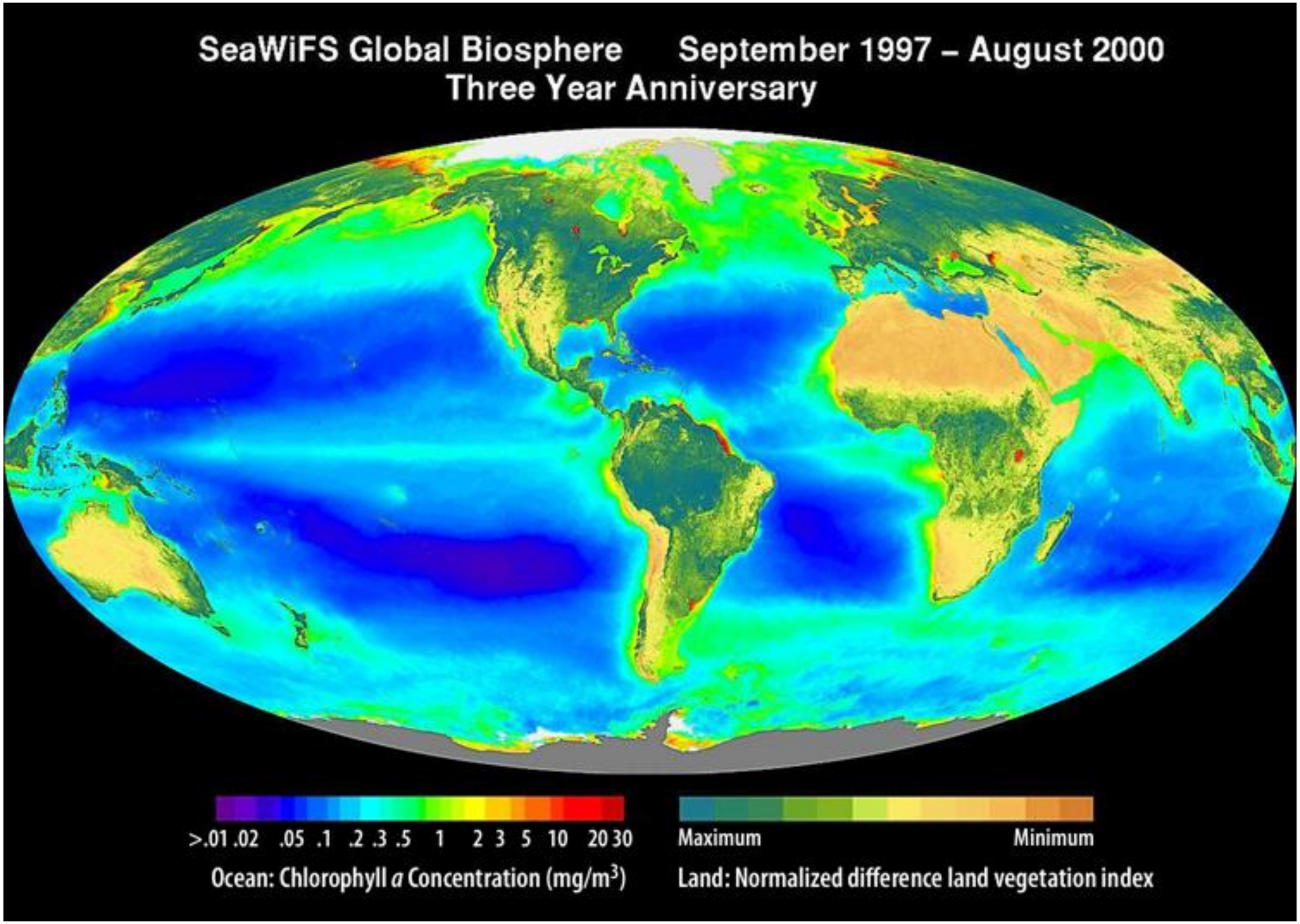
Topic 2

Feedstocks

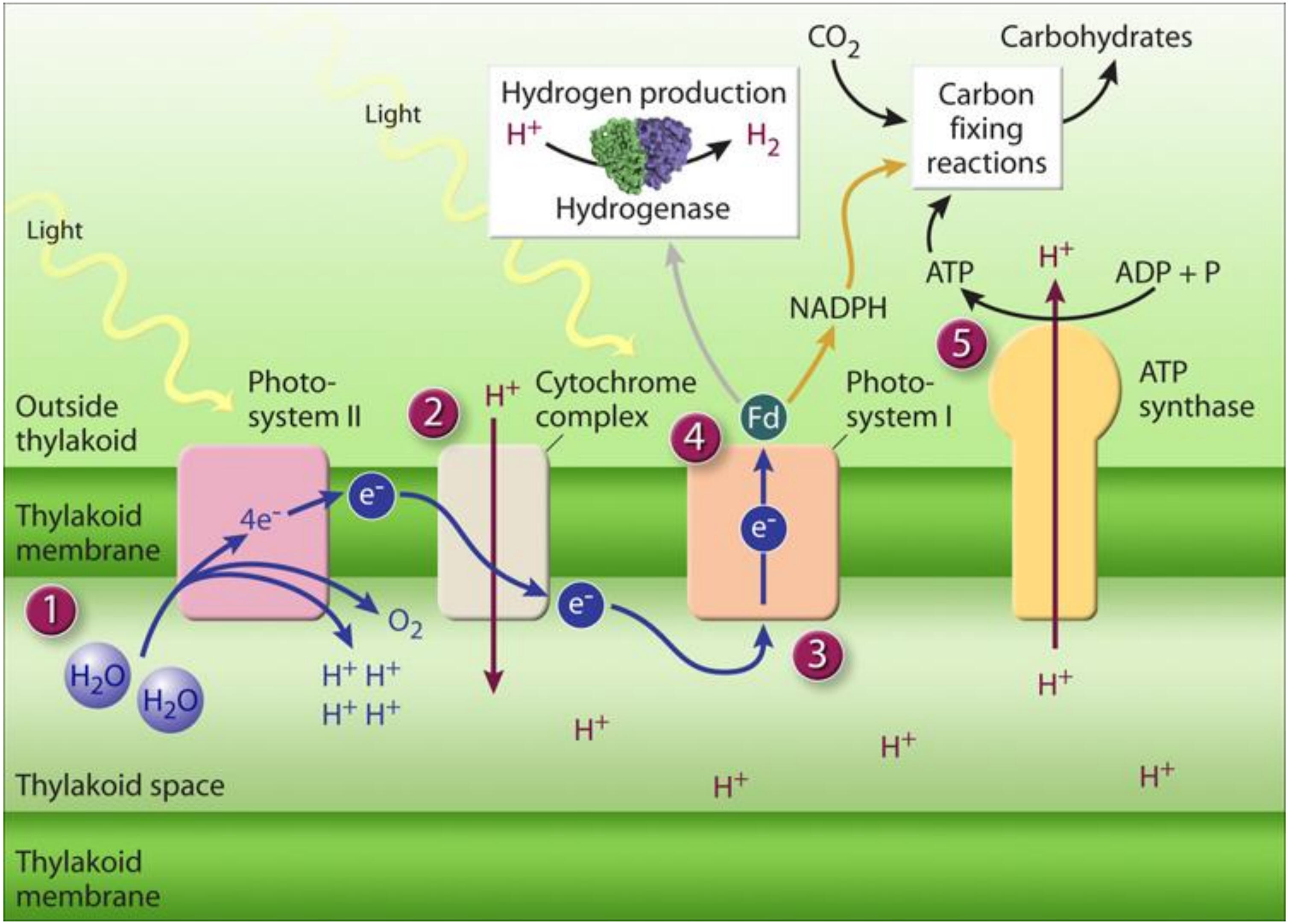
Topic 2

Starch: Synthesis, Properties and Degradation

Photosynthesis



Photosynthesis

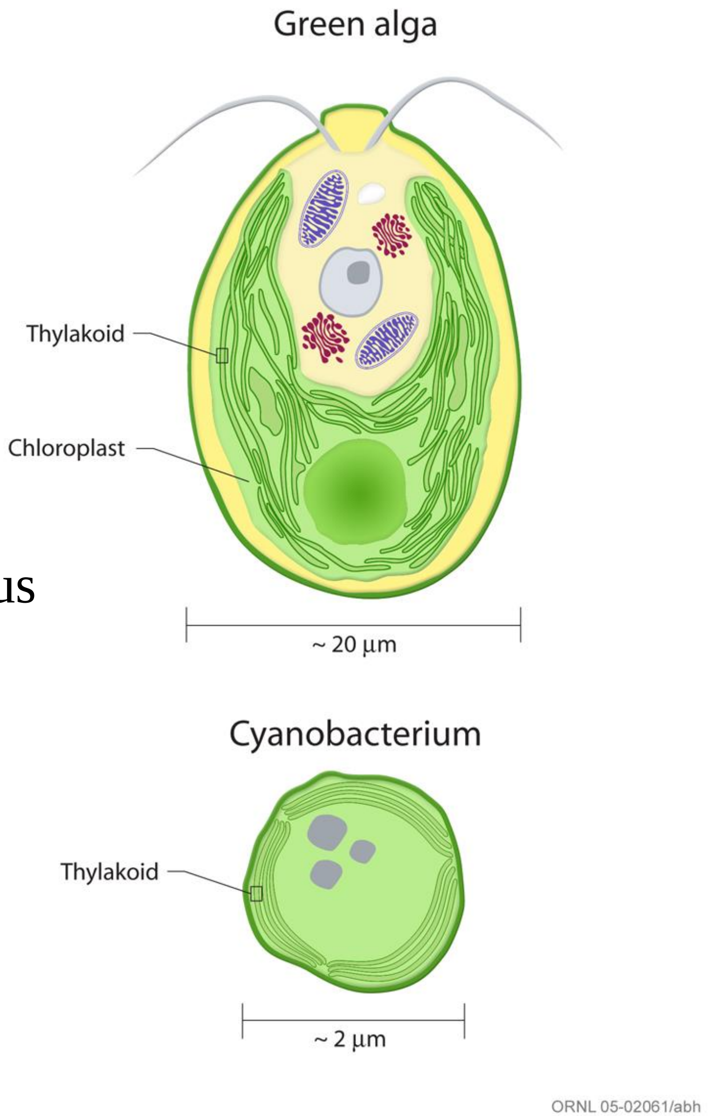


Photosynthesis

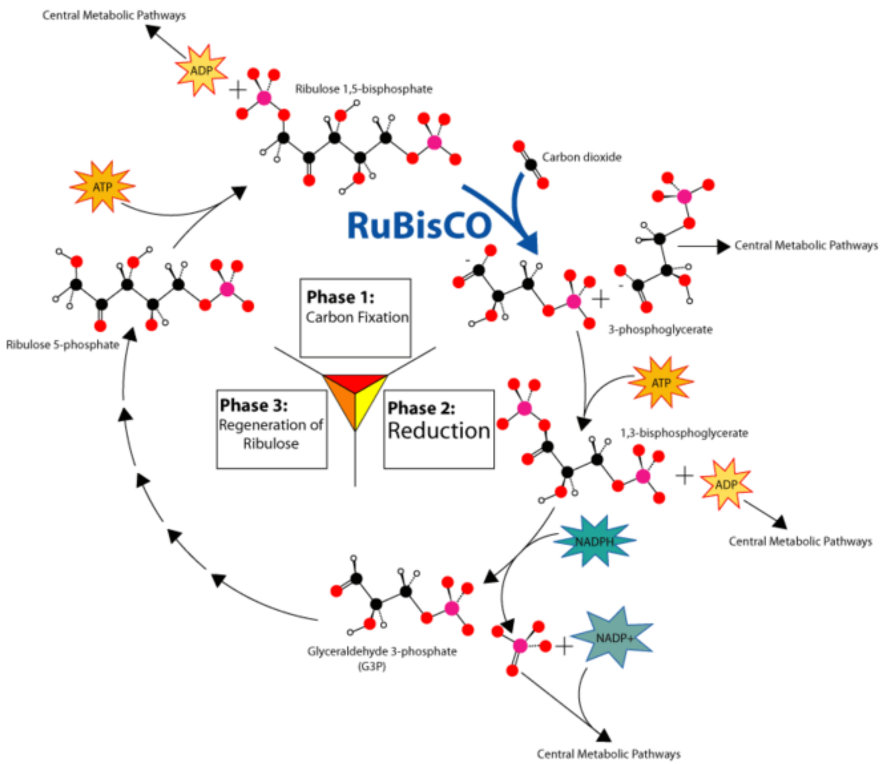
What are the types of Photosynthesis?

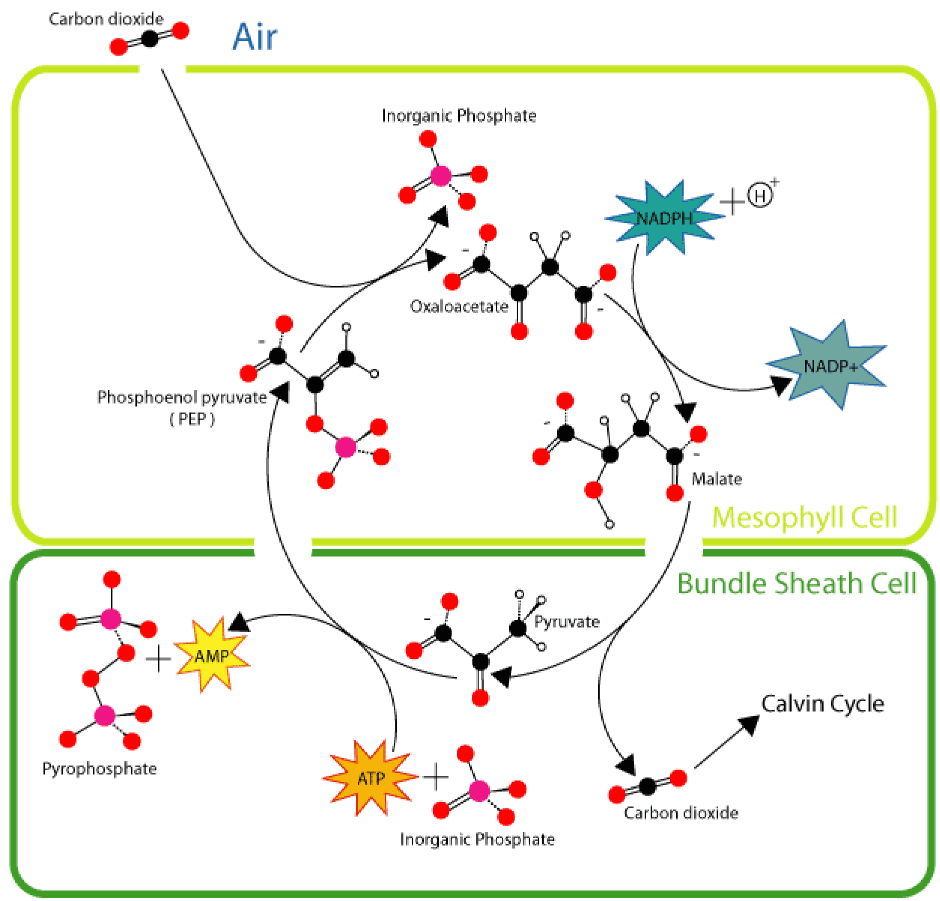
- C3: Most plants
- C4: corn and many summer plants (6X)
- CAM (Crussulacean acid metabolism): cactus

Why do these types of Photosynthesis exist?



C3 and C4 Photosynthesis





C4 plants have evolved to be more efficient in utilizing CO_2 and water. Ex. Maize, sugar cane, sorghum and millets.

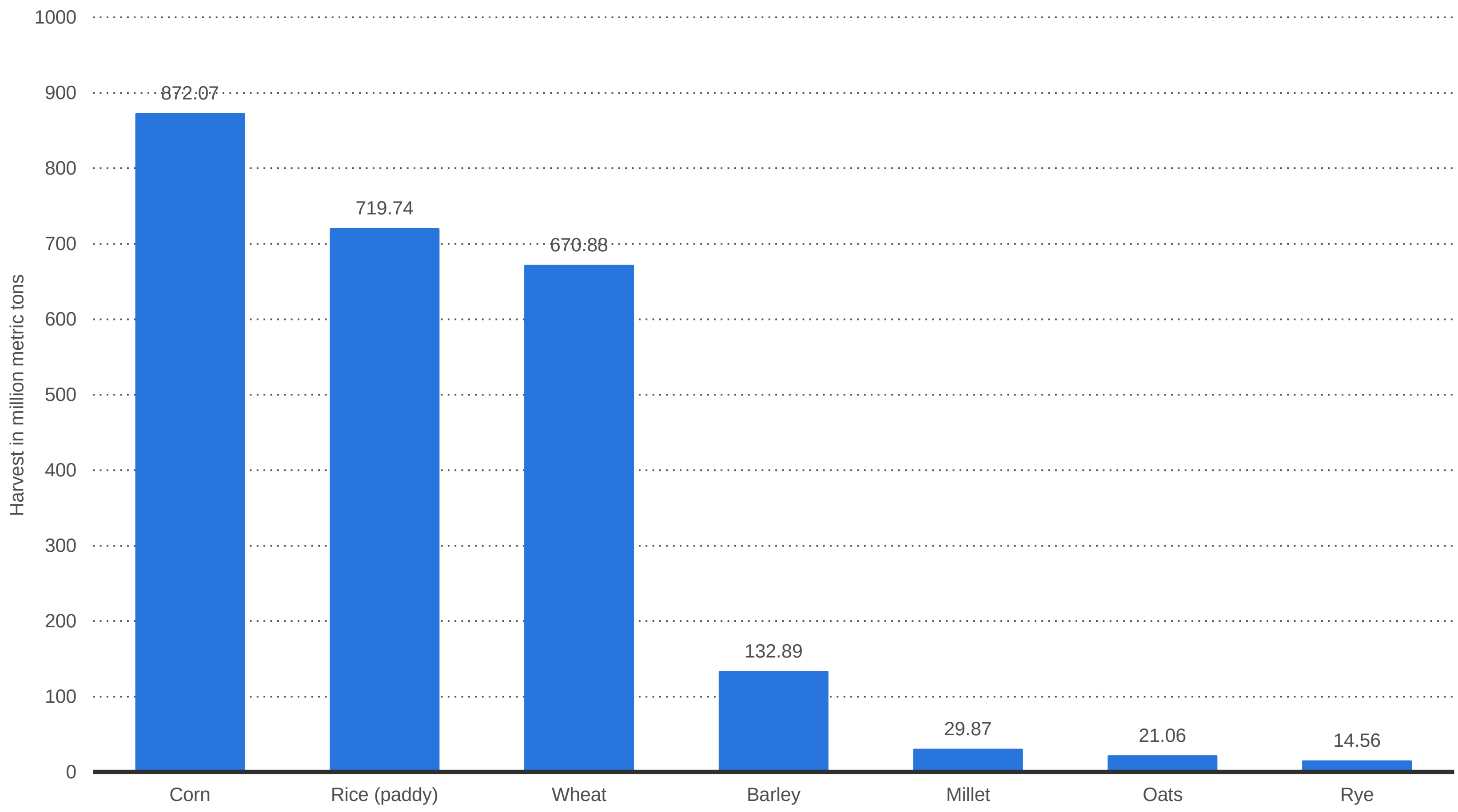
Ref: http://en.wikipedia.org/wiki/File:HatchSlackpathway.png

Feedstocks: Starch

What are the major sources of starch in the world?

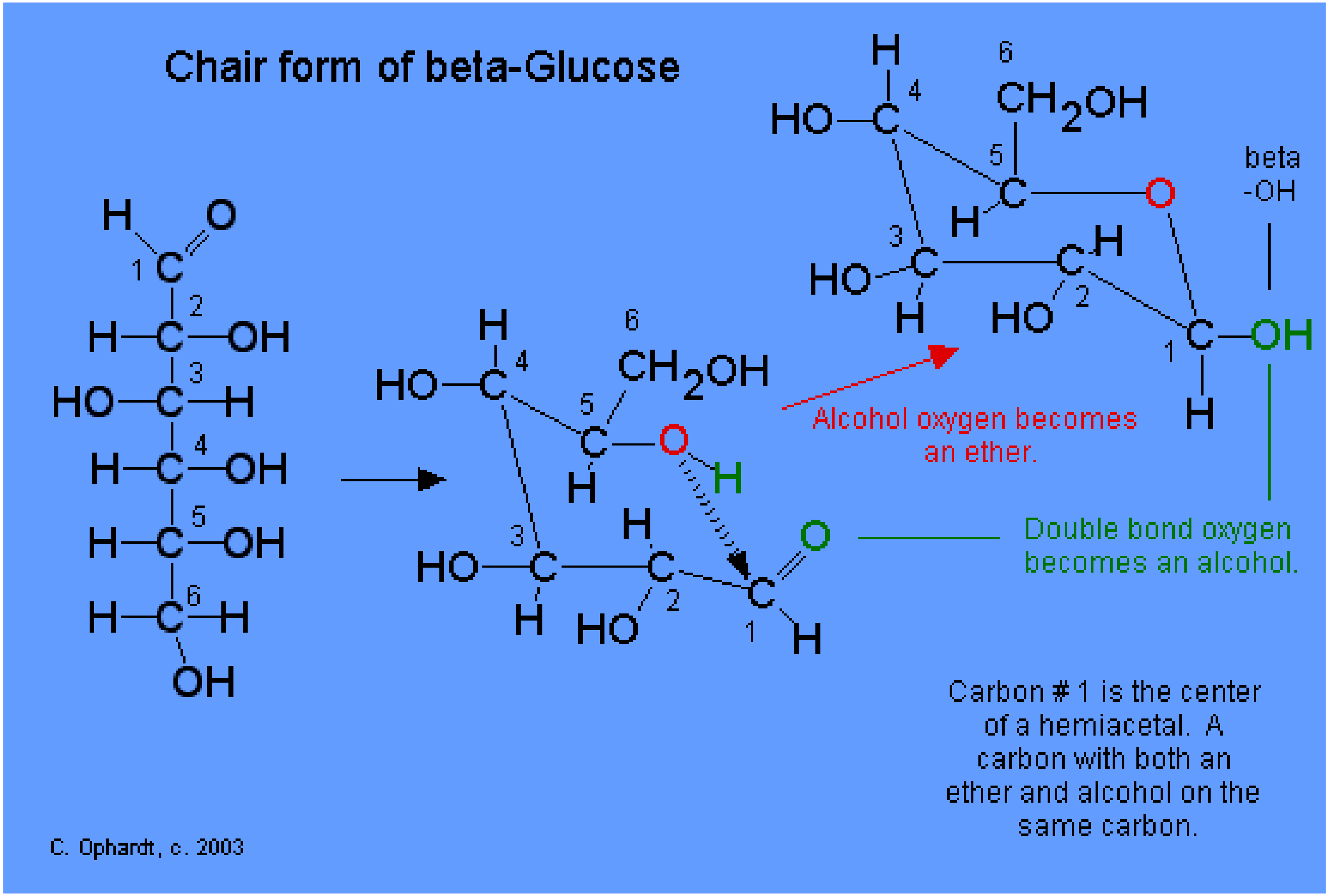
- Cereals: Wheat, corn, rice, barley, oats, sorghum
- Minor grains: Millets, rye, triticale
- Tubers and roots: Potatoes, sweet potatoes, cassava

Worldwide production of grain in 2012 (in million metric tons)



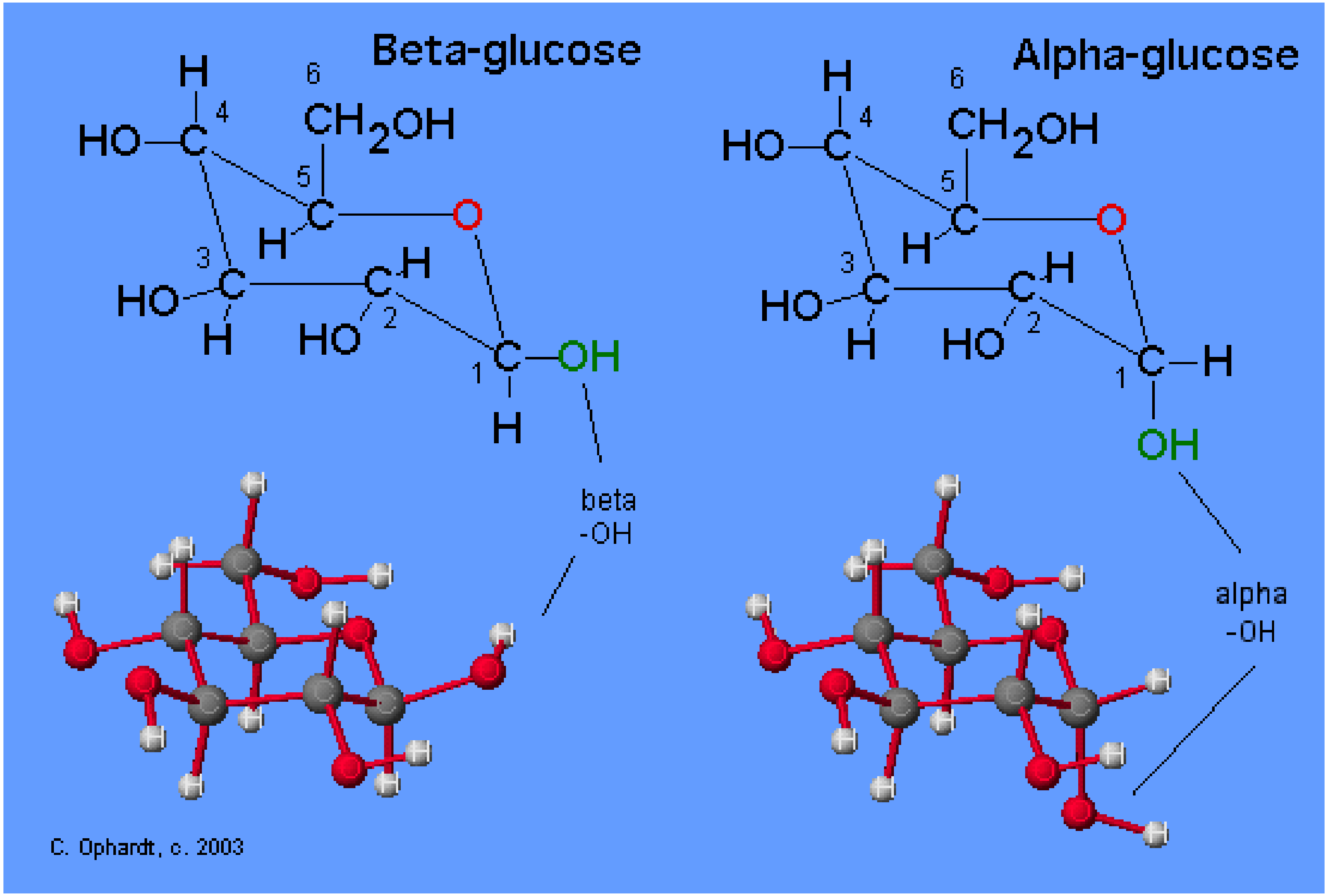
Ref: www.statista.com Source Data: FAO

Ring Structure of Glucose



Ref: http://www.elmhurst.edu/~chm/vchembook/543glucose.html

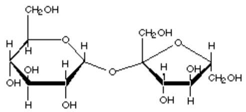
α and β Forms of Glucose



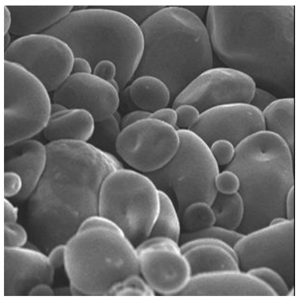
Ref: http://www.elmhurst.edu/~chm/vchembook/543glucose.html

Feedstocks: Starch

How is starch synthesized?

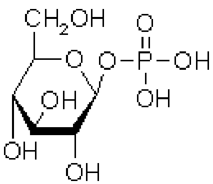


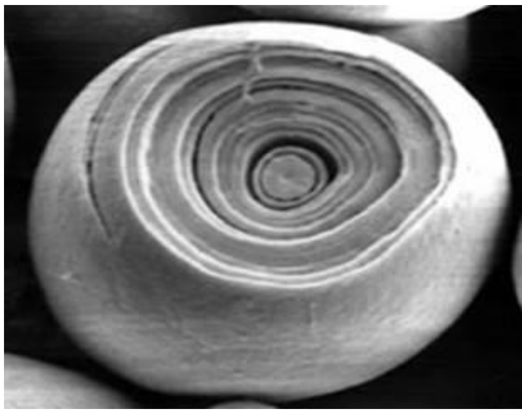
Sucrose
(glucose (α1-->2) fructose)



Sucrose

Glucose-1-phosphate





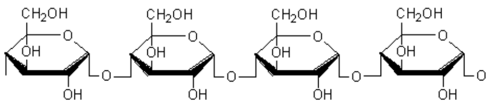
ADP-Glucose

Amyloplast membrane

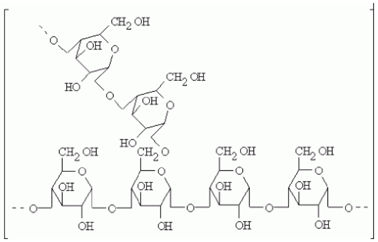
ADP-Glucose

Granule bound starch synthase

Soluble starch synthase



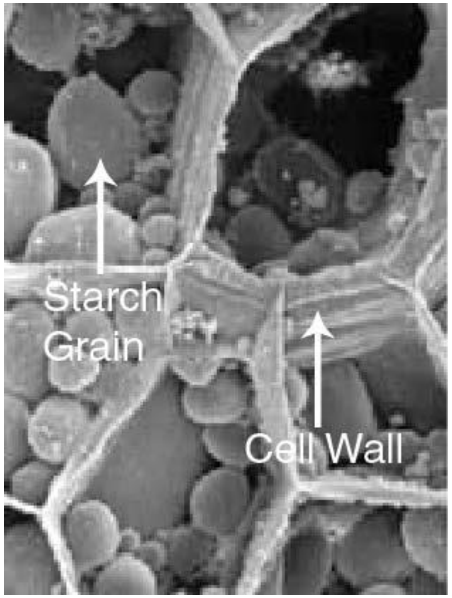
Amylose/ Amylopectin

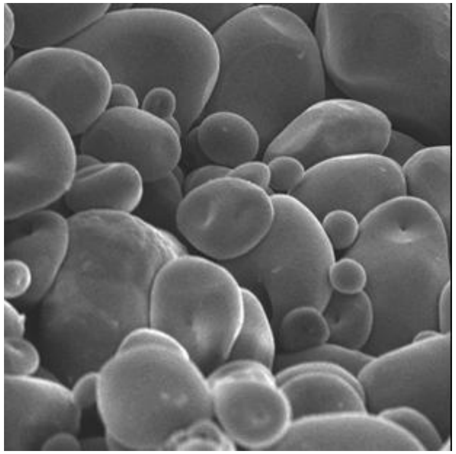


Starch Granule

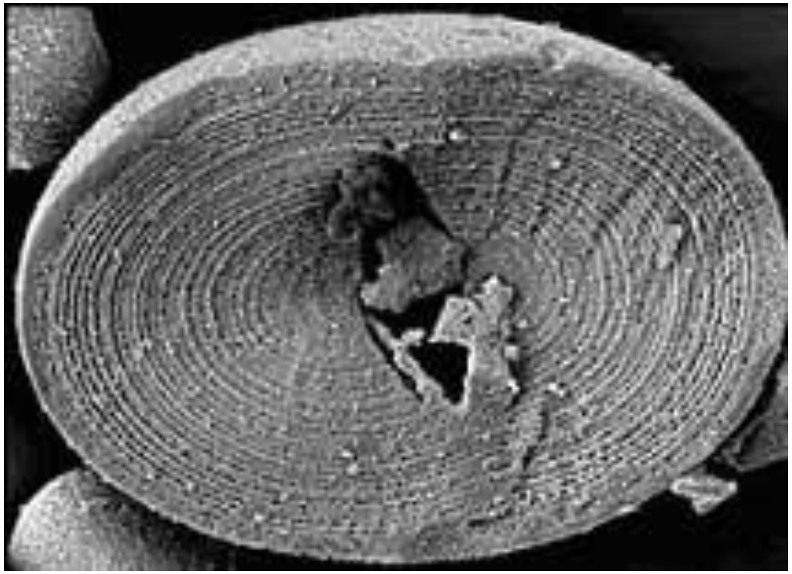
Structures and Relative Length Scales in Starch

Starch granules contained within cell walls





Starch Granules



Starch granule cross-section

Courtesy: Adapted from the slides in BEEMS project

Image Sources: http://www.ansto.gov.au/research/bragg_institute/current_research/scientific_highlights/2007_mar

http://course1.winona.edu/sberg/IMAGES/potatoCell.jpg

Feedstocks: Starch

Starches can be classified based on

X-ray diffraction pattern:

A-type: most cereal starches belong to this class

B-type: Tuber, legume, root, fruits (Banana) and stem starches

C-type Peas and beans (a combination of A- and B-type starches)

Shape: Lenticular (A-type starches, wheat), spherical (B-type, sorghum), polygonal (oats, millets), irregular (amylomaize)

Size distribution: Uni-modal (maize, rice) or Bi-modal (wheat barley, rye)

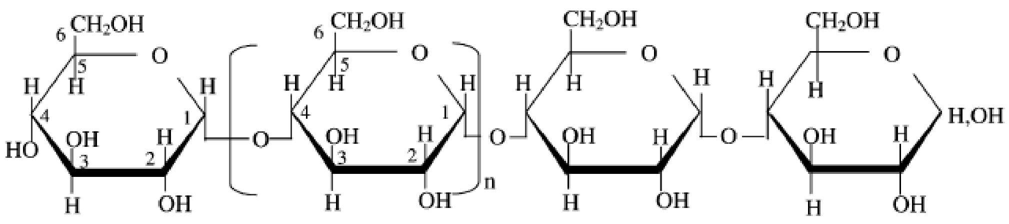
Association: Simple (wheat) or compound (rice, oat)

Composition: α-glucan, lipid, moisture, protein and mineral content

Ref: Tester et al (2004)

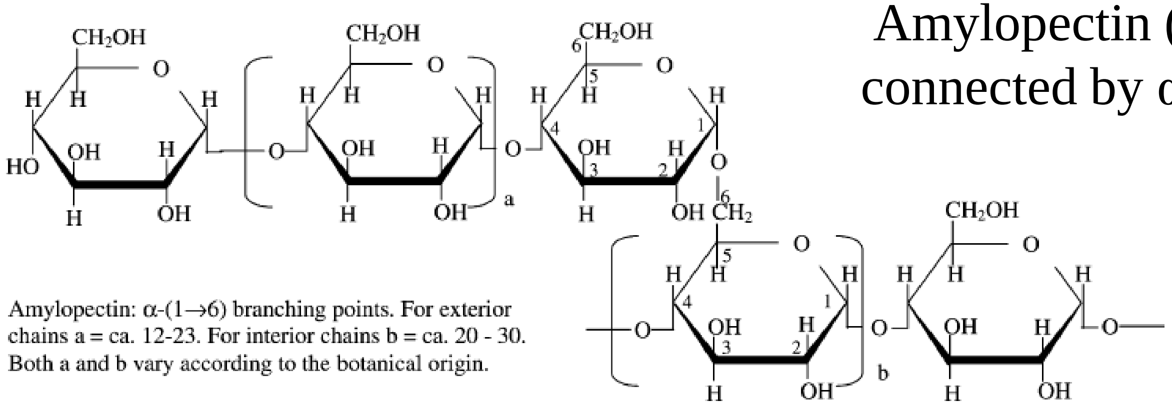
Feedstocks: Starch

Amylose and amylopectin are two polymers of glucose that constitute starch. Depending on biological source, starch can have various amounts of these polymers thus resulting in different physical and chemical properties.



Amylose: α-(1→4)-glucan; average n = ca. 1000. The linear molecule may carry a few occasional moderately long chains linked α-(1→6).

Amylose (glucose molecules linked by α 1→4 bonds)



Amylopectin: α-(1→6) branching points. For exterior chains a = ca. 12-23. For interior chains b = ca. 20 - 30. Both a and b vary according to the botanical origin.

Amylopectin (branches are connected by α 1→6 bonds)

Ref: Tester et al (2004)

Feedstocks: Starch

Amylose

- Linear polymer of glucose
- Glucose molecules linked by α 1→4 bonds (>99% bonds)
- Mol. Wt 1×10^5- 1×10^6
- DP* is 324-4920
- Forms a blue complexes with iodine
- Complexes with lipids, proteins inside starch granules
- Found in amorphous regions of starch granule

Amylopectin

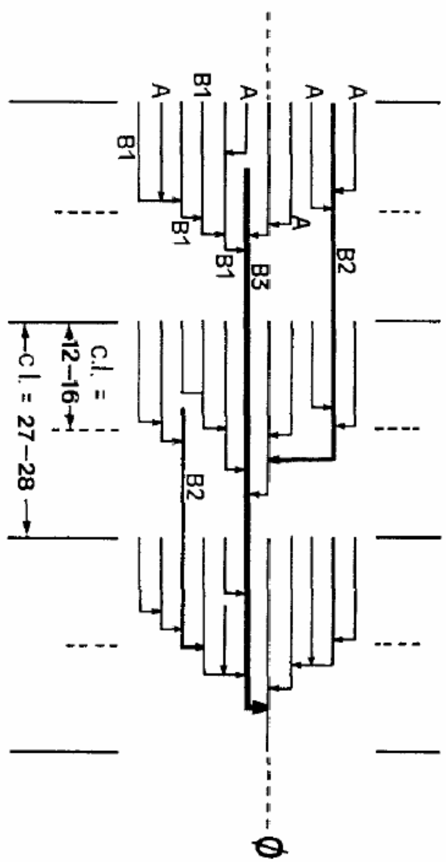
- A highly branched polymer
- Branches are connected by α 1→6 bonds
- Mol. Wt 1×10^7 -1×10^9
- DP* is 9600-15,900
- Branch characteristics are dependent on the biological source
- Crystallinity of starch is determined by amylopectin characteristics

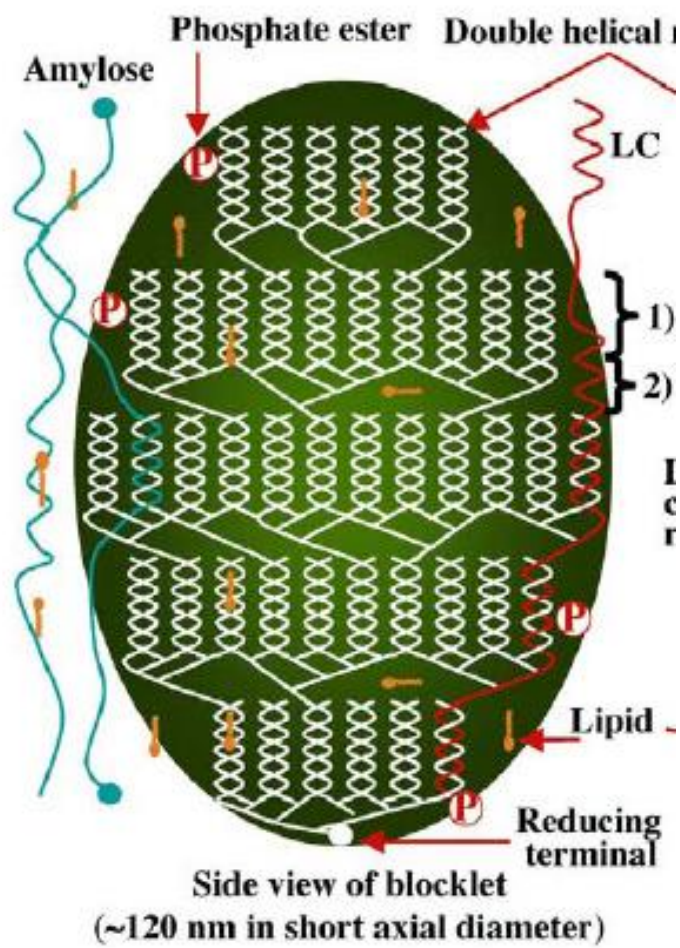
*DP=degree of polymerization. Average number of glucose units for one molecule

Ref: Tester et al (2004)

Feedstocks: Starch

Amylose and amylopectin molecules form double helix structures that form alternate crystalline and amorphous regions

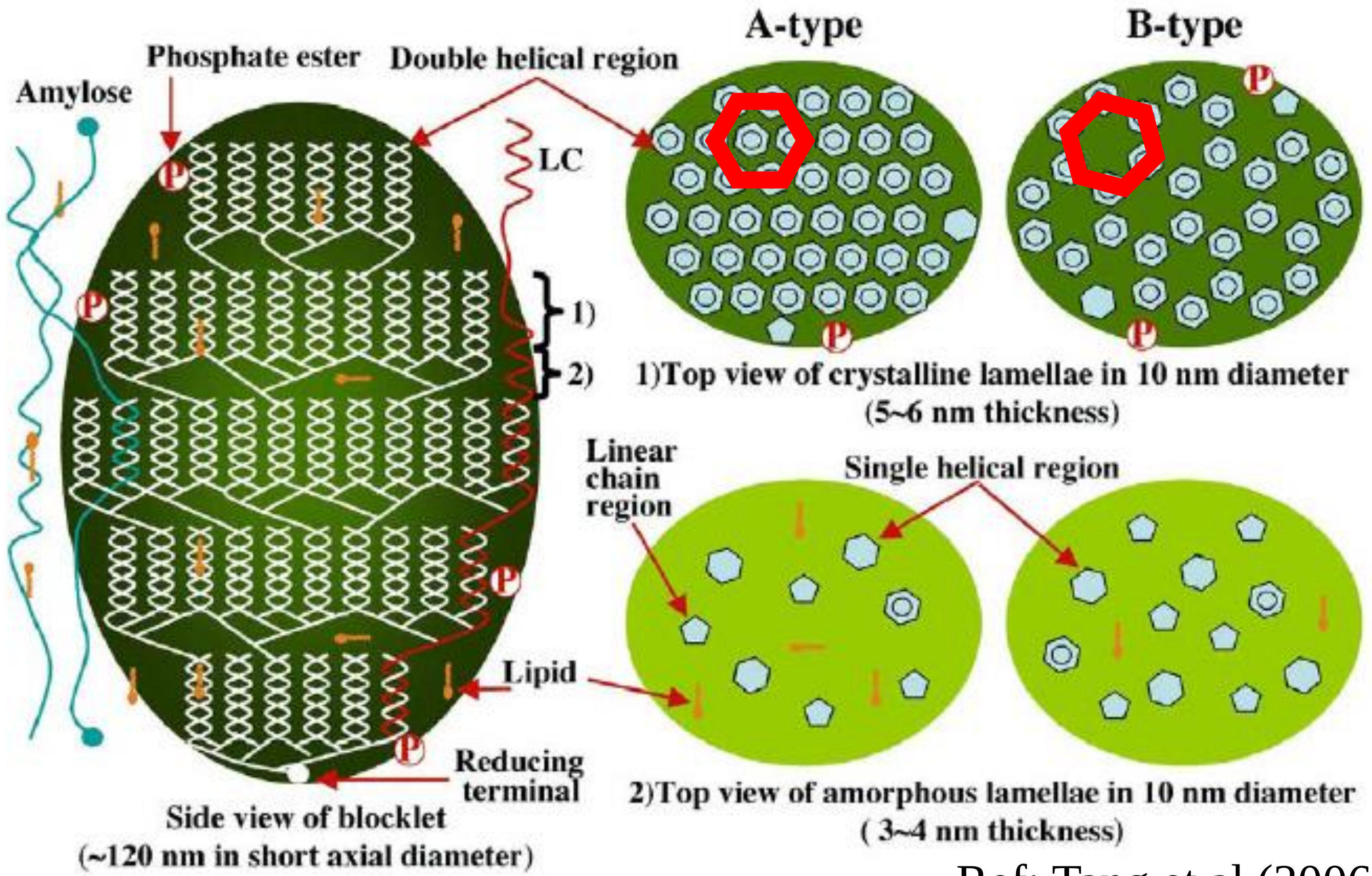




Ref: Hizkuri (1986); Tang et al (2006)

Feedstocks: Starch

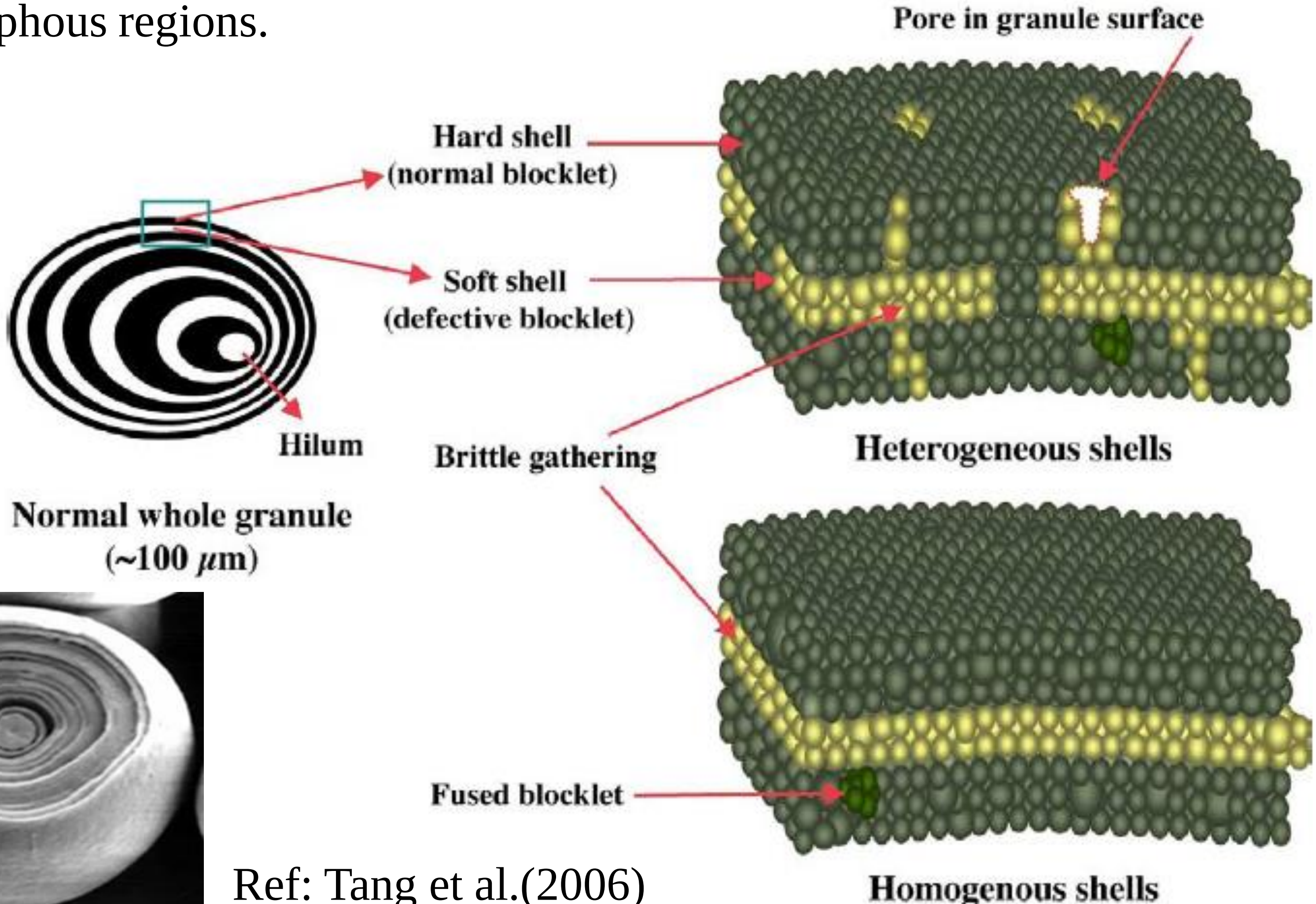
Further, these structures are also responsible for the formation of A and B type starches

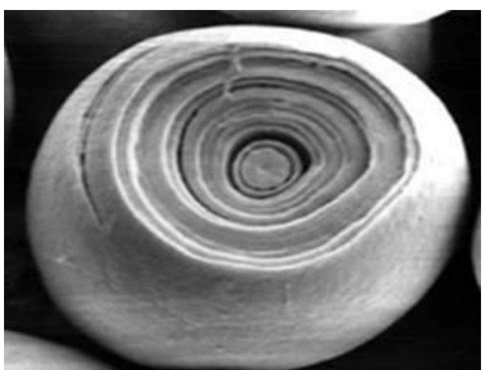


Ref: Tang et al.(2006)

Feedstocks: Starch

Amylpectin and amylose molecules interact, they alternating crystalline and amorphous regions.

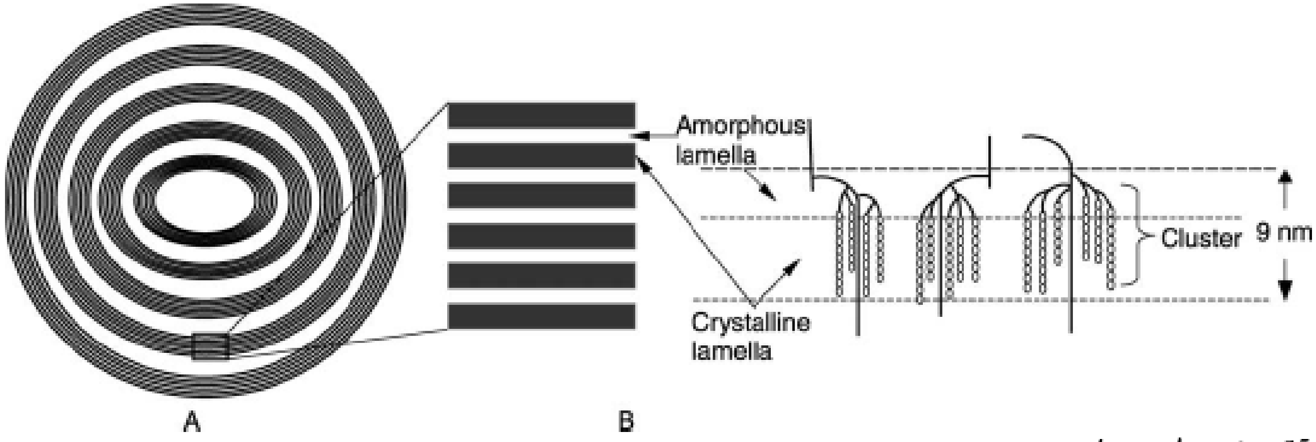


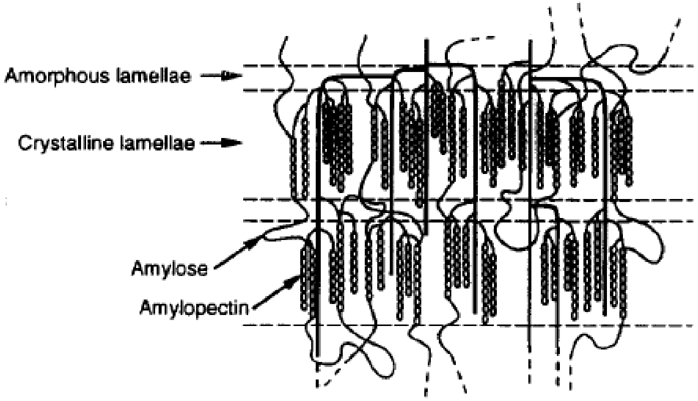


Ref: Tang et al.(2006)

Feedstocks: Starch

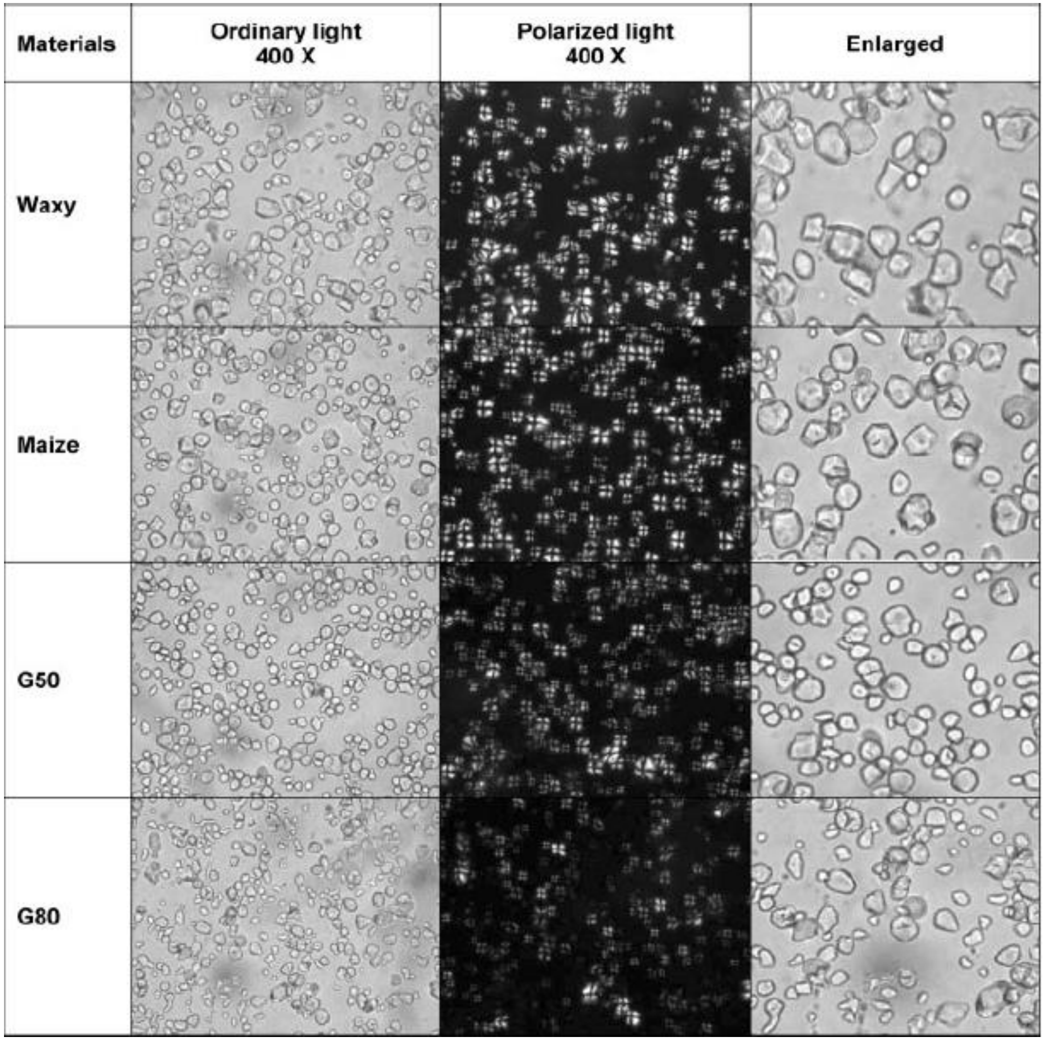
Degree of crystallinity is in the range of 17-51% depending on the preparation, moisture content among other factors. There is no definitive proof that amylose content has significant effect on crystallinity.





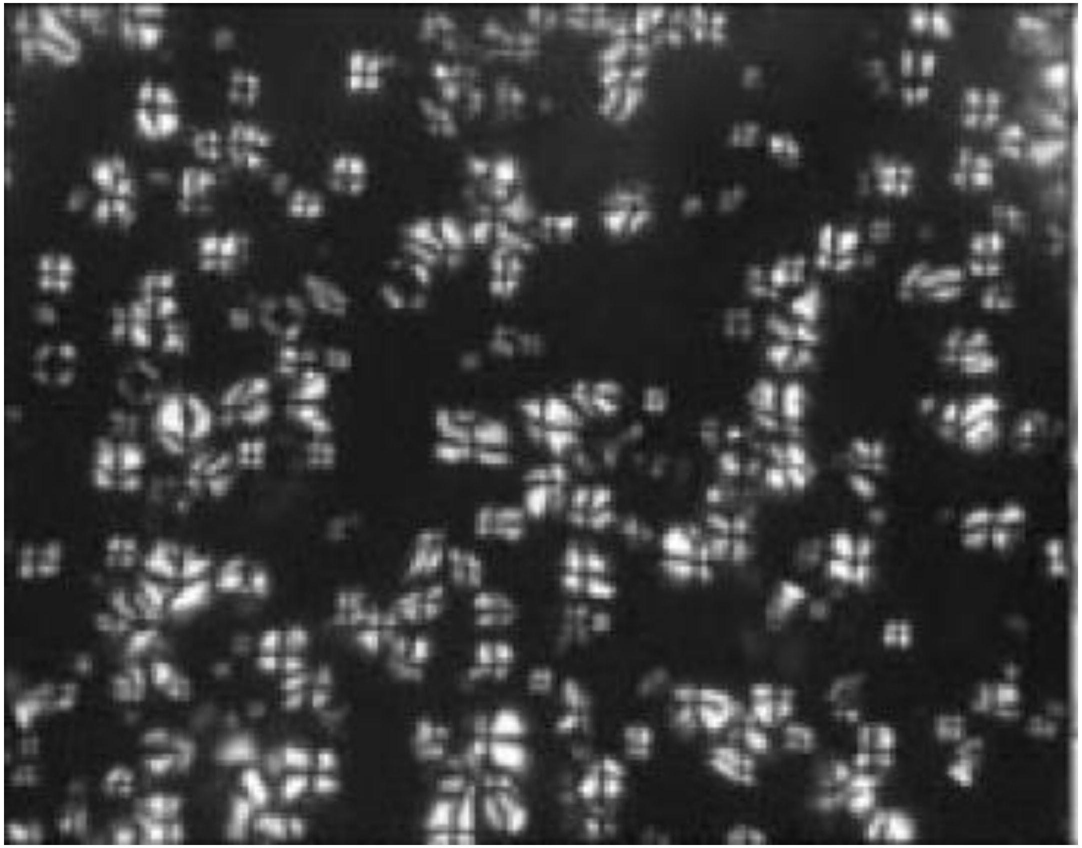
Ref: Tang et al.(2006)

Feedstocks: Starch



Ref: Chen et al.(2006)

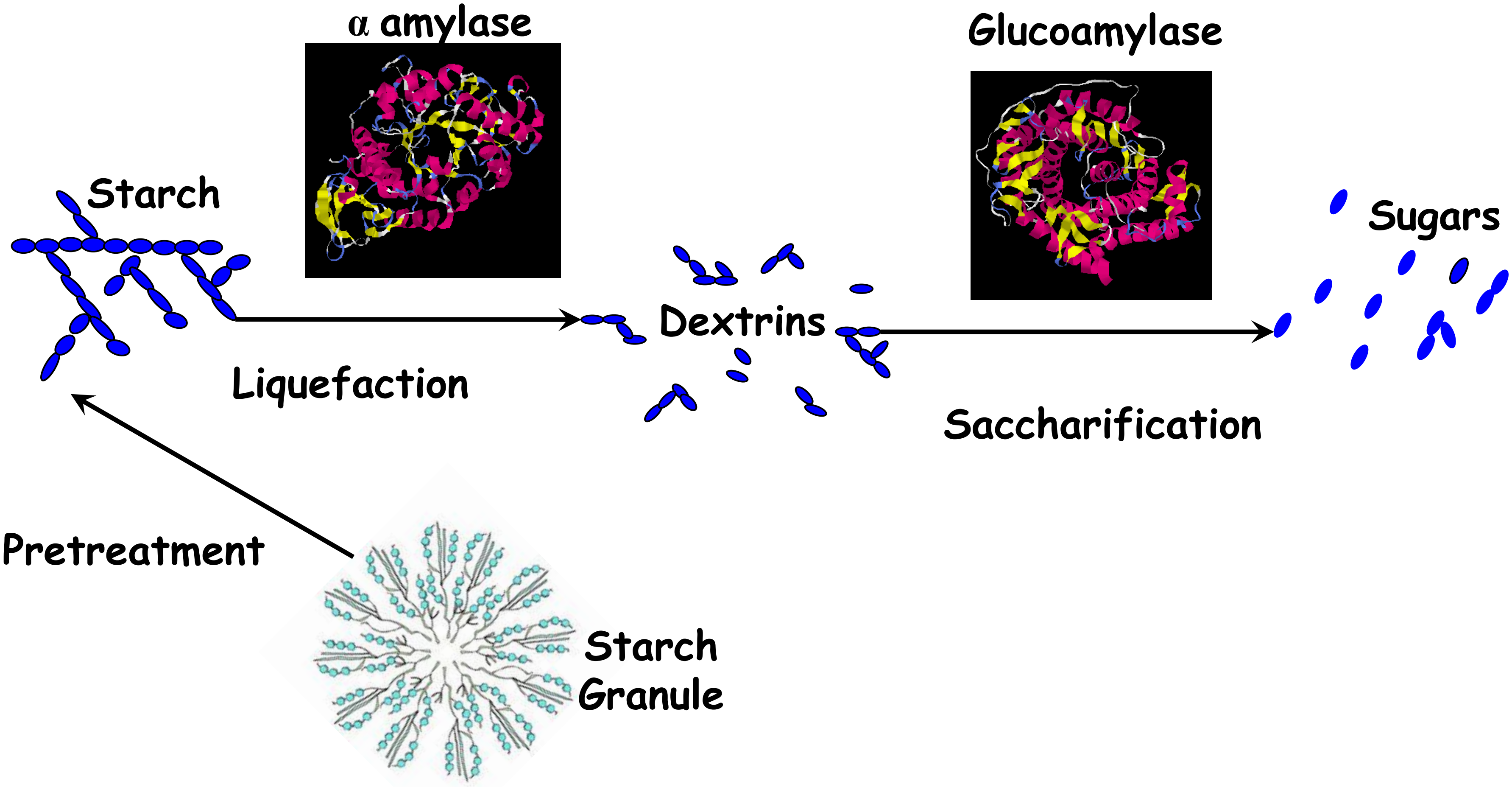
Feedstocks: Starch



Birefringence in wheat starch

Ref: Chen et al.(2006)

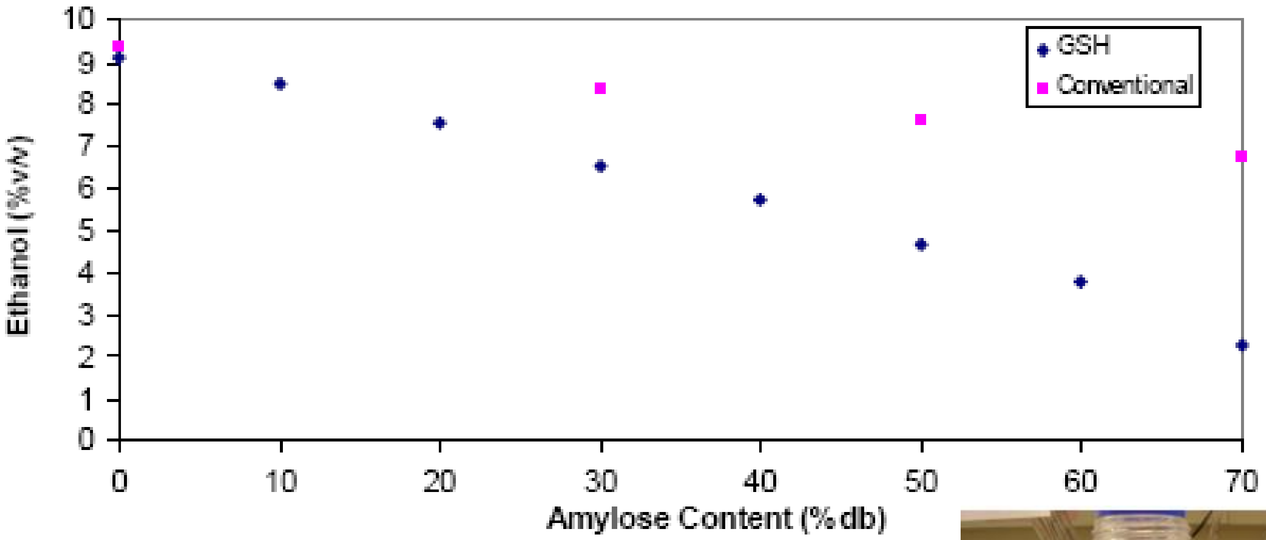
Feedstocks: Starch

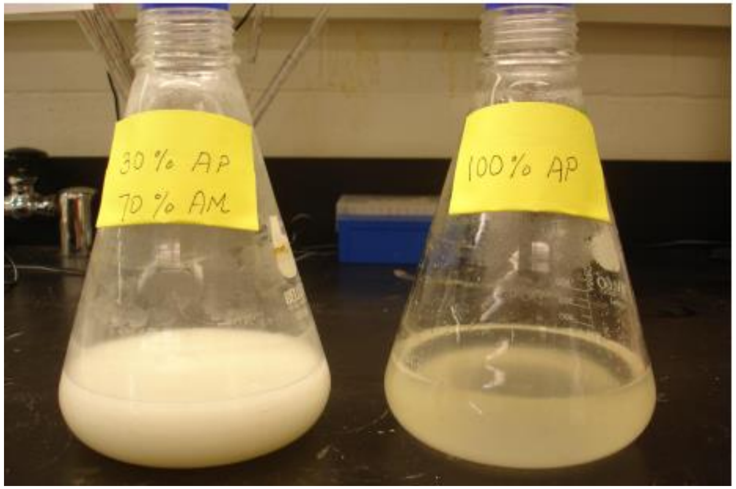


Ref: http://food.oregonstate.edu/learn/starch.html

Feedstocks: Starch

Enzymatic hydrolysis is dependent on the composition of starch.





Ref: Sharma et al. (2006)

Feedstocks: Starch

Amylases

- α-amylase (EC 3.2.1.1) Endo enzyme
- β-amylase (EC 3.2.1.2) 1,4-α-D-glucan maltohydrolase Exo enzyme
- γ-amylase (EC 3.2.1.3) Glucan 1,4-α-glucosidase/ glucoamylase/ amyloglucosidase Exo enzyme

Pullulanases

- EC 3.2.1.41is also known as α-dextrin endo-1,6-alpha-glucosidase (Debranching enzyme). Exo enzyme
- Type I act only on α-1→6 whereas type II can also act on α-1→4

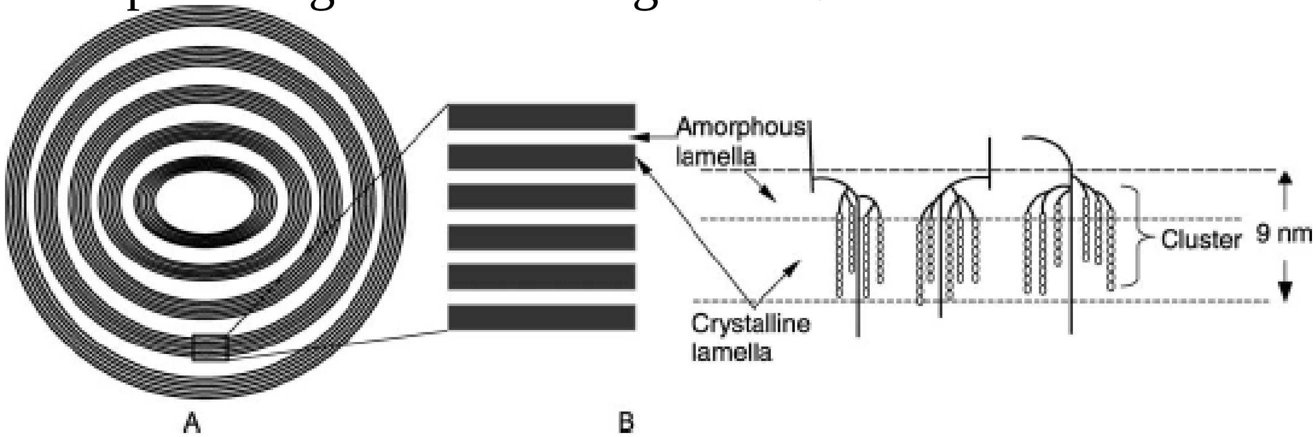
EC 3.2.1.X stands for Enzyme Commission 3(Hydrolases). 2(sugars). 1(glycoside hydrolases). Ex. EC 3.2.1.4 is a cellulase (Endo-1,4-β-D-glucanohydrolase)

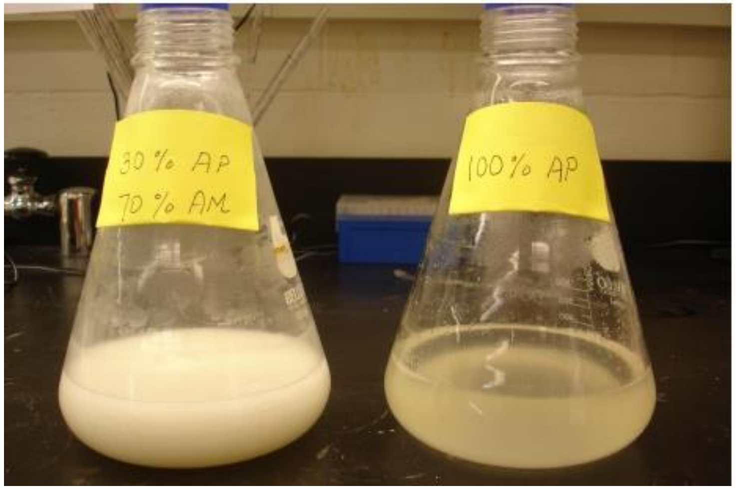
Topic 2

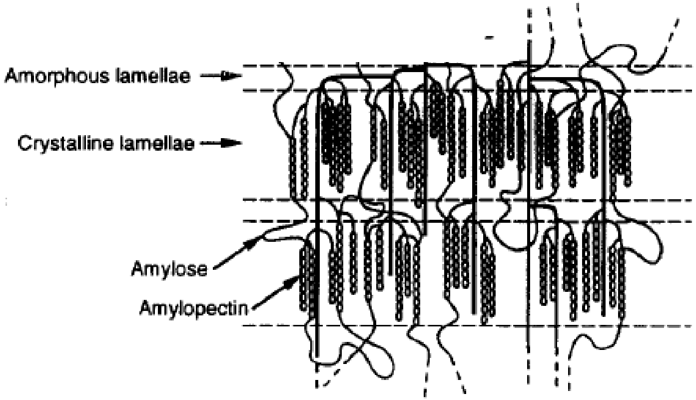
Cellulose, Hemicellulose and Lignin

Summary of Previous Lecture

Synthesis of starch in plants. Amylose and amylopectin. Crystalline and amorphous regions in starch granules.







Enzymatic hydrolysis is dependent on the composition of starch.

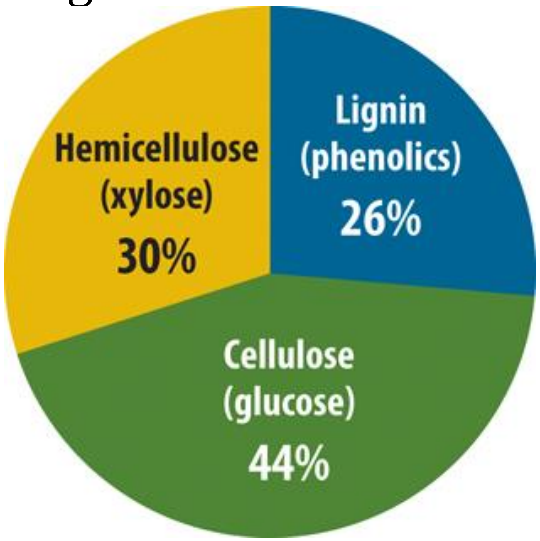
Cellulosic Feedstocks

What are the main constituents of lignocellulosic feedstocks?

- Cellulose: Homopolymer of glucose consisting of β(1→4) bonds. It differs from another polymer of glucose, starch that consists of α(1→4) bonds

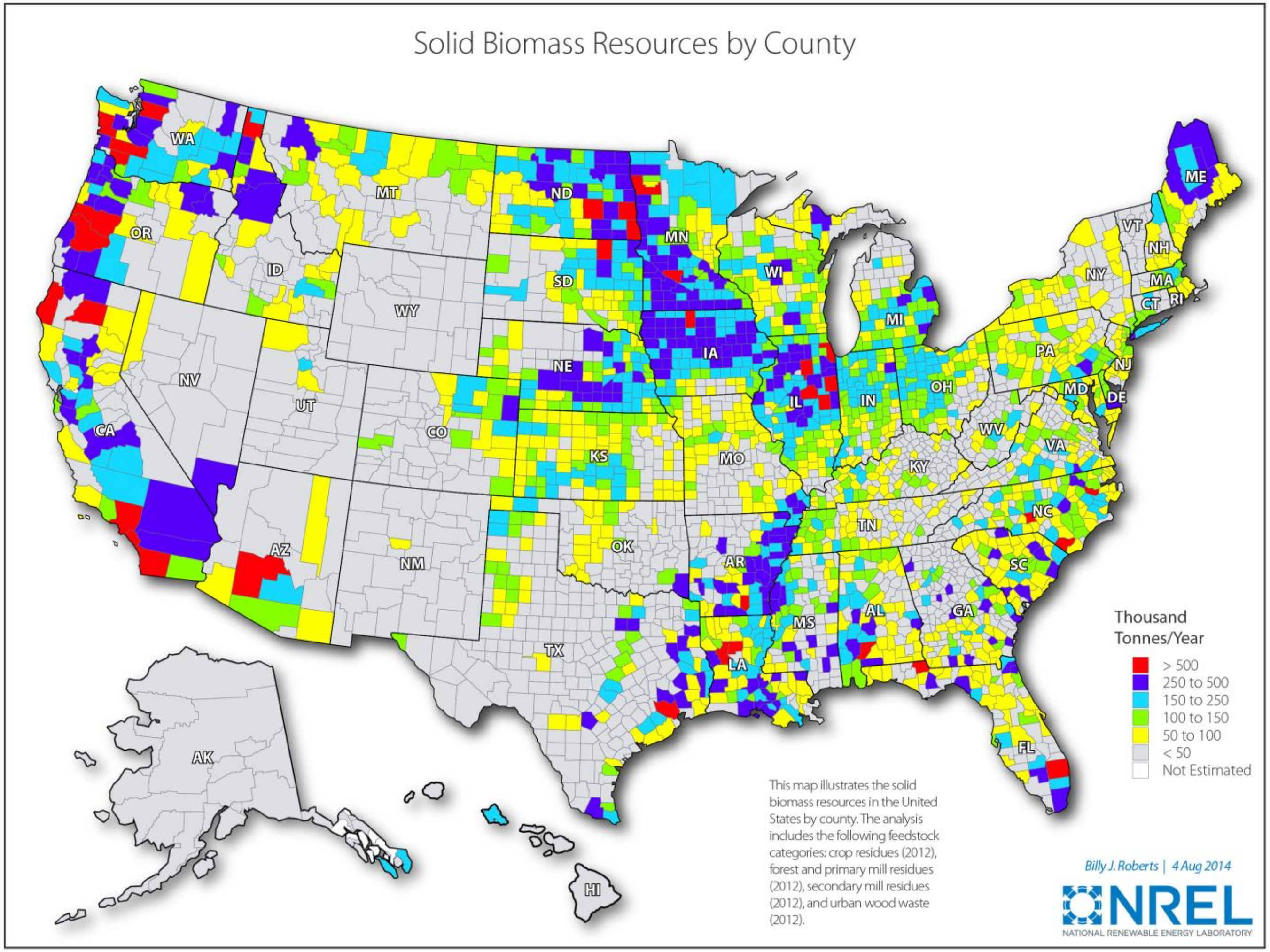
- Hemicellulose: Heteropolymer of pentoses (xylose and arabinose) and hexoses (glucose, galactose, mannose) and sugar acids (acetic). Hardwood hemicelluloses are xylans whereas softwood consists of glucomannans

- Lignin: A racemic, heteropolymer consisting of three hydroxycinnamyl alcohol monomers differing in their degree of methoxylation: p-coumaryl, coniferyl and sinapyl alcohols



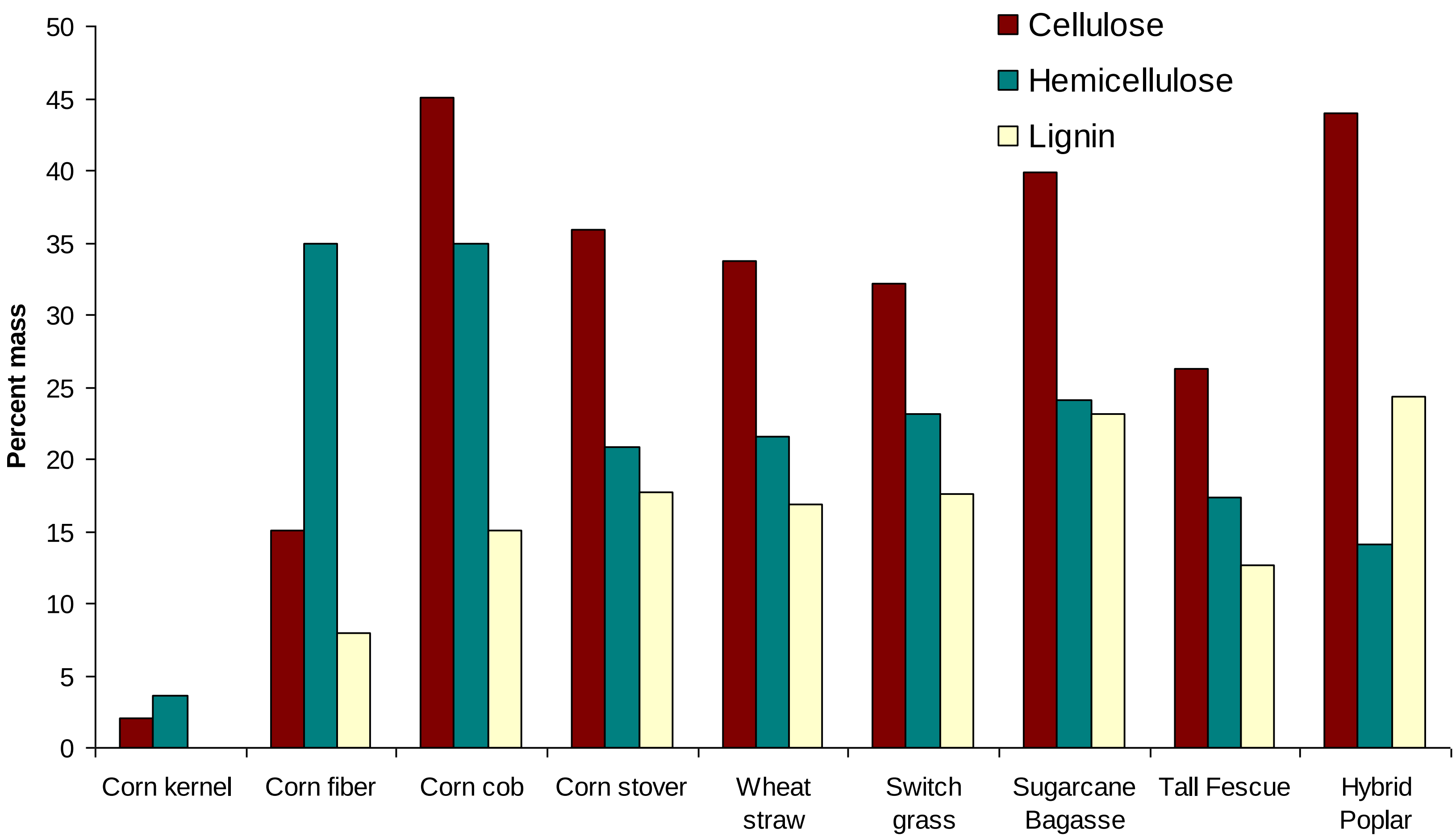
Ref: http://bioenergy.ornl.gov/main.aspx and GMIS, Oak Ridge National Laboratory

Cellulosic Feedstocks



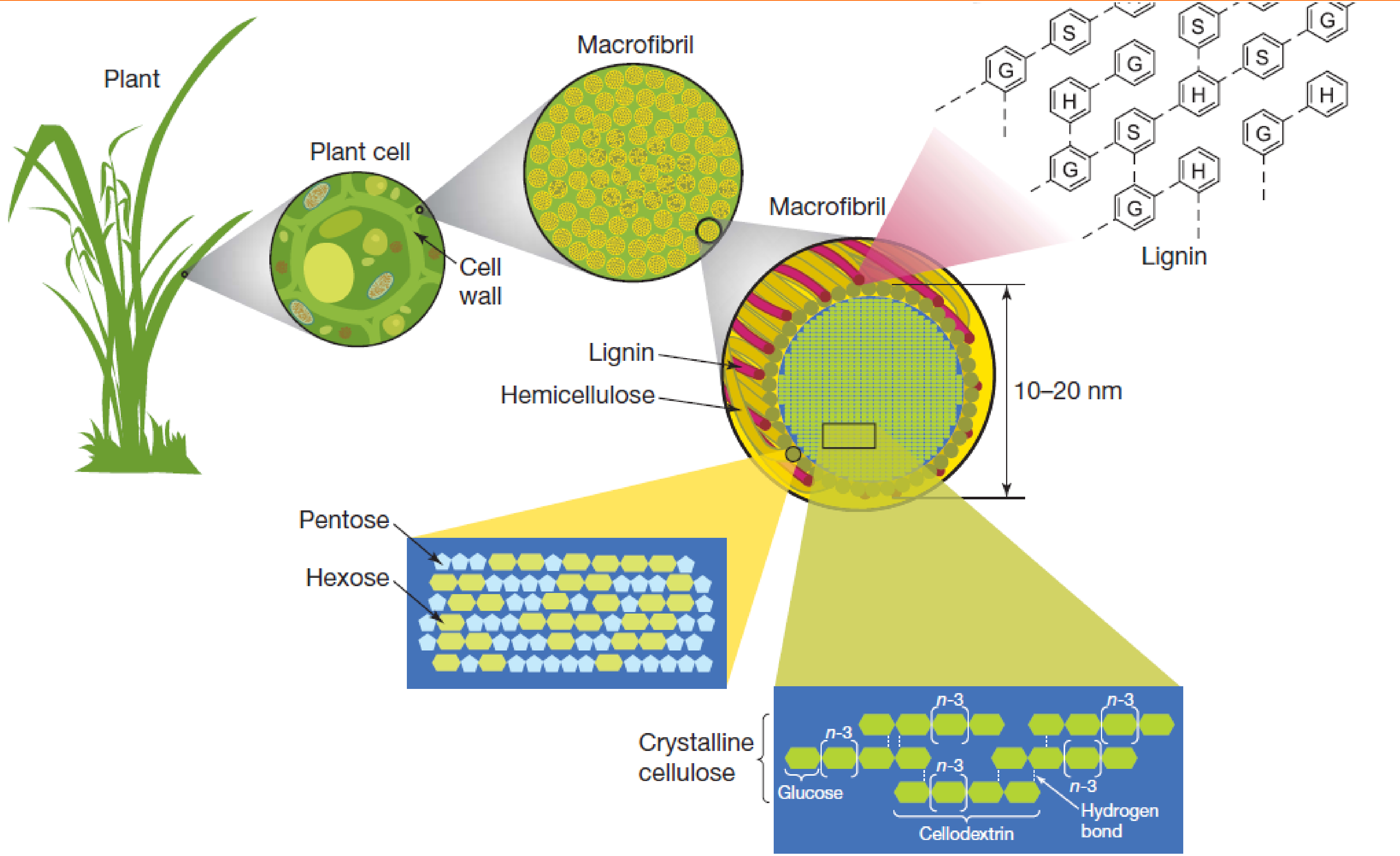
Ref: http://www.nrel.gov/gis/biomass.html

Cellulosic Feedstocks

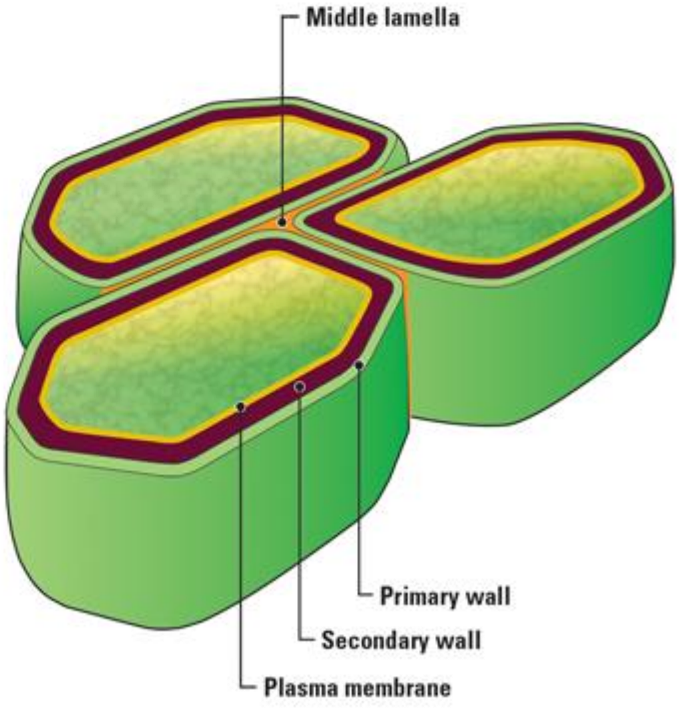


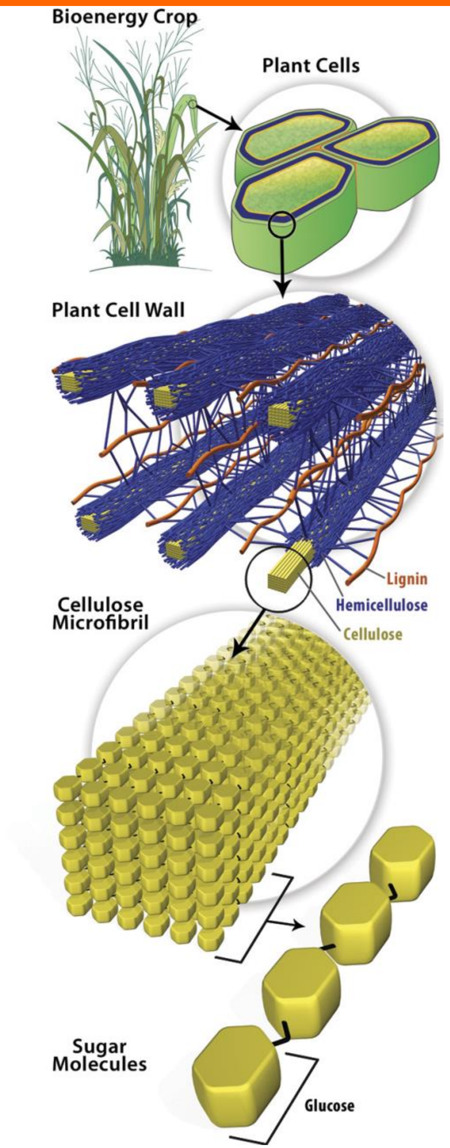
Ref: http://www.nrel.gov/biomass/data_resources.html

Plant Cell Model



Plant Cell Model

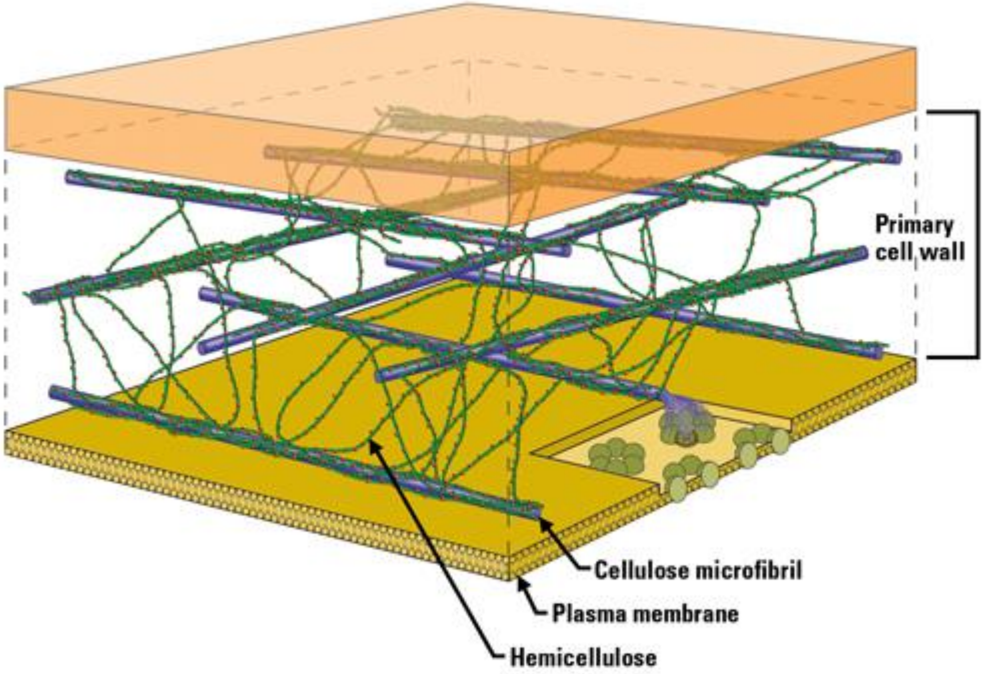




Ref: http://genomicsgtl.energy.gov/biofuels/.

Plant Cell Model

A simplified model of plant cell wall



Ref: http://genomicsgtl.energy.gov/biofuels/.

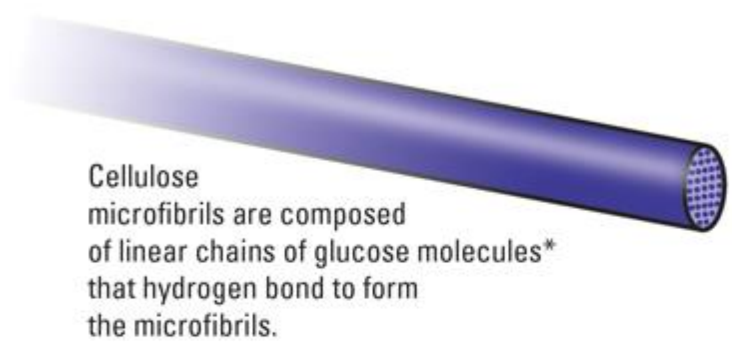
Cellulosic Feedstocks

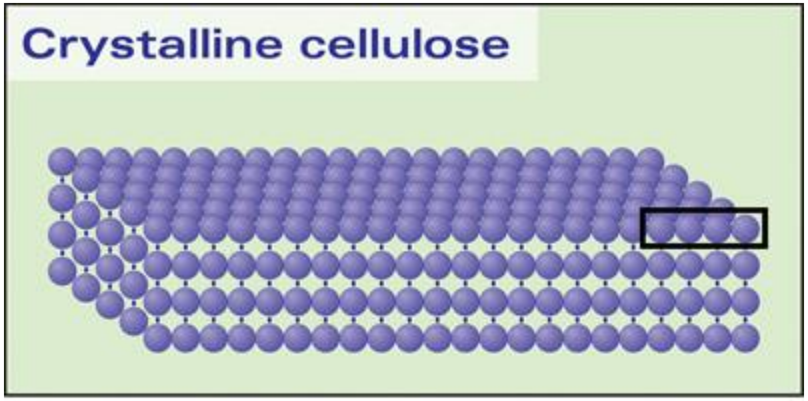
Ref: http://www.eurocode2.info/images/reinforcement.jpg

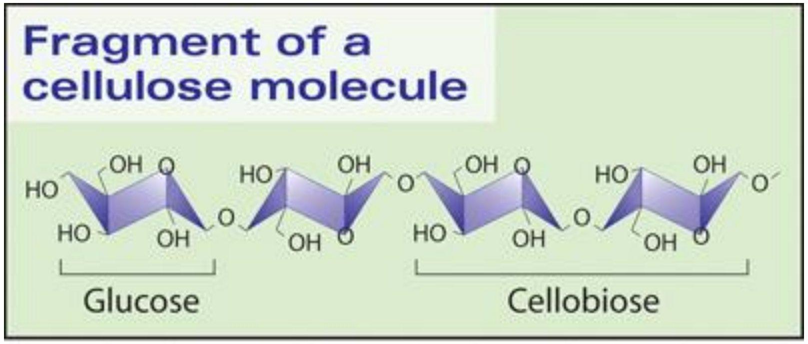
http://pro.corbis.com

Cellulose

Cellulose: Homopolymer of glucose consisting of β(1→4) bonds. It differs from another polymer of glucose, starch that consists of α(1→4) bonds . Callose is another polymer of glucose β(1→3) bonds







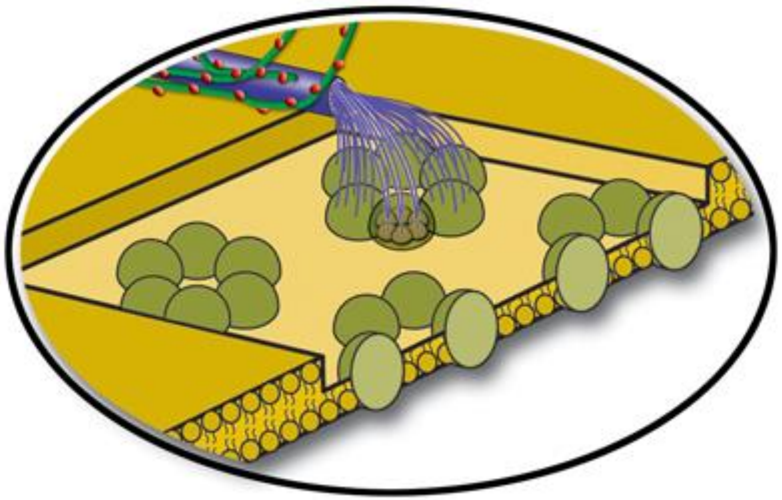
Alternating glucose residues are in an inverted orientation so the cellobiose (a disaccharide) is the repeating structural unit.

Each glucose unit is rotated by approximately 180.

Ref: http://genomicsgtl.energy.gov/biofuels/.

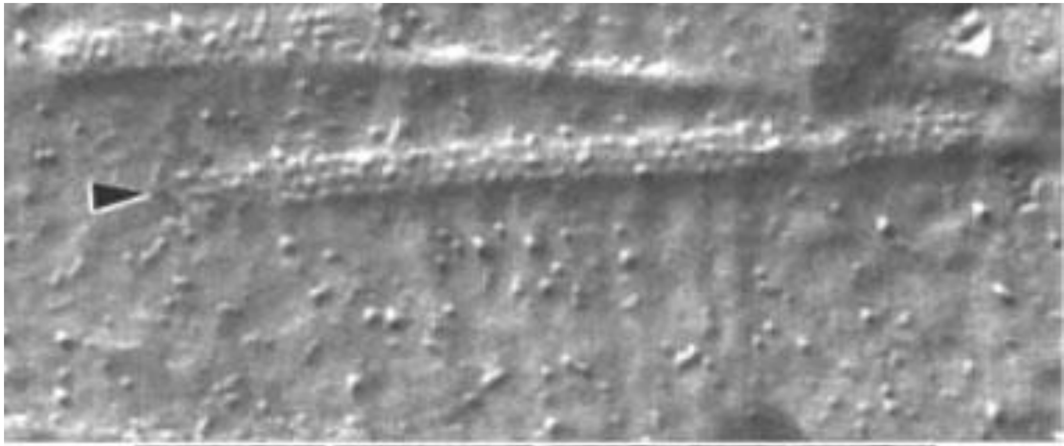
Cellulosic Synthesis

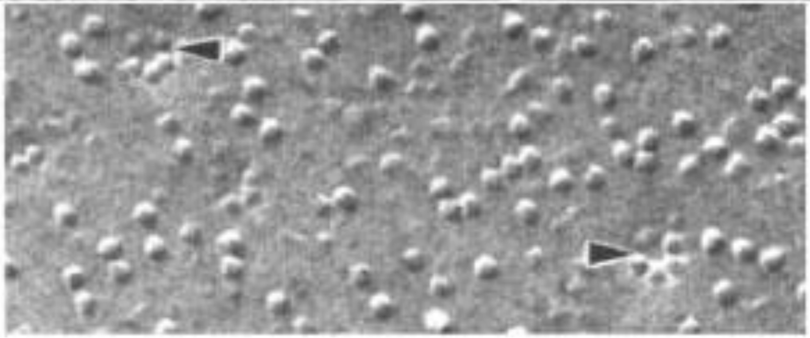
Cellulose synthase complexes are involved in cellulose microfibril synthesis. These complexes called terminal complexes (TC) are arranged in different patterns in the cell membrane.



Cellulose synthase complexes

Linear TC in *Oocystis apiculata*



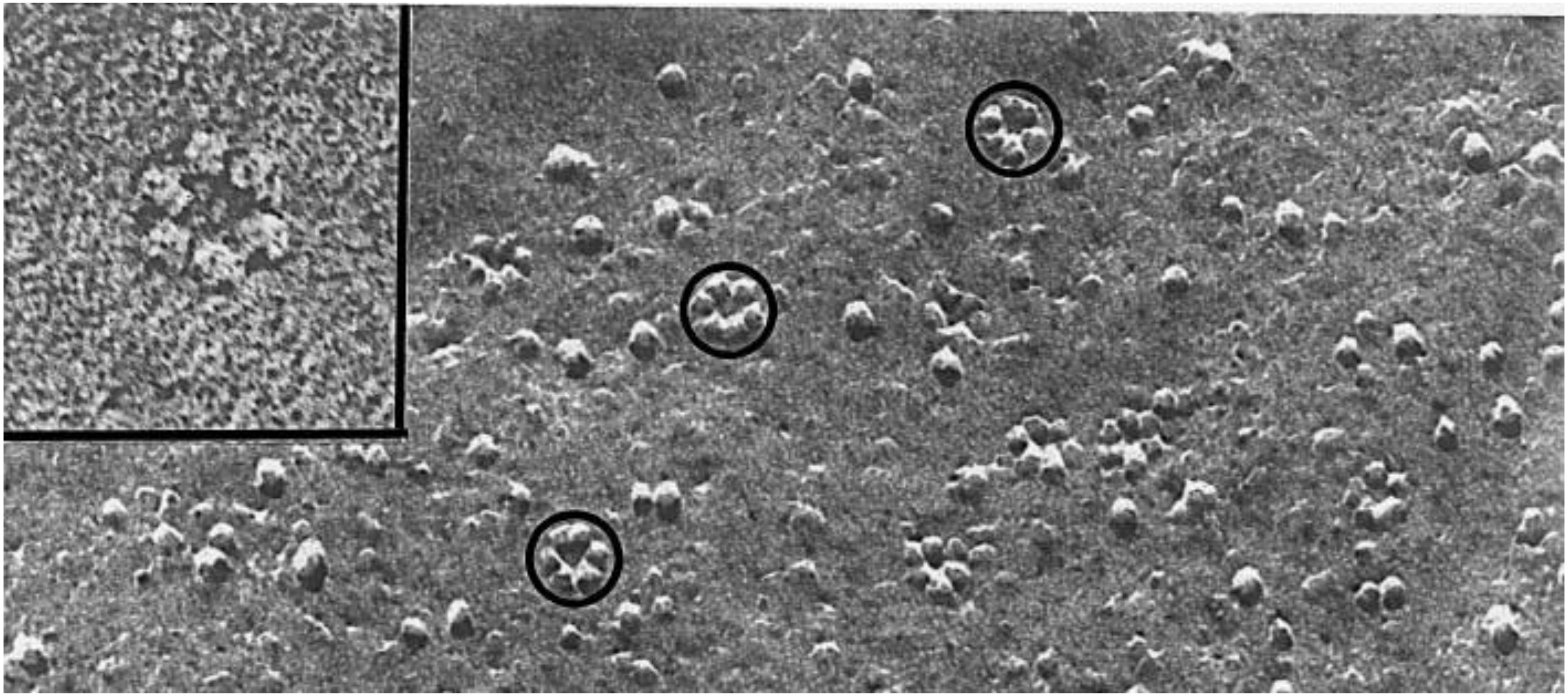


Rosette TC in *Zea mays*

Ref: http://genomicsgtl.energy.gov/biofuels/.

Cellulosic Synthesis

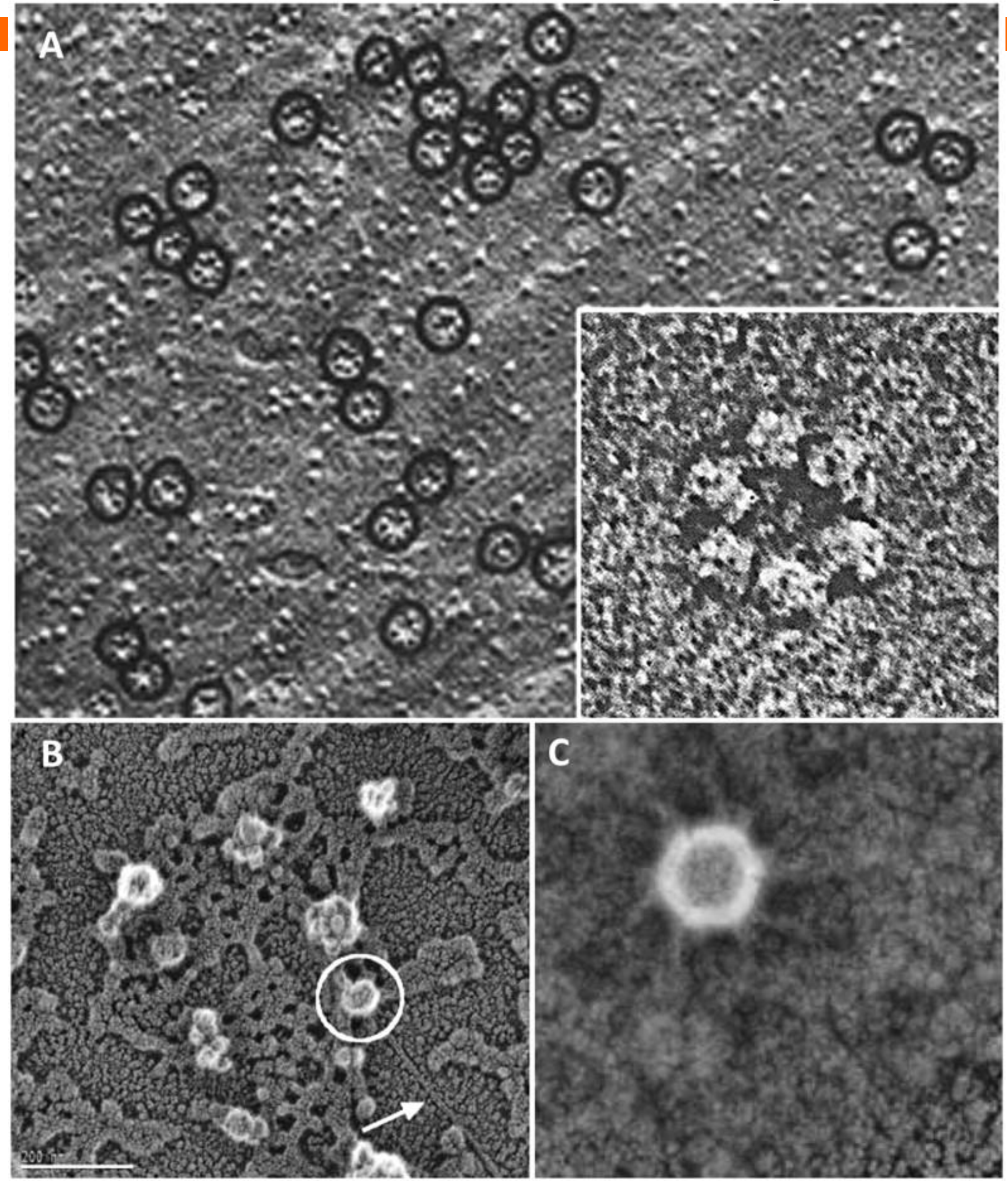
Cellulose synthase complexes are involved in cellulose microfibril synthesis. These complexes called terminal complexes (TC) are arranged in different patterns in the cell membrane.



Rosette TC in *Zinnia elegans*

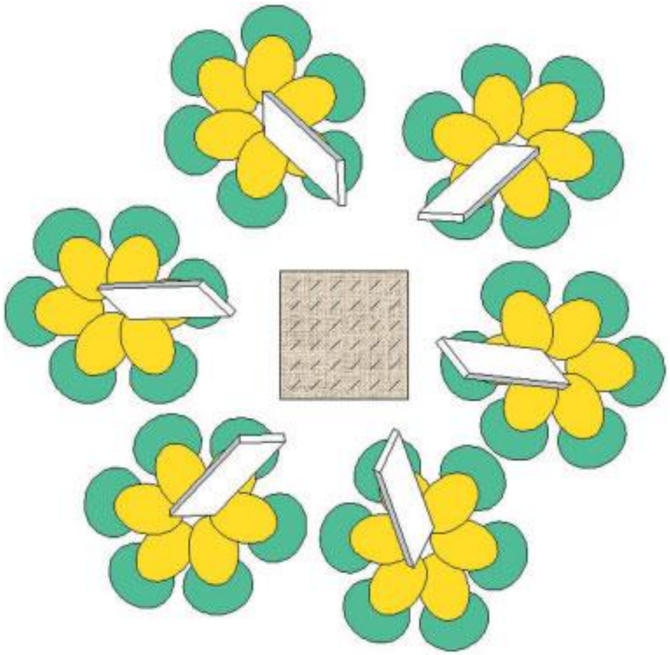
Ref: Delmer 1999.

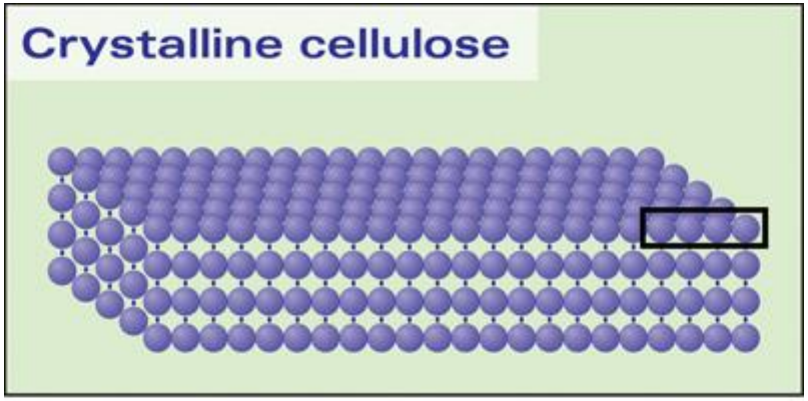
Particle rosette structures associated with cellulose synthesis in angiosperms.



Cellulose Synthesis

Model for microfibril formation with 36 glucan chains (elementary fibril) with each rosette subunit forming a sheet of 6 glucan chains. DP of the chains varies from 2000-25000.





The glucan chains contain thousands of glucose residues.

Ref: http://genomicsgtl.energy.gov/biofuels/. Brown, 2003.

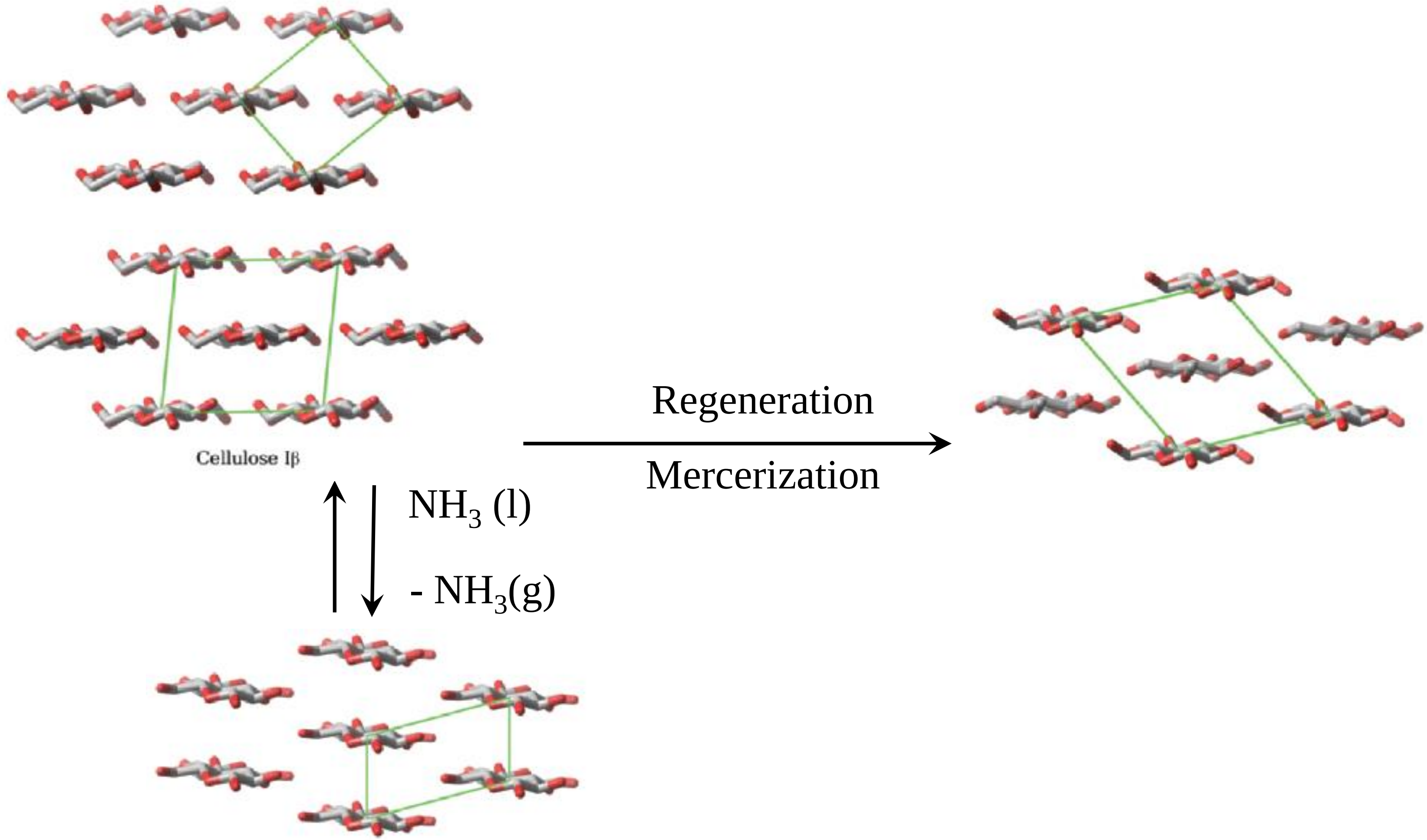
Cellulose Polymorphs

Cellulose: There are six polymorphs of cellulose. Cellulose I occurs naturally while the polymorphs are formed by physical treatments. Cellulose II is thermodynamically most stable form. Polymorphs are identified by X-ray or electron diffraction.

$$\begin{tikzcd}
\text{Cellulose I} \arrow[rr, "\text{Regeneration}", "\text{Mercerization}"'] \arrow[d, shift left, "NH_3\,(l)"] & & \text{Cellulose II} \arrow[d, shift left, "NH_3\,(l)"] \\
\text{Cellulose III}_{I} \arrow[u, shift left, "-\,NH_3(g)"] \arrow[d, "\text{Heat}"] & & \text{Cellulose III}_{II} \arrow[u, shift left, "-\,NH_3(g)"] \arrow[d, "\text{Heat}"] \\
\text{Cellulose IV}_{1} & & \text{Cellulose IV}_{II}
\end{tikzcd}$$

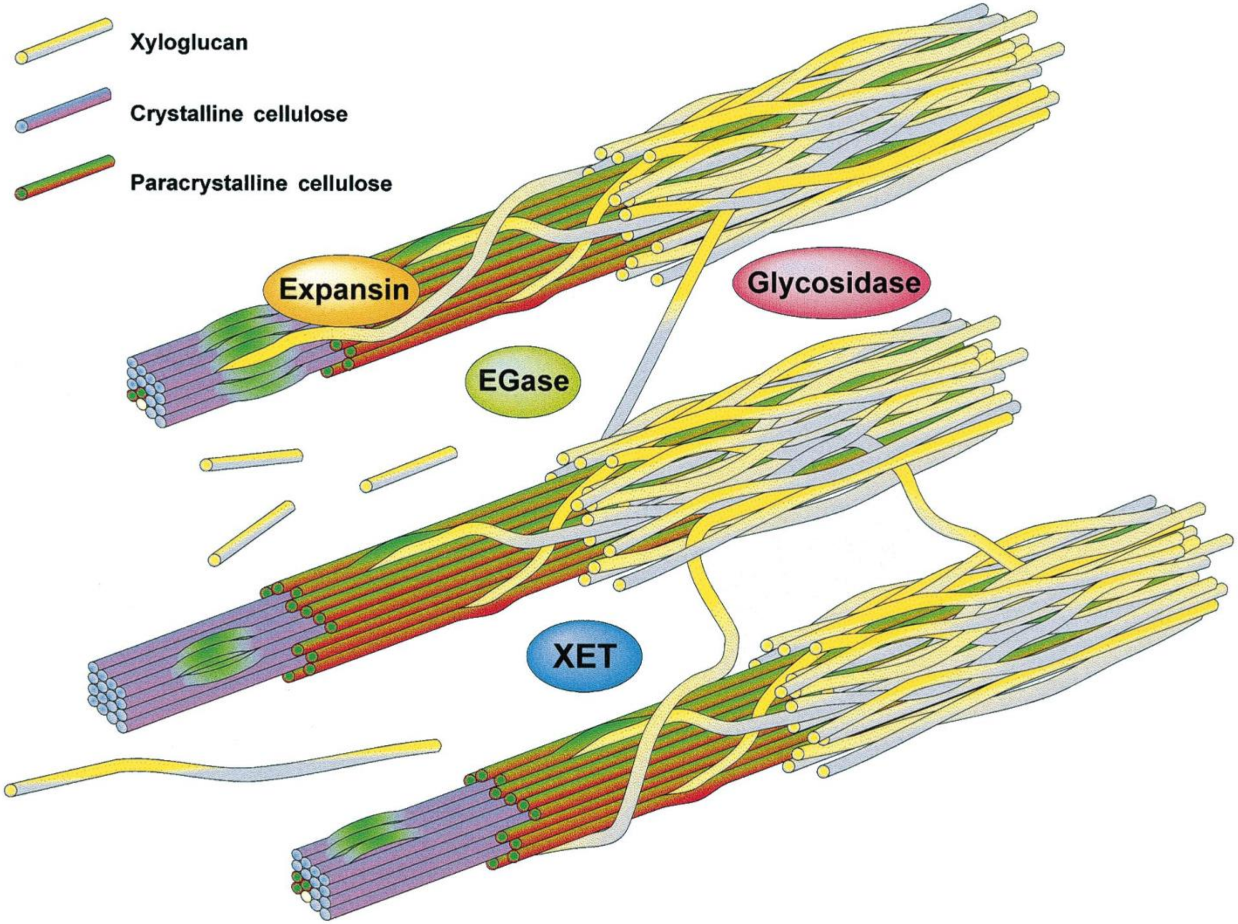
Ref: O'Sullivan (1997)

Cellulose Polymorphs



Ref: French and Johnson (2007)

Cellulose Hemicellulose



Cellulose xyloglucan network

Cellulose

Development of tomato

Hemicellulose

Second most abundant biopolymer on earth. Hemicellulose: Heteropolymer of pentoses (xylose and arabinose) and hexoses (glucose, galactose, mannose) and sugar acids

• Hemicelluloses are made of xylan (xylose, arabinose and glucuronic acid) backbone

• Xylan backbone is highly substituted by side chains consisting of xylose, arabinose and galucuranic acids

• Heteroxylans are highly cross linked with diferulic bridges (with other xylans), isodityrosine bridges (structural proteins)

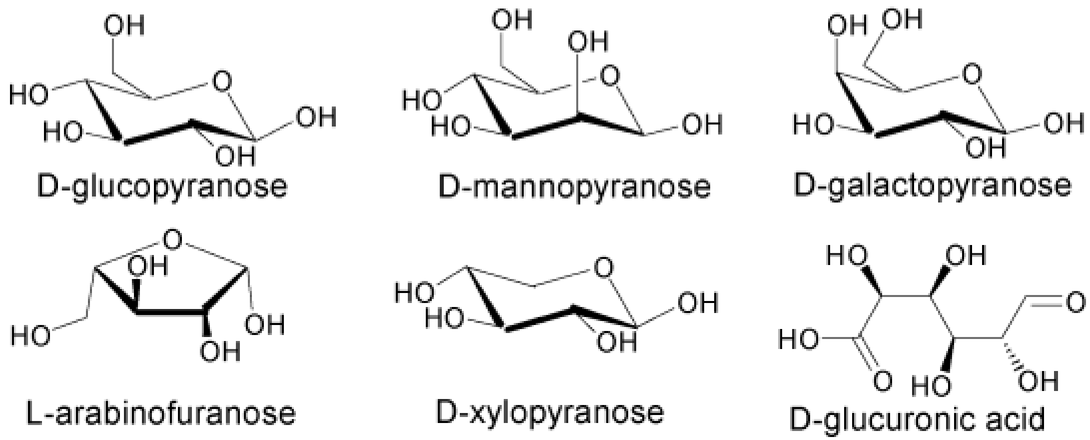
• Hemicellulose can be extracted by alkali or hot water and hydrolyzed by dil. H_2SO_4.

Ref: Saha (2003)

Hemicellulose

Important Monomers of Hemicellulose

1. Xylose
2. Glucose
3. Mannose
4. Galactose
5. Arabinose
6. Glucuronic acid
7. Acetic acid



Ref: Hansen and Plackett (2008)

Hemcellulose

Hemicellulose: Heteropolymer of pentoses (xylose and arabinose) and hexoses (glucose, galactose, mannose) and sugar acids

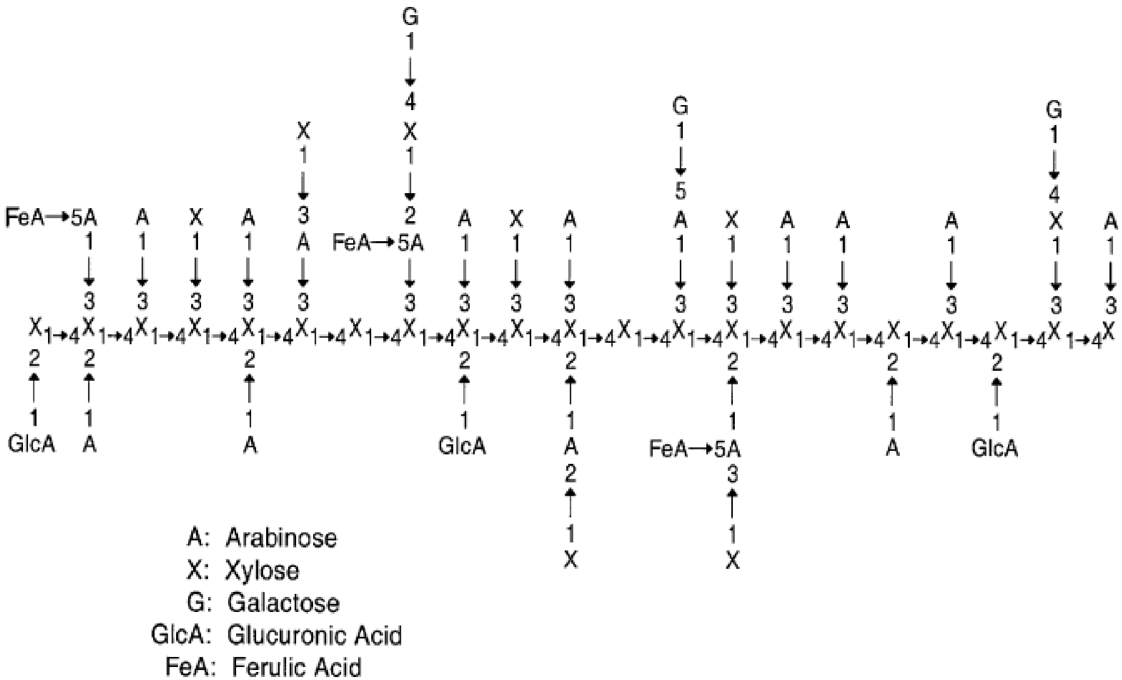
• Hemicelluloses can be classified into four types based on main type of sugar residue:

- Xylans : secondary cell walls of hardwood and herbaceous plants.
- Xyloglucans: primary cell walls of higher plants; bound to cellulose.
- Mannans : Secondary cell walls of conifers (softwood) and *Leguminosae*.
- Mixed linkage β-glucans: Common in Poales (members of grass family) and pteridophytes (vascular plants that do not have flowers and seeds and reproduce by spores).

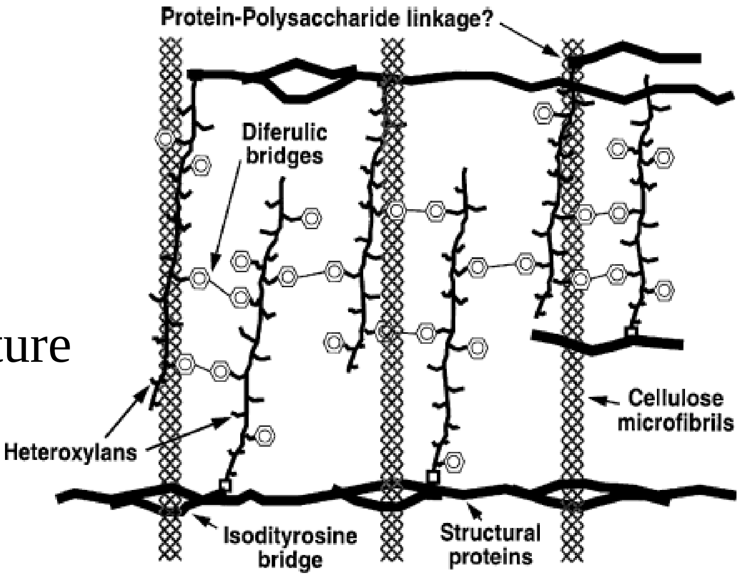
Ref: Schadel et al. (2009)

Hemicellulose

Heteroxylan structure

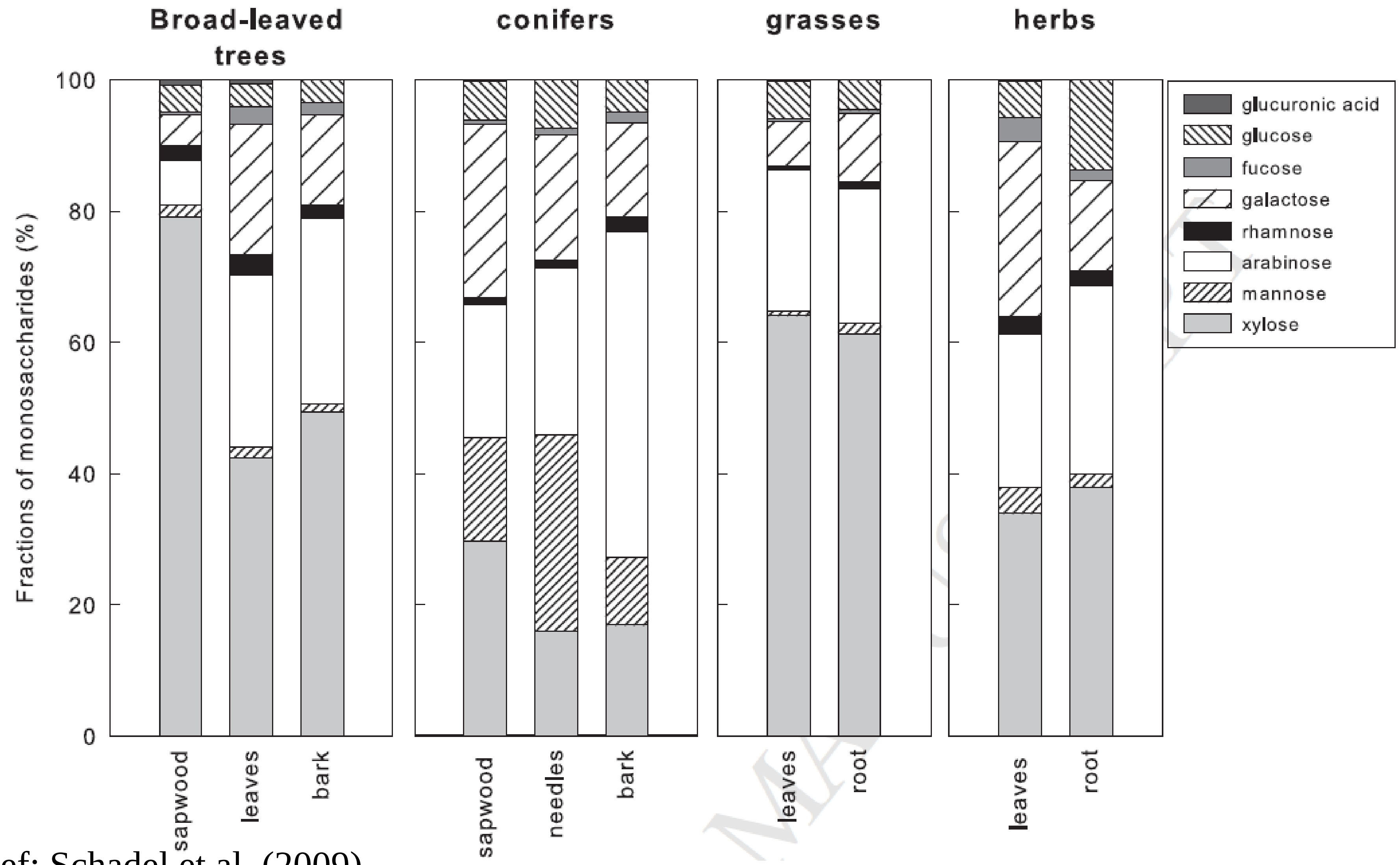


Corn fiber cell structure



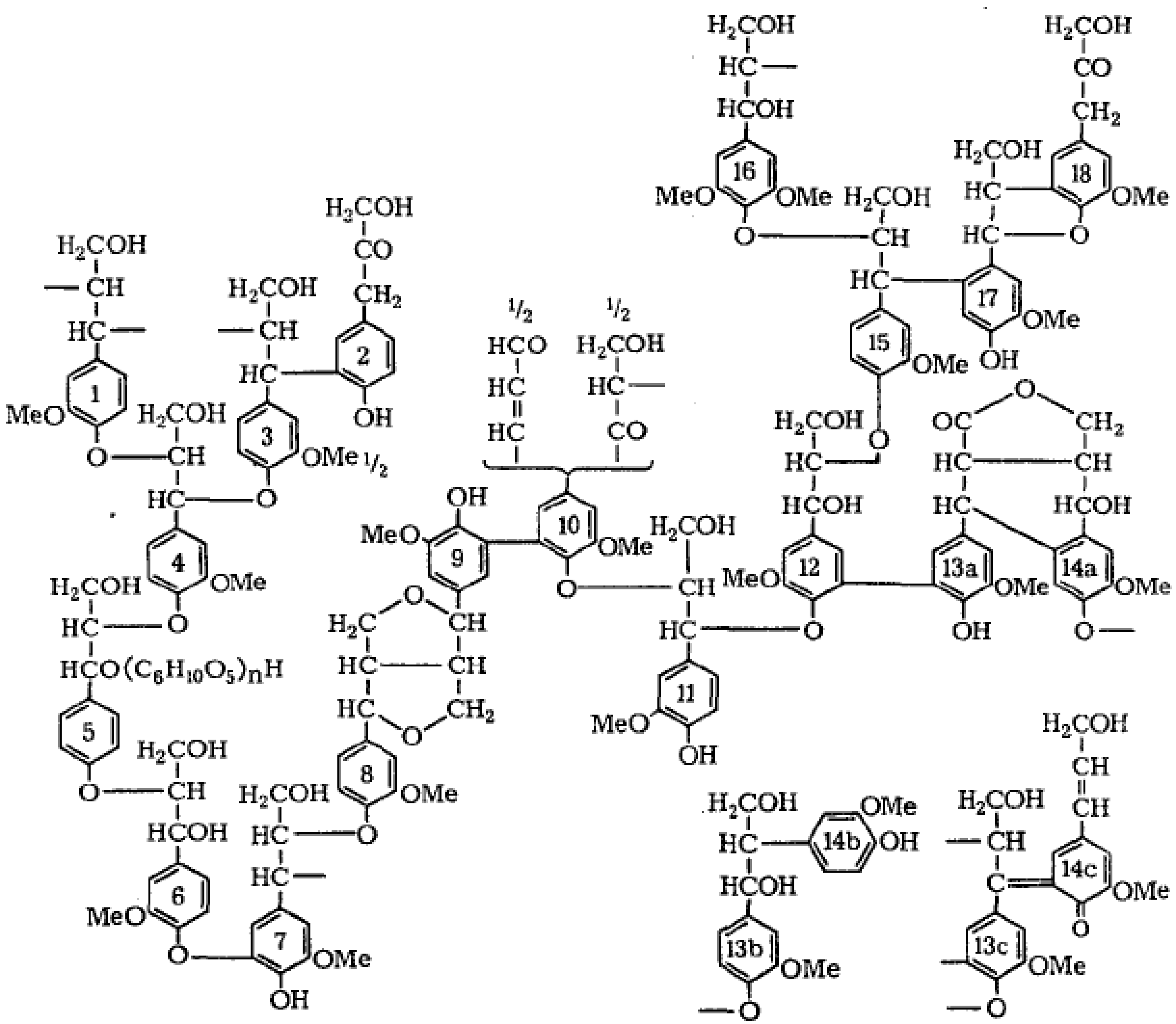
Ref: Saha (2003)

Hemicellulose Composition Variation



Ref: Schadel et al. (2009)

Lignin Composition and Synthesis

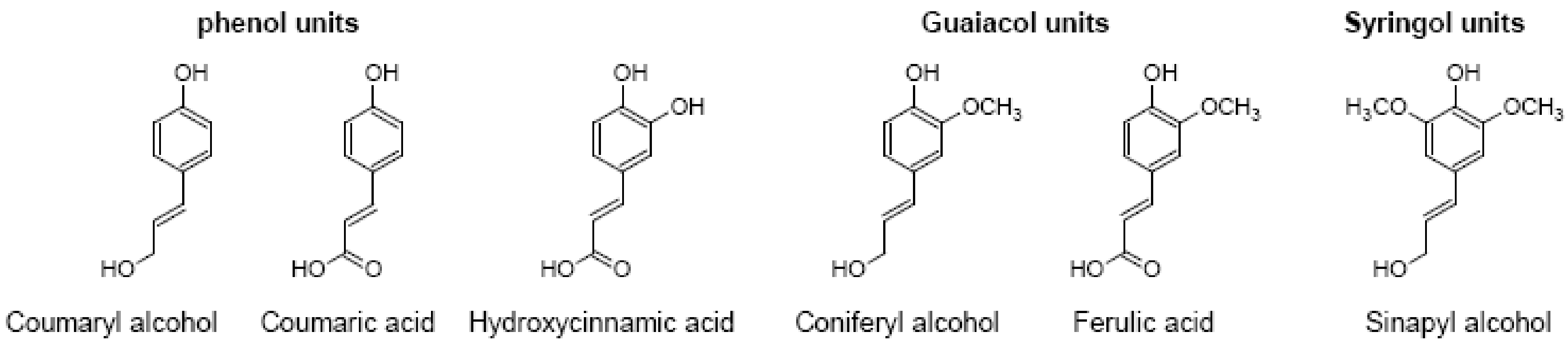


Ref: Eggeling (1983)

Lignin Composition and Synthesis

Lignin: A racemic, heteropolymer consisting of three hydroxycinnamyl alcohol monomers (C9) differing in their degree of methoxylation: p-coumaryl, coniferyl and sinapyl alcohols

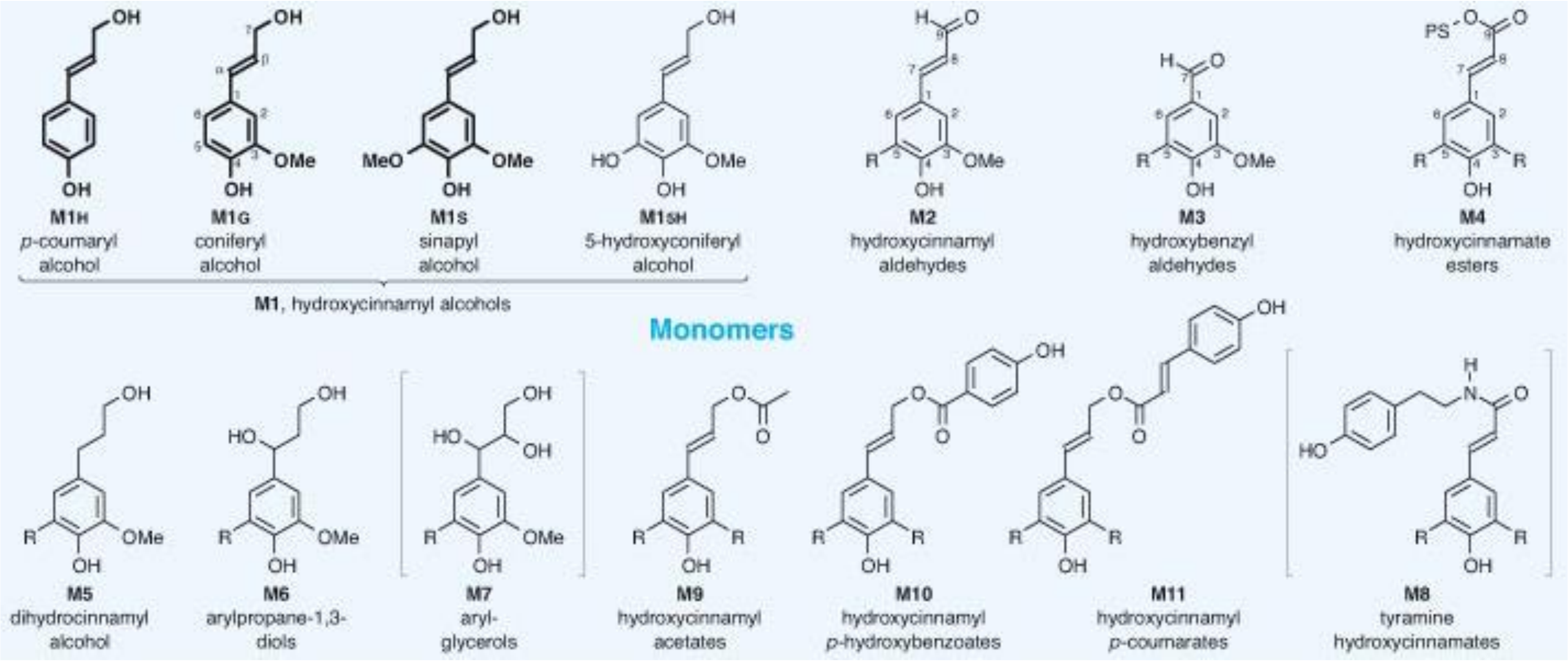
They produce p-hydroxyphenyl H, guaiacyl G, and syringyl S phenylpropanoid units when incorporated into lignin polymer.



Ref: Boerjan at al. (2003) and Holladay et al (2007)

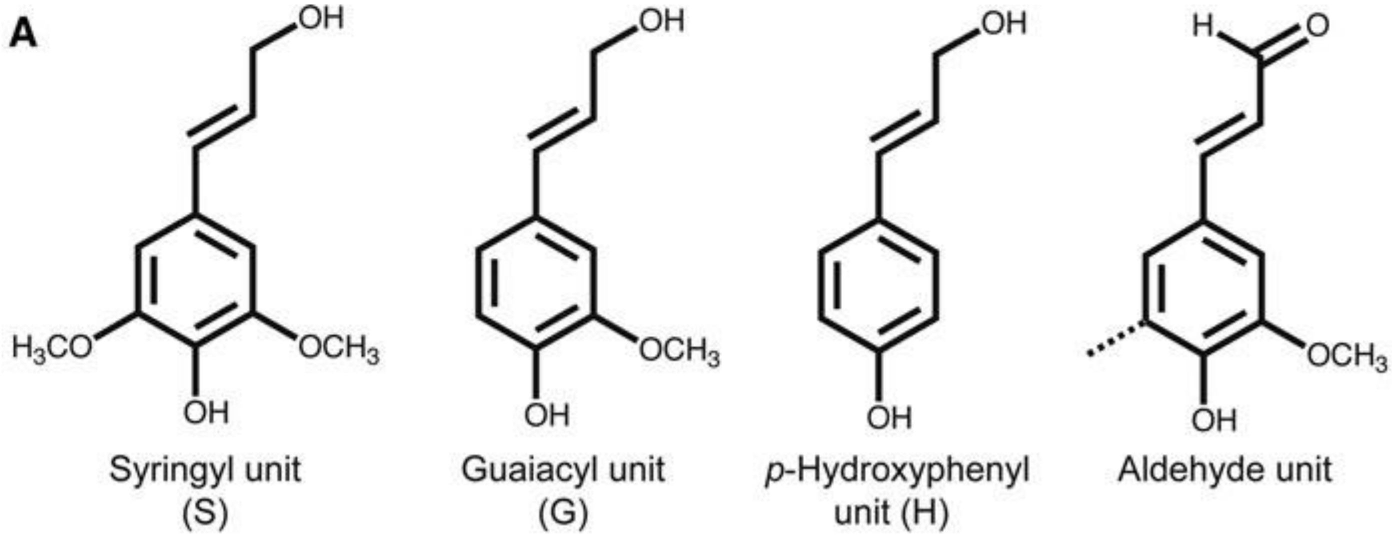
Lignin Composition and Synthesis

There are other monomer units in minor concentrations



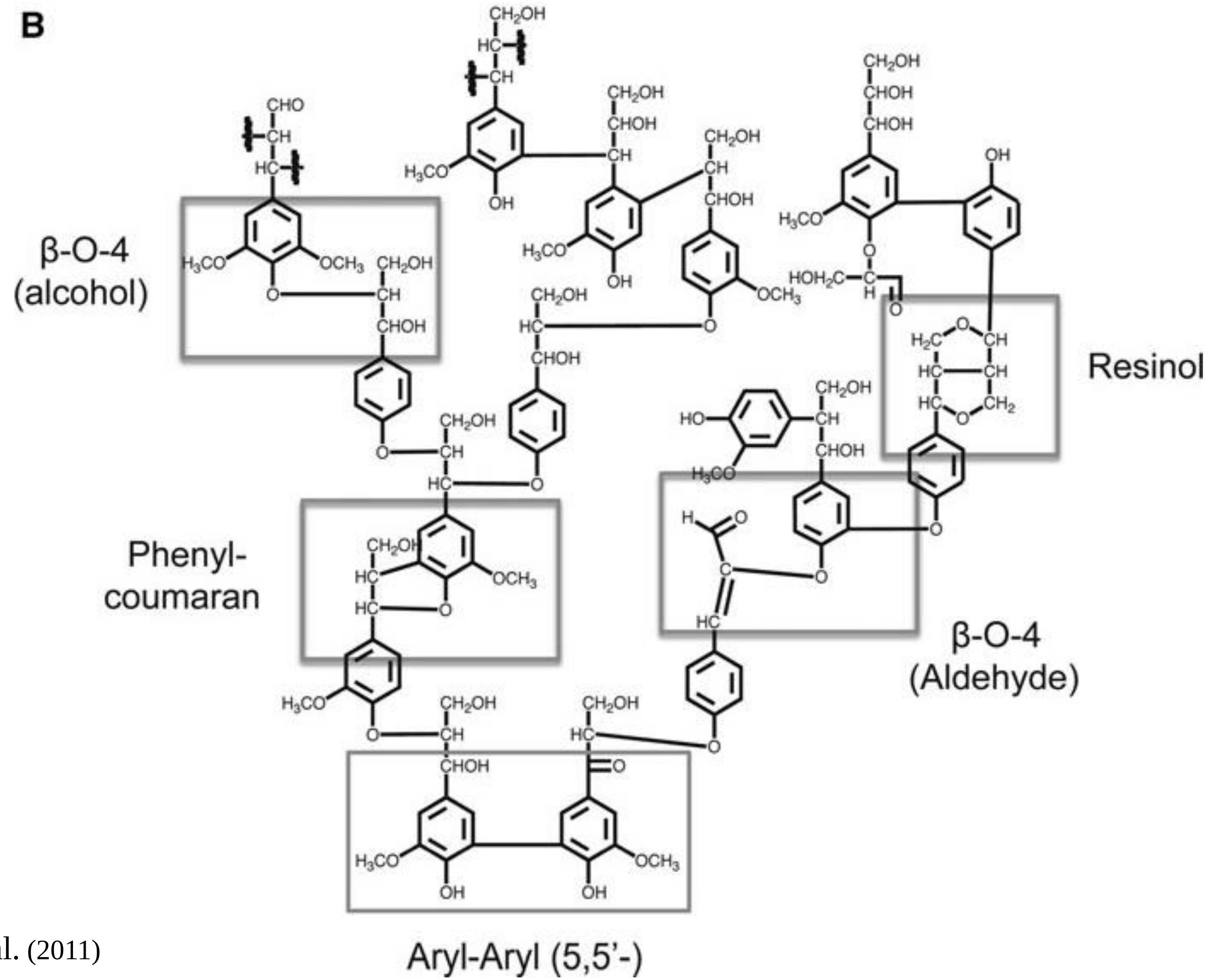
Ref: Boerjan at al. (2003)

Lignin Composition and Synthesis



Ref: Talbot et al. (2011)

Lignin Composition and Synthesis



Ref: Talbot et al. (2011)

Lignin Composition and Synthesis

Composition of Lignin is dependent on the source of feedstocks

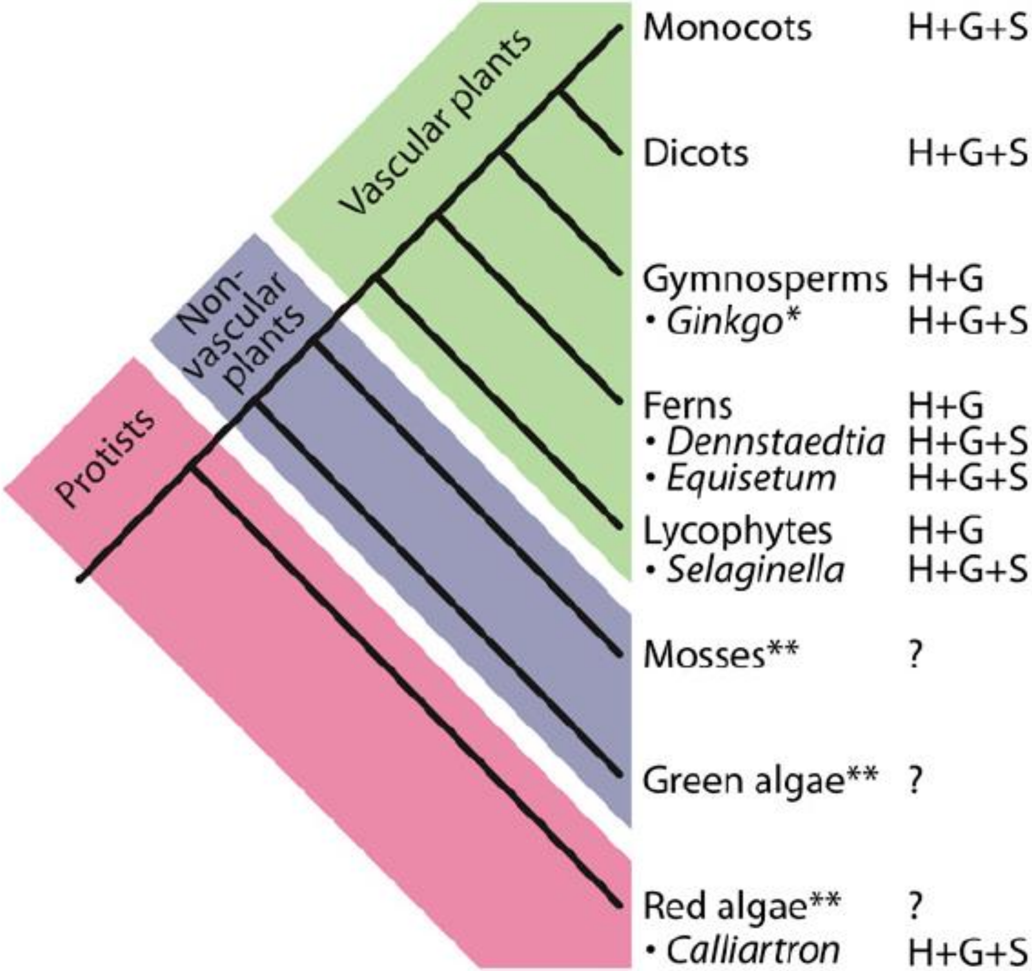
• Softwood lignins are rich in coniferyl alcohol (90%)

• Hardwood lignins are made of about equal amounts of coniferyl and synapyl alcohols

• Grasses consist of coniferyl and synaply alcohol and significant amounts of p-coumaryl alcohol (10-20%)

Ref: Boerjan at al. (2003)

Lignin Composition and Synthesis



Ref: Vanholmet at al. (2010)

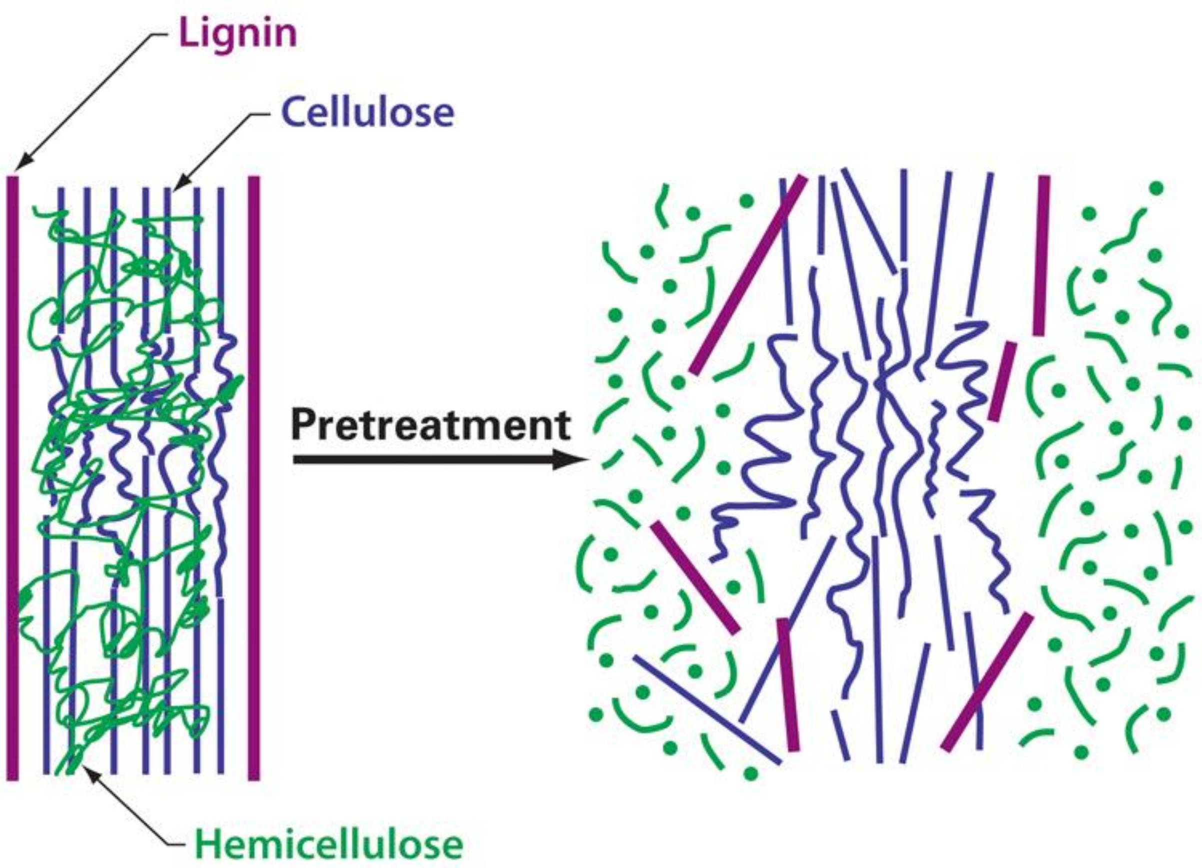
Lignin Composition and Synthesis

Composition of Lignin is dependent is also dependent on the method of extraction.

- Kraft Lignin
- Sulfite Lignin
- Organosolv Lignin
- Pyrolysis Lignin
- Steam explosion Lignin
- Dilute Acid Lignin
- Alkaline Oxidative Lignin
- Ammonia fiber Explosion (AFEX)
- Hot water

Ref: Boerjan at al. (2003)

Cellulosic Ethanol



Ref: http://genomicsgtl.energy.gov/biofuels/.

Thank you