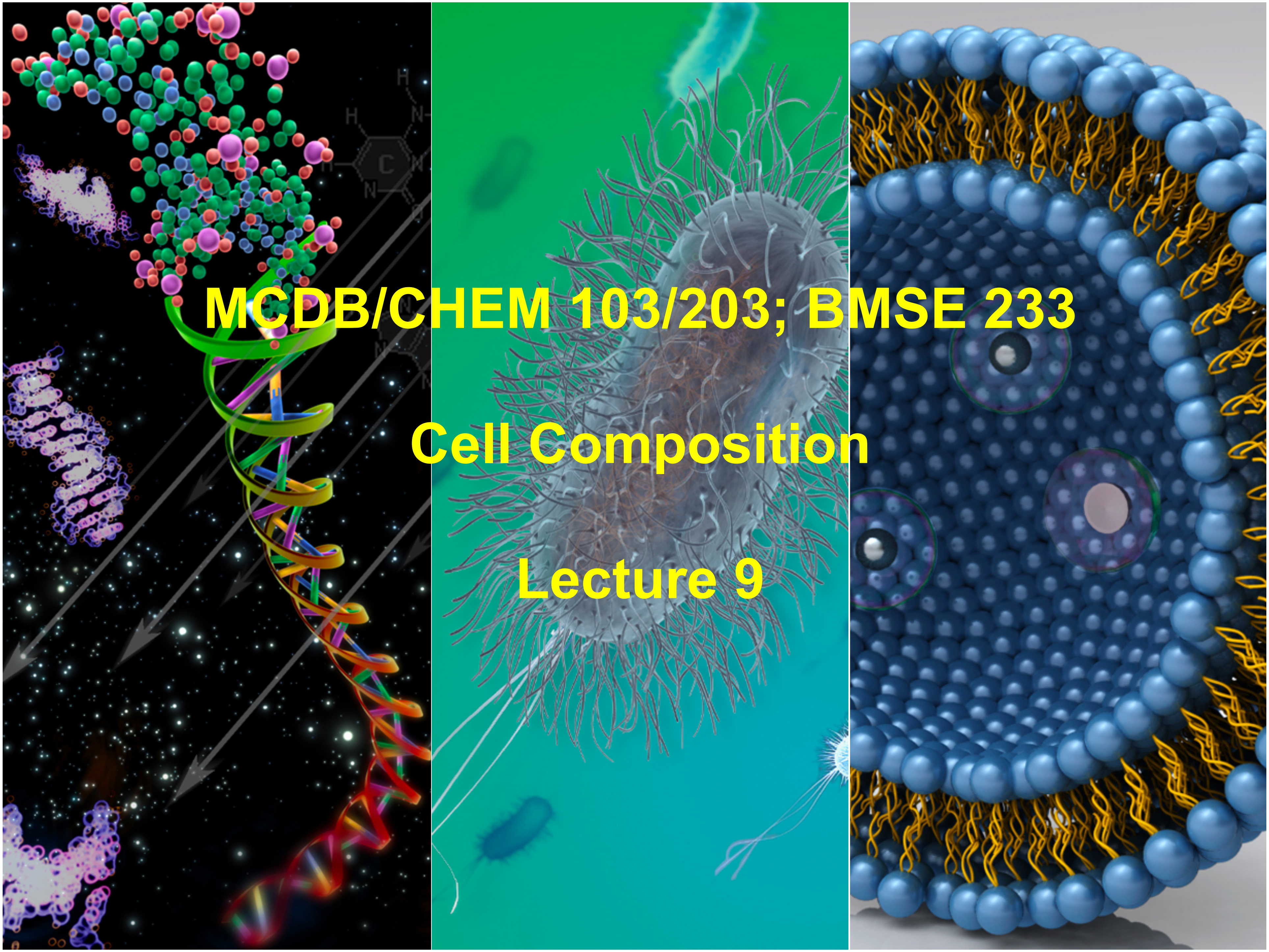
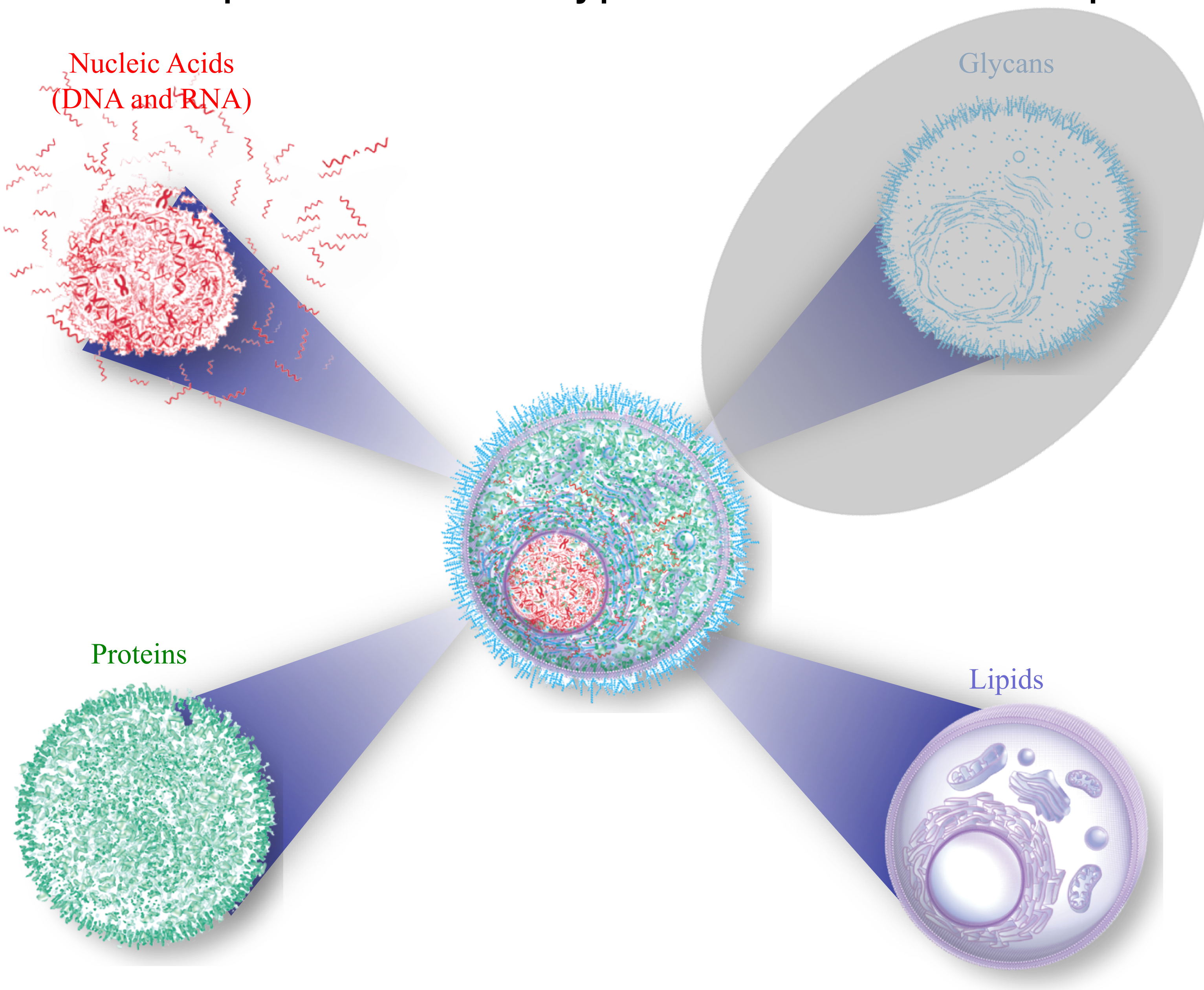


MCDB/CHEM 103/203; BMSE 233

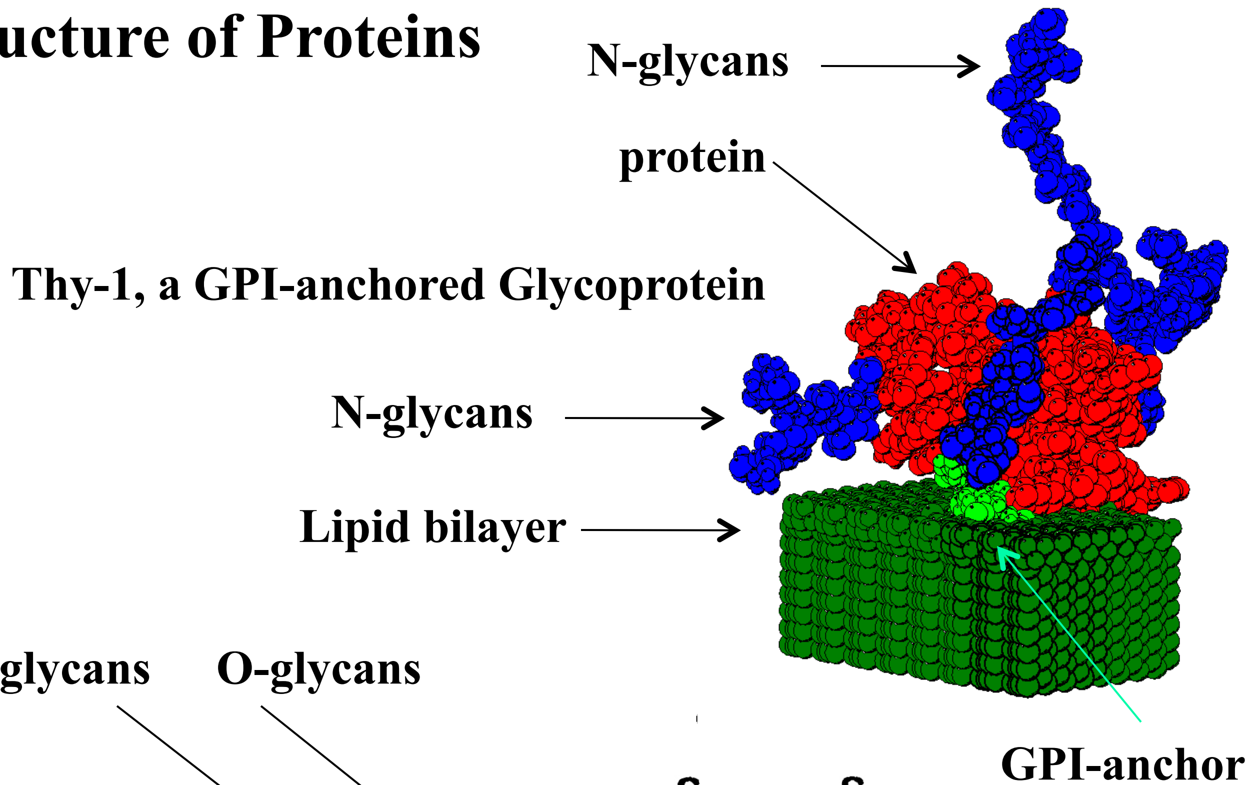
Cell Composition

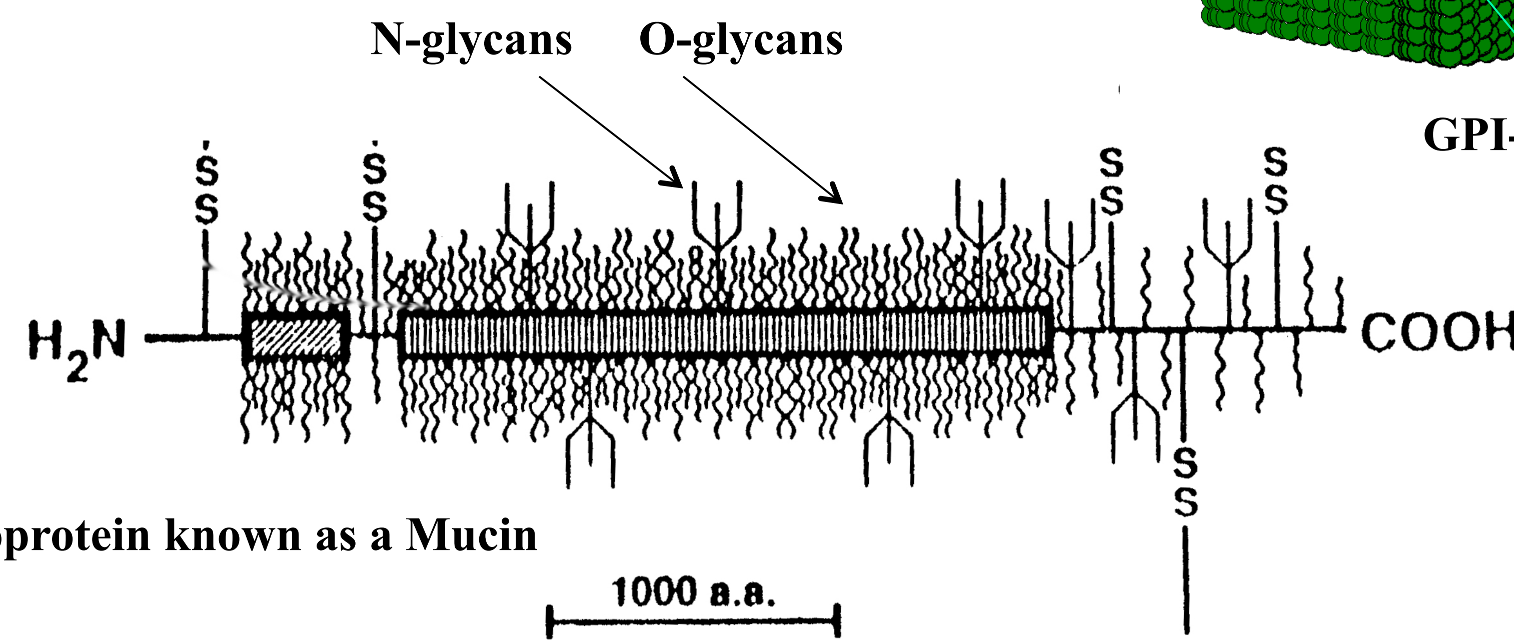
Lecture 9

Cells are Composed of Four Types of Molecular Components



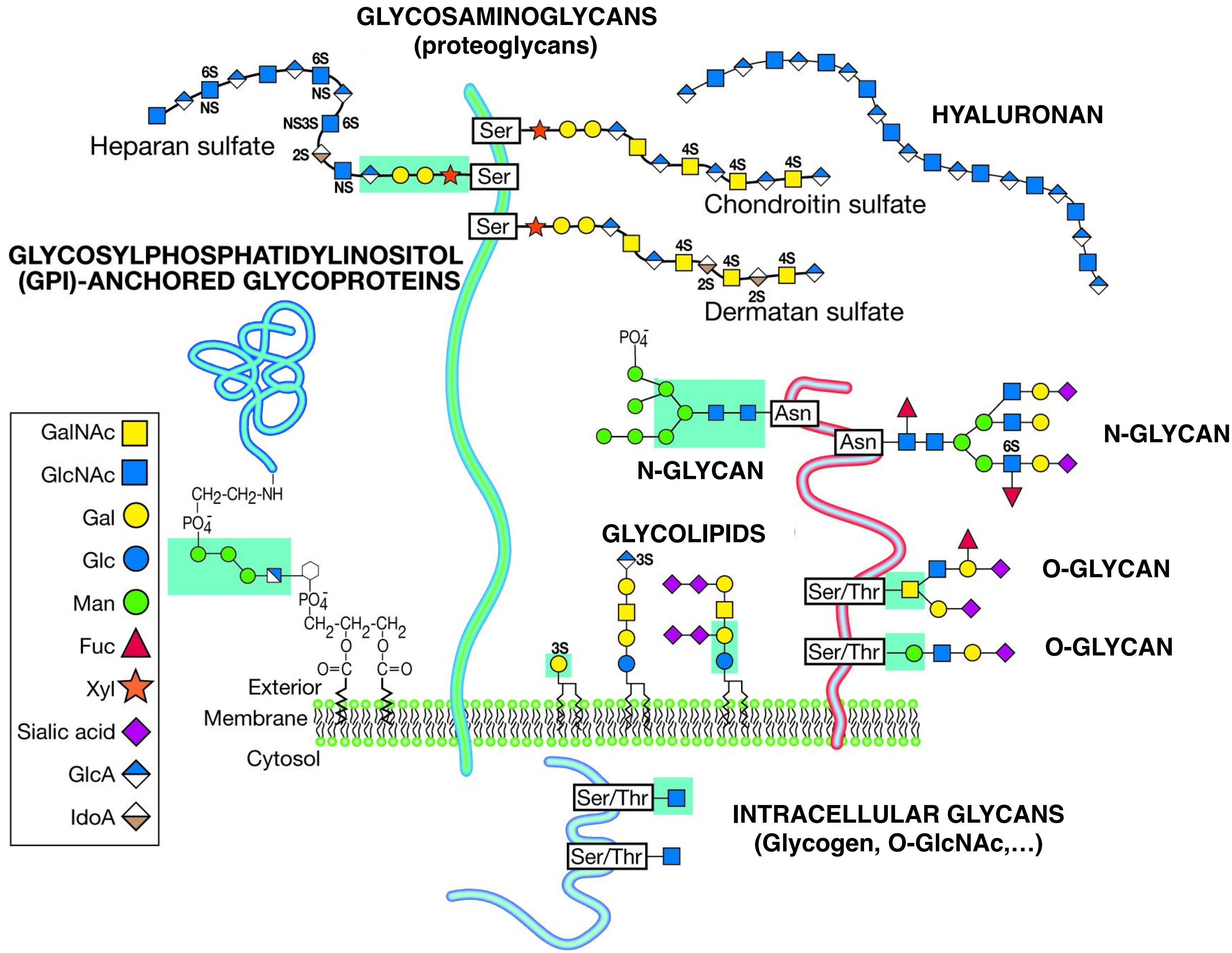
Glycans Can Contribute Extensively to the Mass and Structure of Proteins





Glycoprotein known as a Mucin

Common Classes of Vertebrate Glycans



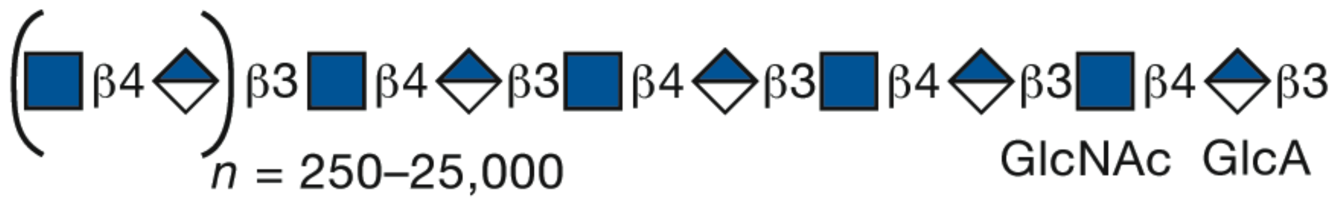
Saccharides Can Be Modified in Glycans by Glycan-Modifying Enzymes

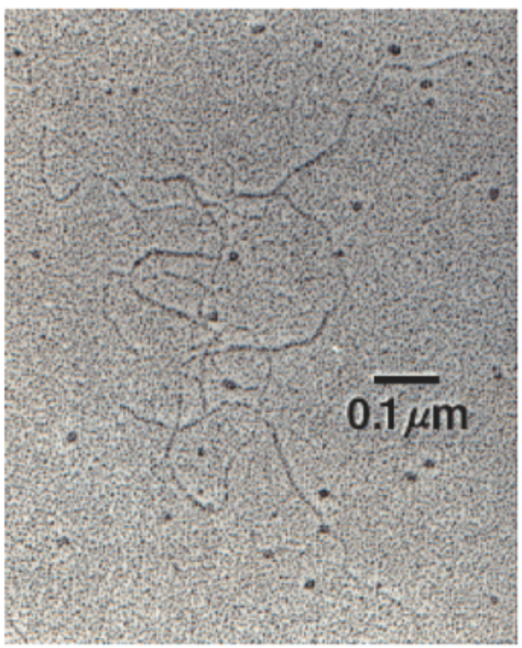
Phosphoadenosine-phosphosulfate (PAPS) + Glycan acceptor	Sulfotransferase →	Sulfated glycan + 3’5’-ADP
Acetyl-coenzyme A + Glycan acceptor	Acetyltransferase →	Acetylated glycan + CoA
S-Adenosylmethionine (SAM) + Glycan acceptor	Methyltransferase →	Methylated glycan + S-Adenosyl-homocysteine
Nucleotide sugar + Glycan/protein acceptor	Phosphotransferase →	Sugar-P-glycan/protein + Mononucleotide
Phosphoenolpyruvate + Glycan acceptor	Pyruvyltransferase →	Pyruvylated glycan + Phosphate
Phosphotidylethanolamine + Glycan acceptor	Ethanolamine phosphate transferase →	Ethanolamine P-glycan + Diacylglycerol

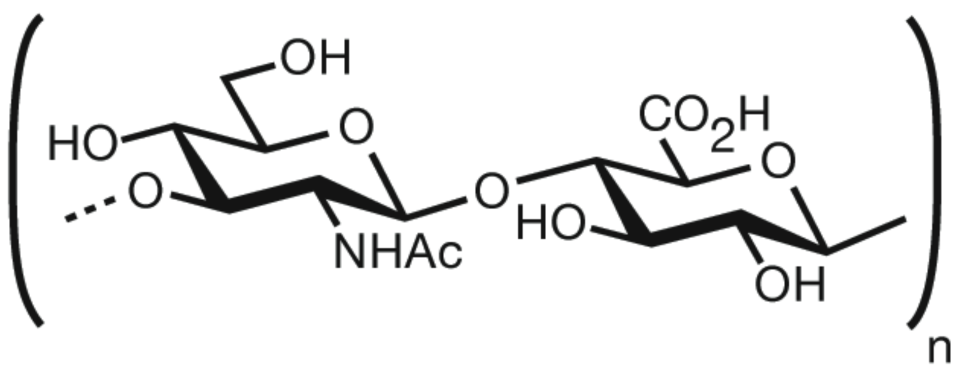
Glycans:

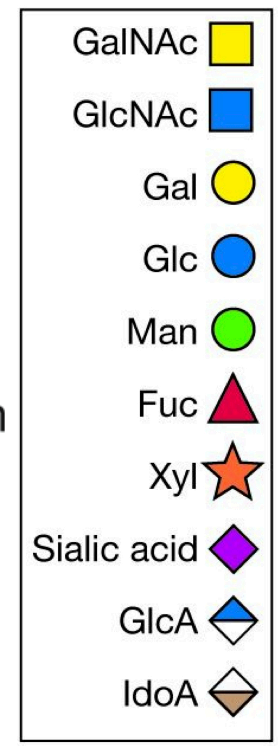
Hyaluronan

Hyaluronan Consists of a Repeating Disaccharide Composed of N-Acetylglucosamine and Glucuronic Acid



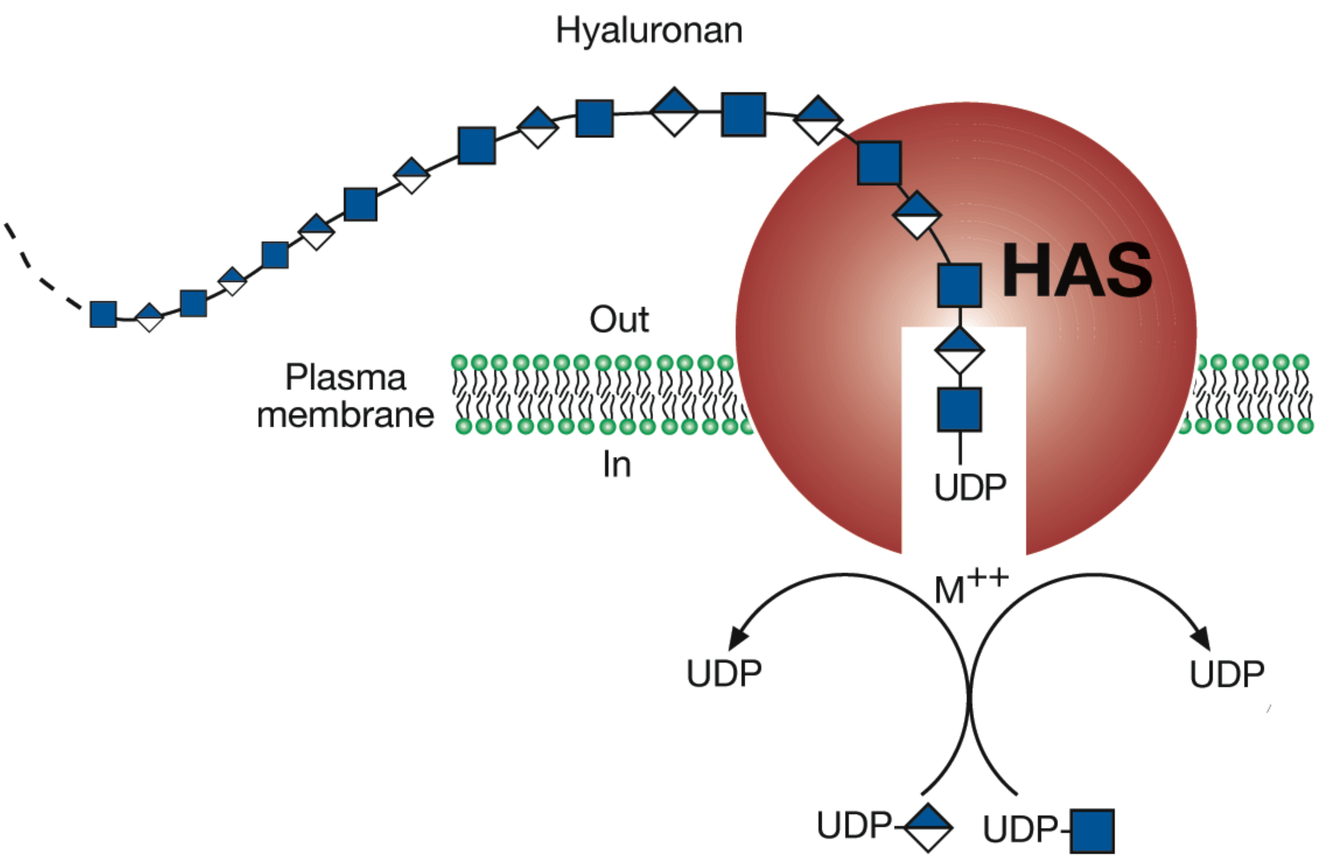


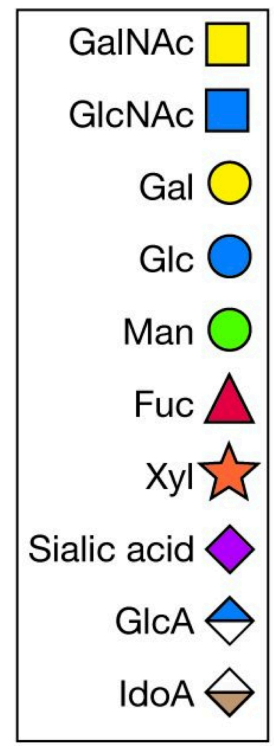




Gel-like, hydration and molecular depot functions

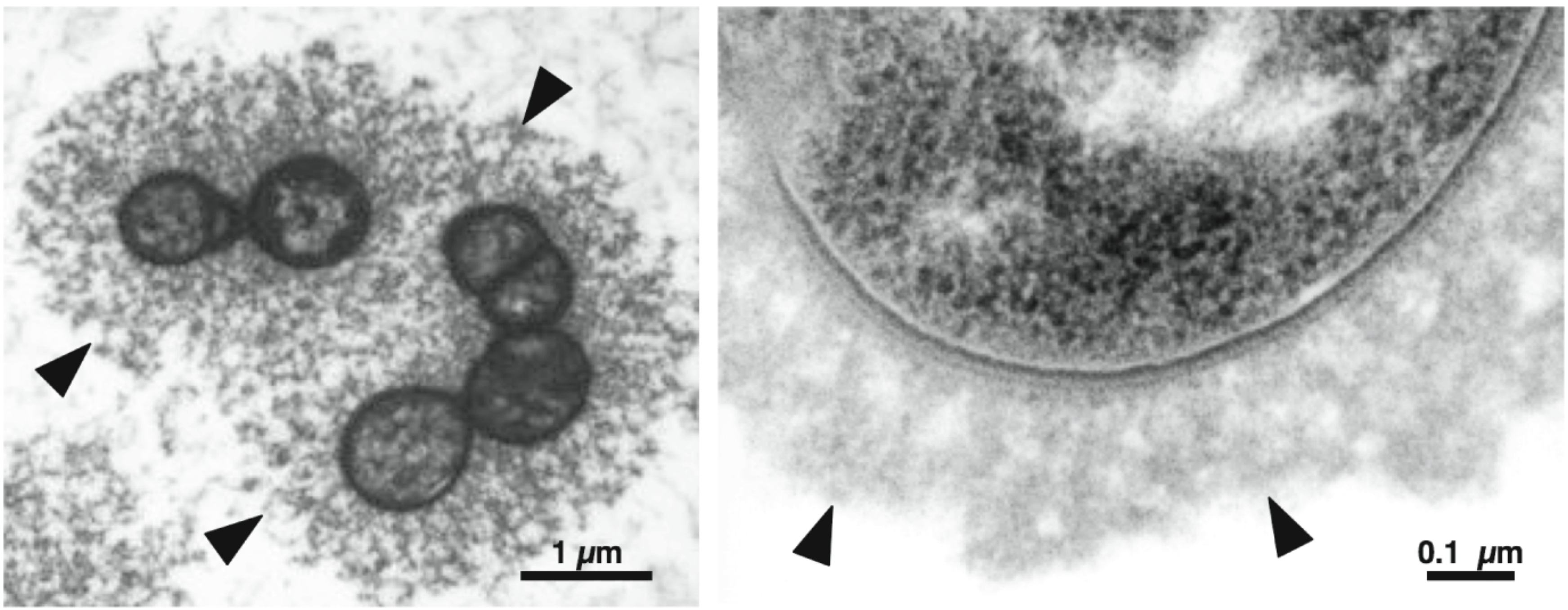
Hyaluronan Biosynthesis





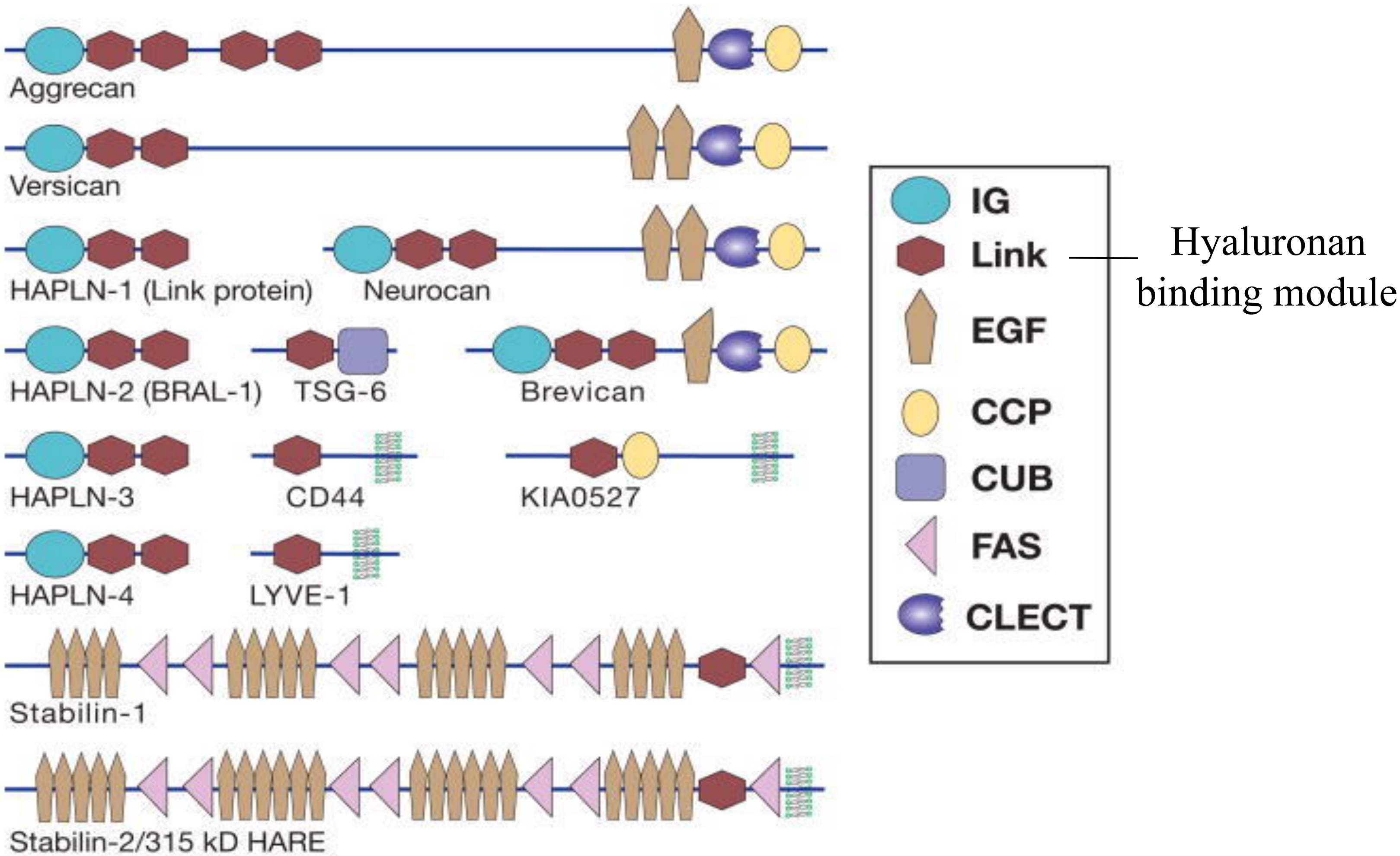
HAS1, HAS2, and HAS3 in mammals.
Unusual and still mysterious biosynthetic process at the plasma membrane.

The Hyaluronan Capsule of *Streptococcus zooepidemicus*



Hyaluronan binds to various extracellular molecules

Examples of Hyaluronan Binding Proteins/Lectins in Animals



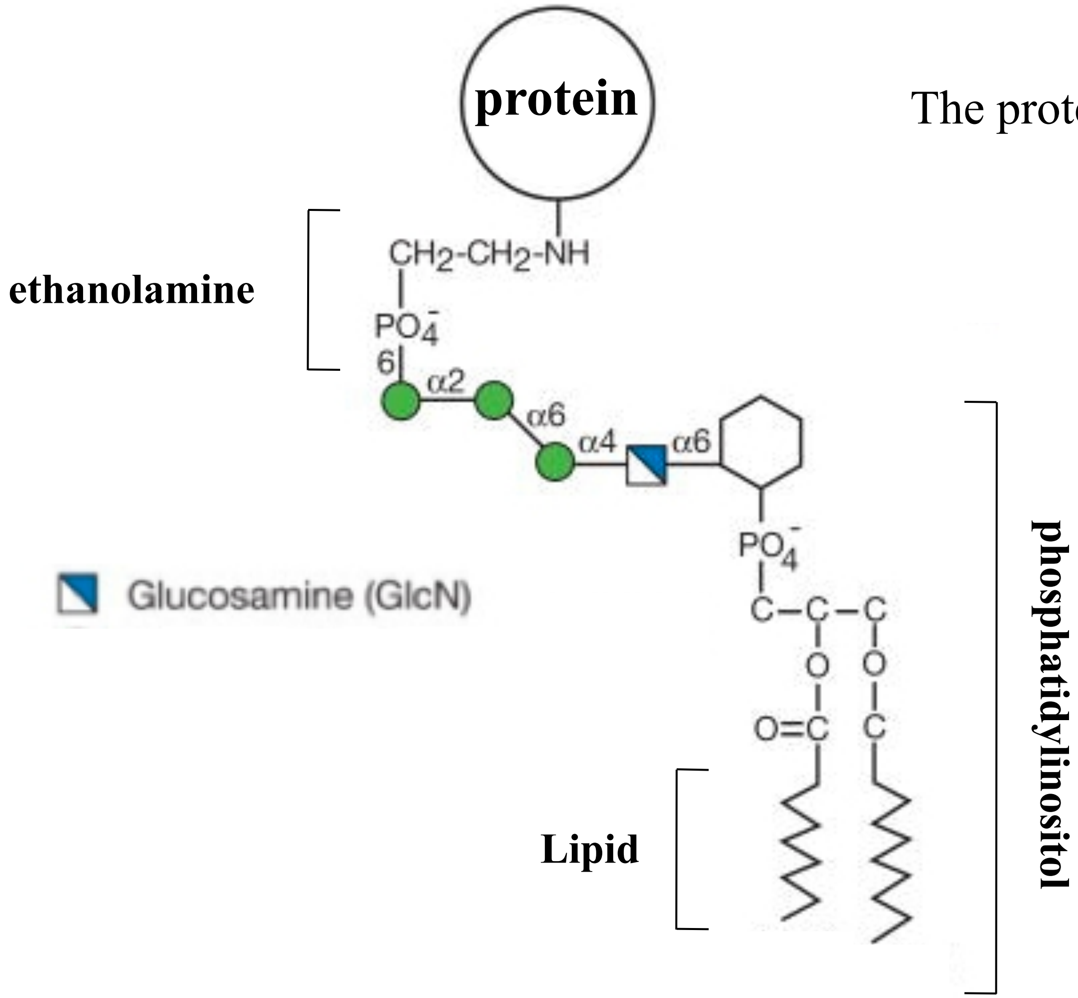
Modular organization of the 'link' module superfamily of hyaluronan-binding proteins. These proteins contain one or more link modules that bind to hyaluronan.

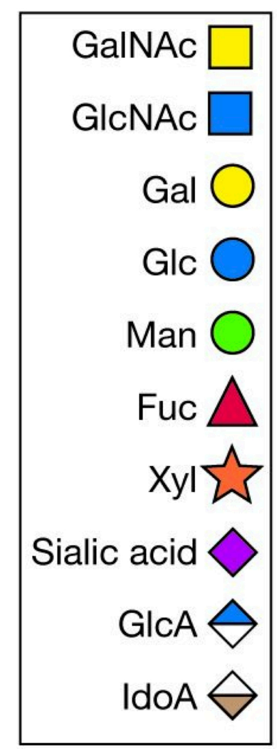
Glycans:

GPI Anchors

General Structure of GPI Anchors

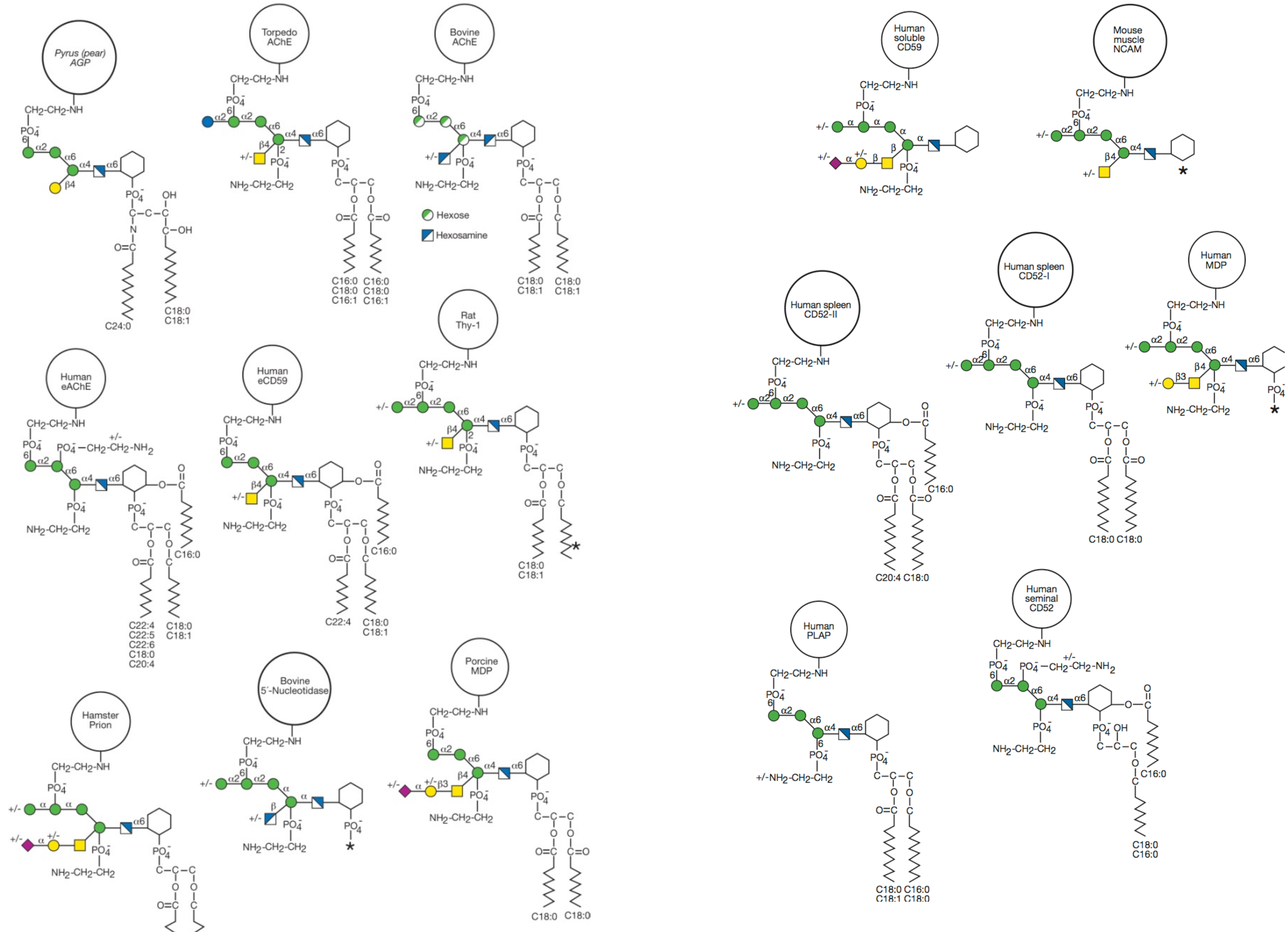
The protein may also be glycosylated





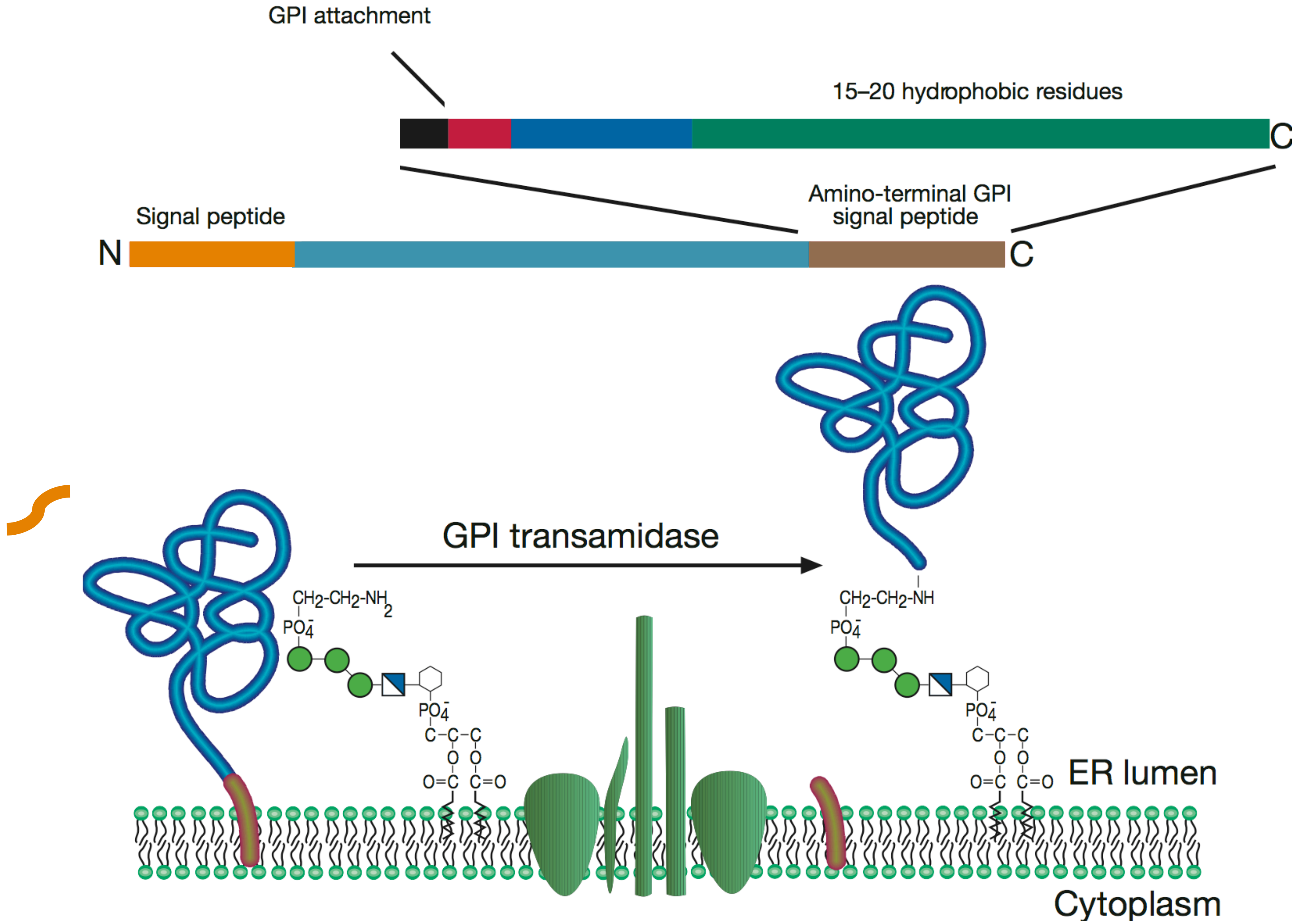
GlcN in GPI anchors is produced by de-acetylation of GlcNAc after GlcNAc is linked to phosphatidylinositol

Varied Structures of GPI anchors
(mammalian examples shown, different from prokaryotes)



(*) unknown lipid type or chain length

Features of GPI-Anchored Proteins and their Processing by GPI Transamidase



GPI Addition Sites Do Not Have a Precise Canonical Amino Acid Sequence in Proteins: Examples of Carboxyl-Terminal Sequences Signaling the Addition of GPI Anchors

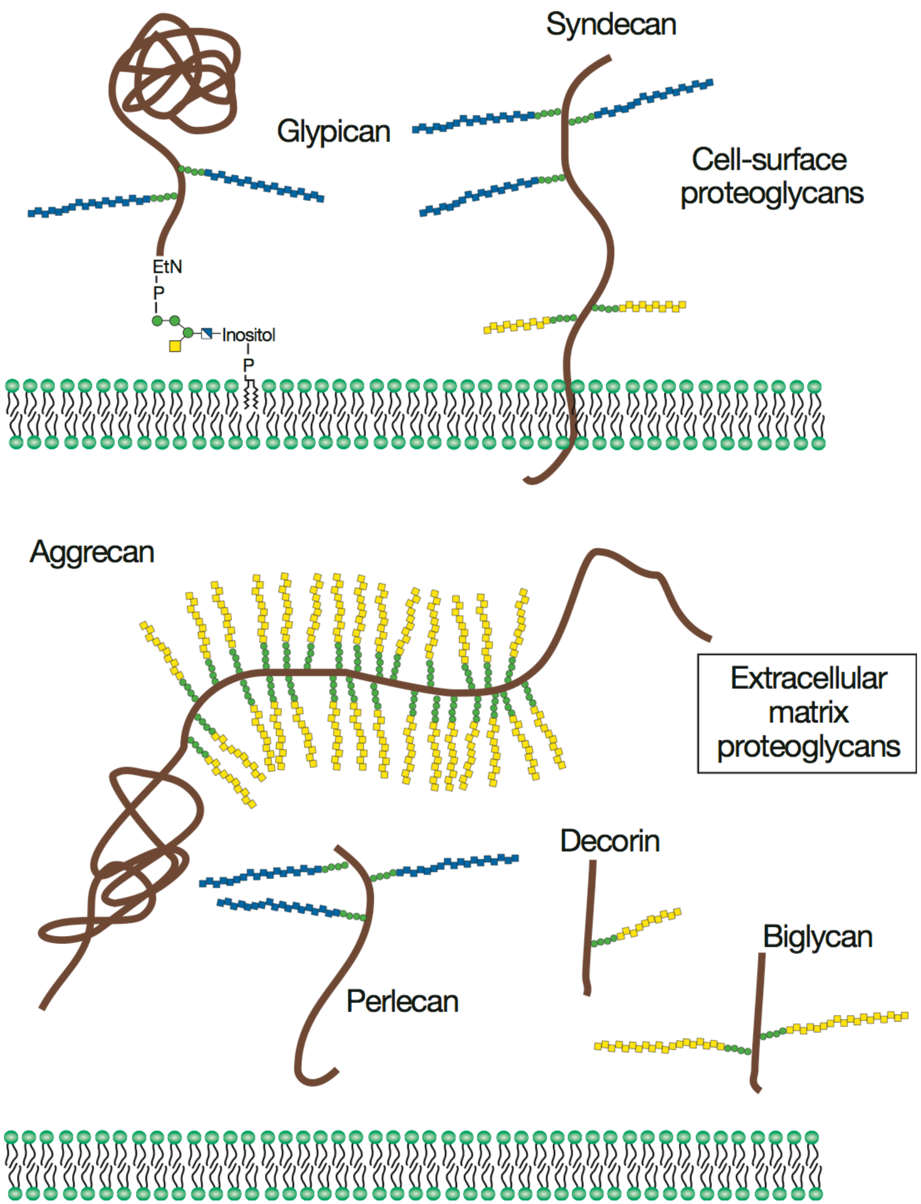
Protein	GPI signal sequence	
Acetylcholinesterase *(Torpedo)*	NQFLPKLLNATA	**C** DGELSSSGTSSSKGIIFYVLFSILYLIFY
Alkaline phosphatasee *(placenta)*	TACDLAPPAGTT	**D** AAHPGRSVVPALLPLLAGTLLLLETATAP
Decay accelerating factor	HETTPNKGSGTT	**S** GTTRLLSGHTCFTLTGLLGTLVTMGLLT
PARP *(T. Brucei)*	EPEPEPEPEPEP	**G** AATLKSVALPFAIAAAALVAAF
Prion protein *(hamster)*	QKESQAYYDGRR	**S** SAVLFSSPPVILLISFLIFLMVG
Thy-1 *(rat)*	KTINVIRDKLVK	**C** GGISLLVQNTSWLLLLLLSLSFLQATDFISI
Variant surface glycoprotein *(T. Brucei)*	ESNCKWENNACK	**D** SSILVTKKFALTVVSAAFVALLF

Boldfaced amino acid is the site of attachment of the GPI. Sequence to the right of the space is cleaved from the protein by the transpeptidase upon anchor addition.

Glycans:

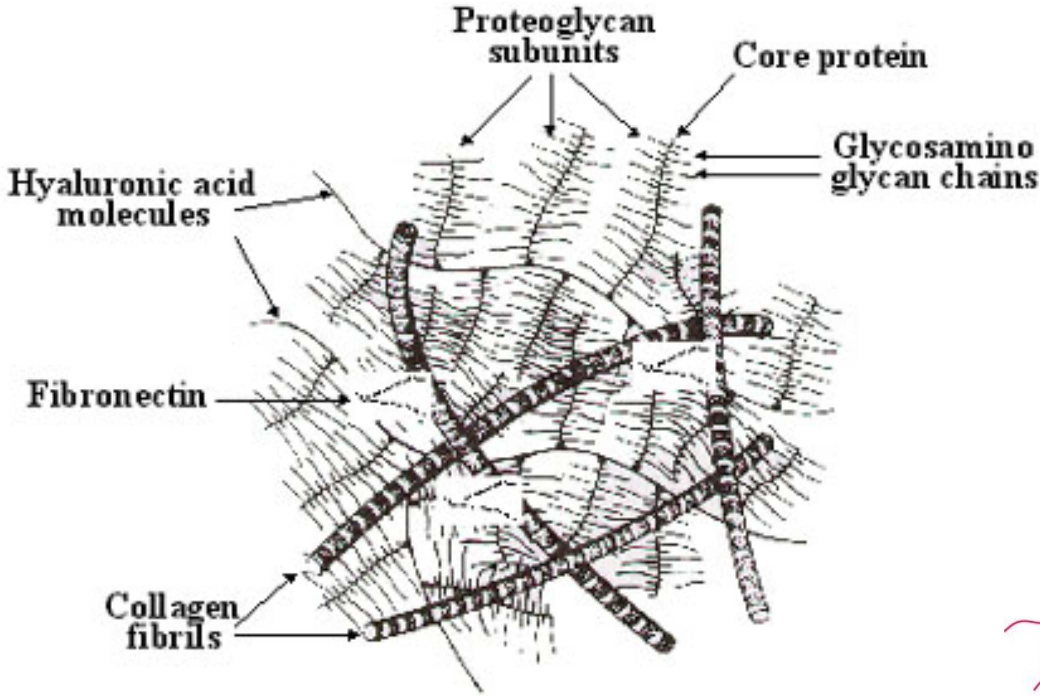
Glycosaminoglycans (Proteoglycans)

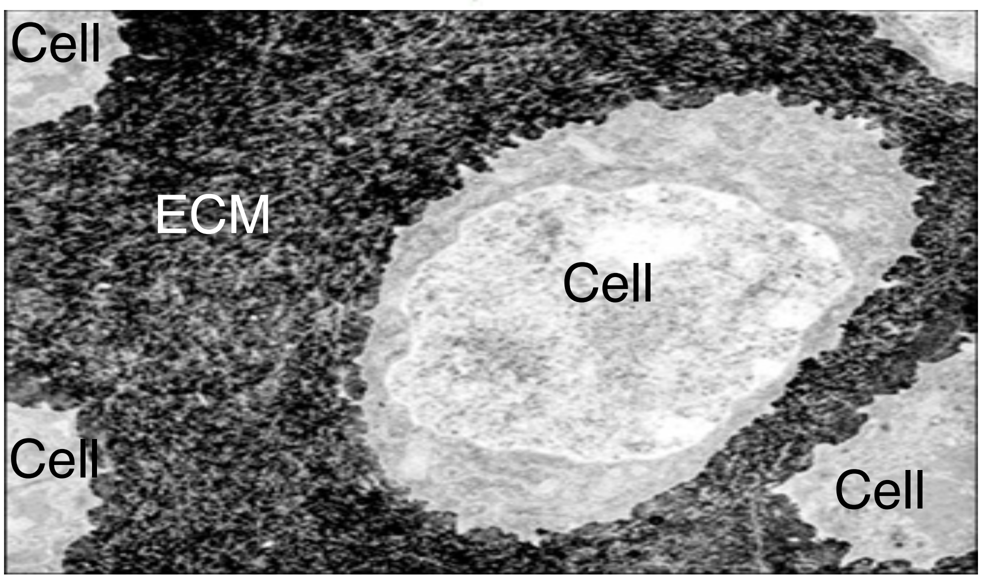
Proteoglycans Consist of a Protein Core and One or More Covalently Attached Glycosaminoglycan Chains

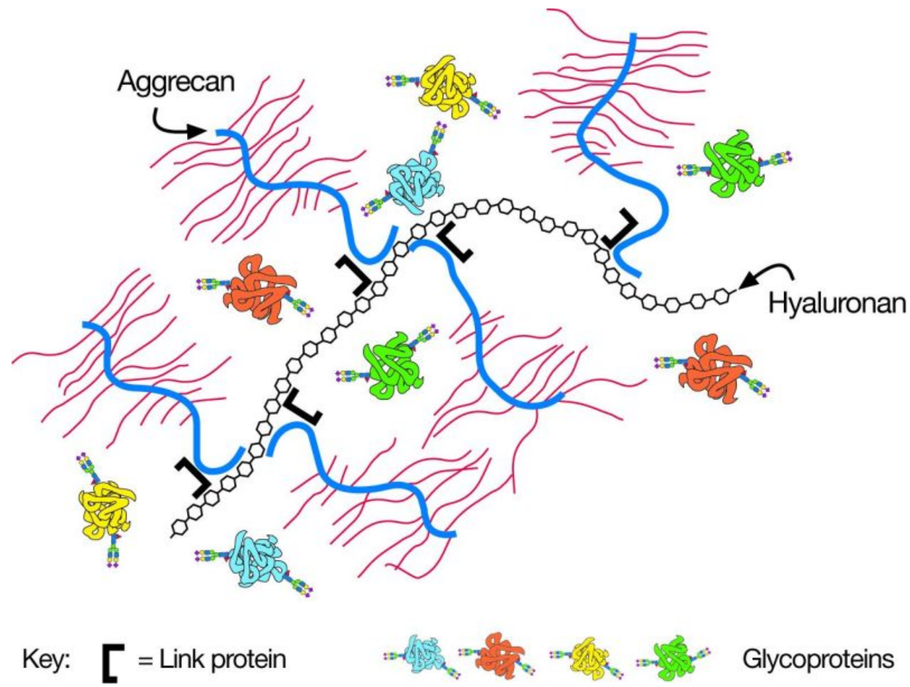


No canonical amino acid consensus sequence exists for glycosaminoglycan addition to proteins

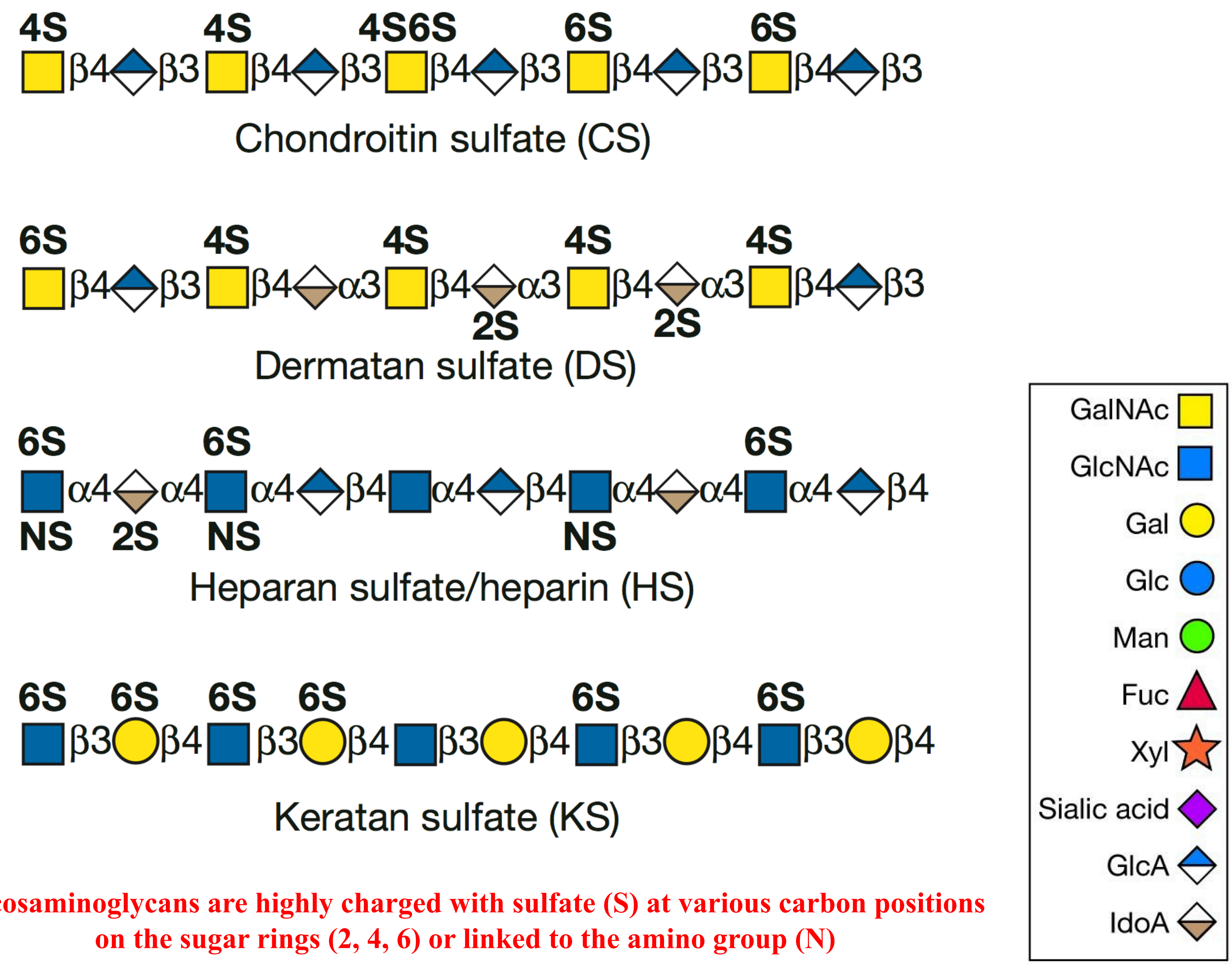
Glycosaminoglycans, Hyaluronan, Collagen and Other Components Make up the Extracellular Matrix (ECM)







Glycosaminoglycans Consist of Repeating & Different Disaccharides



Glycosaminoglycans are highly charged with sulfate (S) at various carbon positions on the sugar rings (2, 4, 6) or linked to the amino group (N)

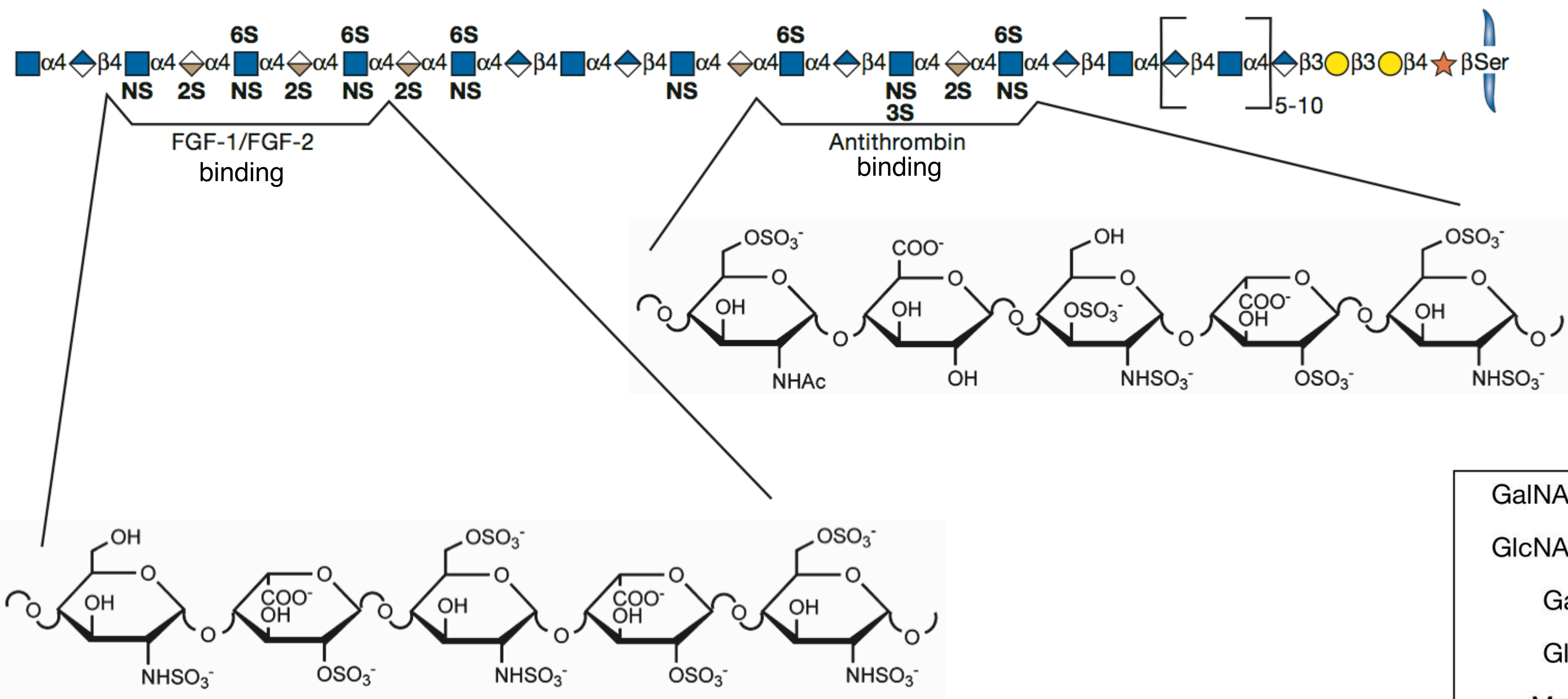
The Distinctions Between Heparin and Heparan Sulfate

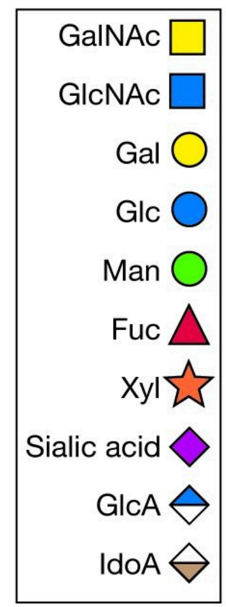
Major differences between heparin and heparan sulfate

Characteristics	Heparan sulfate	Heparin
Soluble in 2 M potassium acetate (pH 5.7,4°C)	yes	no
Size	10–70 kD	7–20 kD
Sulfate/hexosamine ratio	0.8–1.8	1.8–2.6
$GlcNSO_3$	40–60%	≥80%
Iduronic Acid	30–50%	≥70%
Binding to antithrombin	0–0.3%	~30%
Site of synthesis	virtually all cells	connective-tissue-type mast cell

Heparin is a potent anti-coagulant and the most successful drug of all time, prescribed in metric tons each year.

The Heparan Sulfate Glycosaminoglycan Chain Consists of Different Domains that Vary in the Extent of Modification by Sulfation and Epimerization

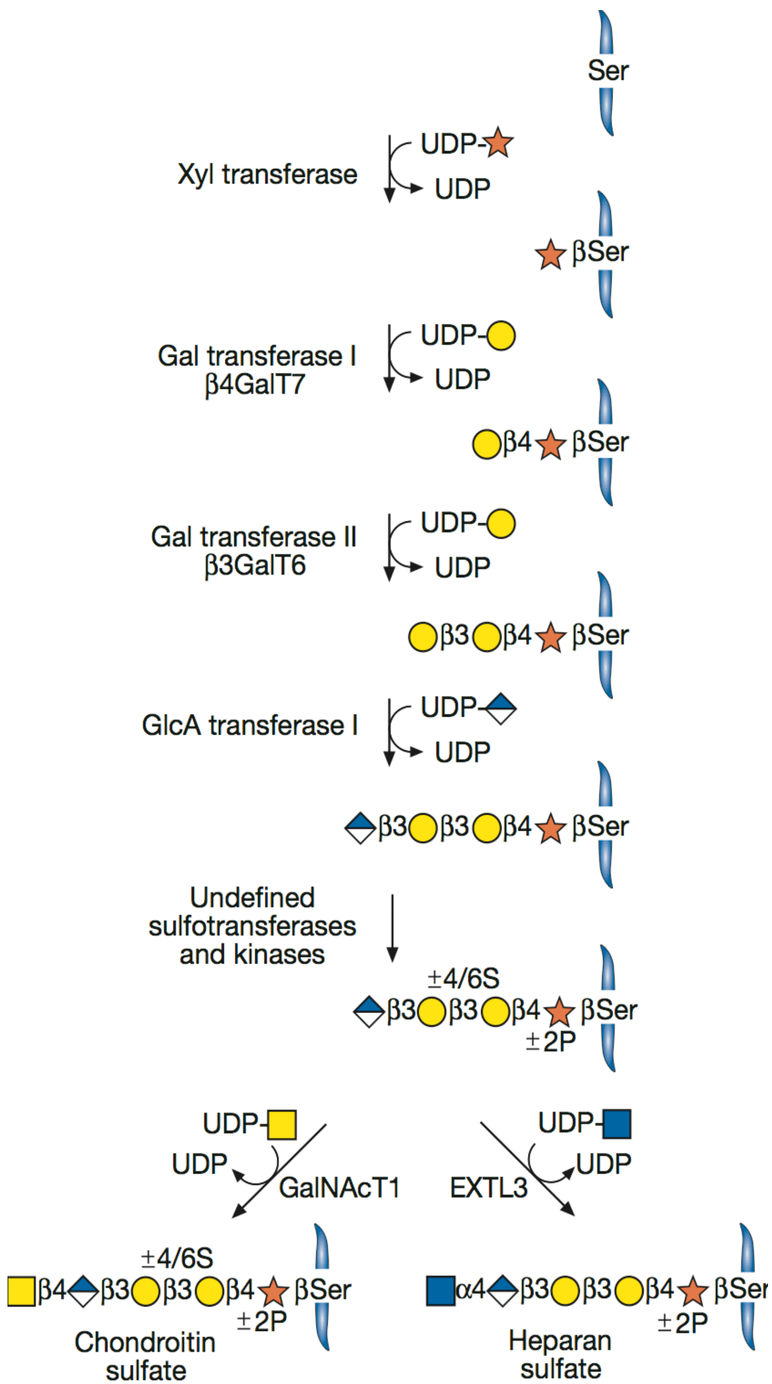


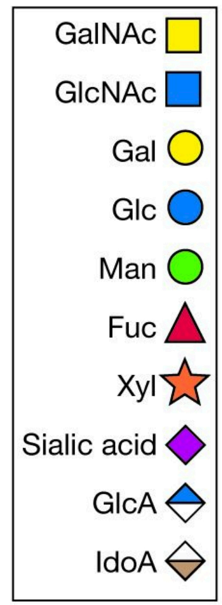


These modifications confer binding activity to various molecules and proteins.

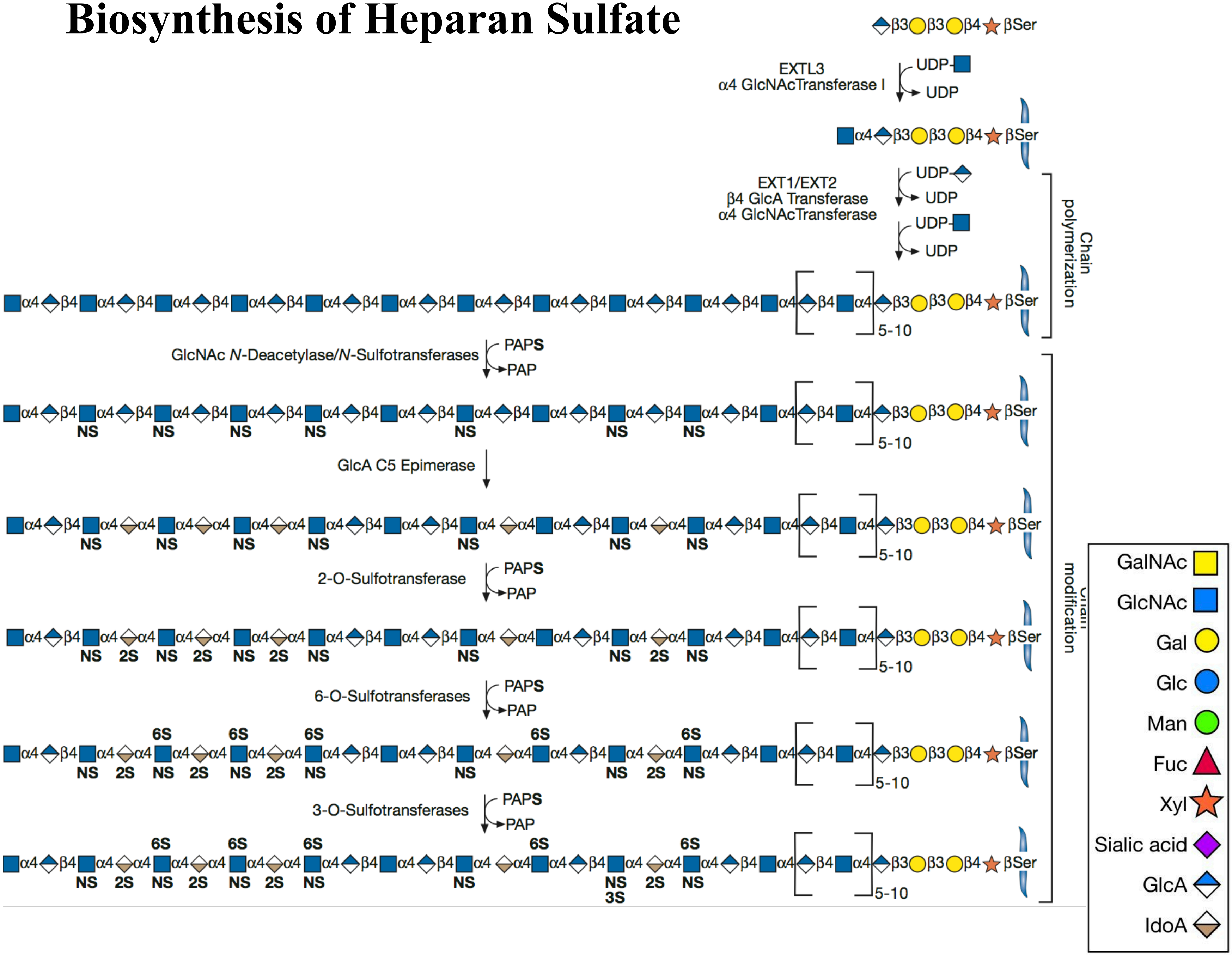
Glycosminoglycans are like a saccharide-sulfate “bar-code” for biological function.

Biosynthesis of Glycosaminoglycans is Initiated by the Formation of a Common Tetrasaccharide

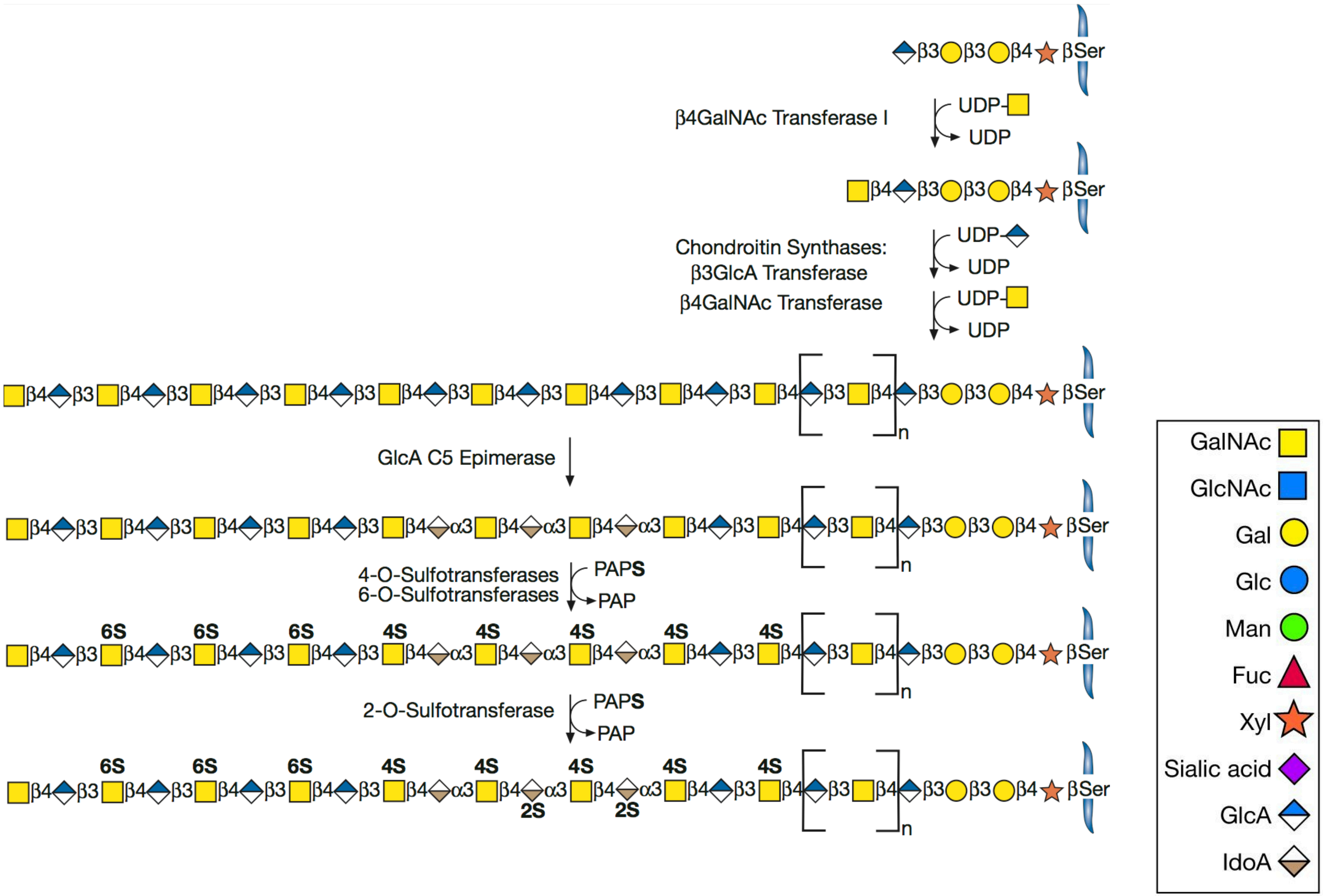




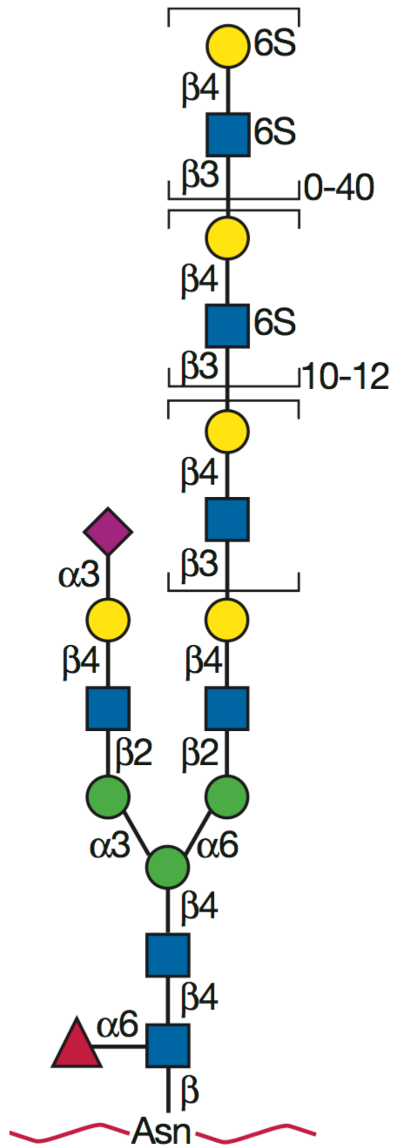
Biosynthesis of Heparan Sulfate



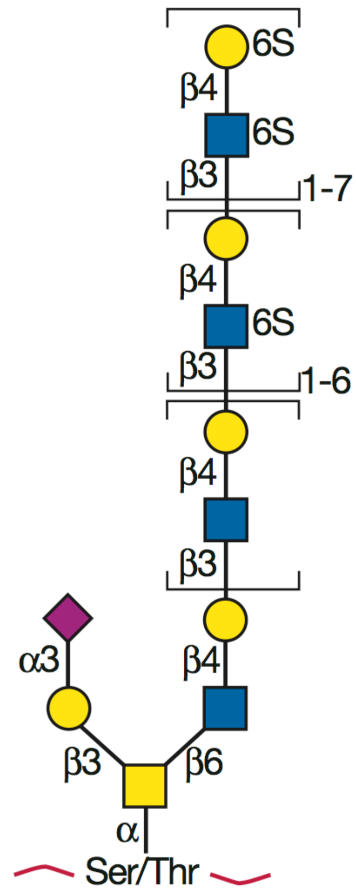
Biosynthesis of Chondroitin Sulfate / Dermatan Sulfate Glycosaminoglycans



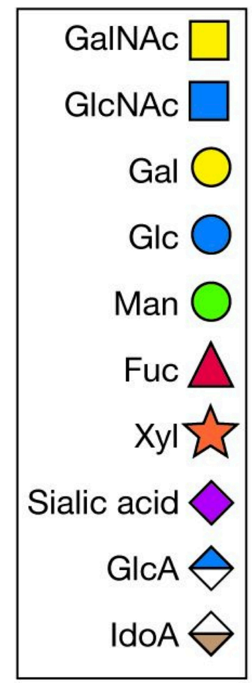
Two Types of Keratan Sulfate: Among N-Glycans and O-Glycans



Keratan sulfate
Type 1



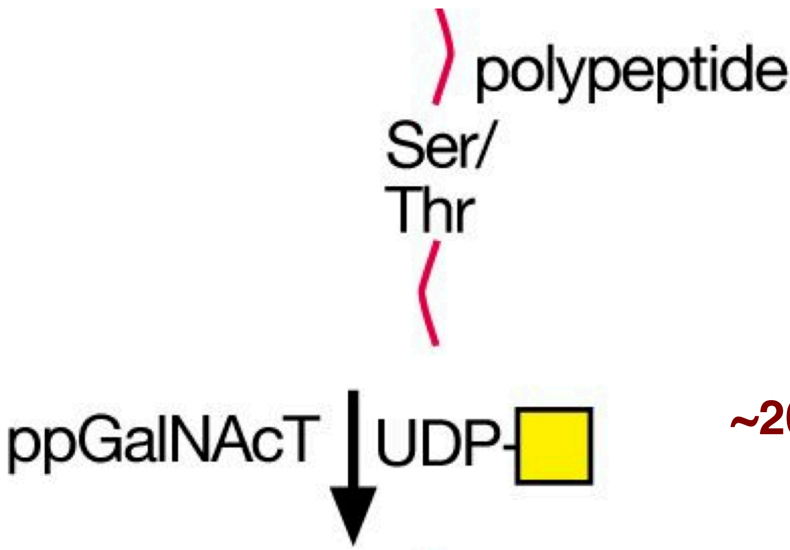
Keratan sulfate
Type 2



Glycans:

O-Glycans

Initiation of Protein O-Glycosylation (O-Glycan Formation) by Multiple Protein Polypeptide (pp) GalNAc Transferases

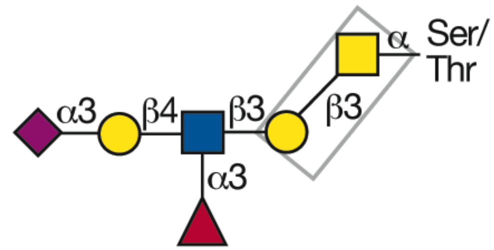


~20 ppGalNAcT enzymes/genes from worms to humans!

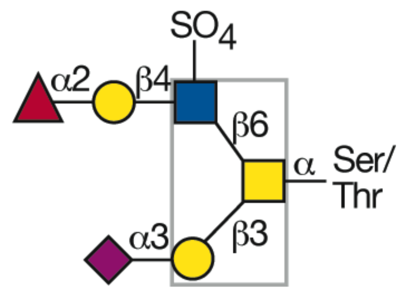
No precise canonical amino acid consensus sequence exists for O-glycan addition to proteins, predictive algorithms are about 80%% accurate.

O-Glycans of Different Core Structures/Classifications

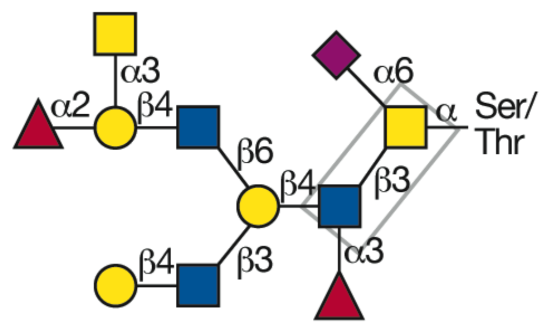
Core 1



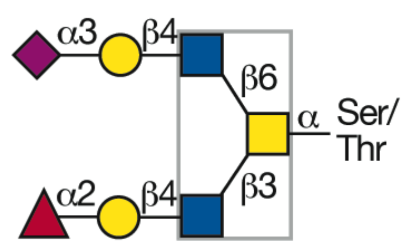
Core 2

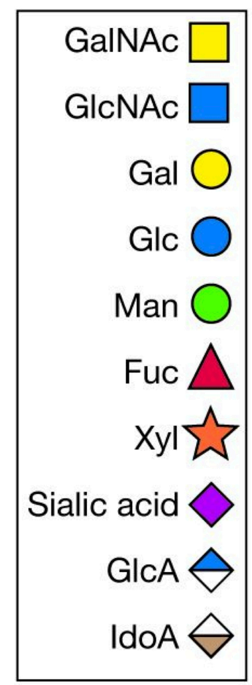


Core 3

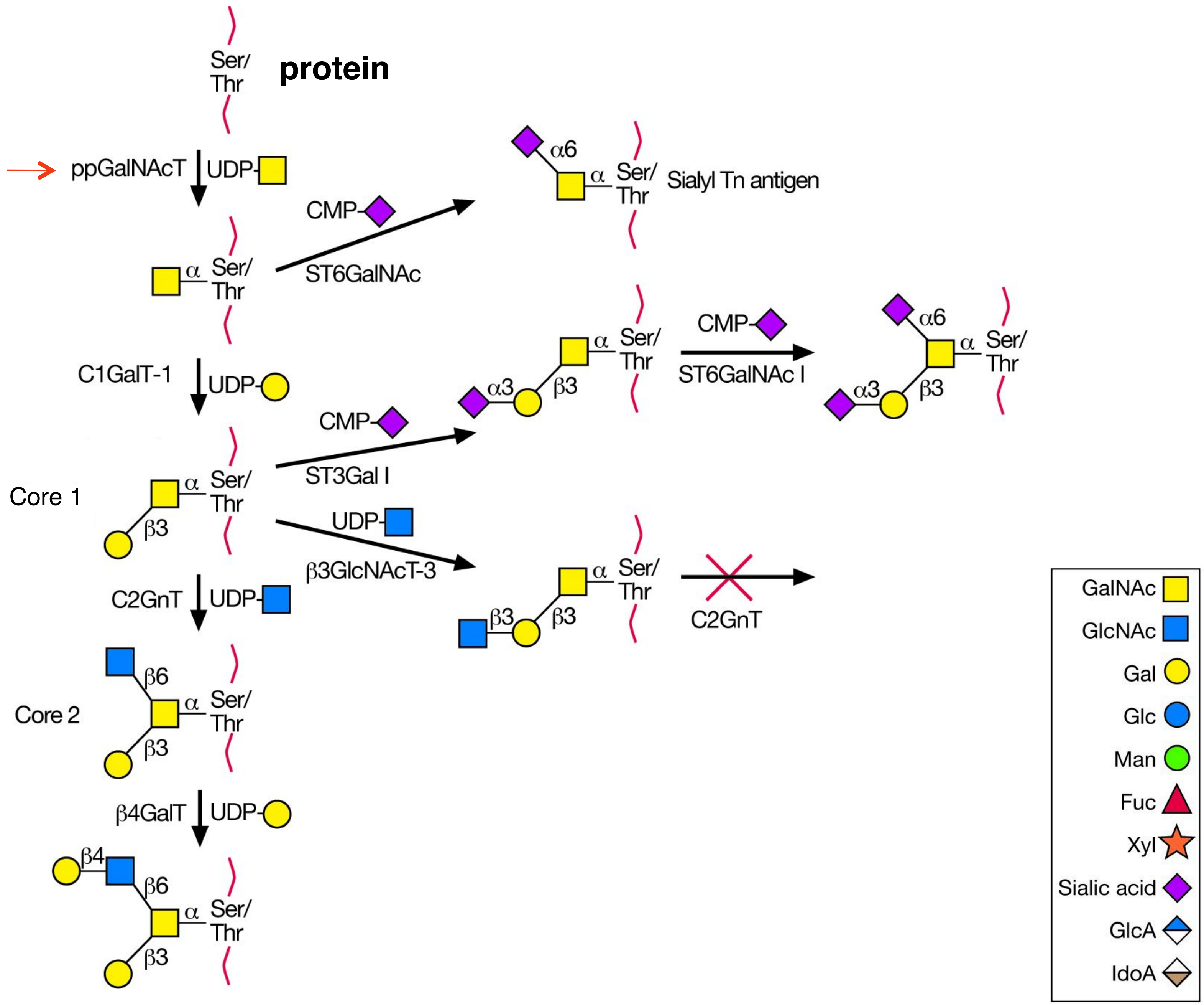


Core 4

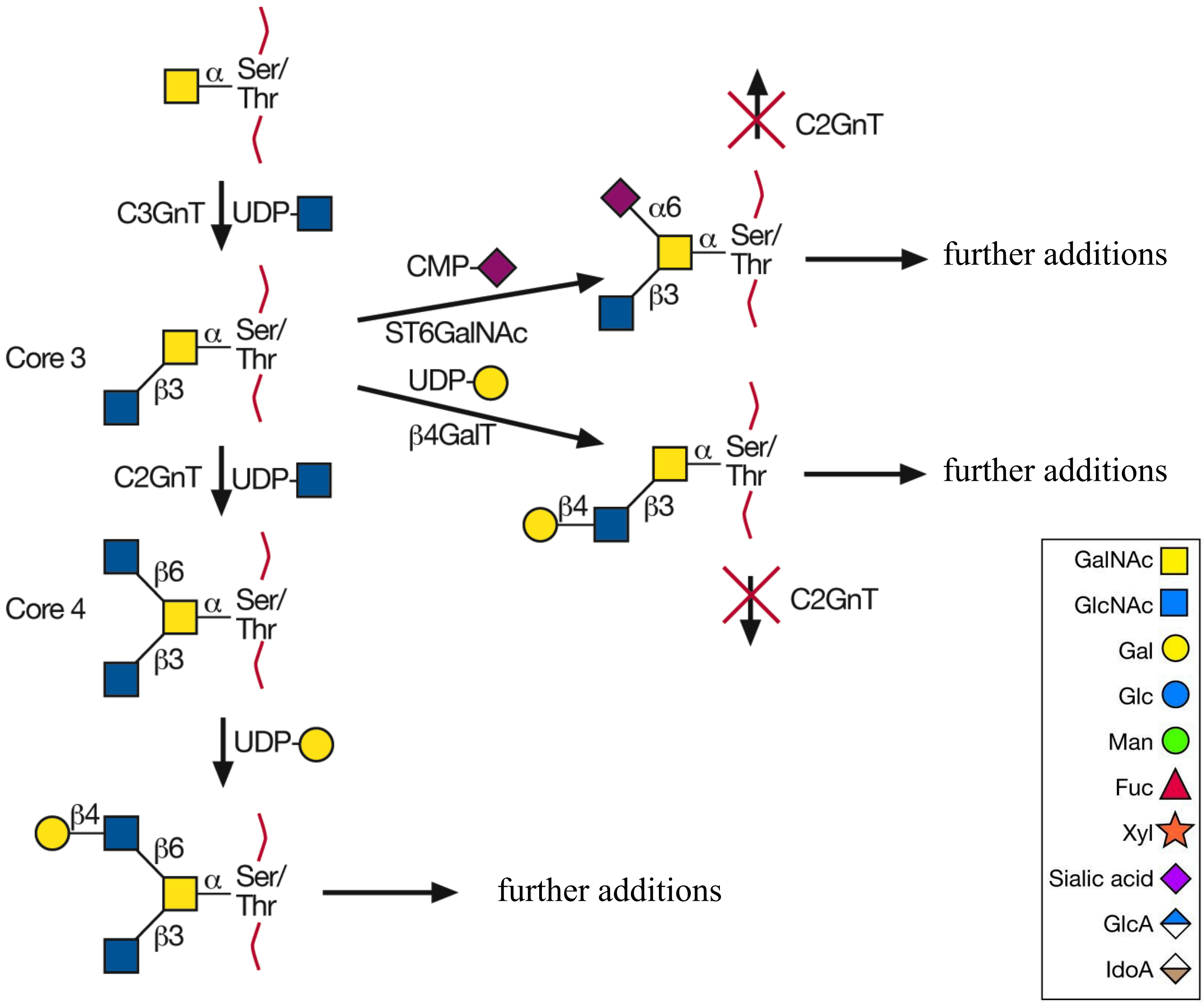




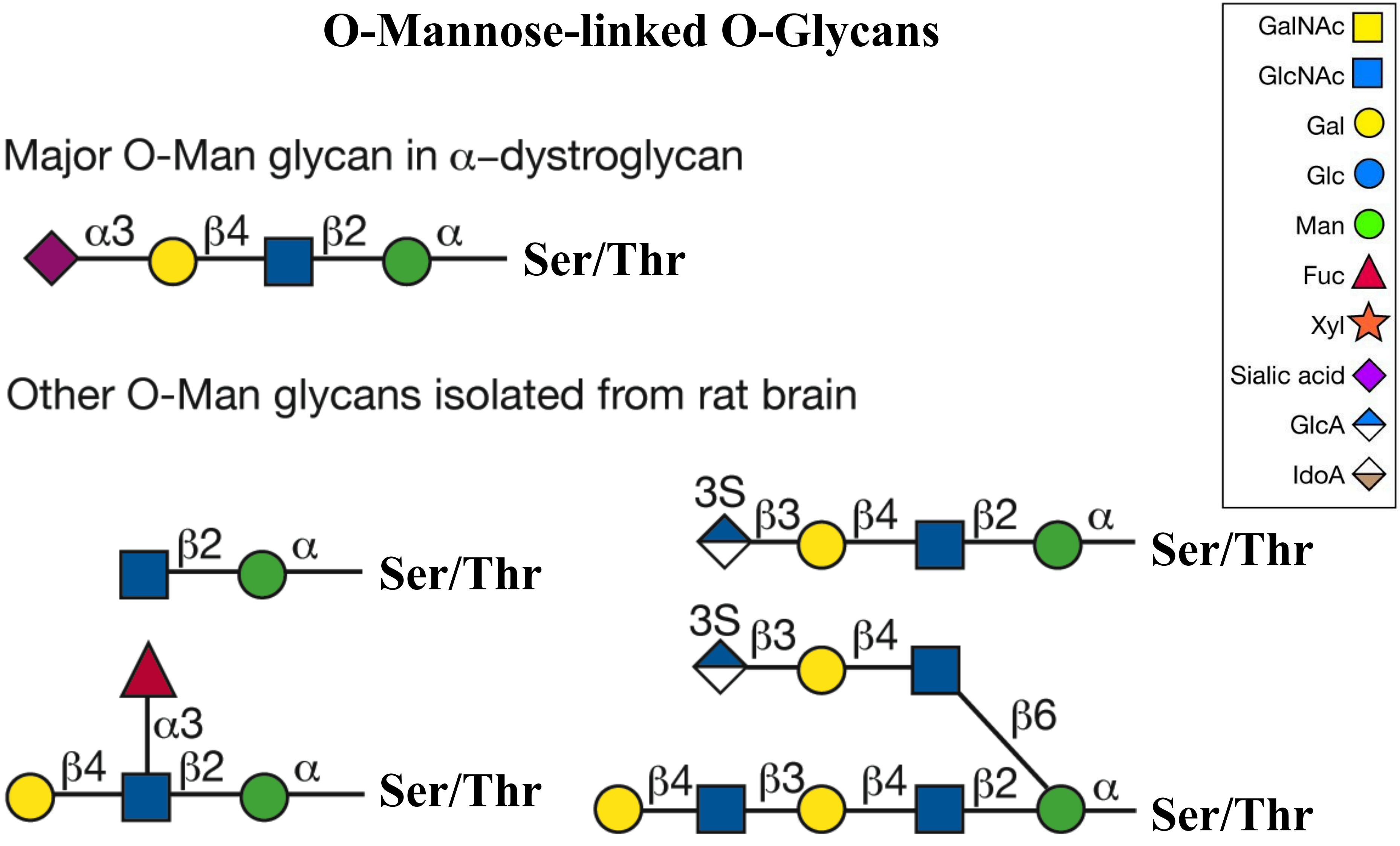
Biosynthesis of Core 1 and Core 2 O-Glycans



Biosynthesis of Core 3 and Core 4 O-Glycans



O-Mannose-linked O-Glycans



Disruption of O-mannose-linked O-glycan biosynthesis is the basis of several muscular dystrophies