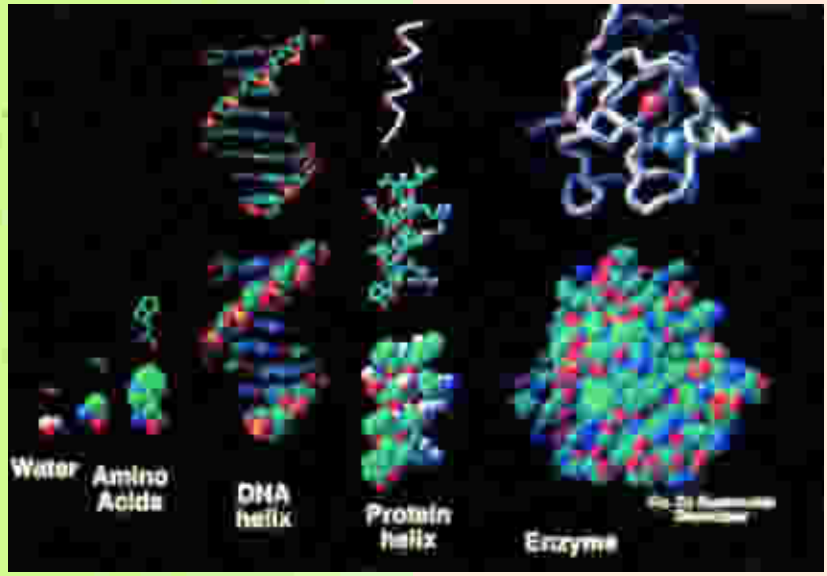
PART

2

Biomolecules



Water, Amino Acids, DNA and Protein Structure

Starting at the far left, we see a water molecule, two common amino acids, alanine and tryptophan, a segment of a DNA double helix, a segment of a protein single helix, and the folded polypeptide chain of the enzyme copper, zinc superoxide dismutase or SOD. With respect to the relative sizes of some of these molecules and structures, the water molecule is roughly half a nanometer (nm) across, the DNA and protein helices are about 2 nm and 1 nm in diameter, respectively, and the SOD, a small, globular protein of about 150 amino acids, is about 6 nm in width. SOD catalyzes the breakdown of harmful, negatively–charged oxygen radicals, thereby protecting people against neurodegenerative diseases such as Lou Gehrig's disease.

"Life is a relationship among molecules and not a property of any one molecule."

—Linus Carl Pauling : The Chemical Bond, 1960

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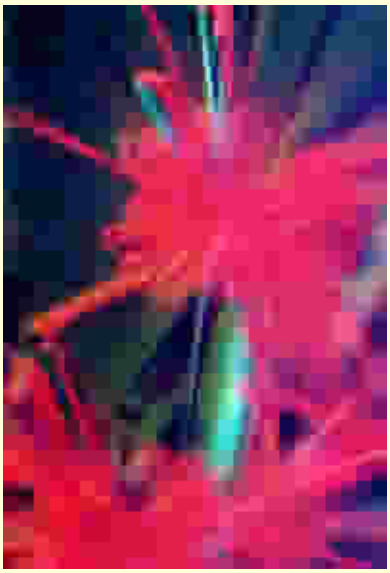
CHAPTER

5

Carbohydrates-I

General Structure of Monosaccharides



Crystals of **glucose**, a key molecule in carbohydrate metabolism, viewed under polarised light.

IMPORTANCE

The carbohydrates, often termed as sugars, are the '*staff of life*' for most organisms. On the basis of mass, they are the most abundant class of biomolecules in nature. Carbohydrates are also known as *saccharides* (*sakcharon*[G] = sugar or sweetness) since many of those of relatively small molecular weight have a sweet taste, although this is not true of those with large molecules. They are widely distributed molecules (*moles*[L] = mass) in both plant and animal tissues. They are indispensable for living organisms, serving as skeletal structures in plants and also in insects and crustaceans. They also occur as food reserves in the storage organs of plants and in the liver and muscles of animals. In addition, they are an important source of energy required for the various metabolic activities of the living organisms; the energy being derived as a result of their oxidation. They also serve to lubricate skeletal joints, to provide adhesion between cells and to confer biological specificity on the surface of animal calls.

Plants are considerably richer in carbohydrates in comparison to the animals. In fact, animal and plant tissues differ widely in the relative abundance of the various major classes of constituent chemicals (Table 5–1).

Table 5–1. Percentage composition of various major classes of constituent chemicals in plants and animals

Organism	*Water*	*Carbohydrates*	*Proteins*	*Lipids*	*Ash*
Animal	60	1	20	15	4
Plant	60	30	5	1	4

NOMENCLATURE AND DEFINITION

The term 'carbohydrate' was originally coined for this class of compounds as most of them were '**hydrates of carbon**' or could be represented by the general formula, $C_x(H_2O)_y$. Later, it was found that some of them, such as deoxyribose ($C_5H_{10}O_4$) and rhamnose ($C_6H_{12}O_5$) do not have the required ratio of hydrogen to oxygen. In addition, certain other carbohydrates are now known to possess nitrogen (*e.g.*, glucosamine, $C_6H_{13}O_5N$), phosphorus or sulfur also and obviously do not coincide with the above general formula. Furthermore, formaldehyde (H.CHO or CH_2O), acetic acid (CH_3.COOH or $C_2H_4O_2$) and lactic acid (CH_3.CHOH.COOH or $C_3H_6O_3$) which have C, H and O and the ratio of H : O is also the same as in water, but are not a carbohydrates. Hence, *the continued usage of the term 'carbohydrate' is for convenience rather than exactness.*

To accommodate a wide variety of compounds, the carbohydrates are nowadays broadly defined as **polyhydroxy aldehydes or ketones and their derivatives** or as substances that yield one of these compounds on hydrolysis.

CLASSIFICATION

Carbohydrates are usually classified in 3 groups :

A. **Monosaccharides** or **Monosaccharoses** (*mono*[G] = one; *sakcharon*[G] = sugar). The monosaccharides, often called *simple sugars*, are compounds which possess a free aldehyde (—CHO) or ketone (= CO) group and 2 or more hydroxyl (—OH) groups. They are, in fact, the simplest sugars and cannot be hydrolyzed into smaller units. Their **general formula** is $C_n(H_2O)_n$ or $C_nH_{2n}O_n$.

The monosaccharides may be subdivided into trioses, tetroses, pentoses, hexoses, heptoses etc., depending upon the number of carbon atoms they possess; and as aldoses or ketoses, depending upon whether they contain aldehyde or ketone group. Some important examples are :

Name	**Formula**	**Aldoses** (*Aldo sugars*)	**Ketoses** (*Keto sugars*)
Trioses	$C_3H_6O_3$	Glycerose	Dihydroxyacetone
Tetroses	$C_4H_8O_4$	Erythrose	Erythrulose
Pentoses	$C_5H_{10}O_5$	Ribose	Ribulose
Hexoses	$C_6H_{12}O_6$	Glucose	Fructose
Heptoses	$C_7H_{14}O_7$	Glucoheptose	Sodoheptulose

Both these characters (*i.e.*, the number of carbon atoms and the nature of functional group present) may also be combined into one. Thus, for example, glycerose (= glyceraldehyde) is an aldotriose; ribulose, a ketopentose and glucose, an aldohexose. It is noteworthy that, *except fructose, ketoses are not as common as aldoses*. The most abundant monosaccharide in nature is the 6-carbon sugar, D-glucose.

Sometimes, a distinction in naming between aldoses and ketoses is also maintained. The suffix

-oses is kept reserved for the aldoses and the suffix -*uloses* is used for ketoses. Thus, glucose is a hexose and fructose, a hexulose. However, a few ketoses are named otherwise, such as fructose (*fructus*^{L} = fruit) as fruits are a good source of this sugar.

Anselme Payen, codiscoverer of diastase, also isolated a compound common to cell walls of higher plants, which he named `cellulose'. His naming of this polysaccharide, in fact, introduced the **-ose** suffix into the nomenclature of carbohydrates.

B. **Oligosaccharides** or **Oligosaccharoses** (*oligo*^{G} = few). These are *compound sugars* that yield 2 to 10 molecules of the same or different monosaccharides on hydrolysis. Accordingly, an oligosaccharide yielding 2 molecules of monosaccharide on hydrolysis is designated as a dissaccharide, and the one yielding 3 molecules of monosaccharide as a trisaccharide and so on. The **general formula** of disaccharides is $C_n(H_2O)_{n-1}$ and that of trisaccharides is $C_n(H_2O)_{n-2}$ and so on. A few examples are :

Disaccharides – Sucrose, Lactose, Maltose, Cellobiose, Trehalose, Gentiobiose, Melibiose

Trisaccharides – Rhamninose, Gentianose, Raffinose (= Melitose), Rabinose, Melezitose

Tetrasaccharides – Stachyose, Scorodose

Pentasaccharide – Verbascose

The molecular composition of the 3 legume oligosaccharides (*viz.*, raffinose, stachyose and verbascose) is shown below :

α-Galactose (1–6) α-Glucose (1–2) β-Fructose — Raffinose

α-Galactose (1–6) α-Galactose (1–6) α-Glucose (1–2) β-Fructose — Stachyose

α-Galactose (1–6) α-Galactose (1–6) α-Galactose (1–6) α-Glucose (1–2) β-Fructose — Verbascose

C. **Polysaccharides** or **Polysaccharoses** (*poly*^{G} = many). These are also compound sugars and yield more than 10 molecules of monosaccharides on hydrolysis. These may be further classified depending on whether the monosaccharide molecules produced as a result of the hydrolysis of polysaccharides are of the same type (homopolysaccharides) or of different types (heteropolysaccharides). Their *general formula* is $(C_6H_{10}O_5)_x$. Some common examples are :

Homopolysaccharides – Starch, Glycogen, Inulin, Cellulose, Pectin, Chitin

Heteropolysaccharides – "Specific soluble sugar" of pneumococcus type III, Hyaluronic acid, Chondrotin

ASYMMETRY

Jacobus H. van't Hoff, the first Nobel Laureate in Chemistry (1901) and Joseph A. Le Bel, in 1894, introduced the **concept of tetrahedral carbon atom**.

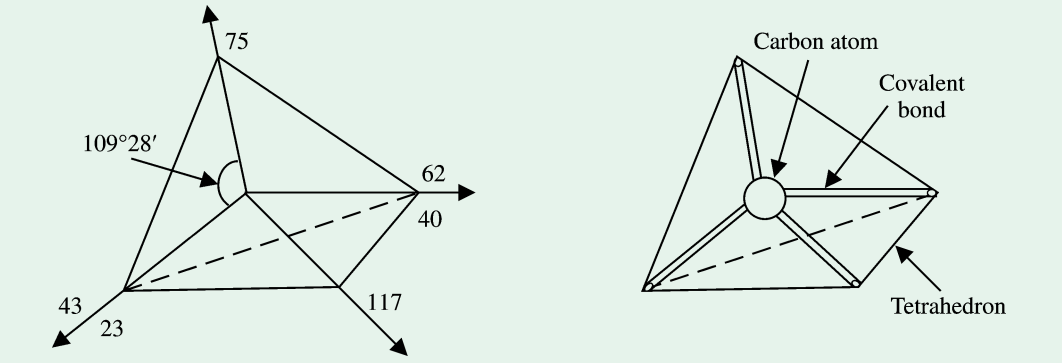


Fig. 5–1. Spatial arrangement of the valences of carbon

JACOBUS HENRICUS VAN'T HOFF
(LT, 1852-1911)

A Dutch chemist. Best known for his hypothesis about the structure of carbon atom. Made notable contributions in the field of Physical Chemistry, particularly on the theory of dilute solutions. Won the first-ever Noble Prize in Chemistry in 1901 for work on rates of reactions, chemical equilibrium and osmotic pressure.

JOSEPH A. LE BEL
(LT, 1847-1930)

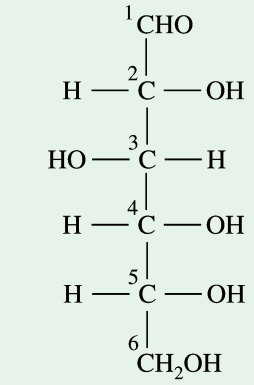
A French chemist. Best known for his stereochemical theory of carbon and its compounds. Co-discoverer of the cause of optical activity with van't Hoff. Synthesised the first optically-active compound with asymmetric nitrogen atom.

It is now recognized that the carbon atom has the shape of a tetrahedron in which the carbon nucleus resides in the centre of this tetrahedron and the 4 covalent bonds extend out to the corners of it. The angle between any two covalent bonds is 109°28′ (Fig. 5–1).

The 5th Incarnation of Carbon

Carbon is one of the most important elements in the universe since *life itself*, as we know it, *is carbon-based.* The versatility of water to form a huge variety of compounds and their stability even under extreme conditions are the secret of carbon acting as the vehicle of life. Versatility of carbon compounds apart, the element itself has become a centre of great attention in recent years. For long, we knew carbon could manifest itself in vividly contrasting styles. It was known to assume a soft and black form as **graphite**; it was known equally well in its hard and sparkling - white incarnation of **diamond** which is the hardest material on earth. Then it was discovered (in 1985) to have had the shape of a soccer ball (called **fullerenes**), and of a cylindrical tube (called **nanotubes**). And now an announcement has been made that carbon can also assume a foamy shape, called **nanofoam.**

This new spongy form of carbon is also a modification of graphitic form. It is extremely light-weighted form and was created when some researchers, at the Australian National University in Canberra, bombarded a piece of graphite with a high-power laser beam. The graphite piece vapourised as temperature rose to more than 10,000°C. On cooling, the vapourised carbon atoms formed a web of tubes. But these tubes instead of being aligned in any particular direction (as happens in the case of nanotubes), were found to be highly dispersed, criss-crossing with one another to form a highly-porous mass. This web of nanotubes is seen as a nonofoam. Surprisingly, the nanofoam, unlike all the other forms of carbon, is *magnetic*. Although the magnetism is ephemeral at room temperature, it is an entirely new and unexpected property in the case of carbon. Because of light weight, foamy nature and magnetic property, nanofoam is being seen as useful for cancer and brain scans through magnetic resonance imaging (MRI). Besides, it might also be useful in the treatment of tumours. Being foam, it would be a bad conductor of heat and if injected into tumours, and heated, it would retain the heat locally and kill tumours almost without affecting other body parts. But it needs confirmation through a series of studies.



Glucose

[The carbon atoms of a sugar are numbered from the end closest to the aldehyde or ketone group.]

In the formula given above for glucose, it may be observed that a different group is attached to each of the 4 valences of carbon atoms 2 to 5. For example, the 4 groups attached at carbon atom 2 are :

— CHO, —H, — OH and HO — C — H

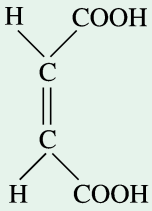
Thus, a carbon atom to which 4 different atoms or groups of atoms are attached is said to be **asymmetric** or **chiral** (*cheir*[G] = hand).

ISOMERISM

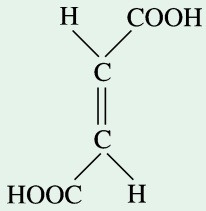
The term **isomer** (*isos*[G] = equal ; *meros*[G] = part) was originally applied by Jönes Jacob Berzelius, in 1827, to different compounds with the same molecular formula, and the phenomenon was called **isomerism**. The presence of asymmetric carbon atoms in the carbohydrates makes possible the formation of isomers in them. It shall, hence, be beneficial at this stage to discuss in brief about isomers.

The isomers are of 2 types : structural isomers and stereoisomers. **Structural isomers** have the same molecular formula but possess different structures. The difference in structure may be exhibited either in the length of the carbon chain (**chain isomers**) or in the position of a substituent group (**positional isomers**) or in possessing different functional groups (**functional-group isomers**).

Stereoisomers, on the other hand, have the same molecular formula and the same structure but differ only in spatial configuration. Stereoisomers are of 2 types : geometrical and optical. **Geometrical** or **cis-trans** (*cis*[L] = same side; *trans*[L] = across) **isomers** arise from peculiar geometry of compounds having a double bond within the carbon chain. It may be illustrated by *cis-trans* pair, maleic and fumaric acids.

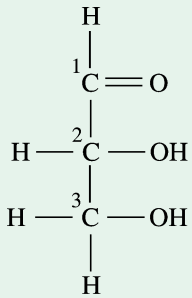


Maleic acid
(*cis* form)

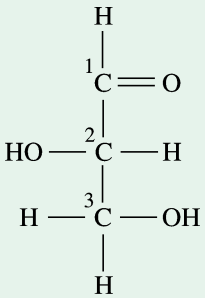


Fumaric acid
(*trans* form)

Optical isomers (= enantiomers) differ from each other in the disposition of the various atoms or groups of atoms in space around the asymmetric carbon atom. These are, in fact, the mirror image of each other. These may also be likened to left- and right-handed gloves. For example, the glyceraldehyde has only one asymmetric carbon atom (numbered as 2) and it can, therefore, exist in 2 isomeric forms :



D-glyceraldehyde



L-glyceraldehyde

One form in which H atom at carbon 2 is projected to the left side and OH group to the right is designated as **D-form** and the other form where H atom is projected to the right side and OH group to the left is called as **L-form** (note the use of small capital letters D and L) Similarly, a compound having 2 asymmetric carbon atoms (*e.g.*, a tetrose) might exist in 4 optical isomeric froms.

The number of possible isomers of any given compound, thus, depends upon the number of asymmetric carbon atoms present in its molecule. According to the **rule of n** (also called as

Le Bel-van't Hoff rule), the total number of optical isomers of a compound will be equal to 2^n, where *n* represents the number of asymmetric carbon atoms present in the molecule.

In fact, in sugars with 2 or more asymmetric carbon atoms, the designation of D- and L- forms depends upon the orientation of the H and OH groups around the lowermost asymmetric carbon atom (*i.e.*, the asymmetric carbon furthest from the aldehyde or keto group). The distribution of the H and OH groups on the other carbon atoms in the molecule is of no importance in this connection. It is interesting to note that *majority of the monosaccharides found in the human body are of D type*. However, some sugars do occur naturally in their L form such as L-arabinose, a pentaaldose.

KILIANI CYANOHYDRIN SYNTHESIS

A method for the synthesis of monosaccharides was first proposed by Heinrich Kiliani in 1886. It is, in fact, a method by which the chain length of a carbohydrate may be increased. The application of Kiliani synthesis to D-glyceraldeyde resulting in the production of 2 tetroses, D-erythrose and D-threose is shown in Fig. 5–2.

The process is based upon the addition of HCN to the carbonyl group of aldehydes (or ketones) of the sugars forming cyanohydrin (Reaction-1). This reaction creates a new asymmetric carbon atom, marked by an asterisk in the figure. Thus, two compounds differing in conformation about the newly-formed asymmetric carbon atom are formed. These two cyanohydrins are, then, hydrolyzed to produce carboxylic acids (Reaction-2) which are later converted to γ-lactones or inner esters (Reaction-3). Finally, the lactones are reduced to the corresponding aldoses, containing one carbon atom more than their parent sugar (Reaction-4).

The process can be repeated and the 4 isomeric D-pentoses may be produced; and from these the 8 isomeric D-hexoses would also result.

The structural relationships of the monosaccharides (the aldoses, for example) may be visualized by the formulae given in Fig. 5–3. It is easy to remember the configurations of the aldoses by recalling the *Rosanoff scheme*, drawn out on the chart and mneumonic—ET; RAXL; AAGMGIGT.

The term **'conformation'** was first introduced by Walter Norman Haworth (1929), an English chemist. In its broadest sense, conformation has been used to describe different spatial arrangements of a molecule which are not superimposable. This means that, in effect, the terms *conformation* and *configuration* are equivalent. In the classical sense, the definition of conformation does not include the internal forces acting on the molecule. The term conformation, however, is the spatial arrangement of the molecule when all the internal forces acting on the molecule are taken into account. In more restricted sense, the term conformation is used to denote different spatial arrangements arising due to twisting or rotation of bonds of a 'given' configuration (used in the classical sense). The terms **rotational isomers** and **constellations** have also been used in the same sense as conformations.

Any two sugars which differ from each other only in the configuration around a single asymmetric carbon atom other than the carbonyl carbon atom are called **epimers.** Glucose and galactose, for example, form an epimeric pair as they differ with respect to carbon 4 only. Similarly, glucose is also epimeric with mannose (differing in C_2 configuration) and allose with altrose (differing also in C_2 configuration).

The two D- and L-forms of a compound constitute a pair of **enantiomers** (*enantios*[G] = opposite) or **enantiomorphs**. Thus, D-erythrose and L-erythrose are enantiomers; also D-glucose and L-glucose. The enantiomers (also called optical isomers or double image isomers) are nonsuperimposable mirror images of each other but are chemically identical in their reactions. They agree in their melting points, solubility etc., but differ in their ability to rotate the plane of polarized light in a polarimeter; a solution of one of the two enantiomers rotates the plane to the right, and a solution of the other to the left.

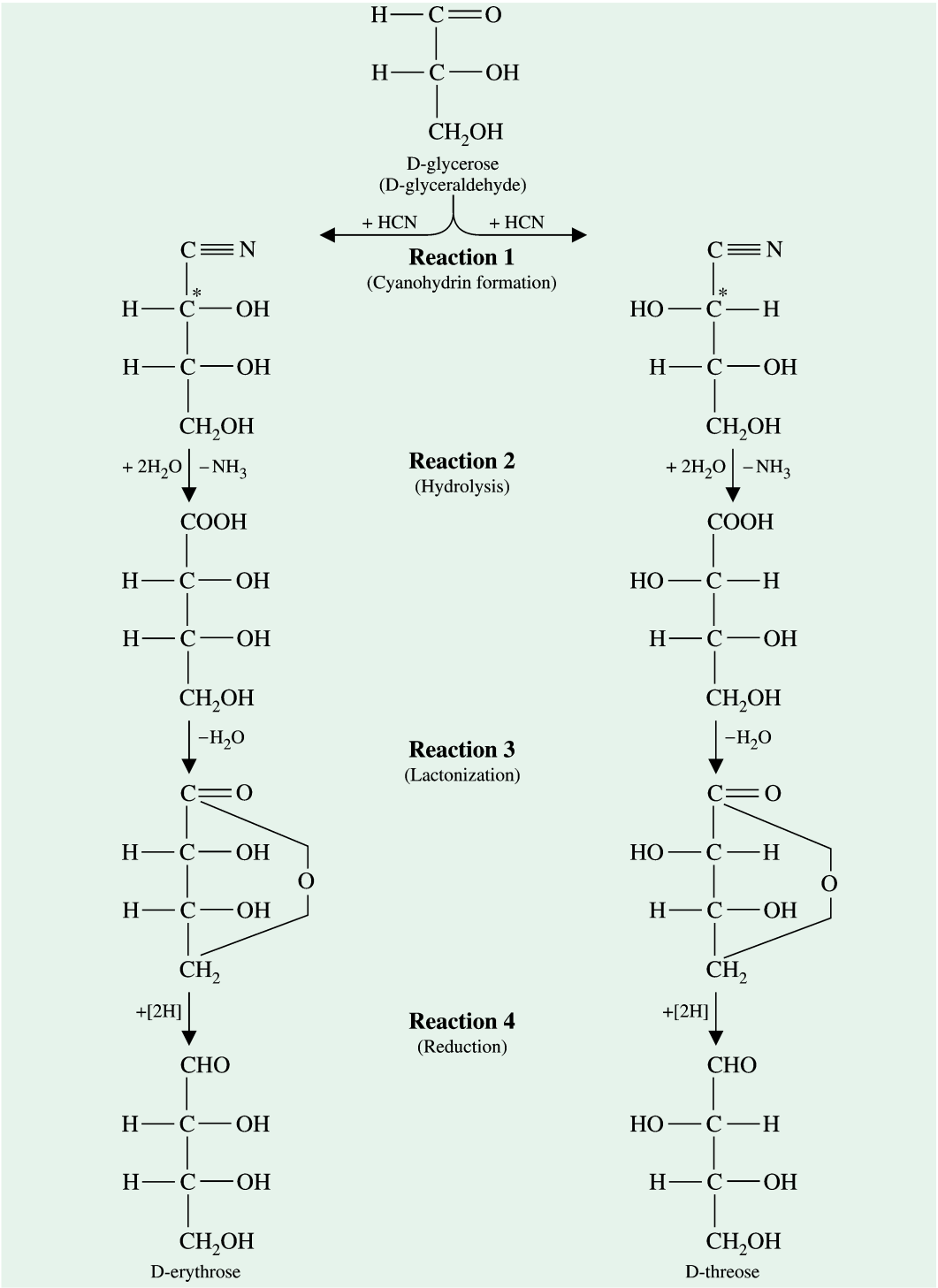


Fig. 5–2. Application of the cyanohydrin synthesis to D-glyceraldehyde

The conversion of D-glyceraldehyde into an aldotetrose yields D-erythrose and D-threose. These are called **diastereoisomers** *i.e.*, isomers but not mirror images of each other. In fact, diastereoisomers are different forms of a compound with two asymmetric centres. It may, hence,

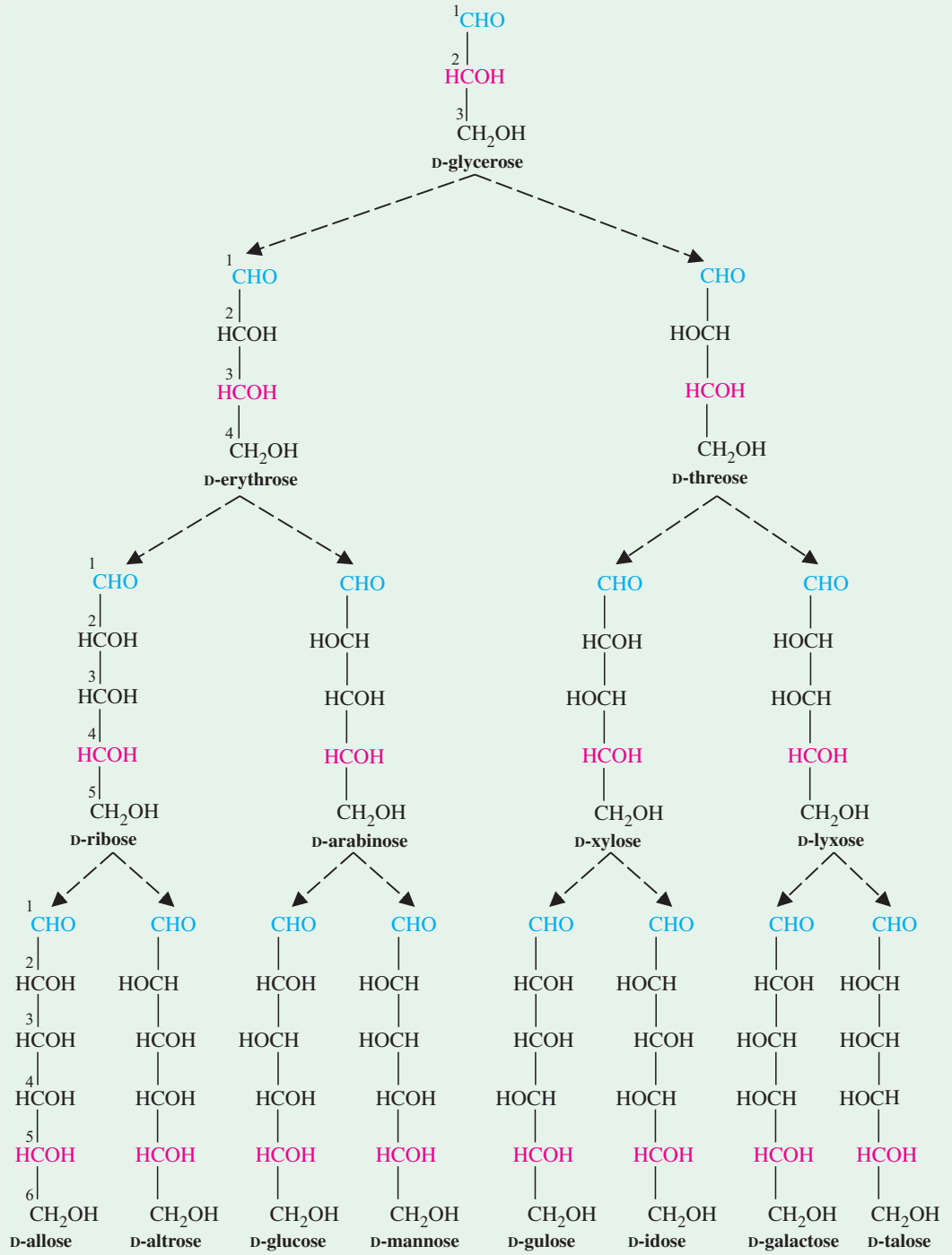


Fig. 5–3. D. Aldoses containing three, four, five and six carbon atoms

Aldoses contain an aldehyde group (shown in blue) and have the absolute configuration of D-glyceraldehyde at the asymmetric centre (shown in red) farthest from the aldehyde group. The numbers indicate the standard designations for each carbon atom. The formulae of L-aldoses are in each case the mirror images of these structures.

be inferred that if two optical isomers are not enantiomers, they are related as diastereoisomers. They differ from each other in their melting points, solubilities and the chemical properties, in general.

OPTICAL ISOMERISM

The presence of asymmetric carbon atoms in the compound also confers optical activity to it. When a beam of polarized light is passed through an optically active solution, it may be rotated either to the right or to the left, depending upon the type of optical isomer present. A compound rotating the plane of polarized light to the right is called as *dextrorotatory* (*dexter*^{L} = right, on the right side; *rotatus*^{L} = pp of *rotare*^{L}, to turn) or clockwise and is designated as *d* or (+) type. If the compound causes rotation of polarized light to the left, it is said to be *levorotatory* (*laevus*^{L} = left, on the left side) or anticlockwise and is denoted as *l* or (–) type. The direction in which the light is rotated (or in other words, the optical rotation) is a specific property of the molecule. It should, however, be emphasized that *the optical rotation is not at all related with the two* D *and* L *forms of a compound*. Thus, D-glucose is dextrorotatory and D-fructose is levorotatory. If it is desired to indicate the direction of rotation, the two names may be written as D(+)-glucose and D(–)-fructose. Similarly, D-erythrose is levorotatory and D-threose, dextrorotatory.

When equal amounts of extrorotatory and levorotatory isomers are present, the resulting mixture becomes optically inactive because the optical activities of each isomer cancel each other. Such a mixture is called a **racemic** or **dl-mixture** or (±)-**conglomerate** and this process of converting an optically active compound into the racemic modification is known of **racemisation** (*racemus*^{L} = grape). Compounds produced synthetically are invariably racemic in nautre because equal chances exist for the formation of two types of optical isomers or the *d* and *l* antipodes.

It may be concluded that all monosaccharides are optically active (except aldobiose and ketotriose) since they contain one or more asymmetric carbon atoms in their molecules.

Mutarotation

When a monosaccharide is dissolved in water, the optical rotatory power of the solution gradually changes until it reaches a constant value (Dubrunfaut, 1846). A freshly-prepared aqueous solution of α-D-glucose, for instance, has a specific rotation, $[\alpha]_D^{20}$ of +112.2°. And when this solution is allowed to stand, the rotation falls to +52.7° and remains constant at this value. The final stage can be attained more quickly either by heating the solution or by adding some catalyst which may be an acid or an alkali. This gradual change in specific rotation is known as **mutarotation** or changing rotation. In fact, this term reflects the discovery of the phenomenon by way of changes in the optical rotation of certain carbohydrates. The terms **multirotation** and **birotation** have sometimes also been used for mutarotation. The value of mutarotation for α-D-glucose is (+112.2°) – (+52.7°) or +59.5°. A fresh solution of β-D-glucose, on the other hand, has a rotation value of +18.7°; on standing, it also changes to the same value, +52.7°. *All reducing sugars (except a few ketoses) undergo mutarotation*. The specific rotations for the anomers of glucose and for the aqueous equilibrium mixture are :

$$\underset{\text{α-D-glucose}}{[\alpha]_D^{20} = +\,112.2^\circ} \longrightarrow \underset{\text{Mutarotational equilibrium mixture}}{[\alpha]_D^{20} = +\,52.7^\circ} \longleftarrow \underset{\text{β-D-glucose}}{[\alpha]_D^{20} = +\,18.7^\circ}$$

These substances (open and ring forms) interconvert due to a dynamic equilibrium. Although the ring form predominates, each molecule spends some time in the open form, when asymmetry is lost. When the open-chain form passes back to the ring form, it is possible for it to give rise to either anomer. Thus, a pure sample of an anomer only remains so when in the solid state. In solution, particularly at alkaline pH, a pure sample of one anomer will change to an equilibrium mixture of the two forms. There need not be equal quantities of the two forms at equilibrium because there are other centres for asymmetry and so anomers (like *allo* and *threo* pairs of amino acids) do not have identical physical properties, including ΔG° of formation. Equilibrium between

the anomers is called mutarotation and it can be followed by observing optical rotation (Fig. 5–4).

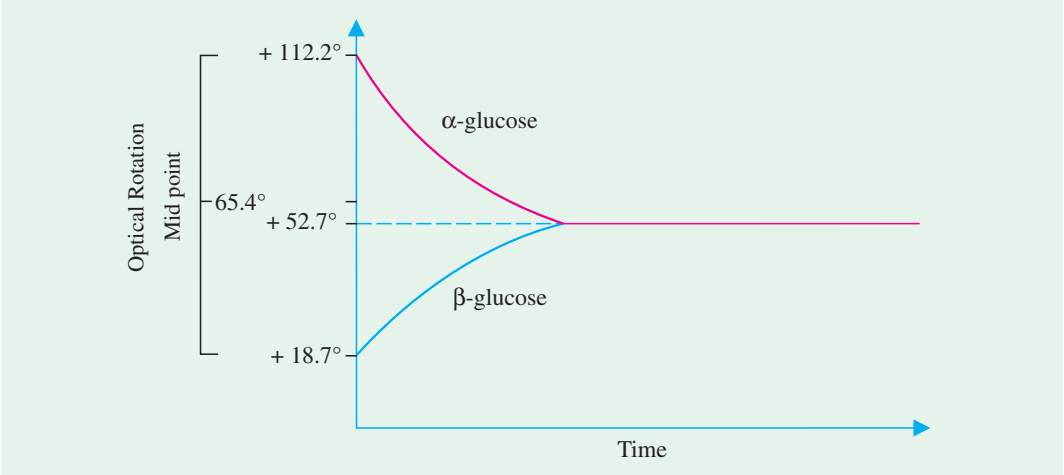


Fig. 5–4. The experimental detection of mutarotation

Mechanism of mutarotation. The phenomenon of mutarotation appears to be due to the changes of α- to β-forms and vice versa via the straight chain aldo or keto form (refer to the subsequent section). When an equilibrium is attained, the characteristic rotation is reached. According to Lowry (1925), mutarotation can take place only in the presence of an amphiprotic solvent (*i.e.*, a solvent which can act both as an acid and a base). Water is one such solvent. Thus, Lowry and Faulkner (1925) showed that mutarotation is checked in cresol solution (an acidic solvent) and in pyridine solution (a basic solvent). It is assumed that when mutarotation takes place, the ring opens and then recloses either in the inverted position or in the original position. The existence of the open chain form in mutarotation is proved by certain evidences. For example, the absorption spectra of fructose and sorbose in aqueous solution indicate the presence of open chain forms ; the aldoses, however, give negative results (Bednarczyk *et al*, 1938).

FORMULATION OF MONOSACCHARIDES

Formula of Glucose—Linear Form

An organic chemist intends to incorporate as many characteristics as possible in writing the formulae (Fig. 5–5). When the molecular formula of glucose, $C_6H_{12}O_6$ is written in a form as propounded by Fittig and Baeyer, it tells us the presence of an aldehyde and 5 hydroxyl groups. Replacement of this formula by Fischer projection formula informs the reader about some more details. For example, the presence of 4 asymmetric carbon atoms in the molecule indicates towards the existence of 2^4 or 16 stereoisomers.

Ring Form

However, even Fischer formula fails to describe certain other properties of glucose. Hence, another method of representing the structure was searched for. The aldehyde or ketone group of a sugar can react with hydroxyl groups of alcohols forming **hemiacetals** or **hemiketals**, respectively. For the larger sugars (n = >4), this happens *within the same molecule* to form a 5- or 6-membered ring.

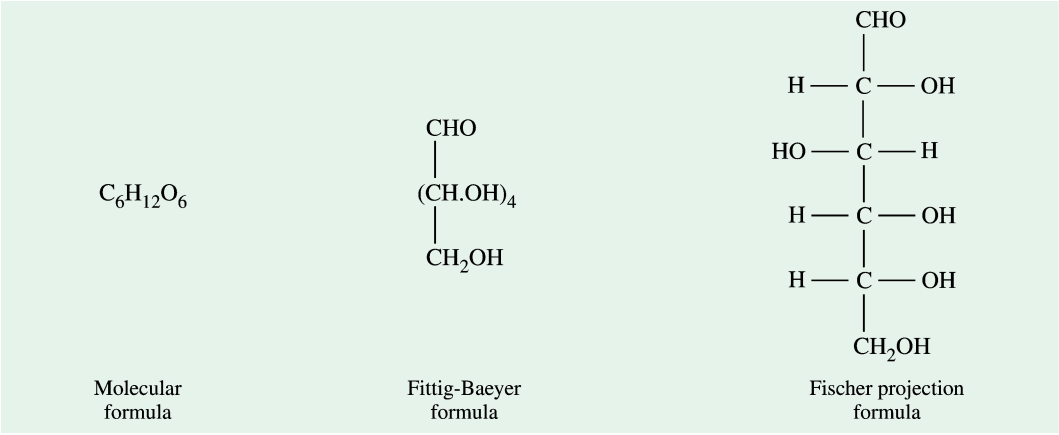
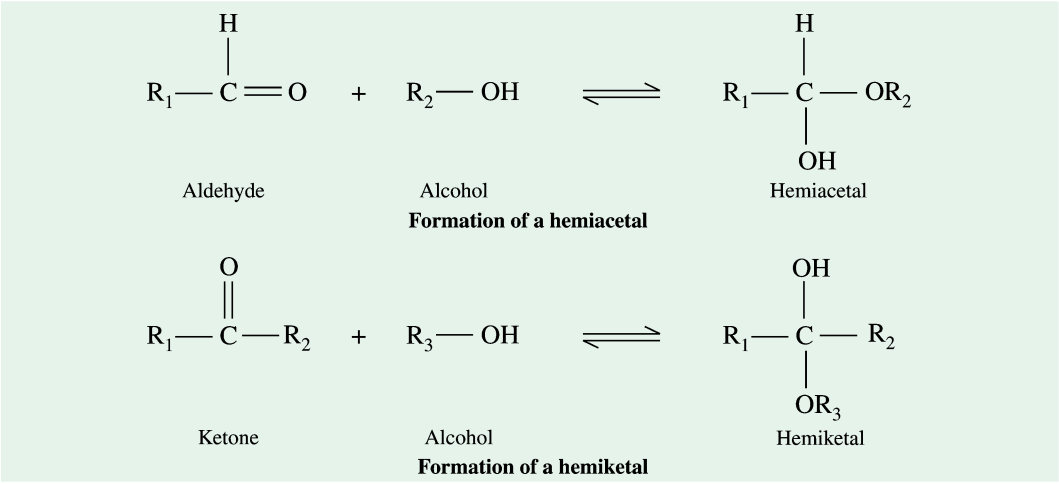
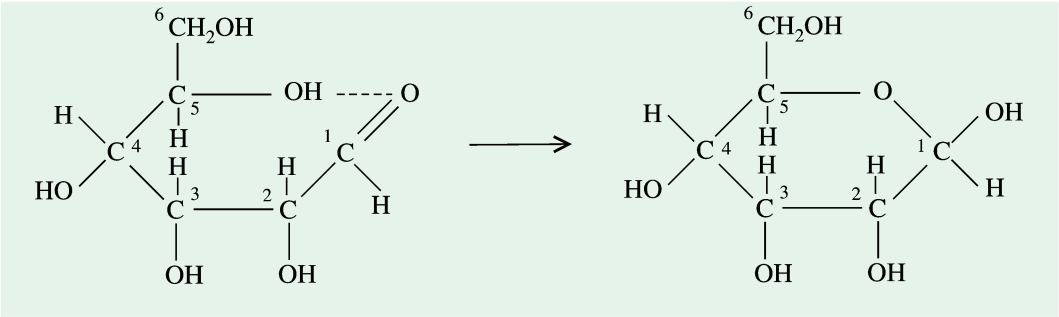


Fig. 5–5. Formulation of glucose

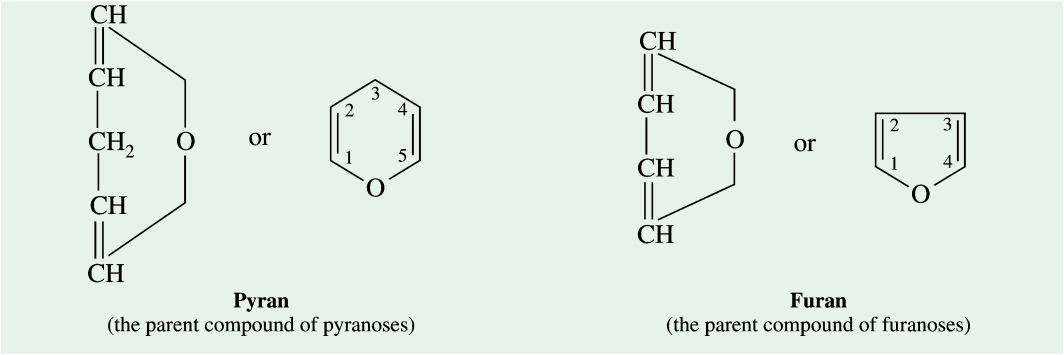
In fact, the angles of the tetrahedral carbon atom tend to bend the glucose molecule forming a ring. The aldehyde group and the alcohol group of carbon 5 or carbon 4 readily approach each



other forming intramolecular *hemiacetals*. Similarly, the keto group of a ketose can also approach the alcohol group of either carbon 5 or carbon 4 forming intramolecular *hemiketals*. This results in the formation of either a 6-membered ring (**pyranose form**) or a 5-membered ring (**furanose form**). A 7-membered ring, however, becomes too strained to allow participation of the OH group of carbon 6 of aldohexoses with the CHO group in ring formation. The pyranose form has a lower ΔG° of formation than the furanose form.



The terminology of such structures is, in fact, based on the 2 simple organic compounds, exhibiting a similar ring structure. These are **pyran** and **furan**. *The pyranose forms of the sugars are more stable than the furanose forms in solution.*



The formation of a ring structure in the glucose molecule creates a new centre of asymmetry, *i.e.*, the carbon 1 (carbon 2 in ketoses). One may, therefore, find that the ring form of glucose can exist as either of the 2 isomeric forms, termed as α and β isomers. These two isomers are, in fact, diastereoisomers rather than enantiomers because the α-form of D-glucose is not the mirror image of the β-form. They are also known as **anomers** (*ano*^{G} = upper) as they differ in configuration only around the hemiacetal or anomeric carbon atom (*i.e.*, carbon 1). The designation of α- and β-forms is based on the suggestion of Freudenberg (1933) who stated that those anomers having the same configuration at both the anomeric and penultimate carbon atoms should be called as α-form while in β-form the configuration would be different at both these carbon atoms. In other words, the configuration of H atoms at both these carbons (anomeric and penultimate) is **erythro** (written on the same side of the structure) in α-form and **threo** (written on opposite sides) in β-form. The α- and β-forms of D-glucose in both pyranose and furanose rings along with the open chain formula are shown in Fig. 5–6.

As shown in the above figure, the hemiacetal formation is of reversible nature. Thus, D-glucose in aqueous solution exists as an equilibrium mixture of 5 compounds. The pyranose form, however, predominates in solution. An equilibrium mixture of glucose contains about one-third (37%) α anomer, two-thirds (62%) β anomer and very little (<< 1%) of the open-chain form. Likewise, the α and β anomers of both the pyranose and the furanose forms of fructose interconvert through the open-chain form. In fact, under physiologic conditions in solution, monosaccharides with 5 or more carbons are typically more than 99% in the ring forms. The distribution between pyranose and furanose forms depends on : (*a*) the particular sugar structure, (*b*) the pH, (*c*) the solvent composition, and (*d*) the temperature. Respresentative data obtained from nuclear magnetic resonance (NMR) studies are shown in Table 5–2. When the monomers are incorporated into polysaccharides, the structure of the polymer may also influence the ring form chosen. As an instance, as Table 5–2 depicts, D-ribose exists in solution as a mixture of the two ring forms. But in biological polysaccharides, specific forms are stabilized. Ribonucleic acid, for example, contains exclusively ribofuranose whereas some plant cell wall polysaccharides have pentoses entirely in the pyranose form.

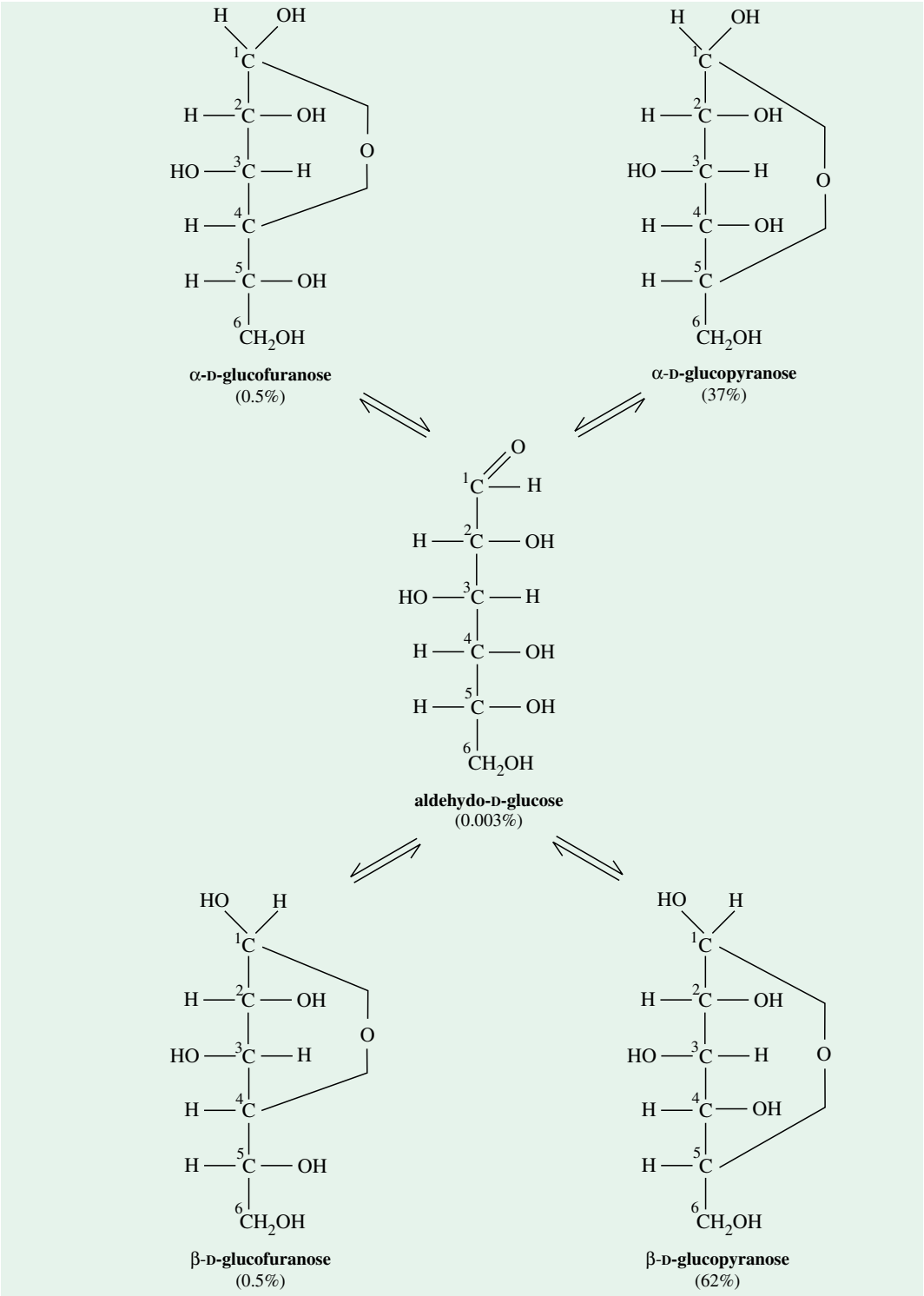


Fig. 5–6. Various forms of d-glucose present in an aqueous solution

[Note that in α-form, the OH group at anomeric carbon 1 is also on the right side like that at the penultimate carbon atom 5 ; whereas in β-form, the OH group at carbon 1 is on the left and that on carbon 5 is on the right side. Figures in parentheses indicate the molecular percentage present in an equilibrium mixture at pH 7.]

Table 5–2. **Relative amounts of tautomeric forms for some monosaccharide sugars at equilibrium in water at 40°C**

Monosaccharide	*Relative Amount* (in%)				
	α *pyranose*	β *pyranose*	α *furanose*	β *furanose*	*Total furanose*
Ribose	20	56	06	18	24
Lyxose	71	29	–[a]	–[a]	<1
Altrose	27	40	20	13	33
Glucose	36	64	–[a]	–[a]	<1
Mannose	67	33	–[a]	–[a]	<1
Fructose	03	57	09	31	40

Note that in all cases, the open-chain form is much less than 1%.
a designates much less than 1%.

Like other kinds of steroisomers, these α and β forms rotate the plane of polarized light differently and can be distinguished that way. The monosaccharides can undergo interconversion between the α and β forms, using the open-chain structure as an intermediate. This process is referred to as **mutarotation.** A purified anomer, dissolved in aqueous solution, will approach the equilibrium mixture, with an accompanying change in the optical rotation of the solution. Enzymes called **mutarotases** catalyze the interconversion of anomeric sugars *in vivo*.

Haworth Perspective Formula

Although Fischer formulae are useful in indicating configurational differences among sugars, they are tedious to write. Haworth (1929), an English chemist, devised another method of representing their structure, resembling a hexagon (pyranose form) or a pentagon (furanose form). In such a projection, the carbon atoms in the ring are not explicitely shown. The approximate plane of the ring is perpendicular to the plane of the paper, with the heavy line on the ring projecting toward the reader. Haworth projections more clearly approximate the predominant "chair" structure of glucose (Fig. 5–7) and other hexose sugars that exist in solutions.

WALTER NORMAN HAWORTH

(LT, 1883–1950)

Walter Norman Haworth, a Briton, had been a student of Otto Wallach and W.H. Perkin at the Universities of Manchester and Göttingen. He was awarded 1937 Nobel Prize in Chemistry for his work on the constitution of carbohydrates and vitamin C, along with Paul Karrer of Switzerland. Haworth's contributions include : methylation of sugars, pyranose and furanose structures of monosaccharides, structure of starch, cellulose and amygdalin, and structure and synthesis of ascorbic acid.

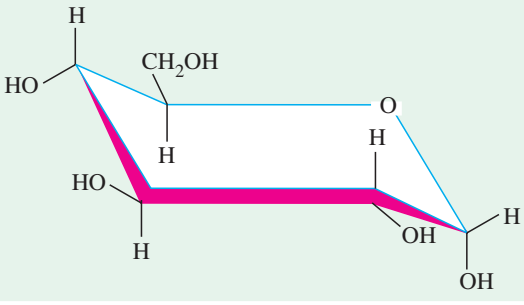


Fig. 5–7. **"Chair" form of α-D-glucopyranose**

In the pyranose form, carbon atoms 1 to 5 of the aldohexoses and the O atom of the ring are represented in the form of a hexagon and the plane of this ring projects from the paper. Bonds that are nearer to the reader are thickened in this type of formula and the carbon atoms are numbered clockwise. Fischer formula may be converted to Haworth formula by twisting it around imaginatively or by constructing models. Certain '**assumptions**' have, however, been followed for conversion. These are :

1. Any group to the right of the carbon chain in Fischer formula is written *down* the plane of the ring in the Haworth formula ; those to the left are written *up*. A notable exception to this assumption is carbon atom 5 where H atom, present on the left side, is shown below the plane of the ring instead of showing it above the plane of the ring. This is as a result of torsion needed to bring about ring closure. The carbon atom 5 rotates so that the O atom of the OH group on the anomeric carbon comes in the plane of the first five carbon atoms. Consequently, the H atom on carbon 5 is shifted to the other side of the carbon chain by rotating through more than 90°.

2. Another important assumption is that the carbon atom (or atoms) not involved in ring formation (*e.g.*, carbon 6 in aldopyranses) will be shown above the plane of the ring, if the oxide ring in Fischer formula is to the right. Conversely, if the oxide ring is to the left, the extra carbon atom (or atoms) would be shown below the plane of the ring in Haworth projection. Two examples (α-D-glucopyranose and β-L-glucopyranose) will illustrate the mode of conversion (Fig. 5–8).

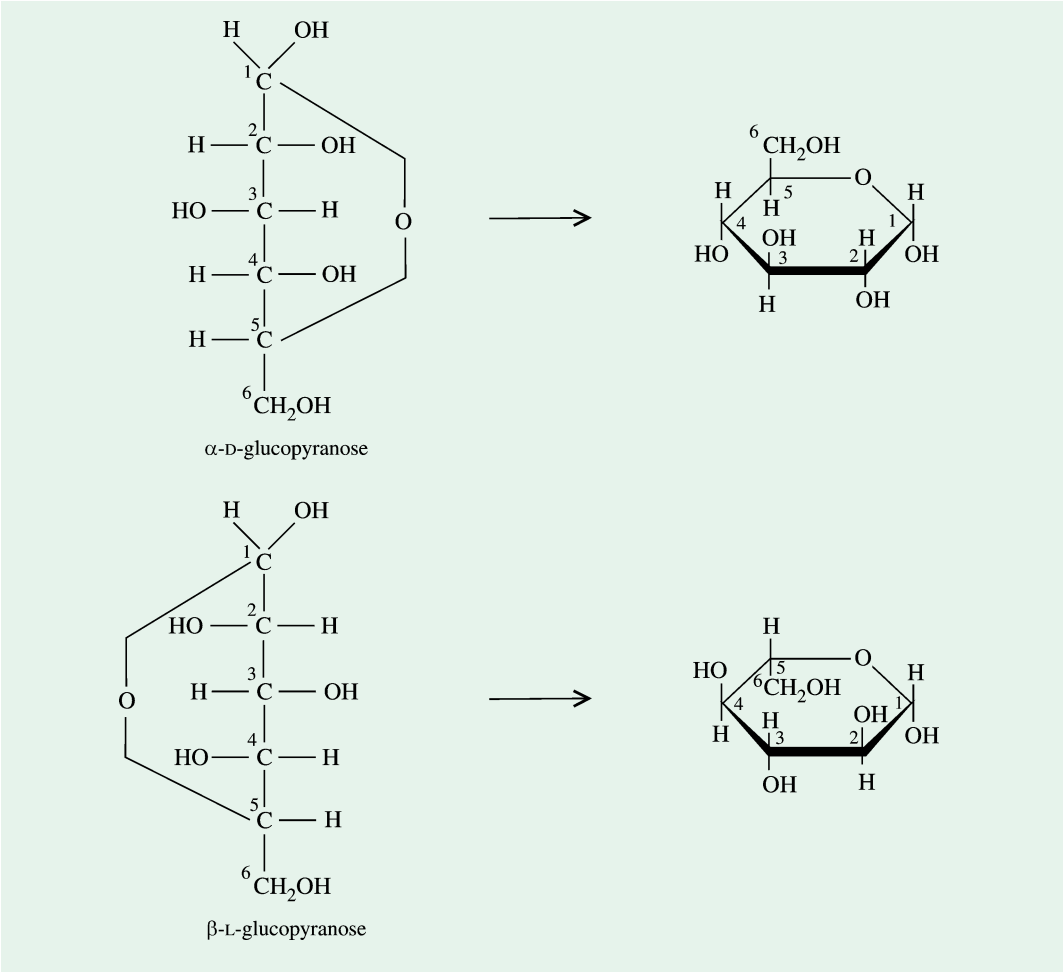


Fig. 5–8. Mode of conversion of two hexose sugars from Fischer projection formula to Haworth perspective formula

For the sake of simplicity, while writing Haworth formulae, the H and OH groups are not written and the bond indicating the single hydrogen atom is not shown. Thickened line in lower half of the hexagon is also omitted. The extra carbon atoms are, however, written as such. Thus, the above two compounds may be simply represented as in Fig. 5–9.

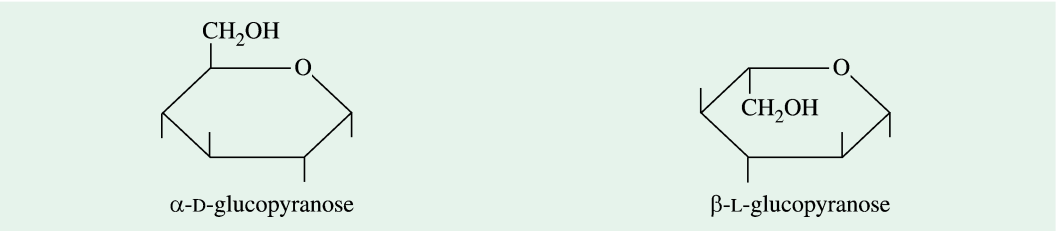


Fig. 5–9. Simplified Haworth formulae for two hexose sugars

The **6-membered pyranose ring**, like cyclohexane, cannot be planar because of the tetrahedral geometry of its saturated carbon atoms. Instead, pyranose rings adopt *chair* and *boat* conformations (Fig. 5–10). The substituents on the ring carbon atoms have two orientations : *axial* and *equatorial*. Axial bonds are nearly perpendicular to the average plane of the ring, whereas equatorial bonds are almost parallel to this plane. Axial substituents emerge above and below the average plane of the ring and sterically hinder each other if they emerge on the same side of the ring. By contrast, equatorial substituents emerge at the periphery and are less crowded. *The chair form of β-D-glucopyranose predominates because all axial positions are occupied by hydrogen atoms*. The bulkier —OH and —CH_2OH groups emerge at the less hindered periphery. On the contrary, the boat form of glucose is highly disfavoured because it is much hindered sterically.

More generally, variations in the 3-'D' structure of biomolecules are described in terms of configuration and conformation. However, great care should be exercised in the use of these terms as they are not synonyms. **Conformation** refers to the spatial arrangement of substituent groups that are free to assume different positions in space, without breaking any bonds, because of the freedom of bond rotation. In ethane (C_2H_6), for example, there is nearly complete freedom of rotation around the single C—C bond. Many different interconvertible conformations of the ethane molecule are, therefore, possible, depending upon the degree of rotation. Two conformations are of special interest: the *staggered* conformation, which is more stable than others and thus predominates, and the *eclipsed* form, which is least stable. It is not possible to isolate either of these conformational forms because *they are freely interconvertible without the breakage of any bonds and are in equilibrium with each other*.

On the contrary, **configuration** denotes the spatial arrangement of an organic molecule that is conferred by the presence of (*a*) double bonds, around which there is no freedom of rotation, or (*b*) chiral centres, around which substituent groups are arranged in a specific sequence. The diagnostic feature of configurational isomers is that they *can be interconverted only by breaking one or more covalent bonds*. For example, maleic acid is a conformational isomer of fumaric acid.

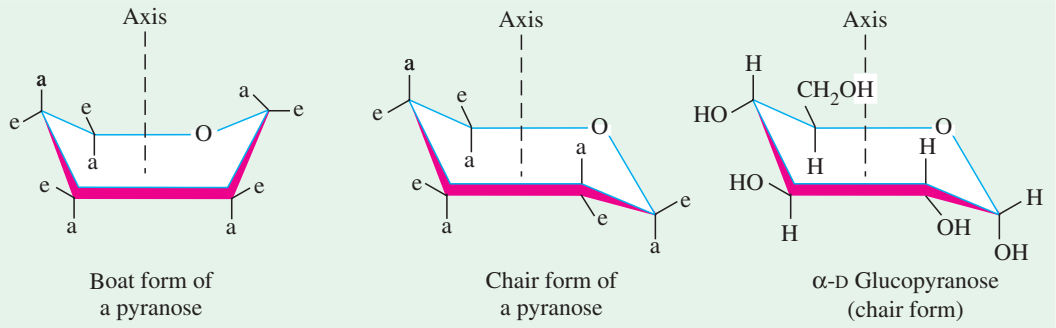


Fig. 5–10. Conformational formulae of the boat and chair forms of the pyranose ring

Substituents on the ring carbons may be either axial (a), projecting almost parallel with the vertical axis through the ring or equatorial (e) projecting almost perpendicular to this axis. For sugars with large equatorial substituents, the chair form is energetically more favourable because it is less hindered. The boat conformation is uncommon except in derivatives with very bulky substituents.

The **5-membered furanose rings,** like pyranose rings, are not planar. They can be puckered so that four atoms are nearly coplanar and the fifth is about 0.5Å away from this plane. This conformation is called *envelope form* because the structure resembles an opened envelope with the back flap raised (Fig. 5–11). In the ribose moiety of most biomolecules, either C-2 or C-3 is out of plane on the same side as C-5. These conformations are called C_2-*endo* and C_3-*endo*. [Note that the sugars in DNA double helix are in the C_2-*endo* form whereas the sugars in RNA are in the C_3-*endo* form.] Furanose rings can interconvert rapidly between different conformational states. *Furanose rings are more flexible than pyranose rings* and possibly this may be the reason for their selection as components of DNA and RNA.

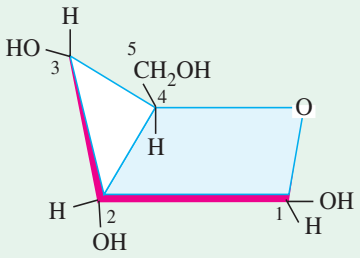


Fig. 5–11. An envelope form of β-d-ribose

The C_3-*endo* conformation is shown. C-3 is out of plane on the same side as C-5.

REFERENCES

See list following Chapter 8.

PROBLEMS

1. Account for the origin of the term *carbohydrate*.

2. Indicate whether each of the following pairs of sugars consists of anomers, epimers, or an aldose-ketose pair :

(a) D-glyceraldehyde and dihydroxyacetone

(*b*) D-glucose and D-mannose

(*c*) D-glucose and D-fructose

(*d*) α-D-glucose and β-D-glucose

(*e*) D-ribose and D-ribulose

(*f*) D-galactose and D-glucose

3. Glucose and other aldoses are oxidized by an aqueous solution of a silver–ammonia complex. What are the reaction products ?

4. The specific rotations of the α and β anomers of D-glucose are + 112 degrees and + 18.7 degrees, respectively. Specific rotation, $[\alpha]_D$, is defined as the observed rotation of light of wavelength 589 nm (the D line of a sodium lamp) passing through 10 cm of a 1 g ml^{-1} solution of a sample. When a crystalline sample of α-D-glucopyranose is dissolved in water, the specific rotation decreases from 112 degrees to an equilibrium value of 52.7 degrees. On the basis of this result, what are the proportions of the α and β anomers at equilibrium ? Assume that the concentration of the open-chain form is negligible.

5. Glucose reacts slowly with hemoglobin and other proteins to form covalent compounds. Why is glucose reactive ? What is the nature of the adduct formed ?

6. Compounds containing hydroxyl groups on adjacent carbon atoms undergo carbon–carbon

bond cleavage when treated with periodate ion (IO_4^-). How can this reaction be used to distinguish between pyranosides and furanosides ?

7. Does the oxygen atom attached to C-1 in methyl α-D-glucopyranoside come from glucose or methanol ?

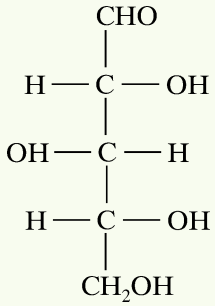
8. Fructose in its β-D-pyranose form accounts for the powerful sweetness of honey. The β-D-furanose form, although sweet, is not as sweet as the pyranose form. The furanose form is the more stable form. Draw the two forms and explain why it may not always be wise to cook with honey.

9. (*a*) Compare the number of reducing ends to nonreducing ends in a molecule of glcogen.

(*b*) Glycogen is an important fuel storage form that is rapidly mobilized. At which end—the reducing or nonreducing—would you expect most metabolism to take place ?

10. Draw Haworth projections for the following :

(*a*) in α-furanose form. Name the sugar.



(*b*) The L isomer of (*a*)

(*c*) α-D-GlcNAc

(*d*) α-D-fructofuranose

11. α-D-galactopyranose rotates the plane of polarized light, but the product of its reduction with sodium borohydride (galactitol) does not. Explain the difference.

12. Provide an explanation for the fact that α-D-mannose is more stable than β-D-mannose, whereas the opposite is true for glucose.

13. What is the natural polysaccharide whose repeating structure can be symbolized by GlcUAβ(1→3)GlcNAc, with these units connected by β(1→4) links ?

14. A freshly prepared solution of the α form of D-galactose (1 g/mL in a 10 cm cell) shows an optical rotation of +150.7°. When the solution is allowed to stand for a prolonged period of time, the observed rotation gradually decreases and reaches an equilibrium value of + 80.2°. In contrast, a freshly prepared solution (1 g/mL) of the β form shows an optical rotation of only + 52.8°. Moreover, when the solution is allowed to stand for several hours, the rotation increases to an equilibrium value of +80.2°, identical to the equilibrium value reached by α-D-galactose.

(*a*) Draw the Haworth perspective formulas of the α and β forms of galactose. What features distinguishes the two forms ?

(*b*) Why does the optical rotation of a freshly prepared solution of the α form gradually decrease with time ? Why do solutions of the α and β forms (at equal concentrations) reach the same optical rotation at equilibrium ?

(*c*) Calculate the percentage composition of the two forms of galactose at equilibrium.

15. An unknown substance containing only C, H, and O was isolated from goose liver. A 0.423 g sample produced 0.620 g of CO_2 and 0.254 g of H_2O after complete combustion in excess oxygen. Is the empirical formula of this substance consistent with its being a carbohydrate ? Explain.