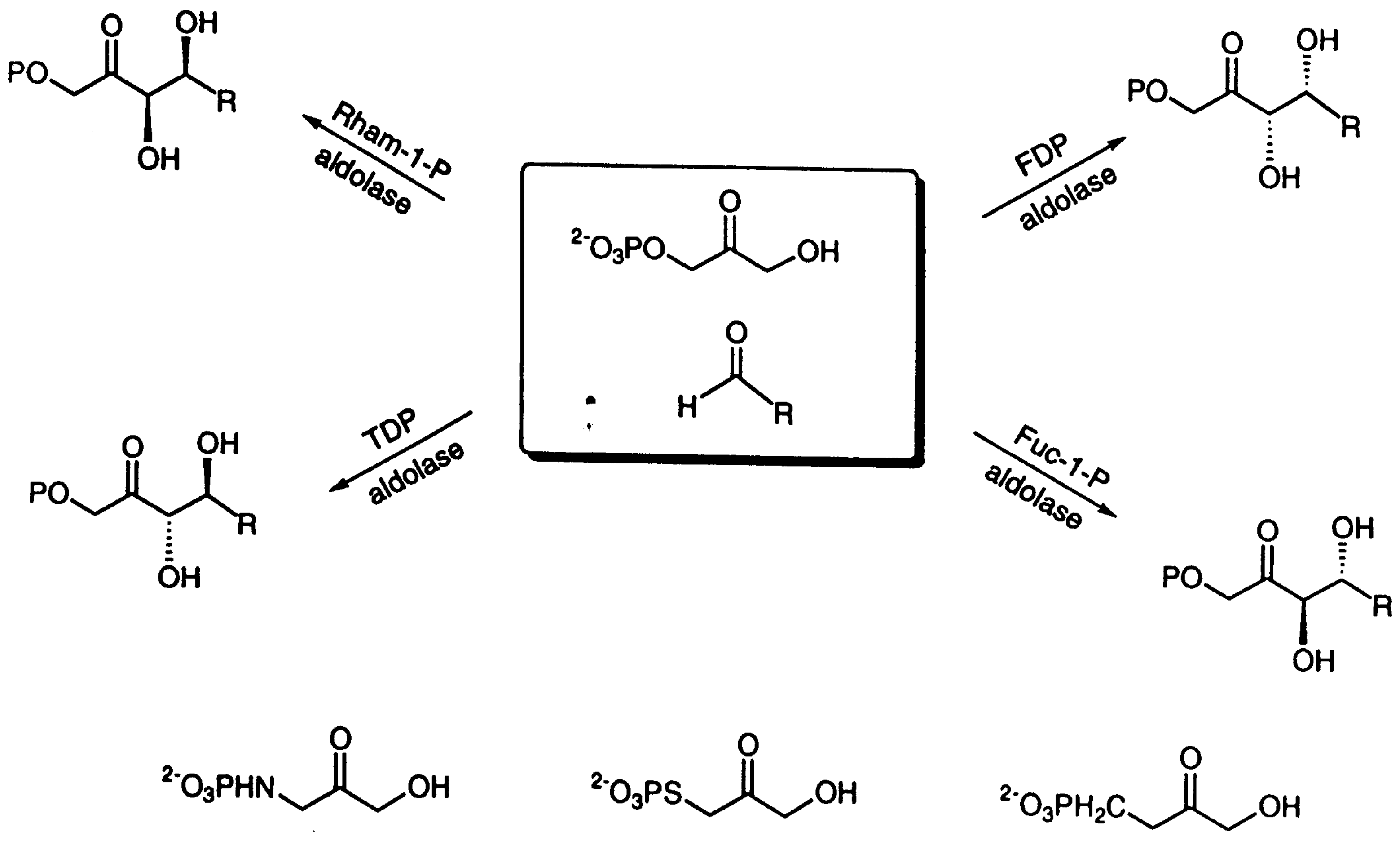
R = CH_3, CH_2CH_3, CH_2F, CH_2Ph, Ph

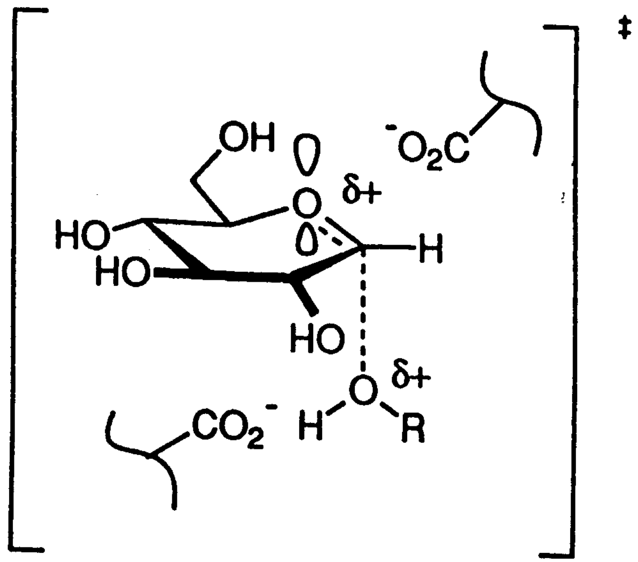




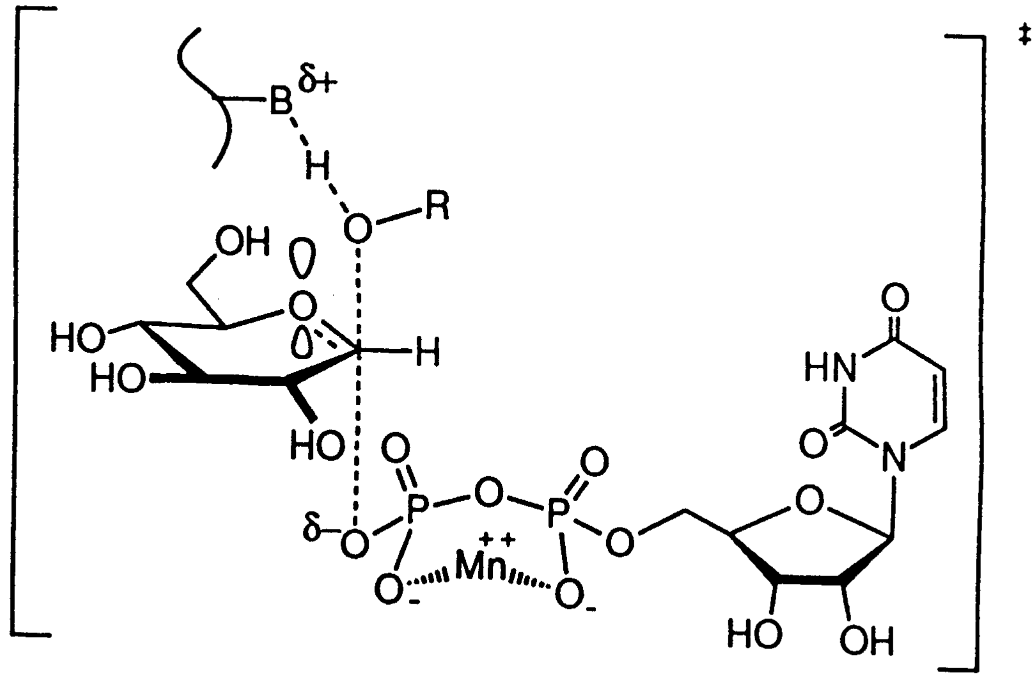
AD-mix-β
AD-mix-α
AD-mix-β
AD-mix-α
OsO_4
FDP aldolase
Rha aladolase
Fuc aldolase
Tag aldolase
Rha aldolase
FDP aldolase
Tag aldolase
Fuc aldolase

Dihydroxyacetone phosphate-dependent Aldolases

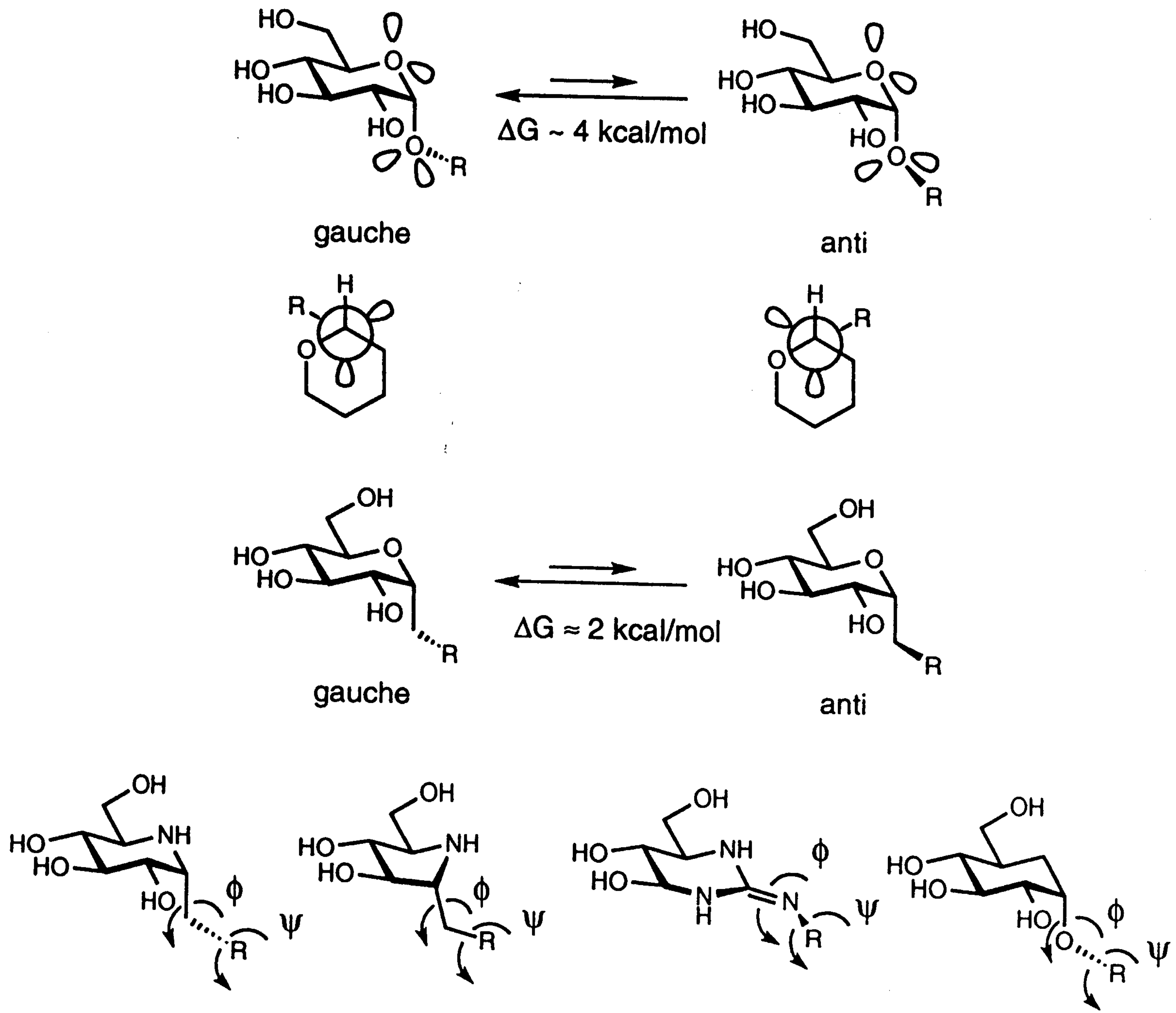




Glycosidase Reaction

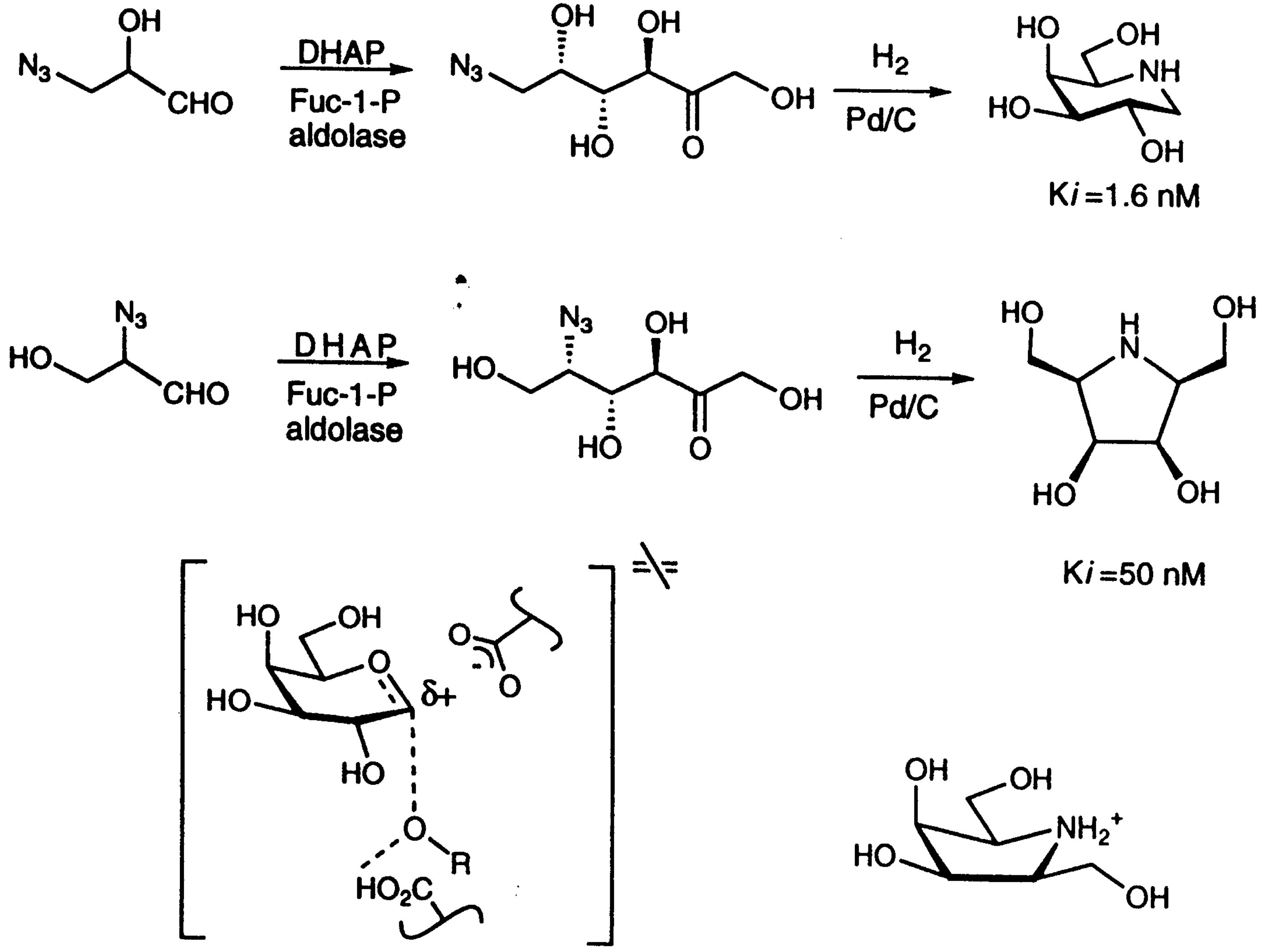


Glycosyltransferase Reaction

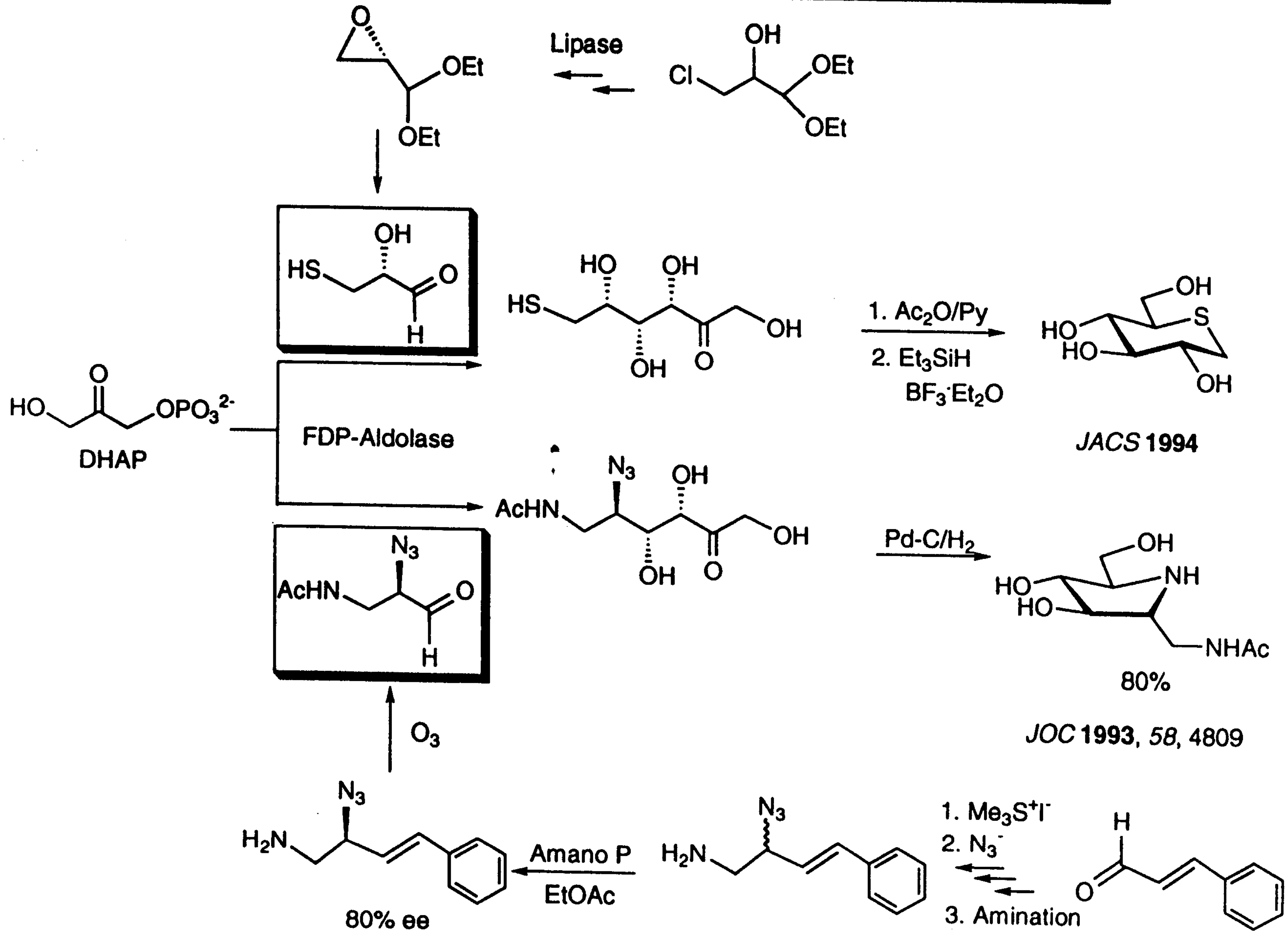


ΔG ~ 4 kcal/mol
gauche
anti
ΔG ≈ 2 kcal/mol
gauche
anti

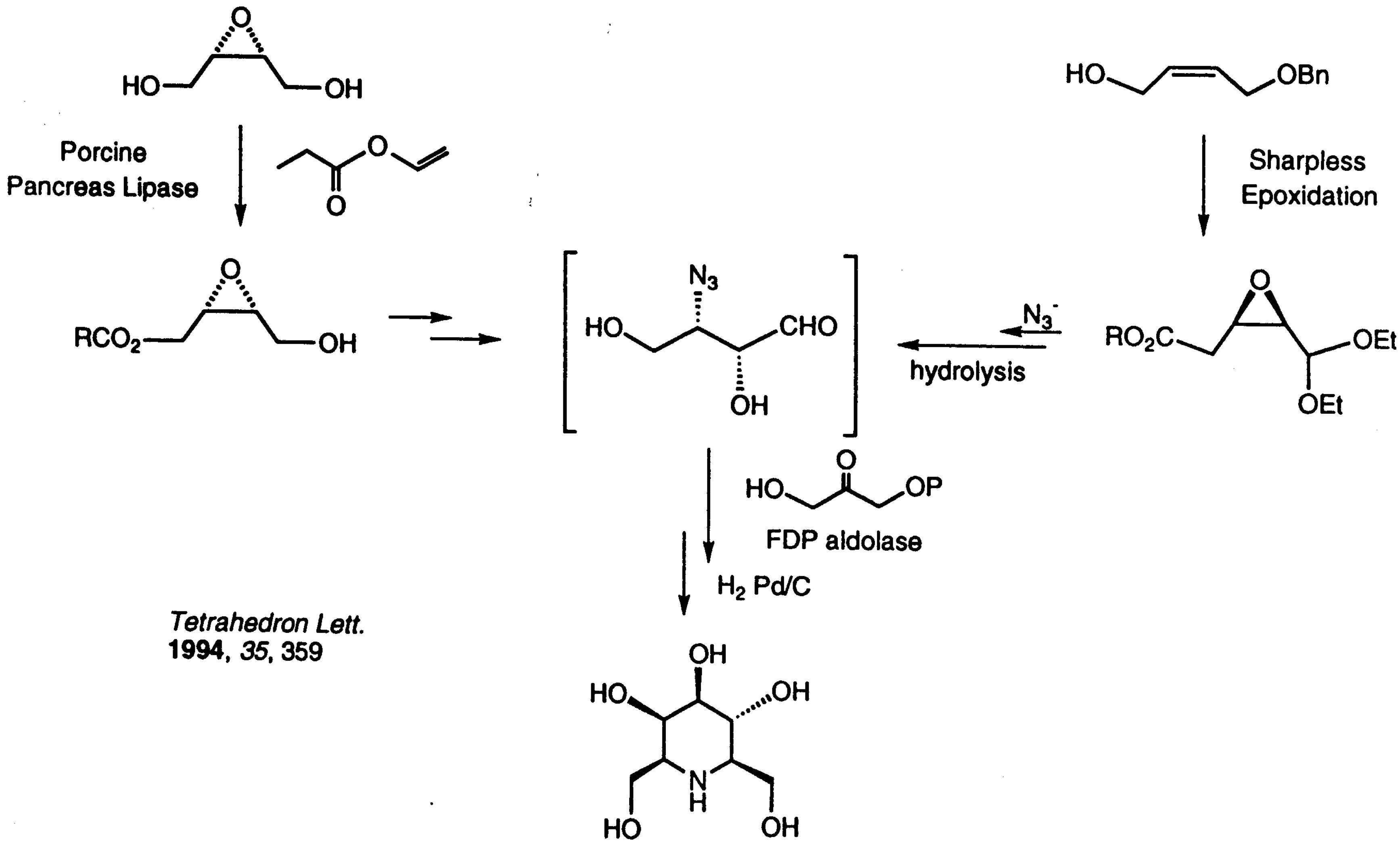
Inhibitors of α-Galactosidase



Aminoazasugar and Thiosugar from FDP-Aldolase

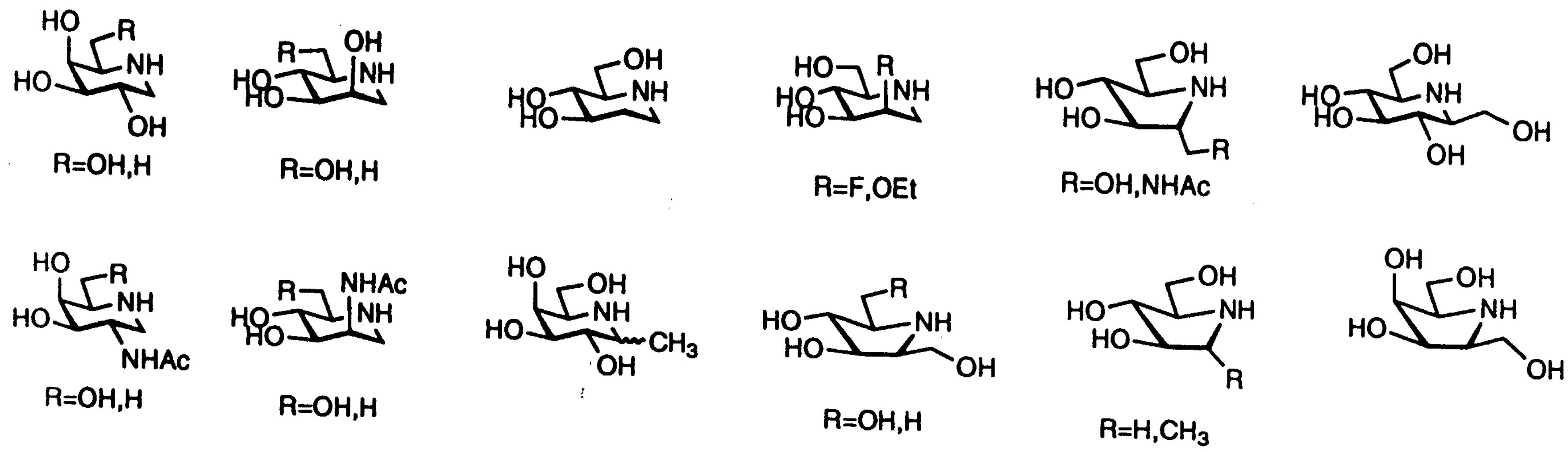


Synthesis of Homoazasugar

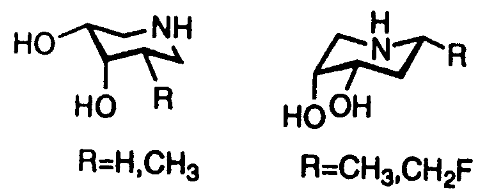


Tetrahedron Lett.
1994, *35*, 359

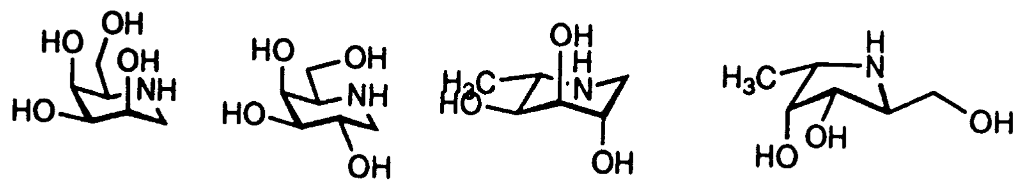
From Fructose-1,6-diphosphate aldolase



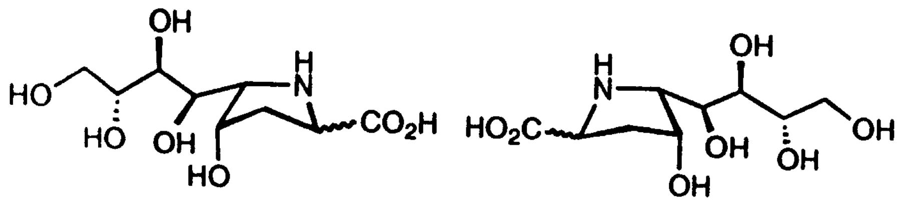
From 2-Deoxyribose-5-phosphate aldolase



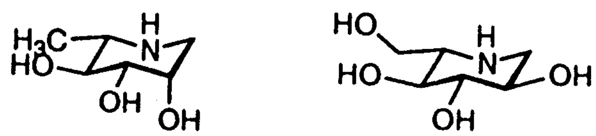
From Fuculose-1-phosphate aldolase



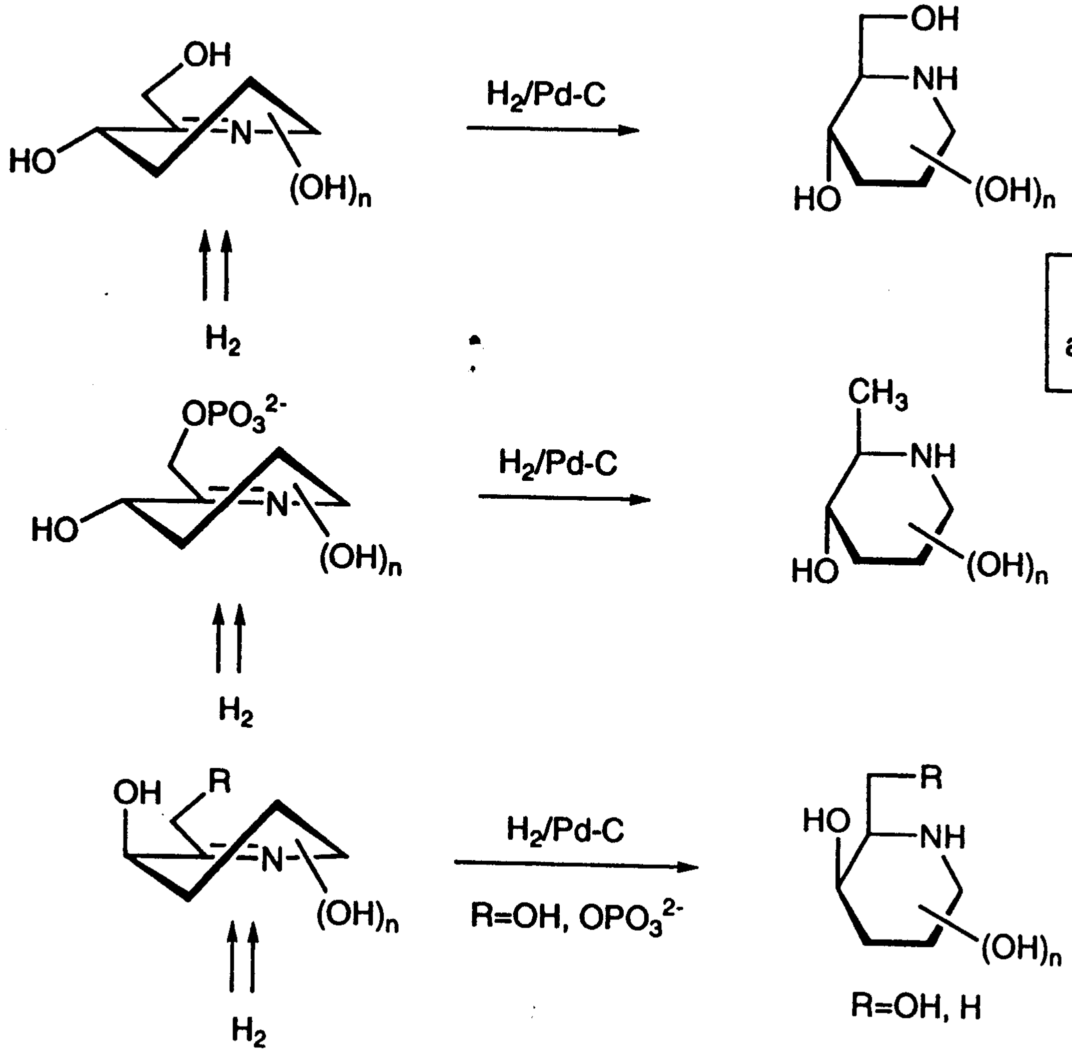
From Sialic acid aldolase



From Rhamnulose-1-phosphate aldolase



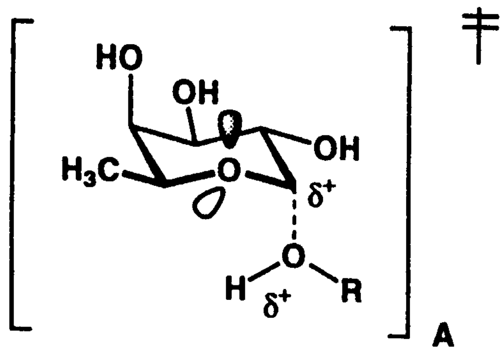
Stereochemistry of Pd-Mediated Reductive Amination



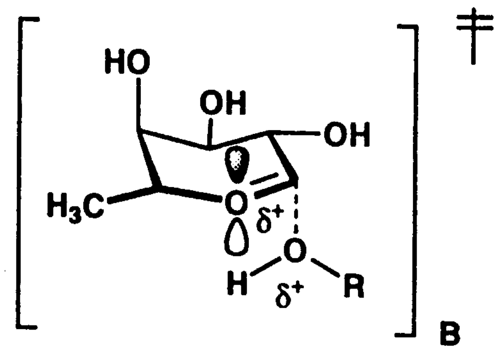
Trans product
avoid tortional strain

Cis product
Steric effect

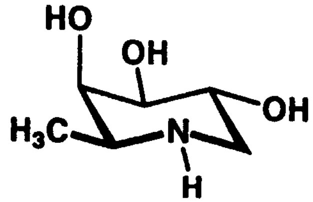
α-Fucosidase-Catalyzed Reaction



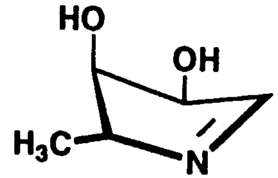
A



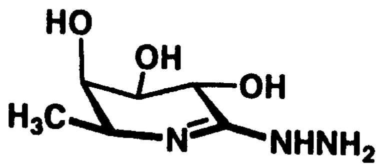
B



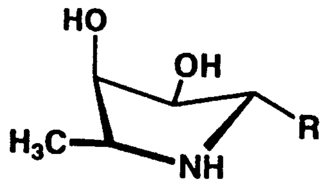
K_I = 10 nM



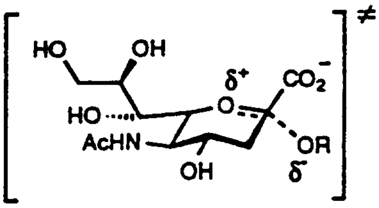
K_I = 160 nM

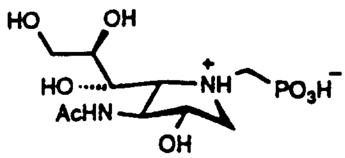


K_I = 820 nM

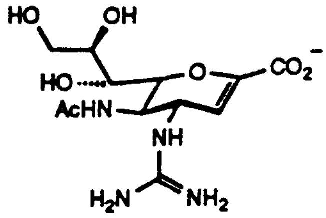


R	K_I (μM)
H	2.0
CH_2OH	1.2
CH_2NH_2	1.9

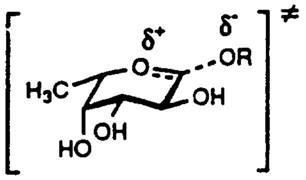


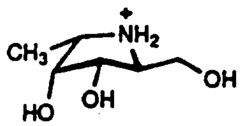


$K_i = 5.3 \times 10^{-5}$ M
sialidase (*V. cholae*)
(ref. 325b)

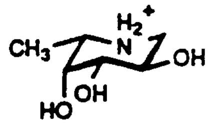


$K_i = 2 \times 10^{-10}$ M
influenza virus sialidase
(ref. 325c)

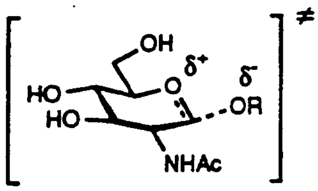


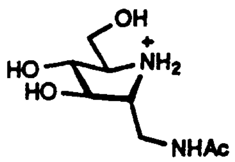


$K_i = 1.4 \times 10^{-8}$ M
α-fucosidase (bovine kidney)
(ref. 54)

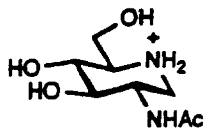


$K_i = 1 \times 10^{-8}$ M
α-fucosidase
(human liver) (ref. 325d)

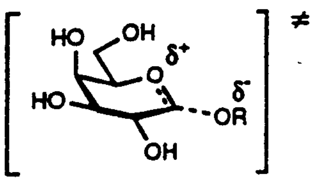


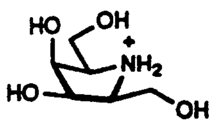


$K_i = 1.9 \times 10^{-6}$ M
β-*N*-acetylglucosaminidase
(Jack beans) (ref. 58)

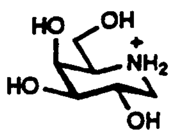


$K_i = 3.8 \times 10^{-7}$ M
β-*N*-acetylglucosaminidase
(bovine kidney) (ref. 51)

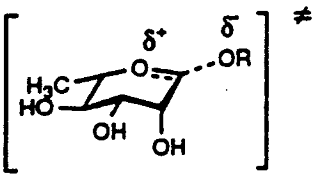


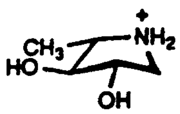


$K_i = 5.0 \times 10^{-8}$ M
α-galactosidase
(green coffee beans) (ref. 325e)

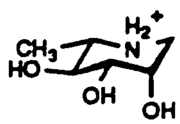


$K_i = 1.6 \times 10^{-9}$ M
α-galactosidase
(green coffee bean) (ref. 325f)





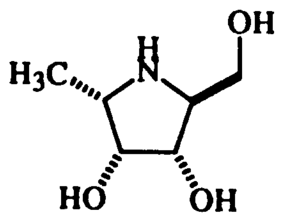
$K_i = 5 \times 10^{-6}$ M
α-rhamnosidase
(ref. 525g)

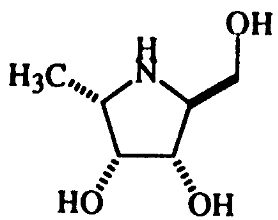


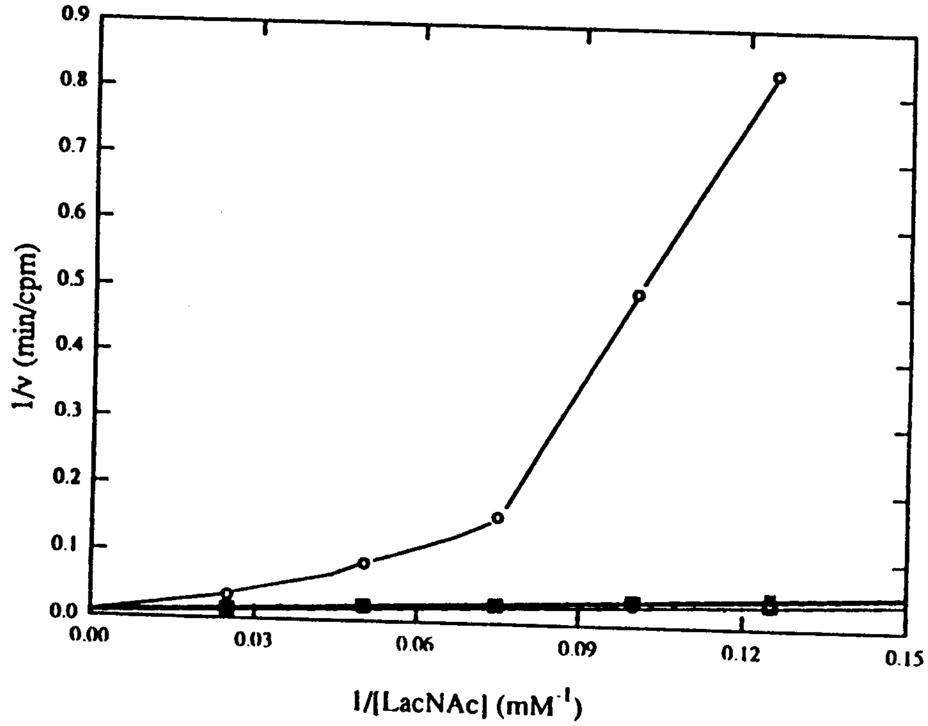
$K_i = 5 \times 10^{-5}$ M
α-rhamnosidase
(ref. 325g)

Synergistic Inhibition of α1,3-Fucosyltransferase

▲ GDP (50 μM)

□  (10 mM)

○ GDP (50 μM)  (10 mM)



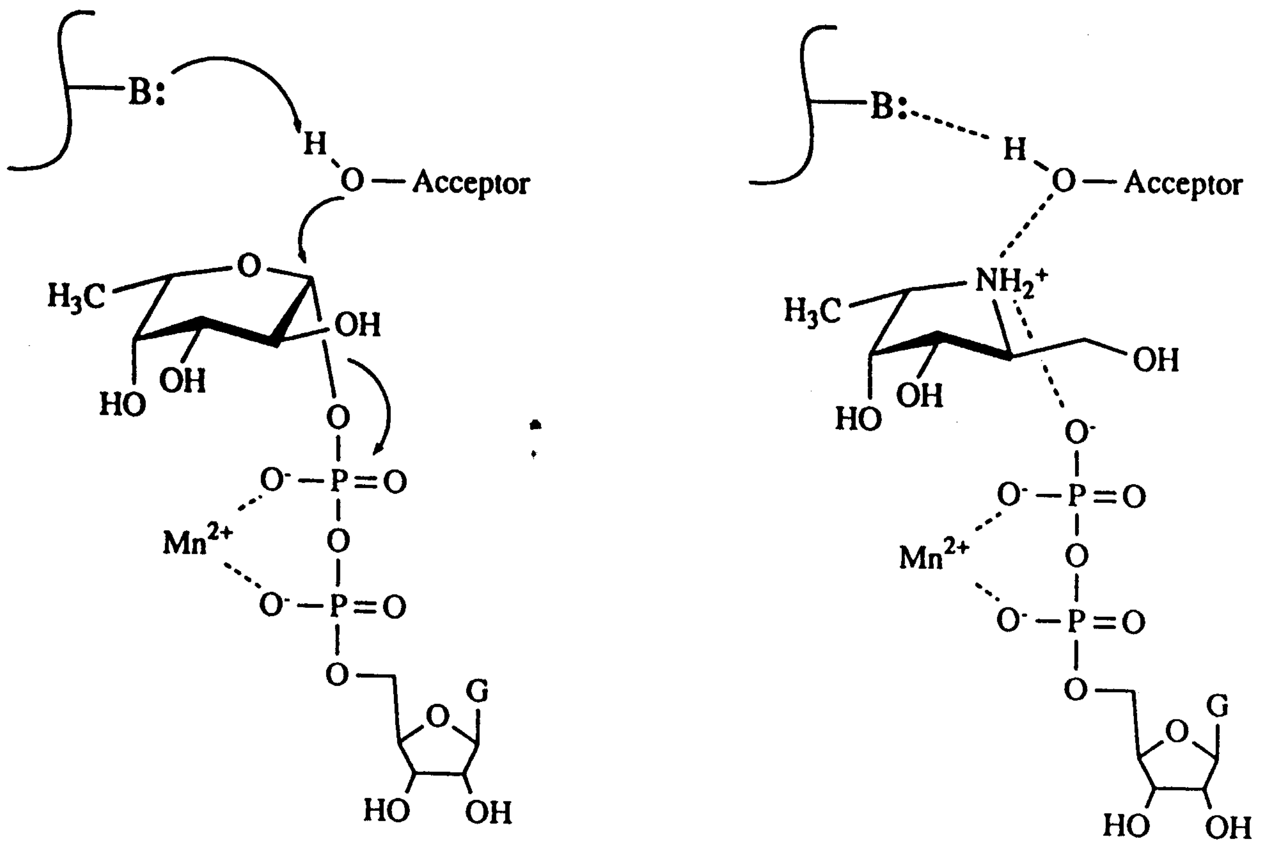
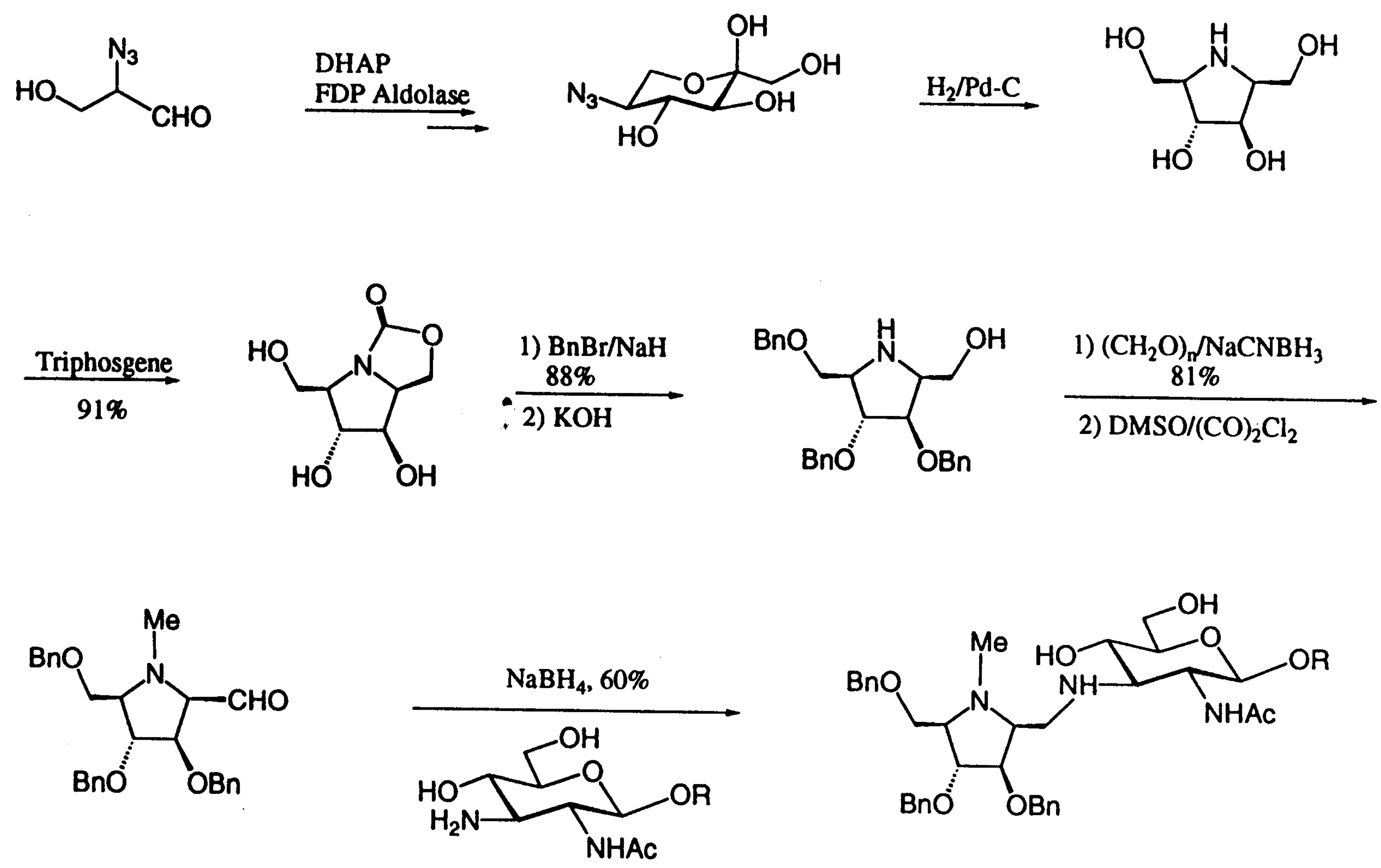
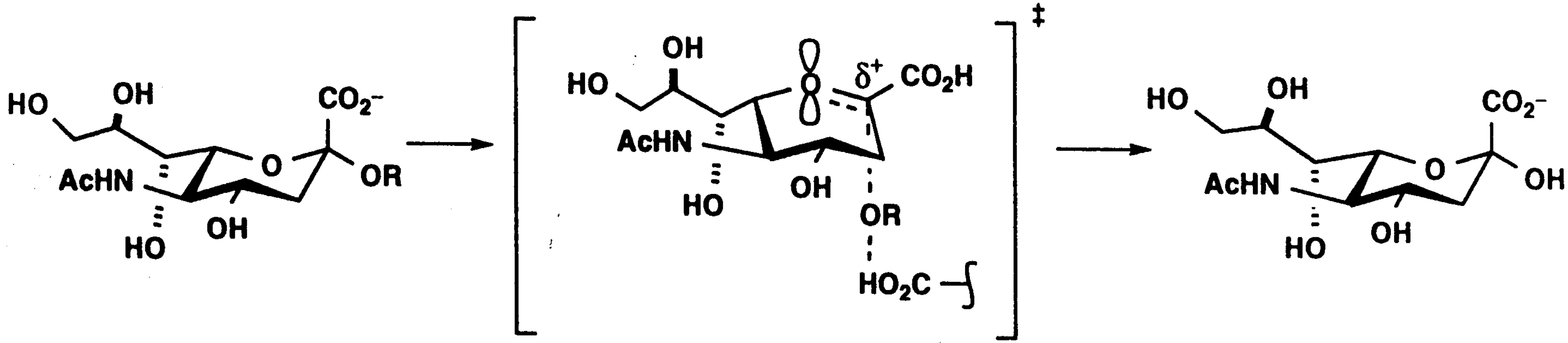


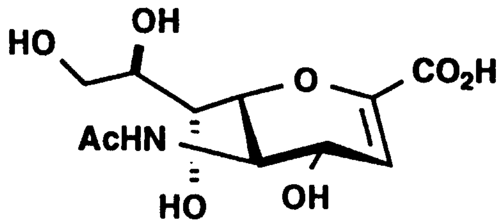
Figure 7. A proposed complex of GDP and a five-membered aza sugar which mimics the transition state of the fucosyltransfer reaction.



HO
N_3
CHO
DHAP
FDP Aldolase
N_3
HO
OH
OH
OH
H_2/Pd-C
HO
H
N
OH
HO
OH
Triphosgene
91%
O
HO
N
O
HO
OH
1) BnBr/NaH
88%
2) KOH
BnO
H
N
OH
BnO
OBn
1) $(CH_2O)_n$/NaCNBH$_3$
81%
2) DMSO/$(CO)_2Cl_2$
Me
BnO
N
CHO
BnO
OBn
NaBH$_4$, 60%
OH
HO
H_2N
OR
NHAc
Me
BnO
N
NH
HO
OH
OR
NHAc
BnO
OBn

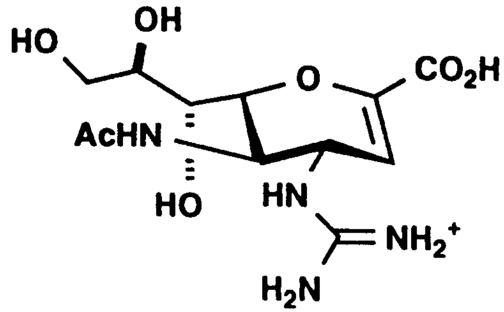
Sialidase (Neuraminidase) Reaction





$K_i = 1.4 \times 10^{-6}$ M

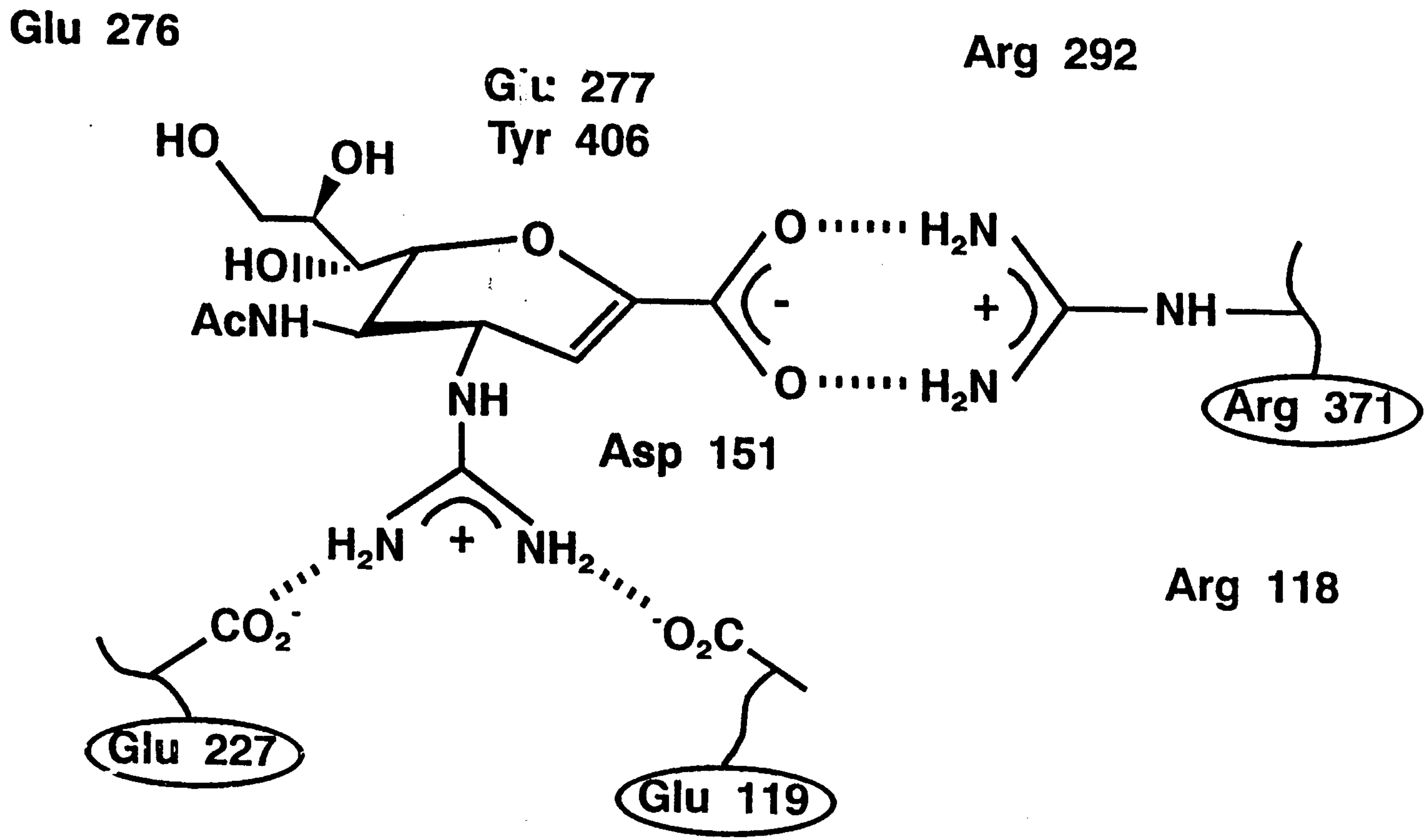
Miller et al.
BBRC **1978**, *83*, 1479



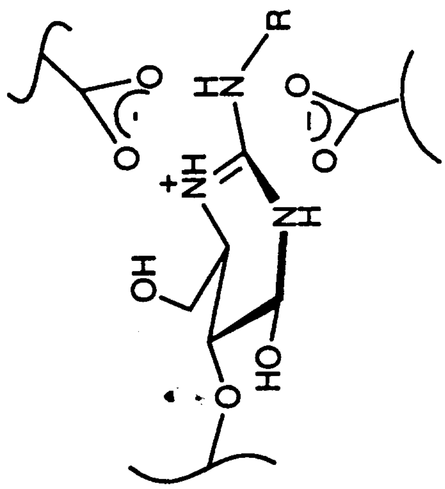
$K_i = 2 \times 10^{-10}$ M

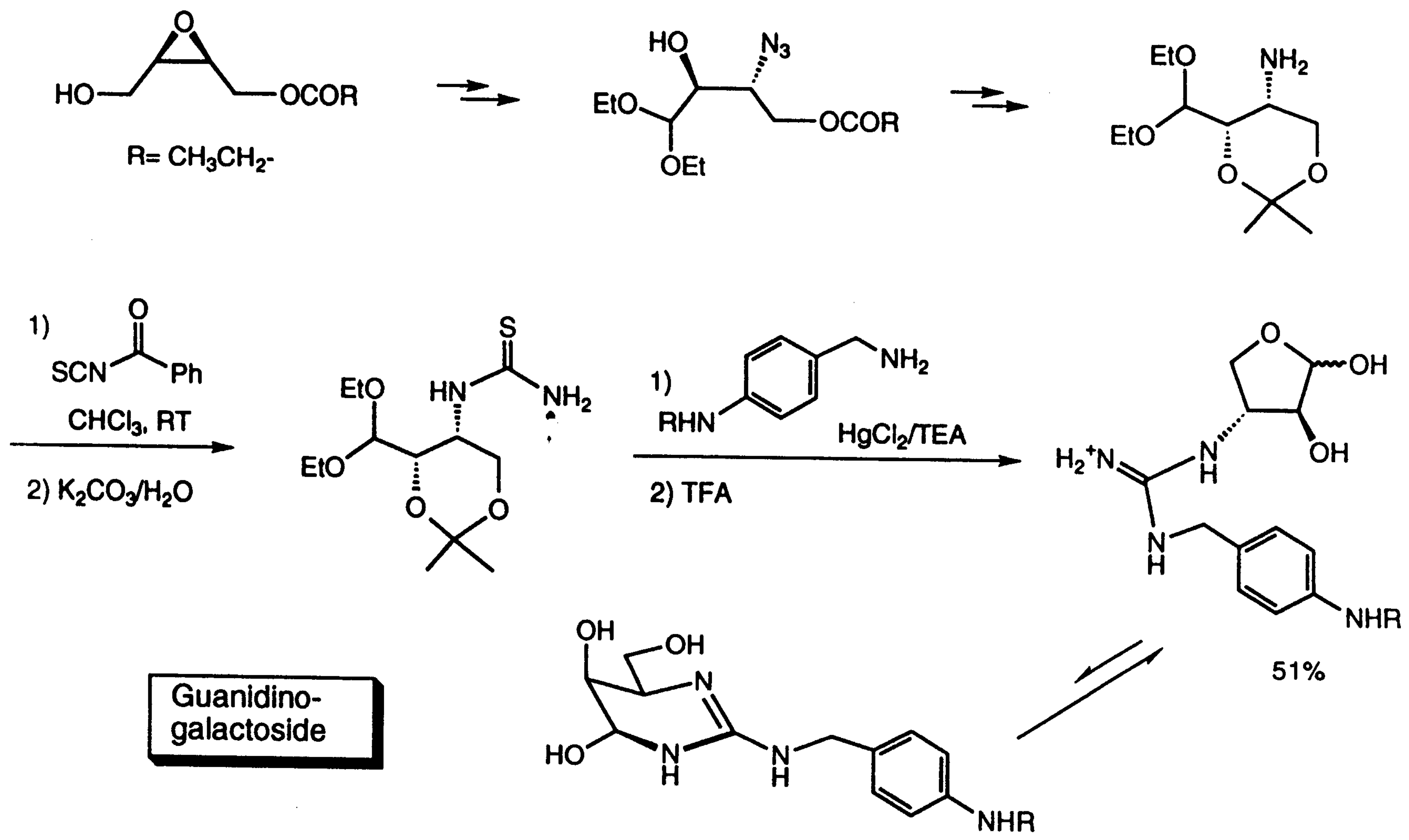
von Itzstein et al.
Nature **1993**, *363*, 418

Influenza Neuraminidase Inhibitor: 2-Deoxy-2,3-Didehydro-D-N-Acetylneuraminic Acid

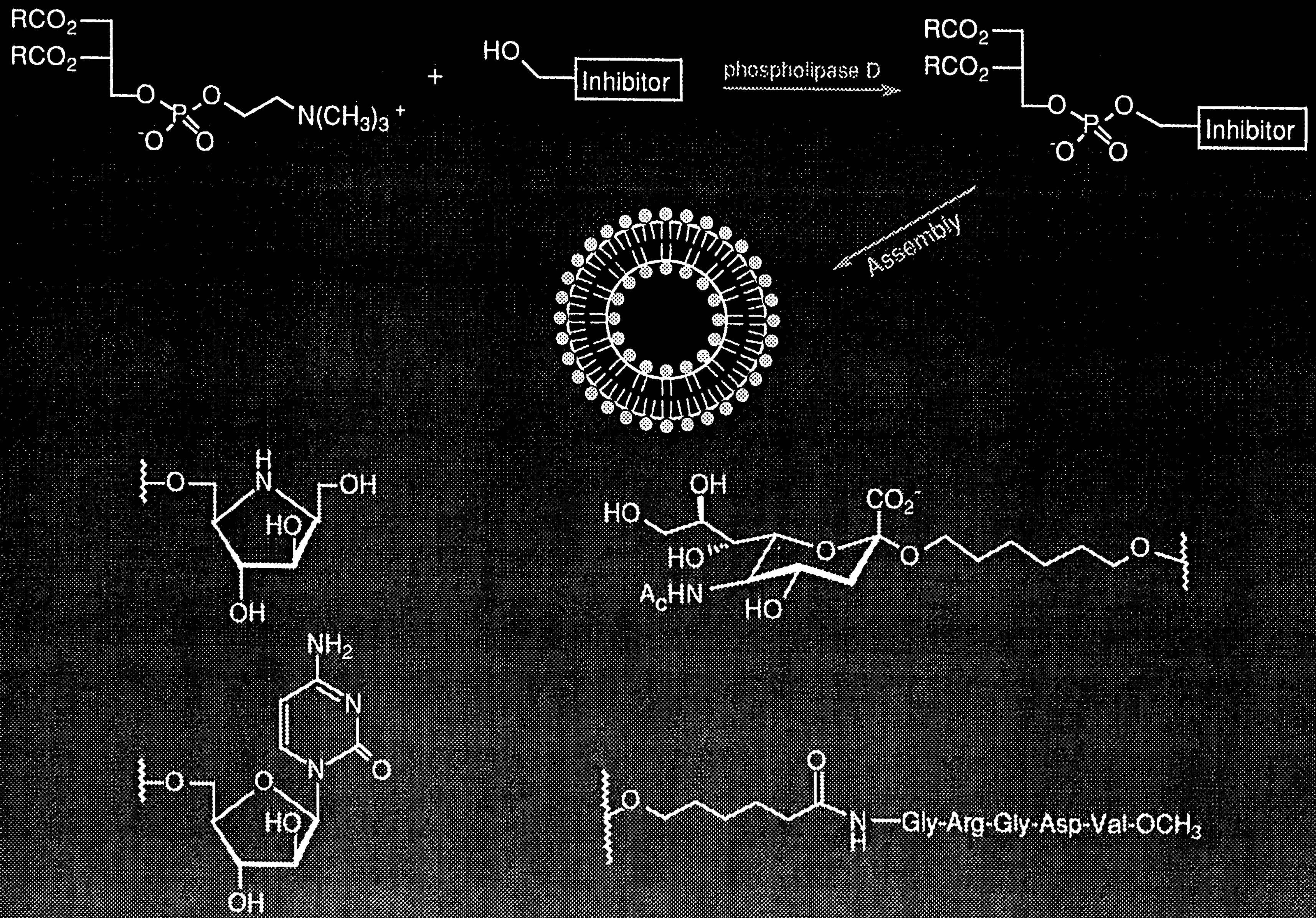


$K_I = 2 \times 10^{-10}$ M *Nature* 1993, 363, 418




HO
OCOR
R= CH3CH2-
HO
N3
EtO
OEt
OCOR
EtO
NH2
EtO
1)
SCN
Ph
CHCl3, RT
2) K2CO3/H2O
EtO
HN
S
NH2
EtO
1)
RHN
NH2
HgCl2/TEA
2) TFA
H2+N
OH
OH
NHR
51%
Guanidino-
galactoside
OH
OH
HO
NHR

Enzymatic Synthesis of Phospholipid-Inhibitor Conjugates



WongC/CHEM/dia1c/062