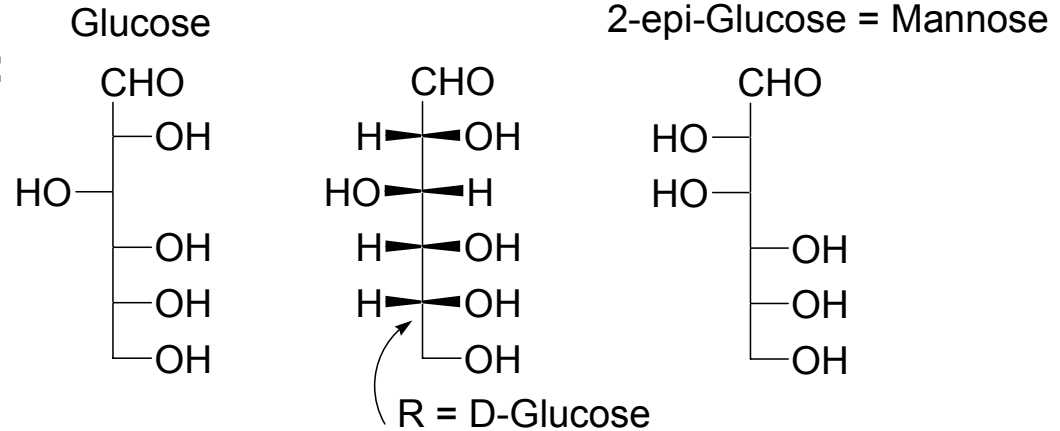


# Naturstoffchemie - Kohlenhydrate

Einführung:

## I. Die Fischer-Projektion:



## II. Stereoisomere:

4 chirale Zentren, kein Symmetrieelement.

→  $2^4 = 16$  Aldohehexosen-Stereoisomere.

→ 8 diastereomere Paare von Enantiomeren.

## III. D/L-Nomenklatur:

Die D/L-Nomenklatur für Monosaccharide definiert die absolute Konfiguration am höchst nummerierten chiralen Zentrum.

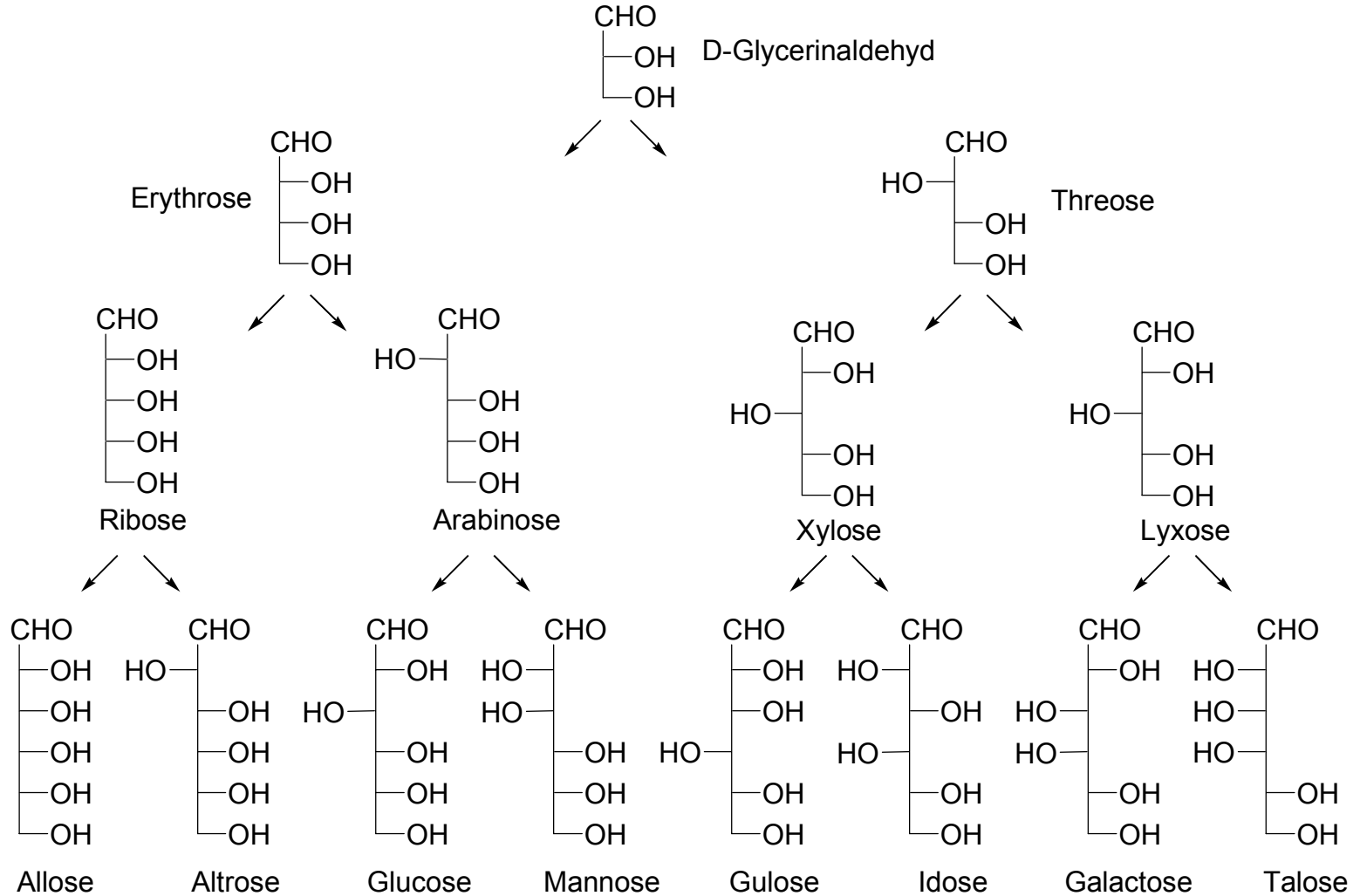
R → D-Zucker

S → L-Zucker

# Naturstoffchemie - Kohlenhydrate

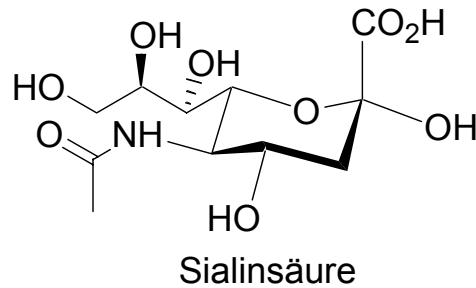
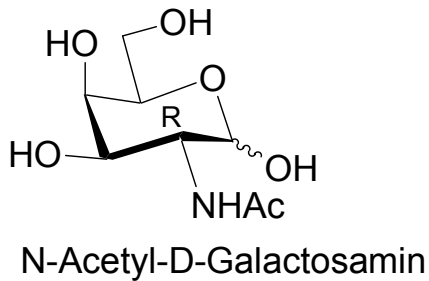
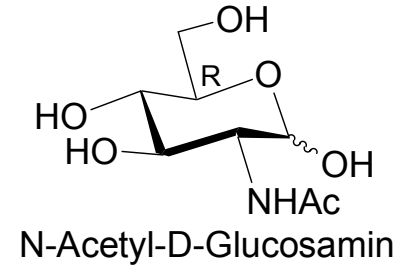
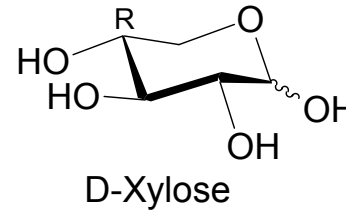
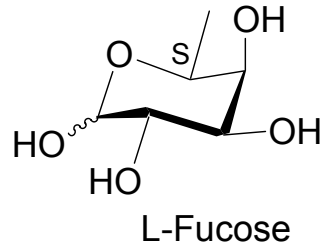
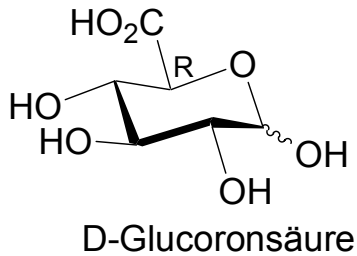
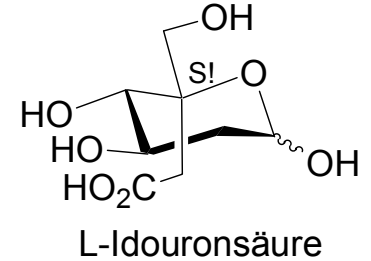
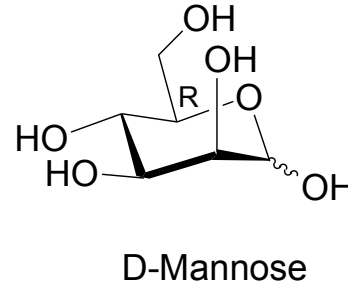
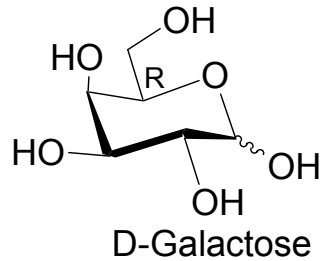
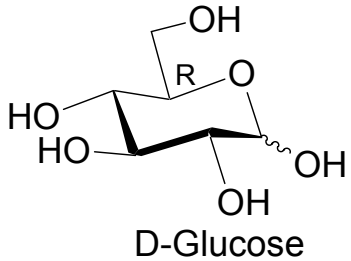
## Monosaccharide – Aldosen

Fischer-Projektion in der D-Serie der Aldosen. E. Fischer: Bestimmung der relativen Konfiguration aller Hexosen.



# Naturstoffchemie - Kohlenhydrate

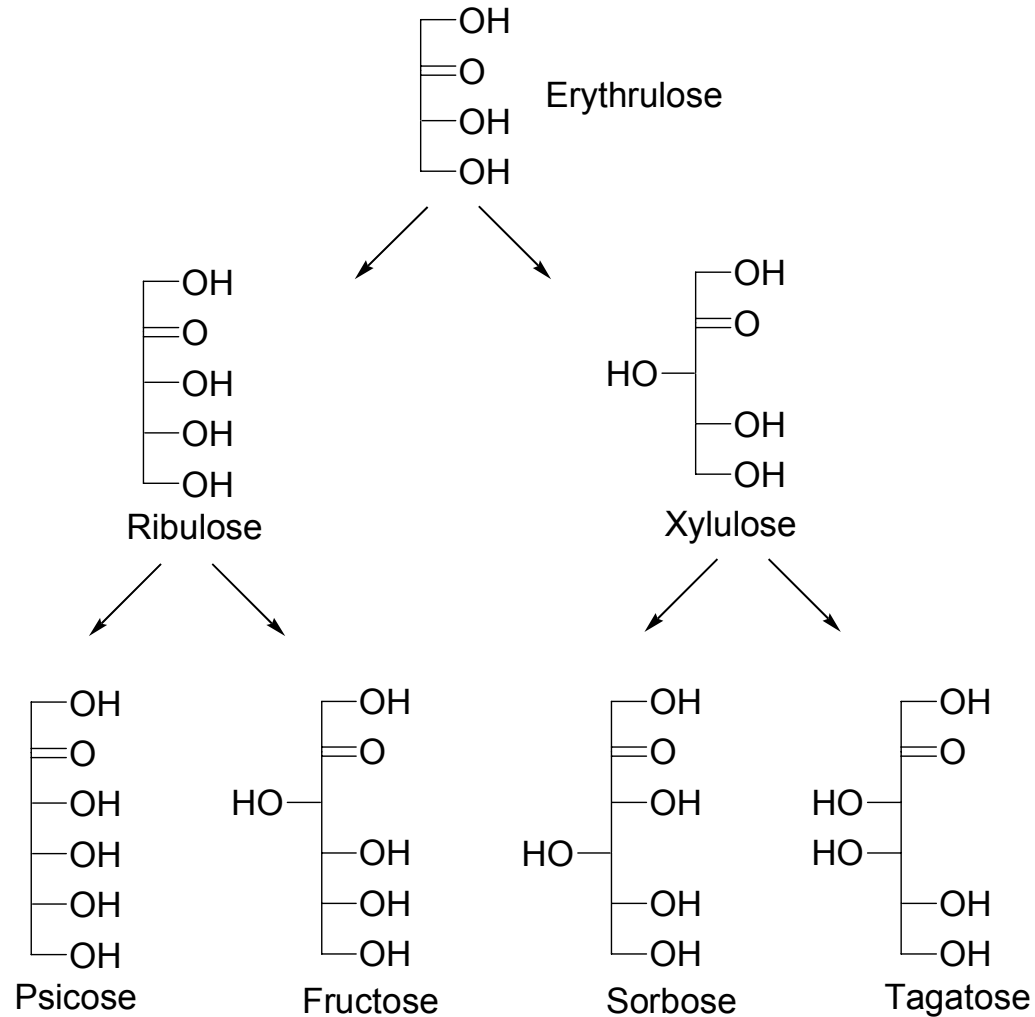
In Säugetieren häufig vorkommende Zucker.



# Naturstoffchemie - Kohlenhydrate

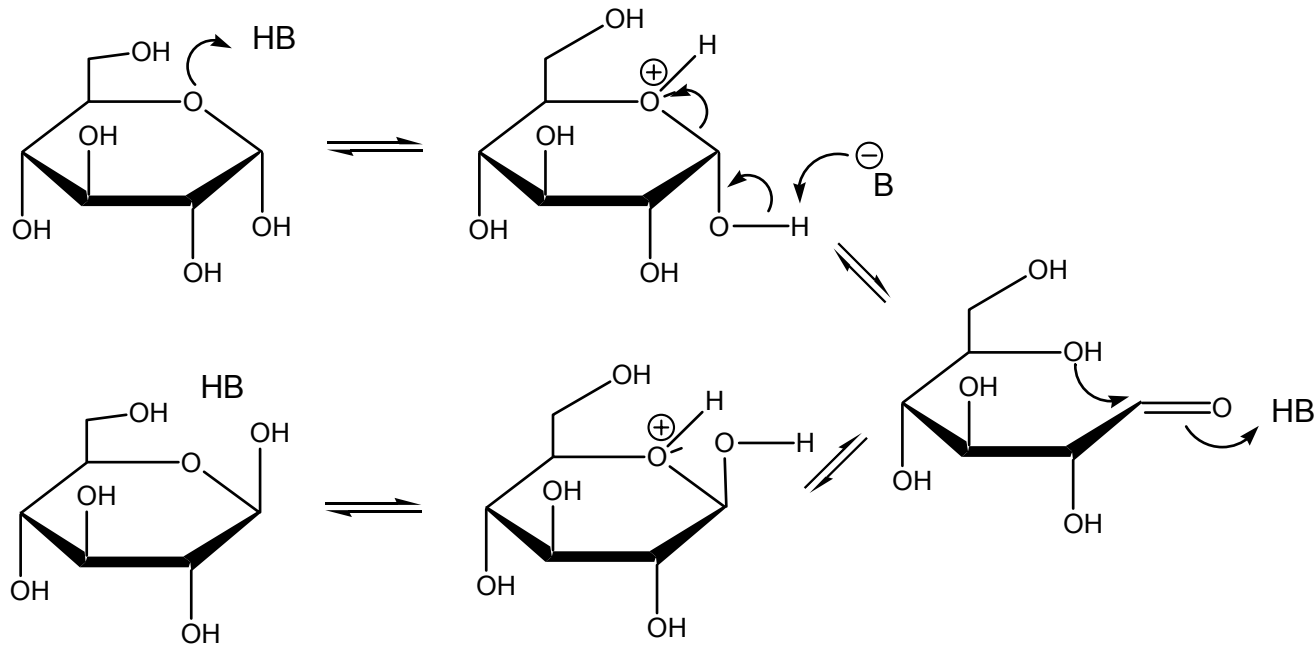
## Monosaccharide – Ketosen

Fischer-Projektion in der D-Serie der Ketosen.



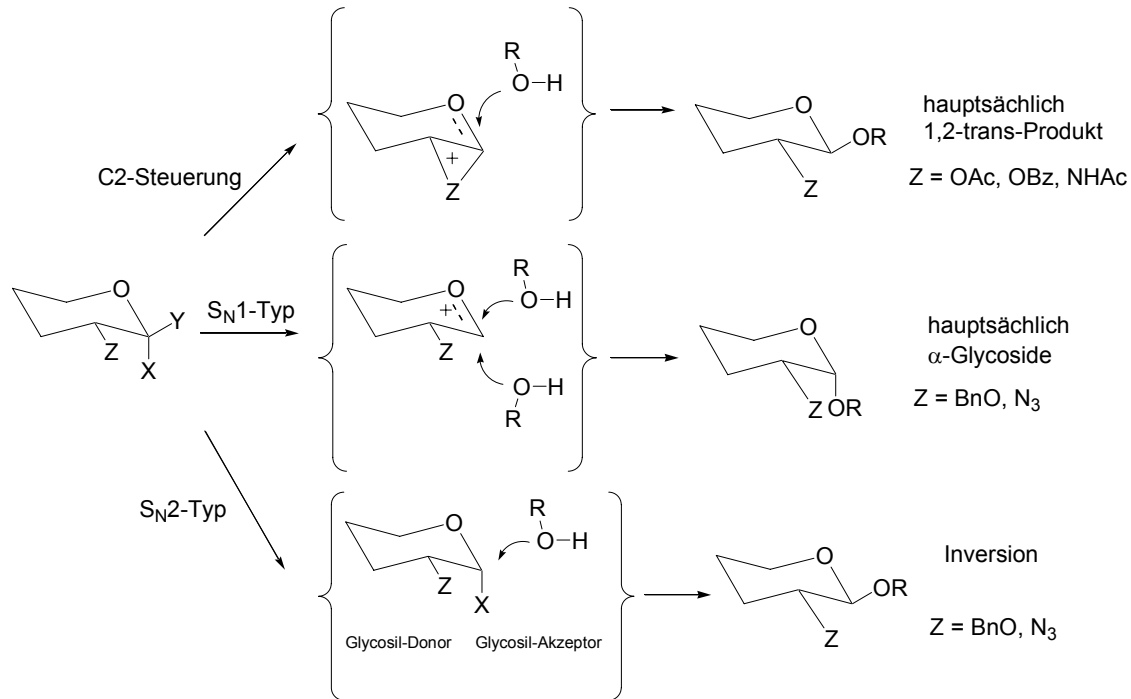
# Naturstoffchemie - Kohlenhydrate

**Mutarotation:** Einstellung eines Gleichgewichtes der Anomeren über eine offenkettigen Zwischenstufe. Allgemeine Säuren-Basen-Katalyse.

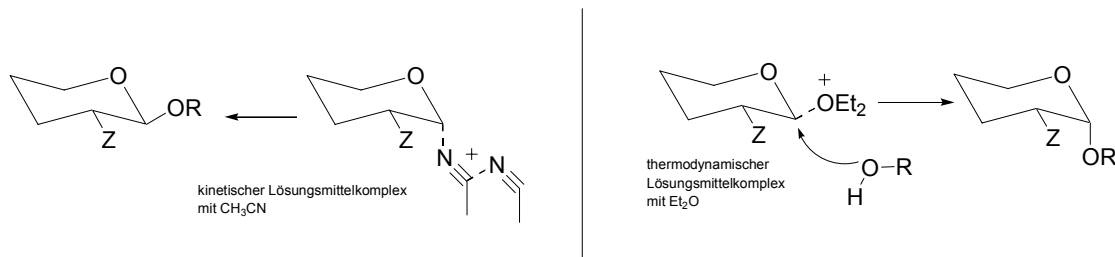


# Naturstoffchemie - Kohlenhydrate

## Glycosid-Synthese via Austausch am anomeren Sauerstoffatom

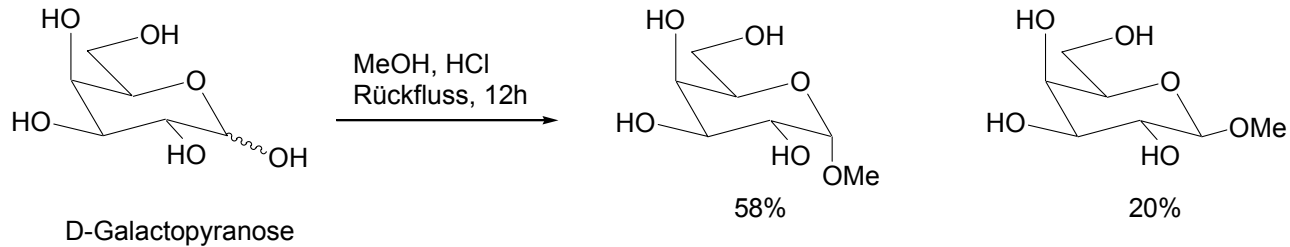


### Lösungsmiteleinflüsse beim $S_N1$ -Mechanismus

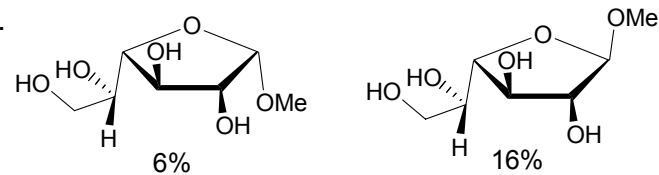


# Naturstoffchemie - Kohlenhydrate

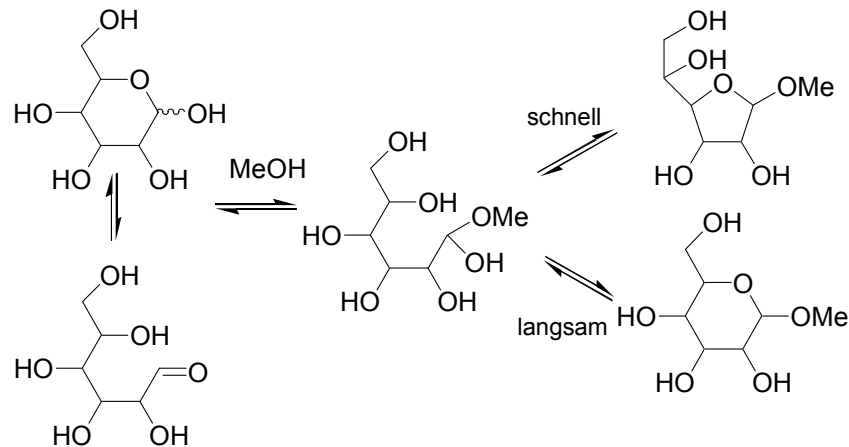
## Fischer-Glycosidierung



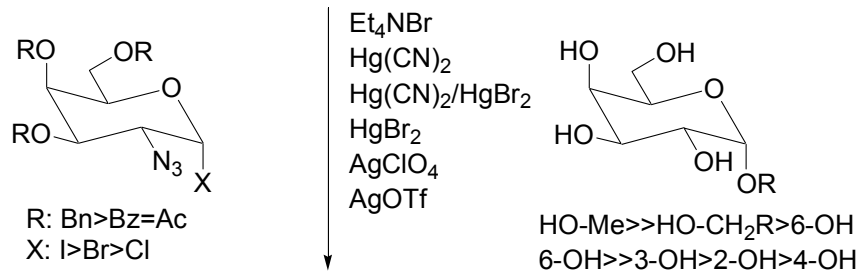
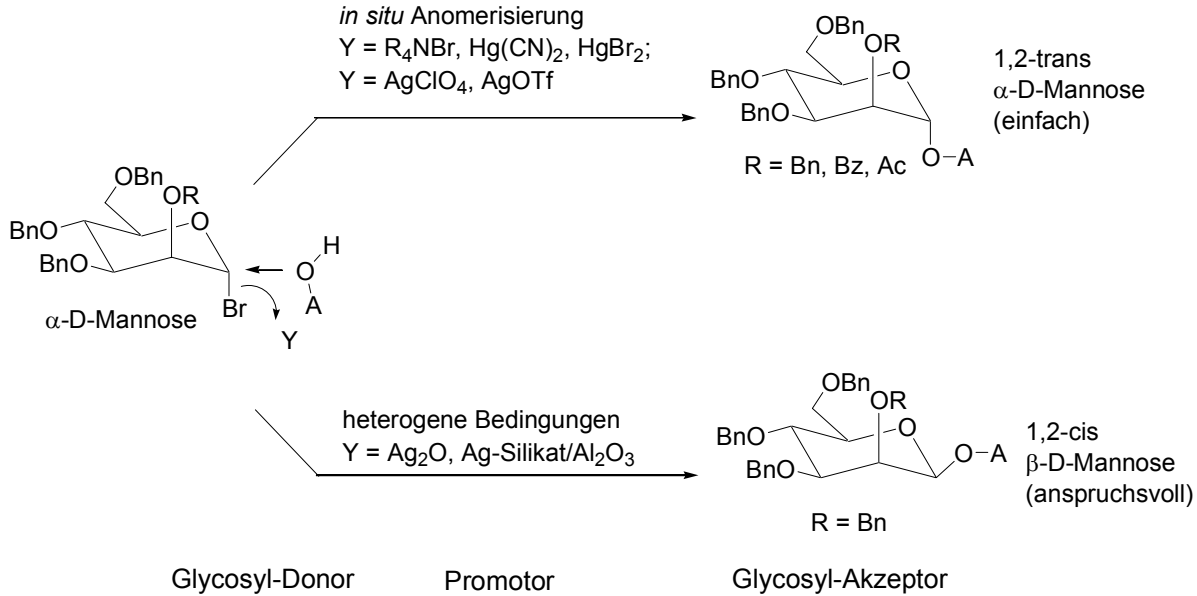
Thermodynamische Bedingungen (12h): hauptsächlich Pyranosen.  
Kinetische Bedingungen (6h): >50% β-D-Galactofuranoside.



Die Fischer-Glycosidierung läuft über die anfängliche Bildung der Furanoside.



# Naturstoffchemie - Kohlenhydrate Die Koenigs-Knorr-Reaktion



Eine Optimierung von Ausbeute und Selektivität verlangt nach einer genauen Einstellung der drei Parameter.

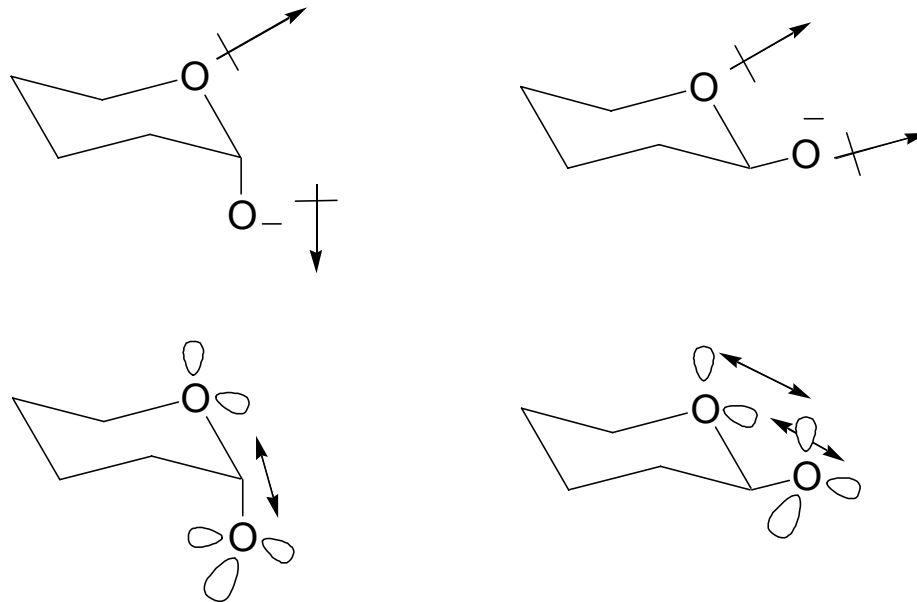
- 1) Reaktivität des Glycosyl-Halogenids - Abhängig von Schutzgruppen.
- 2) Reaktivität des Glycosyl-Akzeptors - Abhängig von Schutzgruppen und räumlichen Anspruch.
- 3) Aktivität des Promoters - Stark variierender Aktivitätsbereich.

# Naturstoffchemie - Kohlenhydrate

## Stereochemie der Trichloracetimidat-Bildung - Der **kinetische** anomere Effekt

$\beta$ -Glycosyloxid-Anionen sind aufgrund des kinetischen anomeren Effektes stärker nucleophil als  $\alpha$ -Glycosyloxid-Anionen.

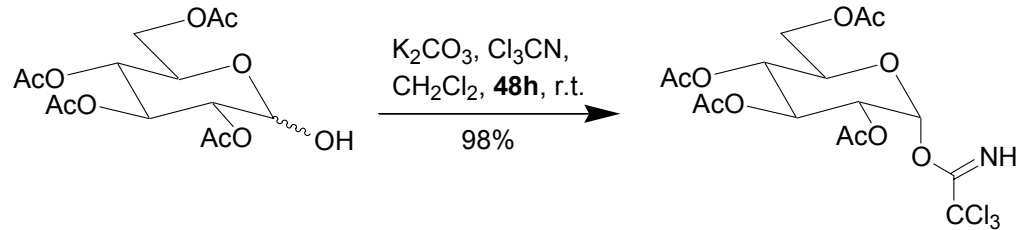
1.  $\beta$ -Glycoside sind sterisch leichter zugänglich (mehr „Platz“ in der äquatorialen Position).
2. Wechselwirkungen zwischen Dipolen und freien Ionenpaaren destabilisieren das  $\beta$ -Glycosyloxid-Anion.



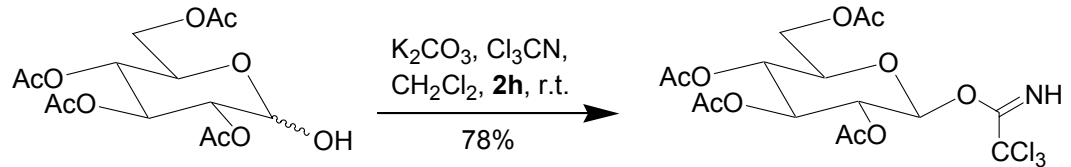
# Naturstoffchemie - Kohlenhydrate

1. Kinetische Bedingungen:  $\beta$ -Trichloracetimidat (kinetischer anomerer Effekt)
2. Thermodynamische Bedingungen:  $\alpha$ -Trichloracetimidat (anomerer Effekt)

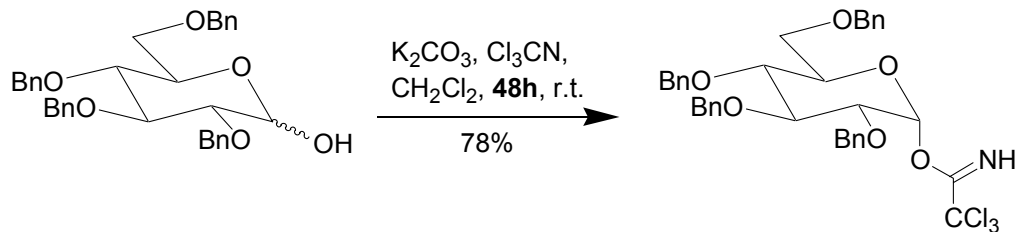
Thermodynamisch



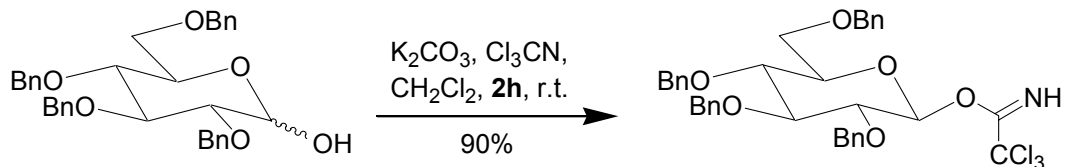
Kinetisch



Thermodynamisch

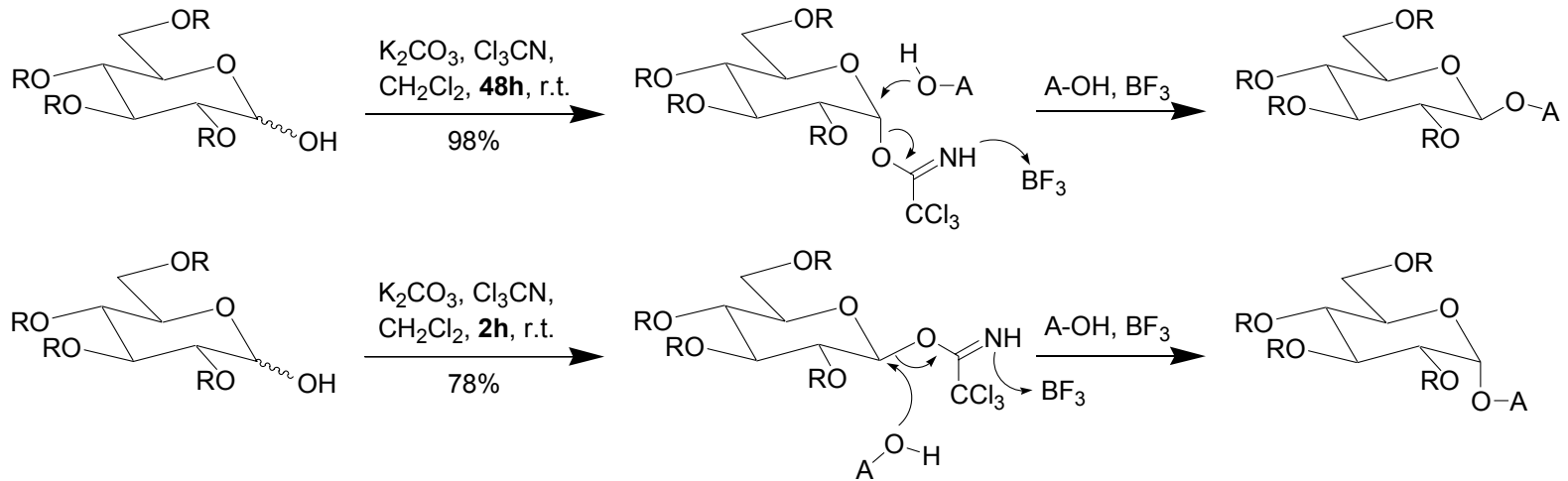


Kinetisch



# Naturstoffchemie - Kohlenhydrate

## Die Trichloracetimidat-Methode

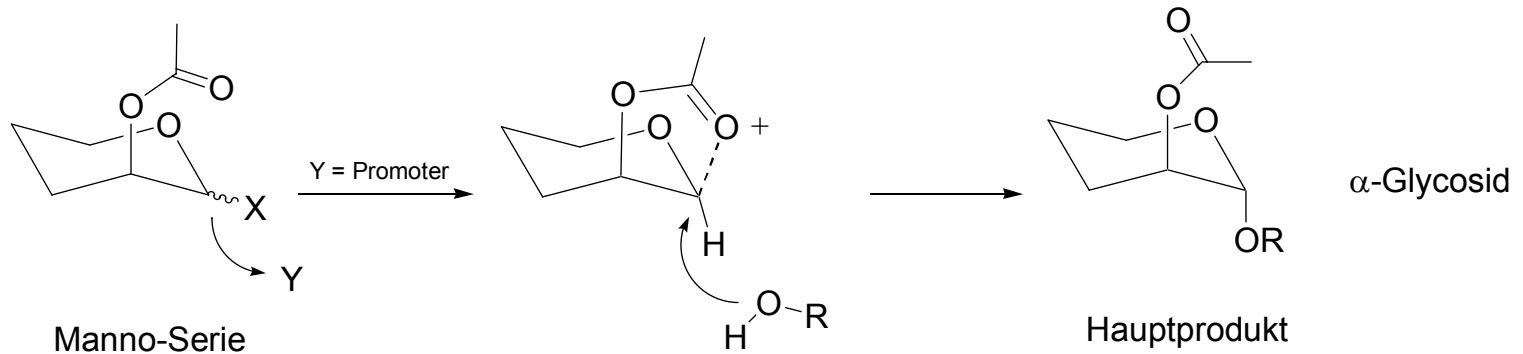
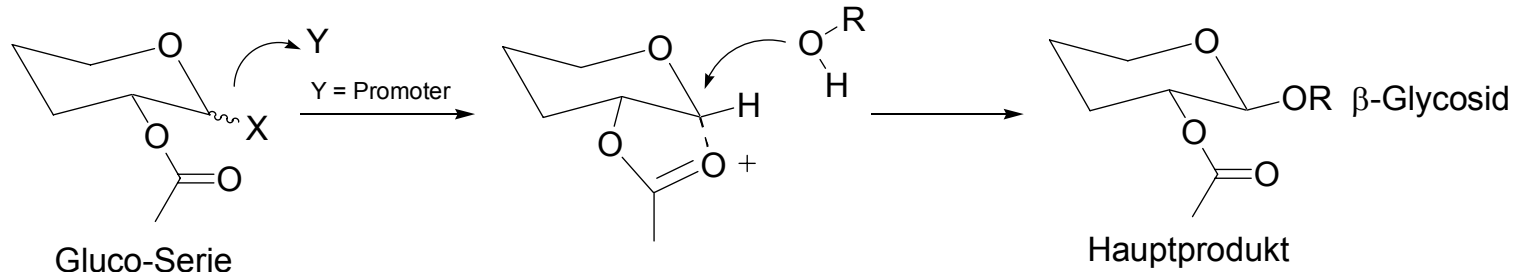


Kontrolle über das anomere Zentrum durch:

1. Nachbargruppen-Effekte ( $R = OAc, OBz, NHAc$ : 1,2-trans-Konfiguration.
2. Keine Nachbargruppen-Effekte ( $R = OBn$ )
  - a) Milde Lewis-Säure ( $BF_3$ ) und tiefe Temperatur: Inversion an C1.
  - b) Starke Lewis-Säure ( $TMSOTf$ ): Bildung der thermodynamischen Glycoside.
  - c) Lösungsmittel-Effekte:  $Et_2O$  begünstigt die  $\alpha$ -Konfiguration;  $MeCN$  begünstigt die  $\beta$ -Konfiguration.

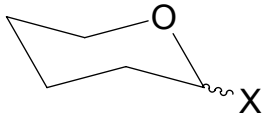
# Naturstoffchemie - Kohlenhydrate

Die Nachbargruppen-Beteiligung an C2 bietet Kontrolle über das anomere Zentrum

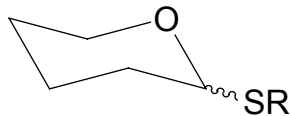


# Naturstoffchemie - Kohlenhydrate

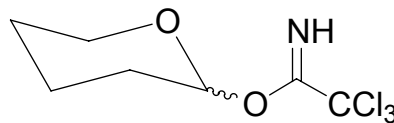
## Häufig verwendete Glycosyl-Donatoren



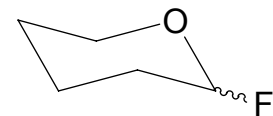
Glycosyl-Halogenide  
(X = Br, Cl)  
Aktivierung durch:  
 $R_4NBr$ ;  $Hg(CN)_2$ ,  
 $AgClO_4$ ,  $AgOTf$



Thio-Glycoside  
(R = Alkyl, Aryl)  
Aktivierung durch:  
NIS/TfOH; DMTST

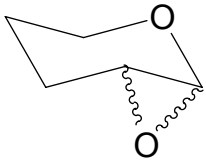


Trichloroacetimidate  
Aktivierung durch:  
 $BF_3 \cdot OEt_2$ , TMSOTf

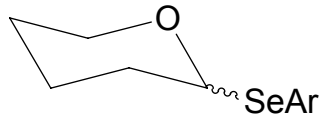


Glycosyl-Fluoride  
Aktivierung durch:  
 $SnCl_2/AgClO_4$ ;  
 $HfCp_2Cl_2$ , ( $AgOTf$ )

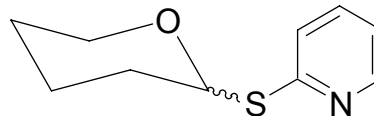
## Weitere Glycosyl-Donatoren



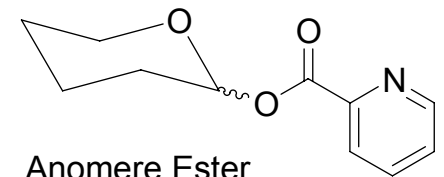
Epoxide  
Aktivierung durch:  
 $ZnCl_2$



Seleoglycoside  
Aktivierung durch:  
 $AgOTf/K_2CO_3$



Pyridylthioglycoside  
 $Hg(NO_3)_2$ ,  $AgOTf$



Anomere Ester  
 $Cu(OTf)_2$ ,  $Sn(OTf)_2$

# Naturstoffchemie - Kohlenhydrate

## Die biologische Bedeutung der Oligo- und Polysaccharide

### Struktur-gebende Eigenschaften:


- Zellulose: Zellwände der Pflanzen, Baumwolle, ...
- Agar: Zellwandmaterial von Seegras.
- Chitin: Skelett der Insekten, ...

### Energiespeicher:

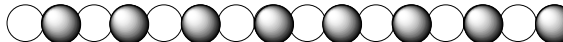
- Glycogen: Energiespeicher in Säugetieren.
- Stärke (Mischung aus Amylose und Amylopektin), Energiespeicher der Pflanzen.

### Träger der biologischen Information:

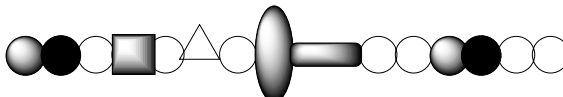
- Zell-Zell-Kontakte/Erkennung, Wachstumskontrolle, Entwicklung des Nervengewebes.
- Antigen-Antikörper-Wechselwirkung.
- Glycokonjugate: Oligosaccharide (Glycan) verknüpft an Biopolymere (Protein oder Lipid).

 A: Lineare Polysaccharide: Cellulose, Amylose (Stärke)

 B: Branched Polysaccharides: Amylopectin, Glycogen (Stärke)

 C: Alternierende Kohlenhydrat-Einheiten: Agarose

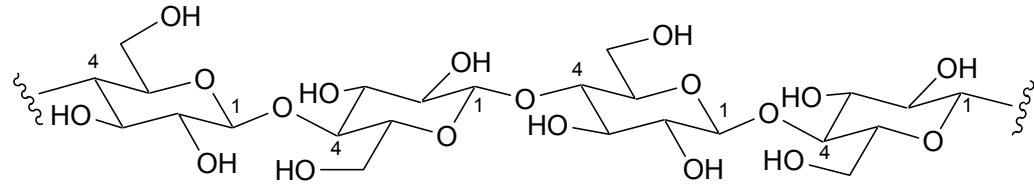
 D: Oligosaccharid-Blöcke: Alginat

 E: Komplexe Lineare Kohlenhydrat-Einheiten: Gellan

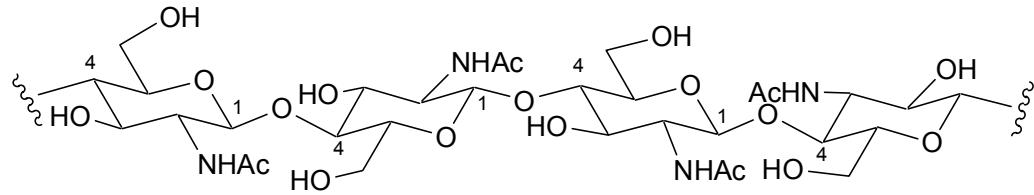
## Polysaccharidstrukturen

# Naturstoffchemie - Kohlenhydrate

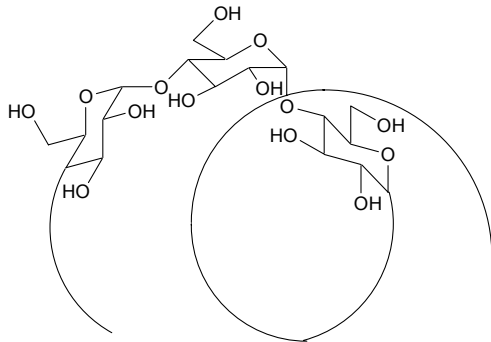
Cellulose:  $\beta$ -(1 $\rightarrow$ 4)-verknüpfte Glucose-Einheiten:



Chitin:  $\beta$ -(1 $\rightarrow$ 4)-verknüpfte N-Acetylglucosamin-Einheiten (GlcNAc):

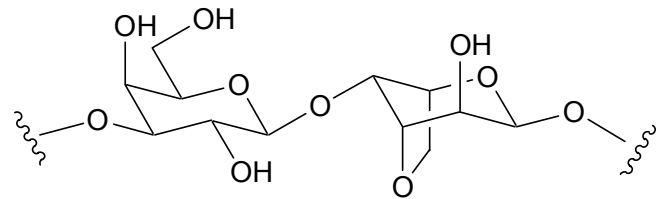


Amylose (eine Komponente von Stärke):  
 $\alpha$ -(1 $\rightarrow$ 4)-verknüpfte Glucose-Einheiten.



Agarose (aus Algen):

Hauptsächlich Galactose/Anhydrogalactose-Einheiten.



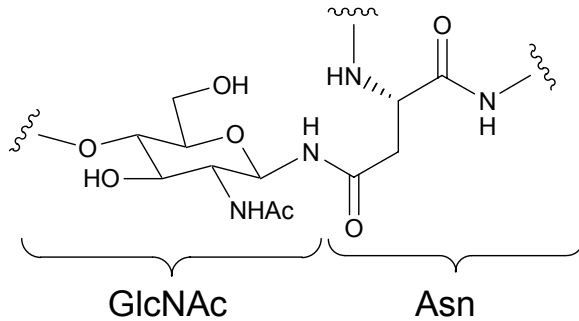
# Naturstoffchemie - Kohlenhydrate

## Glycokonjugate als Träger der biologischen Information

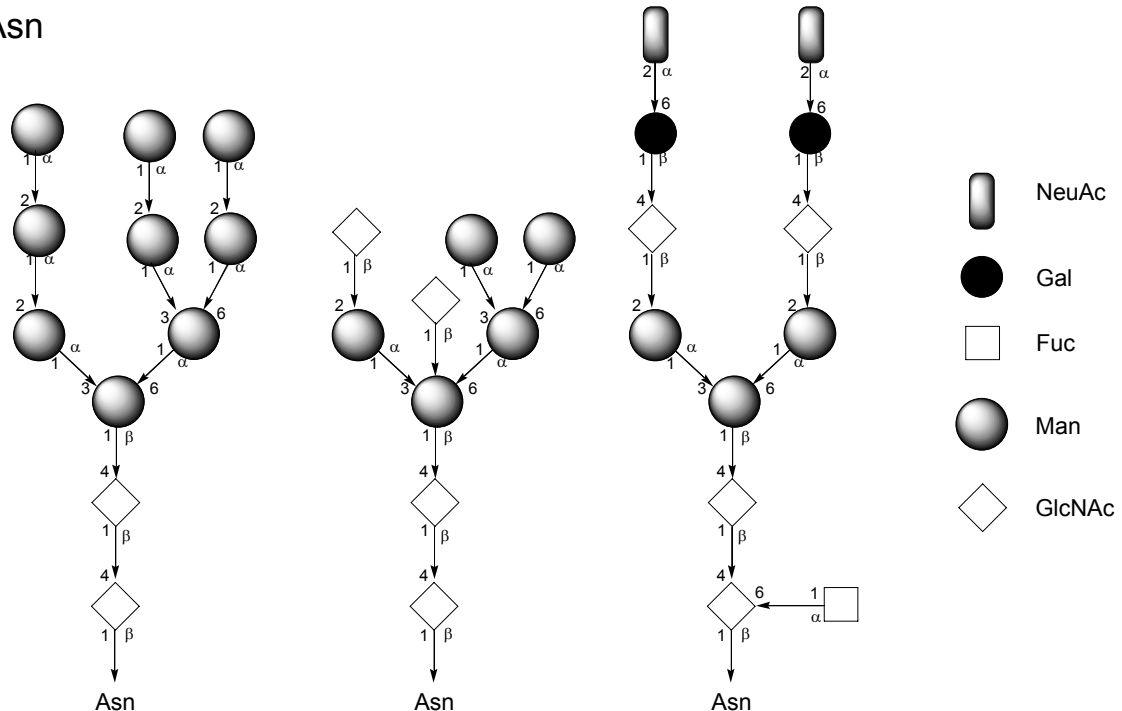
Glycokonjugate = Glycan + Protein/Lipid

### Glycoproteine:

#### N-verknüpfte Glycoproteine

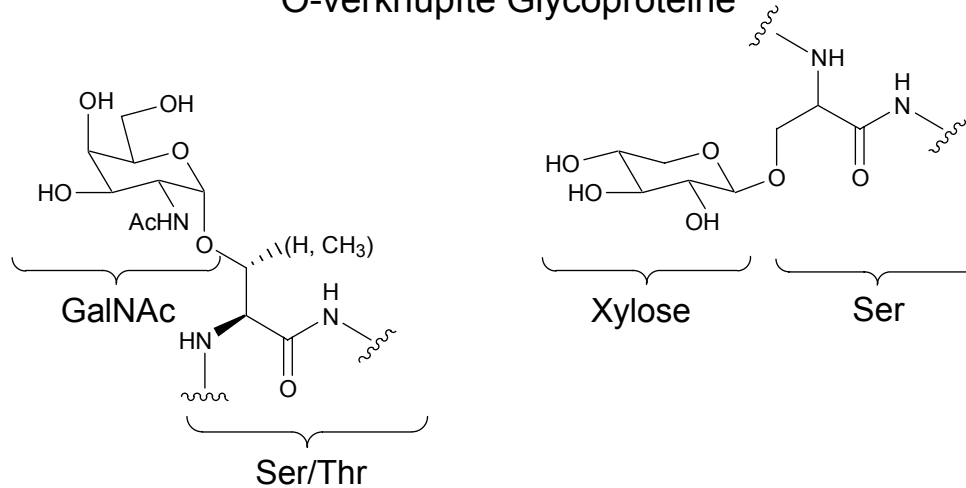


- Verknüpfung des Glycans (GlcNAc) mit der Amid-Gruppe von Asparagin.
- Das Pentasaccharid besitzt immer die selbe Oligomannose-Struktur.
- Beispiele: Serumproteine (Amylase), Plasmaproteine (Fibrinogen), Immunglobuline, Hormone, Membran-Glycoproteine.

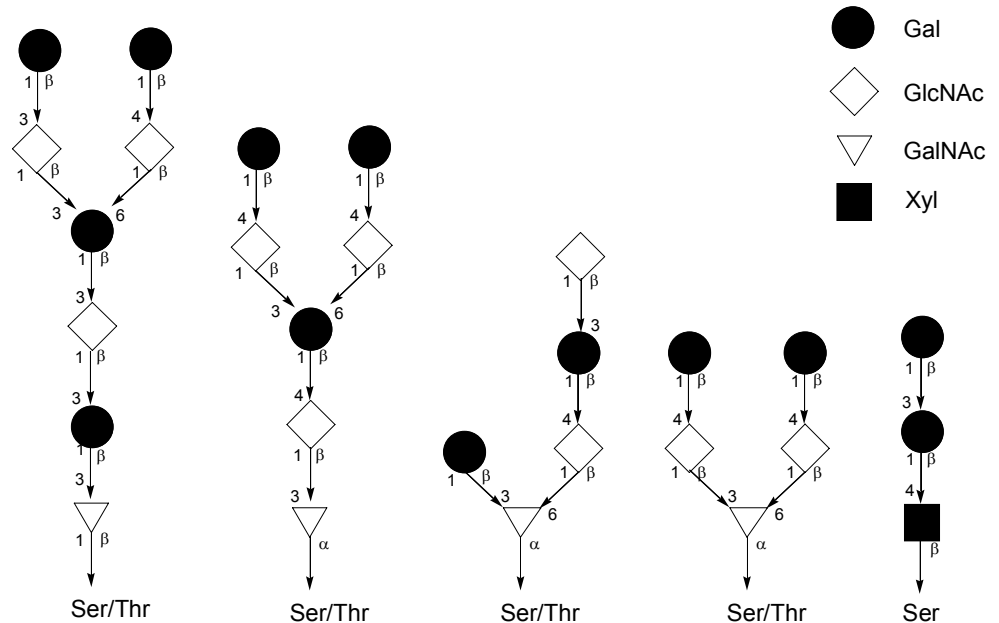


# Naturstoffchemie - Kohlenhydrate

## O-verknüpfte Glycoproteine



Die Strukturen der O-Glycoproteine sind Vielfältiger als die der N-Glycoproteine. Weit verbreitet ist das erste Motif. Beispiele: Serum- und Membran-Glycoproteine (Blutgruppen-Glycoproteine); Heparin.



# Naturstoffchemie - Kohlenhydrate

## Glycosphingolipide:

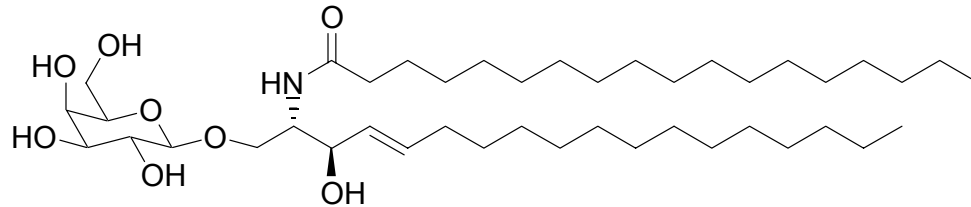
Glycosylierte Sphingosin-Derivate (hauptsächlich Ceramide)

Cerebroside: Eine Kohlenhydrateinheit (oft Glucose oder Galactose)

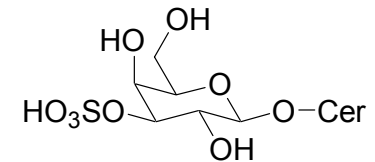
Komplexe Glycosphingolipide: Verzweigte und unverzweigte Oligosaccharide ( $\leq 20$  Monomere)

Ganglioside: Glycosphingolipide mit Sialinsäure

Sulfatide: sulfonylierte Glycosphingolipide.



Galactosylceramid (Galactocerebrosid)

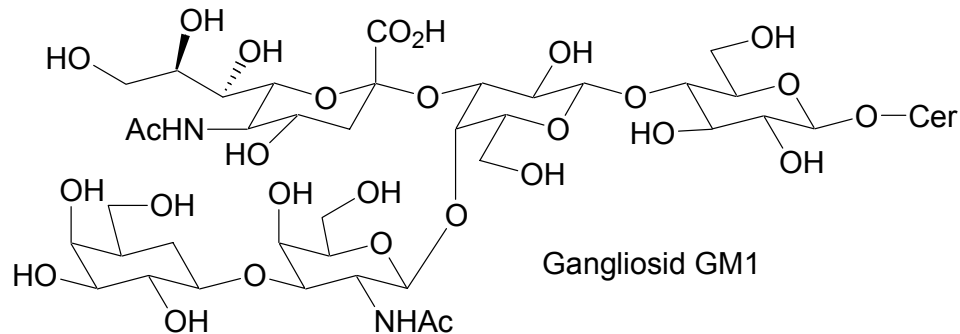


3-Sulfogalactosylceramid

Vorkommen:

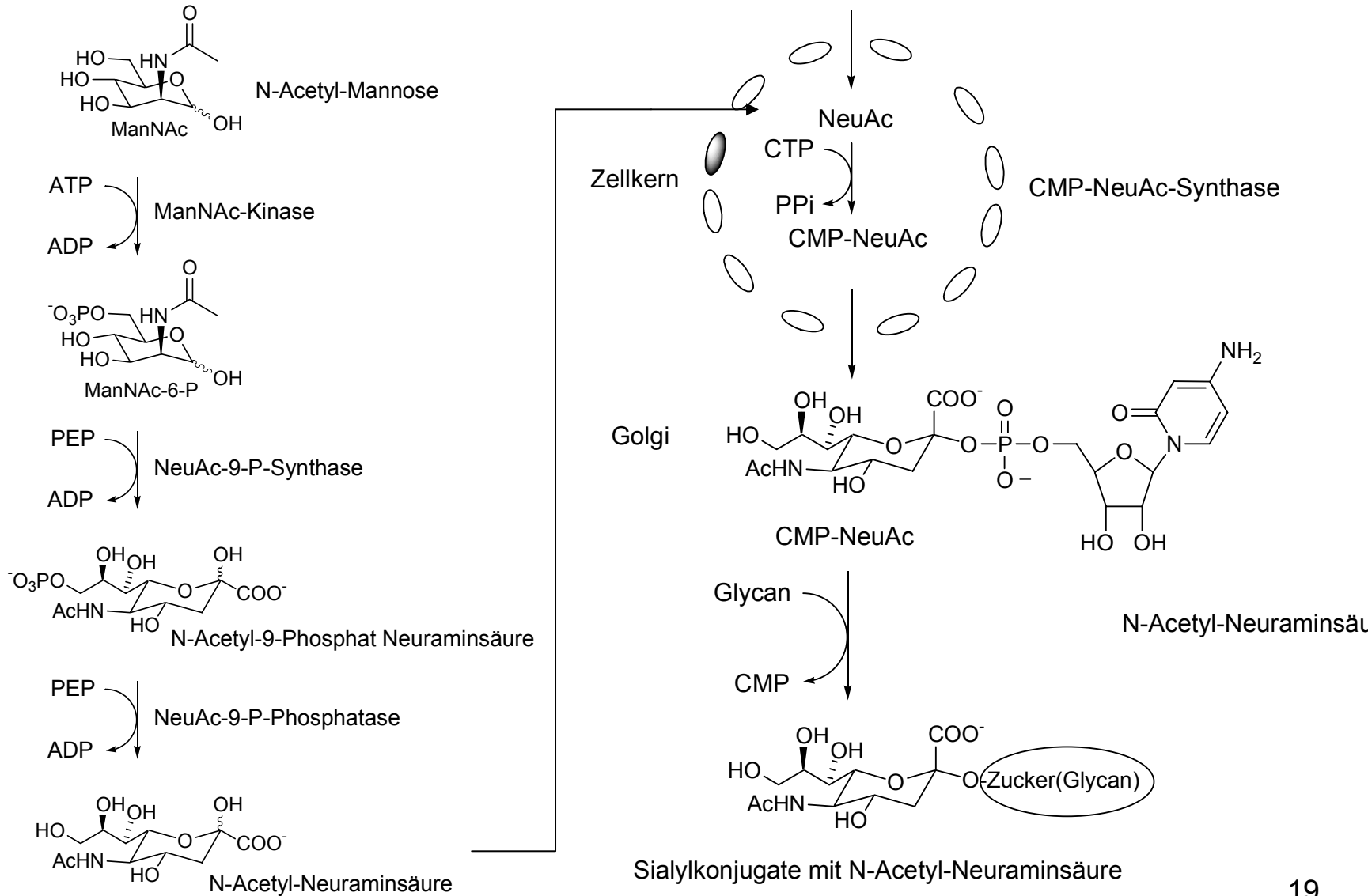
Zellmembran, Nervensystem

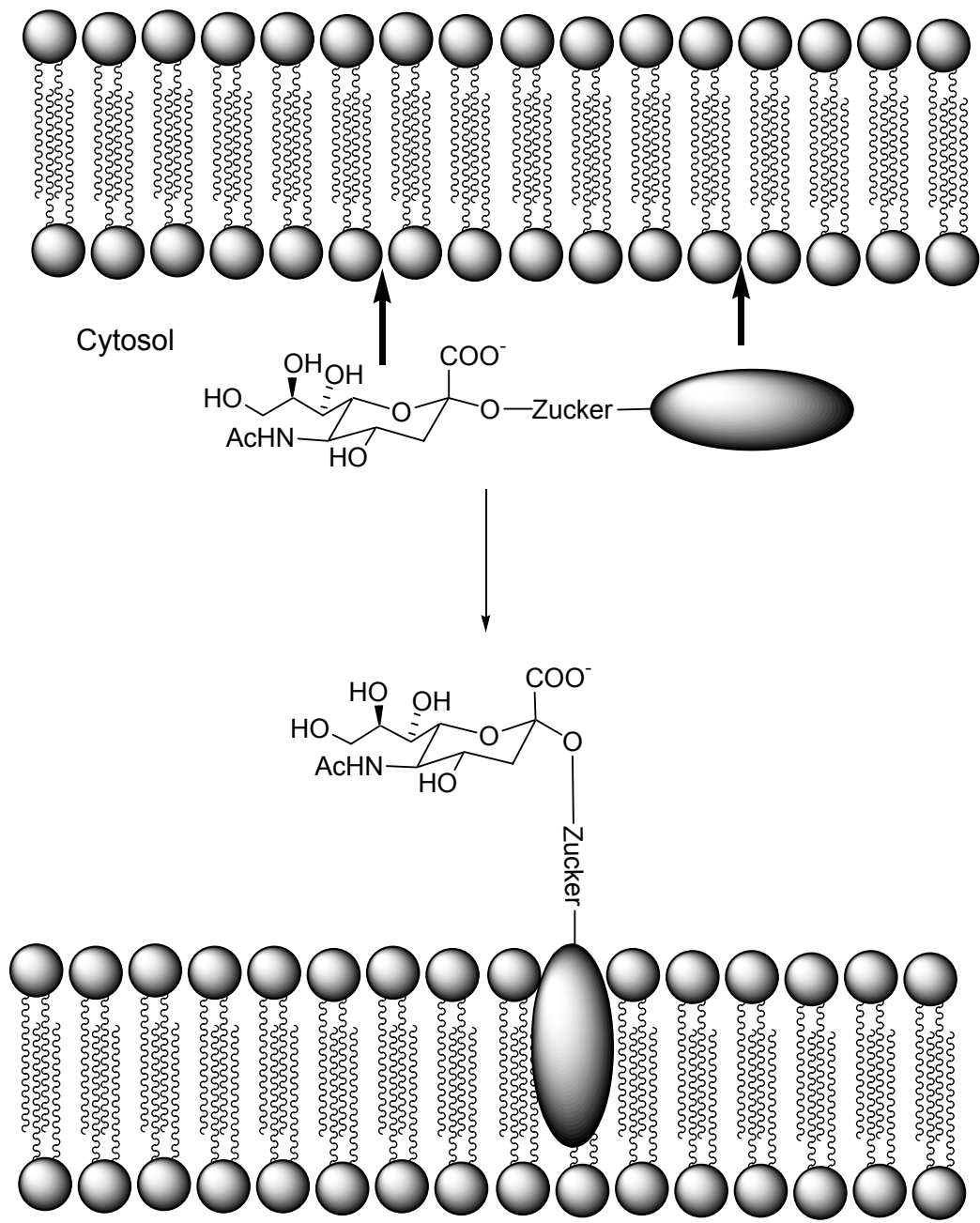
Biologische Funktion: Zell-Adhäsion,  
Zell-Zell-Interaktionen, Zell-Erkennung,  
Zell-Differenzierung, Bildung interneuraler  
Verbindungen, Zell-Differenzierung bei der  
Embryo-Entwicklung,  
Blutgruppen-Antigene.



Gangliosid GM1

# Die Biosynthese von N-Acetyl-Neuraminsäure



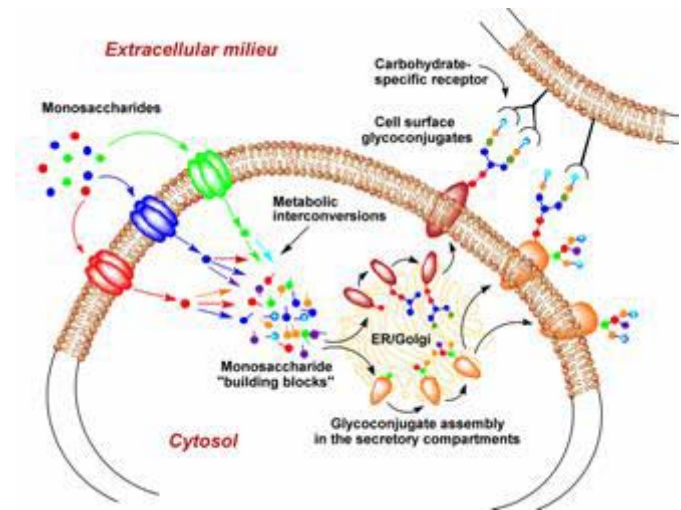
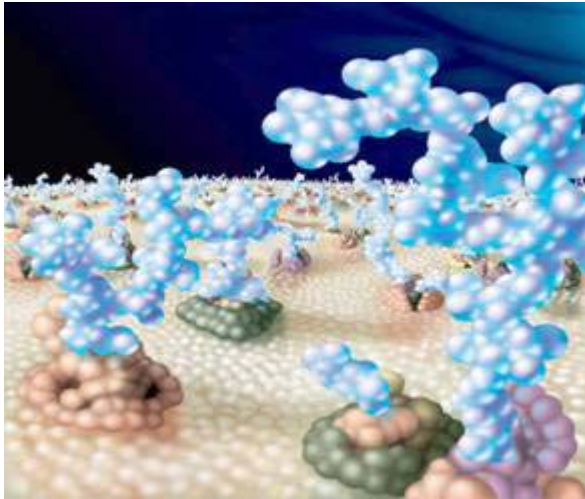




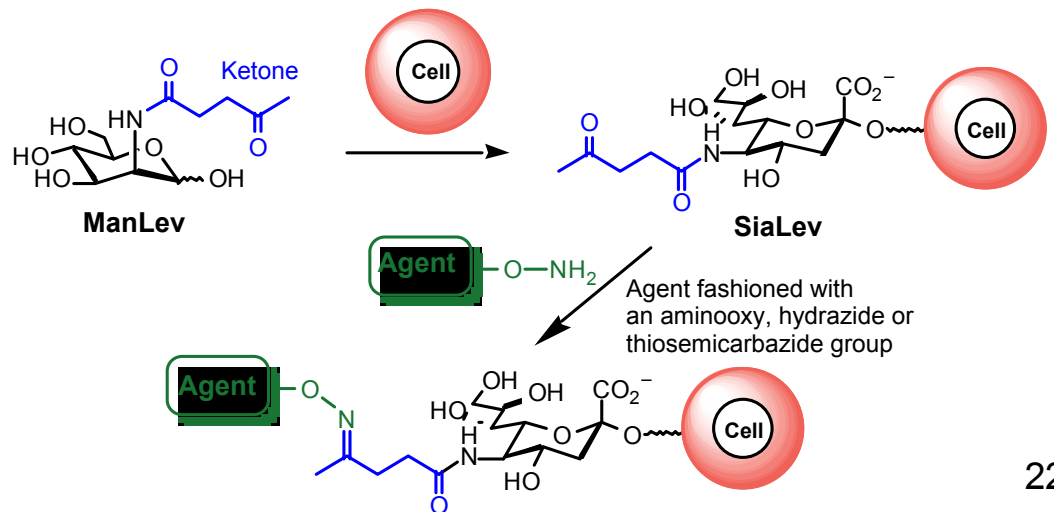
Die folgenden 5 Folien sind eine freundliches Geschenk von Prof. Bertozzi

Ich habe sowohl die Graphiken als auch den Text von Ihrer homepage übernommen.  
Wenn Sie daher von „we“ lesen, ist die Bertozzi-Gruppe gemeint und nicht die von MK.

## Anwendungen von Prof. Bertozzi

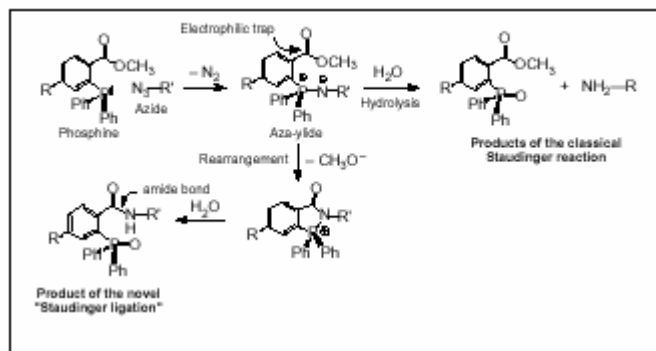


Comparisons have revealed that the human genome is not much larger than those of lower organisms, but vastly more abundant in posttranslational machinery, accounting for numerous complex functions found only in mammals.

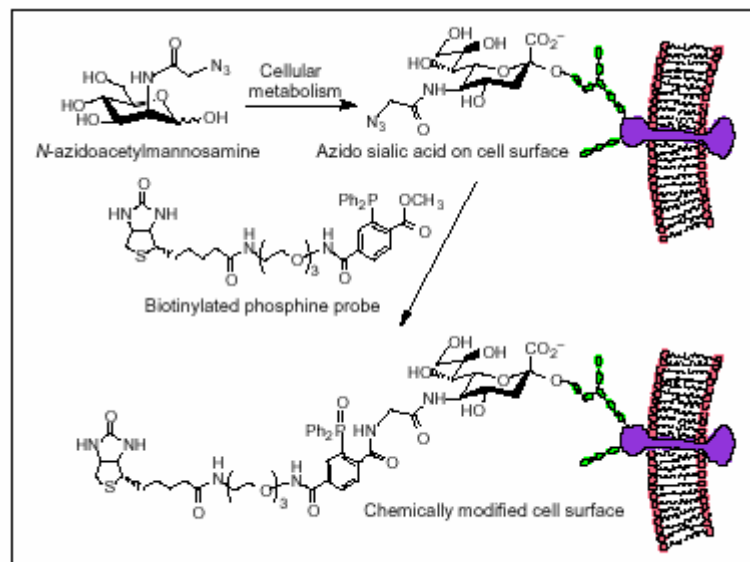


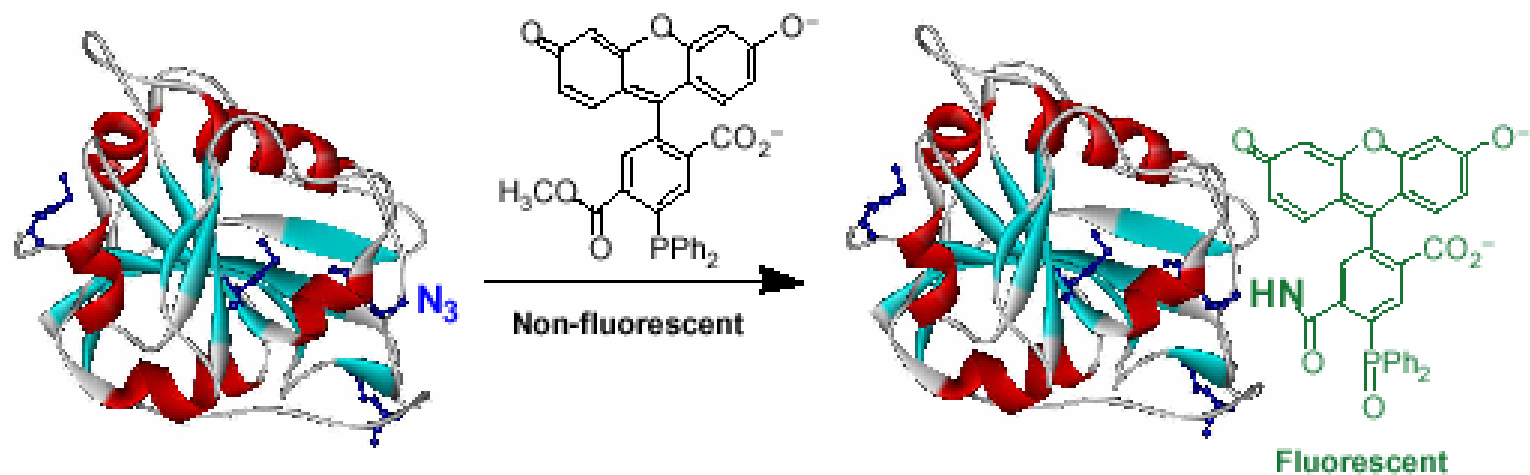
## Other biological applications of the Staudinger ligation

The Staudinger ligation has unique properties that make it well-suited for covalent modification of biomolecules within a complex, highly functionalized environment. The azide and modified triaryl phosphine are both stable in water, but react with each other in a highly chemoselective fashion and are not diverted by any functional groups found in cells or tissues. These qualities have prompted us to expand applications of Staudinger ligation chemistry to include protein tagging within a cellular environment. Azids can be incorporated into proteins via unnatural amino acids and targeted the proteins for covalent modification within cellular lysates.



**Figure 3.** The Staudinger ligation. The reaction is highly chemoselective and can be performed on azido sugars incorporated into cell surface glycans by metabolic processes.





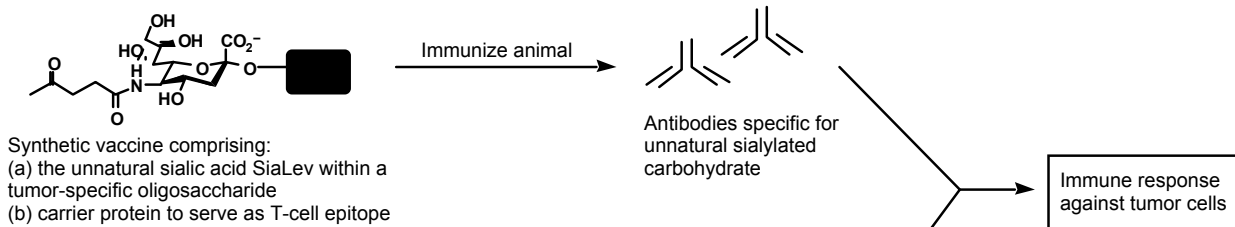
**Figure 4.** Incorporation of azides into proteins via protein expression in the presence of azidohomoalanine and selective protein modification with a pro-fluorescent dye activated by the Staudinger ligation.

# Tumor vaccine therapy

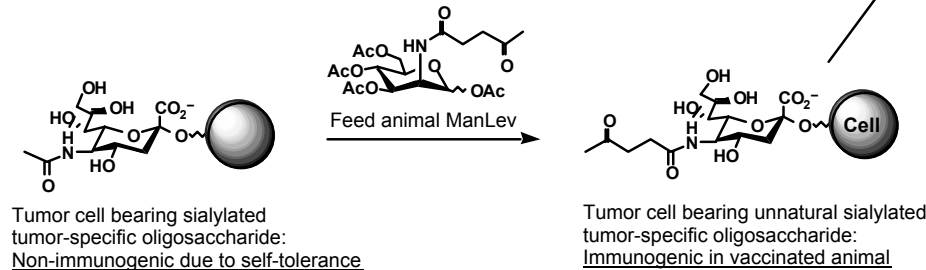
Their close association with malignancy has prompted consideration of tumor-associated oligosaccharides as components of anti-tumor vaccines. However, tumor oligosaccharides themselves usually fail to stimulate an immune response due to immune self-tolerance. We are exploring an approach to tumor immunotherapy that circumvents self-tolerance by creating unnatural modifications to tumor-associated oligosaccharides, rendering them "foreign" and immunogenic.

An immune response to an unnatural variant of a tumor oligosaccharide is engendered by vaccination using a synthetic version of the molecule. The same unnatural modification is induced on tumor cell surfaces by feeding the organism an unnatural metabolic substrate. Once the unnatural epitope is expressed on cells, the immune response is triggered to kill them. The approach is under investigation using the tumor-specific sialyl Tn antigen as a vaccine target.

**A**



**B**



**Figure:** A strategy for tumor vaccine therapy that breaks oligosaccharide self-tolerance through unnatural sialic acid biosynthesis. **A.** The animal is vaccinated with a synthetic glycoconjugate comprising an unnatural sialylated tumor-specific oligosaccharide bound to an antigenic carrier protein. The unnatural sugar structure circumvents self-tolerance, leading to a vigorous antibody response. **B.** A tumor cell bearing a natural oligosaccharide is not normally recognized by the immune system due to self-tolerance. But, exposure to the unnatural metabolic substrate ManLev induces the unnatural modification that renders the oligosaccharide immunogenic in the vaccinated animal. The tumor is then destroyed by the immune system.

## Metabolic disruption of poly- $\alpha$ -2,8-sialic acid, an oligosaccharide associated with metastasis

Finally, we use metabolic processes to reversibly inhibit the cell surface expression of interesting polysaccharide structures. We have focused initially on polysialic acid (PSA), a polymer uniquely found attached to the neural cell adhesion molecule (NCAM). This epitope is found in the normal adult brain in regions of neuronal plasticity, and on numerous tumors where it appears to promote metastasis. The molecular underpinnings of these biological roles are not known. We discovered that unnatural sialic acids generated in cells by metabolism of synthetic precursors are incorporated into the PSA chain where they act as chain terminators during polymer extension.

*N*-Butanoylmannosamine (ManBut), for example, serves as a reversible switch for the expression of PSA on both neurons and tumors, and we are now using this chemical tool to study the effects of PSA expression on cell-cell interactions *in vitro* and *in vivo*. Other metabolic interference projects in the laboratory focus on altering core structures on cell surface glycoconjugates. The salvage pathway for *N*-acetylgalactosamine (GalNAc), for example, can be intercepted with unnatural GalNAc analogs thereby changing the structures and perhaps functions of *O*-linked glycoproteins and chondroitin sulfate proteoglycans.

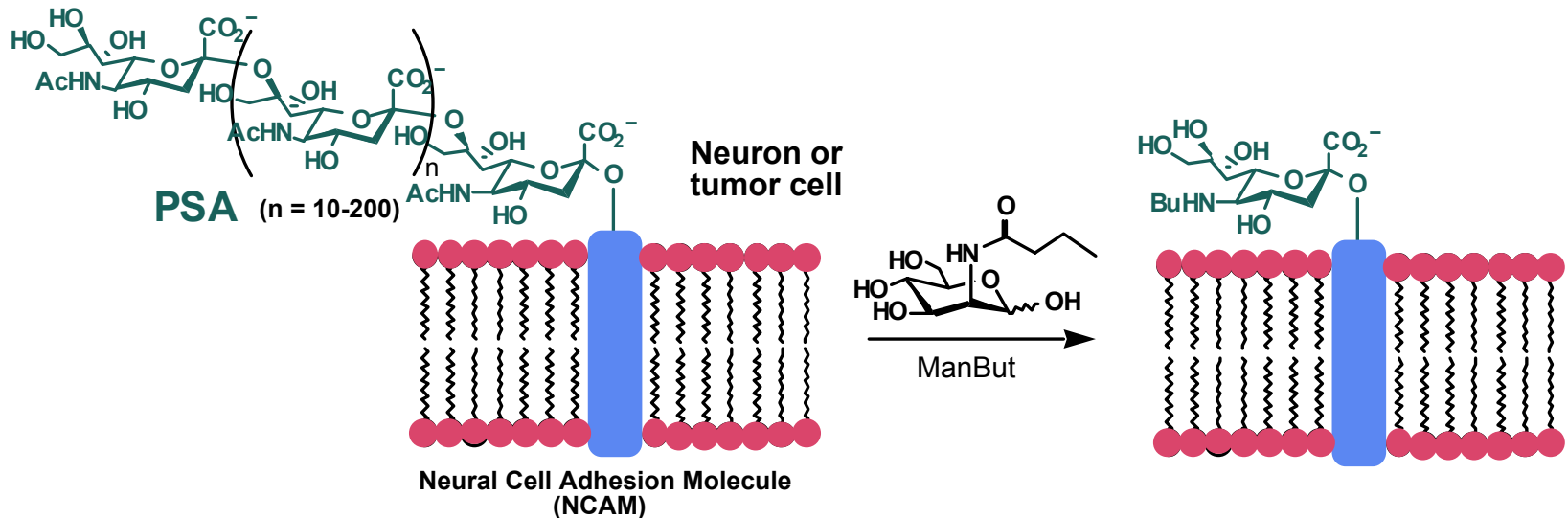


Figure: ManBut is a small molecule switch for polysialic acid expression on tumors and neurons.