Synthesis of Glycosides of Glucuronic, Galacturonic and Mannuronic Acids: An Overview

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Abstract: Uronic acids are carbohydrates present in relevant biologically active compounds. Most of the latter are glycosides or oligosaccharides linked by their anomeric carbon, so their synthesis requires glycoside-bond formation. The activation of this anomeric center remains difficult due to the presence of the electron-withdrawing C-5 carboxylic group. Herein we present an overview of glucuronidation, mannanuronidation and galacturonidation reactions, including syntheses of prodrugs, oligosaccharides and stereochemical aspects.

Keywords: glycosidation; glucuronic acid; galacturonic acid; mannuronic acid; stereoselectivity

1. Introduction

Uronic acids are reducing sugars of biological relevance. They are involved in the metabolism of many drugs and endogenous compounds, and they are found natural products such as glycosaminoglycans, pectins and carragenans, among others, isolated from different sources – mammals, plants and algae.

In uronidation reactions, uronic acids are attached to an aglycone through the anomeric carbon atom forming O-glycosidic bonds. Although sharing all common aspects with general glycosidations, which
has been extensively reviewed [1,2], the synthesis of uronic acid glycosides is particularly challenging, because the presence of the C-5 carboxylic group decreases the reactivity at the anomeric position. Different methodologies have been investigated in order to overcome this drawback and to develop general strategies allowing the synthesis of uronic acid glycosides of biological importance and of complex oligosaccharides in a regio- and stereoselective manner.

Several methods are available for the synthesis of glycosides of glucuronic acids, and these can be placed into two broad categories involving either oxidation of the corresponding glucoside or by carrying out the glycosidation on an activated glucuronic acid. This manuscript covers the second approach.

As glucuronidation has been previously reviewed in 1998 [3,4], the present review will summarize the latest advances on glucuronic, galacturonic and mannanuronic acid glycosylation methodologies, especially those involving the synthesis of metabolites of bioactive molecules and oligosaccharides. As L-iduronic acid has been the subject of a great number of articles dealing to the synthesis of heparin sequences, its reactions are out of the scope of this review.

2. Glucuronidation

2.1. Metabolites Synthesis

Many drugs have been conjugated to D-glucuronic acid (GlcA) in order to obtain the required tools for improving insights on their absorption, metabolism and bioavailability. Moreover, the isolation of the metabolites is often tedious and analytical standards are necessary as reference compounds for quantification of metabolite levels in clinical samples and for further pharmacological evaluation.

Different methods, for the preparation of these standards conjugated to polyphenol residues, have been developed [1,5,6]. The study of metabolites of drugs can contribute to the toxicity, research and safety assessment, taking into account that the biological activity of GlcA conjugates is often similar or even higher than that of the aglycone (drug) [7,8].

A number of GlcA donors have been used for the synthesis of glucuronides. The first report on stereoselective synthesis and characterization of morphine 6-α-D-glucuronide (M6αG), useful as a reference marker for testing the purity and stability of the pharmaceutically important morphine 6-β-D-glucuronide (M6G) was described by Rukhman et al. [9,10] Several groups reported various methodologies for the synthesis of M6G, the preparation of the α-isomer had not been previously reported, and therefore the chemical and biological properties of morphine 6-α-D-glucuronide remained unknown. The synthesis the α-anomer is based on the glycosylation of 3-O-acetylated morphine 2 with methyl 2,3,4-tri-O-acetyl-D-glucopyranosyluronate bromide 1 as glycosyl donor and zinc bromide as catalyst (Scheme 1).

The selectivity of this reaction is controlled by the amount of catalyst, the use of 1.8 equivalents of ZnBr2 afforded M6αG in a 8:1 α/β ratio. After hydrolysis of the acetyl groups, crystallisation gave the α anomer in reasonable yield (63%).
Other examples of Koenigs-Knorr procedures to synthesize metabolites are the preparation of doxorubicin, daunomycin, clenbuterol and edaravone glucuronates. Daunomycinone-7-D-glucuronide (DM7G, 5) and doxorubicinone-7-D-glucuronide (DX7G, 6) were conveniently prepared through the glycosylation at the 7-hydroxyl group of daunomycinone (3) or 14-acetoxydoxorubicinone (4) with \( \alpha\)-glucosyluronate bromide 1 by a Koenigs-Knorr procedure catalyzed by HgBr\(_2\) (Scheme 2), followed by alkaline deacetylation using aqueous LiOH solution and Amberlite cation exchange material [11]. The desired compounds 5-6 were obtained as a 3:7 \( \alpha/\beta \) mixture, the anomers could be separated by flash column chromatography.

Clenbuterol is a sympathomimetic agent that selectively activates \( \beta_2 \)-receptors. It is used as a bronchodilator for the treatment of asthma and chronic obstructive pulmonary disease in both human and veterinary medicines. Two clenbuterol O-glucuronide diastereomers (8, Scheme 3) were synthesized by the Koenigs–Knorr reaction [12] of 1 with racemic 7. HPLC purification allowed to isolate both diastereoisomers albeit in very low yield (1.7%), due to the formation of orthoesters by-products.
During the Koenigs Knorr reaction, orthoesters are frequently produced as by-products. The formation of orthoester derivatives can be explained by the competitive nucleophilic attack of the oxygen atom of the alcohol on the two possible electrophilic sites of the intermediate II, which is obtained from the oxocarbenium ion I (Scheme 4).

Scheme 4. Mechanism of β-glycoside and orthoester formations.

Edaravone, a neuroprotective agent is metabolized to the glucuronate metabolite in humans. Edaravone glucuronate and edaravone sulfate, two metabolites of edaravone, were synthesized in high yields [13]. The edaravone glucuronate 10 was synthesized from glucosyluronate bromide 1 by conjugation with edaravone (9) using silver trifluoromethanesulfonate as promoter (Scheme 5).

Scheme 5. Synthesis of edaravone glucuronate 10.

The investigation of drug metabolism requires a substantial amount of metabolites. As isolation from urine is long and tedious, the material obtained by synthesis is often preferred. In the case of phenolic compounds, the synthesis of glucuronides has been studied for each individual case by Arewang et al [14]. A number of GlcA donors as benzoylated or acetylated glucosyluronate bromides were treated with acceptor 12 under silver triflate promotion. Coupling of the acetylated donor 1 with 12 at 0 °C, gave a α/β-mixture (1:3) in 40% yield, whereas the benzoylated donor 11 produced a mixture of β-glycoside 13 and the corresponding orthoester in similar yield at the same temperature (Scheme 6). Exclusive formation of β-glycoside 13, albeit still only in a moderate yield (40%), was obtained when the coupling between donor 11 and acceptor 12 was carried out at ambient temperature. The use of the other donors did not improve the yield.

For the aglycone 14, the acetylated donor 1 gave the best result. On the other hand, in the case of compound 17 the use of 1-O-acetyl derivative 16 as donor in the presence of BF3 etherate afforded the glucuronide 18 in 67% yield (Scheme 6). These synthetic pathways used easily available glycosyl donors and allowed the preparation of substantial amounts of the target glucuronides. Bromide glucosyluronate donors gave moderate yields and have been compared to imidate glucuronyl donors which are more efficient in most cases.
To study the resveratrol metabolites and their effects on cell viability and on the inhibition of HIV infection, a one-pot synthetic approach using a random glycosylation procedure between resveratrol (19) and methyl acetobromoglucuronate 1 was used leading to resveratrol 3-O-glucuronide 20 and resveratrol-4’-O-glucuronide 21 [15] (Scheme 7). The 3-O- and 4’-O-glycoside derivatives were formed in one-pot. After deprotection, the mixture was purified by HPLC and the desired resveratrol-3-O- and 4’-O-glucuronides 20 and 21 were isolated in 13 and 18% yields, respectively, based on the resveratrol used.

In order to improve the yields of these two conjugates, the glycosylation between the trichloroacetamidate donor 22 and the silylated resveratrol acceptors 23 and 24 was performed in the presence of trifluoromethanesulfonate. Resveratrol-3-O- and 4’-O-glucuronides 20 and 21 were obtained in 94 and 89% yields, respectively (Scheme 7).

Another example is the chemical synthesis of quercetin 3’-glucuronide 29. Previously described by Wagner et al. [16], glucuronidation of 25 with glucosyluronate bromide (1) in the presence of silver
oxide (Ag₂O), only gave 26 in 40% yield from 25. The glucuronidation was accompanied by the formation of the glycal 27 (Scheme 8). The use of imidate donor 28 led exclusively to the quercetin-3’-glucuronide 29 in 11% overall yield. The methyl ester trichloroacetimidate 28 was prepared using a known procedure starting from D-(+)-glucurono-3,6-lactone in 60% yield over four steps [17].

**Scheme 8.** Comparison of bromide and imidate donors for the preparation of quercetin 3’-glucuronides.

A comparison of the reactivity between bromide and imidate donors was studied for the glucuronidation of ABT-751 32 which was evaluated as a treatment for pediatric neuroblastoma [18]. Compound 32 is metabolized in humans to glucuronide 33 and therefore the synthesis of both compounds was required to support Phase II clinical studies. The initial synthesis of glucuronide 33 used p-nitrophenol 30 and the glucosyluronate bromide (1) to form the corresponding glycoside, followed by five further steps to reach the target molecule 33 (Scheme 9).

**Scheme 9.** Glucuronidation of ABT-751.

The Schmidt trichloroacetamidate methodology, promoted by BF₃.Et₂O gave, after deprotection, the glucuronide 33 directly in 60% yield, allowing a faster synthesis of the multigram quantities required for clinical use (Scheme 9).
While methods to synthesize simple glucuronides are relatively well developed, the synthesis of structurally complex glucuronides is not straightforward. The efficiency and scale ability of such syntheses is often limited by low yields or unselective glycosidic couplings, complex protecting group strategies, tedious isolations, or enzymatic reactions. In these examples glucuronate imidate donors showed to be more reactive, leading to better results.

In general, the use of GlcA derived glycosyl donors is often inefficient due to the destabilizing effect of the C-5 electron withdrawing group on the glycosidic bond forming event. A gram-scale synthesis of the glucuronide metabolite of ABT-724, potent selective D4 dopamine receptor agonist, could be obtained from imidate donor 28 and compound 34 in the presence of BF3.Et2O in 75% yield [19]. Compound 35 gave after 6 steps the metabolite of ABT-724 36 in 33% overall yield from 28 (Scheme 10).

**Scheme 10. Preparation of the ABT-724 metabolite.**

The attempts to synthesize 36 directly from the ABT-724 phenolic compound failed whatever the conditions (donors, promoters) used. For other phenolic compounds, the glucuronidation needs to be compatible with the stability of the aglycone ring as flavones or isoflavones. For example, the first efficient synthesis of flavanone glucuronides as potential human metabolites was optimized for 7,4′-di-O-methylerydrcytol (persicogenin, 38) because it did not involve a complex protection/deprotection strategy of the aglycone moiety [20]. Thus, the 2,3,4-triacetyl-D-methyl-glucuronate-N-phenyl)-2,2,2,- trifluoroacetamide donor 37 was treated with 38 in the presence of BF3 etherate to yield the acetylated glucuronide in 41% which gave after deacetylation and deprotection of the methyl ester by an esterase the final compound 39 in 73% yield (Scheme 11).

**Scheme 11. Glucuronidation of persicogenin.**
A high yielding synthesis of isoflavone 7-glucuronides was accomplished by the reaction between the 7-OH of the isoflavone esters and a novel O-acetyl glucuronyl (N-p-methoxyphenyl)-trifluoroacetimidate donor 40 [21]. Treatment of 4-O-hexanoyl-daidzein (41a) and glycitein (41b) with O-acetyl glucuronyl trifluoroacetimidate 40 in CH2Cl2 under the promotion of BF3.Et2O (0.2 equiv) at room temperature led to the desired coupling products, 42a and 42b in 81% and 78% yields, respectively, as only the β anomers (Scheme 12).

Scheme 12. (N-p-Methoxyphenyl)-trifluoroacetimidate donor for glucuronidation of 4-O-hexanoyl-daidzein and glycitein.

The synthesis of pure standards of isoflavone O-glucosides and O-glucuronides were developed for a better understanding of the absorption, the metabolism and the bioavailability of isoflavones, [22]. This methodology was used to prepare the 7-O-glycosides of the three main isoflavones, daidzein, genistein and glycitein.

Scheme 13. Preparation of isoflavone O-glucuronides.

To improve the yields of some glucuronidations of phenolic compounds, different protecting groups on the glucuronate donor were studied. A simple and direct glucuronidation strategy for the urolithin-B 44, the silylated resveratrol 48, and the corresponding hydroxytyrosol derivatives 51, was described (Scheme 13). The critical glycosylation step was optimized using a structurally simple phenol,
urolithin-B, by modification of several reaction parameters (solvent, promoter, and glucuronide donor). Glycosylation of urolithin-B acceptor 44 and glucuronosyl donor 43 was first performed using TMSOTf as the promoter with a moderate yield but a very good stereoselectivity, only the β-monomer was obtained. To improve the yield, the most common promoter used in aromatic glycosylation BF3.OEt2 was used [23] for the reaction of 44 with the glucuronosyl donor 43, producing compound 47 in much higher yield (78%). When the glucuronosyl donors 22 and 28 were reacted with urolithin-B 44, products 45 and 46 were obtained in very high yields (95% and 83%, respectively) with no sign of orthoester formation.

The glucuronidation of silylated resveratrol 48 was performed in 71% yield from the acetylated imidate donor 28. Benzoylated imidate 22 treated with 51 gave a higher yield (84%) than the two other donors 22 and 43 (18 and 53% yield respectively). These results showed the importance of the optimization of the reactions conditions, as well as promoters for each phenolic acceptor/protector donor pair.

Another protected imidate, the isobutyryl imidate 55, has been successfully used in this type of reactions. For example, the synthesis of morphine-3,6-di-β-D-glucuronide was efficiently synthesized from this imidate donor. Previous attempts to couple methyl 2,3,4-tri-O-acetyl-1-O-trichloroacetimidoyl-α-D-glucopyranuronate (28) to 3-acetylmorphone by Lewis acid catalysis, afforded mostly 3,6-diacetymorphine and a small amount of the desired 6-glucuronate. Similar poor results were obtained with morphine 56 [24]. The methyl groups of the sugar acetates was replaced by larger groups in order to increase steric hindrance. Therefore, the rate of nucleophilic attack at the carbonyl, and hence transacylation, was reduced, whereas the rate of glycosylation was relatively unaffected. The isobutyrate group was found to be the best compromise, combining minimal transacylation with ease of hydrolysis. Subsequent use of the tri-isobutyrate 55 led to effective preparations of M3,6dIG 57 and related derivatives, with essentially complete stereoselection for the β-anomers due to participation of the neighbouring C-2 acyl group.

**Scheme 14.** Isobutyryl imidate as donor in the synthesis of morphine-3,6-di-β-D-glucuronide.

By reaction of imidate 55 with dry morphine 56 in dichloromethane in presence of BF3/Et2O catalyst, the morphine 3,6-β-D-glucuronide derivative 57 in crystalline form was obtained with exclusive β-stereochemistry at C-1 of both glucuronates in 60% yield (Scheme 14).

The glucuronidation of a number of important steroidal secondary alcohols, such as androsterone (58), epianandrosterone (59), 17-acetoxyandrostane-3α,17β-diol (60) and 11α-hydroxyprogesterone (61) was studied for different glycosyl donors [25] (Scheme 15).
Glucuronates are well known to be poor glycosyl donors and the reactivity of alcohols 58-61 is rather low. To reduce transacylation, a known side-reaction, reaction of tri-isobutyryl imidate 55 was studied under both ‘normal’ (Method A; viz. adding Lewis acid catalyst to the mixture of alcohol and 55) and ‘inverse’ conditions (Method B; viz. adding imidate to a mixture of alcohol and catalyst). The tri-isobutyryl imidate 55 gave satisfactory results in inverse mode with androsterone 58 and epiandrosterone 59, as compounds 63 and 64 were obtained in 41 and 54% yields respectively, whereas in normal mode the yields were 16 and 34% respectively. Also, imidates 55, 43 and iodide 62 showed to be efficient donors for the β-glucuronidation of a range of steroidal secondary alcohols.

Scheme 15. β-Glucuronidation of steroidal secondary alcohols.

When acetyl protecting groups were used, orthoesters side-products were obtained, whereas with isobutyryl imidate, in “inverse mode”, the yield increased [26]. For example the glucuronation of estradiol derivative 65 with the imidate 55 gave the glucuronide 66 in 77% yield (Scheme 16).

Scheme 16. Preparation of estradiol glucuronide.

Studies on the rapid metabolism of the trioxane derivative artemisinin, dihydroartemisinin (DHA, 70) required its conjugation to GlcA [27]:

Glucuronidation of 67 as acceptor component may work well if the donor can generate a highly stabilized carbonium ion, though at low temperature the 12α-1’β-glucuronide may predominate. So,
when 67 and 68 were treated. Use of TMS triflate-AgClO₄ at −10 °C, the 12α-isomer was obtained in 40% yield as the only product, while the use of BF₃/ET₂O at 20 °C gave mainly an anhydro-DHA. However, ZnCl₂ proved to be an effective catalyst. Thus reaction of 67 with 68 in the presence of ZnCl₂ afforded crystalline 69 in very satisfactory yield after chromatography (31%) (Scheme 17). The tri-O-isobutyryl imidate 55 showed improved stability and reduced transacylation compared to its acetyl protected analog. Thus reaction of 55 and 70 with BF₃,ET₂O gave complete reaction of 70 with noticeably less amounts of the DHA degradation products. By chromatography, the 12α,1'β-glucuronide ester 71 was isolated in excellent purity and 32% yield and 15% of the 12β-isomer 69. Both new esters 69 and 71 gave microanalytically pure material on recrystallization (Scheme 17).

Scheme 17. Conjugation of artemisinin and dihydroartemisinin to GlcA.

A successful synthesis of the glucuronide metabolite 73 was performed using a N-acetylated Soraprazan 72 and tri-iso-butryate trichloroacetamidate donor 55 [28] (Scheme 18), avoiding the formation of orthoesters observed when using the analogous acetyl protected trichloroacetimidate donor [26].

Scheme 18. Synthesis of soraprazan glucuronide.

Activation of the anomeric position of the glucuronate donor can also be achieved with a sulfonyl group. N-glucuronide 78, a major metabolite of 4-(imidazole-1-yl)butanamide derivative KRP-197/ONO-8025, known for its antimuscarinic activity, was synthesized via glucuronidation of
compound 76 using methyl 2,3,4-tri-O-benzoyl-1-methanesulfonyl-α-D-glucopyranuronate (75) [29] (Scheme 19). The latter showed β-selectivity and the glucuronide 77 was obtained in moderate yield (41%). Although this work involved the synthesis of N-glycosides, the strategy has a potential application in the preparation of O-glycosides from nucleophilic hydroxyl containing acceptors.

Scheme 19. Methanesulfonyl-α-D-glucopyranuronate donor.

This methodology, with formation of an anomeric mesylate, could be applicable to O-nucleophiles.

2.2. Prodrug Therapy

Since most of the glucuronides exhibit a weaker biological activity than their corresponding aglycones, the glucuronidation is generally considered as an important detoxification metabolic process in mammals. However, even if the glucuronide has no activity itself, it can undergo an enzymatic hydrolysis catalyzed by β-D-glucuronidase, releasing the corresponding biologically active aglycone. In some cases, the glucuronidation can maintain or even increase the therapeutic effect of the drug [30], probably because the active compound is gradually liberated. Synthesis of prodrugs, via a glucuronidation reaction has been studied for the development of more selective drugs, specially for selective delivery of systemically administrated chemotherapeutic drugs for solid cancers.

Indeed, glucuronides can be selectively activated at the tumoural site since the enzyme β-D-glucuronidase is found at highly elevated concentrations in necrotic tumour tissue [31,32]. The design of a suitable glucuronide prodrug must be based upon four criteria: enhanced water solubility, stability in blood, decreased cytotoxicity and drug release after enzymatic cleavage. Several glucuronide prodrugs have already been synthesized and proved to be selectively activated by β-glucuronidase, either present in high concentration in necrotic tumour areas (PMT) [31] or previously targeted to the tumour sites (ADEPT [33], GDEPT [34]), and consequently demonstrated superior efficacy in vivo compared to standard chemotherapy [35]. For example, two glucuronide prodrugs of the histone deacetylase inhibitor CI-994 81 were synthesized [36]. The β-O-glucuronyl carbamate 80 was synthesized by coupling the methyl glucuronate 67 with commercially available 2-nitrophenyl isocyanate in a very high β-diastereoselectivity in 86% yield (e.d. 97%) using the method developed by Leenders et al. [37] (Scheme 20).
A series of anthracycline prodrugs containing an immolative spacer were synthesized for application in selective chemotherapy. The key step in the synthesis of all prodrugs is the highly β-diastereoselective addition reaction of the anomeric hydroxyl of a glycosyl donor 67 to a spacer isocyanate resulting in the respective β-glycosyl carbamate pro-moieties [38] (Scheme 21).

The synthesis and biological evaluation of novel prodrugs based on the cytotoxic antibiotic duocarmycin was realized from imidate donors 28 and 86. The resulting glucuronide compounds were not isolated and directly coupled with the indole carboxylic acid 88 to afford the corresponding β-glucuronide 89 and 90 in 59% and 43% yields respectively [39] (Scheme 22).
2.3. Antibacterials Inhibitors

In the development of aryl glucuronides as potential probes for heparanase, the acid-catalysed glycosidation between the trichloroacetimidate-activated GlcA 28 and a variety of phenols was investigated. In preliminary studies, the BF$_3$/Et$_2$O-catalysed coupling of imidate 28 with phenols provided the desired aryl glucuronides 91-94 in high yields (61–81%) (Scheme 23). The attempted BF$_3$.Et$_2$O-catalysed glycosidation of 28 with 4-hydroxycinnamic acid 95 did not give the desired glycoside, but instead gave a complex mixture of products 96-99 [40] (Scheme 23).

Scheme 23. Trichloroacetimidate-activated GlcA as donor in the synthesis of aryl glucuronides.

To obtain the O-aryl glucuronide 101 in satisfactory yield, the methyl ester 100 was used as glycosyl acceptor (Scheme 24).

Scheme 24. O-glucuronidation of methyl 4-hydroxycinnamate.

CRM646-A and -B, two fungal glucuronides with a dimeric 2,4-dihydroxy-6-alkylbenzoic acid (orcinol $p$-depside) aglycone showing significant heparinase and telomerase inhibition activities, were synthesized for the first time [41]. The successful approach involved the construction of the phenol glucuronidic linkage, via coupling of the orsellinate derivative 102 with glucosyluronate bromide 1, before assembly of the phenolic ester onto the depside aglycone (Scheme 25). Attempts to perform direct glycosylation of the depside aglycone derivatives were not successful.
Tizoxanide is a potent antibacterial and antiparasitic agent. Metabolism of tizoxanide leads to the \( O \)-aryl glucuronide 106 which was efficiently synthesised in four steps from benzyl salicylate 104 and showed a low antibacterial activity. Koenigs-Knorr reaction of 1 with 104 gave the conjugate 105 in 61% yield which was then converted into 106 [42] (Scheme 26).

### Scheme 26. Synthesis of tizoxanide metabolite.

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\begin{align*}
\text{Scheme 25. Preparation of two fungal glucuronides.} \\
\end{align*}
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\begin{align*}
\text{Scheme 27. 2-\( O \)-Acyl glucuronate or cyanoester donors for \( \beta \)-glucuronidation.}
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\begin{align*}
\text{β-Glucuronated lactone 109 was provided from 2-\( O \)-acetylated donors 28 or cyano-orthoester 112. The anchimeric participation of the acetyl group in 28 guarantees the formation of the \( \beta \)-linkage, whereas the tribenzylated donor 107 gave the glucuronide 110 as a 2:8 anomeric mixture [43] (Scheme 27).}
\end{align*}
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An indirect strategy for the synthesis of glycosides GlcA involved the use of ulosyl bromide 113 (easily obtained in four steps from glucuronolactone). Glycosylation in the presence of insoluble silver
catalyst led to the β-glycoside 114 which could be converted stereoselectively to the gluco isomer 115, whereas selectride reduction afforded the mannuronide derivative [44] (Scheme 28).

**Scheme 28.** Ulosyl bromide donor for β-glucuronidation.

Glucuronyl iodide 62 has been studied as a “disarmed” glycosyl donor and primary or secondary alcohols as acceptors, promotion with NIS/I2 followed by TMSOTf gave the corresponding β-glucuronides in good yields [45] (Scheme 29). For example, 2-phenylethanol was glucuronylated in 88% yield when CuCl was used.

**Scheme 29.** “Disarmed” glucuronyl iodide for β-glucuronidation.

This methodology was applied to the synthesis of disaccharides with the same β-stereoselectivity. Glycosylation of conformationally inverted donors derived from GlcA was studied towards several silyl ethers [46]. 6,1−Lactone derivative 117 has been used in the synthesis of 1,2-cis-glycoside 118, the SnCl₄-catalyzed coupling of silyl ethers with 117 provides α-O-glucuronides in significantly improved yields without loss of stereoselectivity (Scheme 30). This methodology was extended to 2-deoxylactones, which gave α or β-glycosides depending on the structure of the donor.

**Scheme 30.** Stereoselective SnCl₄-catalyzed glucuronidation.

ROH (X) : aliphatic alcohols, sugars
The stereoselectivity observed for both 117 and 119 contrasted with that of the methyl ester 16, which only gave β-glucuronides 120. Similarly, the 2-deoxy-2 iodo donor 121 gave the β-glycosides 122, which is explained by the participation of iodine, better than the 2-O-acetyl group (Scheme 30).

Cyclic imidates can be used as glycosyl donors, and it was observed that 1,2-cis glycosides obtained from the reactions of glycosyl acetates or cyclic imidates, resulted from the anomerisation of initially formed 1,2-trans glycosides [47].

For example, reaction of imidate 123 with phenol in the presence of TMSOTf-SnCl4 (2.5/.5) gave after 1h a mixture of compounds 124, 125 and 126 in 1:1:2.2 ratio, and after 24h only the α-anomer in 63% yield (Scheme 31). The rate of the anomerisation reaction showed to be dependent on the structure of the aglycone [48] and for glucopyranuronic acid the anomerisation is faster than that of glucopyranuronate compounds.

Scheme 31. Cyclic imidates as glycosyl donors.

Microwave-assisted reaction of 6,1-lactone 117 with alcohols in the presence of acidic catalysts has been recently described. Classical Lewis acidic catalysts used in solvent-free conditions under MW irradiation gave different chemoselectivities compared to classical reactions, in shorter reactions times. For example, SnCl4 provided glycosylated-esterified compounds 127, while with FeCl3 or [α1-M(H2O)4P2W17O61]n– (M = Yb, Hf and Zr) Dawson-type polyoxometalate (POM) the chemoselectivity was in favour of esterified products 128 [49,50] (Scheme 32).

Scheme 32. Microwave-assisted glucuronidation.

Other heterogeneous systems were used, sulfuric acid loaded on porous silica (H2SO4/SiG60) and silica-supported Keggin type heteropolyacid. The reaction of the GlcA with different alcohols in the presence of these catalysts, gave glucofuranosidurono-6,3-lactone glycosides 129 in 62–98% yields (Scheme 33).
Scheme 33. Microwave-assisted formation of alkyl glucofuranosidurono-6,3-lactones.

The supported sulfuric catalyst was stable under microwave conditions and could be recovered and reused [51]. The formation of alkyl glucofuranosidurono-6,3-lactones have been already described from unprotected GlcA in heterogeneous media and promoted by Lewis acids [52].

The synthesis of the selectively protected disaccharides glycosides 132 and 133, which are required for further conversion into glycosyl donors for block synthesis of more extended oligosaccharides was studied.

A strategy for the synthesis of the target disaccharides [53], was the selection of the glucosyluronic donor. Selective O-deacetylation of methyl 1,2,3,4-tetra-O-acetyl-β-D-glycopyranuronate using hydrazinium acetate gave methyl 2,3,4-tri-O-acetyl-D-glucopyranuronate 67, followed by treatment with trichloroacetonitrile and 1,8-diazabicyclo[5,4,0]-unde-7-ene (DBU) to form the crystalline imidate 28 in good yield. The coupling of compound 28 with 130, in the presence of trimethylsilyl triflate as the glycosyl promoter and molecular sieves 4 Å in dichloromethane for 2 h at −30 °C, gave the desired disaccharide glycoside 132 in moderate yield (54%), after separation from some accompanying transesterification product of the acceptor, namely allyl 3-O-acetyl-4,6-O-benzylidene-2-deoxy-2-phthalimido-β-D-glucopyranoside 134 (Scheme 34). Coupling of compound 28 with 131 gave the crystalline glycoside 133 in 59% after separation of the side-product 135.

Scheme 34. Stereoselective synthesis of disaccharides glycosides.

Westman et al. prepared the thioethyl donor 136 for glycosylation of methyl 4-O-acetyl-2,6-di-O-benzyl-3-D-galactopyranoside (137) using dimethyl(methylthio)sulfonium triflate (DMTST) as promoter, affording the disaccharide 138 in 87% yield [54] (Scheme 35). Interestingly, this reaction was performed in the absence of an acid acceptor in order to prevent orthoester formation.
Jacquinet’s group reported that O-benzoylated derivatives of GlcA activated through their corresponding trichloroacetimidates were very efficient donors for the preparation of β-D-glucuronides [55]. Condensation of methyl 2,3,4-tri-O-benzoyl-1-O-trichloroacetimidoyl-α-D-glucopyranuronate 22 (1.5 equivalents) with the alcohol 139 in the presence of trimethylsilyl triflate afforded the crystalline trisaccharide derivative 140 in 85% yield (Scheme 36).

Deactivated donors such as GlcA phosphate 141 were found to be highly efficient in reactions with primary or secondary alcohols. Combined with the straightforward synthesis from readily accessible GlcA glycal precursors, the use of 141 as a glycosylating agent provided a direct entry to complex glycan structures [56]. The promoters used are TMS or TBS triflates, and the reactions were performed at low temperatures (−50 °C to −20 °C) affording 143 and 145 in 72% and 84% yield, respectively (Scheme 37).
Interestingly, glycosylations performed with C2-OH donor 146 (Scheme 38) also proceeded with complete β-selectivity. GlcA phosphate 146 was prepared and coupled with 142. The reaction was significantly more rapid than in the case of donor 141 bearing a C2-ester. Disaccharide 147 was obtained as the only product in good yield (65%) (Scheme 38).

**Scheme 38.** Glycosylation with C2-OH GlcA phosphate donor.

During the preparation of synthetic mimics of hyaluronan and its dimerized (Gemini) disaccharides [57] that could act as versatile building blocks with different therapeutic applications, glycosylation of the n-pentenyl glycoside acceptor 148 was carried out with the trichloroacetimidate donor 28 in the presence of TMSOTf, affording the β-(1→3) linked disaccharide 149 in 78% yield (Scheme 39).

**Scheme 39.** Preparation of synthetic mimics of hyaluronan.

Among others, Rele et al. showed that N-acetylglucosamine derivatives are unreactive acceptors [58]. Glycosylation of the n-pentenyl-terminated N-acetyl-D-glucosamine acceptor 151 with either glycosyl donor 1 or 28 was unsuccessful (Scheme 40).

**Scheme 40.** Unreactive N-acetylglucosamine acceptors in glucuronidation reaction.
On the other hand, azido precursors as 153 or 154 were suitable acceptors. Glycosylation of the \( n \)-pentenyl glycoside acceptors was carried out separately using the trichloroacetimidate donor 28 in the presence of acid promoters (Scheme 41). Attempts to glycosylate the sugar alcohol 154 with imidate 28 in the presence of TBSOTf or TMSOTf and molecular sieves produced the undesired ortho ester intermediate 155 as the major product. The authors believe that the molecular sieves, which are aluminosilicates and are inherently basic in nature, hindered acid-catalyzed 1,2-trans glycosidic bond formation. Indeed, in the absence of molecular sieves, boron trifluoride etherate mediated glycosylation of 153 with imidate 28 afforded the thermodynamically favored \( \beta \)-(1,4) disaccharide without a trace of ortho ester, albeit in 30% yield after 14 h. Significantly, TMSOTf-catalyzed conditions provided the most efficient route for coupling the acceptor 154 with glycosyl donor 28, producing the fully protected disaccharide 157 in moderate (60%) yield. The ortho ester 155 could be converted to disaccharide 157 using an excess amount of acid catalyst in the absence of molecular sieves. It was also observed that replacing the 3-\( O \)-benzyl protecting group by a 3-\( O \)-acetyl functionality retarded \( \beta \)-(1,4) glycosidic bond formation. Likely, the electron-donating nature of the 3-\( O \)-benzyl group enhanced the nucleophilicity of the glycosyl acceptor at the 4 position.

Scheme 41. Glucuronidation of \( n \)-pentenyl glycosides.

Thiophenyl glucuronate disaccharide donor 158 was used by Dinkelaar et al. [59] in an iterative strategy for hyaluronic acid (HA)-oligosaccharides assembling (Scheme 42). First, the reducing end glucosamine 159 was condensed with dimer 158 using the \( \text{Ph}_2\text{SO}/\text{Tf}_2\text{O} \) activating system. Although preactivation of the thiodisaccharide proceeded smoothly, the ensuing reaction with acceptor 159 did not go to completion and trisaccharide 160 was isolated in 46% yield. Changing from \( \text{Ph}_2\text{SO}/\text{Tf}_2\text{O} \) to the related \( \text{BSP}/\text{Tf}_2\text{O} \) reagent system significantly improved the outcome of the glycosylation, allowing to obtain the protected hyaluronic acid trisaccharide 160 in 75% yield. \( N \)-iodosuccinimide (NIS)/TfOH as activator system was also examined, giving trisaccharide 160 in 75% yield. Interestingly, the NMR spectrum of 160 revealed a rather small homonuclear coupling constant (\( J^\text{H1'-H2'} \)) for the anomic proton of the glucuronate moiety (H-1') of 4.4 Hz. Upon deprotection of the oligosaccharides the coupling constant changed to 8.4 Hz, indicative of the \( \beta \)-glucuronic acid linkage formed. The small coupling constant for the glucuronate anomic proton suggested that the glucuronate ester takes up a flattened 4C1-chair conformation, when positioned in between two 4,6-\( O \)-
di-tert-butylsilylidene glucosamine residues. To elongate trisaccharide 160, the C3″-O-Lev was deprotected and the resulting alcohol 161 was condensed with dimer 158 (NIS/TfOH activation) and pentamer 162 was obtained in 98% yield. Ensuing delevulinoylation of 162 gave alcohol 163 which was elongated in a subsequent NIS/TfOH mediated glycosylation with building block 158. Heptamer 165 was easily separated from the smaller products in the reaction mixture by size-exclusion chromatography on Sephadex LH-20, and isolated in 61% yield.

Scheme 42. Synthesis of hyaluronic acid-oligosaccharides.

In a investigation of the use of a safety-catch linker for supported synthesis of HA oligosaccharides, de Paz et al. [60] performed the glycosylation of acceptor 166 with donor 165 affording polymer-bound GlcA derivative 167 (Scheme 43). The reaction was repeated to drive it to completion as the first cycle resulted only in partial glycosylation.
Unfortunately, after delevulinoylation, the corresponding alcohol failed as acceptor with a glucosamine donor for disaccharide synthesis. The same drawback was encountered when a glucosamine acceptor was fixed on the resin and glycosylation with 165 was tried. This problem was attributed to the acylsulfonamide linker, and model glycosylations were carried out in solution and on PEG support without the N-acylsulfonamide linker to demonstrate this hypothesis. Thus, polymer acceptor 168 was efficiently glycosylated with trichloroacetimidate 165 to afford bound disaccharide 169. Therefore, the safety-catch linker approach was not suitable for oligosaccharide assembly involving glycosylation of low nucleophilic acceptors with electron-poor donors. It is reasonable to suppose that the chemical nature of the linker, in particular the high acidity of the NH proton, can explain the presence of charged species that hinder coupling reactions mediated by oxocarbenium ions.

The synthesis of glycosaminoglycan oligosaccharides has been the main interest of several research groups. Concerning heparan sulfate oligosaccharides, the synthesis of the key disaccharide building block 172 was smoothly accomplished by TMSOTf-catalyzed reaction (−30°C) of the 2-O-benzoyl-protected GlcA imidate 170 with the azido acceptor 171, providing the desired disaccharide donor 172 in 89% yield [61] (Scheme 44).

Scheme 44. Preparation of a key disaccharide building block for synthesis of oligosaccharides.

This block was used for the synthesis of a tetrasaccharide involved in prion diseases. The enzymatic synthesis of GlcA glycosides by the use of snail (Helix pomatia and Helix aspersa), limpet (Patella vulgata), and bovine glucuronidases was investigated by Nagatsukaa et al. [62].

As the acceptors, GlcA-O-pNP 173 (for selftransglycosylation), 6-O-sulfo-β-V-glucopyranosides (6-O-sulfo-Glc-O-pNP 176 and 6-O-sulfo-Glc-S-pNP 177), and 6-O-sulfo-β-D-galactopyranosides (6-O-sulfo-Gal-OpNP 182 and 6-O-sulfo-Gal-S-pNP 183) were employed, using GlcA O-pNP as the donor substrate (Scheme 45). All of the snail, limpet, and bovine enzymes were able to transfer GlcA from GlcA-O-pNP to the O-2 and O-3 positions of 6-O-sulfo-β-D-glucopyranoside.
Scheme 45. Enzymatic synthesis of oligosaccharides.

The limpet and bovine enzymes can also use the O-3 position of 6-O-sulfo-β-D-galactopyranosides for transglucuronylation. The enzymes also catalyze self-transglycosylation to afford O-2- or O-3-linked disaccharide. The bovine enzyme showed the highest reactivity for providing a practical enzymatic approach to highly functional saccharides like compounds 184 and 185 bearing both carboxyl and sulfate groups in one molecule. The main drawback of this approach is the obtention of mixtures of 2-/3-linked disaccharides, however in the case of the sulfated galactopyranoside derivative the reaction proceeded with complete regioselectivity.

3. Mannuronidation

Stereocontrolled synthesis of homooligomers of mannnuronic acid was performed from thiomannuronic derivatives. In this alginate oligomer, the uronic acid monomers are interconnected through 1,4-interglycosidic linkages that have a 1,2-cis configuration. Preactivation of thiomannuronic donor 186 with NIS/TMSOTf followed by addition of the mannnuronic acceptor gave the β-
disaccharide 188 in 78% yield, which was subjected to the same coupling reaction with donor 186 [63]. The trisaccharide 190 was obtained in 50% yield (Scheme 46).

Scheme 46. Preparation of a trisaccharide via manuronidation.

Conformational studies on the thiomannuronic donors showed that the electron-withdrawing C-5 carboxylate group destabilized the oxocarbenium ion. The oxocarbenium intermediate would adopt a 3H4 half-chair conformation 193a, with the carboxylate group in an axial position. The nucleophilic attack of the acceptor led to 1,2-cis-mannuronate 194 [64-66] (Scheme 47).

Scheme 47. Proposed mechanism for manuronidation from thiomannuronic donors.

The stereodirecting effect of the C-5 glycuronate ester has been demonstrated in the synthesis of a set of oligomers of manuronuric acid, 195, with 1,2-cis linkages (Scheme 48).

Scheme 48. Oligomers with 1,2-cis linkage from manuronuronic esters.
Further studies on conformationally restricted manuronates 196 and 198 were enterprised to explore the stereoselectivity of 1,2-cis-glycosylation [67,68]. The stereoselectivity is dependent on the nature of the protecting groups on the mannose core.

**Scheme 49.** Constrained uronate donors in manuronidation reaction.

The flexible manurononate 191 gave excellent β-selectivity, whereas conformationally constrained uronate donors such as 196 and 198 provided predominantly α-linkage formation in pseudo-disaccharides 198 and 199 (Scheme 49).

4. Galacturonidation

Magaud et al. described for the first time, the stereoselective α-(1→4) glycosylation between two D-galacturonic acid ester derivatives (Scheme 50) giving rise to disaccharides in good yields [69].

**Scheme 50.** D-galacturonic acid ester derivatives as donors and acceptors.

Using N-iodosuccinimide-trifluoromethanesulfonic acid as promoter, the thioglycoside donor 203 reacted smoothly at low temperature (−60 °C), with the glycosyl acceptor 201 to give stereoselectively the α-(1→4) linked product 206 (Scheme 50) in very good yield (Table 1). The coupling between donor 202 and acceptor 200 was achieved under the same conditions, and furnished exclusively the dimer 207.
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Table 1. Results of galacturonidation of thioglycoside donors 202-205.

| Entry | Donor | Acceptor | Product | α/β ratio | Yield (%) |
|-------|-------|----------|---------|-----------|-----------|
| 1     | 203   | 201      | 206     | 95/5      | 91        |
| 2     | 202   | 200      | 207     | > 95/5    | 70        |
| 3     | 204   | 201      | 208     | > 95/5    | 70        |
| 4     | 205   | 201      | 209     | > 95/5    | 78        |

Methyl 3,4-di-O-acetyl-1,2-O-[1-(exo-cyano)ethylidene]-α-D-galactopyranuronate was used as galacturonate donor by Vogel et al. [70]. The synthesis of disaccharides was carried out in the presence of tritylium perchlorate in dichloromethane. The coupling of 210 with 211 and 213 gave the expected β(1→2) and β(1→3)-linked disaccharides 212 (58%) and 214 (57%) containing no more than 1% of the α-anomer (Scheme 51).

Scheme 51. Stereoselective synthesis of β-linked disaccharides from cyano-orthoester donor.

In the case of glycosylation at C-4 (reaction between of 210 and 215) two phenomena were observed. First, 1',2'-trans and 1',2'-cis-disaccharides were formed, with the loss of stereoselectivity. Secondly, partial anomerisation at the glycosidic centre of the 1,2-trans-allyl glycoside 215 occurred.

Homogalacturonanes are interesting targets as they are the main components of plant pectin. Thioglycoside galacturonate donor 202 (Scheme 52) was tested with acceptor 217 using N-iodosuccinimide/silver triflate promotion at −20 °C with rigorous exclusion of moisture. The corresponding α-disaccharide 218 was obtained in 42% yield based on 217, showing that the tert-butyl ester was not stable under the glycosylation reaction conditions.
Scheme 52. Preparation of homogalacturonanes.

During the glycosylation conditions described above using \( p \)-methoxybenzyl derivative 219 as glycosyl donor and compound 220 as acceptor, the \( p \)-methoxybenzyl protective group proved not to be stable and the corresponding disaccharide with a free hydroxyl group in the C-4’-position was isolated in only 25% yield. In order to manage the lability of the \( p \)-methoxybenzyl function, different promotors were checked. The best result was obtained with freshly prepared iodonium di-sym-collidine perchlorate (64% isolated yield based on 220). Silver triflate/silver carbonate promoted glycosylation of glycosyl acceptor 220 with methyl(galactopyranosyluronate)bromide 222 provided the disaccharide 223 in 35% yield based on 220 [71]. This work by Vogel et al. allowed the preparation of di- and tri- galacturonan fragments.

On the other hand, the glycosylation of the galacturonate acceptor 220 with trichloroacetimidate donors in a ratio of 1:1 promoted by trimethylsilyl trifluoromethanesulfonate revealed a peculiar effect of the chosen substitution pattern (Scheme 53) [72]. Thus, the 3,4-di-O-acetyl-\( \alpha \)-D-galacturonate trichloroacetimidate 224 provided 67% yield of the \( \alpha \)-(1→4)-coupled disaccharide 225 and only 5% of the \( \beta \)-coupled disaccharide was detected. By way of contrast, the more active 2,3-di-O-benzyl glycosyl donor 226 coupled with 220 furnished the corresponding disaccharides 227 in a yield of 59% but no \( \beta \)-coupled disaccharide was observed. In earlier experiments the coupling of 220 with 226 in the
presence of boron trifluoride diethyl etherate furnished the corresponding disaccharides 227 in a total yield of 53% and a disappointing α/β ratio of 1.6:1.

Scheme 53. Preparation of oligogalacturonans.

Finally, the methyl group in the O-4 position of the glycosyl donor 228 gave rise to the lowest stereoselectivity (nearly 1:1 for disaccharide 229) with a total yield of 52%. Subsequent experiments shown that the α- or β-configuration of the trichloroacetimidate group at the anomic center of the donors exerted no influence on the outcome of stereoselectivity of the glycosylations investigated. This approach using galacturonates suitable as donors in α-glycosylation reactions can be carried out directly from commercially available D-galacturonic acid avoiding the crucial oxidation step in comparison to an approach involving D-galactose-derived intermediates.

The synthesis of glycosphingolipids is an important challenge. The Seeberger group studied the glycosylation of galacturonic acids 230 and 231 with ceramide A, to yield conjugate 234-236 (Table 2) [73] (Scheme 54). The problem in these reactions is the relatively poor solubility of ceramide A in many solvent systems at low temperature. During these studies, the well-known benefits of ether and the remote anchimeric assistance of C4 esters in galacto-configured systems in obtaining good α-selectivities became again apparent, as omission led to a dramatic increase in β-glycoside formation. A compromise between yield and selectivity was found by employing acetyl-protected thioglycoside 228.
and the NIS/TfOH activator system. Thus, the product 234 was isolated in 85% yield and 4.2:1 selectivity with dioxane/toluene (3:1) as solvent. Both anomers were separated by flash column chromatography.

Scheme 54. Synthesis of a conjugate of ceramide A.

Table 2. Effect of the activator system on galacturonidation of ceramide A.

| Entry | Donor | Conditions | Product | α/β | Yield (%) |
|-------|-------|------------|---------|-----|-----------|
| 1     | 233   | NIS/TBSOTf | 236     | <10 |           |
| 2     | 233   | NIS/TfOH   | 236     | 1:0:1 | 45        |
| 3     | 231   | NIS/TfOH   | 235     | 2:0:1 | 49        |
| 4     | 231   | DMTST      | 235     | <10  |           |
| 5     | 232   | TMSOTf     | 234     | 2:1:1 | 96        |
| 6     | 230   | NIS/TfOH   | 234     | 3:7:1 | 85        |
| 7     | 230   | NIS/TfOH   | 234     | 4:2:1 | 85        |

Fischer glycosylation of free galacturonic acid was recently studied by Allam et al. Tests were first conducted using sulfuric acid as the catalyst [74]. The best yields (80%) were obtained using 10 equiv of octanol at 80 °C during 48 h with a catalytic amount of sulfuric acid. A lower excess of octanol (5 equiv) clearly decreased the overall yield.

The formation of furanosiduronate compounds has already been reported when galacturonic acid was treated with methanol in the presence of Amberlite IR-120H for 48 h at 35 °C in an orbital shaker affording methyl (methyl D-galactofuranosid)uronate as the only product, in a 2.6:1 β:α ratio [75].

Shorter reaction times also resulted in lower yields. A higher temperature than 80 °C led to more important formation of degradation products (dark brown solution) while reaction at 50 °C showed a yield decrease. Several strong organic or mineral acid catalysts were used without noticeable changes in anomer ratio. In each case, the four anomers 237-238, 239-240 were present whatever the reaction conditions and the major one was identified as the β-furanose anomer (Scheme 55). The use of p-toluenesulfonic acid (PTSA) as the acid catalyst gave n-octyl (n-octyl D-galactoside) uronates 237-240 in good yield (83%), suggesting that organic sulfonic acids possess the appropriate acidities to promote the ester condensation. Under these experimental conditions, the ratio of the four anomers was
approximately 50% of β-furanose 237, 15% of α-furanose 238, 5% of β-pyranose 239 and 30% of α-\(\beta\)-yranose 240 as determined by \(^1\)H NMR. After chromatography, the pure major β-furanose isomer 237 can be obtained in about 35% yield.

**Scheme 55.** Fischer glycosylation of free galacturonic acid.

A significant acceleration of the reaction was observed using microwave activation, since all reactions were complete within 10 min. With \(\text{H}_2\text{SO}_4\) as the catalyst, the best results were obtained with a 10-fold excess of octanol (20 equiv) at 100 °C. A lower temperature (80 °C) was detrimental since the yield in \(n\)-octyl (\(n\)-octyl D-galactosid)uronates 237-240 dramatically decreased (25%). On the contrary, with a higher temperature (120 °C), the reaction turned brown with partial degradation of the sugar compounds and lower yield (52%). When compared to thermal activation, yields and product purity were improved since the reaction medium was only slightly yellow after the reaction. Unfortunately, the ratio of the four anomers was only slightly modified by microwave activation: β-furanose 237 60%, α-furanose 238 15%, β-pyranose 239 5%, and α-pyranose 240 20%. In the presence of a catalytic amount of PTSA, \(n\)-octyl (\(n\)-octyl D-galactosid)uronates 237-240 were only obtained as the furanose isomers in 63% yield, the pyranose ones representing about 5% of the mixture. Several attempts to improve the reaction yield and to reduce the ratio of the α-furanose anomer were unsuccessful. A rapid esterification of the carboxylic group with PTSA, which proceeds faster than glycosylation, could favor the formation of the furanose ring.

5. Conclusions

Uronic acid derivatives are poor donors, due to the deactivating effect of the electron-withdrawing carboxylate group. Peracetylated bromides are easy to prepare and have been widely used as donors, however they are less reactive than imidates and can lead to the formation of by-products as glycals or orthoesters. In addition, trichloroacetimidates allow glycosylation conditions compatible with a large variety of protecting groups. Thioglycosides, iodides and phosphates have been also employed as donors, but they are less developed. Benzoyl protected donors prevents orthoester formation, whereas isobutyryl derivatives reduce the risk of transacylation to the acceptor, often observed when using acetyl protected donors. The use of 6,1-lactones is an alternative approach to GlcA glycosides, either in “classical” conditions or by MW-assisted procedures. Glucuronidation of aminosugars is especially difficult. NHAc-containing derivatives fail in these reactions, the most useful acceptors are azido precursors or NHTCA derivatives. Phenolic hydroxyls required the development of specific methods. As observed in general glycosylation reactions, both reactivity and stereoselectivity is highly dependent on the electronic and steric character of protecting groups. Finally, mannuronic and
galacturonic acid glycosidations are less developed, probably due to their relative lower abundance in natural products, however analogous methodologies have been applied leading to comparable results.

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