Biochemistry Fundamentals

CARBOHYDRATES

This is a Tutorial/Silent Lecture

What is a Tutorial/Silent Lecture?

a sequence of "slides" formatted to guide you through the exploration/study of the topic

you are the main actor in this active learning experience
think of it as working with a tutor without having to pay for it

as the slide sequence unfolds, you will get opportunities to engage with the material

- **by thinking about/answering questions**,
  (my answer is always provided on the next slide).
- **by completing a "short assignment"**
  (it never will take more than a few minutes, if at all that long),
- by **watching a short video/clip**
  (the embedded links will take you to my YouTube@VsBCBMB channel;
  key moments are captured as still and are shown in the slide-deck, in case you don't want to watch the videos)

of course, you can skip the active learning aspect and look at the answers right away.

Why Give This a Go?

- **benefits: you set the pace** taking as much or as little time as you need.
- you **can turn tutorials/silent lectures into fully immersive experiences** (eg playing your favourite music while working through the content),
- **or invite friends to over the Q&A structured/guided materials together,** discussing the questions before looking at answers.

each of these features help you to hold on to the material.

Setting the Stage

while this short primer on carbohydrate biochemistry can stand on its own, you will achieve a greater gain if you looked over the following handouts (all free downloads):

- From Cell Biology to Social Media, the Internet and More
- What Do You See?
- How Do Molecules See Part 1 & 2
- Biochemistry Fundamentals - LIPIDS

building on those
the Silent Lectures on "Biochemistry – Fundamentals" will look at the chemical inventory of living systems from an
“engineering“ point of view
because that strategy will help you to
understand **why**
each class of biological macromolecules is uniquely suited for fulfilling certain roles.

where we started:
from the first chapter listed above, you learned that “life“ depends on partioning of matter across boundaries
➔ the most logical first “pit stop“ in exploring the chemical basis of "life" therefore was to begin looking at “boundary formation“

to do that, we looked at "lipids" to start answering the following questions:
what types of molecules can form boundaries?
are all boundaries alike?
what causes these boundaries to form?
(summary brief answer in "lipid chapter")

Goals of This Silent Lecture

by the end of working through this silent lecture you will

- have **reviewed why lipids can form boundaries**
- **that – counterintuitively – carbohydrates can form boundaries** as well
- that boundaries formed by carbohydrates are **very** different from boundaries formed by lipids
- you will **understand basic principles of carbohydrate structure**
- you will **understand how idiosnycratic properties of carbohydrate structure allow them to play very different roles in biology**
(boundaries, energy storage, connective tissue, joint lubricants)
- you will be **able to distinguish carbohydrates from other biologically important classes of molecules**

Recap: Boundaries Through Self-Assembly

the narrative of the Silent Lecture about LIPIDS was centered on the consequences of placing hydrophobic/amphiphilic lipid molecules in water.

if you did work through that "lipids" lecture then test your recall by trying to fill (some of) the bullet points yourself first before looking at the summary on the next slide:

- ➢ .
- ➢ .
- ➢ .
- .

Recap: Boundaries Through Self-Assembly

the narrative of the Silent Lecture about LIPIDS was centered on the consequences of placing hydrophobic/amphiphilic lipid molecules in water.

here are the key takeaways from that lecture

- **key observation**: the **hydrophobic effect** **drives a spontaneous phase separation** when poorly water soluble (organic) compounds encounter an aqueous environment.

- depending on the type of hydrophobic compound – **phase separation creates an emulsion,** (micelle – we didn't cover that**), or bilaye**r, in which favorable **weak interactions** between molecules are maximized.
-
- life **exploits both** – emulsions and bilayers – to **compartmentalize** space and function

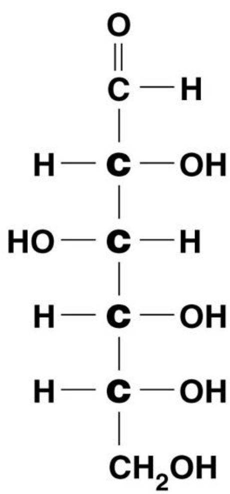
- **bilayers** are a particularly useful and **versatile structural template for implementing physical barriers** that surround cells and many of their intracellular compartments, thus allowing for the fundamental **asymmetry** that enables life processes to be maintained.

You may notice words highlighted in "pink". Use of, and highlighting these words is **<u>not</u>** random/arbitrary. Rather, these "pink" terms represent fundamental **CONCEPTS** (= abstract, overarching ideas, that allow you to easily connect, to structure, and to understand the fundamental "Whys" of (molecular) biology.

If you are interested in learning how Concepts make studying sciences a lot easier, then consider registering for a 50 min online workshop: "Conceptual Frameworks" (QR Code to access list of workshops)

Non-Lipid Boundaries – An Unexpected Beginning

moving on from lipids, not all boundaries are made from lipids.
in fact, some types of boundaries are built from molecules like the one shown here that appear the least likely to create a physical barrier based on what we covered



what do you see <u>right away</u>?

…*try to find the answer … it's not very difficult …*
and just needs you to count a few things ….

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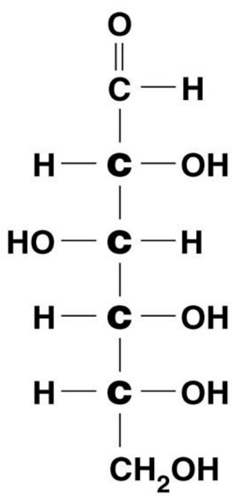

what do you see right away?

6C, 6O, 12H; 5 hydroxyl groups, 1 aldehyde group

based on what we covered in the Lipids Lecture

why does it seem unlikely that this molecule could form a physical barrier?

...try to answer

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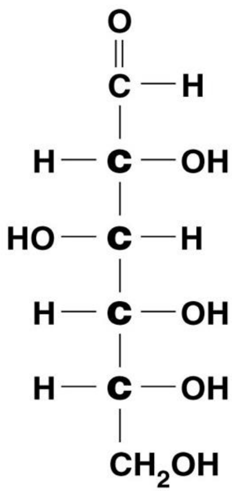

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molecule is extremely hydrophilic/polar
➔ it can readily form H-bonds with water → it should be very soluble in water

→ well…..it is!
→ **how then can this make a barrier?**

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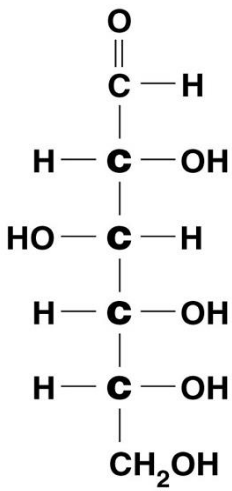

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→ well…..it is!
→ **how then can this make a barrier?**

Answer: not obvious…need to explore further to understand it

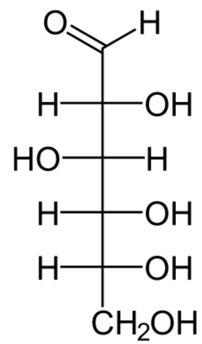
→let's continue looking at the molecule to see if we can discover more things about it.

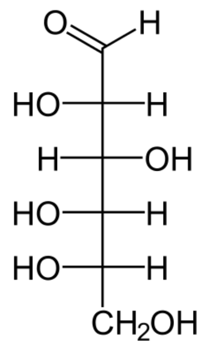
Molecular Asymmetry

closer look - compare the two molecules shown to the right, are they the same?

6C, 6O, 12H; 5 hydroxyl groups, 1 aldehyde group

→ so? are they?





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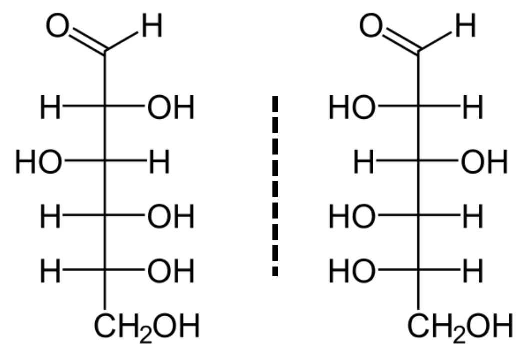

Answer
chemically – yes
but
structurally they seem to be mirrors of each other.

does this matter?

…try to answer

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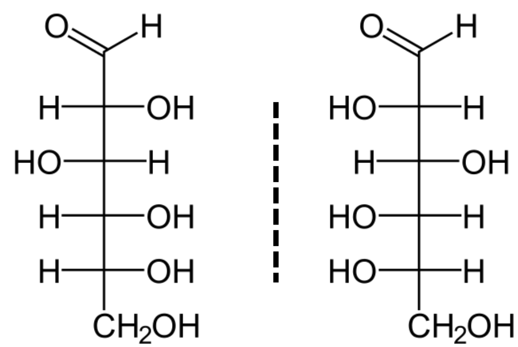
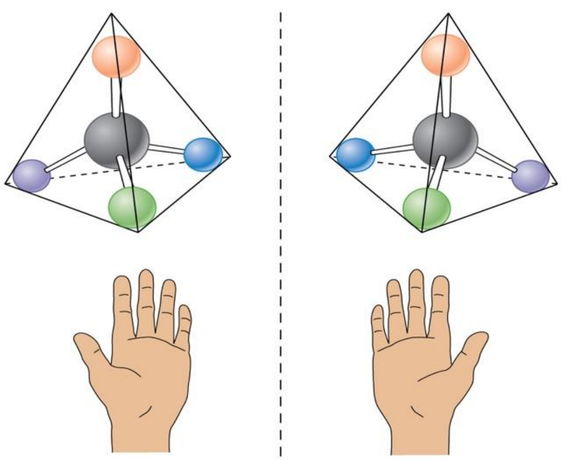


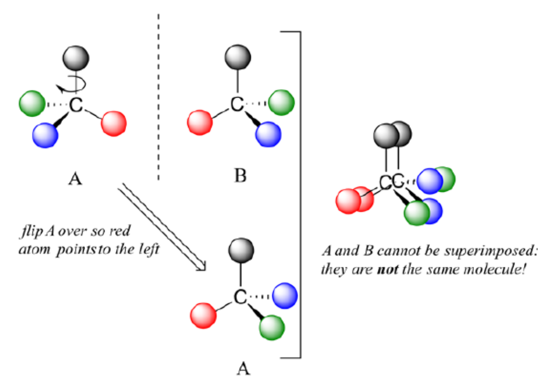
Answer: sure does – why?

→ matters because in these molecules, some carbons are sp^3-hybridized <u>and</u> carry four different substituents.

Such carbons are called asymmetric because the **compound** and its mirror image **cannot be superimposed through simple translations and rotations**.



chiral from greek χειρ (*kheir*), = hand



https://www.khanacademy.org/science/organic-chemistry/stereochemistry-topic/chirality-r-s-system/v/chiral-achiral-jay

Molecular Asymmetry

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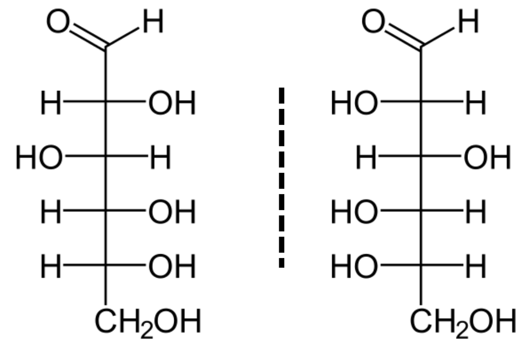

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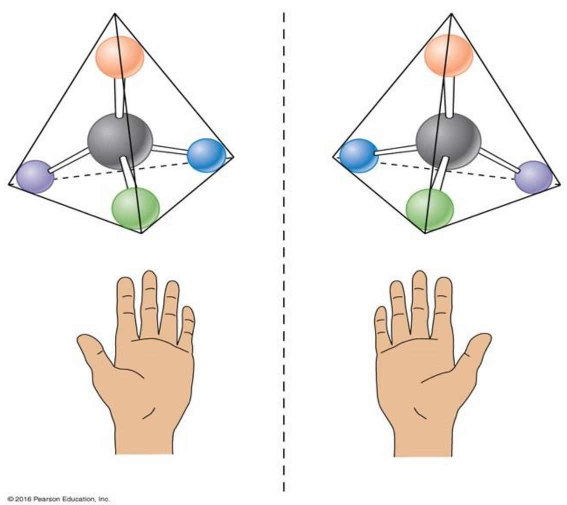


organic compounds that are **identical in composition and connectivity** but differ from each other by the orientation of atoms in space are called **stereoisomers.**

if the two molecules behave like **mirror images**, they are called **enantiomers** (eg as shown in the image above)

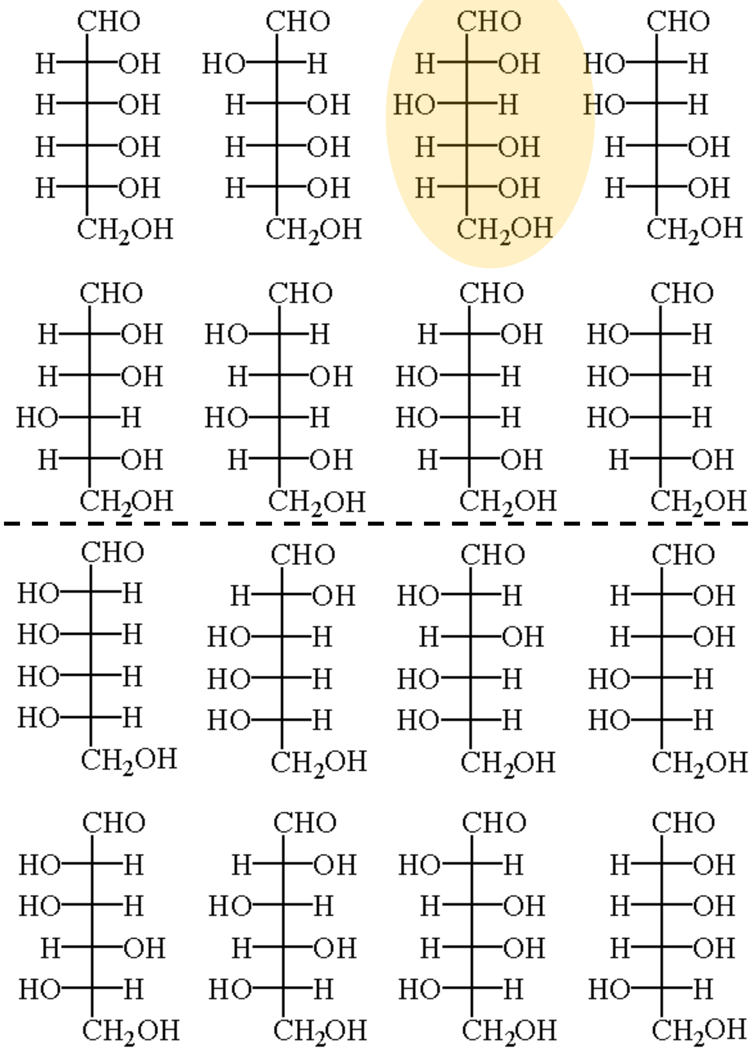
the presence of more than one asymmetric carbon causes 2^n stereoisomers; not all of which relate to each other like simple mirror images, and you will learn a great deal more about their nomenclature and the consequences of stereoisomerism in your orgo classes.

Molecular Asymmetry

for instance: for a "hexose" that has 4 asymmetric C-atoms and an aldehyde group at C1 (the "top" of the molecule), this will look like this

The highlighted compound is glucose

16 Aldohexoses



Definitions

molecules that have equimolar ratios of Carbon and Water - $(CH_2O)_n$ - are collectively called **carbohydrates** (= "carbon + water")

for instance: $C_6H_{12}O_6$

if the carbohydrate has an aldehyde functional group, it belongs to the "aldose" family

(example shown here for aldose with 6 carbons = hexose)

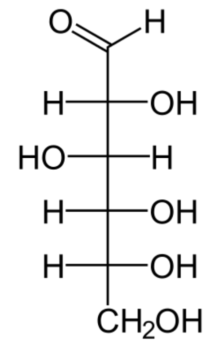
if the carbohydrate has a ketone functional group, it belongs to the "ketose" family

(not covered in this Lecture, but an example would be Fructose)

Carboyhdrate Structure – Ring Formation

chirality of the molecule, called glucose, is interesting but

what, if anything, has that to do with the ability of glucose to form boundaries/barriers?



Answer:
A LOT

but to understand it, we need to think more about structure of glucose (and molecules like it)...so I ask your patience as we work through the different levels of the answer.

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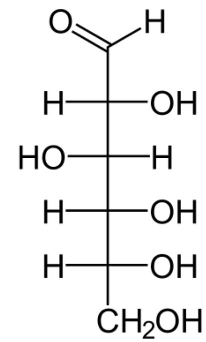

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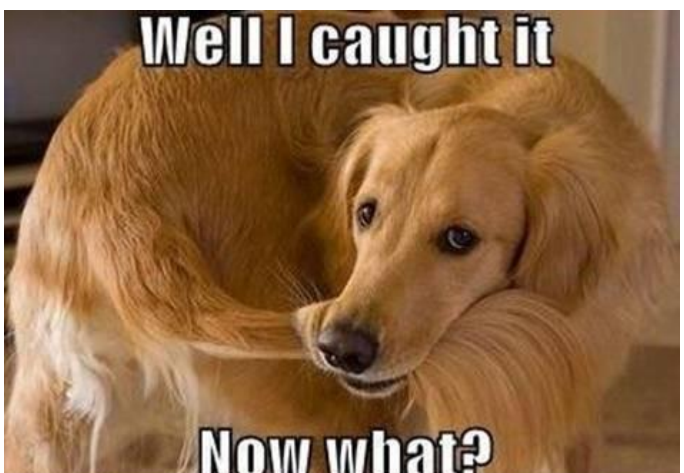
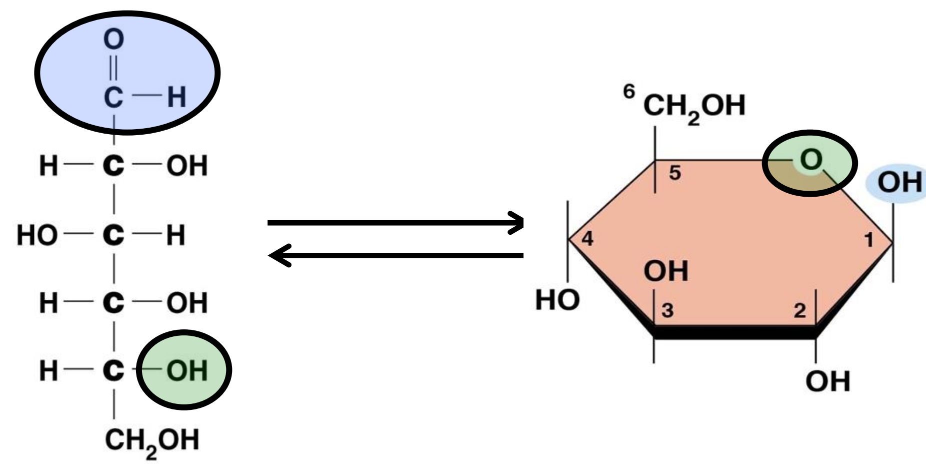
starting right here: carbohydrates can exist in linear and cyclic forms ... why?

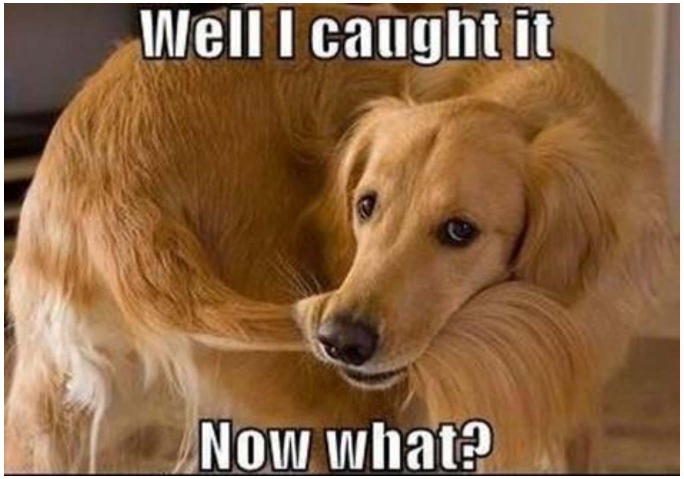


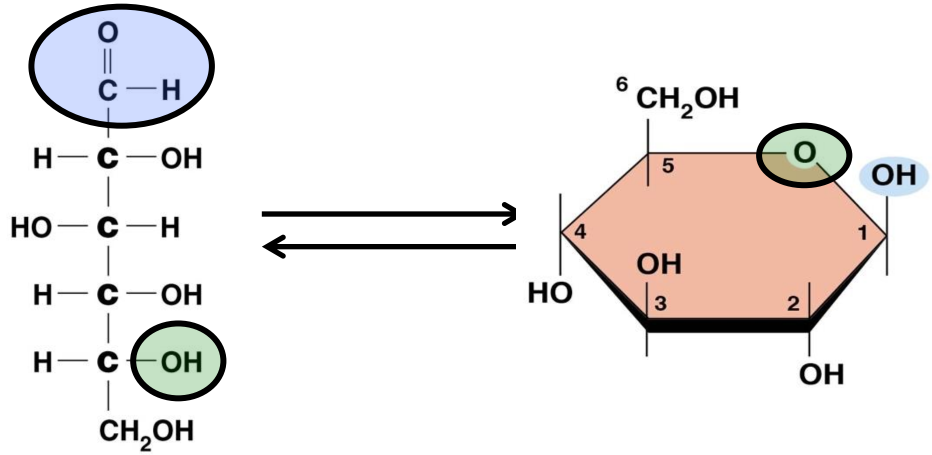


Carboyhdrate Structure – Ring Formation

Answer

the **“flexibility”** (free rotation around single bonds) **of the carbon backbone in** glucose and molecules like it **allows** an **intra**molecular **ring closure** reaction between one of the hydroxyl groups and the carboyhydrate’s carbonyl function.





the rings typically have 5 or 6 atoms, including the ring oxygen – which gives rise to their family name pentose (5 membered ring), hexoses (6 membered ring)….

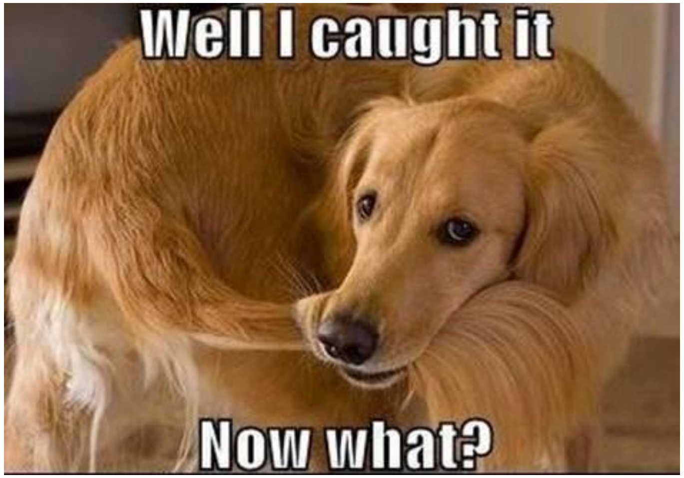
(you will learn more about it in the "Advanced Biochemistry" Lectures)

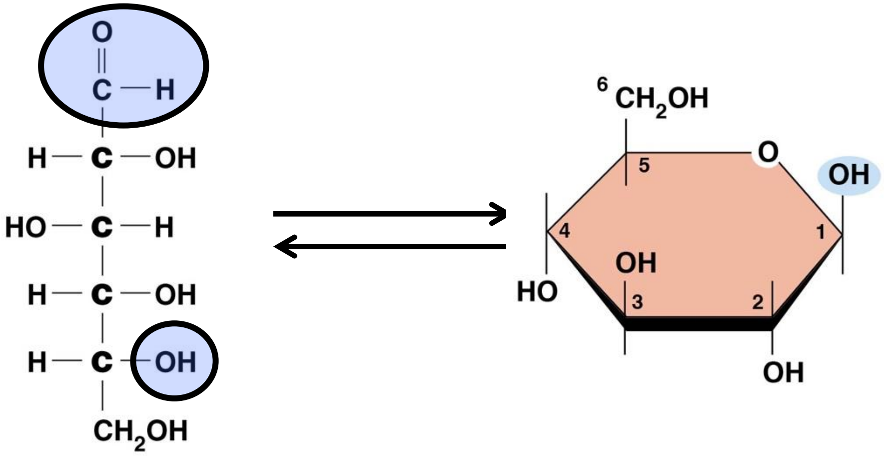
here we just want to focus on the aspects that help understand how glucose can form barriers

Carbohydrate Structure - Conformation

writing the ring this way is convenient but also makes an **important omission ….what could that be?**

.....if you already took "Organic Chemistry" ... then you should be able to answer this .... try!





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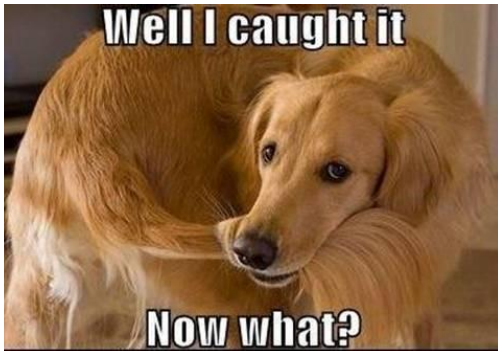
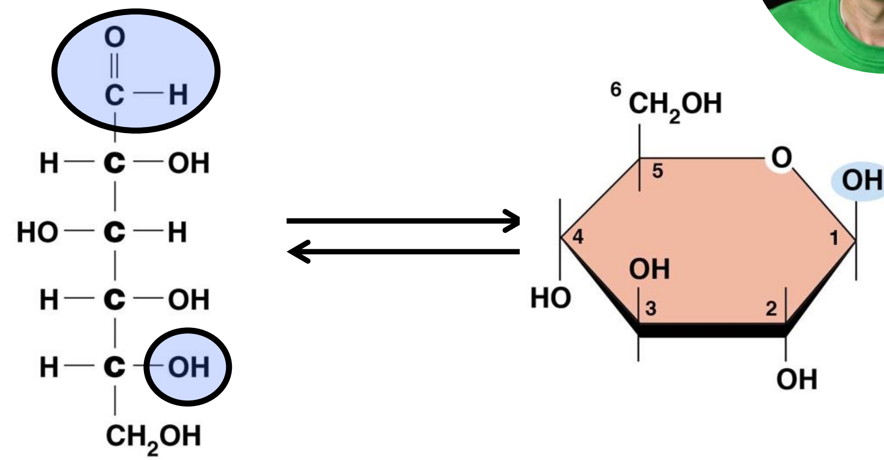
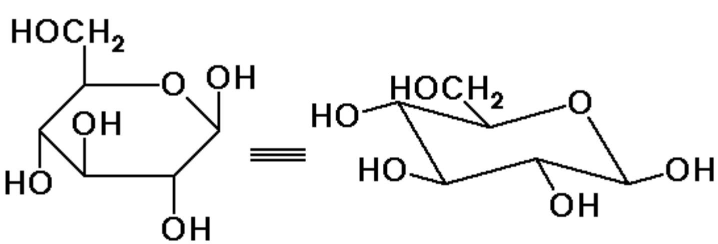




Answer:
it ignores that the **carbons are sp^3-hybridized** … meaning **tetrahedral geometry.**

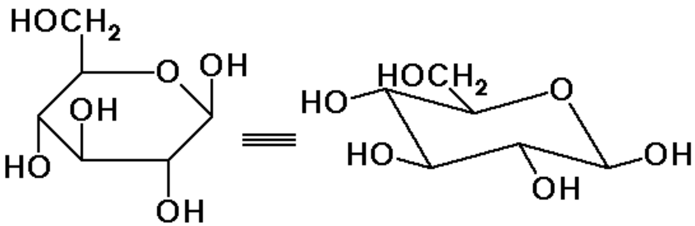
→ accounting for that, the **overall shape of glucose is similar to cyclohexane** ... → we looked at that in the context of mechanisms of molecular recognition ("How Do Molecules See? Pt2, slide 10) .
that is ...a representation closer to the "truth" would look like this:



which of the two ways of drawing it do you think people prefer?
→ the one on the left
→ understandable but also makes it easy to forget about "spatial" realities.

Carbohydrates Beyond Monomers

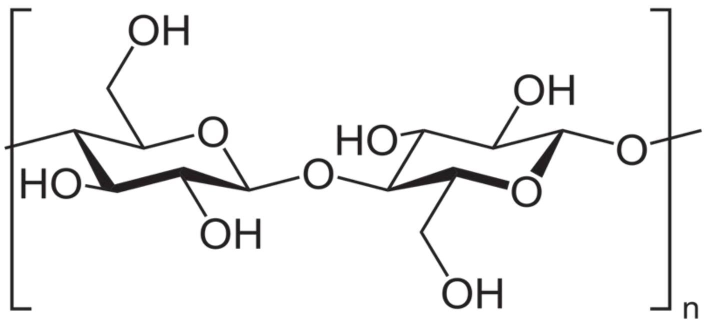
getting close to finish line now…..but why?
what does **the "chair" conformation have to do with water solubility of glucose?**



Answer: nothing IF you deal with a single glucose molecule, **and EVERYTHING** once you make a glucose polymer.

important point we learn here:
the chemical and physical properties of a polymer **<u>can but need not be</u>** reflective of the chemical and physical properties of the constituent monomers

→ in other words: polymer formation imparts **emergent properties**...!



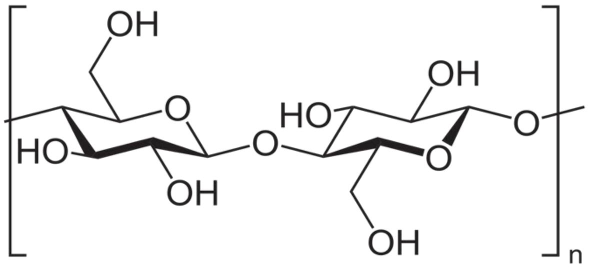
Cellulose, n = 10,000 – 15,000

point in case
glucose polymers…..let's begin with cellulose
(…yep, there is >1 type of glucose polymer)

first thing to notice: **structural "blueprint" is fundamentally different from** the polymer that is formed in the **aggregation of phospholipids? how?**

***....**try to answer before going on …..*

Carbohydrates Beyond Monomers



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Answer:
covalent bonds between monomers.
in other words: cellulose is a first example for a **covalent polymer.**
the polymerization reaction (in this case, elimination of water to make a bond between two glucose molecules) is **<u>not</u>** spontaneous, fundamentally setting it apart from the spontaneous aggregation of phospholipids.

the lack of spontaneity in the reaction goes along with increased control "where" and "when" a boundary is formed, but does not explain how cellulose can serve to form boundaries in the first place….

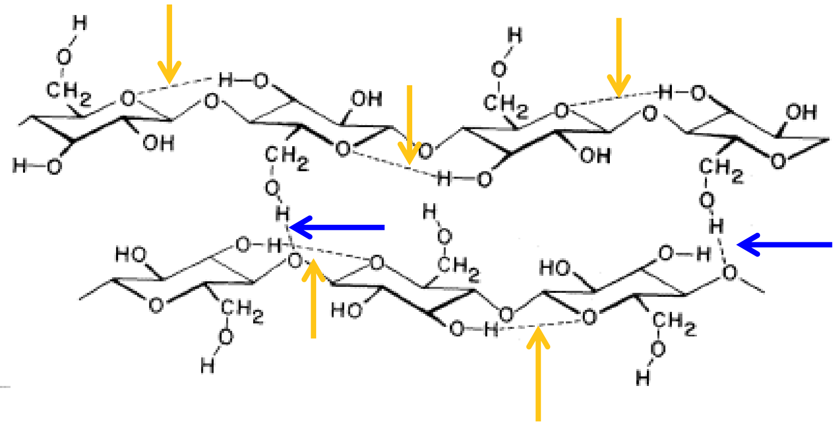
Cellulose

….**very rigid boundaries** in fact → found in the **cell walls of plants.**

understanding of the **water insolubility and rigidity of cellulose based boundaries** comes from looking at the detailed structure:

the **stereochemistry and chair conformation** of the glucose monomers allow this polymer to form long linear H-bond stabilized **strands** that **laterally aggregate through non-covalent (!) H-bonding between strands**. This results in “cables” that are so tightly packed that **water molecules cannot penetrate** the assembled structure.

→ if water cannot get there, then there is no “solvation” and ….yep: the polymer will be completely insoluble in water.



https://publishing.cdlib.org/ucpressebooks/view?docId=ft796nb4n2&chunk.id=d0e311&toc.depth=1&toc.id=d0e311&brand=ucpress

Chitin

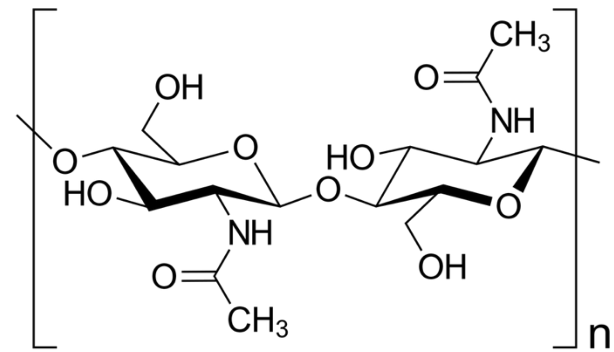
is cellulose the only polysaccharide that can form amazingly stable and resilient physical barriers?

Answer

no

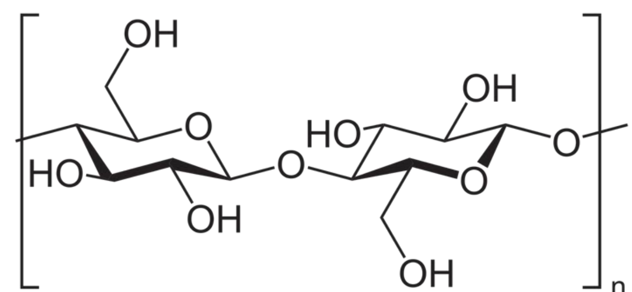
the hard exoskeletons of insects or crustaceans are another example.

The underlying polysaccharide, chitin, has this structural formula:



do you notice what is different between cellulose (shown to the right) **and chitin?**

…try to describe in your own words…..



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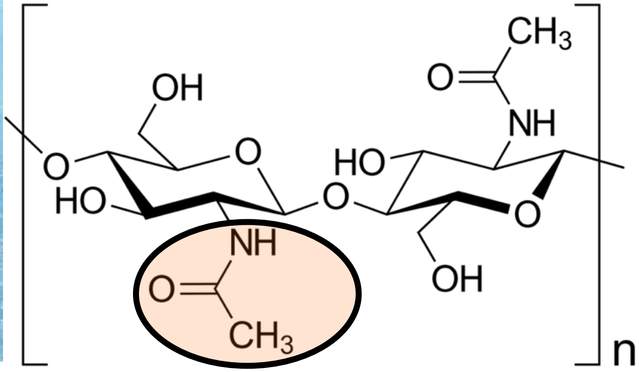
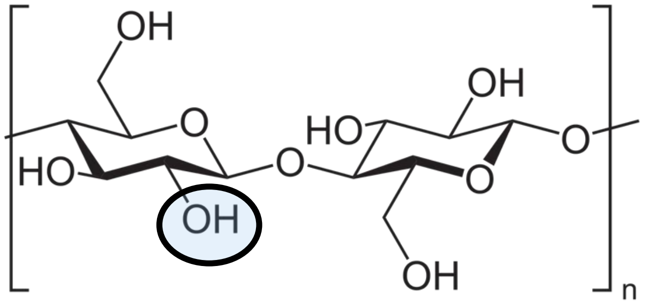




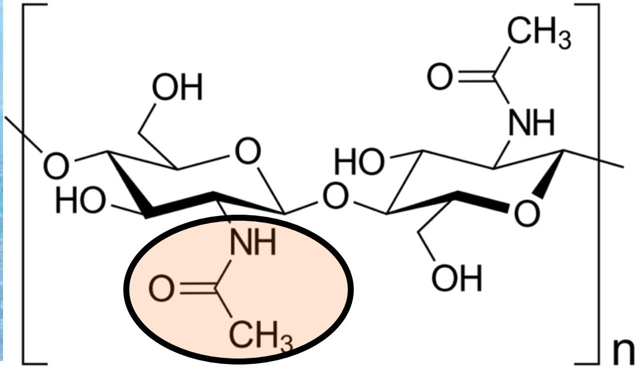
Answer:

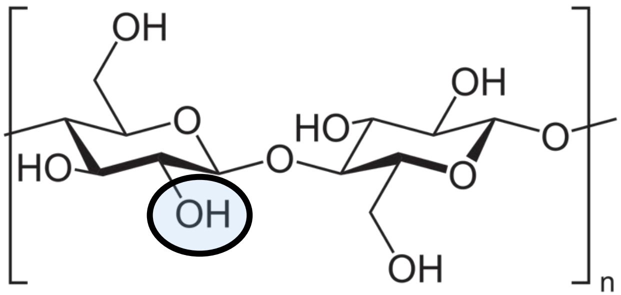
chitin is based on a chemically modified glucose molecule where one of the hydroxyl groups (blue underlay) was replaced by an acetylated amino group (N-acetyl).

although this modification causes the chemical formula of N-acetylglucosamine to deviate from the empirical $(CH_2O)_n$ formula that globally defines carbohydrates, N-acetylglucosamine and other chemical variants are still categorized as carbohydrates because they share key chemical, structural and functional properties.

Chitin

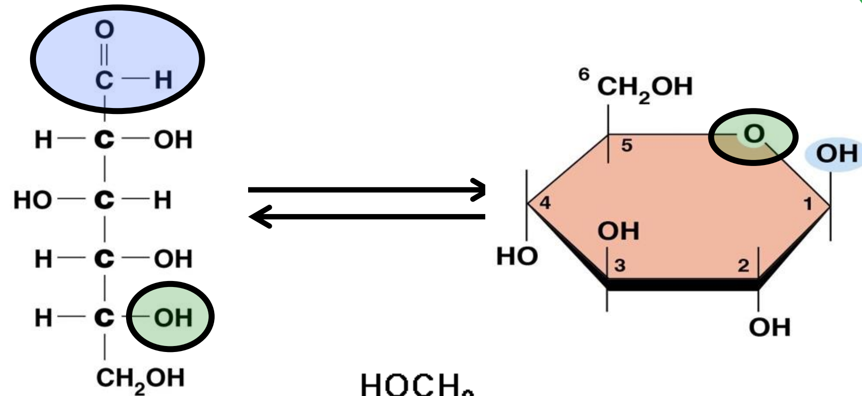
most importantly: the comparison between cellulose and chitin showcases the **important concept**: redundancy = the "tweaking" of a common template (here: glucose) to implement different biological solutions (here: cell wall, exoskeleton)

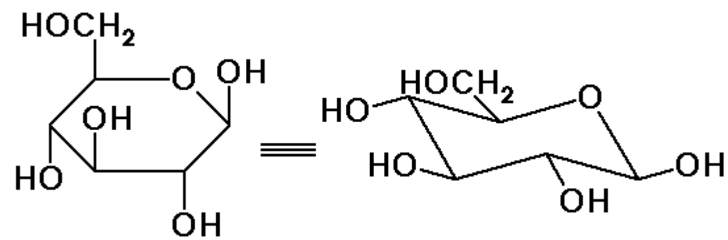


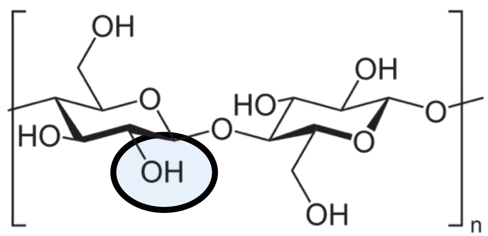


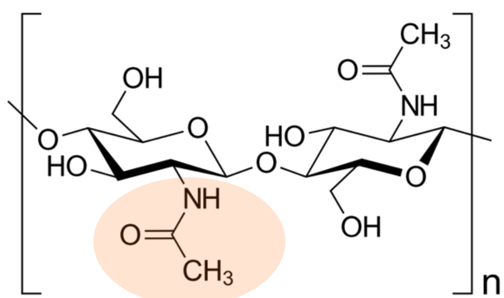
RECAP

carbohydrate: $(CH_2O)_n$ [some deviations are allowed]









if you want to solidify your understanding before moving on, then use this "RECAP" slide as template to give a short verbal summary of how carbohydrates can form very stable boundaries, and why – at first sight – this is counterintuitive

you could, for instance, start with the sentence: "Sugar is sweet …until it isn't….."

Glucose Polymers – Devil in the Details

now that you have seen how glucose can be used to generate a physical barrier – the question becomes: **do ALL glucose polymers form rigid, cell-wall like boundaries?**

….what do you think? …

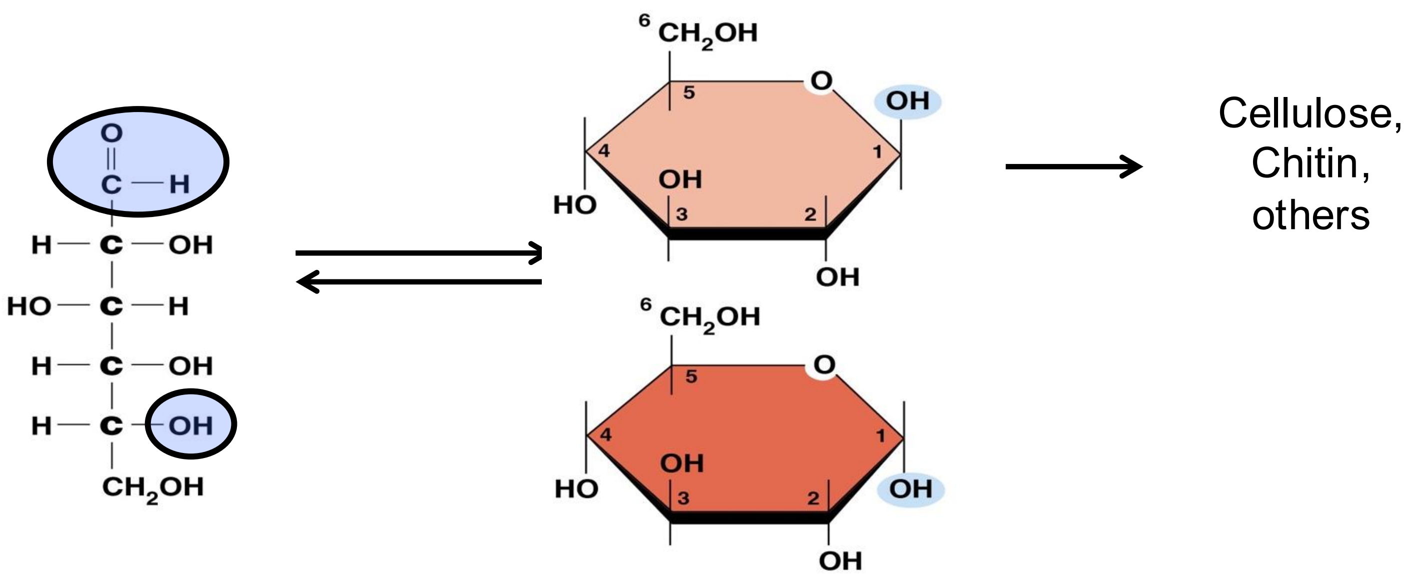
Glucose Polymers – Devil in the Details

now that you have seen how glucose can be used to generate a physical barrier – the question becomes: **do ALL glucose polymers form rigid, cell-wall like boundaries?**

Answer:

No – "of course" not. (this was obviously a leading question...)

to make sense of this, we want to revisit the ring formation of glucose...because there is one important detail that was not brought up earlier:

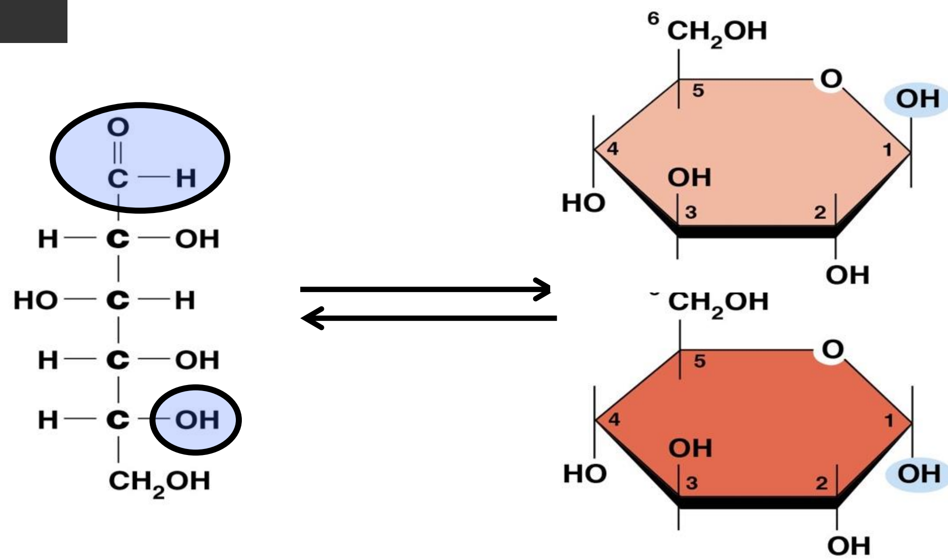


the free rotation around C-C single bonds combined with the easy reversibility of the ring closure reaction (equilibrium!) means that the reaction can have two different outcomes.

what is the big deal about it = why would you want to care?

....try to come up with a guess (or answer if you know it) ....

Glucose Polymers – Devil in the Details



→ Cellulose, Chitin, others

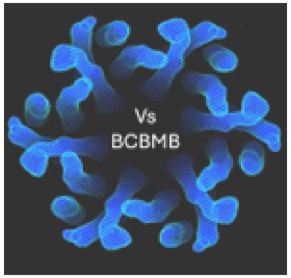
the free rotation around C-C single bonds combined with the easy reversibility of the ring closure reaction (equilibrium!) means that the reaction can have two different outcomes.

what is the big deal about it = why would you want to care?

Answer

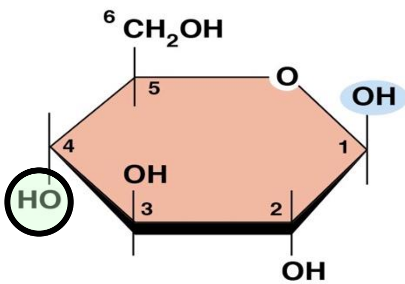
if you apply what we learned so far, you realize that the ring closure creates **two stereoisomers**. in carbohydrate chemistry…this newly formed asymmetric carbon is “special” and is called anomeric carbon.

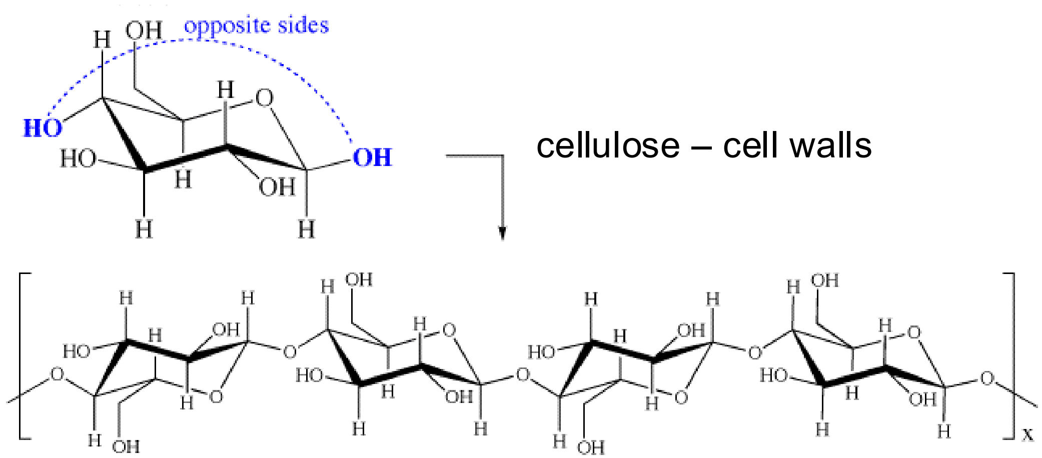
→ The existence of **anomers** has enormous significance in Nature ....



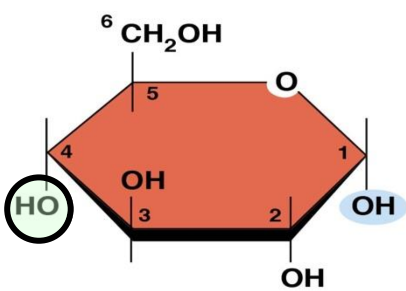
Wall …or no Wall …
It's All About "Up" or "Down"

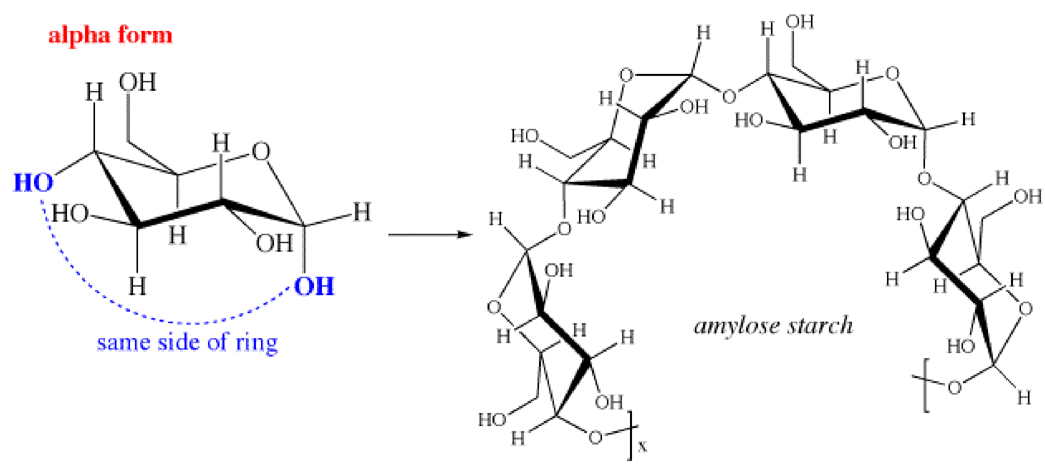
….for instance when it comes to what polymers of glucose look like, and are used for by biology……





Linear polymer





Spiral shaped
polymer

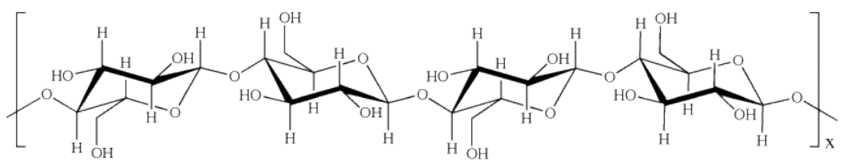
Structure Determines EVERYTHING….

comparing just amylose and cellulose let's us derive an important general statement: **STRUCTURE DETERMINES FUNCTION**

→ this simple statement is a **fundamental paradigm in all of biology**.

based on what we covered in this and previous lectures, you should be able to understand why this paradigm holds…. structure determines shape → shape determines what is presented to the environment → what is presented determines what it can see/recognize/interact with, and it also determines what chemistry can occur if the environment is suitable.

…the differences in the structures (and consequently function/role) of cellulose and amylose are entirely due to the fact that they are build from different anomers of glucose.

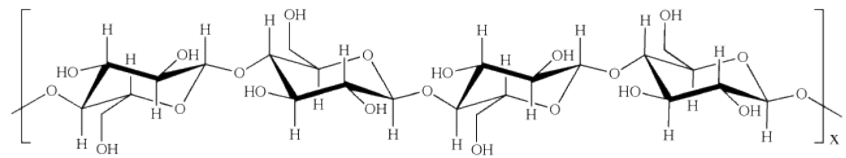


→ **important insight: STEREOCHEMISTRY MATTERS IN BIOLOGY!**

→ true not only in the case of amylose vs cellulose, but in general because stereochemistry defines the patterning/presentation of functional groups ➔ stereochemistry is key for molecular recognition (= way how molecules "see") and the **specificity** that characterizes biological processes

….Though Physics is Important Too

one question that may linger on your mind considering that starch evidently is not cellulose why isn't amylose used to form cell walls too?



Answer

here ... any physics/engineering background comes in handy.

What has higher tensile strength…..cable or spring?

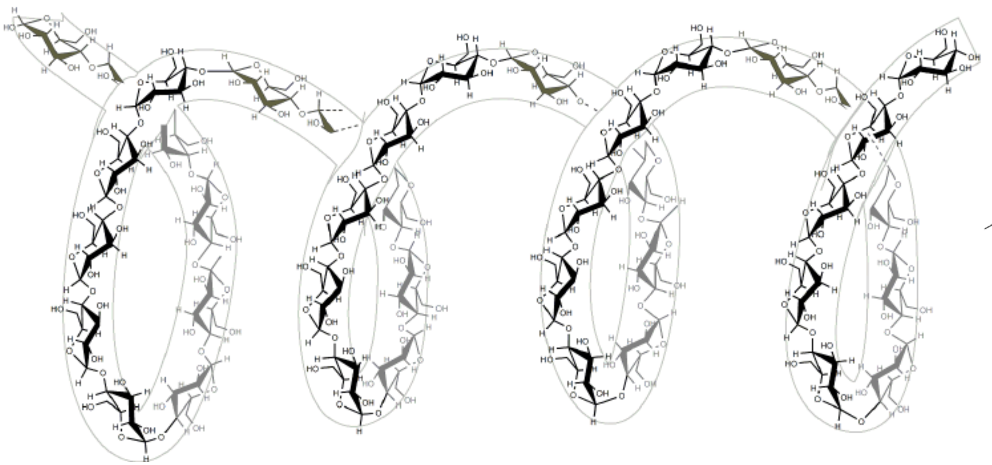
- **Cellulose;** n=10,000-15,000; polymer is maximally extended = can't stretch more without breaking covalent bonds; tensile strength further increased by parallel alignment of many polymer chains that are held together through weak interactions → those add up causing a register that will not "slip" even under heavy load; layering cables at different angles on top of each other makes it even stronger (**cellulose fibers can be used to generate fibrils that are stiffer and stronger than steel**)
- **Amylose;** n averages ~100-1000; polymer is coiled; if at all - only few H-bonds form between turns, and lateral packing of independent spirals through direct H-bonds is far less favorable/prevalent than between cellulose strands.

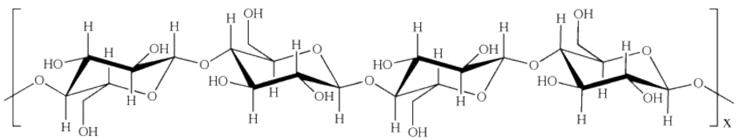
→ amylose is fairly "flimsy" molecule and not able to resist big loads without deforming.

yet: the one property it shares with cellulose is ….insolubility in water …why????

. *…make a guess based on what was said about cellulose earlier on ….*

….Though Physics is Important Too





→ amylose (left) is fairly “flimsy” molecule and not able to resist big loads without deforming.

yet: the one property it shares with cellulose is ….insolubility in water …why????

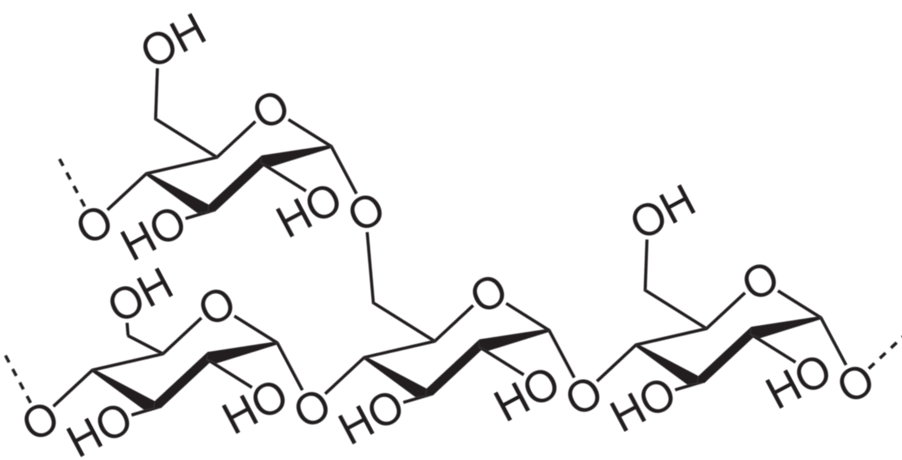
Answer

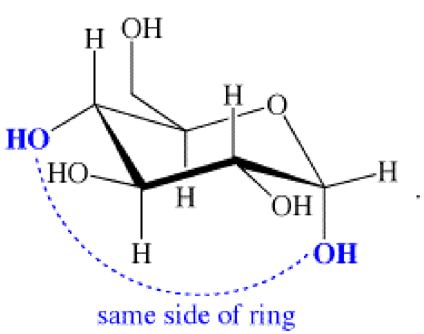
as open as the spirals may look …if you think of them as a string wrapped around a “cylinder”, then all the –OH groups project parallel to the cylinder surface (no good for water to infiltrate) → the inside of the cylinder is quite hydrophobic + space is so tight that the van der Waals surfaces of the C, and H atoms fill it in.

Polysaccharides – Water Soluble at Last!!

Cellulose, Chitin, Amylose …. all water insoluble.
this raises the question:
are <u>all</u> polymers of glucose water insoluble?

Answer: no.
sticking with plant starch ….while amylose is water insoluble, a second glucose polymer in starch is actually water soluble. Its name: amylopectin.





what's happening here?

***...your turn**, look at it and describe in your own words what you observe ….what is different here from the examples we saw so far?*

Polysaccharides – Water Soluble at Last!!

Cellulose, Chitin, Amylose …. all water insoluble.
this raises the question:
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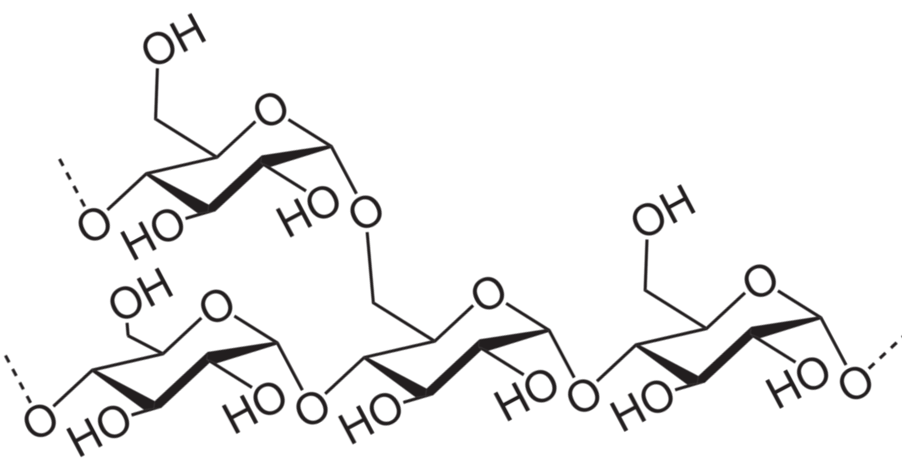
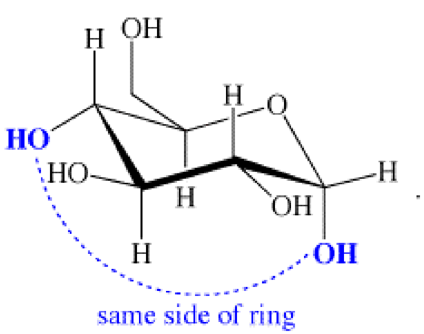


Answer
the chain is still made from the same glucose anomer than amylose – **BUT**, every 24-30 monomers along the chain this **polymer branches!** And each branch can spawn off additional branches.

→ The branching prevents coiling, and results in a much more open structure that water can access.

→ **water soluble** (….at last!)



Among covalent biological polymers …branching of the polymer <u>backbone</u> (= at the level of the basic unit that allows the molecules to bond together) is unique to carbohydrates
(you will learn more about this in the "Advanced Biochemistry" Lectures)

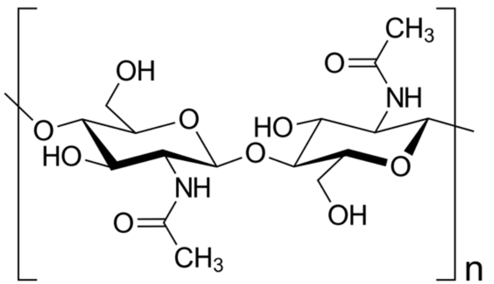
….it is, however, not the only way how you can make a polysaccharide water soluble ......

Polysaccharides – "Supercharging" Solubility

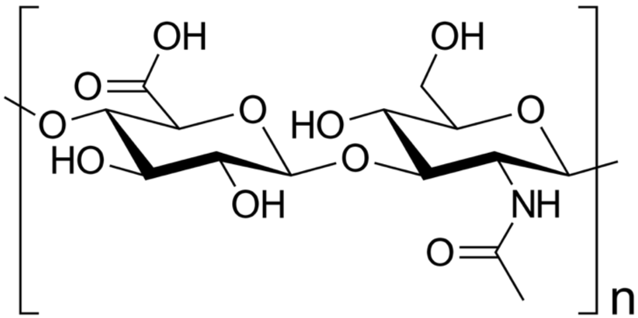
,,,,if the key to water solubility is to minimize weak interaction **within** the polymer (and maximize H-bonding on the surface) then besides steric hindrance, you should be able to exploit chemical properties too….

→ **redundancy = tweak a template to make it work in a different context**
(eg cellulose (all glucose) and chitin (all N-acetylglucosamine))

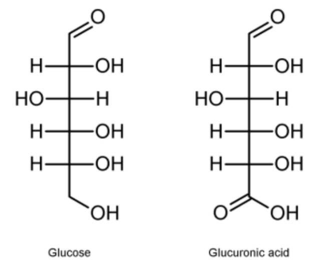
→ shown below, we encounter a different strategy ….**what is different?**
.
….*look at the structures below and verbalize what you notice/observe/see*



Chitin

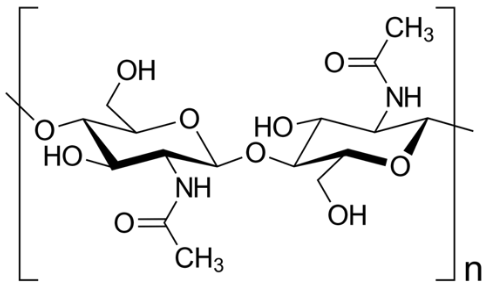


Hyaluronic Acid; n ~20,000

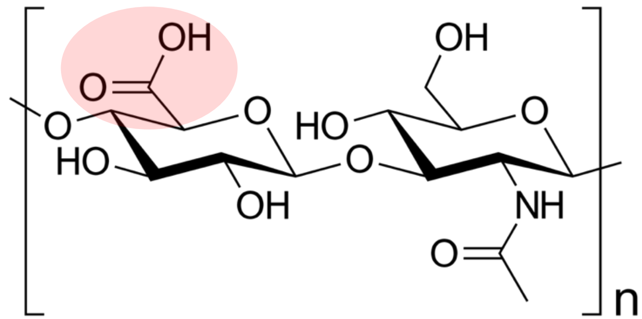


Polysaccharides – “Supercharging” Solubility

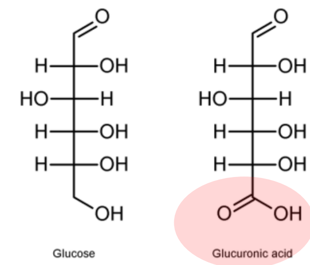
→ ….. different strategy ….what is different?



Chitin



Hyaluronic Acid; n ~20,000



Answer:

water **insoluble chitin** = N-acetylglucosamine **homopolymer**;
water soluble **hyaluronic acid = heteropolymer** of **glucoronic acid:N-acetylglucosamine** (glucoronic acid: oxidized glucose with a -COOH instead of $–CH_2OH$). Hyaluronic acid is water soluble because the acidic carboxyl groups dissociate in water, leaving a negatively charged carboxylate anion ($-COO^-$)

→ high negative charge density on hyaluronic acid prevents lateral association between polymer chains (electrostatic repulsion) and provides a very good hydrophilic surface to interact with water molecules.

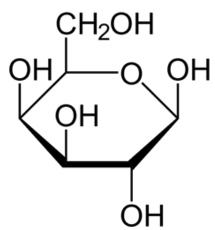
under physiological conditions: hyaluronic acid is a gel
used in connective tissue and as joint lubricant;
the ability to bind and hold water molecule also makes hyaluronic acid a popular ingredient in skincare products (keeps skin hydrated and “plump”, smoothing out fine lines/wrinkles)

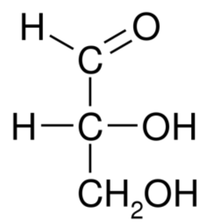
POP QUIZ……

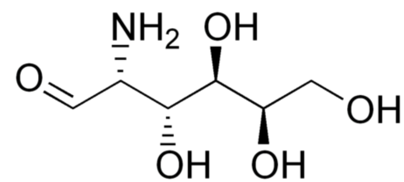
let's finish by seeing how well you recognize characteristics of carbohydrates….

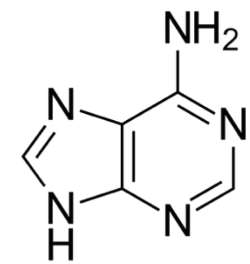
look at the following structures and decide

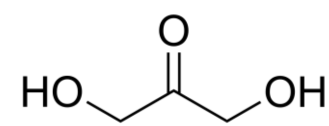
- **whether they are carbohydrates and**
- **whether or not they are water soluble**

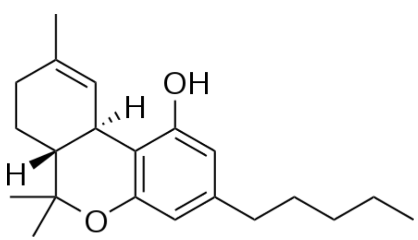


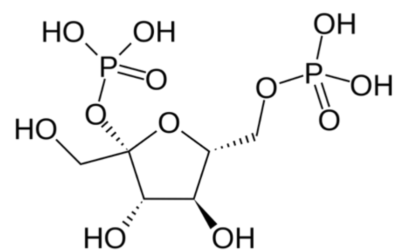


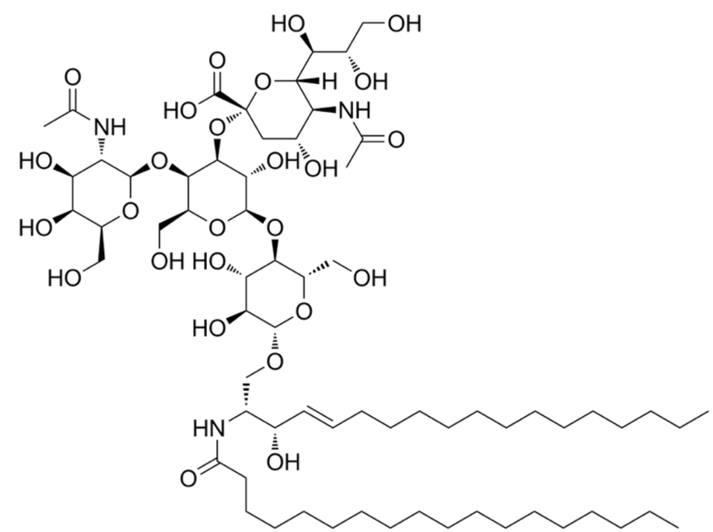


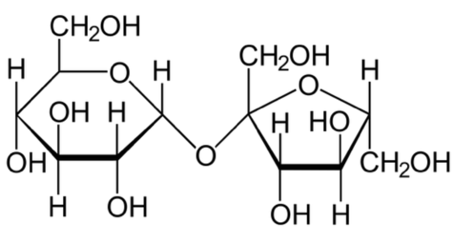


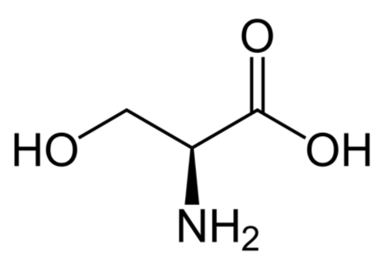


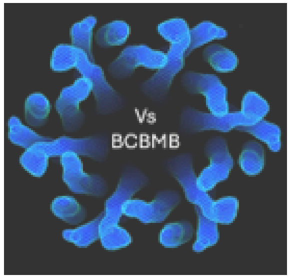










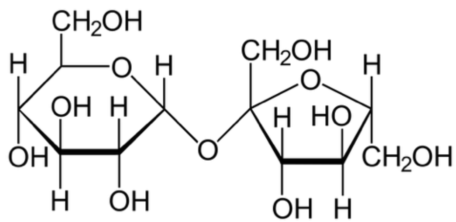


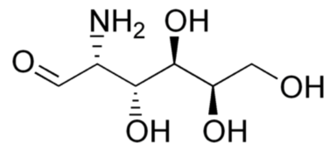
POP QUIZ……

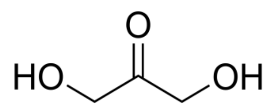
look at the following structures and decide

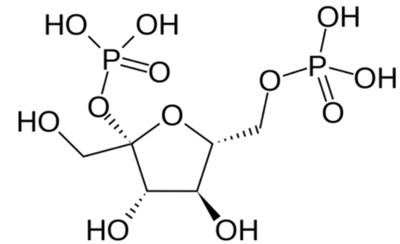
- whether they are carbohydrates and
- whether they are water soluble.

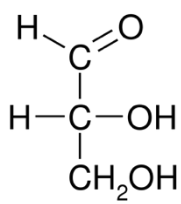
yes/yes

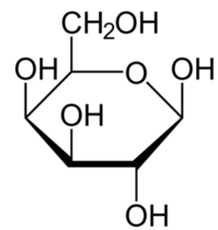


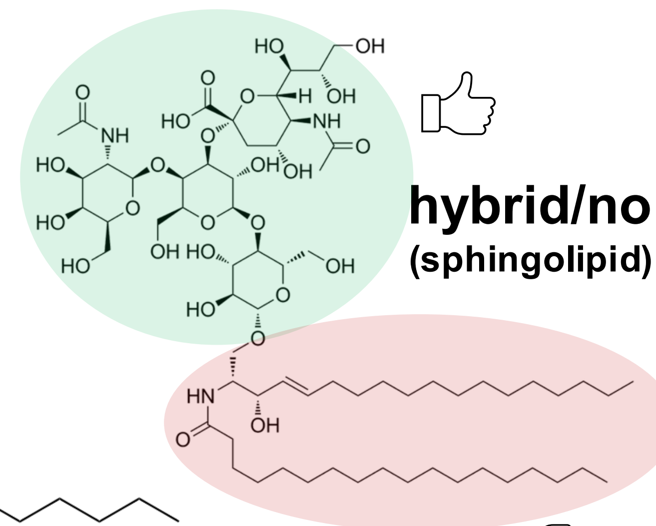




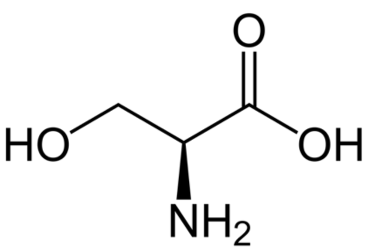




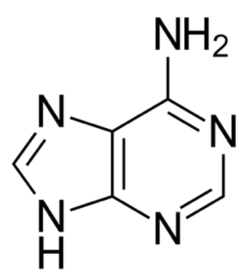




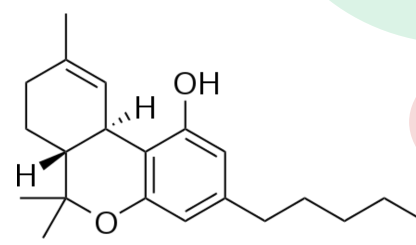
hybrid/no
(sphingolipid)



no/yes
(amino acid)



no/poorly
(nucleobase)



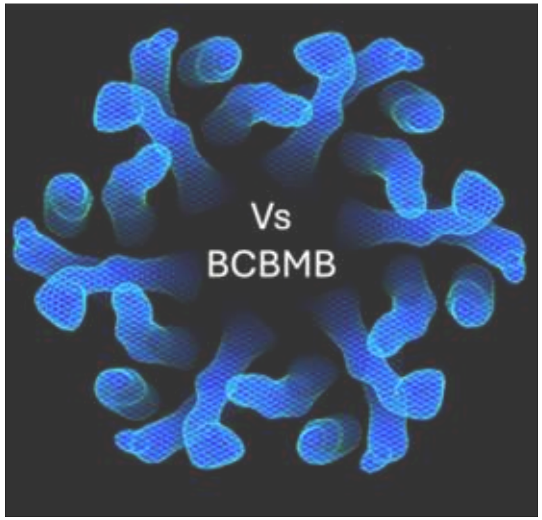
no/no
(lipid)

SUMMARY

- **carbohydrates** are **globally defined** as molecules made from equimolar amounts of carbon and water = **$(CH_2O)_n$**
- in their **monomeric** form, carbohydrates are **readily water soluble**
- just as for other types of biological macromolecules, **chemical modifications** of the basic parent compounds are **common**. The resulting molecules are still thought of as carbohydrates, as long as their overall composition are within reason of the generic description $(CH_2O)_n$
- with **<u>very</u> few exceptions** that are all limited to carbohydrates with 3 carbon atoms, carbohydrates are asymmetric molecules that **contain one or more chiral carbon atom**.
- presence of one or more asymmetric carbon atoms gives rise to **complex stereochemistry**.
- a **characteristic structural property** of monomeric carbohydrates is the ability of their linear forms to spontaneously and reversibly **form closed 5- and/or 6-membered rings**.
- for any given sugar (= monomeric carbohydrate), **ring closure** of a sugar gives rise to a pair of stereoisomers – referred to as **anomers** - because the reaction creates an additional asymmetric carbon atom.
- in their monomeric state, anomers have very similar solubility in water. However, the **structural, physical, and chemical properties of carbohydrate polymers – called polysaccharides - vary tremendously depending on which anomer** of a sugar forms the basic unit of a polymer
- **polysaccharides are covalent polymers**, which sets them apart from the non-covalent polymers that form when lipids are exposed to water

the emergent properties of polysaccharides are exploited for a broad spectrum of biological functions, ranging from cell wall formation in plants, to energy storage (eg starch), or lubrication of joints.

adding two **Mantras/Paradigms** beyond "**Everything is based on Equilibria**": **STEREOCHEMISTRY MATTERS** and **STRUCTURE DETERMINES FUNCTION**



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