Handbook of Chemical Glycosylation

Advances in Stereoselectivity and Therapeutic Relevance

Edited by
Alexei V. Demchenko

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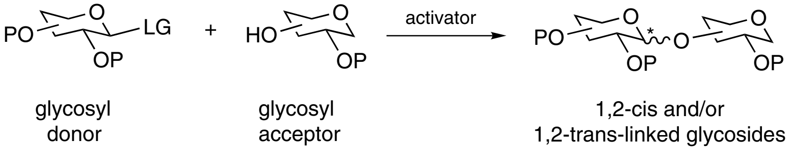
Preface

Carbohydrates are the most abundant biomolecules on Earth. Although information about these fascinating natural compounds is not yet complete, we have already learned about some crucial aspects of the carbohydrate involvement in damaging cellular processes such as bacterial and viral infections, development and growth of tumors, metastasis, septic shock that are directly associated with deadly diseases of the twenty-first century, such as AIDS, cancer, meningitis and septicemia. The tremendous medicinal potential of glycostructures has already been acknowledged by the development of synthetic carbohydrate-based vaccines and therapeutics. The elucidation of the mechanisms of carbohydrate involvement in disease progression would be further improved if we could rely on the detailed knowledge of the structure, conformation and properties of the carbohydrate molecules. Therefore, the development of effective methods for the isolation and synthesis of complex carbohydrates has become critical for the field of glycosciences. Although significant improvements of the glycoside and oligosaccharide synthesis have already emerged, a variety of synthetic targets containing challenging glycosidic linkages cannot yet be directly accessed.

A vast majority of biologically and therapeutically active carbohydrates exist as polysaccharides (cellulose, chitin, starch, glycogen) or complex glycoconjugates (glycolipids, glycopeptides, glycoproteins) in which monosaccharide units are joined via glycosidic bonds. This linkage is formed by a glycosylation reaction, most commonly a promoter-assisted nucleophilic displacement of the leaving group (LG) of the glycosyl donor with the hydroxyl moiety of the glycosyl acceptor. Other functional groups on both the donor and the acceptor are temporarily masked with protecting groups (P). These reactions are most commonly performed in the presence of an activator: promoter or catalyst. As the new glycosidic linkage creates a chirality center, particular care has to be taken with regard to the stereoselectivity. Although in the natural environment specificity and selectivity of an enzyme ensure the stereoselectivity of glycosylation, synthesis of synthetic carbohydrate faces a major challenge in comparison to the synthesis of other natural biopolymers, that is proteins and nucleic acids.



Although mechanistic studies of the glycosylation reaction are scarce, certain conventions have already been established. Pioneering mechanistic work of Lemieux was enriched by recent studies by Bols, Boons, Crich, Gin, Kochetkov, Schmidt, Whitfield and others. 1,2-*trans* Glycosides are often stereoselectively obtained with the assistance of the 2-acyl neighboring participating group. In case of ether-type nonparticipating substituents, the glycosylation proceeds with poorer stereocontrol that results in mixtures of diastereomers, which makes the synthesis of 1,2-*cis* glycosides a notable challenge.

Since the first attempts at the turn of the twentieth century, enormous progress has been made in the area of the chemical *O*-glycoside synthesis. However, it is only in the past two–three decades that the scientific world has witnessed a dramatic improvement in the methods used for glycosylation. Recently, an abundance of glycosyl donors that can be synthesized under mild reaction conditions and that are sufficiently stable toward purification, modification and storage have been developed. Convergent synthetic strategies enabling convenient and expeditious assembly of oligosaccharides from properly protected building blocks with the minimum synthetic steps have also become available.

As it stands, many of the recent developments in the area of chemical glycosylation still remain compromised when applied to the stereoselective synthesis of difficult glycosidic linkages. These special cases include the synthesis of 1,2-*cis* glycosides, especially β-mannosides and *cis*-furanosides, 2-amino-2-deoxyglycosides, 2-deoxyglycosides and α-sialosides. In spite of the considerable progress and the extensive effort in this field, no universal method for the synthesis of targets containing these types of linkages has yet emerged. Therefore, these difficult cases will be discussed individually.

This book summarizes the recent advances in the area of chemical glycosylation and provides updated information regarding the current standing in the field of synthetic carbohydrate chemistry. An expansive array of methods and strategies available to a modern synthetic carbohydrate chemist is discussed. The first chapter (Chapter 1) discusses major principles of chemical glycosylation, reaction mechanisms, survey methods for glycosylation and factors influencing the reaction outcome, as well as describes the strategies for expeditious synthesis of oligosaccharide. Each subsequent chapter discusses a certain class of glycosyl donors. Methodologies developed to date are classified and discussed based on the type of the anomeric leaving group: halogens (Chapter 2), oxygen-based derivatives (Chapter 3) and sulfur/selenium-based derivatives (Chapter 4). Bicyclic compounds, 1,2-dehydro derivatives, miscellaneous glycosyl donors and indirect synthetic methods are discussed in Chapter 5. Each chapter will discuss the following aspects of a particular methodology or approach, wherever it is applicable:

(1) Introduction (relevant to this class of glycosyl donors/methods)
(2) Synthesis of glycosyl donor
(3) Glycosylation (major activators/promoters, particulars of the reaction mechanism, examples of both 1,2-*cis* and 1,2-*trans* glycosylations)
(4) Application to target/total synthesis (oligosaccharides, glycoconjugates, natural products)
(5) Special topics (synthesis of β-mannosides, furanosides, sialosides, glycosides of aminosugars and deoxysugars, if applicable)
(6) Conclusions and future directions
(7) Typical experimental procedures
(8) References.

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January, 2008

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1
General Aspects of the Glycosidic Bond Formation

Alexei V. Demchenko

1.1
Introduction

Since the first attempts at the turn of the twentieth century, enormous progress has been made in the area of the chemical synthesis of *O*-glycosides. However, it was only in the past two decades that the scientific world had witnessed a dramatic improvement the methods used for chemical glycosylation. The development of new classes of glycosyl donors has not only allowed accessing novel types of glycosidic linkages but also led to the discovery of rapid and convergent strategies for expeditious oligosaccharide synthesis. This chapter summarizes major principles of the glycosidic bond formation and strategies to obtain certain classes of compounds, ranging from glycosides of uncommon sugars to complex oligosaccharide sequences.

1.2
Major Types of *O*-Glycosidic Linkages

There are two major types of *O*-glycosides, which are, depending on nomenclature, most commonly defined as α- and β-, or 1,2-*cis* and 1,2-*trans* glycosides. The 1,2-*cis* glycosyl residues, α-glycosides for D-glucose, D-galactose, L-fucose, D-xylose or β-glycosides for D-mannose, L-arabinose, as well as their 1,2-*trans* counterparts (β-glycosides for D-glucose, D-galactose, α-glycosides for D-mannose, etc.), are equally important components in a variety of natural compounds. Representative examples of common glycosides are shown in Figure 1.1. Some other types of glycosides, in particular 2-deoxyglycosides and sialosides, can be defined neither as 1,2-*cis* nor as 1,2-*trans* derivatives, yet are important targets because of their common occurrence as components of many classes of natural glycostructures.

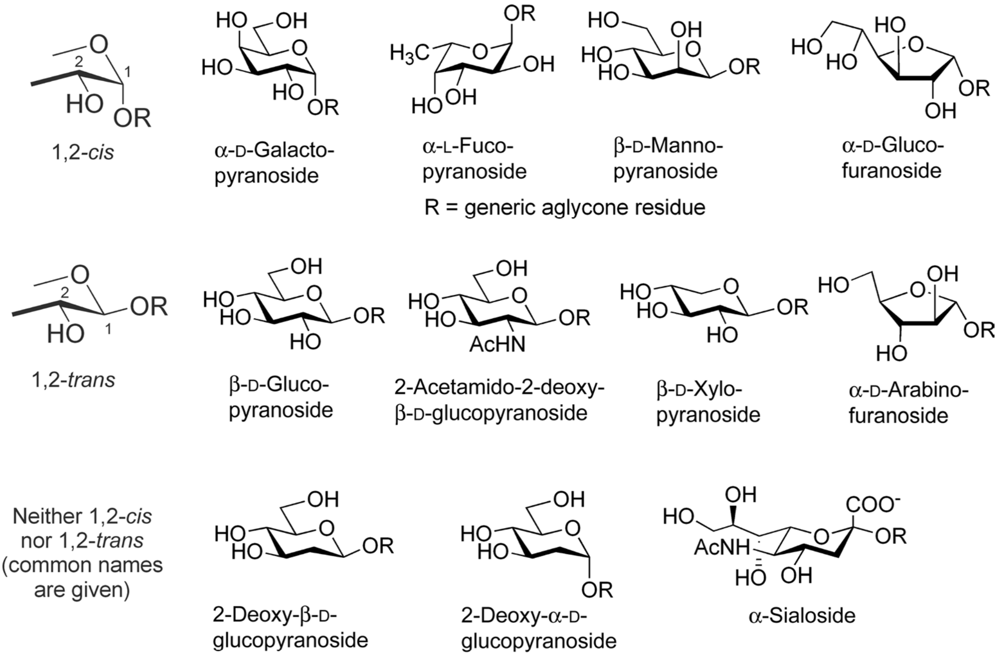


Figure 1.1 Common examples of *O*-glycosides.

1.3
Historical Development: Classes of Glycosyl Donors

The first reactions performed by Michael (synthesis of aryl glycosides from glycosyl halides) [1] and Fischer (synthesis of alkyl glycosides from hemiacetals) [2] at the end of the nineteenth century showed the complexity of the glycosylation process. The discovery of the first controlled, general glycosylation procedure involving the nucleophilic displacement of chlorine or bromine at the anomeric center is credited to Koenigs and Knorr [3]. The glycosylations were performed in the presence of Ag_2CO_3, which primarily acted as an acid (HCl or HBr) scavenger. At that early stage, glycosylations of poorly nucleophilic acceptors such as sugar hydroxyls were sluggish and inefficient; hence, even the synthesis of disaccharides represented a notable challenge. The first attempts to solve this problem gave rise to the development of new catalytic systems that were thought to be actively involved in the glycosylation process [4]. Thus, Zemplen and Gerecs [5] and, subsequently, Helferich and Wedermeyer [6] assumed that the complexation of the anomeric bromides or chlorides with more reactive, heavy-metal-based catalysts would significantly improve their leaving-group ability. This approach that has become a valuable expansion of the classic Koenigs–Knorr method made it possible to replace Ag_2CO_3 or Ag_2O by more active mercury(II) salt catalysts. The early attempts to improve the glycosylation process have revealed the necessity to find a delicate balance between the reactivity and stereoselectivity [7,8]. Indeed, it was noted that faster reactions often result in a decreased stereoselectivity. At around the same time, the first attempts to involve other classes of anomeric leaving groups (LGs) resulted in the investigation of peracetates as glycosyl donors [9].

Seminal work of Lemieux [10] and Fletcher and coworkers [11,12] has led to the appreciation that the reactivity of the glycosyl halides and the stereoselectivity of glycosylation are directly correlated to the nature of the protecting groups, especially at the neighboring C-2 position. From early days, it has been acknowledged that peracylated halides often allow stereoselective formation of 1,2-*trans* glycosides. Later, this phenomenon was rationalized by the so-called participatory effect of the neighboring acyl substituent at C-2. Although occasionally substantial amounts of 1,2-*cis* glycosides were obtained even with 2-acylated glycosyl donors, the purposeful 1,2-*cis* glycosylations were best achieved with a nonparticipating ether group at C-2, such as methyl or benzyl. Further search for suitable promoters for the activation of glycosyl halides led to the discovery of Ag-silicate that proved to be very efficient for direct β-mannosylation, as these reactions often proceed via a concerted S_N2 mechanism [13,14].

For many decades classic methods, in which anomeric bromides, chlorides, acetates or hemiacetals were used as glycosyl donors, had been the only procedure for the synthesis of a variety of synthetic targets ranging from simple glycosides to relatively complex oligosaccharides (Figure 1.2). Deeper understanding of the reaction mechanism, driving forces and principles of glycosylation have stimulated the development of other methods for glycosylation, with the main effort focusing on the development of new anomeric leaving groups [15,16]. During the 1970s to early 1980s, a few new classes of glycosyl donors were developed. The following compounds are only the most representative examples of the first wave of the leaving-group development: thioglycosides by Ferrier *et al.* [17], Nicolaou *et al.* [18], Garegg *et al.* [19] and others [20]; cyanoethylidene and orthoester derivatives by Kochetkov and coworkers [21,22]; *O*-imidates by Sinay and coworkers [23] and Schmidt and Michel [24]; thioimidates including *S*-benzothiazolyl derivatives by Mukaiyama *et al.* [25]; thiopyridyl derivatives by Hanessian *et al.* [26] and Woodward *et al.* [27] and glycosyl fluorides by Mukaiyama *et al.* [28] (Figure 1.2). Many glycosyl donors introduced during that period gave rise to excellent complimentary glycosylation methodologies. Arguably, trichloroacetimidates [29,30], thioglycosides [31–33] and fluorides [34,35] have become the most common glycosyl donors nowadays.

A new wave of methods arose in the end of the 1980s, among which were glycosyl donors such as glycosyl acyl/carbonates [36–38], thiocyanates [39], diazirines [40], xanthates [41], glycals [42,43], phosphites [44,45], sulfoxides [46], sulfones [47], selenium glycosides [48], alkenyl glycosides [49–51] and heteroaryl glycosides [52] (Figure 1.2). These developments were followed by a variety of more recent methodologies and improvements, among which are glycosyl iodides [53], phosphates [54], Te-glycosides [55], sulfonylcarbamates [56], disulfides [57], 2-(hydroxycarbonyl) benzyl glycosides [58] and novel thio- [59,60] and *O*-imidates [61,62] (Figure 1.2). In

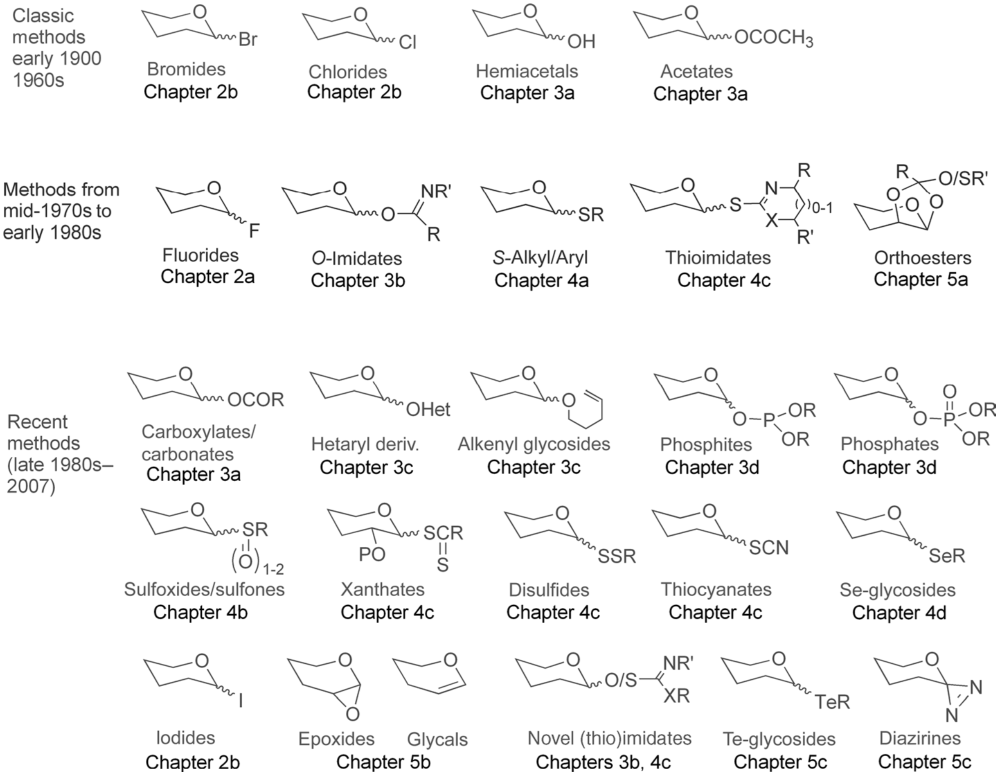
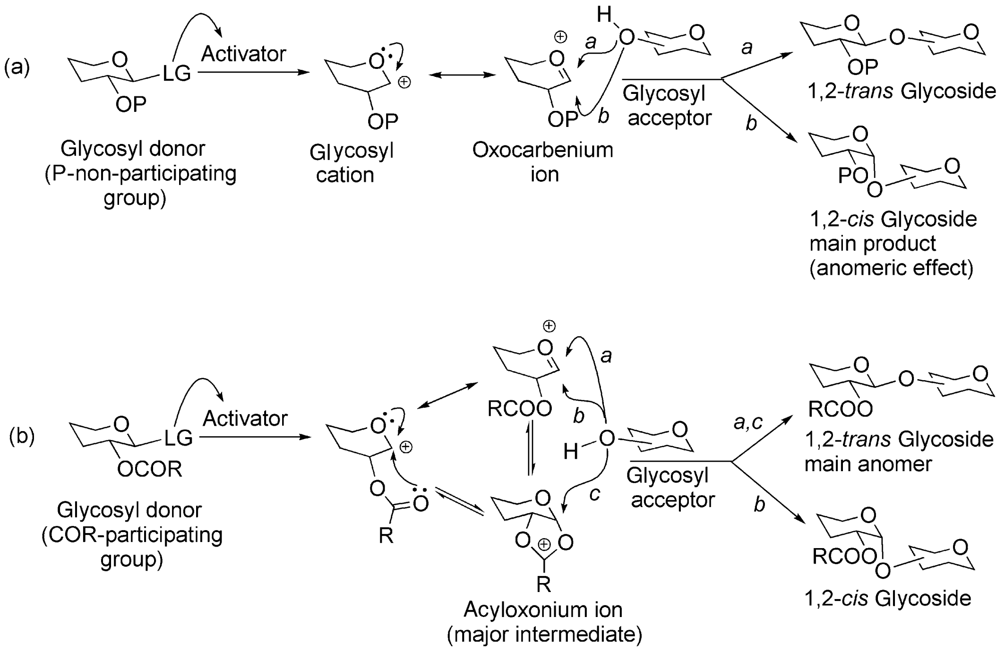


Figure 1.2 Survey of glycosyl donors.

addition, a variety of new recent methodologies bring the use of classic glycosyl donors such as hemiacetals to entirely different level of flexibility and usefulness [63]. These innovative concepts will be discussed in the subsequent chapters dealing with particular classes of clycosyl donors..

1.4
General Reaction Mechanism

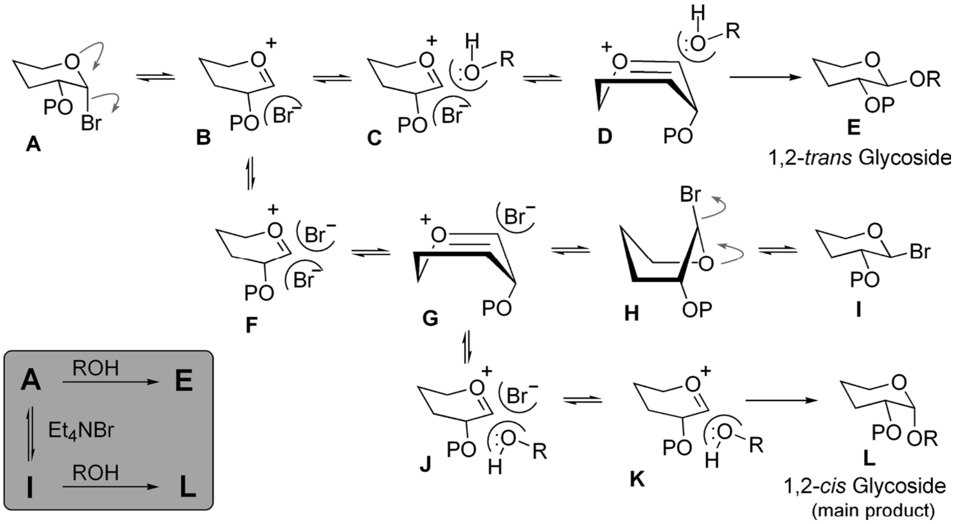
Detailed glycosylation mechanism has not been elucidated as yet; therefore, speculations and diagrams presented herein are a commonly accepted prototype of the glycosylation mechanism. Most commonly, the glycosylation reaction involves nucleophilic displacement at the anomeric center. As the reaction takes place at the secondary carbon atom with the use of weak nucleophiles (sugar acceptors), it often follows a unimolecular S_N1 mechanism. Glycosyl donors bearing a nonparticipating and a participating group will be discussed separately (Scheme 1.1a and b, respectively). In most cases, an activator (promoter or catalyst) assisted departure of the anomeric leaving group results in the formation of the glycosyl cation. The only



Scheme 1.1

possibility to intramolecularly stabilize glycosyl cation formed from the glycosyl donor bearing a non-participating group is by resonance from O-5 that results in oxocarbenium ion (Scheme 1.1a). The most commonly applied nonparticipating groups are benzyl (OBn) for neutral sugars and azide (N_3) for 2-amino-2-deoxy sugars; however, other moieties have also been occasionally used. The anomeric carbon of either resonance contributors is sp^2 hybridized; hence, the nucleophilic attack would be almost equally possible from either the top (*trans*, β- for the D-gluco series) or the bottom face (*cis*, α-) of the ring. Even though the α-product is thermodynamically favored because of the so-called anomeric effect (discussed in the subsequent section) [64], a substantial amount of the kinetic β-linked product is often obtained owing to the irreversible character of glycosylation of complex aglycones. Various factors such as temperature, protecting groups, conformation, solvent, promoter, steric hindrance or leaving groups may influence the glycosylation outcome (discussed below) [65,66].

1,2-*trans* Glycosidic linkage can be stereoselectively formed with the use of anchimeric assistance of a neighboring participating group, generally an acyl moiety such as *O*-acetyl (Ac), *O*-benzoyl (Bz), 2-phthalimido (NPhth) and so on [67–69]. These glycosylations proceed primarily via a bicyclic intermediate, the acyloxonium ion (Scheme 1.1b), formed as a result of the activator-assisted departure of the leaving group followed by the intramolecular stabilization of the glycosyl cation. In this case, the attack of a nucleophile (alcohol, glycosyl acceptor) is only possible from the top face of the ring (pathway c), therefore allowing stereoselective formation of a 1,2-*trans* glycoside. Occasionally, substantial amounts of 1,2-*cis*-linked products are also



Scheme 1.2

formed, most often when unreactive alcohols are used as the substrates and/or poorly nucleophilic participatory substituents are present at C-2. In these cases, glycosylation assumingly proceeds via oxocarbenium ion, via pathways a and b (Scheme 1.1b), resulting in the formation of 1,2-*trans* and 1,2-*cis* glycosides, respectively, or most commonly mixtures thereof.

Seminal work by Lemieux on the halide-ion-catalyzed glycosidation reaction involved extensive theoretical studies that gave rise to a more detailed understanding of the reaction mechanism [70]. Thus, it was postulated that a rapid equilibrium could be established between a relatively stable α-halide **A** and its far more reactive β-counterpart **I** by the addition of tetraalkylammonium bromide (Et_4NBr, Scheme 1.2). In this case, a glycosyl acceptor (ROH) would preferentially react with the more reactive glycosyl donor (**I**) in an S_N2 fashion, possibly via the tight ion-pair complex **K**, providing the α-glycoside **L**. It is likely that the energy barrier for a nucleophilic substitution **I** → **L** (formation of the α-glycoside) is marginally lower than that for the reaction **A** → **E** (formation of a β-glycoside). If the difference in the energy barrier were sufficient, it should be possible to direct the reaction toward the exclusive formation of α-anomers.

Therefore, to obtain complete stereoselectivity, the entire glycosylation process has to be performed in a highly controlled manner. In this particular case, the control is achieved by the use of extremely mild catalyst (R_4NBr), although very reactive substrates and prolonged reaction at times are required.

Other common approaches to control the stereoselectivity of glycosylation will be discussed in the subsequent sections. In addition to the apparent complexity of the glycosidation process, there are other competing processes that cannot be disregarded. These reactions often cause the compromised yields of the glycosylation products and further complicate the studies of the reaction mechanism.

Elimination, substitution (formation of unexpected substitution products or hydrolysis at the anomeric center), cyclization (inter- and intramolecular orthoesterification), migration and redox are only a few to mention [71].

1.5
Anomeric Effects

A basic rule of conformational analysis known from the introductory organic chemistry is that an *equatorial* substituent of cyclic six-membered hydrocarbons is energetically favored. Hence, it is more stable owing to 1,3-diaxial interactions that would have occurred if a large substituent were placed in the *axial* position (Figure 1.3). For sugars, this rule is only applicable to hemiacetals (1-hydroxy derivatives) that are stabilized in β-orientation via intramolecular hydrogen-bond formation with O-5. Other polar substituents such as halide, OR or SR attached to the anomeric center of pyranoses/pyranosides prefer the axial orientation, which would be exclusive if the equilibrium at the anomeric center could be achieved. This phenomenon, which was first observed by Edward [72] and defined as *anomeric effect* by Lemieux [73], is partially responsible for the stereochemical outcome of processes taking place at the anomeric center of sugars [64,74,75].

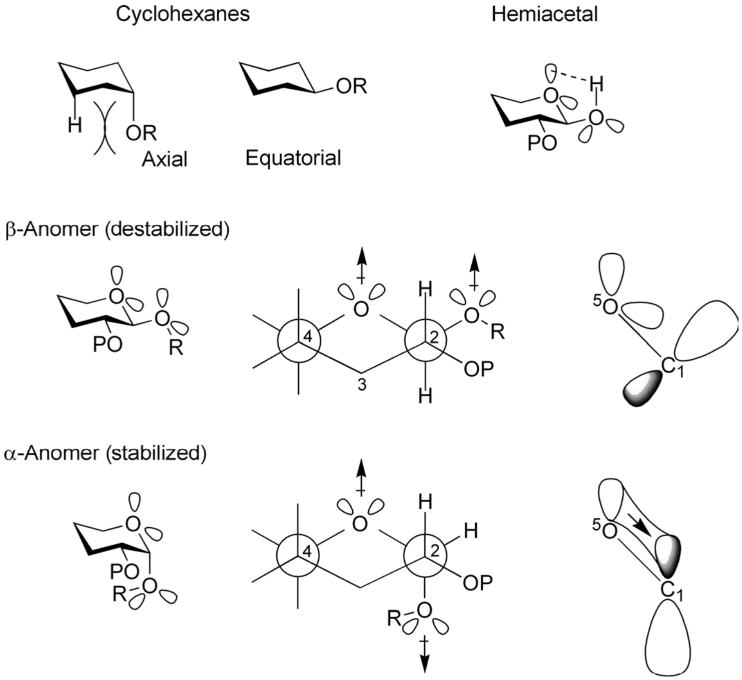


Figure 1.3 Anomeric effect.

What are the origins of the anomeric effect, sometimes referred to as *endo*-anomeric effect? In this context, the so-called *exo*-anomeric effect dealing with the stabilization of the β-anomer is of somewhat lesser influence on the overall process and will not be discussed herein [64]. One factor is that the substituent on the atom bonded to the ring at C-1 has lone-pair electrons, which would have repulsive interactions with those of the ring oxygen (O-5) if the anomeric substituent is in the equatorial position (β-position for D-sugars in $^4C_1$ conformation) but not if it is in the axial position (Figure 1.3). In addition, an electron-withdrawing axial substituent (α-anomer for D-sugars in $^4C_1$ conformation) is stabilized via hyperconjugation owing to the periplanar orientation of both nonbonding orbital of O-5 and antibonding orbital of C-1. This does not occur with the β-anomer, as the nonbonding orbital of O-5 and antibonding orbital of C-1 are in different planes and therefore are unable to interact.

1.6
Stereoselectivity of Glycosylation

As noted above, it is a general experience of carbohydrate synthesis that stereoselective preparation of 1,2-*cis* glycosides is more demanding than that of 1,2-*trans* glycosides. The formation of 1,2-*trans* glycosides is strongly favored by the neighboring-group participation (generation of intermediate acyloxonium ion). Typically, the use of a participating substituent at C-2 is sufficient to warrant stereoselective 1,2-*trans* glycosylation.

One of the factors affecting the stereochemical outcome of glycosidation of glycosyl donors bearing a nonparticipating substituent at C-2 is the anomeric effect, which favors α-glycoside formation (1,2-*cis* for the D-gluco series). However, because of the irreversible character of glycosylation, the role of the anomeric effect is diminished and other factors affecting the orientation of the new glycosidic bond (discussed below) often come to the fore. Although variation of reaction conditions or structural elements of the reactants may lead to excellent 1,2-*cis* stereoselectivity, no successful comprehensive method for 1,2-*cis* glycosylation has emerged as yet.

1.6.1
Structure of the Glycosyl Donor

1.6.1.1 Protecting Groups

The most powerful impact on the stereoselectivity is produced from the neighboring group at C-2. Neighboring-group participation is one of the most powerful tools to direct stereoselectivity toward the formation of a 1,2-*trans*-linked product. The neighboring substituent at C-2 is also responsible for the 'armed–disarmed' chemoselective glycosylation strategy [76]. The effects of the remote substituents are of lesser importance; however, there is strong evidence that a substituent at C-6 position may influence the stereochemical outcome of glycosylation dramatically. Although experimental proof has not emerged as yet, a possibility for the long-range 6-*O*-acyl or carbonate group assistance resulting in the preferential formation of α-glucosides cannot be overruled [77–81]. It was also found that the steric bulkiness or strong electron-withdrawing properties of a substituent at C-6 are beneficial for 1,2-*cis* glucosylation, most likely because of shielding (sterically or electronically) the top face of the ring and, therefore, favoring the nucleophilic attack from the opposite side [14,82–88].

Although the effect of the C-6 substituent was found to be of minor importance for the derivatives of the D-galacto series [89], a remote effect is sufficiently strong when a participating moiety is present at C-4 [90,91]. Thus, the use of *p*-methoxybenzoyl (anisoyl) [91] and diethylthiocarbamoyl [81] groups was found to be exceptionally beneficial for the formation of α-galactosides. Similar effects (including C-3 participation) were also detected for the derivatives of the L-fuco [92,93], L-rhamno [94], D-manno and D-gluco [14,82,95] series [96]. It was noted that when the unprotected hydroxyl is present at C-4 of the sulfamidogalacto donor, the expected β-glycosyl formation occurs. However, when the hydroxyl group is blocked with benzyl or acyl, the process unexpectedly favors α-glycoside formation. This phenomenon was rationalized via the formation of the intramolecular hydrogen bonding (C4—O—H · · · O—C5), destabilizing oxocarbenium ion contribution to the reaction mechanism that favors α-glycosylation (pathway b, Scheme 1.1a). Torsional effects induced by the cyclic acetal protecting groups may also strongly affect the stereoselectivity at the anomeric center; however, these effects remain unpredictable at this stage [88,97–99].

1.6.1.2 **Leaving Group**

There are a large number of publications describing the comparison of various glycosylation methods applied for particular targets. However, only few principles could be reliably outlined. It has been unambiguously demonstrated that halides activated in the presence of a halide ion (from, e.g. Bu_4NBr) often provide the highest ratios of α-/β-glycosides [100–104]. Since in most cases the glycosylation reactions proceed via unimolecular S_N1 mechanism, the orientation of the leaving group at the anomeric center is of lesser importance. However, the glycosylation reactions occasionally proceed via bimolecular S_N2 mechanism with the inversion of the anomeric configuration. In this context, glycosyl donors with 1,2-*cis* orientation form 1,2-*trans* glycosides: for example glycosyl halides with insoluble catalysts (also used for β-mannosylation) [105], α-imidates in the presence of boron trifluoride etherate (BF_3–Et_2O) at low temperature [106] and 1,2-anhydro sugars [107]. Conversely, 1,2-*trans*-oriented glycosyl donors stereospecifically afford 1,2-*cis* glycosides, for example highly reactive β-glucosyl halides [70], glycosyl thiocyanates [39,108] and anomeric triflates formed *in situ* were found superior for the synthesis of β-mannosides [109,110].

1.6.2

Structure of the Glycosyl Acceptor

1.6.2.1 **Position of the Hydroxyl**

Alcohol reactivity is typically inversely correlated with the 1,2-*cis* stereoselectivity – the most reactive hydroxyls give the lowest α-/β-ratios – the stronger the nucleophile, the faster the reaction, and hence the more difficult it is to control. Regarding the sugar or aliphatic glycosyl acceptors, the general rule normally states that glycosylation of more reactive primary hydroxyl provides poorer stereoselectivity in comparison to that when the secondary hydroxyls are involved [111]. The same principles are applicable for the synthesis of glycopeptides; thus, the glycosylation of the secondary hydroxyl of threonine typically gives higher α-stereoselectivity than when primary hydroxyl group of serine is glycosylated with 2-azido-2-deoxy-galactosyl bromide or trichloroacetimidates [112,113]. Occasionally, primary hydroxyls provide somewhat higher 1,2-*cis* stereoselectivity in comparison to that of the secondary hydroxyl groups. This can serve as an indirect evidence of the glycosylation reaction proceeding via the bimolecular mechanism, at least partially.

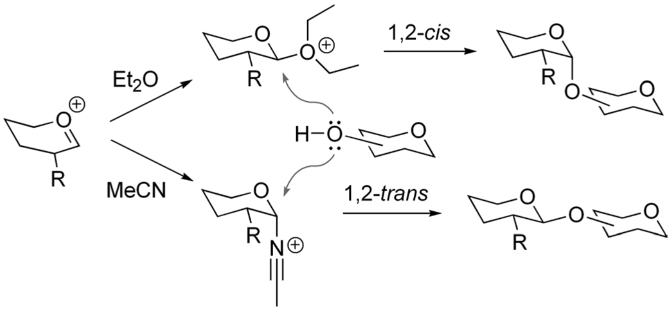
1.6.2.2 **Protecting Groups**

It is well established that ester-electron-withdrawing substituents reduce electron density of the neighboring hydroxyl group by lowering its nucleophilicity [88,105,114]. This may improve stereoselectivity, as the reaction can be carried out in a more controlled manner. As an example, glycosylation of axial 4-OH of galactose often gives excellent 1,2-*cis* stereoselectivity, especially in combination with electron-withdrawing substituents (e.g. *O*-benzoyl, OBz) [115]. However, poorly reactive hydroxyls can lose their marginal reactivity completely when surrounded by the deactivating species, resulting in lower glycosylation yields.

1.6.3
Reaction Conditions

1.6.3.1 **Solvent Effect**

Another important factor that influences the stereoselectivity at the anomeric center is the effect of the reaction solvent. In general, polar reaction solvents increase the rate of the β-glycoside formation via charge separation between O-5 and β-O-1. If the synthesis of α-glycosides is desired, CH_2Cl_2, $ClCH_2CH_2Cl$ or toluene would be suitable candidates as reaction solvents. However, there are more powerful forces than simple solvation that have to be taken into consideration. The so-called participating solvents, such as acetonitrile and diethyl ether, were found to be the limiting cases for the preferential formation of β-D- and α-D-glucosides, respectively [78]. These observations were rationalized as follows: if the reactions are performed in acetonitrile, the nitrilium cation formed *in situ* exclusively adopts axial orientation, allowing stereoselective formation of equatorially substituted glycosides (Scheme 1.3). This approach allows obtaining 1,2-*trans* glucosides with good stereoselectivity even with glycosyl donors bearing a nonparticipating substituent. On the contrary, ether-type reaction solvents such as diethyl ether, tetrahydrofuran [116] or dioxane [117] can also participate in the glycosylation process. Differently, in these cases the equatorial intermediate is preferentially formed, leading toward the axial glycosidic bond formation [85,86,118–120]. Nitroethane was also employed as a suitable solvent for 1,2-*cis* glycosylation [121].



Scheme 1.3

1.6.3.2 **Promoter (Catalyst), Additions**

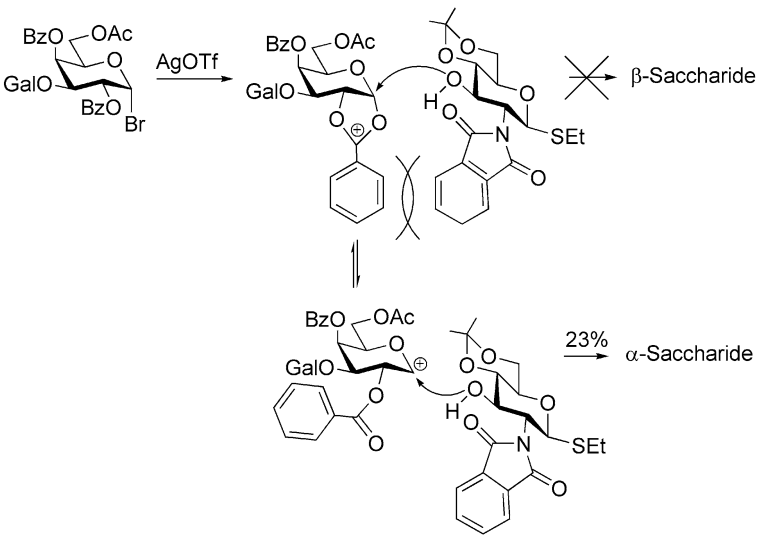
Milder activating conditions are generally beneficial for 1,2-*cis* glycosylation. Thus, halide-ion-catalyzed reactions give the best results for the glycosylation with glycosyl halides [70]; thioglycosides perform better when activated with a mild promoter, such as iodonium dicollidine perchlorate (IDCP) [122,123]; whereas trichloroacetimidates are best activated with the strong acidic catalysts, such as trimethylsilyl trifluoromethanesulfonate (TMS-triflate, TMSOTf) or triflouromethanesulfonic acid (triflic acid, TfOH) [106]. Various additions to the promoter systems often influence the stereochemical outcome of the glycosylation. Among the most remarkable examples is the use of perchlorate ion additive that was found to be very influential in 1,2-*cis* glycosylations [118,124].

1.6.3.3 **Temperature and Pressure**

High pressure applied to the reactions with participating glycosyl donors further enhances 1,2-*trans* selectivity [125]; when the high-pressure conditions were applied for glycosylations with a nonparticipating glycosyl donor, remarkable increase in the reaction yield was noted with only marginal changes in stereoselectivity [126]. Kinetically controlled glycosylations at lower temperatures generally favor 1,2-*trans* glycoside formation [100,120,127–130], although converse observations have also been reported [131,132].

1.6.4
Other Factors

Unfavorable steric interactions that occur between glycosyl donor and acceptor in the transition state or other factors or conditions may unexpectedly govern the course and outcome of the glycosylation process. One of the most remarkable effects, the so-called ‘double stereodifferentiation’ takes place when stereochemical interactions between bulky substituents in glycosyl donor and glycosyl acceptor prevail the stereodirecting effect of a neighboring participating group. The pair of reagents where these interactions occur is called a ‘mismatched pair’. Thus, only α-linked product was unexpectedly formed with 2-phthalimido glycosyl acceptor (Scheme 1.4). [133]. A coupling of the same glycosyl donor with conformationally



Scheme 1.4

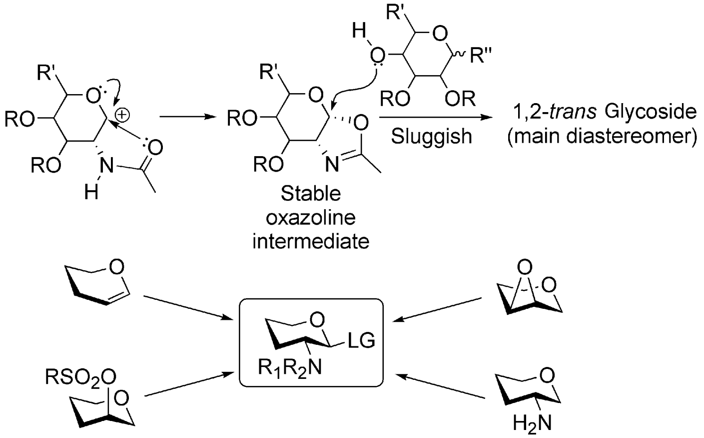
modified 1,6-anhydro acceptor afforded β-linked oligosaccharide with 75% yield. It was also demonstrated that if this effect takes place with a sugar of the D-series, its L-enantiomer forms a matched pair with the same glycosyl acceptor.

1.7
Special Cases of Glycosylation

This section outlines special cases of glycosylation, not necessarily uncommon, which do not follow general conventions discussed above. It is not unusual when general glycosylation methods do not work or cannot be applied to the synthesis of glycosides described herein. The synthesis of each of these classes of compounds requires careful selection of techniques, their modification or design of conceptually new approaches. Sometimes special indirect or total synthesis based technologies have been developed and applied specifically to the synthesis of these targets.

1.7.1
Aminosugars

Glycosides of 2-amino-2-deoxy sugars are present in the most important classes of glycoconjugates and naturally occurring oligosaccharides, in which they are connected to other residues via either 1,2-*cis* or, more frequently, 1,2-*trans* glycosidic linkage [134–136]. In particular, 2-acetamido-2-deoxyglycosides, most common of the D-gluco and D-galacto series, are widely distributed in living organisms as glycoconjugates (glycolipids, lipopolysaccharides, glycoproteins) [134], glycosaminoglycans (heparin,



Scheme 1.5

heparin sulfate, dermatan sulfate, chondroitin sulfate, hyaluronic acid) [137] and so on [138,139]. Special efforts for the synthesis of glycosyl donors of 2-amino-2-deoxy sugars have been focusing on the development of simple, efficient, regio- and stereoselective procedures.

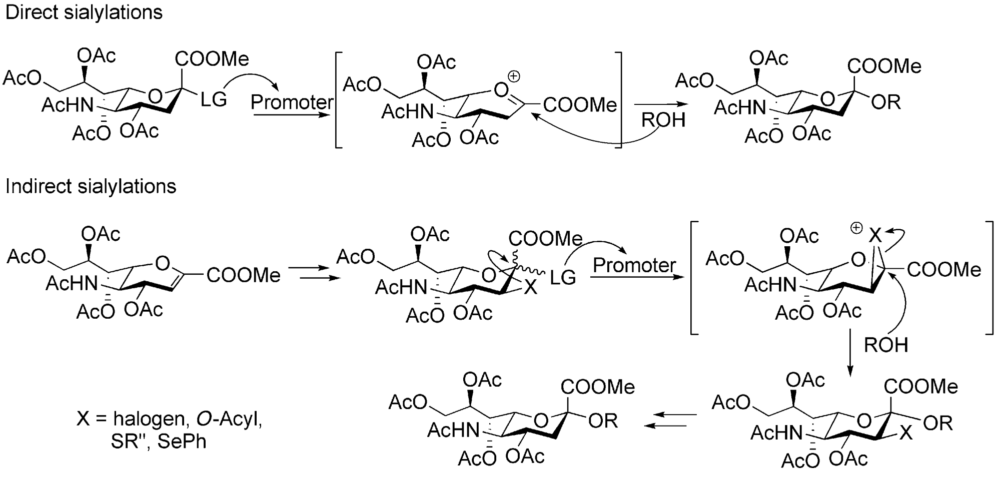
As a vast majority of naturally occurring 2-amino-2-deoxy sugars are *N*-acetylated, from the synthetic point of view, a 2-acetamido-2-deoxy substituted glycosyl donor would be desirable to minimize protecting-group manipulations. For this type of glycosyl donors, however, the oxocarbenium ion rearranges rapidly into an oxazoline intermediate (Scheme 1.5). Even under harsh Lewis acid catalysis, this highly stable oxazoline intermediate does not exert strong glycosyl-donor properties. Although the synthesis of 1,2-*trans* glycosides is possible with the use of this type of glycosyl donors, the synthesis of 1,2-*cis* glycosides is a burden. As a matter of fact, the participating nature of the *N*-acetyl moiety presents an obvious hindrance when the formation of the α-linkage is desired. A minimal requirement for the synthesis of 1,2-*cis* glycosides would be the use of a C-2 nonparticipating moiety.

Nowadays, a variety of synthetic approaches to the synthesis of 2-amino-2-deoxyglycosides have been developed, and progress in this area has been reviewed [140–142]. These syntheses are started either from a glycosamine directly or by the introduction of nitrogen functionality to glycose or glycal derivatives. To this end, various glycosamine donors with modified functionalities have been investigated; in particular, those bearing an N-2 substituent capable of either efficient participation via acyloxonium, but not (2-methyl) oxazoline, intermediate for 1,2-*trans* glycosylation or a nonparticipating moiety for 1,2-*cis* glycosylation.

1.7.2
Sialosides

Sialic acids are nine-carbon monosaccharides involved in a wide range of biological phenomena. Their unique structure is characterized by the presence of a carboxylic group (ionized at physiological pH), deoxygenated C-3, glycerol chain at C-6 and differently functionalized C-5. Among the 50 derivatives reported so far, *N*-acetylneuraminic acid (5-acetamido-3, 5-dideoxy-D-*glycero*-D-*galacto*-non-2-ulopyranosonic acid, Neu5Ac) is the most widespread. The natural equatorial glycosides and their unnatural axial counterparts are classified as α- and β-glycosides, respectively. In spite of extensive efforts and notable progress, the chemical synthesis of sialosides remains a significant challenge [143–146]. The presence of a destabilizing electron-withdrawing carboxylic group and the lack of a participating auxiliary often drive glycosylation reactions toward competitive elimination reactions, resulting in the formation of a 2,3-dehydro derivative and in poor stereoselectivity (β-anomer). To overcome these problems, a variety of leaving groups and activation conditions for direct sialylations have been developed. It was also demonstrated that the *N*-substituent at C-5 plays an influential role in both stereoselectivity of sialylation and the reactivity of sialyl donors [147].

Along with these studies, a variety of indirect methods for chemical sialylation have been developed. Several glycosyl donors derived from Neu5Ac have been prepared that possess an auxiliary at C-3. This auxiliary should control the anomeric selectivity of glycosylation by neighboring-group participation, leading to the formation of 2,3-*trans*-glycosides [143]. Thus, α-glycosides are favored in the case of equatorial auxiliaries (Scheme 1.6), whereas β-glycosides are preferentially formed when the participating auxiliary is axial. The auxiliaries also help in preventing 2,3-elimination that often constitutes a major side reaction in the direct *O*-sialylations. One of the most important requirements is that an auxiliary should be easily installed prior to, and removed after, glycosylation. Usually, the auxiliaries are introduced by a chemical modification of the readily accessible 2,3-dehydro derivative of Neu5Ac [148]. These transformations can be performed



Scheme 1.6

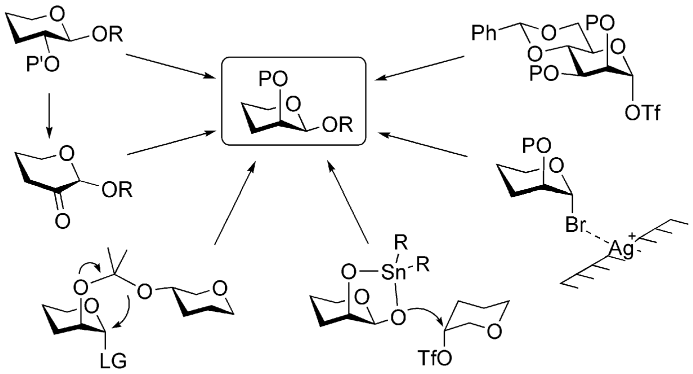
either through a 2,3-oxirane derivative or by a direct addition reaction to the double bond.

1.7.3
Synthesis of 2-Deoxyglycosides

2-Deoxyglycosides are important constituents of many classes of antibiotics. The development of reliable methods for stereoselective synthesis of both α- and β-2-deoxyglycosides has become an important area of research and development of new classes of drugs and glycomimetics [149,150]. It should be noted that because of the lack of anchimeric assistance of the substituent at C-2, the synthesis of both types of linkages represents a notable challenge. On one hand, the direct glycosylation of 2-deoxy glycosyl donors often results in the formation of anomeric mixtures. Similar to that of conventional glycosylation, the solvent and promoter effects play important stereodirecting roles in the synthesis. On the other hand, similar to that discussed for the sialosides, a participating auxiliary can be used to add to the stereoselectivity of glycosylation. Usually this moiety is introduced through 1,2-dehydro derivatives with concomitant or sequential introduction of the anomeric leaving group. The methods employing both axial and equatorial substituents are known to result in the formation of 1,2-*trans* glycosides, which upon 2-deoxygenation can be converted into the respective targets. Although this latter approach requires additional synthetic steps, it is often preferred because it provides higher level of stereocontrol.

1.7.4
Synthesis of β-Mannosides

β-Mannosyl residues are frequently found in glycoproteins. The chemical synthesis of 1,2-*cis*-β-mannosides cannot be achieved by relying on the anomeric effect that would favor axial α-mannosides at the equilibrium. In addition, it is further disfavored by the repulsive interactions that would have occurred between the axial C-2 substituent and the nucleophile approaching from the top face of the ring. For many years the only direct procedure applicable to β-mannosylation – Ag-silicate promoted glycosidation of α-halides – was assumed to follow bimolecular S_N2 mechanism [13,14]. The difficulty of the direct β-mannosylation was addressed by developing a variety of indirect approaches such as C-2 oxidation-reduction, C-2 inversion, anomeric alkylation and intramolecular aglycone delivery (Scheme 1.7) [151–155]. This was the standing in this field before Crich and coworkers discovered that 4,6-*O*-benzylidene protected sulfoxide [109] or thiomannoside [110] glycosyl donors provide excellent β-manno stereoselectivity. Mechanistic and spectroscopic studies showed that anomeric α-*O*-triflates generated *in situ* as reactive intermediates can be stereospecifically substituted. On a similar note, 2-(hydroxycarbonyl)benzyl glycosides have proven to be versatile glycosyl donors for the synthesis of β-mannosides via anomeric triflate intermediates [58].



Scheme 1.7

1.7.5
Synthesis of Furanosides

In comparison to their six-membered ring counterparts, furanosides are relatively rare. Nevertheless, their presence in a variety of glycostructures from bacteria, parasites and fungi makes this type of glycosidic linkage an important synthetic target [156,157]. The synthesis of 1,2-*trans* furanosides is relatively straightforward and, similar to that of pyranosides, can be reliably achieved with the use of glycosyl donors bearing a participating group at C-2. In contrast, the construction of 1,2-*cis*-glycofuranosidic linkage is difficult, even more so than with pyranosides, because the stereocontrol in glycofuranosylation cannot be added by the anomeric effect owing to the conformational flexibility of the five-membered ring. In fact, both stereoelectronic and steric effects favor the formation of 1,2-*trans* glycofuranosides. Despite some recent progress, stereoselective synthesis of 1,2-*cis* glycofuranosides has been one of the major challenges of synthetic chemistry. General glycosylation methods, involving glycosyl fluorides [158], trichloroacetimidates [159], and thioglycosides [156,160] along with less common and indirect techniques [161–164], were applied to 1,2-*cis* furanosylation. More recently, a notable improvement in stereoselectivity of 1,2-*cis* furanosylation was achieved by using glycosyl donors in which the ring has been locked into a single conformation. These examples include 2,3-anhydro [165–169], 3,5-*O*-(di-*tert*-butylsilylene) [170,171] and 3,5-*O*-tetraisopropyldisiloxanylidene [172] protected bicyclic glycosyl donors.

1.8
Glycosylation and Oligosaccharide Sequencing

Stereoselective glycosylation is only a part of the challenge that synthetic chemists confront during the synthesis of oligosaccharides. Regardless of the efficiency of a single glycosylation, a traditional stepwise approach requires subsequent conversion of the disaccharide derivative into the second-generation glycosyl acceptor or glycosyl