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Synthesis of carbohydrate derivatives using solid-phase work-up and scavenging techniques

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Methods to allow the clean preparation of oligosaccharides were investigated using techniques that do not require conventional column chromatography or an aqueous work-up. The route was designed to provide rapid access to oligosaccharides and is suitable for automation and parallel library formation. The research has focused on the glycosidations of a range of glycosyl acceptors with various selenophenyl glycosyl donors using iodine as an activator in the presence of DTBMP, a hindered organic base. Hydroxyl-containing contaminants were removed by scavenging with polymer-supported tosyl chloride.

The abundance of carbohydrates in nature and their diverse involvement in biological systems make them attractive molecules for chemical and biological research. However, due to the large number of monomer possibilities, stereocontrol at the anomeric carbon, arduous protecting group strategies, and hydrolysis by-products, the synthesis of oligosaccharides is still an extremely complicated process. The aim of this work has been to investigate facile methods for oligosaccharide synthesis by exploiting the benefits of both solution-phase reactions and solid-phase work-ups using polymer-supported scavengers.

Advantages of utilizing these methods[1] in lieu of polymer-supported substrates[2] are elimination of additional steps due to compound linkage and cleavage from the polymer, compatibility of reaction monitoring with conventional solution-phase methods like TLC, NMR, HPLC, and LCMS, possibility of convergent synthesis, and ease of work-up by filtration. It would also be beneficial to develop these clean techniques that do not require conventional column chromatography or aqueous work-up to allow the possibility of automation, parallel library formation, and combinatorial synthesis. Kirschning and co-workers[3] incorporated polymer-bound iodate(I) complexes to activate thioglycosides for the preparation of deoxyglycosides. Kirschning[4] also synthesised deoxyglycosides by using a polymer-supported bis(acetoxy)-iodate(I) complex to transform glycals into 2-iodoglycosyl acetates which were then activated by polymer-supported silyl triflate.

In this work, we have focused on selenophenyl glycosides due to their overall stability and versatility as glycosyl donors.[5] Selenophenyl glycosides are similar to thioethyl glycosides in their activation steps, but are more reactive to most activation systems. One set of conditions investigated for glycosidations with selenophenyl glycosides as donors was iodine in the presence of anhydrous potassium carbonate. Field and co-workers[6] first studied this system for thioglycosidic donors in order to minimize the number of reagents present in the reaction mixture. They reacted a variety of acceptors with different thioglycosides activated by iodine using potassium carbonate to remove any hydroiodic acid that was formed in the process. An aqueous work-up of these afforded clean products as mixtures of anomers. Here, we focus on the work-up of these reactions by a protocol that allows removal of side products including unreacted free hydroxyl groups. This methodology was also applied for work-ups of glycosidations with a thioethyl mannopyranoside also activated by iodine and a mannopyranosyl fluoride activated by $AgOTf–Cp_2HfCl_2$.[7] The procedure is designed for parallel synthesis of carbohydrates using polymer-supported scavengers or solid-phase extraction methods.

Side products in these selenophenyl glycosidations are diphenyl diselenide and iodine, which are formed when two molecules of phenylselenyl iodide react. A similar occurrence is observed with phenylthiol glycosides which produce diphenyl disulfide upon glycosidation. Kirschning[8] incorporated a borohydride exchange resin to convert diphenyl disulfide into thiophenol which can then be scavenged by a sulfide resin. In our work, several conditions for scavenging the diphenyl diselenide were attempted, although none of the systems scavenged efficiently enough to be widely applicable. It was discovered, however, that solid-phase extractions remove diphenyl diselenide simply by flushing of the reaction mixture through a commercially available pre-packed Bond Elut™ silica cartridge, obtained form Varian, with either dichloromethane or hexanes. All carbohydrate-containing compounds remained on the solid support until they were easily eluted with ethyl acetate or methanol.

During glycosidation reactions, hydrolysis of the glycosyl donor is often observed. This results in hydrolysed donor products and unreacted acceptor being present in the product solution, a fairly typical occurrence in glycosidation reactions. In order to remove these free hydroxyl containing side products, polymer-supported tosyl chloride (PS-TsCl) was used as a scavenger in the presence of pyridine and 4-dimethylaminopyridine (DMAP) in dichloromethane. After filtration, the desired glycosidation product was obtained in high purity.

The reaction conditions described by Field and co-workers[6] were incorporated with the new work-up procedure when the C6 position of the glycosyl donor was protected with a *t*-butyldimethylsilyl (TBDMS) group. This was intended to be the first step in the synthesis of a trisaccharide. Reaction with seleno-α-D-mannopyranoside **1** as the glycosyl donor and glycosyl acceptor **2** was performed using iodine in the presence of potassium carbonate. During glycosidation, some TBDMS group removal was observed as described by Field,[9] presumably due to inefficient neutralization of iodic acid. An anomeric ratio of 3 : 1 (α : β) was obtained while the yield was only 78% due to the deprotected product **4** being scavenged with the other side products containing free hydroxyl groups, including unreacted acceptor **2** and hydrolysed donor **3** (Scheme 1).

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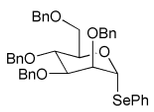
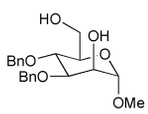
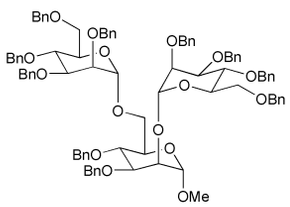
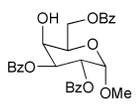
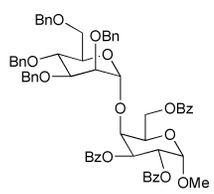
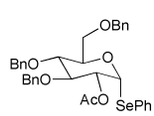
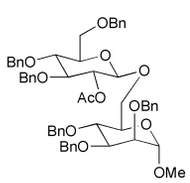
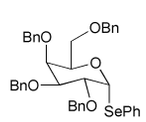
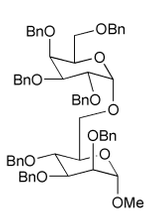
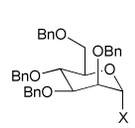
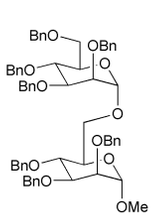
Table 1 Glycosyl donors and acceptors applied to this method

Donor	Acceptor	Product	Yield
 **6**	 **11**	 **13**	96%[a]
6	 **12**	 **14**	[b]
 **7**	**2**	 **15**	75% all β
 **8**	**2**	 **16**	88% all α
 **9** X = SEt **10** X = F	**2**	 **17**	**9** = 73% 4.4 : 1 (α : β) **10** = 84% 1.5 : 1 (α : β)

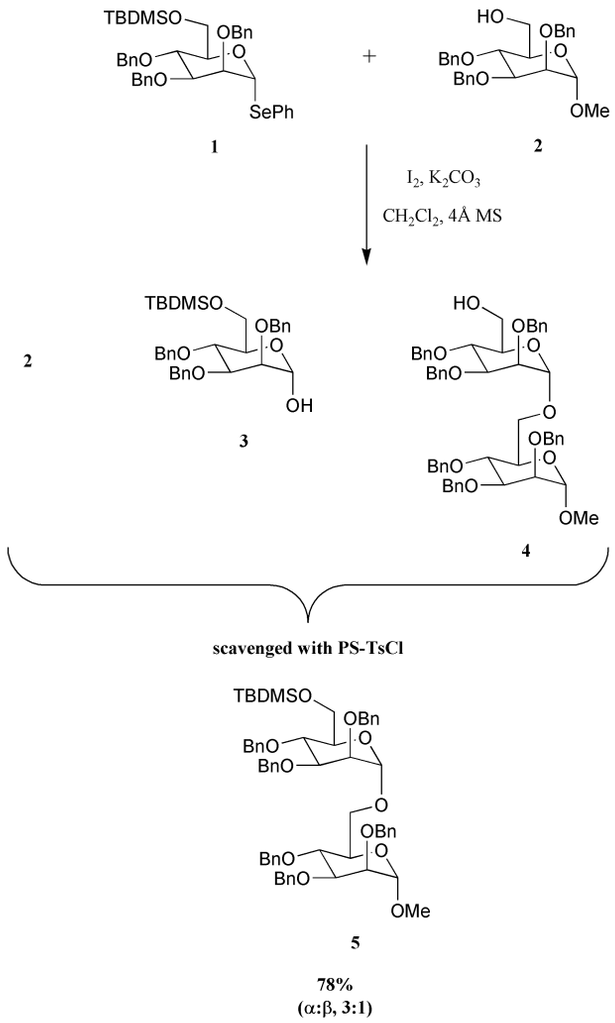
[a] Could not determine anomeric ratios. [b] Product could not be separated from by-products.

Other bases, pyridine and 2,6-lutidine, were studied in an attempt to discover a more effective acid neutralizer than potassium carbonate. No TBDMS group removal was detected when pyridine was used, but most of the donor that did not react with acceptor **2** was observed as a complex with pyridine. Although 2,6-lutidine is a less nucleophilic base and no hydrolysed donor was observed, the reaction was not complete after 24 hours. The only successful base integrated into this reaction was 2,6-di-*t*-butyl-4-methylpyridine (DTBMP). The disaccharide product **5** was obtained in 98% yield with an anomeric ratio of 3 : 1 (α : β).[10] This methodology has been applied to various glycosidations (Table 1).

A branched trisaccharide **13** was synthesised using a dihydroxy acceptor **11** in high yield as an anomeric mixture. The anomeric ratios at the anomeric carbons could not be determined due to the complexity of the NMR spectra. When the glycosyl acceptor **12** was deactivated by benzoyl protecting groups, the yield could not be determined; coupling was effective by LCMS, but the unreacted, deactivated acceptor **12** could not be scavenged with PS-TsCl. Fully deactivated galactose donor gave no coupling while C2 deactivated glucose **7** coupled well to give pure β-product. Fully activated galactose donor **8** produced only α-product in 88% yield. Ethylthiol mannopyranosyl donor **9** reacted in 73% yield with a good anomeric ratio. However, this donor took longer to react than the phenyl selenides. Mannopyranosyl fluoride donor **10** coupled in 84% yield. The best anomeric ratio of 1.5 : 1 was only obtained when the glycosidation was done at −78 °C. The reagents ($AgOTf–Cp_2HfCl_2$) were removed by the Bond Elut™ cartridge.

The trisaccharide **18** was synthesized by deprotection of the TBDMS protected disaccharide **5** with Amberlyst-15 (A-15) in methanol. The deprotected disaccharide **5** was obtained by simple filtration in 92–99% yield. This acceptor **4** was reacted with donor **6** using the afore described techniques to produce the trisaccharide **18** in 89% yield (Scheme 2). The product was obtained as a mixture of anomer combinations.

In summary, polymer-supported scavengers in combination



Scheme 1 Glycosidation using I_2 as an activator in the presence of K_2CO_3.

with solid-phase extractions allow the clean synthesis of oligosaccharides without conventional column chromatography or aqueous work-up. We plan to develop fully automated, robotic syntheses utilizing this method for the synthesis of carbohydrate libraries.

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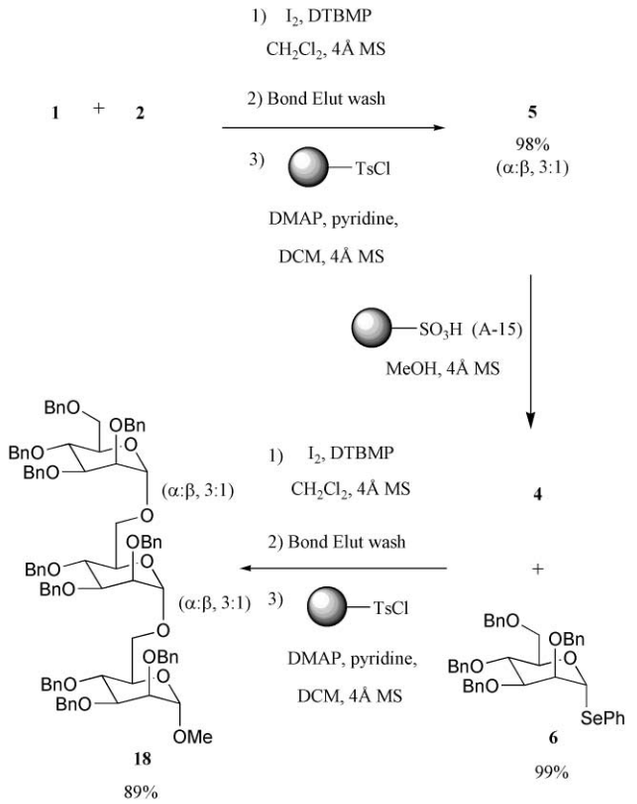
References

1 (*a*) S. V. Ley, I. R. Baxendale, R. N. Bream, P. S. Jackson, A. G. Leach, D. A. Longbottom, M. Nesi, J. S. Scott, R. I. Storer and S. J. Taylor, *J. Chem. Soc., Perkin Trans. 1*, 2000, **23**, 3815–4196; (*b*) A. Kirschning, H. Monenschein and R. Wittenberg, *Angew. Chem., Int. Ed.*, 2001, **40**, 650–679.

2 For examples of oligosaccharides synthesised on solid support see (*a*) P. H. Seeberger, *Solid Supported Oligosaccharide Synthesis and Combinatorial Carbohydrate Libraries*, Wiley, New York, 2001; (*b*) H. M. I. Osborn and T. H. Khan, *Tetrahedron*, 1999, **55**, 1807–1850.

3 For a recent, excellent example in oligosaccharide assembly, see J. Jaunzems, G. Sourkouni-Argirusi, M. Jesberger and A. Kirschning, *Tetrahedron Lett.*, 2003, **44**, 637–639.



Scheme 2 Trisaccharide synthesis using I_2 an activator in the presence of DTBMP.

4 A. Kirschning, M. Jesberger and A. Schonberger, *Org. Lett.*, 2001, **3**, 3623–3626.

5 (*a*) B. Fraser-Reid, P. Konradsson and U. E. Udodong, *Tetrahedron Lett.*, 1990, **31**, 4313; (*b*) J. H. van Boom, G. H. Veeneman and S. H. van Leeuwen, *Tetrahedron Lett.*, 1990, **31**, 275; (*c*) J. Pietruszka, S. V. Ley, P. Grice, H. W. M. Priepke and E. P. E. Walther, *Synlett*, 1995, 781–784.

6 R. A. Field, K. P. Ravindranathan Kartha, P. Cura, M. Aloui, K. S. Readman and T. J. Rutherford, *Tetrahedron: Asymmetry*, 2000, **11**, 581–593.

7 K. Suzuki, K. Maeta, T. Matsumoto and G. Tsuchihashi, *Tetrahedron Lett.*, 1988, **29**, 3567–3570.

8 J. Jaunzems, E. Hofer, M. Jesberger, G. Sourkouni-Argirusi and A. Kirschning, *Angew. Chem., Int. Ed.*, 2003, **42**, 1166–1170.

9 R. A. Field and K. P. Ravindrathan Kartha, *Synlett*, 1999, **3**, 311–312.

10 *Glycosidation procedure*: Donor **1** (423 mg, 0.61 mmol) and acceptor **2** (278 mg, 0.6 mmol) were combined with DTBMP (464 mg, 1.83 mmol) and 4 Å molecular sieves in CH_2Cl_2 with stirring under argon. After 15 minutes, iodine (155 mg, 0.61 mmol) was added. After 2.75 hours, the reaction was quenched with solid sodium thiosulfate, filtered through a Varian Bond Elut™ cartridge (10 g, 60 ml), washed with 50 ml each of CH_2Cl_2, EtOAc, and MeOH consecutively. The EtOAc fraction was concentrated *in vacuo*. *Scavenging procedure*: The mixture was dissolved in CH_2Cl_2 (10 ml) and combined with DMAP (6.3 mg, 0.051 mmol), pyridine (2 ml), PS-TsCl (1.01 g, 1.53 mmol g^{-1}), and 4 Å molecular sieves. After being shaken overnight under argon, the reaction mixture was filtered through Celite™ and washed with CH_2Cl_2. The resulting filtrate was concentrated *in vacuo*. *Deprotection procedure*: The clean TBDMS protected disaccharide **4** was then dissolved in MeOH (5 ml) and A-15 (1.29 g, 1.0 mmol g^{-1}) was added. Shaking was maintained for an hour under argon before the reaction mixture was filtered through Celite™ and washed with CH_2Cl_2. The resulting filtrate was concentrated *in vacuo* to give the deprotected disaccharide **5**. The glycosidation with donor **8** and scavenging were repeated to give the trisaccharide product **7**.