Oxidative Activation of C–S Bonds with an Electropositive Nitrogen Promoter Enables Orthogonal Glycosylation of Alkyl over Phenyl Thioglycosides

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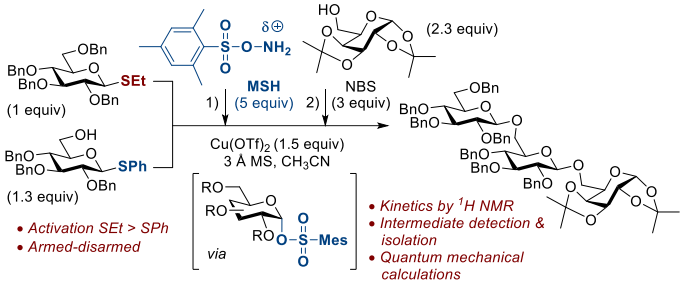


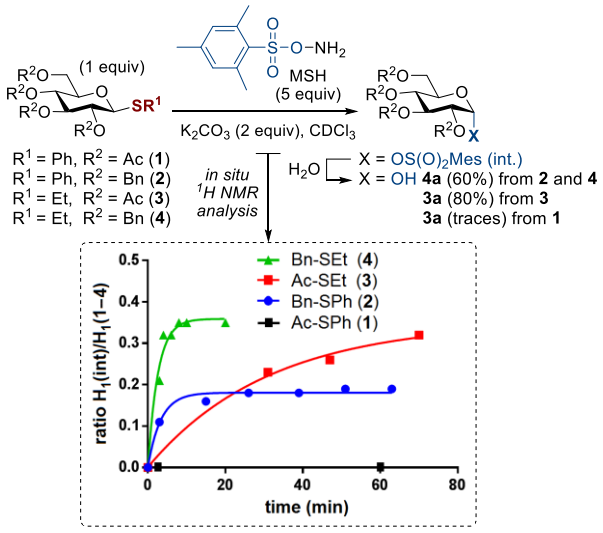
ABSTRACT: A method for the selective activation of thioglycosides that uses the N⁺-thiophilic reagent *O*-mesitylenesulfonylhydroxylamine (MSH) as a promoter is presented. The reaction proceeds *via* anomeric mesitylensulfonate intermediates, which could be isolated and fully characterized by placing a fluorine atom at the C-2 position. In the presence of a soft Lewis acid, glycosylation reaction proceeds at ambient temperature with good yields. It is further demonstrated that it is possible to orthogonally activate *S*-ethyl in the presence of *S*-phenyl donors enabling the design of sequential glycosylation strategies.

Carbohydrates represent one of the largest groups of key biomolecules since they are involved in many essential biological processes.[1] For a better understanding of their roles in biological systems, as well as for the development of carbohydrate-based therapeutics and vaccines,[2] it is key to access chemically defined oligosaccharides. However, their isolation from natural sources in pure form is difficult. Thus, efforts have been devoted to the development of efficient methods that allow their controlled synthesis.[3] While many methods are available to perform glycosylation reactions, their outcome is largely dependent on a number of factors, including reactant concentrations, nature of protecting groups, promoter, solvent effects or the presence of counter ions/additives.[4] Thioglycoside donors are often used in glycosylation reactions since they are stable under various conditions and allow the ready manipulation of existing protecting groups. Furthermore they are easily activated with thiophilic promoters (soft Lewis acids) such as heavy metal salts, halonium/organosulfur reagents or by single electron transfer methods.[5] Despite their enormous potential, the selective activation of *S*-alkyl *vs.* *S*-aryl donors (or *vice versa*), resulting in orthogonal glycosylation reactions is scarce.[6] In this context, the choice of a suitable promoter able to differentiate between the subtle electronic properties of alkyl *vs.* aryl thioglycoside donors is critical for the success of this transformation. We hypothesized that by inverting the normal polarity of the NH_2 group (hard Lewis base) to a soft Lewis acid by using the N⁺-thiophilic reagent *O*-mesitylenesulfonylhydroxylamine (MSH), this would allow the activation of soft alkyl thioglycosides (match scenario) in the presence of the less activated thiophenyl counterparts. MSH reactivity with sulfur species proceeds *via* direct S-to-N nucleophilic attack and typically affords sulfilimine [$R^1R^2(S{=}NH)$] and/or sulfoximine [$R^1R^2S(O)(=NH)$] derivatives. Moreover, it has been shown to promote the oxidative elimination of cysteine to dehydroalanine[7] and the activation of *S*-alkyl thioglycosides.[8] This encouraged us to examine this activation method further because of its potential to be applied in orthogonal glycosylation strategies. We systematically evaluated the ability of MSH to activate a series of

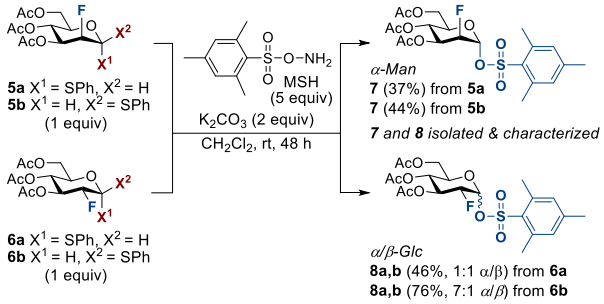
thioglycosyl donors and demonstrated the influence of the leaving group (SEt *vs.* SPh), protecting groups (Ac, Bn), and different configurations (Glc, Gal) using a combination of experimental (intermediate detection and isolation), kinetic (*in situ* ^{1}H NMR), and computational methods (quantum mechanical calculations).

We started our investigation by monitoring the reaction of a series of thioglycosyl donors **1**–**4** with MSH in $CDCl_3$ using *in situ* ^{1}H NMR (Scheme 1). Interestingly, we observed the disappearance of the anomeric proton signal *H*1 at around 4.5 ppm ($J_{1,2}$ ~10 Hz) of starting 1-β-thioglycosides **2**–**4** and the appearance of a new set of signals tentatively assigned to a common α-1-*O*-sulfonylmesitylene intermediate with the anomeric proton *H*1 shifted downfield to ~5.9 ppm ($J_{1,2}$ ~4 Hz), which upon hydrolysis from residual water ultimately results in the formation of corresponding hemiacetals **4a** (60% from **2** and **4**) and **3a** (80% from **3** and traces from **1**). Similar glycosyl sulfonate intermediates have also been described by Bennett (tosyl)[9] and Taylor (mesyl).[10] Indeed, no *syn*-elimination by-products, typically obtained with MSH,[11] were detected under the conditions tested. We found a reactivity profile (Bn-SEt>Bn-SPh>Ac-SEt>Ac-SPh) that correlates with a primary protecting group-based armed-disarmed effect (Ac *vs.* Bn)[12] with a leaving group contribution (SEt *vs.* SPh).[13] Moreover, our findings indicate the SEt group is readily activated with MSH probably *via* charged sulfonium ion intermediates [$^+$S(NH$_2$)Et], whilst SPh activation involves a first step to form a "latent" [$^+$S(NH$_2$)Ph][14] species that temporary protects the leaving group. This moiety only evolves to the activation product in a second, irreversible step upon addition of a base (K_2CO_3) probably *via* the neutral sulfilimine [R^{1}R^2(S=NH)], which is indeed structurally similar to an imidate [R^{1}O(C=NH)R^2] [15] and can be considered the *N*-version of a sulfoxide (S=NH *vs.* S=O).

Scheme 1. *In situ* ^{1}H NMR analysis of the activation of thioglycosides 1–4 with MSH.



Scheme 2. Activation of 2-deoxy-2-fluoro-1-thioglycosides 5a,b and 6a,b with MSH.



Next, to gain further insight into the nature of the proposed intermediates, we decided to perform the same experiments using 2-deoxy-2-fluoro-thioglycosides[16] with D-manno **5** and D-gluco **6** configurations to substantially increase their stability (Scheme 2).[17] Unlike other examples using non-fluorinated thioglycosides,[18] the activation of **5** and **6** proceeded smoothly regardless the anomer used (and without *syn*-elimination) and the resulting intermediates were purified by SiO_2 flash column chromatography and fully characterized. While both 2-F-mannose derivatives **5a,b** afforded α-1-*O*-sulfonylmesitylene intermediate **7** (37–44%) as the sole anomer, activation of 2-F-gluco **6a** gave **8a,b** (46%, 1:1 α/β) and **6b** furnished **8a,b** (76%, 7:1 α/β). Moreover, to further demonstrate 1-*O*-Mes-intermediates are competent in glycosylation reactions, **8a,b** (7:1 α/β) was treated with Cu(OTf)$_2$, 3 Å molecular sieves (MS), and MeOH in dry CH_3CN at room temperature for 16 h to afford complete conversion to a 1:1 inseparable mixture of expected β-methyl glycoside **S1** together with β-methyl 6-OH by-product **S2**, arising from partial deprotection of the 6-OAc moiety in **S1** under the conditions tested (see Supporting Information (SI)). These results reinforce our hypothesis that 1-*O*-sulfonylmesitylene intermediates are also involved in the non-fluoro series. The superior stability of 2-deoxy-2-fluoro-1-*O*-sulfonylmesitylene intermediates compared to their 2-oxygenated counterparts could be tentatively explained by a stronger hyperconjugative effect, particularly in the 2-F-mannose derivative **7** (Figure 1), and/or the unfavored formation of fluorinated oxonium intermediates. However, natural bond orbital (NBO) quantum mechanical calculations on the 1-*O*-sulfonylmesitylene intermediates derived from **1**, **5**, and **6** did not reveal a significant difference on either the anomeric or gauche effects (see SI). Nevertheless, transition state calculations on abbreviated models reproduced the higher reactivity of non-fluorinated intermediates towards hydrolysis (Figure 1). Hence, transition states (TS) α-Me$_4$Glc-1-OMs_TS$_{hyd}$ (related to derivatives **2** and **4**) and α-Ac$_4$Glc-1-OMs_TS$_{hyd}$ (related to derivatives **1** and **3**), were calculated to be ~4 kcal mol^{-1} lower in energy than fluorinated counterparts α-2-F-Ac$_3$Man-1-OMs_TS$_{hyd}$ (related to derivatives **5a** and **5b**) and α-2-F-Ac$_3$Glc-1-OMs_TS$_{hyd}$ (related to derivatives **6a** and **6b**), thus making the reaction ~850 times faster. In such studies, different explicit solvation models were evaluated, and at least two water molecules were necessary to locate the hydrolysis transition structures (TS).

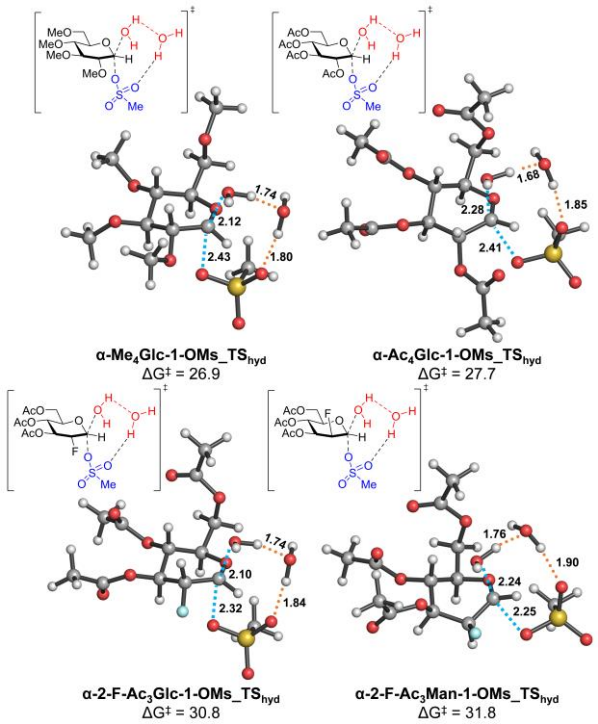
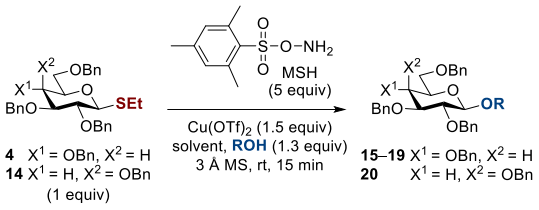


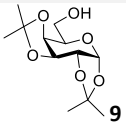
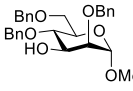
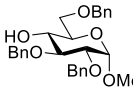
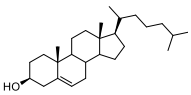
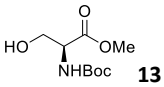
Figure 1. Transition structures calculated with PCM(CH_2Cl_2)/M06-2X/6-31G(d,p) level for the hydrolysis of 1-*O*-sulfonylmesitylene (Mes) intermediates. Models for the α-1-*O*-Mes-anomers with two reacting water molecules are shown. Activation free energies ($\Delta G^{\ddagger}$) are in kcal mol^{-1}.

These TS's involve an asynchronous concerted C1–OS bond cleavage and C1–OH bond formation in which one additional water molecule assists proton transfer to the released methanesulfonic acid. The presence of the 2-F atom in equatorial position (D-Glc) destabilizes the partial positive charge developing at the C1 carbon of the TS; this makes the hydrolysis TS to be earlier than the 2-OMe and 2-OAc analogues in terms of the cleaving C1–OS bond distance and significantly raises the activation barrier. Additionally, when the 2-F substituent is in an axial position (D-Man), the TS adopts a more encumbered, high-energy boat-like geometry to avoid repulsion with the incoming water.

We next evaluated the scope of the MSH-promoted glycosylation using selected acceptors **9**–**13** (Table 1). Surprisingly, we could not observe any glycosylation product using the original activation conditions (MSH, K_2CO_3 in CH_2Cl_2,) probably because of the low reactivity of the α-1-*O*-Mes intermediate towards the attack of poorly reactive *O*-acceptors under the conditions tested. Performing the reaction under S_N2 conditions upon generation of the intermediate and using the more nucleophilic alkoxide from **9** (NaH, 15-crown-5 in 1,4-dioxane) furnished **15** in very low yields (<5%) as was also the case for the corresponding 1-*S*-Ac product **S3** (35%) when the "soft" KSAc was used (18-crown-6 in CH_2Cl_2) (see SI). Nevertheless, these experiments suggest the intermediacy of a covalent α-1-*O*-Mes intermediate in the absence of external additives. We first screened reaction conditions including commonly used promoters/additives (AgOTf, $Cu(OTf)_2$, and $LiClO_4$),[19] α- *vs.* β-selective solvents (CH_2Cl_2, Et_2O, and CH_3CN), and reaction temperature (0 ºC *vs.* room temperature) (entries 1–5).

Table 1. Reaction scope[a]



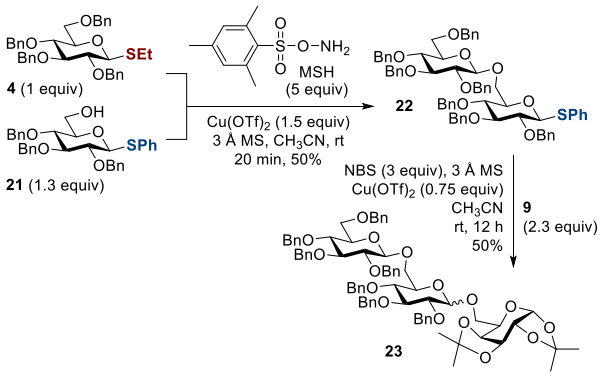
entry	donor	ROH	solvent	product	yield (%)[b]	α/β ratio[c]
1[d]	**4**	 **9**	CH_2Cl_2	**15**	26	1.3:1
2	**4**	**9**	Et_2O	**15**	34	1.7:1
3[e]	**4**	**9**	Et_2O	**15**	54	5:1
4[e,f]	**4**	**9**	Et_2O	**15**	40	2.5:1
5	**4**	**9**	CH_3CN	**15**	71	1:5
6[g]	**4**	**9**	CH_3CN	**15**	41	1:1.7
7[h]	**4**	 **10**	toluene	**16**	40	>20:1[i]
8	**4**	 **11**	toluene	**17**	22	1.2:1
9	**4**	 **12**	CH_3CN	**18**	35	1:3.7
10	**4**	 **13**	CH_3CN	**19**	50	1:2.2
11	**14**	**9**	CH_3CN	**20**	50	1:2.9
12	**14**	**9**	Et_2O	**20**	26	2.5:1

[a]General conditions: 1-thioglycoside donors **4**, **14** (1 equiv), ROH (1.3 equiv), MSH (5 equiv), $Cu(OTf)_2$ (1.5 equiv) and 3 Å MS in dry solvent (0.01 M) unless otherwise indicated. [b]Isolated yield. [c]Determined by integration of the anomeric proton signals in the ^1H NMR spectrum of the crude reaction mixture. [d]AgOTf (4 equiv) used as a promoter. [e]$LiClO_4$ (1 equiv) used as an additive. [f]Conducted at 0 ºC for 6 h. [g]$Cu(OTf)_2$ (1 equiv). [h]The solvent was further optimized for secondary glycosyl acceptors (see SI). [i]Only the α-anomer was detected after purification by SiO_2 flash column chromatography.

The best results were obtained with stoichiometric amounts of $Cu(OTf)_2$, which has been suggested to act as an "extra" triflate source promoting a OMes to OTf exchange, especially with >1 equiv (entry 5 *vs.* 6).[20] The reaction can be performed at ambient temperature and it is typically complete after only 15 min.[21] Notably, control experiments demonstrate that a successful glycosylation necessitates MSH to be added to a mixture of donor/$Cu(OTf)_2$ (see SI). Next, the acceptor scope was expanded to secondary glycosyl acceptors **10** (D-Man), **11** (D-Glc) as well as models of natural aglycones **12** and amino acids **13** to afford **16** (40%), **17** (22%), **18** (35%), and

19 (50%) (entries 7–10). 1-Thioglycosyl donor with D-Gal configuration **14** was also tested; it provided **20** in moderate yield (up to 50%) and α/β-selectivity (1:2.9 in CH_3CN and 2.5:1 in Et_2O) (entries 11 and 12) as expected for donors bearing non-participating groups at C-2. Finally, we designed a proof-of-principle glycosylation strategy that enabled the preparation of a trisaccharide, which took advantage of the orthogonal activation of SEt over SPh donors with MSH (Scheme 3). Thus, a mixture of **4** and **21** was treated with MSH/$Cu(OTf)_2$ under our optimized conditions to afford disaccharide **22** (50% after SiO_2 flash column chromatography). The successful activation of the more reactive SEt group in **4** gave **22** while the SPh group of **21** remained intact. Finally, **22** was converted to the model Glc(1→6)Glc(1→6)Gal trisaccharide **23** (50%, 1:1 α/β) by activation of the remaining SPh group with NBS/$Cu(OTf)_2$, thus demonstrating the orthogonal activation of SEt over SPh leaving groups with MSH at ambient temperature. This might find useful applications in one-pot oligosaccharide synthesis.

Scheme 3. Sequential preparation of trisaccharide 19



In summary, the selective activation of different 1-thioglycoside donors by the N^+-thiophilic reagent *O*-mesitylenesulfonylhydroxylamine (MSH) as a promoter has been thoroughly studied. The resulting 1-*O*-sulfonylmesitylene intermediate species were detected by ^1H NMR for monosaccharides **2**–**4** and isolated/characterized in the presence of a fluorine atom at C-2 in the D-mannose and D-glucose series. We showed that MSH is the thiophilic species, but a soft Lewis acid such as $Cu(OTf)_2$ is necessary for a successful glycosylation reaction. Furthermore, a proof-of-principle study demonstrated the specific activation of anomeric *S*-ethyl leaving groups in the presence of *S*-phenyl groups and this enabled the sequential preparation of a trisaccharide. Since this differentiation can be performed at ambient temperature, this protocol may find utility for one-pot oligosaccharide synthesis.

ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website.

Detailed experimental procedures and characterization data (PDF)

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Notes

The authors declare no competing financial interest.

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