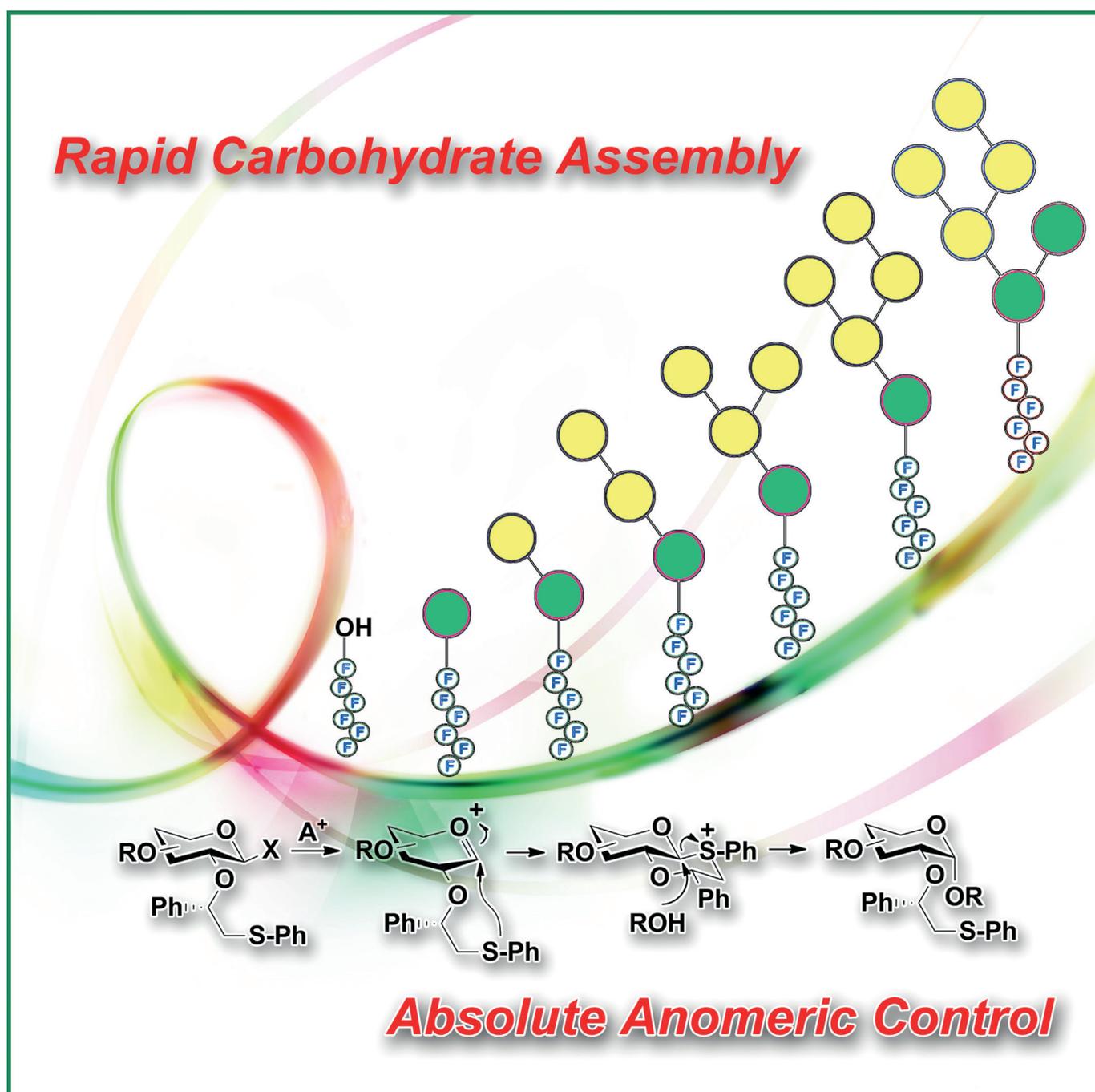


Synthetic Methods

Assembly of a Complex Branched Oligosaccharide by Combining Fluorous-Supported Synthesis and Stereoselective Glycosylations using Anomeric Sulfonium Ions

Wei Huang,^[a, b] Qi Gao,^[a] and Geert-Jan Boons^{*[a, b]}



Abstract: There is an urgent need to develop reliable strategies for the rapid assembly of complex oligosaccharides. This paper presents a set of strategically selected orthogonal protecting groups, glycosyl donors modified by a (*S*)-phenylthiomethylbenzyl ether at C-2, and a glycosyl acceptor containing a fluoros tag, which makes it possible to rapidly prepare complex branched oligosaccharides of biological importance. The C-2 auxiliary controlled the 1,2-*cis* anomeric selectivity of the various galactosylations. The orthogonal protecting groups, 2-naphthylmethyl ether (Nap) and levulinic ester (Lev), made it possible to generate glycosyl acceptors and allowed the installation of a crowded branching

point. After the glycosylations, the chiral auxiliary could be removed using acidic conditions, which was compatible with the presence of the orthogonal protecting groups Lev and Nap, thereby allowing the efficient installation of 1,2-linked glycosides. The light fluoros tag made it possible to purify the compounds by a simple filtration method using silica gel modified by fluorocarbons. The set of building blocks was successfully employed for the preparation of the carbohydrate moiety of the GPI anchor of *Trypanosoma brucei*, which is a parasite that causes sleeping sickness in humans and similar diseases in domestic animals.

Introduction

It is now well-established that a dense layer of complex carbohydrates covers the surface of all prokaryotic and eukaryotic cells. These carbohydrates have been implicated in a wide range of biological processes such as protein folding, fertilization, embryogenesis, host-guest interactions, and cell differentiation and mobility.^[1] In addition, overwhelming data supports the relevance of glycosylation in pathogen recognition, inflammation, innate immunity, and the development of autoimmunity and cancer.^[2] Although the importance of cell-surface carbohydrates in health and disease is widely appreciated, advances in glycoscience have been slow, due to the staggering complexity of the glycome.^[3] This complexity makes it difficult to define glycan structures expressed by a given cell type and complicates the identification of specific glycan recognition determinants of glycan-binding proteins.^[4] Libraries of well-defined glycans will make it possible to address these difficulties.

The need for diverse collections of complex glycans has stimulated the development of fast and convenient methods for their synthesis.^[5] For example, several synthetic strategies make it possible to assemble complex oligosaccharides from carefully selected monosaccharide building blocks using a minimal number of chemical steps.^[6] Among these strategies, one-pot multi-step glycosylations, in which several glycosyl donors are sequentially reacted in the same flask, are particularly attractive and can furnish target oligosaccharides without the need for protecting group manipulations and isolation and purification of synthetic intermediates.^[6c] Within the past few years, automated solid-phase oligosaccharide synthesis has

also advanced substantially.^[7] A host of glycosylating agents, new linker systems, different solid supports, and a variety of protecting groups have been carefully evaluated and these efforts have resulted in the first commercially available glycan synthesizer.

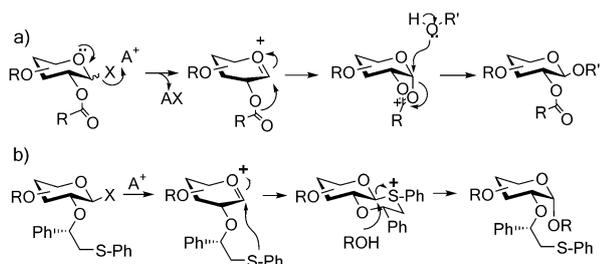
Soluble light fluoros tags offer another attractive means to simplify the process of oligosaccharide synthesis. In this case, tagged carbohydrates can easily be separated from nonfluorous-tagged side products by solid-phase extraction using silica gel modified by fluorocarbons.^[8] This generic procedure, which more closely resembles filtration than chromatography, depends primarily on the presence or absence of a fluoros tag and not on the polarity or other molecular features of the compound. Unlike solid-phase supported synthesis, light fluoros technology does not require large excesses of reagents to drive the reactions to completion. Fluorous-tagged compounds can easily be analyzed by standard spectroscopic methods, thereby providing control over the synthesis. Furthermore, efforts are underway to develop a liquid handler to automate fluoros supported oligosaccharide synthesis.^[9] Several fluoros versions of protecting groups have been developed for a variety of functional groups, and thus tags can easily be installed.^[10] Additionally, it is possible to array fluoros-tagged glycans, thereby eliminating the necessity to install reactive functional groups for glycan immobilization.^[11]

Despite the promise of fluoros-supported oligosaccharide synthesis, it has mainly been employed for the preparation of relatively simple linear compounds.^[10] This limited application is most likely due to the difficulties of controlling anomeric selectivities in glycosylations and challenges to install branching points in high yield.^[5a,6a,11] In this respect, many complex oligosaccharides are branched and, due to steric crowding, the corresponding glycosylations are often low-yielding. Furthermore, 1,2-*trans*-glycosides, such as β -glucosides and β -galactosides, can reliably be introduced by neighboring-group participation of an ester-protecting group at C-2 of a glycosyl donor (Scheme 1 a). On the other hand, the installation of 1,2-*cis* glycosidic linkages, such as α -glucosides and α -galactosides, requires glycosyl donors that have a non-assisting functionality at C-2, and often these coupling reactions result in mixtures of anomers.^[5a,11] Low-yielding glycosylations and the formation of

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Scheme 1. Control of anomeric selectivity in glycosylations. a) Neighboring-group participation by C-2 esters to give a five-membered ring oxocarbenium ion intermediate to form selectively 1,2-*trans*-glycosides. b) Neighboring group participation by chiral auxiliary to give a *trans*-decalin anomeric sulfonium ion intermediate to provide 1,2-*cis*-glycosides.

anomers defeat the purpose of fluoros support synthesis that relies on simple filtration protocols for purification.

Recently, we introduced a stereoselective glycosylation approach based on neighboring-group participation by a (*S*)-phenylthiomethylbenzyl moiety at C-2 of a glycosyl donor, which can readily provide 1,2-*cis*-glycosides (Scheme 1 b).^[12] Upon activation of the donor and formation of an oxocarbenium ion, the thiophenyl moiety of the C-2 auxiliary participates, resulting in the formation of an intermediate sulfonium ion having a *trans*-decalin configuration. This stereoisomer is strongly favored because of the absence of unfavorable gauche interactions. Furthermore, the alternative *cis*-decalin system places the phenyl-substituent in an axial position, thereby inducing unfavorable steric interactions. Displacement of the anomeric sulfonium ion by a sugar alcohol then results in the formation of a 1,2-*cis*-glycoside.

We describe here that the use of glycosyl donors modified by a C-2 (*S*)-phenylthiomethylbenzyl ether or ester-protecting group to stereoselectively introduce 1,2-*cis*- or 1,2-*trans*-glycosides, respectively, and glycosyl acceptors modified by a fluoros tag can readily provide highly complex branched oligosaccharides of biological importance. The strategy was applied to the preparation of the carbohydrate moiety of the GPI anchor of *Trypanosoma brucei* (Figure 1), which is the parasite that causes sleeping sickness in humans and similar diseases in domestic animals.^[13] The oligosaccharide is composed of a branched tri-mannoside core, which is a structurally conserved motif of GPI anchors of many different organisms. It is further elongated by α -galactosides that are unique to *T. brucei*. It is expected that synthetic carbohydrates of different compositions will aid in the development of therapeutics and diagnostic for infections caused by this pathogen.^[14] Previous attempts to prepare such oligosaccharides entailed low yielding galactosylations and provided anomeric mixtures.^[15]

Results and discussion

The synthesis of building blocks

We envisaged that building blocks 2–7 and fluoros-tag-modified benzyl alcohol 8 (Figure 1) would make it possible to assemble target compound 1. Levulinic ester (Lev)^[16] and 2-

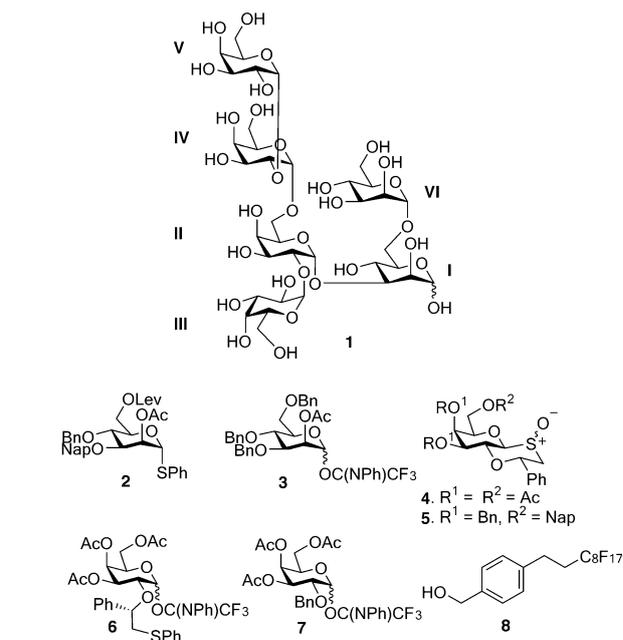
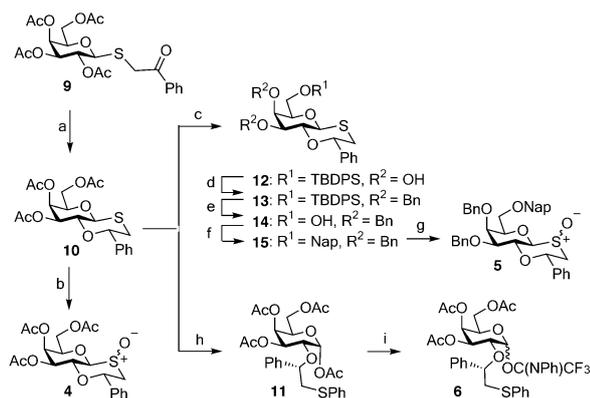


Figure 1. The structure of hexasaccharide 1 of the GPI anchor of *T. Brucei* and the monosaccharide building blocks required for its assembly.

Naphthylmethyl ether (Nap)^[17] were employed as a convenient set of orthogonal protecting groups for glycosyl acceptor formation and branching-point installation. The donors 2 and 3, having participating esters at C-2, were used to install the mannosyl moieties. Furthermore, it was anticipated that galactosyl donors 4–6, having a chiral auxiliary at C-2, could be employed for the stereoselective introduction of the challenging α -galactosides.

First, attention was focused on the preparation of galactosyl donors 4–6 (Scheme 2). It was expected that activation of a trifluoro-*N*-phenyl imidate of 6 would result in the formation of an oxocarbenium ion which undergoes neighboring-group participation by the (*S*)-(phenylthiomethyl)benzyl ether leading to a 1,2-*trans* anomeric sulfonium ion. Nucleophilic displacement of the anomeric sulfonium ion by a sugar alcohol will then provide an α -galactoside.^[12a] Alternatively, arylation of the 1,2-oxathiane of compounds such as 4 and 5 will also provide anomeric sulfonium ions and such a transformation can easily be accomplished by activation the sulfoxide with triflic anhydride followed by reaction with 1,3,5-trimethoxybenzene.^[18] An attractive feature of the 2-oxathianes is that they can be converted into compounds such as 7 by treatment with benzyne, which leads to a derivative having a (*S*)-(phenylthiomethyl)benzyl ether at C-2 and an acetate at the anomeric center.^[19] Standard procedures can then be employed to install an anomeric imidate for glycosylations.^[20] Thus, it was anticipated that 2-oxathiane 11 would be an appropriate precursor for the synthesis of glycosyl donors 4–6.

Thus, thioglycoside 9 was prepared by sequential treatment of per-*O*-acetyl-galactose with thiourea and 2-bromoacetophenone. The acetyl esters of 9 were cleavage with sodium methoxide in methanol and the resulting tetraol was treated with



Scheme 2. Preparation of building blocks for the GPI anchor carbohydrate moiety of *T. brucei*. Reagents and conditions: a) MeONa, MeOH, RT, 1 h, then *p*-TSA, MeOH, RT, 18 h; then acetic anhydride, pyridine, RT, 3 h, 73% (for 3 steps), then TiCl₄, Et₃SiH, DCM, 0 °C, 8 h, 83%; b) *m*-CPBA, DCM, −15 °C, 30 min, 96%; c) NaOMe, MeOH, RT, 1 h, then TBDPSCI, imidazole, DMF, 0 °C, 2 h, 98%; d) BnBr, NaH, DMF, 0 °C, 1 h, 75%; e) HF-pyridine in pyridine, RT, 18 h, 61%; f) NaH, NapBr, DMF, 0 °C, 5 h, 95%; g) *m*-CPBA, DCM, −15 °C, 30 min, 72%; h) Pb(OAc)₄, 1-aminobenzotriazole, DCM, −78 °C, 1 h, 95%; i) NH₂NH₂-AcOH, DMF, 50 °C, 4 h; then 2,2,2-trifluoro-*N*-phenylacetimidoyl chloride, DBU, DCM, RT, 1 h, 71%.

methanol in the presence of camphorsulfonic acid (CSA) to form a 1,2-oxathiane ketal. Due to the poor solubility of the latter compound, it was not purified and immediately treated with trimethylsilyl trifluoromethanesulfonate (TMSOTf) or BF₃OEt₂ in the presence Et₃SiH to reduce the ketal to a 1,2-oxathiane ether. Although the latter reaction proceeded smoothly for glucose derivatives,^[18a,21] in the case of galactose no reaction occurred. Fortunately, the use of TiCl₄ as the Lewis acid in the presence of Et₃SiH gave, after *O*-acetylation with acetic anhydride in pyridine, the target compound **10** in a yield of 83%. Oxidation of compound **10** using *meta*-chloroperoxybenzoic acid (*m*CPBA) in dichloromethane (DCM) at −15 °C gave the galactosyl donor **4**. Compound **11** was readily prepared by treatment of **10** with 1-aminobenzotriazole and Pb(OAc)₄ to generate benzyne for arylation of the 1,2-oxathiane. The latter compound was treated with hydrazine acetate to remove the anomeric acetate and the resulting lactol was converted into an *N*-phenyl trifluoroacetimidate (**6**) using 2,2,2-trifluoro-*N*-phenylacetimidoyl chloride in the presence of 1,8-diazabicyclo[5.4.0]undec-7-ene (DBU).^[22]

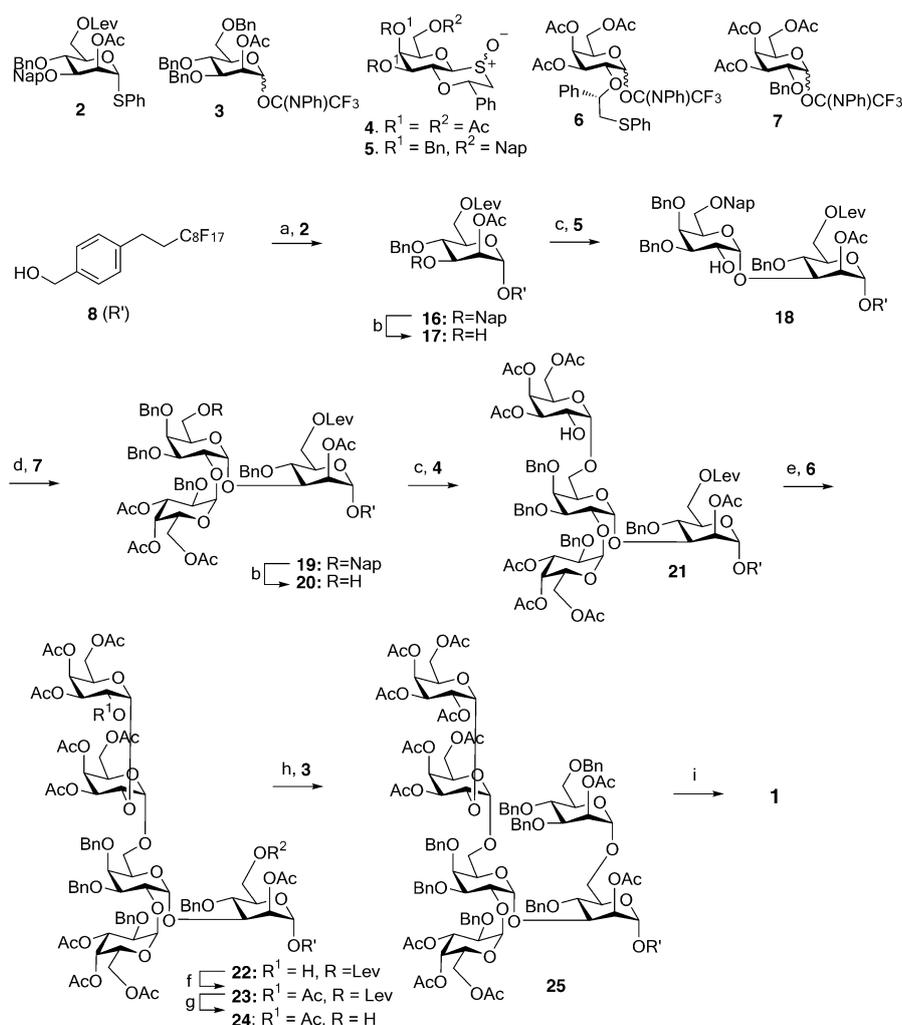
The selectively protected galactosyl donor **5** was synthesized by removal of the acetyl esters of **10** followed by selective silylation of the primary hydroxyl using *tert*-butyl(chloro)diphenylsilane (TBDPSCI) in the presence of imidazole in DMF to give **12**. The latter compound was benzylated under standard conditions (→**13**) followed by removal of the TBDPS ether using HF-pyridine to give **14**, which was converted into Nap ether **15** by alkylation with NapBr in the presence of sodium hydride in dimethylformamide (DMF). Prior to glycosylation, the 1,2-oxathiane **15** was oxidized to the corresponding sulfoxide **5** using *m*CPBA. The mannosyl donors **2** and **3** were prepared by standard protecting group manipulations, as detailed in the Supporting Information.

Assembly of the carbohydrate moiety of the GPI anchor of *T. brucei*

First, target compound **1** was prepared by a conventional purification protocol using silica gel or size exclusion column chromatography (Scheme 3). In this case, each intermediate was carefully characterized by two-dimensional NMR spectroscopy and mass spectrometry. After establishing an appropriate synthetic protocol, the target compound was resynthesized in a rapid manner by employing fluoruous solid phase extraction and, in this case, only the fully assembled oligosaccharide was characterized. The attraction of this approach is that a streamlined synthetic protocol for **1** can easily be adapted for the preparation of many analogs.

Thus, glycosyl donor **2** was coupled with 4-(1*H*,1*H*,2*H*,2*H*-perfluorodecyl)benzyl alcohol (**8**) using *N*-iodosuccinimide (NIS) and triflic acid (TfOH) as the activator^[23] at −25 °C to give, after a reaction time of 30 min, fluoruous-tagged mannoside **16** in high yield. As expected, only the α-anomer was formed due to neighboring-group participation of the acetyl ester of **2**. Next, the Nap ether of **16** was removed by oxidation with 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) in the mixture of DCM and water to give glycosyl acceptor **17**, which was coupled with glycosyl donor **5** to provide, after acid mediated removal of the C-2 auxiliary, disaccharide **18**. In this glycosylation, **5** was arylated by treatment with a stoichiometric amount of triflic anhydride (Tf₂O) and 1,3,5-trimethoxybenzene (TMB) in the presence of 2,6-di-*tert*-butyl-4-methylpyridine (DTBMP) in DCM at −40 °C to form a sulfonium ion intermediate. Next, glycosyl acceptor **17** was added and the reaction mixture was allowed to warm to room temperature and, after a reaction time of 11 h and purification by silica gel column chromatography, a glycoside product was obtained, having a (trimethoxyphenylthiomethyl)benzyl ether moiety at C-2. The latter functionality was cleaved by treatment with 10% trifluoroacetic acid (TFA) in DCM to give glycosyl acceptor **18**. Careful analysis by ¹H NMR spectroscopy confirmed that only the expected α-anomer had formed.

The installation of the α(1,2)-linked galactoside of **1** proved challenging. Preactivation of **4** followed by the addition of acceptor **18** did not lead to glycoside formation. A TMSOTf-mediated coupling of **6** with **18** gave only a trace amount of product, as shown by MALDI-TOF mass spectrometry. The use of 5 equiv of **6** provided the corresponding trisaccharide in a disappointing yield of 25%. We reasoned that the failures of these glycosylations was due to the rather low reactivity of C-2 hydroxyl of **18** and the bulky nature of the C-2 auxiliary of glycosyl donors **4** and **6**.^[24] Therefore, a smaller and more reactive glycosyl donor was required for this glycosylation. Indeed, a triflic acid mediated coupling of **7** with **18** led to the formation of trisaccharide **19** in an isolated yield of 71% and, fortunately, only a trace amount of the unwanted β-anomer was detected. Removal of Nap ether of **19** to give glycosyl acceptor **20** was accomplished by oxidation with DDQ in a mixture of DCM and water. In this reaction, care had to be taken to avoid oxidative removal of one of the benzyl ethers and in particular the use of only a small excess of recrystallized DDQ was critical



Scheme 3. The assembly of the GPI anchor moiety of *T. brucei*. Reagents and conditions: a) NIS, TfOH, DCM, -25°C , 30 min, 89%; b) DDQ, DCM:H₂O = 10:1, RT, 2 h, **17**: 82%; **20**: 77%; c) Tf₂O, TMB, DTBMP, -40°C to RT, then 10% TFA in DCM, RT, 1 h; (**18**: 87%, α only; **21**: 67%, α only); d) TfOH, DCM, -25°C to RT, 3 h, 71%, α : β > 20:1; e) TfOH, DTBMP, DCM, -60°C to RT, 18 h, then 10% TFA in DCM, RT, 1 h, 76%, α only; f) Ac₂O, pyridine, DMAP, RT, 4 h; g) NH₂NH₂-AcOH, pyridine, RT, 1 h; h) TMSOTf, DCM, -25°C to RT, 1 h, 51% over three steps; i) H₂, Pd/C, AcOH, MeOH, RT, 24 h, then MeONa, MeOH, RT, 1 h, 65%.

to avoid overoxidation.^[25] α -Galactosylation of **20** was easily accomplished by preactivation of **4** using Tf₂O and TMB in the presence of DTBMP in DCM at -40°C followed by the addition of glycosyl acceptor **20**. The remnant of the auxiliary of the resulting tetrasaccharide was cleaved by treatment with 10% trifluoroacetic acid (TFA) in DCM to give glycosyl acceptor **21** in an overall yield of 67% as only the α -anomer. Surprisingly, a glycosylation of **21** with **4** gave a pentasaccharide in a disappointing yield of 20%. Fortunately, a TMSOTf mediated glycosylation of **21** with **6** in DCM gave, after cleavage of the auxiliary, pentasaccharide **22** in an overall yield of 76% as only the α -anomer. The HSQC data of **22** showed that all H¹-C¹ coupling constants were in the range of 171 to 176 Hz confirming the α -configurations of the glycosidic linkages.

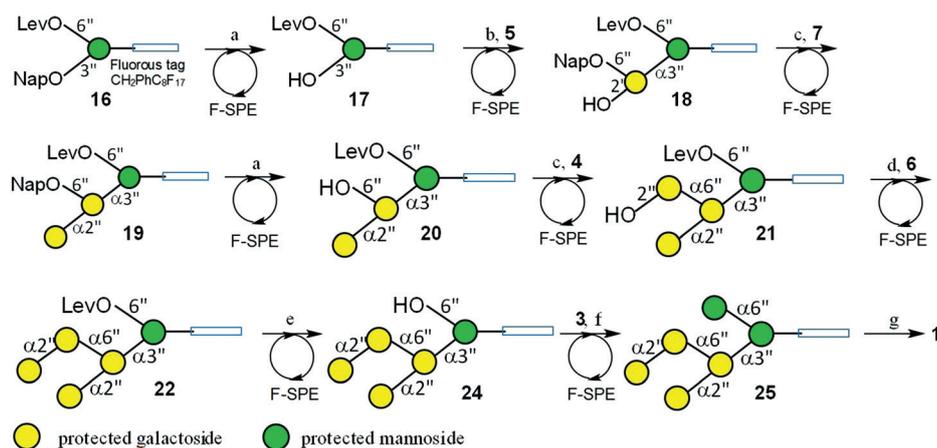
The hydroxyl of **22** was acetylation and the Lev ester of the resulting compound (**23**) was removed using hydrazine acetate to give glycosyl acceptor **24**, which was coupled with mannose donor **3** using TMSOTf as the catalyst to provide hexasac-

charide **24** in an excellent overall yield of 51% (three steps). In this case, only the α -anomeric product was formed due to neighboring-group participation of the acetyl ester at C-2 of the glycosyl donor. The overall yield of the assembly of the hexasaccharide, starting from the monomeric building blocks, was 9%. Finally, hexasaccharide **24** was converted into target compound **1** by hydrogenation over Pd/C, followed by removal of the acetyl esters using sodium methoxide in methanol.

Fluorous-assisted target glycan assembly

Having established a robust synthetic approach for the preparation of **1**, the synthesis of this compound was performed using a purification-protocol-based fluorous solid-phase extraction (F-SPE) using 20% water in methanol as the eluent to remove untagged compounds and the desired compound was isolated by elution with acetone.

Next, acceptor **17** was coupled with **5** using the standard preactivation protocol and, as expected, aqueous workup and solid-phase extraction resulted in the removal of hydrolyzed donor and other nonfluorous byproducts. The glycosylation was repeated and the remnant of the auxiliary was removed using 10% TFA in DCM to give, after standard fluorous solid phase extraction, disaccharide **18**. The latter compound was coupled twice with donor **4** using triflic acid as the promoter to provide trisaccharide **19**, which was subjected to DDQ oxidation to remove the NAP ether to provide acceptor **20**. Next, the α (1-6)-galactoside was installed by preactivation of **4** using Tf₂O, TMB, and DTBMP, followed by glycosylation with **20** and, after repeating the coupling protocol, the remnant of the auxiliary was removed by treatment with 10% TFA in DCM to give tetrasaccharide acceptor **21**. This compound was coupled twice with donor **6** using a standard preactivation protocol to give, after removal of the C-2 auxiliary and passing the material through a F-SPE cartridge, pentasaccharide **22**. The hydroxyl of



Scheme 4. The assembly of the GPI anchor moiety of *T. brucei* by fluororous solid-phase extraction. Reagents and conditions: a) DDO, DCM:H₂O = 10:1, 2 h; b) Tf₂O, TMB, DTBMP, -40 °C to RT, then 10% TFA in DCM, 1 h; c) TfOH, DCM, -25 °C to RT, 3 h; d) TfOH, DTBMP, DCM, -60 °C to RT, 18 h, then 10% TFA in DCM, 1 h; e) Ac₂O, pyridine, DMAP, 4 h, then NH₂NH₂-AcOH, pyridine, 1 h; f) TMSOTf, DCM, -25 °C to RT, 1 h; g) H₂, Pd/C, AcOH, MeOH, 24 h, then NaOMe, MeOH, 1 h.

22 was acetylated and the resulting compound was treated with hydrazine acetate to remove the Lev ester to give an acceptor which was subjected to a double coupling with mannosyl donor **2**. After each step, the product was isolated by solid-phase extraction and immediately used in the next reaction step. Homogeneous hexasaccharide **25** was obtained after purification by silica gel and LH-20 size-exclusion column chromatography. This compound was obtained in an overall yield of 16.7%, which corresponds to an 85% yield per reaction step. The assembly of the hexasaccharide could be completed within six days. Standard deprotection of **25** gave target compound **1**, the analytical data of which were identical to the compound prepared by the conventional approach described above.

After establishing a protocol for the efficient fluororous-supported synthesis of **1**, it could easily be adapted to the preparation of structurally related compounds and, for example, a pentasaccharide was assembled by appropriate protecting group manipulations and sequential coupling of **2** with **8** to give a product that was further extended with **5**, **4**, **4**, and **3**, respectively. The preparation of this compound was completed within five days.

Conclusion

We demonstrate here that a set of strategically selected orthogonal protecting groups, glycosyl donors modified by a chiral auxiliary and glycosyl acceptors containing a fluororous tag, make it possible to rapidly prepare complex branched oligosaccharides of biological importance. After the glycosylations, the chiral auxiliary could be removed using moderately strong acidic conditions, which were compatible with the presence of the orthogonal protecting groups Lev and Nap, thereby allowing efficient installation of 1,2-*cis*-linked glycosides. Previously, the auxiliary-mediated methodology was employed for the installation of α -glucosides,^[12a,c,18] and it is shown here that it can easily be extended to other monosaccharides such as galactosides. An exploratory study was required to identify

potential synthetic problems. For example, due to the bulky nature of the auxiliary, a glycosylation of a sterically hindered acceptor site was challenging and, in this case, a conventional donor had to be used. The attraction of the fluororous-supported methodology is that after establishing a successful synthetic approach, target compounds can rapidly be resynthesized by routine procedures. Also, it allows for fast preparation of structural analogs and, for example, the approach for fluororous-supported synthesis of **1** could easily be adapted to the preparation of structurally related compounds. Efforts are under-

way to develop a liquid handling system to automate fluororous-supported synthesis,^[9] which will make it possible to further speedup the process of oligosaccharide assembly.

Experimental Section

General procedure for the preparation of sulfoxide donors **4** and **5** from their corresponding oxathianes **10** and **15**

m-CPBA ($\leq 77\%$, 1.05 equiv) was dissolved in DCM and the resulting solution was slowly added to a cooled (-78 °C) solution of oxathiane in DCM. The reaction mixture was stirred at -78 °C for 30 min, diluted with DCM (20 mL) and then poured into 10% Na₂S₂O₃ aqueous solution. The organic layer was washed with aq. saturated NaHCO₃, dried (MgSO₄), and filtered; the filtrate was then concentrated in vacuo. The residue was purified by silica gel column chromatography.

General glycosylation procedure for oxathiane donors with various acceptors

Oxathiane donor (1.2 equiv), 1,3,5 trimethoxybenzene (2.5 equiv) and 2,6-di-*tert*-butyl-4-methylpyridine (3.0 equiv) were dissolved in DCM. Molecular sieves (4 Å) were added and the resulting suspension was cooled to -15 °C. Trifluoromethanesulfonic anhydride (1.2 equiv) was added dropwise to the solution and stirring was continued for 10 min. The reaction mixture was further cooled to -40 °C, and a solution of acceptor (1.0 equiv) in DCM, which was dried over molecular sieves (4 Å), was added dropwise. After a reaction time of 30 min, the reaction was quenched with aq. saturated NaHCO₃ (30 mL). The organic phase was washed with brine (30 mL), dried (MgSO₄), filtered, and the filtrate was concentrated in vacuo. The residue was purified by silica gel column chromatography or Sephadex LH-20 size exclusion chromatography (DCM/MeOH = 1:1, 0.2 mL min⁻¹).

General procedure for the removal of a C-2 auxiliary

Trifluoroacetic acid was added dropwise to a solution of the glycosylation product in DCM at 0 °C, adjusting the final concentration to 10% (v/v). The reaction mixture was stirred for 3 h until TLC in-

dicated complete consumption of starting material. The reaction mixture was diluted with DCM and poured into saturated NaHCO₃. The organic layer was dried (MgSO₄), filtered, and the filtrate was concentrated in vacuo. The residue was purified by silica gel column chromatography or Sephadex LH-20 size exclusion chromatography (DCM/MeOH = 1:1, 0.2 mL min⁻¹).

General fluoros-supported purification protocol

F-SPE cartridges (FluoroFlash SPE Cartridges, 10 grams, 20 cc tube) were purchased from Fluorous Technologies, Inc. The fluoros-tagged compound (200 mg compound per 1 g resin) was loaded on a F-SPE cartridge using a minimum amount of mixture of water and DMF (9:1, v:v). The order of elution was 20% water and methanol (3 × 20 mL), hexane (3 × 20 mL). The desired fluoros-tagged compound was obtained by elution with acetone (3 × 20 mL). The formation of the desired compound was determined by TCL and MALDI-TOF. The product containing fractions were concentrated in vacuo.

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Keywords: auxiliary · fluoros tag · stereoselective glycosylations · sulfonium ion

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