

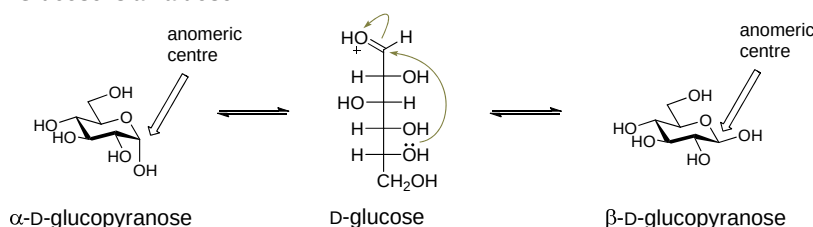
Workshop 5: Biochemistry of carbohydrates and fats – Spencer Williams

Dietary carbohydrates

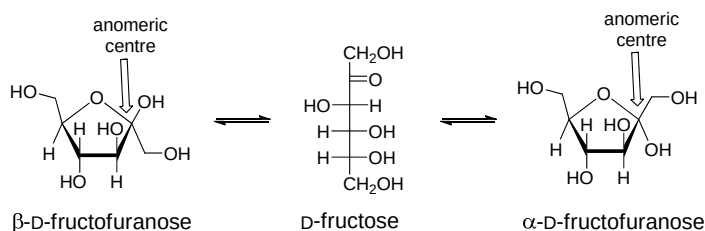
- *Comparison of glucose, fructose, sucrose and the artificial sweetener aspartame with reference to their structures and energy content*

Introduction to Carbohydrates. Carbohydrates are poly-hydroxylated compounds that often possess the general formula $C_m(H_2O)_n$. The individual carbohydrate fragments are termed *monosaccharides*. Monosaccharides usually exist as cyclic forms, but can interconvert freely to acyclic forms. For issues of naming (nomenclature), the acyclic forms are usually considered, despite the fact that they occur only fleetingly, and in trace amounts. Carbohydrates derived from an aldehyde are termed aldoses (e.g. glucose), and those from ketones are termed ketoses (e.g. fructose). Simple carbohydrates possessing an OH at the anomeric centre (a functional group called a hemiacetal) can interconvert between two stereoisomers termed anomers (a contraction of *anomeric isomers*), and which are denoted by α and β . The stereochemical descriptor D (or L) relates to the absolute configuration of the sugar.

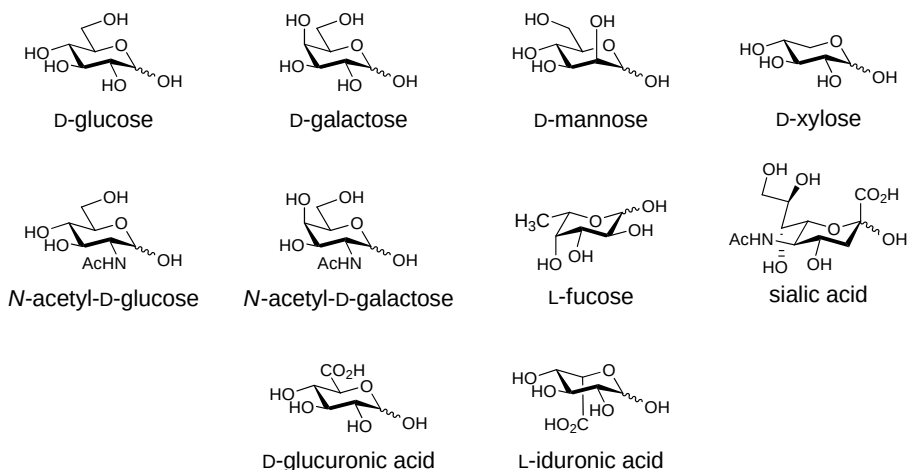
D-Glucose is an aldose:



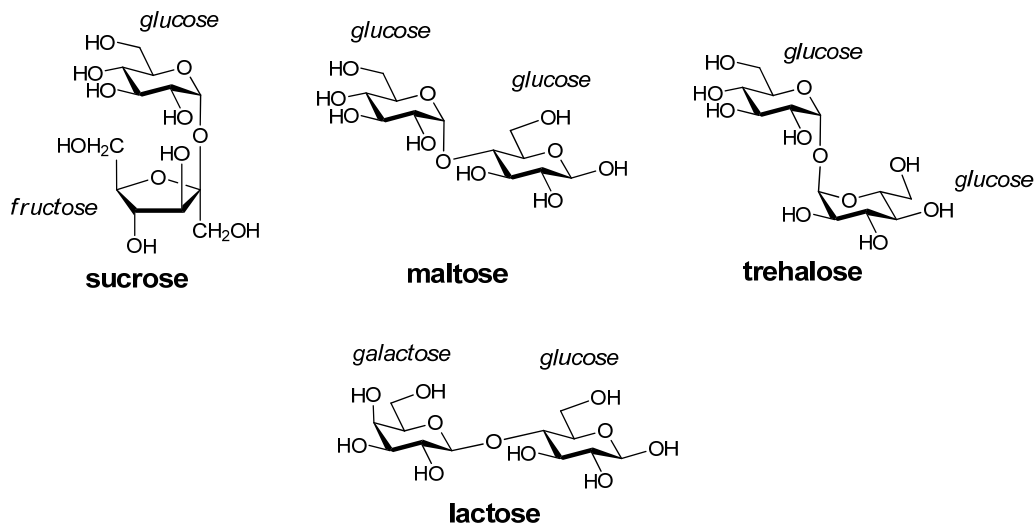
D-Fructose is a ketose:



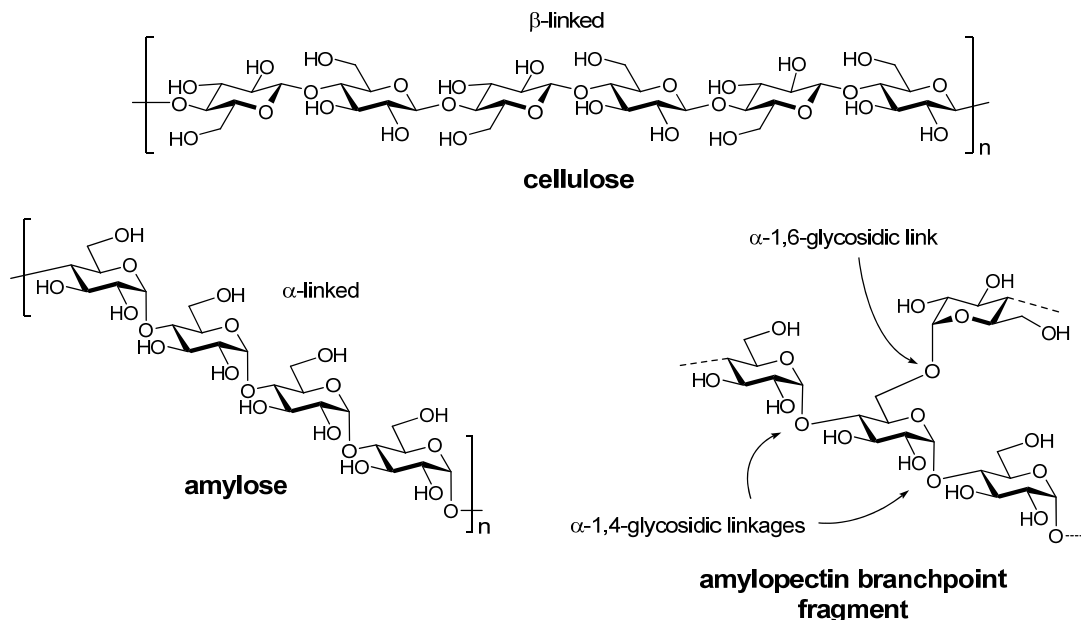
While in theory, owing to the number of stereocentres, there are a large number of possible monosaccharides, in practice in nature there are much fewer carbohydrates relevant to humans. In fact, there are just 10 monosaccharides present in mammalian biology. These are assembled from 9 building blocks, with L-iduronic acid formed after assembly of the glycoconjugate.



Oligosaccharides. Monosaccharides can be joined to one other to make more complex oligo- and polysaccharides. Important disaccharides include sucrose (table sugar), maltose (malt, eg cereals, beer, pasta, potatoes), lactose (milk), and trehalose (major sugar for flight in insects!).



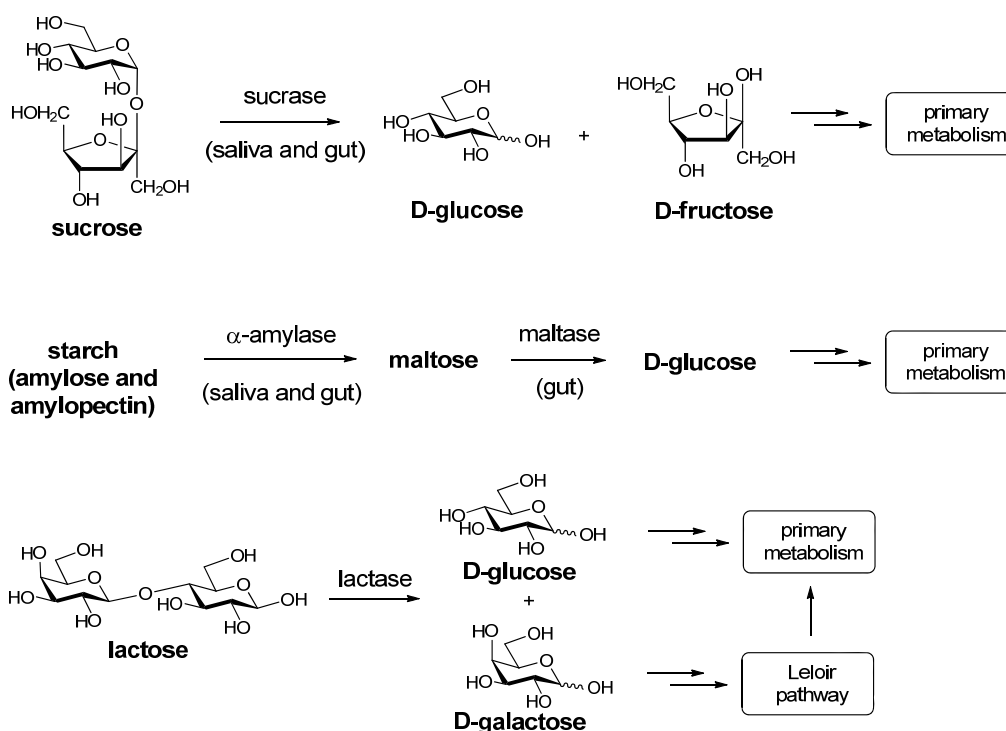
Polysaccharides. Important polysaccharides include starch, amylopectin and cellulose. All three of these polymers are constructed from the same monosaccharide building block, glucose, but possess dramatically different physical properties and are altogether different sources of nutrition. Starch is comprised of amylose and amylopectin and is produced by most plants and is common in seeds. Cellulose is the major structural material of trees.



Metabolism of carbohydrates. Important dietary carbohydrates include glucose, fructose, sucrose, lactose, starch, amylose and (to a small extent) trehalose. In general, the oligo and polysaccharides are broken down in our mouth and gastrointestinal tract by oligo/polysaccharide degrading enzymes including salivary and pancreatic α -amylase, sucrase, trehalase, maltase, and lactase. Some of these enzymes act in concert, with α -amylase degrading the polysaccharide of starch to smaller oligosaccharides, which are in turn degraded by maltase to monosaccharides. The resultant

monosaccharides, glucose, fructose and galactose, are imported through the brush border of the intestine where they enter basic metabolic pathways including glycolysis and the citric acid cycle; the pentose phosphate pathway; and for energy storage in glycogen. Galactose is unusual in that it must proceed through a complex pathway termed the Leloir pathway, which interconverts galactose and glucose.

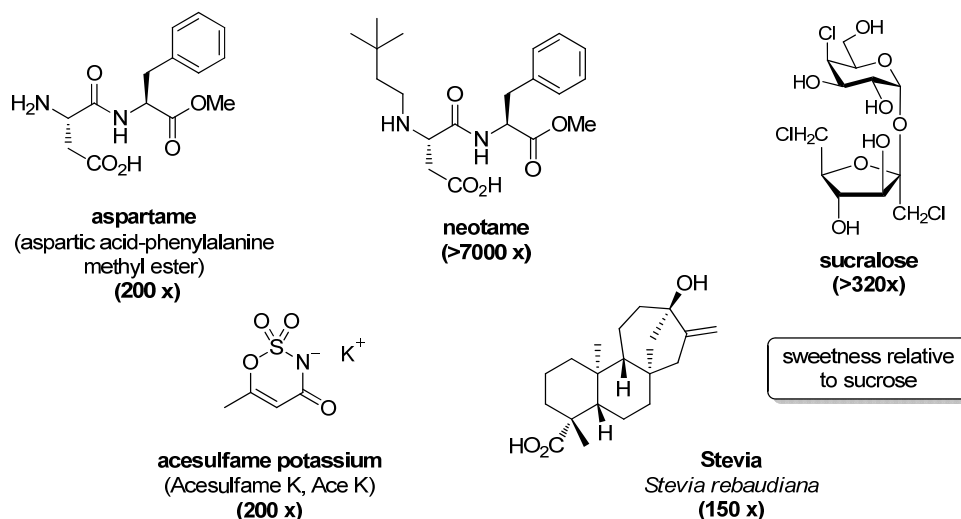
Glucose, galactose and fructose can be interconverted in the body. Glucose is converted to glucose-6-phosphate, which can be isomerized to fructose-6-phosphate in the first three steps of the glycolysis pathway. Galactose can be converted to glucose by the Leloir pathway, which involves conversion of galactose to galactose-1-phosphate, and then UDP-galactose, and finally isomerisation to UDP-glucose.



Lactose is an unusual dietary sugar in that it is the only dietary sugar with a β -linkage. Lactose comprises 2-8% of milk, dependent on the species of mammal. Lactose is the preponderant milk sugar in all lactating mammals and is found at 55-70 g/L in human milk and 40-50 g/L in cow's milk. Lactose is synthesized in response to the hormone prolactin, which results in production of α -lactalbumin. This binds to a glycosyltransferase and results in the production of lactose during lactation.

Sweeteners: Carbohydrate and 'artificial'. Humans crave sweetness. The average adult will generally consider sweeter foods more pleasant up to a threshold equivalent to 10 teaspoons of sucrose in a cup of tea, beyond which it is considered sickly sweet. Babies have no threshold for 'sickly' sweetness. It is recommended that the average person limit their daily intake of added sugar. One approach to do so is through the use of artificial sweeteners. Commonly used artificial sweeteners include aspartame, sucralose, neotame, acesulfame, and stevia (which is a natural product and is not artificial). Most of these compounds have remarkable histories describing their discovery.

- *Aspartame* was discovered in 1965 by James M. Schlatter. He accidentally discovered its sweet taste when he licked his finger, which had become contaminated with aspartame, to lift up a piece of paper.
- The sweetness of *acesulfame* was discovered in 1967 by Karl Clauss and Harald Jensen. After accidentally dipping his fingers into the chemicals with which he was working, Clauss licked them to pick up a piece of paper.
- Leslie Hough and Shashikant Phadnis first synthesized *sucralose* while researching ways to use sucrose and its synthetic derivatives for industrial use. Hough asked Phadnis to "test" a chlorinated sugar compound. Phadnis thought Hough asked him to "taste" it, so he did.



The ester-containing (O–C=O) sweeteners aspartame and neotame are unstable to high pH and elevated temperature, which lead to ester cleavage. This limits their use in baked goods and long shelf-life products. As well, aspartame can be broken down to phenylalanine, and subjects with the rare genetic disorder phenylketonuria that are unable to metabolize phenylalanine must be legally advised to avoid aspartame, although such trace amounts are used in sweetening applications that the risk is much lower than other common dietary components. Sucralose is much more stable to heat and can be used in most baking applications. Whilst aspartame provides energy through gluconeogenesis, the energy content of artificial sweeteners is irrelevant as their powerful sweetening properties result in their use in only trace amounts in the diet.

Digestible carbohydrates and the glycaemic index.

- *explanation of the ability of all humans to hydrolyse starch but not cellulose, and of differential ability in humans to hydrolyse lactose; glycaemic index (GI) of foods as a ranking of carbohydrates based on the hydrolysis of starches (varying proportions of amylose and amylopectin) to produce glucose in the body*

Despite the similarity of structure and the identity of the component monosaccharide glucose, distinct enzymes are required for the degradation of the two polysaccharides starch and cellulose. Mammals produce amylase, which is able to degrade starch and it is specific for the α -glucoside linkage. Mammals do not produce enzymes that degrade the β -glucoside linkage, and thus cellulose and other related polysaccharides are inert.

Lactose is special in that it contains a β -galactoside linkage. Lactose is digested through the action of *lactase*, which is essential in infancy when milk acts as the main source of nutrition. Pre-term babies have very low levels of lactase and must use other sugars, typically glucose, as a source of nutrition until sufficient lactase is produced to allow milk-feeding. This is usually supplied by tube, as pre-term babies have a poor swallowing response. Most mammals, including humans, undergo a natural decline in lactase throughout development. In Asian and black groups, levels decline in early childhood, whereas in most Caucasians decline occurs in later childhood or adolescence. In certain Northern Europeans, lactase activity persists into adulthood.

Lactose intolerance typically arises from low levels of lactase (hypolactasia). This leads to incomplete digestion of lactose and a rise in osmotic load in the colon after milk consumption, causing diarrhoea. In addition, lactose-metabolizing organisms may colonize the GI tract, resulting to gas production and excessive flatulence. The symptoms may be managed by reducing consumption of lactose-containing foods. Certain mammals such as sheep and cattle (ruminants) have special digestive tracts that can be colonized by bacteria with the capacity to degrade cellulose to glucose, which can be used by the host. An unfortunate side-effect of the anaerobic digestion process is the production of large amounts of methane, which contributes in a significant way to global warming. Contrary to many media reports, methane only comes out the front end of the cow!

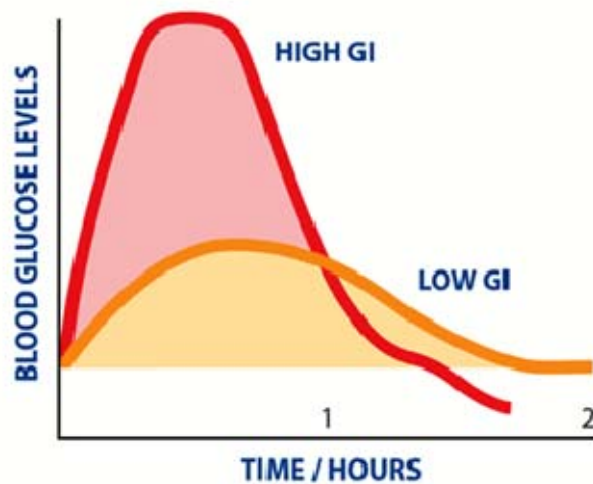
The **glycaemic index (GI)** is a way of measuring the effects of food on blood sugar levels. After a carbohydrate-containing food is eaten there is a rise, and subsequent decrease, in blood sugar (glucose). This phenomenon is known as the glycaemic response.

The glycemic index of a food is measured as the incremental area under the two-hour blood glucose response curve (area-under-the-curve, AUC) following a 12-hour fast and ingestion of a food with a certain quantity of available carbohydrate (usually 50 g). The AUC of the test food is divided by the AUC of the standard (either glucose or white bread, giving two different definitions) and multiplied by 100. The average GI value is calculated from data collected in 10 human subjects.

A practical limitation of the glycemic index is that it does not measure insulin production due to rises in blood sugar, which is the underlying parameter that glycaemic index is seeking to quantify. Two foods may have the same glycemic index, but produce different amounts of insulin, and therefore have a different effect upon health.

The glycemic effect of foods depends on a number of factors, such as the type of starch (amylose versus amylopectin), physical entrapment of the starch molecules within the food, fat and protein content of the food and organic acids or their salts in the meal — adding vinegar, for example, will lower the GI. The presence of fat or soluble dietary fibre can slow the gastric emptying rate, thus lowering the GI. In general, coarse, grainy breads with higher amounts of fibre have a lower GI value than white breads.

However, most breads made with 100% whole wheat or wholemeal flour have a GI not very different from endosperm only (white) bread. Many brown breads are treated with enzymes to soften the crust, which makes the starch more digestible, leading to a higher GI.

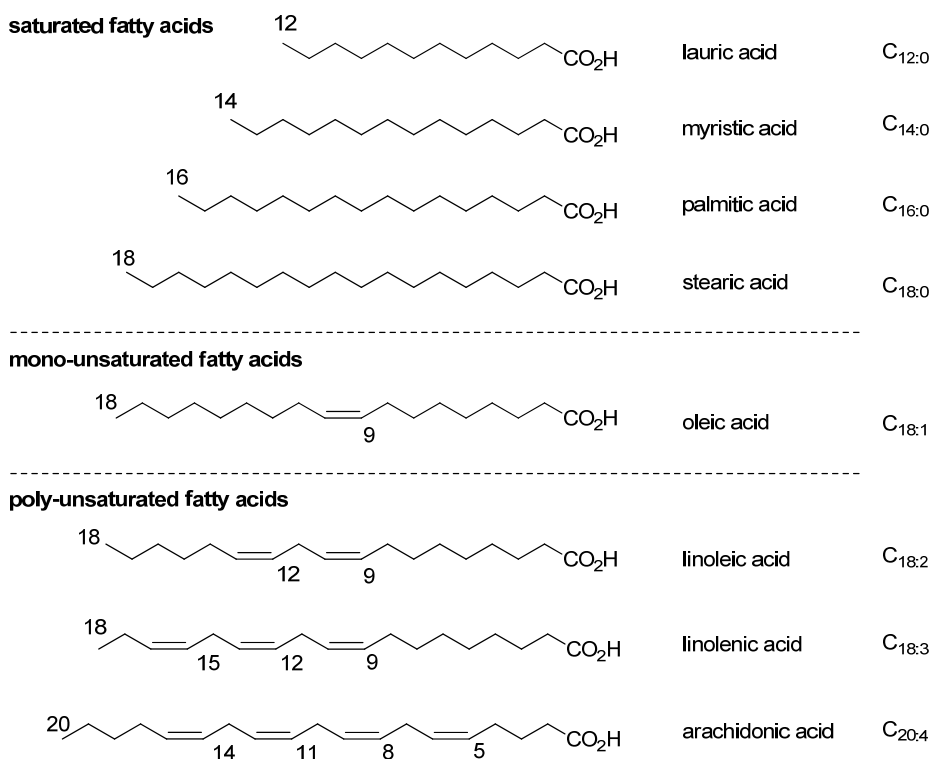


Certain starch molecules are resistant to metabolism. These are termed recalcitrant and retrograded starches. The structural basis of their resistance to degradation is not well understood.

Fatty acids: essential and non-essential

- distinction between essential and nonessential fatty acids; and structural differences between omega-3 fatty acids and omega-6 fatty acids

A selection of common fatty acids is shown below: many are present in a common diet or can be obtained from plant or animal sources:



The common fatty acids have some important features:

- They have straight chains with no branching.
- They have even numbers of carbon atoms.
- They may be saturated, with no double bonds, or unsaturated, in which case they are usually *cis* (Z) alkenes. If there is more than one C=C double bond they are usually not conjugated – there is normally one saturated carbon atom between them.

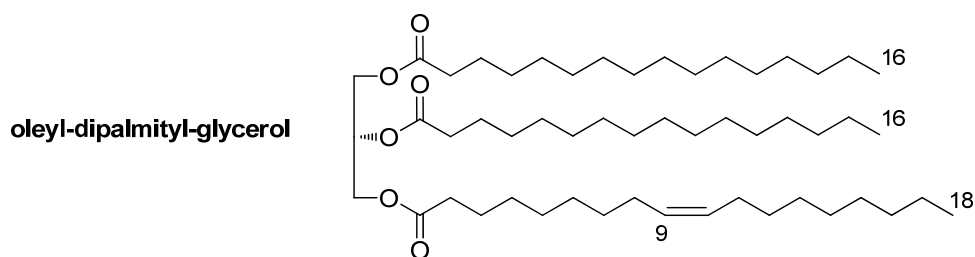
Palmitic acid (C₁₆ saturated) is the most common fatty acid in living things. Oleic acid (C₁₈ mono-unsaturated) is the major fatty acid in olive oil. Arachidonic acid (C₂₀ tetra-unsaturated) is a rare fatty acid, but is an important precursor of signalling molecules including prostaglandins, resolvins, thromboxanes and leukotrienes.

The prevalence of fatty acids with even numbers of carbon atoms is suggestive of a biogenesis from a two carbon precursor. In 1907, it was ingeniously suggested by John Collie¹ that they are made by the head-to-tail linking of acetic acid units.

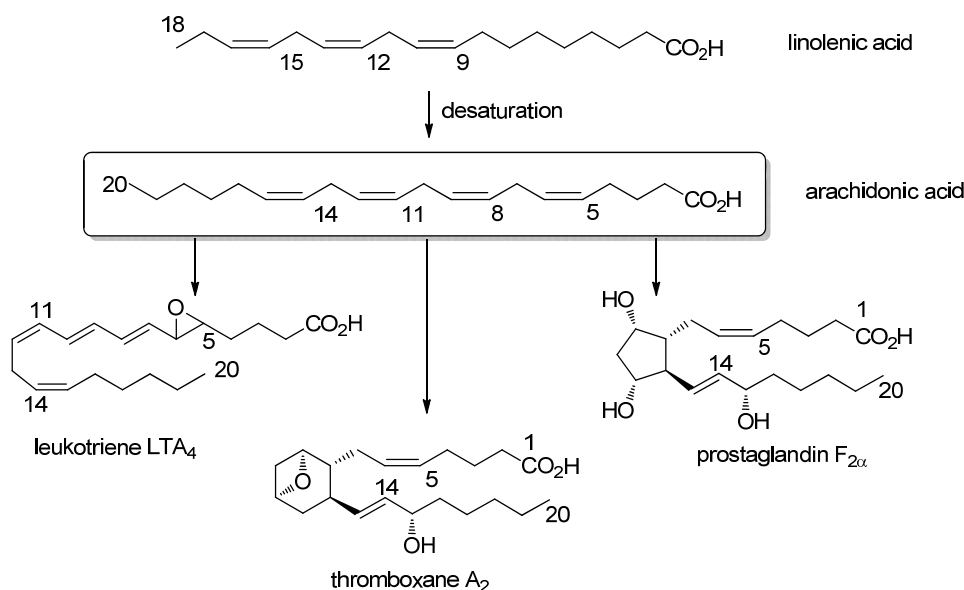
Fatty acids are mainly found as esters in combination with glycerol as *triglycerides*. These are called fats or oils, depending upon whether they are solids or liquids at room temperature. *Simple triglycerides* are comprised of the same three fatty acids, whereas a *mixed triglyceride* contains two or more fatty acids. Fats and oils represent long term stores of energy for most organisms, and are subjected to oxidative metabolism when required. Animal fats contain a higher proportion of glycerides and tend to

¹ John Norman Collie (1859-1942), Professor University College London. Aside from insightful research in chemistry, Collie was a keen mountaineer and claimed 21 first ascents!

be solids (at room temperature), whilst those from plants and fish contain predominantly unsaturated fatty acid esters and tend to be liquids. Partial hydrogenation of unsaturated oils is sometimes used to increase the proportion of saturated fatty acids and consequently the melting temperature. Partial hydrogenation can lead to double bond isomerisation forming *trans* fats, the consumption of which correlates with coronary heart disease, and in most places in the world *trans* fats have either been banned or minimized.



Biosynthesis of unsaturated fatty acids. Mammals can insert a double bond into fatty acyl chains providing it is no further from the carbonyl than C9. Thus stearic acid is converted to oleic acid. Mammals cannot introduce additional double bonds into oleic acid, and thus we obtain linoleic and linolenic acids from our diet. They are essential fatty acids, and extensive studies have shown that exclusion of these fatty acids from the diet leads to death. Linoleic and linolenic acids are produced by plants and bacteria, and then consumed in our diet. Humans are able to convert linolenic acid to the essential prostaglandin precursor arachidonic acid:

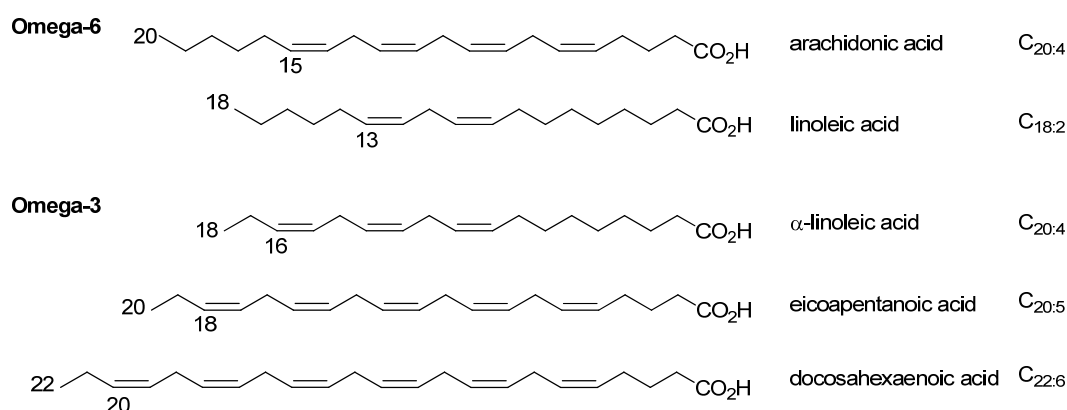


Arachidonic acid and its products are referred to as *eicosanoids* because they are derived from C_{20} unsaturated fatty acids.

Omega-3/-6 fatty acids. The 'Omega-n' notation refers to the location of a fatty acid double bond counted from the terminus of the fatty acid. Omega-6 fatty acids include *linoleic acid* and *arachidonic acid*. Omega-3 fatty acids include α -*linolenic acid* (ALA) (found in plant oils), *eicosapentaenoic acid* (EPA), and *docosahexaenoic acid* (DHA) (both commonly found in marine oils).

Some researchers have suggested that elevated levels of Omega-3 fatty acids are beneficial to health and that high levels of Omega-6 fatty acids are detrimental. Most of these studies have not withstood the assault of high-powered meta-analyses, and all available evidence suggests that supplementation of the diet with Omega-3 fatty acids does not lower the risk of all-cause mortality. Omega-3 fatty acids are commonly touted as valuable dietary supplements by the 'alternative' healthcare

industry, which is characterized by its avoidance of scrutiny and consistent failure to support its claims through placebo-controlled clinical trials and peer-reviewed publication.



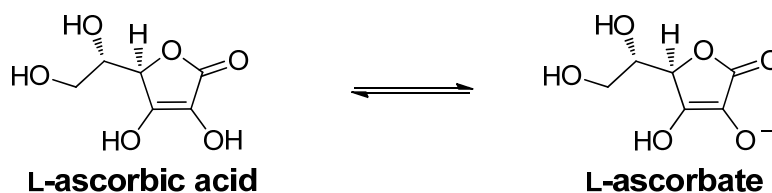
Vitamins

- *vitamins: inability of humans to synthesise most vitamins (except Vitamin D) making them essential dietary requirements; comparison of structural features of Vitamin C (illustrative of a water-soluble vitamin) and Vitamin D (illustrative of a fat-soluble vitamin) that determine their solubility in water or oil.*

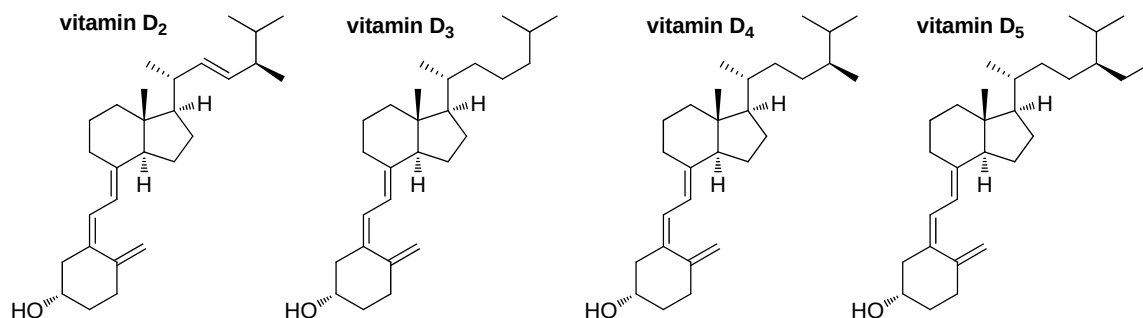
A special group of essential nutrients have historically been given the privileged status of the moniker vitamin. Like essential fatty acids and essential amino acids, these vitamins are essential for humans.

Vitamin C is an essential nutrient in a small number of mammals including humans, bats and guinea pigs. It is a co-factor, required for the catalytic activity of many enzymes, including those responsible for the synthesis of collagen. Scurvy occurs when deficient in vitamin C, resulting in incomplete formation of collagen and related connective tissues. The ability of most animals to synthesize vitamin C allows the use of fresh meat as a cure for scurvy, which was extensively used by explorers such as Scott in the Antarctic. The enantiomer of vitamin C, D-ascorbate, has identical antioxidant effects, yet is not able to substitute for vitamin C, underscoring that the essential properties of vitamin C are related to its effect in stereospecific enzymatic reactions. As a polar molecule, which is predominantly deprotonated and ionic at physiological pH, vitamin C is predominantly found in aqueous compartments in the body.

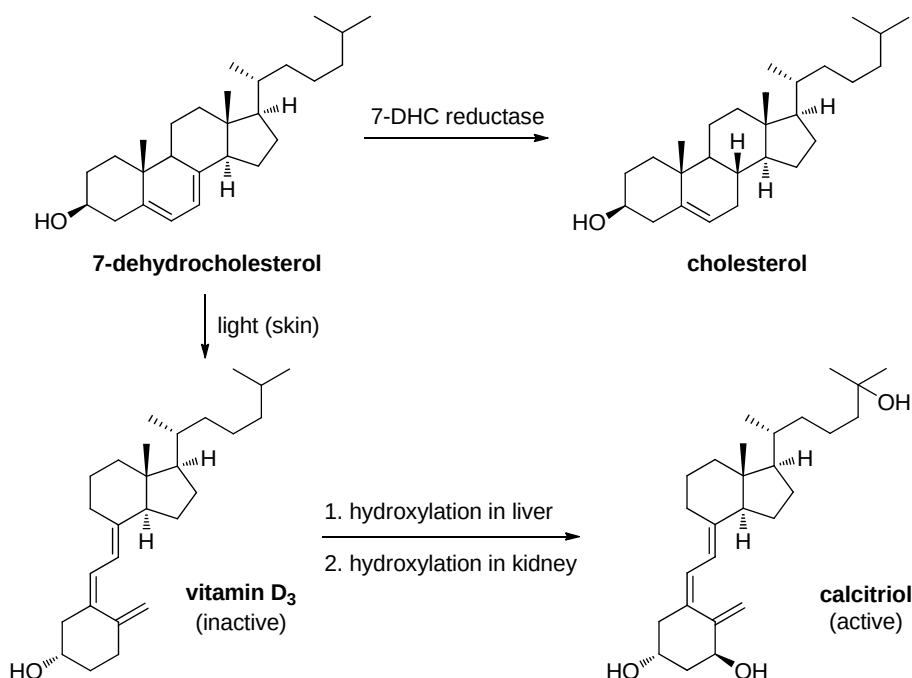
Vitamin C



'Vitamin D' describes a group of steroids that assist in absorption of calcium, iron, zinc and magnesium. The D vitamins are synthesized from endogenous steroids including ergosterol and 7-dehydrocholesterol. Very few foods contain vitamin D. As a lipidic molecule, vitamin D is partitioned into lipophilic compartments in the body including cellular membranes.



Synthesis of vitamin D₃ occurs in the skin and in the liver. In the skin sunlight acts to break one of the rings of the parent sterol 7-dehydrocholesterol (which is an intermediate in the synthesis of cholesterol) to form a secosteroid (i.e. a steroid with one broken ring). The resulting secosteroid is vitamin D₃, which is inactive as a vitamin, and must undergo hydroxylation in the liver and then the kidney to give the active species, calcitriol, which binds at the vitamin D receptor. One school of thought offers that since vitamin D is synthesized in the body, it should be called a hormone, and not a vitamin.

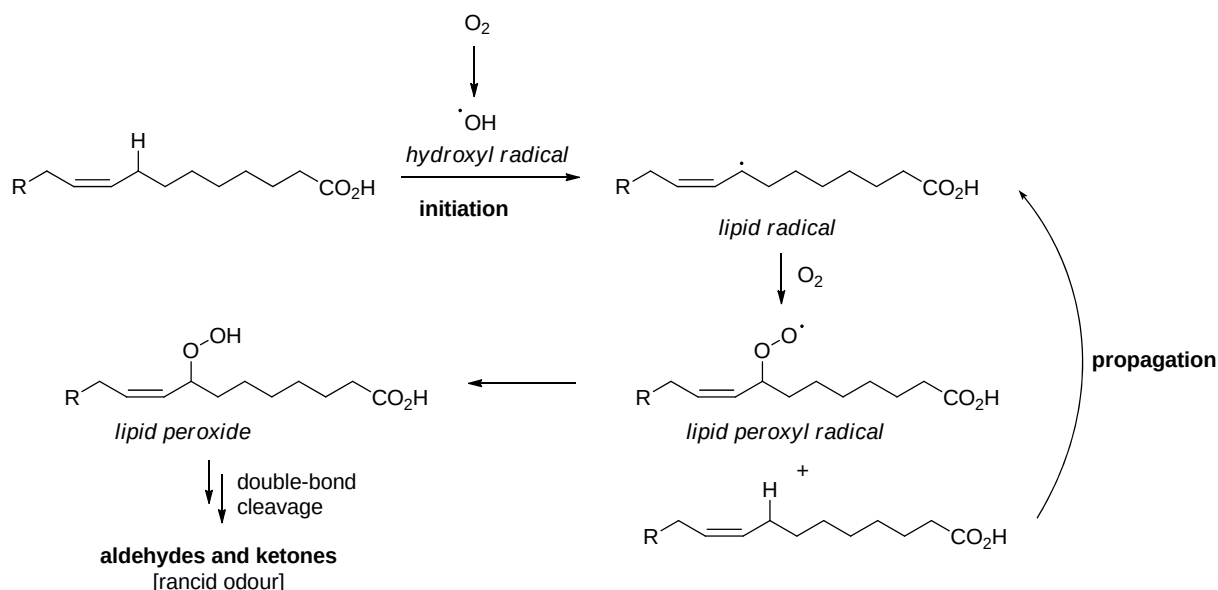


Introduction of hydroxyl groups into vitamin D₃ to form calcitriol results in an increase the water solubility of the latter.

Oxidative rancidity

- oxidative rancidity with reference to chemical reactions and processes, and the role of antioxidants in lowering the rate of oxidative rancidity

Rancidity refers to the process of reaction of fats and oils with oxygen and water to produce short-chain aldehydes and ketones that are objectionable in taste and odour. Oxidative rancidity refers specifically to those processes that involve oxygen. Oxygen through a range of processes can form the hydroxyl radical ($\cdot\text{OH}$), which has a propensity to react at the carbon next to a double bond to form a lipid radical. The lipid radical reacts with another molecule of oxygen to form a lipid peroxy radical. These are then able to react with another lipid to form a new lipid radical, thereby propagating the reaction. The resulting lipid peroxides are unstable, and can decompose with cleavage of the double bond to make various odiferous and objectionable aldehydes and ketones. The requirement for oxygen explains why frozen meats still go off, and why vacuum packaging, where oxygen is excluded from a foodstuff, is a useful preservative method.



Antioxidants are species that can break the chemical reactions involved in oxidative rancidification. Generally, these compounds act not by preventing the initiation step, the formation of the lipid radical, but rather the propagation step, whereby a lipid peroxy radical reacts with a new lipid to continue the oxidation process. Antioxidants are able to intercept the lipid peroxy radical by donating a hydrogen atom. Thus small amounts of lipid peroxides are still formed, but the rapid consumption of the starting lipid is prevented. Common antioxidants include the vitamins C and E (a fat soluble vitamin!), as well as various food additives such as butylated hydroxytoluene (BHT).

Antioxidants:

