# Carbohydrate Chemistry from Fischer to Now

*N R Krishnaswamy*

**N R Krishnaswamy was initiated into the world of natural products by T R Seshadri at University of Delhi and has carried on the glorious traditions of his mentor. He has taught at Bangalore University, Calicut University and Sri Sathya Sai Institute of Higher Learning.**

**The story of carbohydrate chemistry from its embryonic stage to the present day high profile research bridging organic chemistry and the life sciences is like a fascinating travelogue through space and time. In this brief article, this intriguing field of natural products chemistry is presented with appropriate illustrations, with the hope that it will kindle further interest in the young readers to whom this is primarily addressed. We begin our journey with Emil Fischer and quickly traverse some areas of classical and modern organic chemistry. In the process we come across some familiar landmarks as well as visit a few exotic places before ending on the borders of biology. Beyond this is a region full of promise inviting further exploration.**

## Introduction

Among organic compounds the most well known, even to laymen, are the carbohydrates, produced by plants. Green leaves produce glucose using atmospheric carbon dioxide and water with the help of chlorophyll and sunlight. Several molecules of glucose are then condensed together to form cellulose, which serves as a structural material, and starch which acts as a source of food.

Glucose, sucrose, cellulose and starch are household names even if the common man may not know that glucose is a constituent of the other three, two of which are polymers! Within this group, one comes across a wide range of molecular sizes (from monomers to oligomers to polymers), and shapes. The predominant functional group is the hydroxyl, several of which occur in a carbohydrate. Another key functional group is the carbonyl group, which plays a pivotal role in the chemical behavior of carbohydrates.

**Keywords**
Carbohydrates, mutarotation, Fischer–Kiliani synthesis, cyclodextrins, end-group analysis, oligosaccharides, glycosidation reaction, glycocode and glycotherapy.

Emil Fischer and his students were responsible for elucidating the structures and stereochemistry of the monosaccharides. The synthesis of glucose achieved by them in 1890 is considered as one of the important milestones in the development of organic chemistry. This was preceded by the discovery of phenyl hydrazine by Fischer in 1875. He used this reagent to explore the chemistry of glucose and related compounds. In the course of these studies, Fischer developed the mode of molecular representation now known as the Fischer projection formula.

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With the discovery of complex oligosaccharides and polysaccharides of natural origin, the focus shifted to the biological importance of these compounds. Secrets of this aspect of the carbohydrates are being gradually revealed and active research to unravel the role of carbohydrates in living organisms is in progress. For example, it is now known that in eukaryotic organisms, oligosaccharides occurring as conjugates with proteins and lipids on cell surfaces have a key role in cellular communications.

For the elucidation of the structures of such complex oligosaccharides, classical conventional chemical methods proved inadequate. Progress in this area became possible only after instrumental methods such as GC-MS and NMR spectroscopy became more powerful and effective as a consequence of advances in these techniques. For confirmation of the structures thus deduced it also became imperative to develop synthetic methods similar to those used for the synthesis of polypeptides. In the following paragraphs, these developments beginning with the pioneering studies of Fischer and others and culminating in present day research, are briefly described

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## Classification

Carbohydrates are primarily classified according to their molecular size. Monosaccharides are monomers. The most important member of this group is glucose, which is an aldohexose as it has six carbon atoms, five hydroxyl groups (one primary and the other four secondary) and an aldehyde function at one end, as in the

Oligosaccharides are made up of two or more monosaccharide units; for example, disaccharides, such as sucrose, lactose and maltose, are hydrolysable to yield two monosaccharide units.

Fischer representation. Fructose, which is an isomer of glucose, has a keto carbonyl function and is known as a ketohexose. Monosaccharides having fewer carbon atoms are also known. For example, arabinose and ribose are aldopentoses, that is, they are C5 compounds with an aldehyde group and four hydroxyls.

Oligosaccharides are made up of two or more monosaccharide units; for example, disaccharides, such as sucrose, lactose and maltose, are hydrolysable to yield two monosaccharide units. In the case of sucrose, the monomers obtained are glucose and fructose. Raffinose, which can be isolated from molasses, is a trisaccharide. This compound on hydrolysis yields one molecule each of glucose, galactose, another aldohexose, and fructose. As already mentioned, cellulose and starch are polysaccharides, being polymeric compounds. Another example of a polysaccharide is glycogen, commonly known as animal starch.

Carbohydrates which do not conform to the general formula $C_n(H_2O)_m$ include deoxy sugars and amino sugars.

**Monosaccharides**

The optical activity exhibited by (+)-glucose was first observed by Biot in the year 1817. Two years earlier he had recorded that sucrose was optically active. However, the stereochemistry of glucose and other monosaccharides remained obscure until Fischer began his pioneering studies. The molecular formula, formation of a pentaacetate and reduction of Tollen's reagent established that glucose is a pentahydroxy aldehyde having six carbon atoms. The presence of the aldehyde group could be confirmed by oxidation with bromine water, the product being gluconic acid. Glucose cyanohydrin, on hydrolysis followed by reduction with hydriodic acid gave *n*-heptanoic acid showing that glucose is a straight-chain aldohexose. On catalytic hydrogenation over a nickel catalyst glucose yielded glucitol or sorbitol, which is 1,2,3,4,5,6-hexahydroxyhexane. However, structure (**1**) that emerged from the above mentioned reactions. could not account for all the known properties of glucose. On the basis of structure

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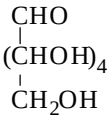
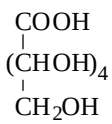
(**1**), which has only historical significance, for glucose, gluconic acid can be formulated as (**2**) and glucitol as (**3**).



**1**



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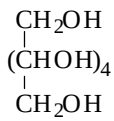
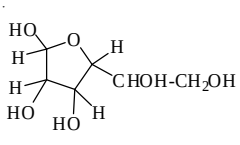


**3**

One property which could not be explained by structure (**1**) is the mutarotation exhibited by aqueous solutions of glucose. The initial specific rotation of ordinary glucose in water is $[\alpha]_D$ = +112°. However, it changes over a period of time and finally reaches the value of +52.3°. A thorough investigation of this phenomenon, discovered by Dubrunfaut, showed that all monosaccharides exhibit this property which could be attributed to the existence of two stereoisomers which are interconvertible. These were designated as $\alpha$- and $\beta$- forms. To account for this phenomenon, Tollens suggested a five-membered cyclic oxide structure (cyclic hemiacetal) (**4**) for glucose involving the aldehyde group at position 1 and the hydroxyl at position 4. However, when Tollens made this proposition in 1883, there was no experimental evidence available to support it. Only 12 years later, Tanret could provide this crucial evidence by isolating the two forms of (+)-glucose. Several years later, as a result of the studies of Haworth and others, the ring structure of (+)-glucose was corrected to a six-membered cyclic hemiacetal structure (**5**), which incorporates the correct configurations at all the chiral centres, which had earlier been determined by Fischer and his coworkers. In this Haworth projection[1] formula, the hydroxyl group at position 1, which is known as the anomeric carbon atom, is on the top in the $\beta$-form, whereas it is oriented downwards in the $\alpha$-form, as shown in **6** and **7** respectively.



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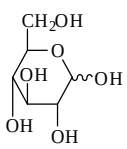

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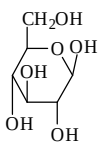
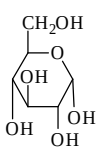


**6**



**7**

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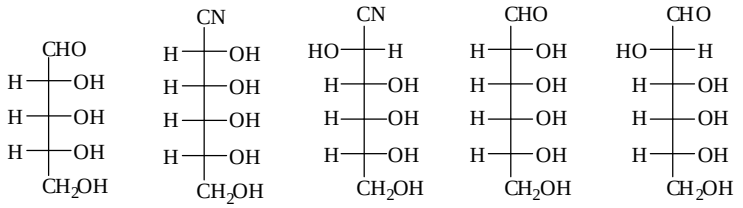
[1] **Haworth Projection:** The mode of two-dimensional representation of the cyclic structures of sugar molecules is known as the Haworth projection, and was developed by Sir Walter Norman Haworth. The method developed by him for the preparation of methyl ethers of sugars using dimethyl sulphate was an important early step in structural studies on carbohydrates.

Fischer used the Kiliani synthesis to convert an aldose into its next higher homologue.

As mentioned above, prior to this development, Fischer and his coworkers had elucidated the stereochemistry of glucose and other aldohexoses in a series of exquisitely planned and elegantly executed experiments. Fischer used the Kiliani synthesis[2] to convert an aldose into its next higher homologue. For example, if an aldopentose having the Fischer structure (**8**) is treated with HCN it will yield two isomeric cyanohydrins (**9**) and (**10**). These are separately hydrolysed and the resulting carboxylic acid lactones reduced with sodium amalgam to obtain two epimeric aldohexoses, (**11**) and (**12**).

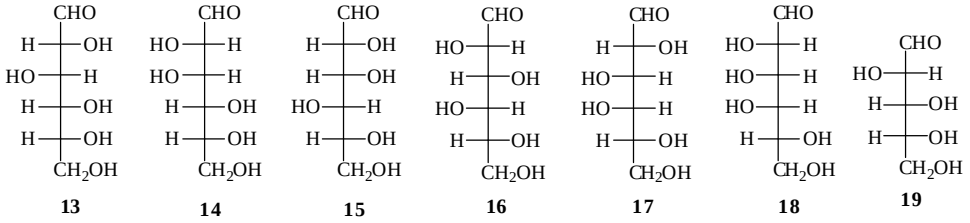



**8** **9** **10** **11** **12**

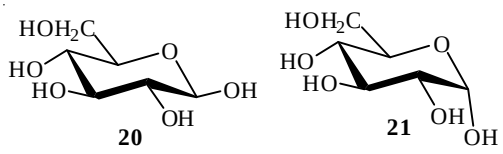
This reaction was effectively used by Fischer in his studies on glucose and its stereoisomers. Since the Fischer structure has four asymmetric carbon atoms, 16 possible configurations, representing eight pairs of enantiomers, are possible. For one set of aldohexoses, designated as D-aldohexoses, the possible structures are **11** to **18**, one of them being the structure of D-(+)-glucose. Fischer showed that by using the Kiliani synthesis the aldopentose (-)-arabinose could be converted into a mixture of (+)-glucose and (+)-mannose. Therefore, the first problem was to establish the configuration of D-(-)-arabinose and this was done using oxidation reactions and optical activity measurements. Thereby, (-)- arabinose was shown to have the structure (**19**). The structure **8** given in the previous paragraph is that of D-ribose. It follows, therefore, that (+)-glucose and (+)-mannose should be **13** and **14** or vice versa. Further experiments proved that D-(+)-glucose is indeed **13**.

Fischer showed that by using the Kiliani synthesis the aldopentose (-)-arabinose could be converted into a mixture of (+)-glucose and (+)-mannose. The first problem was to establish the configuration of D-(-)-arabinose and this was done using oxidation reactions and optical activity measurements.

[2] **Kiliani Synthesis:** This synthesis, named after Heinrich Kiliani and Emil Fischer, begins with an aldose whose cyanohydrin is converted into the corresponding aldonic lactone. The latter is finally reduced to obtain the next higher aldose. The original procedure has undergone several modifications in order to improve the yield of the final product.




The Haworth representation is also not perfect in the sense that it does not reflect the correct conformation of the six-membered cyclic hemiacetal ring. Being analogous to cyclohexane, this ring can also assume several conformations of which the chair form is the most stable. Therefore, D-(+)-$\beta$-glucose should be correctly represented as **20** and its $\alpha$-anomer as **21**.




D-Glucosamine or 2-amino-2-deoxy-D-glucopyranose (**22**) is an important member of the group classified as modified monosaccharides. Its N-acetyl derivative is the sole constituent of the polysaccharide, chitin, which occurs in the shell of the lobster, the cockroach and also in plants.

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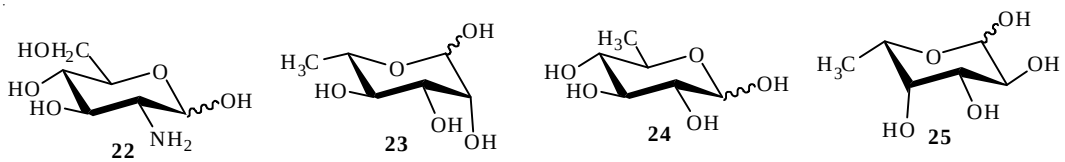
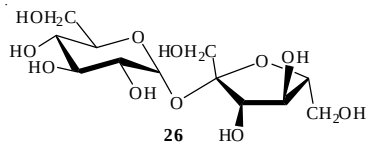
Modified monosaccharides also include deoxy sugars such as L-rhamnose (**23**), which is 6-deoxy-L-mannopyranose, quinovose (**24**) (6-deoxy-D-glucopyranose) and L-fucose (**25**), which is 6-deoxy-L-galactopyranose.



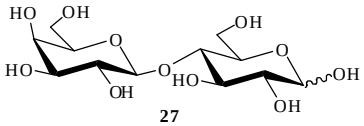
The best known disaccharide is sucrose or cane sugar. Glucose and fructose are combined through their anomeric carbon atoms that are C-1 of glucose and C-2 of fructose as shown in structure **26**.

## Disaccharides

The best known disaccharide is sucrose or cane sugar. As mentioned earlier, on hydrolysis it gives one molecule each of D-(+)-glucose and D-(-)-fructose. Since it does not reduce Tollen's reagent or react with phenylhydrazine, it is evident that it does not have a free carbonyl group. Nor does it exhibit mutarotation. Therefore, it is obvious that glucose and fructose are combined through their anomeric carbon atoms that are C-1 of glucose and C-2 of fructose as shown in structure **26**. This linkage is known as the glycoside bond. The configuration at the anomeric carbon atom of the glucose unit is $\alpha$-, whereas that at the corresponding position in the fructose part is $\beta$.




**26**

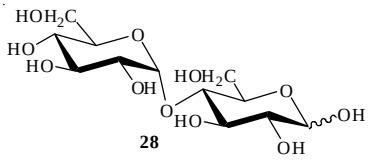



**27**

In contrast to sucrose, (+)-lactose, which is the milk sugar, reduces Tollen's reagent, exhibits mutarotation and reacts with phenylhydrazine to form an osazone derivative. On acidic or enzymatic hydrolysis (brought about by the action of emulsin which specifically cleaves $\beta$- glycosidic linkages), one molecule each of D-(+)-glucose and D-(+)-galactose are obtained. The observation that lactosazone on hydrolysis gives galactose and glucosazone shows that in lactose, the glucose unit retains its anomeric hydroxyl group. Further experiments involving methylation followed by hydrolysis show that the anomeric carbon atom (C-1) of galactose is linked through an oxide bond to C-4 of glucose as shown in **27**.

Maltose (**28**) and cellobiose (**29**) are both diglucosides, each being made of two glucose units.

Maltose (**28**) and cellobiose (**29**) are both diglucosides, each being made of two glucose units. Both are reducing sugars. In both the compounds, C-1 of one glucose unit is linked to C-4 of the other unit through an oxide bond. The only difference is the configuration of the glucosidic bond; in maltose it is $\alpha$-, whereas in cellobiose it is $\beta$. Maltose forms the structural unit of starch, while cellobiose has a similar function in cellulose.



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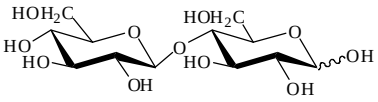

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## Higher Oligosaccharides

The antibiotic, streptomycin (**30**), can be considered as a trisaccharide. The human milk is a source of some complex oligosaccharides which possess an immunostimulising effect. One of these is the trisaccharide, L-fucosyllactose (**31**).

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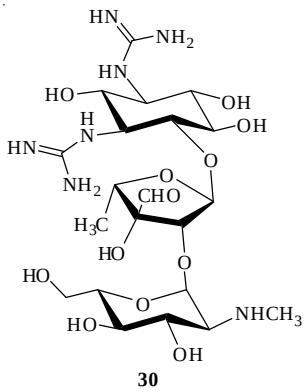

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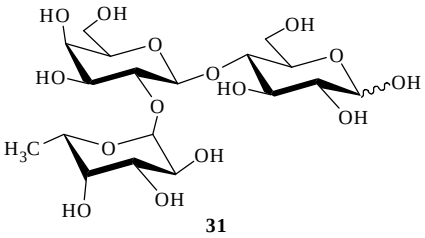

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## Cyclodextrins

These cyclic oligosaccharides are produced when starch is acted upon by amylolytic enzymes present in *Bacillus macerans* and other microorganisms. $\alpha$-Cyclodextrin is made up of six glucose units linked together by $\alpha$-glycosidic bonds. The $\beta$- and $\gamma$-forms contain 7 and 8 glucose units respectively. The exterior surface of these cyclodextrins is hydrophilic, whereas the interior space is hydrophobic. In one possible arrangement of $\beta$-cyclodextrin hendecahydrate, the hydroxyl group on C-2 of a glucose unit is involved in hydrogen bonding with the hydroxyl on C-3 of the neighbouring glucose moiety as shown in structure **32**. The partners involved in this type of intramolecular hydrogen bonding, designated as flip-flop hydrogen bonding, keep changing, resulting in a stabilized structure which is continuously in a

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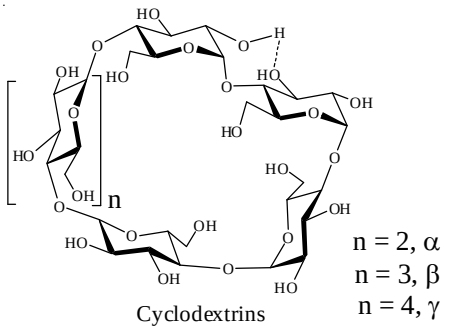
rocking mode. The interior space is large enough to accommodate a variety of other molecules to form inclusion complexes. This property has been exploited to facilitate a wide range of reactions. For example, in a Diels–Alder reaction[3] between cyclopentadiene and acrylonitrile, the addition of $\beta$-cyclodextrin increases the rate of the reaction by several orders compared to the rate in the usual organic solvents. This is the consequence of the cyclodextrin molecule gathering the two reactants inside its cavity and of the rocking motion mentioned above which facilitate interactions between the diene and the dienophile. This reaction is not catalysed by $\alpha$-cyclodextrin which shows that the size of the internal cavity is a crucial factor.




Cyclodextrins

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## Polysaccharides

Among polysaccharides the best known are cellulose, starch and chitin. As mentioned earlier, the monomeric unit in both cellulose and starch is D-glucose but the glucosidic bond in cellulose is $\beta$ and in starch it is $\alpha$. Apart from this important difference, cellulose and starch differ from each other in several other respects. In cellulose, where the disaccharide unit is cellobiose, several molecules of the latter combine in a linear manner to form the polymer. Further, parallel strands of the polysaccharide thus formed link together by hydrogen bonding. The resulting rope-like structure makes cellulose a strong structural material.

The best known polysaccharides are cellulose, starch and chitin. The monomeric unit in both cellulose and starch is D-glucose but the glucosidic bond in cellulose is $\beta$ and in starch it is $\alpha$.

Starch, on the other hand, is not a homogeneous substance; it can be separated into the water-soluble amylose and water-insoluble

[3] **The Diels–Alder Reaction:** Among the many name reactions used for the synthesis of a wide variety of naturally occurring organic compounds, the Diels–Alder reaction occupies a prime place by virtue of being a regio- and stereo-specific reaction. It was first developed in 1928 by the German chemists, Otto Diels and Kurt Alder. It is a cycloadditon reaction involving a diene and a dienophile and comes under the category of pericylic reactions. Orbital symmetry rules have been applied to elucidate the mechanism of this reaction.

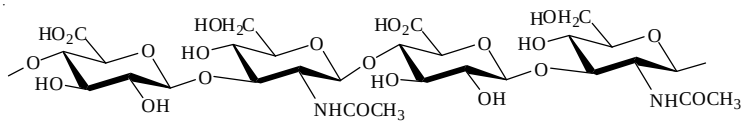
amylopectin. Amylose is a polymer of maltose. On the basis of end group analysis and physical methods it has been estimated that 1000 to 4000 glucose molecules are linked together through 1,4-$\alpha$-glucosidic bonds to form an amylose molecule. Physical methods indicate a higher molecular weight than that given by chemical end group analysis showing that in the latter process some amount of degradation is occurring. The fully formed polymeric structure assumes a spiral, spring-like form in which iodine molecules, for example, get entrapped and form a blue-coloured complex.

Starch, on the other hand, is not a homogeneous substance. On the basis of end group analysis and physical methods it has been estimated that 1000 to 4000 glucose molecules are linked together through 1,4-$\alpha$-glucosidic bonds to form an amylose molecule.

Amylopectin is also made up of maltose units, but unlike amylose, there are several cross linking bonds between these units, making its overall structure much more complex. The resulting highly branched structure is responsible for its insolubility in water.

Chitin is a polymer of N-acetylglucosamine. Its structure is very similar to that of cellulose. Like the latter, it is resistant to solvents. It can however be broken down by the enzyme chitinase which occurs in the intestinal tract of snails.

N-acetylglucosamine is a constituent of the biologically important polysaccharide, hyaluronic acid which functions as a lubricant and shock absorber in animal joints. The other monosaccharide unit in this polysaccharide is $\beta$-D-glucuronic acid. The repeat unit in this is a disaccharide acid in which the anomeric carbon of a glucuronic acid unit is glycosidically linked to position 3 of N-acetylglucosamine, the anomeric carbon atom of which, in turn, is linked to position 4 of the neighbouring glucuronic acid moiety, as shown in the partial structure **33**.




**33**

Chitin is a polymer of N-acetylglucosamine. Its structure is very similar to that of cellulose.

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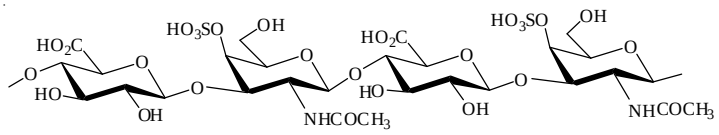
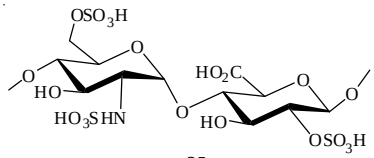
compounds are chemically related to hyaluronic acid with the following differences. In place of N-acetylglucosamine, N-acetylgalactosamine (2-acetamido-2-deoxy-D-galactose) is one of the monosaccharide units in these compounds. Further, the hydroxyl at position 4 of each N-acetylgalactosamine moiety is esterified with sulphuric acid (see partial structure **34**). The anticoagulant, heparin (**35**), which occurs in the liver, heart and other tissues, is also chemically related to the chondroitin sulphates. Its constituents are glucosamine and glucuronic acid in the ratio 1:1. Within each repeat disaccharide unit, there are two O-sulphate and one N-sulphate groups as shown in structure **35**.




**34**




**35**

Using a 750 MHz NMR instrument, with multiple (as many as 32) scans, the structure of an oligosaccharide consisting of 22 monosaccharide units, isolated from *Salmonella enterica* ssp Typhimurium 1135 LPS could be completely elucidated using just 2 mg of the compound.

## Determination of Structures of Complex Oligo- and Polysaccharides

With the introduction of methylation analysis in the 1970s, it became possible to determine the structures of complex oligosaccharides isolated from bacterial sources. The value of this technique was enhanced when advanced physical techniques were used in tandem. For example, using a 750 MHz NMR instrument, with multiple (as many as 32) scans, the structure of an oligosaccharide consisting of 22 monosaccharide units, isolated from *Salmonella enterica* ssp Typhimurium 1135 LPS could be completely elucidated using just 2 mg of the compound. Even less quantity (0.14 mg) was sufficient to record a 550 MHz NMR spectrum using a nanoprobe. Both $^{1}H$ and $^{13}C$ NMR spectral data are extensively used in these studies.

## Synthesis of Oligosaccharides

As already mentioned, the variables in a oligosaccharide structure are the number of monomeric units, the points of attachment of the different components, the configuration of the glycosidic linkages and also the ring size (pyranose or furanose) of the monomeric units. All these factors together make the synthesis of an oligosaccharide a challenging task, demanding a high degree of regio- as well as stereoselectivity. The strategy used is similar to that used in polypeptide synthesis. For example, the use of selective protective groups plays a vital role. As in peptide synthesis, the current trend is to assemble an oligosaccharide sequence on a solid support. A decade ago, the first automated solid phase synthesis of an oligosaccharide was effected. Since then, several innovations have been introduced with the result that a complex nonasaccharide antigen found on tumour cells could be synthesised within a day.

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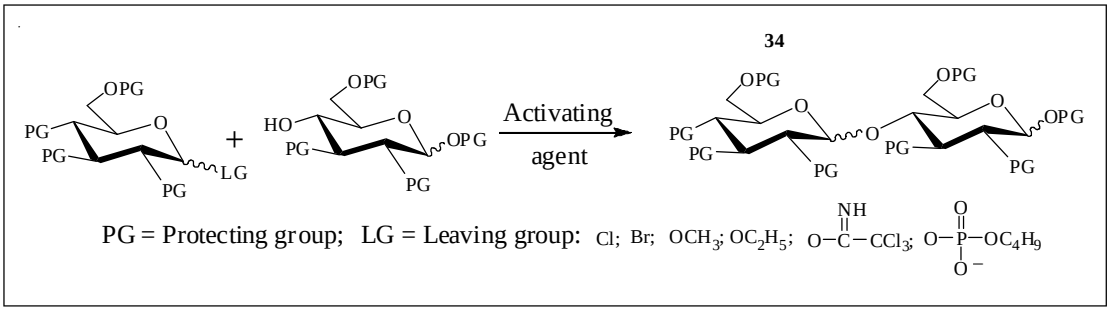
Whatever the strategy, the most important single step involved in the synthesis of an oligosaccharide is the glycosylation reaction, in which two sugar units are linked through an acetal bond. The key to success in this reaction is the nature of the leaving group on the anomeric carbon atom of a sugar molecule. In the classical era, the leaving groups most widely used were bromide, chloride and methoxide groups. More effective leaving groups are the thioether, the phosphate and the trichloroacetimidate groups. The reagent used for introducing the last mentioned group is trichloroacetonitrile. In *Scheme* 1, a regiospecific formation of a diglucoside in which the anomeric carbon atom of one glucose

***Scheme 1. Preparation of disaccharide.***



moiety is linked to position 3 of the other is shown, where PGs are protective groups and LGs are leaving groups. A wide range of protective groups have been used for selective protection of the various hydroxyl groups. These include the acetyl, benzyl, allyl and 4-nitrobenzoyl groups, among others. Several steps, usually not less than five, are needed to prepare an appropriately protected monosaccharide. A number of these derivatives are now available. One solid support commonly used in oligosaccharide synthesis is polyethyleneglycol $\omega$-monomethyl ether (MPEG) attached to $\alpha,\alpha$'-dioxyxylyl diether (DOX).

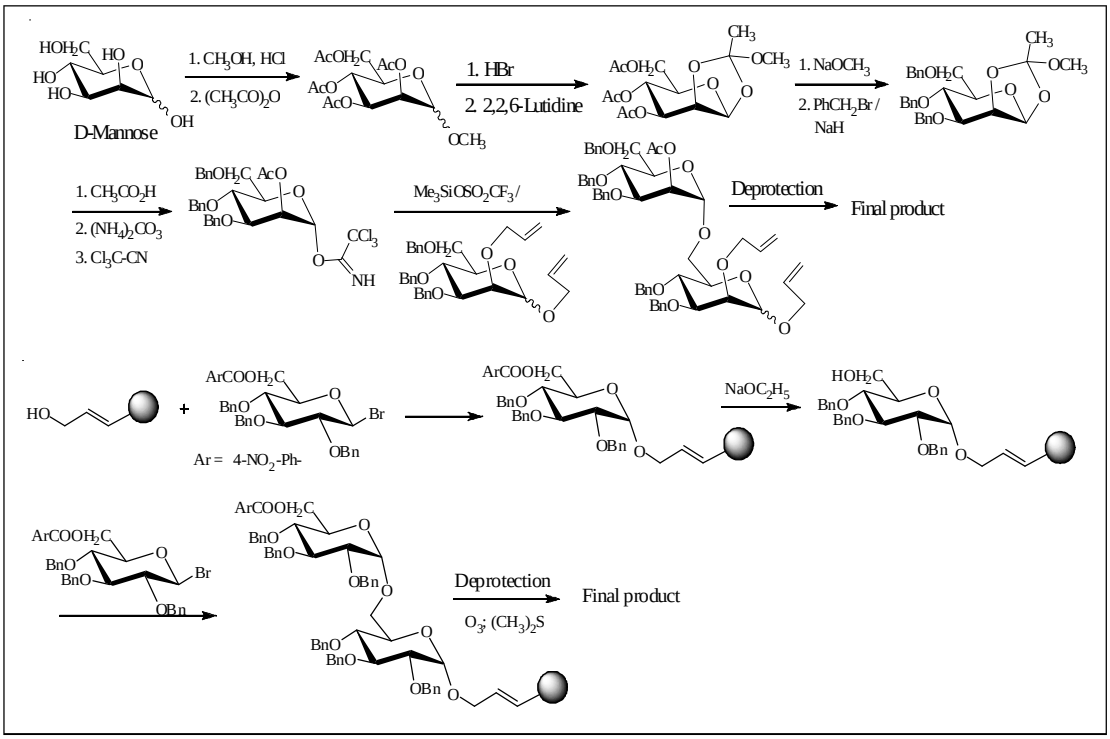
As an alternative to traditional organic synthetic methods, specific enzymes have been used to build oligosaccharides. These enzymes, known as glycosyl transferases, act on nucleotide diphospho sugars in aqueous media to produce complex oligosaccharides without the need for any protective functionalities.

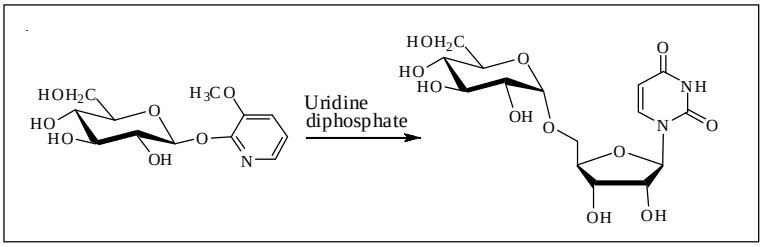
One-pot methods[4] have been developed, for example using thioglycosides as building blocks to prepare oligosaccharide chains from the non-reducing end to the reducing end. Using this technique a library of linear as well as branched oligosaccharides has been prepared.

A synthesis of a 1,6-linked di-D-glucoside as well as that of a 1,6-linked di-D-mannoside, involving some of the principles mentioned above, are given in *Scheme* 2. These two examples are chosen to illustrate a solid-phase synthesis and a solution-phase synthesis using different protecting and leaving groups.

Before leaving this topic of synthesis of oligosaccharides and the glycosylation reaction, it is pertinent to mention about the development of a glycosylation reaction using unprotected glycosyl donors. This reaction discovered by Hanessian and his coworkers involves the use of specially designed anomeric leaving groups. One example is given in *Scheme* 3.

[4] **One-Pot Method:** The strategy of bringing about a multi-step synthesis of an organic compound in one reaction vessel is known as one-pot synthesis. By doing so, time-consuming separations and lengthy work-up procedures are eliminated and the final desired product is obtained in quicker time and better yields as compared to conventional procedures. For example, 7-hydroxycoumarin-3-carboxylic acid has been prepared in an aqueous medium from 2,4-dihydroxybenzaldehyde and malononitrile (see F Fringuelli *et al*, *J.Chem*.Ed., Vol.81, p.874, 2004).



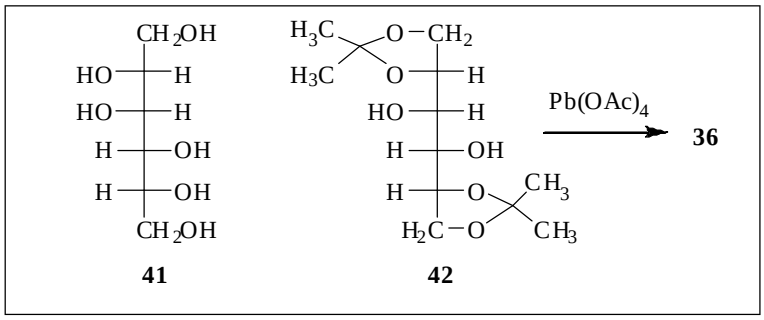




***Scheme 2 (top). Preparation of 1,6-linked disaccharides.***

***Scheme 3 (bottom). Glycosylation using a special leaving group.***

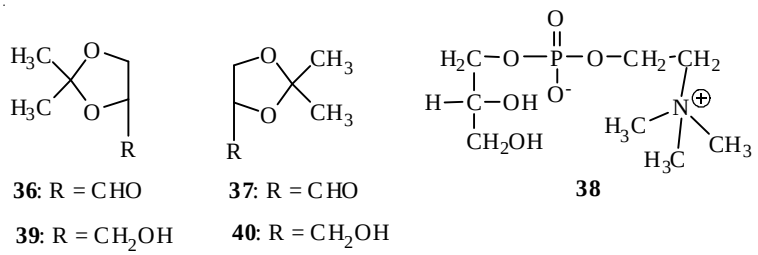
## Carbohydrates as Chirons

By virtue of possessing several asymmetric centres with defined configurations, sugar derivatives can be used as chirons in the asymmetric synthesis of a variety of natural products. We shall describe here only a couple of illustrative examples. A versatile C-3 chiron is 2,3-O-isopropylideneglyceraldehyde. Both enantiomers of this compound, namely, the D- or (*R*)-isomer (**36**) and the L- or (*S*)-form (**37**) have been used in the synthesis of a wide range of biologically important compounds. For example, **36** has been used in the synthesis of L-$\alpha$-glycerylphosphorylcholine



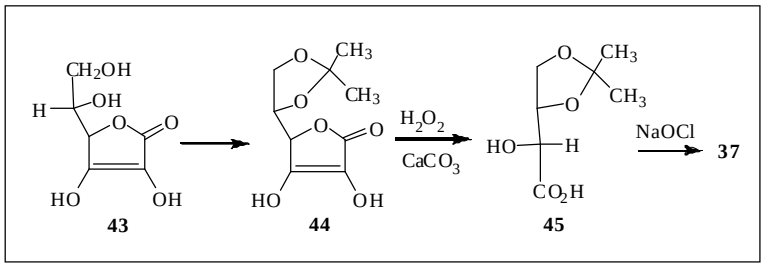

***Scheme 4. Preparation of (R)-2,3-O-isopropylidene-glyceraldehyde.***

(**38**). Reduction of **36** and **37** using Raney nickel or sodium borohydride yields 1,2-O-isopropylidene L- or (*S*)-glycerol (**39**) and its enantiomer (**40**), which are also widely used as chiral building blocks.

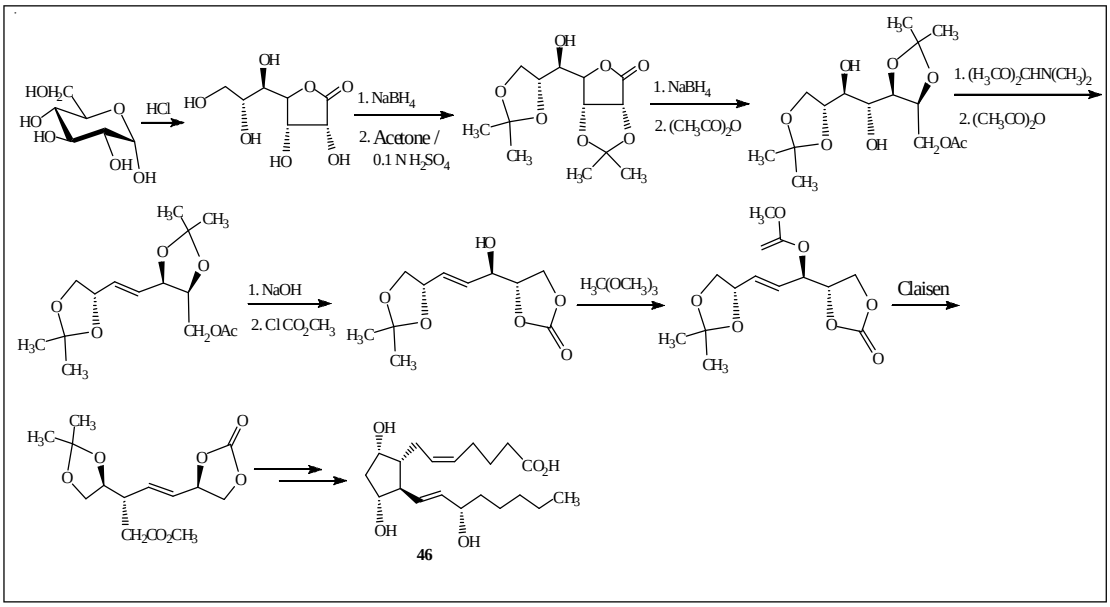



D-Mannitol (**41**) is the starting material for the preparation of **36**. It is first converted into its diacetonide (**42**) which is then oxidatively cleaved with lead tetraacetate to obtain **36** as outlined in *Scheme* 4. Other reagents, such as sodium periodate, bismuth derivatives and meta-iodoxybenzoate, have also been used for the second step in this reaction sequence.

For the preparation of L- or (*S*)-2,3-O-isopropylidene-glyceraldehyde (**37**) and the corresponding alcohol **40**, L-ascorbic acid (**43**) is used as a starting material as shown in *Scheme* 5. The 5,6-O-




***Scheme 5. Preparation of (S)-2,3-O-isopropylidene-glyceraldehyde.***



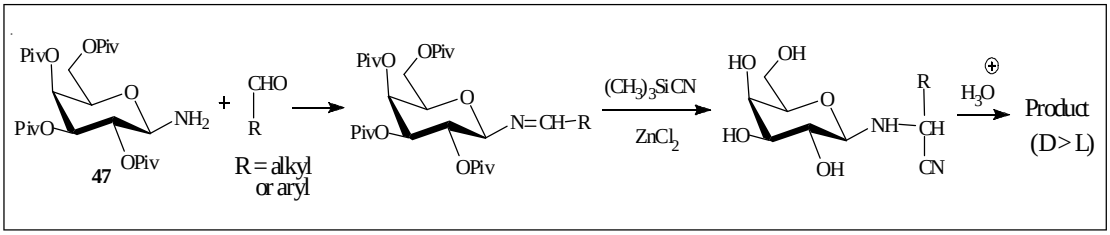

***Scheme 6. Synthesis of prostaglandin F2α.***

isopropylidene derivative **44** of **43** is oxidised with hydrogen peroxide and the resulting **45** is treated with sodium hypochlorite to get **37**.

In an ingenious synthesis of prostaglandin F2$\alpha$ (**46**), Stork and co-workers incorporated some of the stereochemical features of $\alpha$-D-glucose in a part of the final product as shown in *Scheme* 6. As can be seen, a section of the lower part of **46**, with the double bond between 13 and 14 positions having the *E*-configuration, is derived from $\alpha$-D-glucose.

Sugar derivatives, such as, for example, 2,3,4,6-tetrapivaloyl-D-galactosamine (**47**) have been used as chiral templates for the stereoselective preparation of D-$\alpha$-amino acids by the Strecker synthesis[5] as outlined in *Scheme* 7.

***Scheme 7. Preparation of D-α-amino acids by the Strecker synthesis.***

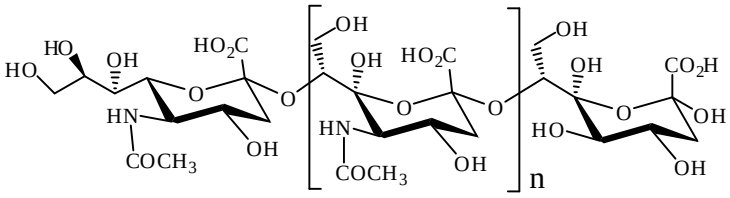


## Biological Aspects

With the elucidation of the structures of several complex oligosaccharides and the availability of synthetic analogues, the focus on carbohydrates has shifted to their biological activities and potential as therapeutic agents. A decade ago, Oxford University established the Oxford Glycochemistry Centre (OGC), wherein active research is in progress on glycocode[6] and glycotherapeutics[7], along with other aspects.

Glycoconjugates, such as carbohydrate-protein and carbohydrate-lipid complexes, occur both in soluble form and as sticky nano-dimensional layers on cell surfaces. The latter, known as glycocalyx, are associated with several important biological phenomena like immune response, intracellular recognition, cellular adhesion, cell growth regulation and inflammation.

One important area of ongoing research in this field is concerned with tumour-associated carbohydrate antigens, or TACAs, as for example p-sial 2 (**48**). These oligosaccharides can serve as molecular markers on cancer cell surfaces and can lead to the development of newer and more effective anti-cancer agents.



**48**

One of the earliest carbohydrate-based drugs to be used in clinical practice was the anticoagulant, heparin, which was mentioned in an earlier section of this article. Since its introduction in the 1940s, several modifications have been effected in order to produce an anticaoagulant without the side effects of heparin. As a result of these studies, in the early 1980s a low-molecular weight heparin became available. This compound produced by chemical and enzymatic fragmentation of the original heparin has longer half-life, greater bioavailability and fewer side effects.

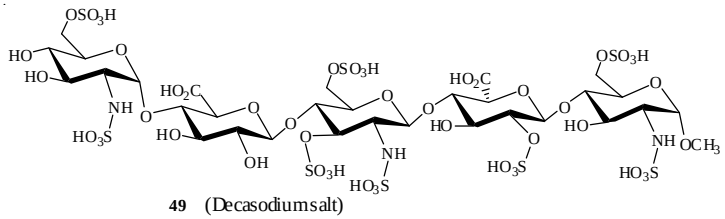
Sugar derivatives, such as, for example, 2,3,4,6-tetrapivaloyl-D-galactosamine (**47**) have been used as chiral templates for the stereoselective preparation of D-$\alpha$-amino acids by the Strecker synthesis.

[5] **Strecker Synthesis:** This method of synthesis of $\alpha$-amino acids was first devised by Adolph Strecker, a student of Justus Liebig in the year 1850. In this method, an aldehyde is made to react with potassium cyanide and ammonium chloride. The resulting amino nitrile is subsequently hydrolysed to obtain the desired $\alpha$-amino acid. A wide range of aldehydes have been used in this reaction. Several procedural modifications are now available including one-pot methods (see, for example, P Fontaine *et al.*, *Org.Lett.*, Vol.10, pp.1509–1512, 2008).

[6] Glycocode refers to the communication mediated by carbohydrates which plays a vital role in cell biological processes.

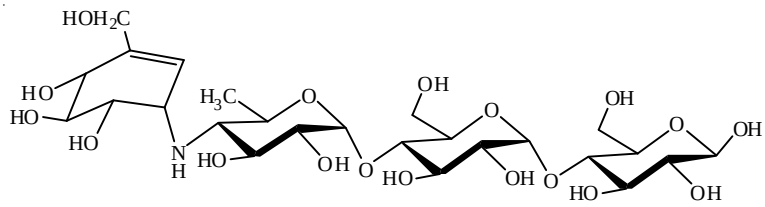
[7] Glycotherapeutics deals with the treatment of a wide range of pathological conditions, such as cancer, bacterial and viral infections, diabetics, etc., using carbohydrate-based drugs.

Later, synthetic analogues were prepared and based on structure-activity studies, a pentasaccharide, named Fondaparinux (**49**) was introduced as an anticoagulant drug. This drug was first marketed in 2002 under the trade name Arixtra.



**49** (Decasodium salt)

Based on the fact that carbohydrate foods have to be enzymatically broken down in the intestinal tract before they can be utilized for nourishment, inhibitors of $\alpha$-glucosidases and amylases have been studied as potential candidates for the treatment of diabetes. One such compound, surprisingly, is a pseudo oligosaccharide. This compound, acarbose (**50**) is of microbial origin and is produced by fermentation of *Actinoplanes* species.



**50**

Two factors were responsible for carbohydrates to be explored as potential vaccines against a number of diseases. (1) The sugars present on the cell surfaces of parasites are quite distinct from those occurring in their hosts. (2) Unlike proteins, carbohydrates are evolutionarily more stable and therefore the sugars of a host are less likely to be changed by parasitic action. As a consequence, several carbohydrate-based vaccines have been developed in recent years to combat a wide range of bacterial, viral and parasitic infections including meningitis, HIV and malaria. The structure of a malaria vaccine contains a carbohydrate derivative of structure **51**.

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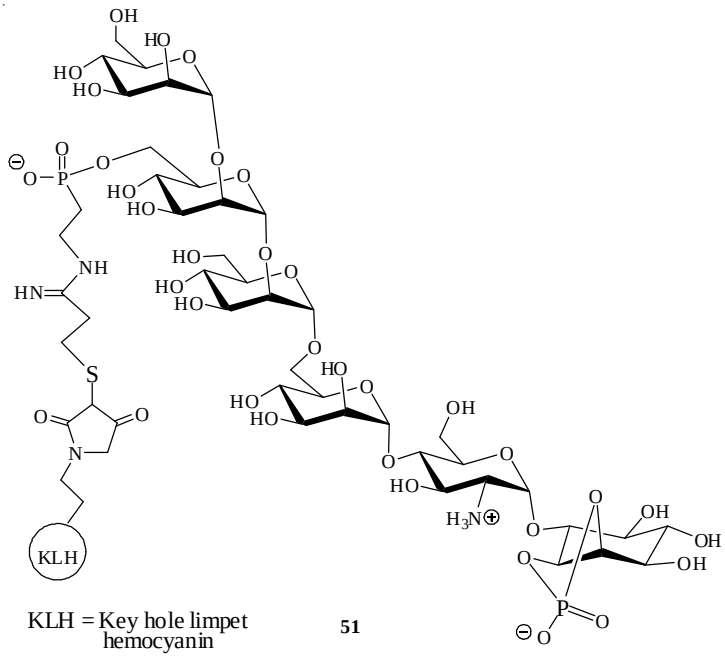
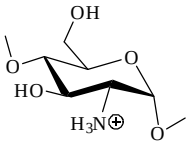



KLH = Key hole limpet hemocyanin

**51**

Please note the fifth ring from the top:



## Conclusions

As a postscript, a quotation from the review by P H Seeberger and D B Werz sums up the future of carbohydrates, "We are still beginning to understand the importance of sugars in our lives beyond pasta, cake and chocolate. There is mounting evidence that the future of medicine will be a sweet one".

Thus, a branch of science initially explored by Emil Fischer has not only traversed the full length of chemistry but has crossed over to the other side forming a firm bridge between the physical sciences and the life sciences.

## Suggested Reading

**[1]** **E A Davidson, *Carbohydrate Chemistry*, Holt, Rinehart and Winston, New York, 1967.**

[2] J O Duus, P M St Hillaire, M Meldal and K Bock, Carbohydrate Chemistry. Synthetic and structural challenges towards the end of the 20th century, *Pure Appl. Chem*., Vol.71, pp.756–765, 1999,

[3] P H Seeberger and D B Werz, Automated synthesis of oligosaccharides as a basis for drug discovery, *Nature reviews/Drug Discovery*, Vol.4, pp.751–763, 2005.

[4] T K Lindhorst, *Essentials of Carbohydrate Chemistry and Biochemistry*, 3rd Edition, Wiley-VCH, Weinjheim, 2007.

[5] A P Rauter and T D Lindhorst, Eds., *Carbohydrate Chemistry*: *Chemical and Biological Approaches*, Royal Society of Chemistry, 2010

[6] M Flice, J M Guisan and J M Palomo, Recent Trends in regioselective protection and deprotection of monosaccharides, *Current Organic Chemistry*, Vol.14, pp.516–532, 2010.

[7] The Web site of Oxford Glycochemistry Centre.

[8] Web site of the Hanessian group, Department of Chemistry, University of Montreal, Canada.

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## A Detailed Description of the Cover Page Figure

Collage of 3D structural representation of a few biomolecules with the collection potrayed as a garden. All the depicted 3D structures have been solved using X-ray crystallography and deposited in the protein data bank and are represented as cartoons. The main tree-like structure is that of a bacterial hemolysin. The glycyl residues in this protein are represented in space-fill model in red to mimic the appearance of fruits. The butterfly-like structure in the right is that of human cystatin which is a dimeric protein. The yellow coloured flower-like structure in the right bottom is that of an isomerase from *E.coli*. The brown coloured flower-like structure in the left is that of $\gamma$-B-crystallin from bovine eye lens. This folds into two domains of similar flower-like 3D structure. The circular structure in the left bottom is that of β-cyclodextrin, a carbohydrate. All the cartoon representations of 3D structures have been generated using the pyMOL software.

This figure was created by Ms. G Sudha, Molecular Biophysics Unit, Indian Institute of Science, Bangalore.