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Article

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Synthetic routes towards acidic pentasaccharide related to the *O*-antigen of *E. coli* 120 using one-pot sequential glycosylation reactions

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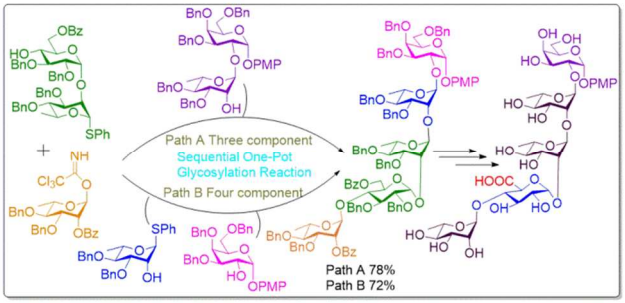


Abstract

Concise syntheses of the acidic pentasaccharide, related to the *O*-antigenic polysaccharide of *Escherichia coli* 120, as its *p*-methoxyphenyl glycoside, have been achieved utilizing one-pot sequential glycosylation technique. The glycosylations have been accomplished either by the activation of the thioglycosides using NIS in the presence of $FeCl_3$ or by a preactivation by Ph_2SO, TTBP, Tf_2O and the activation of the trichloroacetimidates using $FeCl_3$ alone or TMSOTf. Most of the intermediate steps are high yielding, and the stereo outcomes of the glycosylation steps were excellent. The syntheses of the targeted pentasaccharide have been performed with both three- and four-component one-pot sequential glycosylation reactions, and in both cases the orthogonal glycosylations are carried out utilizing catalytic activity of $FeCl_3$. A

late stage TEMPO-mediated regioselective oxidation has been performed to achieve the required uronic acid motif.

Introduction

Being a major component of the cell wall of Gram-negative bacteria, the *O*-antigens often play important roles during host infections, subsequent immune responses in the host and in controlling its virulence property. The *O*-antigen is one of the most variable cell constituent that consists of a polysaccharide chain with a number of repeats of an oligosaccharide. *Escherichia coli (E. coli)* are a facultative gram-negative bacteria present predominantly in the human and animal kingdoms. Identification of *E. coli* clones including the commensal and pathogenic strains are normally done by the combination of their somatic (O), flagellar (H) and occasionally by capsular (K) antigens.[1] More than 180 *O*-antigen forms of *E. coli* have been recognized so far.[2] The pathogenic *E. coli* strains in general cause three common infections like (i) enteric/diarrhoeal (ii) septicaemia/meningitis and (iii) urinary track infections.[3] The virulent *E. coli* strains causing diarrhea are classified in six classes as (i) enteropathogenic *E. coli* (EPEC), (ii) enterotoxigenic *E. coli* (ETEC), (iii) enteroinvasive *E. coli* (EIEC), (iv) diffusely adherent *E. coli* (DAEC), (v) enteroaggregative *E. coli* (EAEC) and (vi) enterohaemorrhagic *E. coli* (EHEC).[4]

The enterohaemorrhagic *E. coli* (EHEC) causes food-borne diseases with life threatening complications like haemorrhagic colitis (HC) and haemolytic-uraemic syndrome (HUS) in human and animal kingdoms worldwide.[4] Because of their toxic effect on the cultured Vero cells, these EHEC strains are also called 'verotoxigenic *E. coli* (VTEC)'. They are also termed as Shiga toxin producing *E. coli* (STEC) as they produce a bacteriophage-mediated Shiga-like

toxin.[5] The strains belonging to *E. coli* O120 isolated from their reservoirs like swine feces, cattle and beef products are identified as STEC.[6,7] Recently, Knirel et al. elucidated the structure of the repeating unit of the *O*-antigen from *E. coli* 120 and found to contain an acidic hexasaccharide repeating unit (Figure 1).[8]

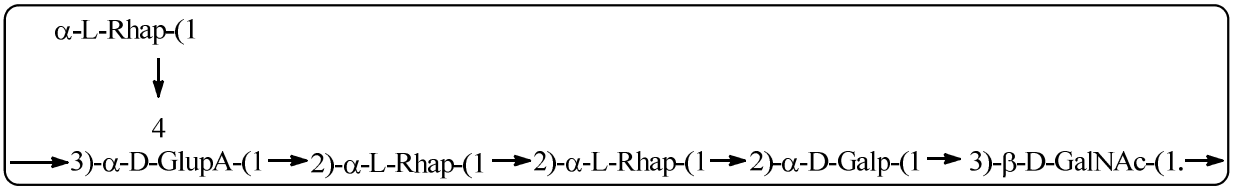


Figure 1 Hexasaccharide repeating unit of the *O*-antigen of *E. coli* type 120.

It has been well established that bacterial *O*-antigens regulate immuno-chemical activity of glyco-vaccines which make them attractive targets to the synthetic organic chemists for the development of glycoconjugate vaccine candidates.[9] Recently a number of reports have appeared for the synthesis of glycoconjugate vaccines and their evaluation against bacterial interactions.[9] Synthesis of an oligosaccharide with a temporary protecting group at the reducing end would be useful for its easy removal whenever necessary as it is often required to attached the oligosaccharide with a carrier protein through a spacer linker toward synthesis of glycoconjigates.[10] In this direction the synthesis of the oligosaccharide related to the repeating unit of the *O*-antigen from *E. coli* 120 was only reported by Mukhopadhyay *et al*,[11] using a conventional step-wise approach. However introduction of a step economic multistep one-pot total synthesis is still necessary and needs refinement of the entire synthetic protocol. The stepwise oligosaccharide syntheses[12] demand extensive protecting group manipulation and purification after each step making them expensive, time consuming, and tedious procedures.

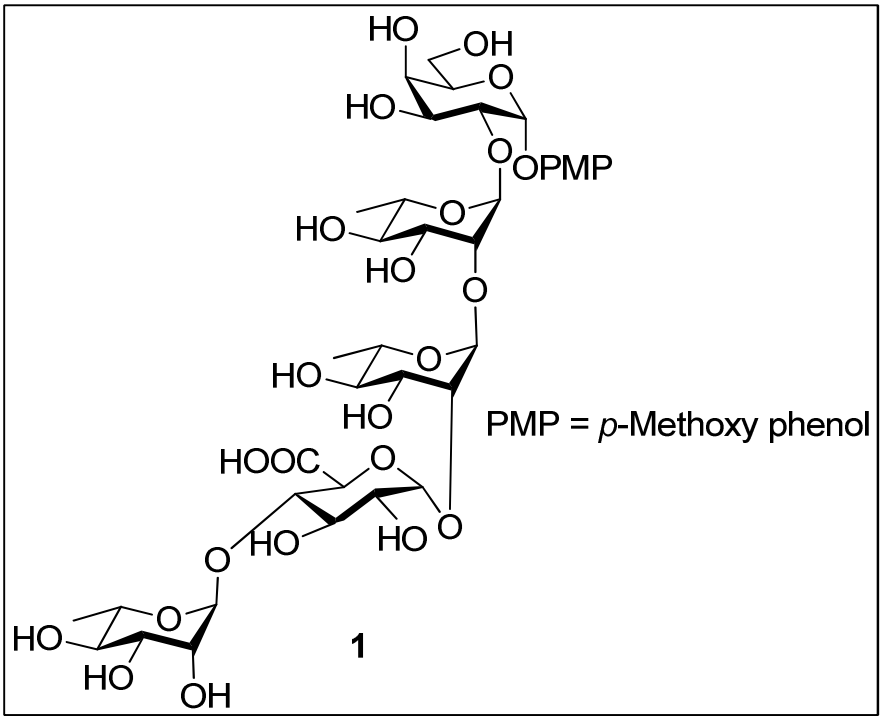


Figure 2 Structure of the target pentasaccharide.

In contrast, the one-pot sequential oligosaccharide syntheses are step economic, comparatively environment friendly, cost effective, and expeditious. We report herein concise synthesis of the acidic pentasaccharide part of the hexasaccharide repeating unit in the form of its 4-methoxyphenyl glycoside *via* both three and four component one-pot sequential glycosylation reactions (**1**, Figure 2). The convergent strategy will give the scope for the preparation of this important oligosaccharide structure in the pure form and in quantity that will pave the way for understanding its role in the pathogenic cycle. Moreover, a selective oxidative removal of the *p*-methoxyphenyl aglycon group of the pentasaccharide derivative using ceric ammonium nitrate (CAN) followed by formation of the corresponding trichloroacetimidate derivative will allow the formation of glycoconjugate with suitable aglycons targeting potential vaccine candidates against this deadly pathogen.

Results and discussion

The sequential one-pot glycosylation technique was applied for these total syntheses. Application of this in both three- and four-component one-pot reactions requires different sets of chemically distinct glycosyl donors requiring different activation conditions. For the convergent synthesis of the targeted acidic pentasaccharide **1** two different pathways were contemplated, one of which was a three-component sequential glycosylation i.e *via* [1+2+2] approach and the other one was a four-component sequential glycosylation reactions i.e *via* [1+2+1+1] approach.

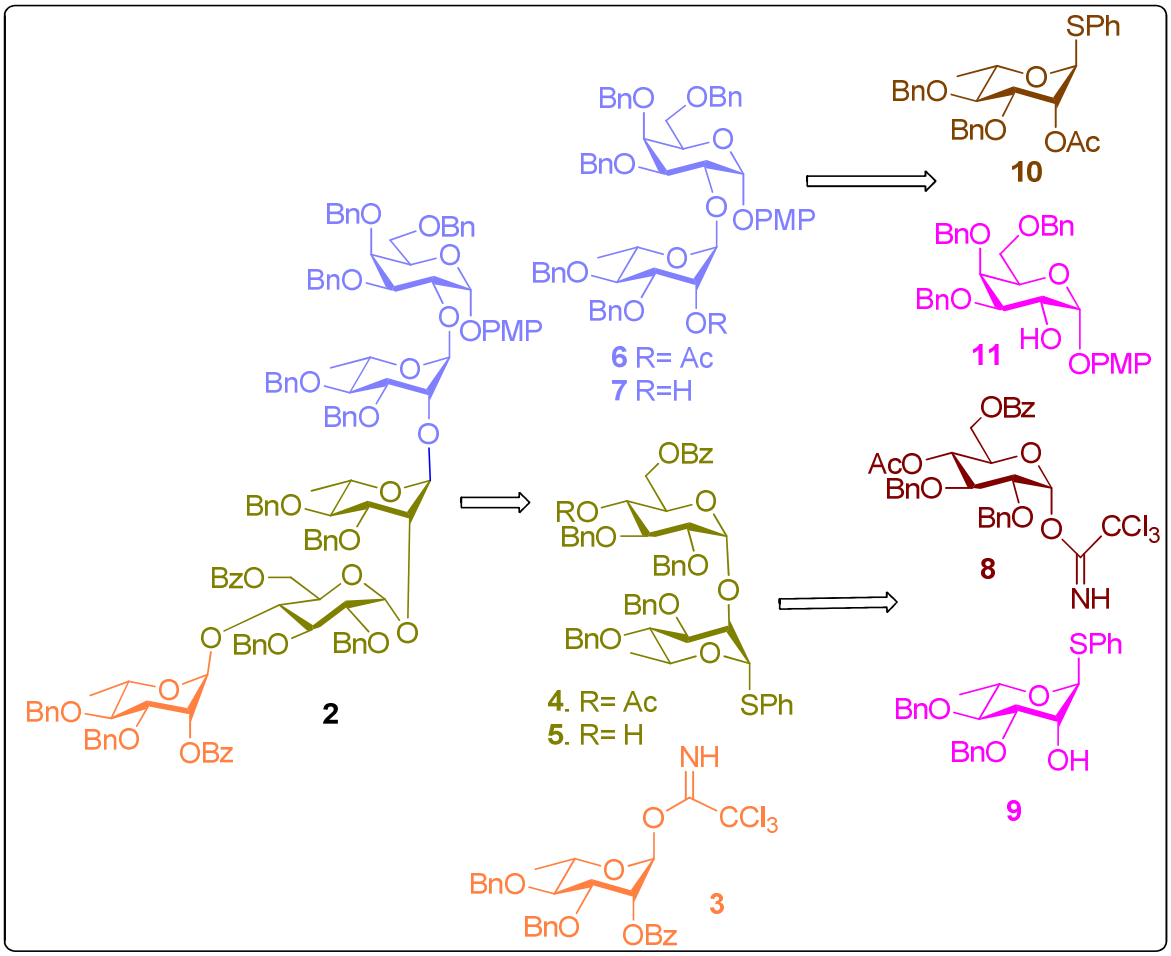
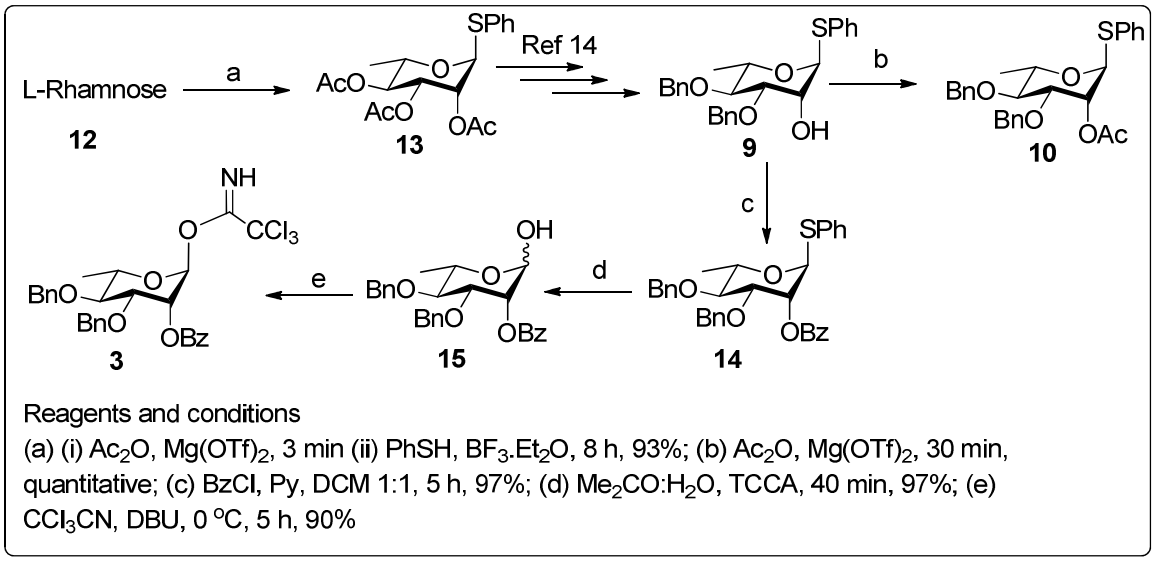


Figure 3 A retrosynthetic analysis of the pentasaccharide derivative **2**.

For each pathway, a retro-synthetic analysis of the fully protected pentasaccharide derivative **2** led to two common building blocks, one is 2-*O*-benzoyl-3,4-di-*O*-benzyl-L-rhamnopyranosyl trichloroacetimidate donor **3** and other is the disaccharide acceptor **5**, which can be prepared from the monomeric units **8** and **9.** Whereas the retrosynthetic analysis for the second approach [1+2+1+1] of the total synthesis guided us for use of the monomeric units **10** and **11**, the first synthetic approach [1+2+2] indicated us to deal with another disaccharide segment **7** to be prepared from **10** and **11** (Figure 3)**.** The disaccharide building blocks **5** and **7** can be obtained from their corresponding parent disaccharides **4** and **6** *via* selective removal of acetate protection.

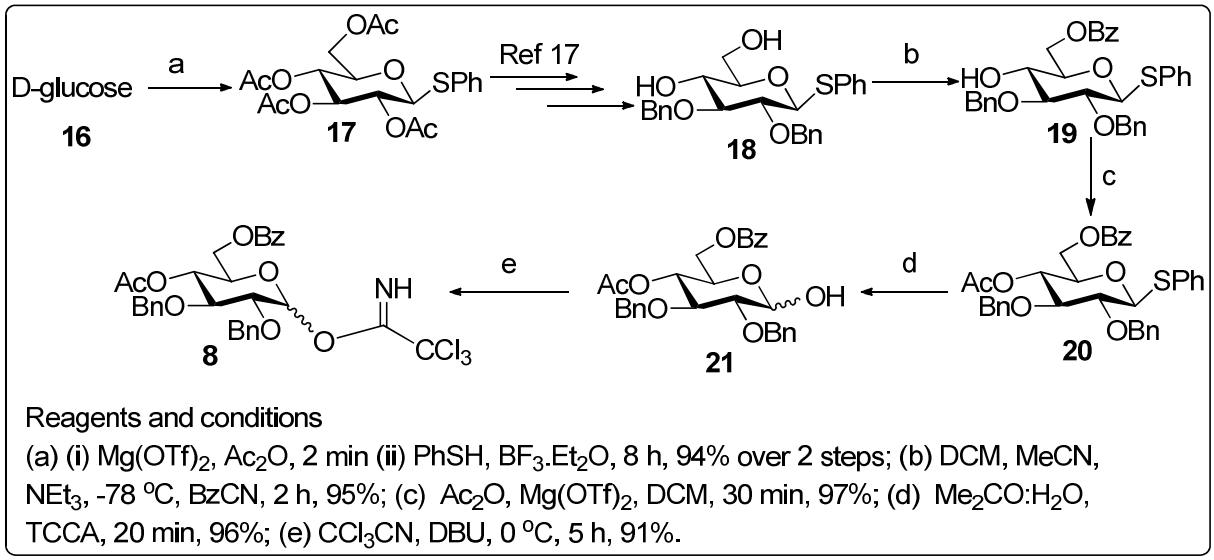
Scheme 1 Synthesis of L-rhamnose based building blocks.



Dry L-rhamnose was acetylated by acetic anhydride and a catalytic amount of magnesium (II) trifluromethanesulfonate [$Mg(OTf)_2$][13] in neat condition; after full consumption of the starting material (checked by TLC) into the same reaction vessel thiophenol followed by $BF_3.Et_2O$ were added to furnish phenyl 2,3,4-tri-*O*-acetyl-1-thio-*α*-L-rhamanopyranoside **13** in

93% yield over two steps after purification by column chromatography. This phenyl 2,3,4-tri-*O*-acetyl-1-thio-*α*-L-rhamanopyranoside **13** was converted to phenyl 3,4-di-*O*-benzyl-1-thio-*α*-L-rhamanopyranoside **9** according to a reported procedure.[14] Compound **9** was acetylated quantitatively using $Mg(OTf)_2$ and acetic anhydride to achieve phenyl 2-*O*-acetyl-3,4-di-*O*-benzyl-1-thio-*α*-L-rhamanopyranoside **10.**[15] On the other hand benzoylation of **9** produced phenyl 2-*O*-benzoyl-3,4-di-*O*-benzyl-1-thio-*α*-L-rhamanopyranoside **14.**[14] Thioglycoside hydrolysis of **14** was carried out following the method developed by us using trichloroisocyanuric acid (TCCA)[16] in aqueous acetone to give compound **15**. Reaction of **15** with trichloroacetonitrile and DBU in dry DCM afforded 2-*O*-benzoyl-3,4-di-*O*-benzyl-L-rhamnopyranosyl trichloroacetimidate donor **3** in 90% yield. Thus L-rhamnose based monosaccharide units **3, 9** and **10** were gathered (Scheme 1).

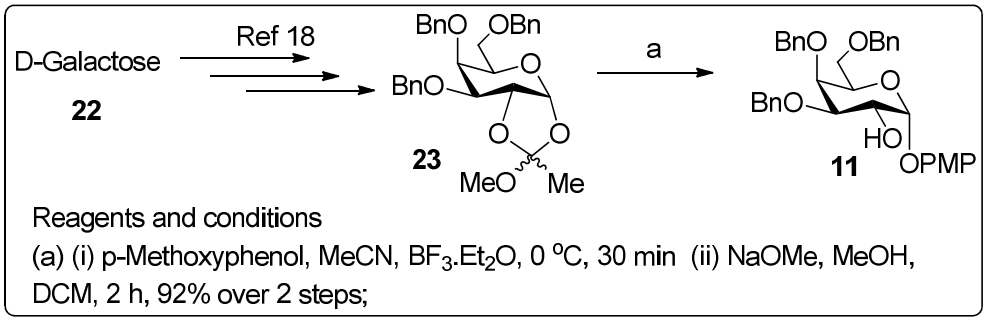
Scheme 2 Synthesis of D-glucose based donor **8**.



D-glucose was used as the preliminary starting material to synthesize the glucose based monomeric building block **8** (Scheme 2). Dry D-glucose was acetylated using acetic anhydride

and catalytic $Mg(OTf)_2$[13] in neat condition, after completion of the reaction (checked by TLC) into the same reaction vessel thiophenol followed by $BF_3.Et_2O$ were added to produce phenyl 2,3,4,6-tetra-*O*-acetyl-1-thio-*β*-D-glucopyranoside **17** in 94% over all yield. Compound **17** was transformed to phenyl 2,3-di-*O*-benzyl-1-thio-*β*-D-glucopyranoside **18**[17] according to the reported procedure. Then compound **18** was chemo-selectively benzoylated using 1:1:0.5 DCM:MeCN:NEt_3 and benzoyl cyanide at -78 ^oC for 2 hours to afford phenyl 6-*O*-benzoyl-2,3-di-*O*-benzyl-1-thio-*β*-D-glucopyranoside **19** in 95% yield. Quantitative acetylation of **19** with $Mg(OTf)_2$ and acetic anhydride produced phenyl 4-*O*-acetyl-6-*O*-benzoyl-2,3-di-*O*-benzyl-1-thio-*β*-D-glucopyranoside **20**. Thioglycoside hydrolysis of **20** with TCCA in wet acetone followed by base catalyzed formation of trichloroacetimidate furnished the desired glycosyl donor **8** (Scheme 2).

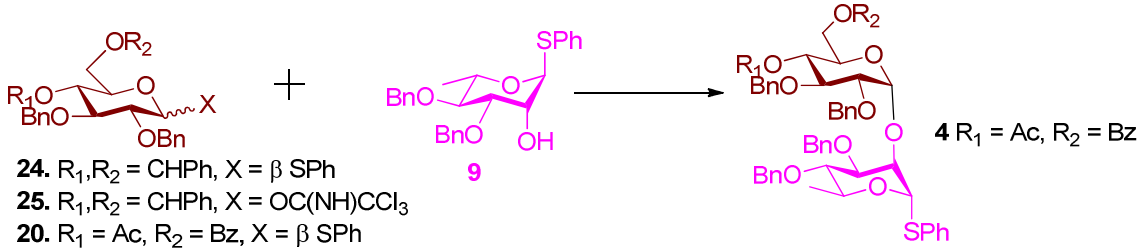
Scheme 3 Synthesis of D-galactose based acceptor **11**.



Dry D-galactose was converted to 3,4,6-tri-*O*-benzyl-*α*-D-galactopyanosyl-1,2-(methyl orthoacetate) **23**[18] according to the literature reported process. Then the reaction of this benzylated orthoester **23** with *p*-methoxyphenol using $BF_3.Et_2O$ in dry MeCN and this followed by Zemplén deacetylation[19] gave the acceptor *p*-methoxyphenyl 3,4,6-tri-*O*-benzyl-*α*-D-galactopyanoside **11** in 92% overall yield after two steps (Scheme 3). The *α*-configuration of the

glycoside **11** was established from both ^{1}H NMR spectrum (δ 5.50, H_1, $J_{1,2}$ = 3.5 Hz) and ^{13}C spectrum (δ 98.9, $J_{C\text{-}H}$ = 169.8 Hz).

Table 1 Optimization of *α*-D-glucosylation on acceptor **9**.



24. R_1,R_2 = CHPh, X = β SPh
25. R_1,R_2 = CHPh, X = OC(NH)CCl_3
20. R_1 = Ac, R_2 = Bz, X = β SPh
8. R_1 = Ac, R_2 = Bz, X = OC(NH)CCl_3

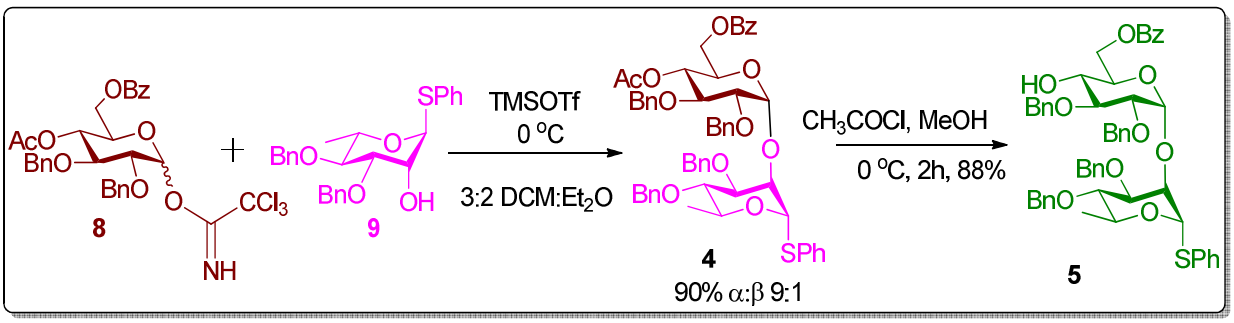
Entry	Donor(equiv)	Promoter	Solvent DCM:Et_2O	Temperature	Yield (α:β)
1.	**24**(1.2)	BSP, TTBP, Tf_2O	1:0	-60 °C to -78 °C to -40 °C	decomposition
2.	**24**(1.2)	Ph_2SO, TTBP, Tf_2O	1:0	-60 °C to -78 °C to rt	decomposition
3.	**25**(1.0)	TMSOTf	1:0	0 °C	decomposition
4.	**25**(1.2)	TMSOTf	1:0	-30 °C	No reaction
5.	**25**(1.2)	TMSOTf	1:0	-10 °C	decomposition
6.	**20**(1.2)	BSP, TTBP, Tf_2O	1:0	-60 °C to -78 °C to -40 °C	decomposition
7.	**8**(1.0)	TMSOTf	1:0	-10 °C	58% (3:2)
8.	**8**(1.2)	TMSOTf	1:0	0 °C	94% (3:2)
9.	**8**(1.2)	TMSOTf	2:1	0 °C	90% (5:2)
10.	**8**(1.2)	TMSOTf	3:2	0 °C	90% (9:1)

Glycosylation of acceptor **9** was first attempted with phenyl 2,3-di-*O*-benzyl-4,6-*O*-benzylidene-1-thio-*β*-D-glucopyranoside donor **24**[20] based on a pre-activation technique using 1-benzenesulfinylpiperidine (BSP),[20] 2,4,6-tri-tert-butylpyrimidine (TTBP) and Tf_2O in dry DCM under standard reaction condition (entry 1, Table 1). Instead of formation of glycosylated

product decomposition of the glycosyl donor was observed under this condition. Similar fate was observed when the preactivation based glycosylation was attempted using diphenyl sulfoxide (Ph_2SO), TTBP and Tf_2O in dry DCM (entry 2, Table 1).[21] Changing the donor **24** with the more reactive 2,3-di-*O*-benzyl-4,6-*O*-benzylidene-D-glucopyranosyl trichloroacetimidate **25** was next ventured. Coupling of **25** with rhamnopyranosyl acceptor **9** was explored using TMSOTf[22] as activator at different temperatures like 0 °C, -30 °C and -10 °C (entries 3, 4 and 5, Table 1). Unfortunately in all the cases either decomposition of the reaction mixture was observed or unreacted starting materials were recovered. Again another pre-activation based glycosylation technique using BSP, TTBP, Tf_2O was attempted on phenyl 4-*O*-acetyl-6-*O*-benzoyl-2,3-di-*O*-benzyl-1-thio-*β*-D-glucopyranoside **20**, but the result was similar as before (entry 6, Table 1).

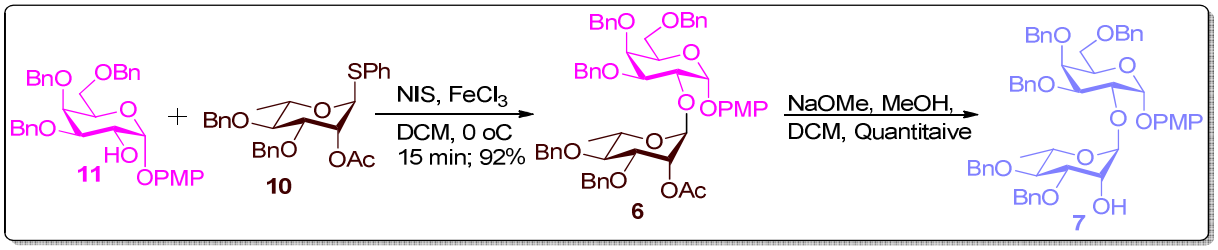
Gratifyingly, when 4-*O*-acetyl-6-*O*-benzoyl-2,3-di-*O*-benzyl-D-glucopyranosyl trichloroacetimidate donor **8** was allowed to couple with **9** using TMSOTf at -10 ℃ the desired coupling product was obtained in 58% yield and α:β = 3:2 ratio (entry 7, Table 1). Pleasingly, proper tuning of the reaction condition (entry 8, Table 1) allowed the isolation of the coupling product in good yield but with poor steroselectivity. Then the glycosylations were run in a mixture of DCM and Et_2O, the ratio (2:1 to 3:2) of which was optimized (entries 9 and 10, Table 1) to comply with solubility requirements on the one hand and glycosylation stereoselectivity at the other hand.

Scheme 4 Synthesis of disaccharide acceptor **5**.



Finally under the optimized condition 4-*O*-acetyl-6-*O*-benzoyl-2,3-di-*O*-benzyl-D-glucopyranosyl trichloroacetimidate **8** and the glycosyl acceptor phenyl 3,4-di-*O*-benzyl-1-thio-*α*-L-rhamanopyranoside **9** were allowed to couple using TMSOTf[22] at 0 °C in 3:2 DCM: Et_2O to produce the desired disaccharide in 90% yield with 9:1 *α*:*β* ratio. The NMR spectra of disaccharide **4** showed signals characteristic of the presence of both donor and acceptor moieties, while an *α*-configuration of the inter-glycosidic linkage was confirmed from the ^1H and ^{13}C NMR spectra. A chemo-selective deacetylation in the presence of 6-OBz of **4** using acetyl chloride in dry MeOH at 0 °C produced the disaccharide acceptor **5** (Scheme 4) in 88% yield. Compared to the NMR spectra of **4**, the disappearance of one signal of carbonyl carbon from 169.6 ppm in the corresponding ^{13}C spectrum and the sharp singlet signal from 1.99 ppm in the ^1H spectrum in **5** clearly indicated the selective removal of the acetate protecting group in the presence of 6-OBz.

Scheme 5 Synthesis of disaccharide acceptor **7**.

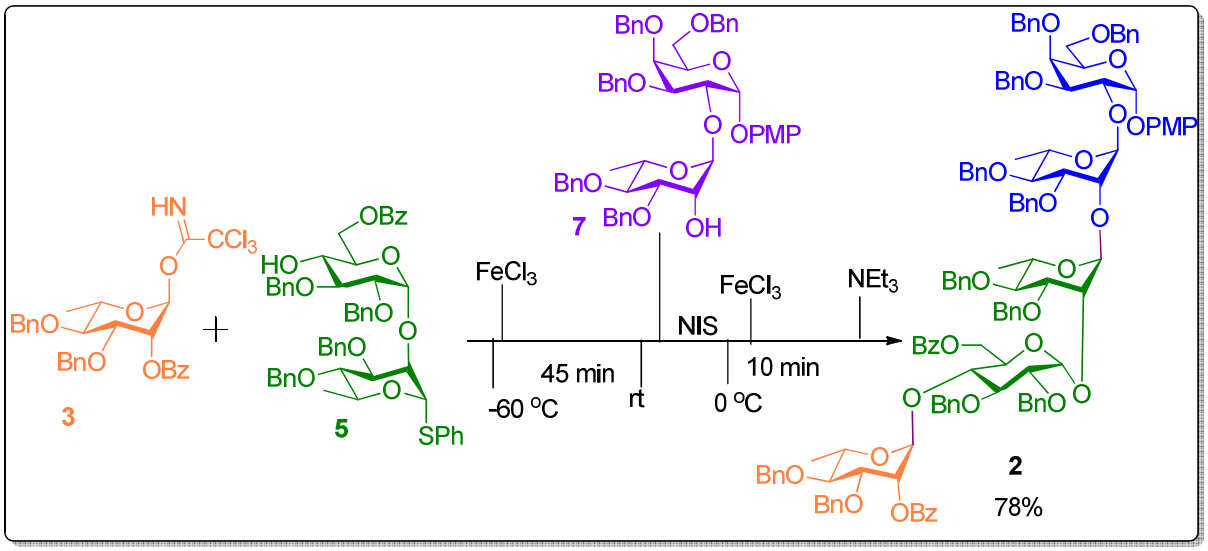


A mixture of thioglycoside donor **10** and glycosyl acceptor *p*-methoxyphenyl 3,4,6-tri-*O*-benzyl-*α*-D-galactopyanoside **11** in dry DCM at 0 °C was treated with NIS and $FeCl_3$[23] to produce disaccharide **6** in 92% yield. The NMR spectra of disaccharide **6** showed signals characteristic of the presence of both donor and acceptor moieties, while an *α*-configuration of the interglycosidic linkage was confirmed from the corresponding ^1H (δ 5.01, H_1', $J_{1,2}$ = 0.8 Hz) and ^{13}C NMR (δ 97.8) spectra. Deacetylation of **6** with NaOMe - MeOH and dry DCM furnished

quantitatively the disaccharide acceptor **7**, necessary for the one-pot sequential glycosylation *via* an [1+2+2] approach (Scheme 5).

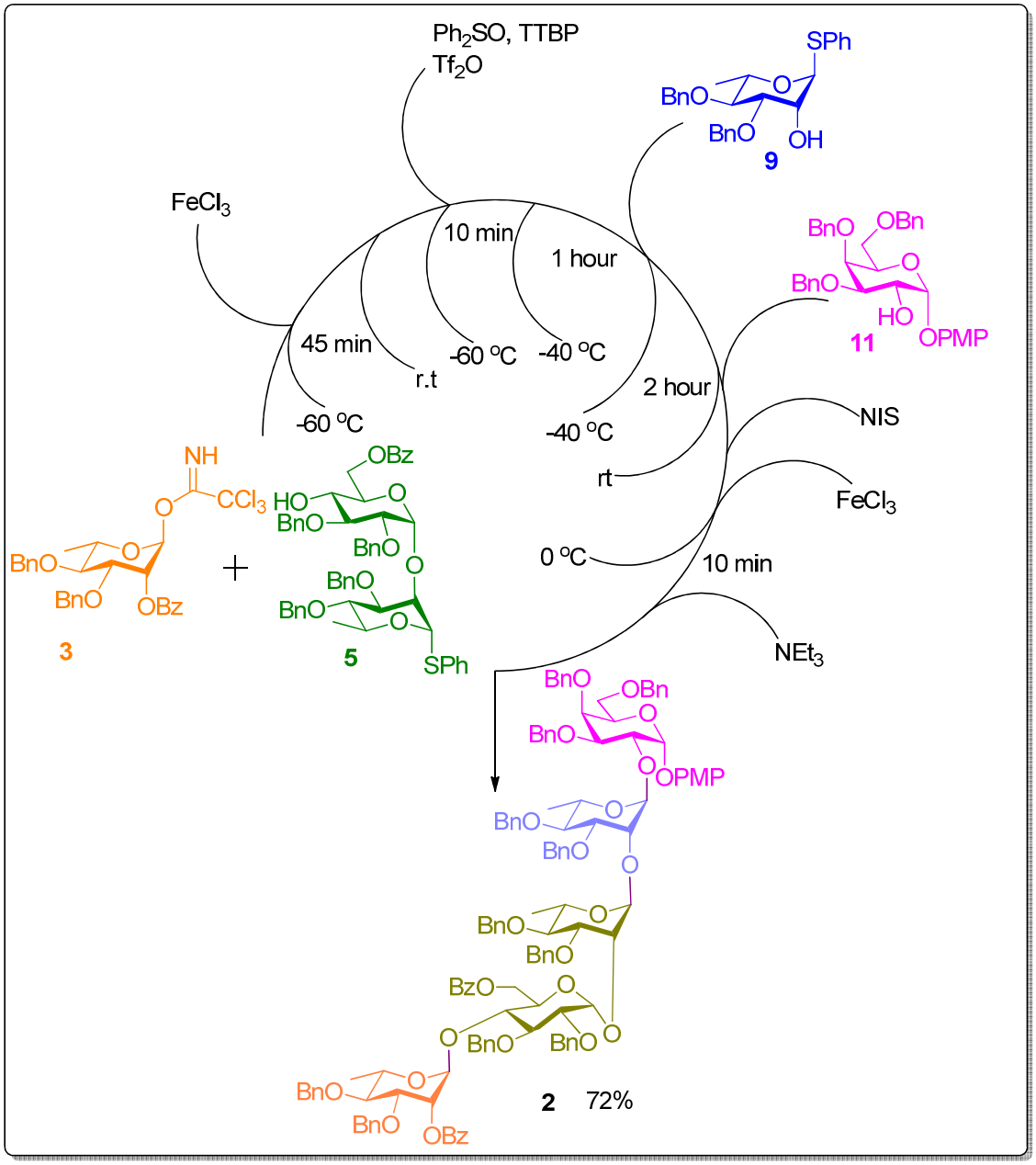
With the present set of donors and acceptors in hand we set out for the one-pot glycosylation reactions towards targeted pentasaccharide *via* a [1+2+2] approach first. Disaccharide acceptor **5** was glycosylated with glycosyl donor **3** at -60 ^{o}C to room temperature using 10 mole% of $FeCl_3$.[23] After full consumption of the starting materials (checked by TLC), into the same pot second disaccharide acceptor **7** followed by NIS were added. The reaction mixture was cooled down to 0 ^{o}C and another 10 mole% of $FeCl_3$ was added, TLC after 10 minutes showed complete consumption of the starting materials. Thus the targeted pentasaccharide was prepared *via* a three-component one pot sequential glycosylation technique in 78% yield (Scheme 6). The formation of the pentasaccharide derivative **2** was confirmed by NMR spectroscopic techniques (^{1}H-, ^{13}C-, COSY, HSQC, HMBC, NOESY) and also by HRMS.

Scheme 6 Synthesis of pentasaccharide derivative **2** *via* a three-component one-pot sequential glycosylation technique.



The anomeric protons of compound **2** appeared at δ 4.68 (bs, H_1'''), 5.03 (2 bs, H_1' and H_1''), 5.21 (bs, H_1'''') and 5.23 (d, J 3.5 Hz, H_1) ppm and the corresponding carbons at 94.1(C_1'''), 98.2 (C_1'), 101.6 (C_1''), 97.9 (C_1'''') and 98.0 (C_1) ppm, respectively, and the signal of the carbonyl carbons appeared at 165.8 and 166.1 ppm.

Scheme 7 Synthesis of pentasaccharide derivative **2** *via* a four-component one-pot sequential glycosylation technique.

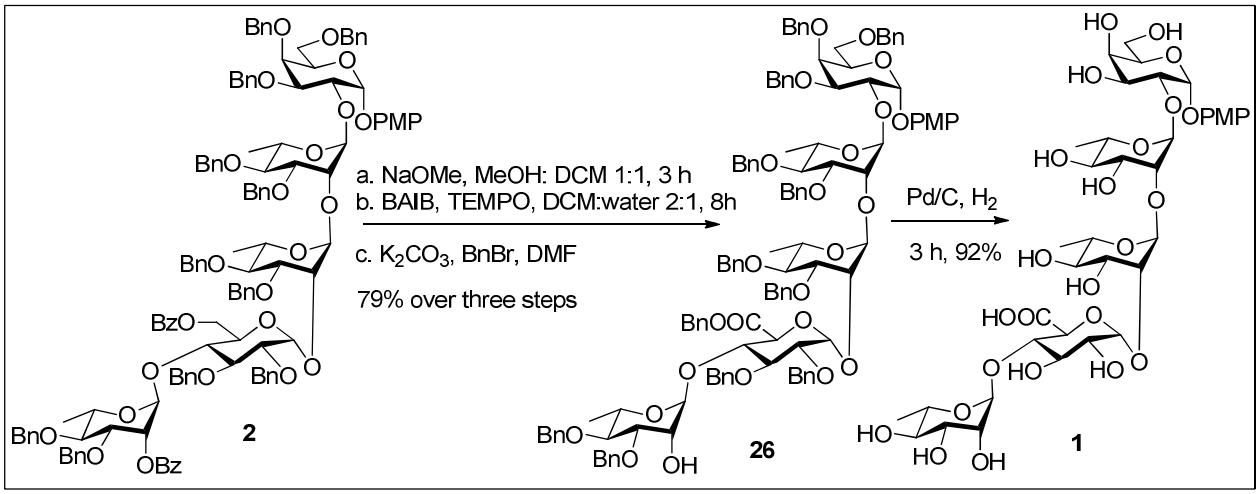


After successfully achieving a synthetic route towards pentasaccharide derivative **2** *via* a three-component one-pot sequential glycosylation technique we sought to synthesize the same in a more step economic way. A four-component one-pot sequential glycosylation reactions for synthesis of the target **2** were then explored. For this, disaccharide acceptor **5** was first glycosylated with 2-*O*- benzoyl-3,4-di-*O*-benzyl-L-rhamnopyranosyl trichloroacetimidate donor **3** under previously standardized procedure using 10 mole% of $FeCl_3$ at -60 ^oC to room temperature.[23] After the reaction showed a clear conversion towards our desired trisaccharide derivative (indicated by TLC) the reaction mixture was again cooled to -60 ^oC. To this cold mixture diphenyl sulfoxide (Ph_2SO), TTBP and triflic anhydride (Tf_2O) were added, and the mixture was kept at that temperature for 10 minutes, and then the temperature was raised to -40 ^oC. After 1 hour at that temperature the second glycosyl acceptor **9** in dry DCM was injected into the cold reaction vessel, and the reaction mixture was allowed to attain room temperature. After consumption of both the starting materials (checked by TLC) the last acceptor **11** and NIS were added into the same reaction vessel. Lowering the temperature to 0 ^oC into this mixture $FeCl_3$ was added again, and the resulting mixture was kept for 10 minutes at that temperature, and after complete conversion towards desired pentasaccharide derivative **2** (indicated by TLC) the reaction was quenched with NEt_3. Thus the targeted pentasaccharide was prepared *via* a four-component one-pot sequential glycosylation technique in 72% yield (Scheme 7).

Finally debenzoylation under Zemplén condition, then regeoselective oxidation of primary hydroxyl group in the presence of a secondary one using TEMPO, bis-acetoxy iodobenzene (BAIB) in DCM water followed by benzylation using K_2CO_3, benzyl bromide in dry DMF afforded the fully benzylated derivative of the acidic pentasaccharide derivative **26** in 79% yield over three steps (Scheme 8). Finally a global debenzylation using hydrogen and

palladium-charcoal in a mixture of ethyl acetate, water and methanol ultimately afforded the desired pentasaccharide **1** in 92% yield (Scheme 8). Compound **1** was characterized by NMR spectroscopic techniques (^{1}H-, ^{13}C-, COSY) and also by HRMS.

Scheme 8 Conversion of pentasaccharide derivative **2** to the target pentasaccharide **1**.



Conclusion

In conclusion, we have developed an expeditious strategy for the synthesis of the acidic pentasaccharide, related to the *O*-antigen of *E. coli* 120, in the form of its *p*-methoxyphenyl glycoside (**1**) *via* one-pot sequential glycosylation techniques. The synthesis of the target compound is achieved through suitable protecting group manipulations on commercially available monosaccharides and stereo-selective glycosylations. Protecting group manipulation like per-*O*-acetylation-thioglycosidation was performed in one-pot. The glycosylations were achieved either by the activation of the thioglycosides using NIS in the presence of $FeCl_3$ or by a preactivation using Ph_2SO, TTBP, Tf_2O and the activation of the trichloroacetimidates using $FeCl_3$ alone or TMSOTf. The targeted pentasaccharide syntheses were performed with both

three- and four- component one-pot sequential glycosylation reactions, and in both cases the orthogonal glycosylations were carried out utilizing the catalytic activity of $FeCl_3$.

Experimental

General procedure

All reactions were performed in flamed-dried flasks fitted with rubber septa under a positive pressure of argon, unless otherwise stated. DCM was refluxed with P_2O_5 and distilled before use and stored over 4Å molecular sieves. Traces of water in the starting materials were removed by co-evaporation with toluene. Flash column chromatography was performed employing Silica Gel 60 Sorbent (40-63 μm, 230-400 mesh). Thin-layer chromatography (analytical and preparative) was performed using Merck silica gel plates (60-F254) to monitor the reactions and visualised under UV (254 nm) and/or by charring with 5% ethanolic solution of sulfuric acid. ^1H and ^{13}C NMR spectra were recorded on a Bruker DPX-300 (300 MHz), a Bruker DPX-400 (400 MHz), a Bruker DPX-500 (500 MHz), or a Bruker DPX-600 (600 MHz) spectrometer at ambient temperature in $CDCl_3$ or D_2O and assigned using 2D-methods (COSY, HSQC). Optical rotations were measured using Jasco P-1020 digital polarimeter. High Resolution Mass Spectra (HRMS) were measured in a QTOF I (quadrupole-hexapole-TOF) mass spectrometer with an orthogonal Z-spray-electrospray interface on Micro (YA-263) mass spectrometer (Manchester, UK).

2-*O*-Benzoyl-3,4-di-*O*-benzyl-α-L-rhamnopyranosyl trichloroacetimidate (3)

To a solution of compound **14** (2 g, 3.7 mmol) in aqueous acetone (4:1), TCCA (1.2 g, 3.7 mmol) was added at 0 °C, and the reaction mixture was kept on stirring for 40 minutes. Then the white precipitate was filtered, and the bed was washed with DCM (3×5 mL). The combined

filtrate and washings was evaporated, and the resulting mixture was again dissolved in DCM. This organic part was washed subsequently with saturated $NaHCO_3$ solution (200 mL) and water (200 mL). The organic layer was dried over anhydrous Na_2SO_4 and evaporated under vacuum to furnish compound **19**. Column filtration of the crude product furnished pure compound as white foam (**15**, 1.6 g, 97 %). Then to this solution of compound **15** (1 g, 2.23 mmol) and CCl_3CN (0.34 mL, 3.34 mmol) in dry DCM (15 mL) DBU (0.1 mL, 0.67 mmol) was added at -5 °C, and the reaction mixture was kept on stirring at that temperature. After 5 hours, excess solvent was removed, and the resulting mixture was purified through flash column chromatography (PE/EA, 5:1) to furnish pure compound **3** as colorless syrup (1.18 g, 90 %).; $[\alpha]^{25}_{D}$ 4.2 (*c* 1.15, $CHCl_3$). ^{1}H-NMR (300 MHz, $CDCl_3$): δ 8.67 (s, 1H, N*H*), 8.11-8.09 (d, *J* = 8.1 Hz, 1H, Ar*H*), 7.64-7.26 (m, 13H, Ar*H*), 6.31 (s, 1H, H_1), 5.72 (s, 1H, H_2), 4.93 (d, *J* = 10.8 Hz, 1H, Bn*H*), 4.80 (d, *J* = 11.4 Hz, 1H, Bn*H*), 4.66 (d, *J* = 11.1 Hz, 1H, Bn*H*), 4.61 (d, *J* = 11.5 Hz, 1H, Bn*H*), 4.11 (dd, *J* = 3.0, 9.4 Hz, 1H, H_3), 4.00 (m, 1H, H_5), 3.65 (app t, *J* =9.5 Hz, 1H, H_4), 1.39 (d, *J* = 6.2 Hz, 3H, CH_3).; ^{13}C-NMR (75 MHz, $CDCl_3$): δ 165.5 (*C*=O), 160.2 (*C*=NH), 138.1, 137.6, 133.4, 130.0, 129.7, 128.5, 128.4, 128.3, 128.1, 127.9, 127.8, 95.4 (C_1), 90.9, 79.4, 77.4, 75.6, 71.9, 70.8, 68.1, 18.2 (*C*H_3).; HRMS (ESI-TOF): Calculated for $C_{29}H_{28}Cl_3NO_6Na$ (M+Na) 614.088 found 614.0882.

Phenyl 6-*O*-benzoyl-2,3-di-*O*-benzyl-1-thio-*β*-D-glucopyranoside (19)

To a solution of phenyl 2,3-di-*O*-benzyl-1-thio-*β*-D-glucopyranoside **18** (3 g, 6.64 mmol) in 1:1:0.5 mixture of DCM:MeCN:NEt_3 (10 mL) benzoyl cyanide (0.9 mL, 7.3 mmol) diluted in 2 mL of dry DCM was added at -78 ^{o}C, and the reaction mixture was stirred at that temperature for 2 hours. The reaction mixture was diluted in DCM and washed subsequently with saturated $NaHCO_3$ solution (200 mL) and water (200 mL). The organic layer was dried over anhydrous

Na_2SO_4 and evaporated under vacuum to furnish crude product **19**. Purification of **19** by silica gel column chromatography (PE/EA, 3:1) yielded phenyl 6-*O*-benzoyl-2,3-di-*O*-benzyl-1-thio-*β*-D-glucopyranoside as white solid (**19**, 3.5 g, 95%).; mp (EA/PE) 90-92 °C.; $[\alpha]^{25}_D$ -21.8 (*c* 1.65, $CHCl_3$).; ^{1}H-NMR (300 MHz, $CDCl_3$): δ 8.09-8.06 (d, *J* = 7.3 Hz, 2H, Ar*H*), 7.61-7.14 (m, 18H, Ar*H*), 4.95-4.92 (m, 2H), 4.81-4.70 (m, 3H), 4.64 (bs, 1H), 3.52-3.59 (m, 4H), 3.51 (m, 1H).; ^{13}C-NMR (75 MHz, $CDCl_3$): δ 166.9 (*C*=O), 138.3, 137.9, 133.5, 133.3, 132.2, 129.9, 129.8, 128.9, 128.7, 128.5, 128.4, 128.3, 128.1, 128.0, 127.7, 87.5 (C_1), 85.9, 80.4, 77.8, 75.7, 75.4, 70.1, 63.9.; HRMS (ESI-TOF): Calculated for $C_{33}H_{32}O_6SNa$ (M+Na) 579.1818 found 579.1819.

Phenyl 4-*O*-acetyl-6-O-benzoyl-2,3-di-*O*-benzyl-1-thio-*β*-D-glucopyranoside (20)

Acetic anhydride (0.56 mL, 5.94 mmol) and $Mg(OTf)_2$ (9.4 mg, 0.027 mmol) were added to a solution of compound **19** (3 g, 5.39 mmol) in dry DCM (15 mL) at 0 °C, and the resulting mixture was kept on stirring for 30 minutes. After completion of the reaction (indicated by TLC), excess acetic anhydride was removed using rotatory evaporator. The resulting syrup was dissolved in DCM and washed subsequently with saturated $NaHCO_3$ solution (200 mL) and water (200 mL). The organic layer was dried over anhydrous Na_2SO_4 and concentrated to afford the crude product. It was purified by column chromatography on silica gel. Column elution by PE/EA, 4:1 furnished pure compound **20** as white solid (3.13 g, 97%).; mp (EA/PE) 84-86 °C.; $[\alpha]^{27}_D$ -27.8 (*c* 2.8, $CHCl_3$).; ^{1}H-NMR (300 MHz, $CDCl_3$): δ 8.08-8.05 (d, *J* = 7.5 Hz, 2H, Ar*H*), 7.60-7.10 (m, 18H, Ar*H*), 4.74-4.64 (m, 3H, H_1, H_6, Bn*H*), 4.54 (d, *J* = 12.1 Hz, 1H, Bn*H*), 5.14 (t, *J* = 9.7 Hz, 1H, H_4), 4.93-4.82 (m, 2H, Bn*H*), 4.32 (m, 1H, H_6), 3.77-3.68 (m, 2H, H_3, H_5), 3.58 (t, *J* = 9.3 Hz, 1H, H_2), 1.93 (s, 3H, COCH_3).; ^{13}C-NMR (75 MHz, $CDCl_3$): δ 169.7 (*C*=O), 166.2 (*C*=O), 138.0, 137.8, 133.2, 133.1, 132.4, 129.9, 129.8, 128.9, 128.5, 128.4, 128.3, 128.1,

127.9, 127.8, 87.5 (C_1), 83.9, 80.6, 76.0, 75.6, 69.9, 63.2, 20.8 (CH_3CO).; HRMS (ESI-TOF): Calculated for $C_{35}H_{34}O_7SNa$ (M+Na) 621.1923 found 621.1940.

4-*O*-Acetyl-6-*O*-benzoyl-2,3-di-*O*-benzyl-D-glucopyranose (21)

To a solution of compound **20** (2 g, 3.3 mmol) in aqueous acetone (4:1), TCCA (1.2 g, 3.3 mmol) was added at ambient temperature, and the mixture was kept on stirring for 20 minutes. Then the white precipitate was filtered, and the bed was washed with DCM (3×15 mL). The combined filtrate and washings was evaporated, and the resulting mixture was again dissolved in DCM. This organic part was washed subsequently with saturated $NaHCO_3$ solution (200 mL) and water (200 mL). The organic layer was dried over anhydrous Na_2SO_4 and evaporated under vacuum to furnish compound **21**. Column filtration of the crude product furnished white foam of pure compound as mixture of anomers (**21**, 1.62 g, 96 %).; HRMS (ESI-TOF): Calculated for $C_{29}H_{30}O_8Na$ (M+Na) 529.1839 found 529.1840.

4-*O*-Acetyl-6-*O*-benzoyl-2,3-di-*O*-benzyl-D-glucopyranosyl trichloroacetimidate (8)

To a solution of compound **21** (1.5 g, 2.96 mmol) and CCl_3CN (0.46 mL, 4.44 mmol) in dry DCM (15 mL) DBU (0.14 mL, 0.9 mmol) was added at -5 °C, and the reaction mixture was kept on stirring at that temperature. After 5 hours, excess solvent was removed, and the resulting crude product was purified through flash column chromatography (PE/EA, 5:1) to furnish pure compound **8** as colorless syrup (1.75 g, 91 %).; ^{1}H-NMR (300 MHz, $CDCl_3$): δ 8.63 (s, 1H, N*H*), 8.02-8.00 (d, *J* = 8.0 Hz, 2H, Ar*H*), 7.55-7.25 (m, 13H, Ar*H*), 6.84 (d, *J* = 3.0 Hz, 1H, H_1), 5.21 (t, *J* = 10.0 Hz, 1H, H_4), 4.89 (d, *J* = 11.5 Hz, 1H, Bn*H*), 4.76-4.67 (m, 3H, H_5, Bn*H*), 4.47 (d, *J* = 12.0 Hz, 1H, Bn*H*), 4.27 (dd, *J* = 12.5, 5.0 Hz, 1H, H_6), 4.21 (dd, *J* = 10.5, 4.5 Hz, 1H, H_6), 4.02 (t, *J* = 9.5 Hz, 1H, H_3), 3.83 (dd, *J* = 9.5, 3.5 Hz, 1H, H_2), 1.97 (s, 3H, COCH_3).; ^{13}C-NMR (75 MHz, $CDCl_3$): δ 169.6 (*C*=O), 166.2 (*C*=O), 162.3, 160.9, 138.2, 137.7, 133.1, 129.8, 128.5,

128.4, 128.3, 128.0, 127.9, 127.8, 127.7, 93.9 (C_1), 91.1, 79.2, 78.4, 75.4, 73.1, 70.5, 69.2, 62.6, 20.8 (CO*C*H_3).; Calculated for $C_{31}H_{30}NCl_3O_8Na$ (M+Na) 672.0935 found 672.0930.

***p*-Methoxylphenyl 3,4,6-tri-*O*-benzyl-*α*-D-galactopyranoside (11).**

To a solution of 3,4,6-tri-*O*-benzyl-*α*-D-galactose 1,2-(methyl orthoacetate) **23** (2.50 g, 4.94 mmol), in dry CH_3CN (30 mL) containing 4 Å molecular sieves, *p*-methoxyphenol (1.23 g, 9.96 mmol) were added at room temperature. $BF_3.Et_2O$ (1.9 mL, 14.94 mmol) was added to this mixture on ice-bath and kept for 30 minutes. The reaction mixture was filtered through a celite bed, and the residue was washed with DCM (3×15 mL). The combined filtrate and washings was washed subsequently with cold 5% aqueous NaOH (250 mL) followed by water (100 mL). The organic layer was dried over anhydrous Na_2SO_4 and concentrated. The crude residue was used for further reaction. To a solution of this crude material in a mixture of dry DCM (10 ml) and dry MeOH (8 mL) 1 M methanolic NaOMe (2 mL) solution was added, and the reaction mixture was stirred for 2 hours at room temperature. The reaction mixture was then neutralized with Dowex-50W cation exchange resin (H^+) and filtered. The resin was washed with MeOH, and the combined filtrate and washings was concentrated under reduced pressure. The residue was purified by column chromatography on silica gel (PE/EA, 9:1) to afford **11** as white solid (2.56 g, 92% over 2 steps).; mp (EA/PE) 96-98 °C.; $[\alpha]^{29}_D$ +100.6 (*c* 2.1, $CHCl_3$).; ^{1}H-NMR (300 MHz, $CDCl_3$): δ 7.43-7.26 (m, 15H, Ar*H*), 7.07-7.04 (d, *J* = 8.9 Hz, 2H, Ar*H*), 6.83-6.79 (d, *J* = 8.9 Hz, 2H, Ar*H*), 5.51 (d, *J* = 3.5 Hz, 1H, H_1), 4.76 (d, *J* = 11.3 Hz, 1H, Bn*H*), 4.84-4.75 (ABq, *J* = 11.5 Hz, 2H, Bn*H*), 4.62 (d, *J* = 11.3 Hz, 1H, Bn*H*), 4.51-4.35 (m, 3H, O*H*, Bn*H*), 4.17 (app t, *J* = 6.3, 6.1 Hz, 1H, H_6), 4.10 (bs, 1H, H_4), 3.92 (dd, *J* = 9.9, 2.4 Hz, 1H, H_3), 3.77 (s, 3H, OCH_3), 3.69-3.56 (m, 3H, H_2, H_5, H_6).; ^{13}C-NMR (75 MHz, $CDCl_3$): δ 155.3, 150.8, 138.5, 138.2, 137.9, 128.6, 128.4, 128.3, 128.2, 127.8, 127.76, 118.7, 114.6, 98.9 (C_1, $J_{C\text{-}H}$ = 169.8),

79.6, 74.8, 74.0, 73.5, 72.6, 70.4, 69.0, 68.8, 55.6 (O*C*H_3).; HRMS (ESI-TOF): Calculated for $C_{34}H_{36}O_7Na$ (M+Na) 579.2359 found 579.2379.

Phenyl 2-*O*-acetyl-6-*O*-benzoyl-2,3-di-*O*-benzyl-*α*-D-glucopyranosyl-(1→2)-3,4-di-*O*-benzyl-1-thio-*α*-L-rhamnopyranoside (4)

To a solution of 2-*O*-acetyl-6-*O*-benzoyl-2,3-di-*O*-benzyl-*α*-D-glucopyranosyl trichloroacitimidate **8** (200.0 mg, 0.31 mmol) and phenyl 3,4-di-*O*-benzyl-1-thio-*α*-L-rhamnopyranoside **9** (120.9 mg, 0.28 mmol) in 3:2 (v/v) mixture of dry DCM:Et_2O (30 mL) activated molecular sieves (4Å) were added, and the reaction mixture was kept on stirring under argon atmosphere for 45 minutes. Then the reaction vessel was placed in a 0 °C cold bath, and TMSOTf (10 µL, 0.06 mmol) was added to it *via* a micro syringe. After 10 minutes complete consumption of both the starting materials was observed. The reaction mixture was quenched by Et_3N, and then filtered through celite bed, and the bed was washed with DCM (3×15 mL). The combined filtrate and washings was washed subsequently with saturated $NaHCO_3$ (2x50 mL) and water (2x50 mL). The organic layer was dried over anhydrous Na_2SO_4 and concentrated to furnish a syrupy compound. The crude product was purified by flash column chromatography (eluent: PE/EA, 3:1) to afford the desired disaccharide derivative **4** as white foam (230.6 mg, 90%).; $[\alpha]^{28}_D$ +11.8 (c 3.6, $CHCl_3$).; ^{1}H-NMR (400 MHz, $CDCl_3$): δ 8.08-8.06 (d, *J* = 8.0 Hz, 2H, Ar*H*), 7.99-7.18 (m, 28H, Ar*H*), 5.48 (s, 1H, H_1), 5.18 (app t, *J* = 9.6, 10.0 Hz, 1H, H_4'), 5.04-4.89 (m, 3H, H_1', Bn*H*), 4.82-4.61 (m, 5H, Bn*H*), 4.54 (d, *J* = 11.6 Hz, 1H, Bn*H*), 4.38-4.23 (m, 4H, H_2, H_5, H_5', H_6'), 4.06 (t, *J* = 9.6 Hz, 1H, H_3'), 3.97-3.89 (m, 2H, H_3, H_6'), 3.77 (t, *J* = 9.2 Hz, 1H, H_4), 3.76 (dd, 1H, *J* = 9.6, 2.8 Hz, H_2'), 1.99 (s, 3H, COCH_3), 1.45 (d, 3H, *J* = 6.0 Hz, CH_3).; ^{13}C-NMR (75 MHz, $CDCl_3$): δ 169.6 (*C*=O), 166.2 (*C*=O), 138.4, 138.6, 138.2, 138.0, 134.6, 133.1, 131.7, 129.9, 129.8, 129.1, 128.5, 128.4, 128.1, 127.9, 127.8, 127.7, 127.5,

127.3, 94.6 (C_1), 84.9 (C_1'), 80.4, 79.9, 79.2, 78.7, 75.3, 75.2, 74.5, 73.0, 72.3, 69.7, 69.4, 67.8, 62.5, 20.9 (CO*C*H_3), 17.9 (*C*H_3).; Calculated for $C_{55}H_{56}O_{11}SNa$ (M+Na) 947.3441 found 947.3457.

Phenyl 6-*O*-benzoyl-2,3-di-*O*-benzyl-*α*-D-glucopyranosyl-(1→2)-3,4-di-*O*-benzyl-1-thio-*α*-L-rhamnopyranoside (5)

Compound **4** (200 mg, 0.22 mmol) was dissolved in dry MeOH (15 mL), and the solution is cooled to 0 ^oC on an ice-bath. Acetyl chloride (0.16 mL, 2.2 mmol) was added drop wise, with continuous stirring, and the solution was kept on stirring under anhydrous condition for 2 hours. After completion of the reaction (indicated by TLC), excess MeOH and acetyl chloride were removed *in vacuo*. The resulting syrup was dissolved in DCM (15 mL), and the solution was washed subsequently with saturated aqueous $NaHCO_3$ (200 mL) and brine (100 mL). The organic layer was dried over Na_2SO_4 and concentrated. This was purified by column chromatography on silica gel (PE/EA, 4:1) to afford **5** as colorless syrup (168 mg, 88%).; $[\alpha]^{25}_D$ +48.5 (*c* 1.0, $CHCl_3$).; ^{1}H-NMR (400 MHz, $CDCl_3$): δ 8.07-8.05 (d, *J* = 7.6 Hz, 2H, Ar*H*), 7.24-7.61 (m, 28H, Ar*H*), 5.51 (s, 1H, H_1), 5.04-4.98 (m, 2H, Bn*H*), 4.96 (d, *J* = 3.4 Hz, 1H, H_1'), 4.87 (d, *J* = 11.2 Hz, 1H, Bn*H*), 4.78-4.63 (m, 5H, Bn*H*), 4.46 (dd, *J* = 12.0, 4.0 Hz, 1H, H_6'), 4.37 (bs, 1H, H_2), 4.26-4.20 (m, 3H, H_5, H_5', H_6'), 3.97 (t, *J* = 9.2 Hz, 1H, H_4'), 3.90 (dd, *J* = 2.6, 9.2 Hz, 1H, H_3), 3.75 (t, *J* = 9.6 Hz, 1H, H_3'), 3.62-3.56 (m, 2H, H_2', H_4), 1.43 (d, *J* = 6.0 Hz, 3H, C*H*$_3$).; ^{13}C-NMR (75 MHz, $CDCl_3$): δ 166.9 (*C*=O), 138.7, 138.4, 138.3, 137.9, 133.2, 131.5, 129.8, 129.7, 129.1, 128.54, 128.45, 128.4, 128.3, 128.1, 128.0, 127.9, 127.8, 127.7, 127.6, 127.4, 95.2 (C_1), 85.1 (C_1'), 80.6, 80.4, 79.7, 79.2, 75.4, 75.1, 72.7, 72.4, 70.0, 69.4, 63.4, 17.9 (C*H*$_3$).; Calculated for $C_{53}H_{54}O_{10}SNa$ (M+Na) 905.3336 found 905.3337.

_p_-Methoxylphenyl 2-_O_-acetyl-3,4-di-_O_-benzyl-_α_-L-rhamnopyranosyl-(1→2)-3,4,6-tri-_O_-benzyl-_α_-D-galactopyranoside (6)

To a mixture of phenyl 2-*O*-acetyl-3,4-di-*O*-benzyl-1-thio-*α*-L-rhamnopyranoside **10** (189.3 mg, 0.39 mmol) and *p*-methoxyphenyl 3,4,6-tri-*O*-benzyl-*α*-D-galactopyranoside **11** (200 mg, 0.36 mmol) in dry DCM (15 mL), flame activated molecular sieves (4Å) were added. It was stirred at room temperature under argon atmosphere. After 40 minutes the mixture was cooled to 0 °C, and NIS (90 mg, 0.39 mmol) was added to it. Then $FeCl_3$ (13.0 mg, 0.08 mmol) was added. After 15 minutes when the acceptor was consumed completely (checked by TLC) reaction mixture was filtered off through celite bed. The filtrate was diluted with DCM and washed subsequently with saturated sodium thiosulphate (100 mL), $NaHCO_3$ solution (100 mL), and water (100 mL). The organic layer was dried over anhydrous Na_2SO_4 and concentrated to afford the glycosylated product. The residue was purified by flash column chromatography (PE/EA, 5:1) to afford the title compound **6** as colorless syrup (305.8 mg, 92%).; $[\alpha]^{25}_{D}$ +19.9 (*c* 6.5, $CHCl_3$).; ^{1}H-NMR ($CDCl_3$, 400 MHz): δ 7.42-7.21 (m, 25H, Ar*H*), 7.01-6.99 (m, 2H, Ar*H*), 6.78-6.75 (d, *J* = 9.2 Hz, 2H, Ar*H*), 5.58 (dd, *J* = 2.8, 2.0 Hz, 1H, H_2'), 5.50 (d, *J* = 3.6 Hz, 1H, H_1), 5.10 (d, *J* = 0.8 Hz, 1H, H_1'), 4.96 (d, *J* = 11.2 Hz, 1H, Bn*H*), 4.86 (d, *J* = 10.8 Hz, 1H, Bn*H*), 4.79-4.68 (m, 3H), 4.59-4.48 (m, 3H, Bn*H*), 4.43-4.35 (ABq, *J* = 11.6 Hz, 2H, Bn*H*), 4.31 (dd, *J* = 10.0, 3.6 Hz, 1H, H_2), 4.13-4.04 (m, 3H, H_3, H_4, H_6), 3.95 (dd, *J* = 9.6, 3.2 Hz, 1H, H_3'), 3.78 (m, 1H, H_5'), 3.75 (s, 3H, OCH_3), 3.53-3.60 (m, 2H, H_5, H_6), 3.37 (app t, *J* = 9.6, 9.2 Hz, 1H, H_4'), 2.13 (s, 3H, COCH_3), 1.00 (d, *J* = 6.4 Hz, 3H, CH_3).; ^{13}C-NMR ($CDCl_3$, 75 MHz): δ 170.1 (*C*=O), 155.0, 151.0, 138.6, 138.4, 138.3, 138.1, 137.9, 128.5, 128.4, 128.3, 128.1, 127.8, 127.7, 127.6, 118.1, 114.6, 99.9 (C_1'),

97.9 (C_1), 79.9, 78.3, 77.9, 76.3, 75.1, 75.0, 74.8, 73.4, 73.2, 71.7, 69.9, 68.6, 68.4, 55.6 (O*C*H$_3$), 21.1 (CO*C*H$_3$), 17.7 (*C*H$_3$).; Calculated for $C_{56}H_{60}O_{12}Na$ (M+Na) 947.3983 found 947.4022.

_p_-Methoxylphenyl 3,4-di-*O*-benzyl-*α*-L-rhamnopyranosyl-(1→2)-3,4,6-tri-*O*-benzyl-*α*-D-galactopyranoside (7)

To a solution of **6** (250 mg, 0.27 mmol) in dry DCM (5 mL) and dry MeOH (5 mL) 1 M methanolic NaOMe (0.2 mL) solution was added, and the reaction mixture was stirred for 3 hours at room temperature. The solvent was removed under pressure and the crude mixture was diluted with DCM (10 mL), and subsequently washed with brine solution (100 mL), and washings were concentrated under reduced pressure. The residue was purified by column chromatography on silica gel (PE/EA, 5:1) to give glassy syrupy product **7** (236 mg, 99%).; $[\alpha]^{27}_D$ +21.9 (*c* 3.12, $CHCl_3$).; ^{1}H-NMR (400 MHz, $CDCl_3$): δ 7.31-7.22 (m, 25H, Ar*H*), 7.05-7.02 (d, *J* = 8.8 Hz, 2H, Ar*H*), 6.79-6.77 (d, *J* = 9.2 Hz, 2H, Ar*H*), 5.57 (d, *J* = 3.2 Hz, 1H, H_1), 5.13 (s, 1H, H_1'), 4.96 (d, *J* = 11.6 Hz, 1H, Bn*H*), 4.83 (d, *J* = 11.2 Hz, 1H, Bn*H*), 4.85 -4.70 (ABq, *J* = 11.6 Hz, 2H, Bn*H*), 4.69-4.62 (ABq, *J* = 11.6 Hz, 2H, Bn*H*), 4.61-4.58 (d, *J* = 11.2 Hz, 2H, Bn*H*), 4.44-4.36 (q, *J* = 11.6 Hz, 2H, Bn*H*), 4.31 (dd, *J* = 10.0, 3.6 Hz, 1H, H_2), 4.13-4.04 (m, 4H, H_3/ H_3' H_4, H_2', H_5'), 3.87 (dd, *J* = 3.0, 9.0 Hz, 1H, H_3/H_3'), 3.80 (dd, *J* = 9.6, 6.4 Hz, 1H, H_5), 3.75 (s, 3H, OCH_3), 3.62 (m, 1H, H_6), 3.54 (dd, *J* = 9.2, 6.0 Hz, 1H, H_6), 3.41 (t, *J* = 9.2 Hz, 1H, H_4'), 0.95 (d, *J* = 6.0 Hz, 3H, CH_3).; ^{13}C-NMR (75 MHz, $CDCl_3$): δ 154.9, 151.0, 138.6, 138.5, 138.4, 138.0, 137.9, 128.6, 128.5, 128.4, 128.32, 128.3, 127.9, 127.82, 127.8, 127.7, 127.4, 118.0, 114.6, 101.8 (C_1'), 97.8 (C_1), 79.9, 79.8, 78.1, 77.4, 75.1, 74.9, 73.4, 73.1, 72.0, 69.9, 68.8, 68.7, 68.0, 55.6 (O*C*H$_3$), 17.6 (*C*H$_3$).; Calculated for $C_{54}H_{58}O_{11}Na$ (M+Na) 905.3877 found 905.3943.

_p_-Methoxyphenyl 2-_O_-benzoyl-3,4-di-_O_-benzyl-_α_-L-rhamnopyranosyl-(1→4)-6-_O_-benzoyl-2,3-di-_O_-benzyl-_α_-D-glucopyranosyl-(1→2)-3,4-di-_O_-benzyl-_α_-L-rhamnopyranosyl-(1→2)-3,4-di-_O_-benzyl-_α_-L-rhamnopyranosyl-(1→2)-3,4,6-tri-_O_-benzyl-_α_-D-galactopyranoside (2).

A mixture of **3** (73.6 mg, 0.13 mmol) and **5** (100.0 mg, 0.113 mmol) and flame activated 4Å molecular sieves were stirred in dry DCM (10 mL) for 40 minutes at room temperature under argon atmosphere. The mixture was cooled to -60 ºC, and $FeCl_3$ (2.2 mg, 0.013 mmol) was added, and the reaction mixture was allowed to achieve room temperature. After 45 minutes complete consumption of both the starting materials was observed (Checked by TLC). Acceptor **7** (89.7 mg, 0.10 mmol) and NIS (25.4 mg, 0.113 mmol) were then added to the same vessel. After the addition reaction vessel was placed in a 0 °C cold bath and another 10 mol% of $FeCl_3$ (2.2 mg, 0.013 mmol) was added to it. The second step of the reaction was completed within 10 minutes (indicated by TLC). The reaction mixture was quenched by Et_3N and then filtered through celite bed. The bed was washed with DCM (3×15 mL). The combined filtrate and washings was washed subsequently with saturated sodium thiosulphate, aqueous $NaHCO_3$ (2x50 mL) and water (2x50 mL). The organic layer was dried over anhydrous Na_2SO_4 and concentrated to furnish a syrupy compound. The crude product was purified by flash column chromatography (eluent: toluene/Et_2O, 9.5:0.5) to afford the desired fully protected pentasaccharide **2** as white foam (184.5 mg, 78%).; $[\alpha]^{27}_D$ +28.1 (c 1.1, $CHCl_3$).; ^{1}H-NMR (500 MHz, $CDCl_3$): δ 8.06-8.03 (d, *J* = 7.0 Hz, 2H, Ar*H*), 8.01-7.98 (d, *J* = 7.5 Hz, 2H, Ar*H*), 7.57-7.48 (m, 2H, Ar*H*), 7.43-7.03 (m, 56H, Ar*H*), 7.01-6.98 (d, *J* = 9.0 Hz, 2H, Ar*H*), 6.77-6.74 (d, *J* = 9.0 Hz, 2H, Ar*H*), 5.53 (d, *J* = 3.5 Hz, 1H, H_1), 5.46 (t, *J* = 2.5 Hz, 1H, H_2""), 5.21 (bs, 1H, H_1""), 5.03 (bs, 2H, H_1', H_1''), 4.97-4.93 (m, 3H, Bn*H*), 4.87-4.75 (m, 5H, Bn*H*), 4.73-4.53 (m,

10H, H_1''', Bn*H*), 4.43 (d, *J* = 12.0 Hz, 1H, Bn*H*), 4.41-4.33 (ABq, *J* = 11.5 Hz, 2H, Bn*H*), 4.32-4.24 (m, 3H, H_2, H_6''', Bn*H*), 4.19-4.12 (m, 4H, H_2', H_2'', H_6''', H_5''''), 4.06 (app t, *J* = 6.5, 6.0 Hz, 1H, H_5'''), 4.04-4.01 (m, 2H, H_4, H_3''''), 3.97-3.86 (m, 5H, H_3, H_3'', H_3''', H_4''', Bn*H*), 3.84-3.78 (m, 1H, H_5'', 1dd, *J* = 10.0, 3.0 Hz, 1H, H_3'), 3.75-3.68 (1s, OCH_3, m, 1H, H_5'), 3.64 (app t, *J* = 7.0 Hz, 1H, H_4''), 3.61-3.49 (m, 4H, H_5, H_6, H_6, H_4'''), 3.40 -3.34 (m, 2H, H_4', H_2'''), 1.27 (d, *J* = 6.5 Hz, 3H, CH_3''''), 1.19 (d, *J* = 6.0 Hz, 3H, CH_3''), 0.94 (d, *J* = 6.0 Hz, 3H, CH_3').; ^{13}C-NMR (125 MHz, $CDCl_3$): δ 166.1 (*C*=O), 165.8 (*C*=O), 155.1, 151.2, 138.9, 138.87, 138.8, 138.7, 138.6, 138.5, 138.4, 138.3, 138.2, 133.1, 133.0, 130.3, 130.26, 130.0, 129.9, 128.7, 128.69, 128.64, 128.5, 128.4, 128.31, 128.3, 128.2, 128.1, 127.9, 127.89, 127.86, 127.83, 127.77, 127.7, 127.65, 127.60, 127.5, 127.4, 127.1, 118.2, 114.8, 101.6 (C_1''), 98.2 (C_1'), 98.0 (C_1), 97.9 (C_1''''), 94.1 (C_1'''), 80.62, 80.6, 80.5, 80.1, 79.8, 78.2, 78.1, 78.0, 75.6, 75.5, 75.34, 75.3, 75.2, 75.13, 75.10, 74.3, 73.5, 73.2, 73.1, 72.6, 72.4, 71.9, 71.6, 70.1, 69.8, 69.04, 69.0, 68.8, 68.7, 68.67, 62.9, 55.8 (O*C*H_3), 18.3 (*C*H_3), 18.1 (*C*H_3), 17.9 (*C*H_3).; Calculated for $C_{128}H_{132}O_{26}Na$ (M+Na) 2107.8905 found 2107.8908.

_p_-Methoxyphenyl 2-_O_-benzoyl-3,4-di-_O_-benzyl-α-L-rhamnopyranosyl-(1→4)-6-_O_-benzoyl-2,3-di-_O_-benzyl-α-D-glucopyranosyl-(1→2)-3,4-di-_O_-benzyl-α-L-rhamnopyranosyl-(1→2)-3,4-di-_O_-benzyl-α-L-rhamnopyranosyl-(1→2)-3,4,6-tri-_O_-benzyl-α-D-galactopyranoside (2).

A mixture of **3** (73.7 mg, 0.13 mmol) and **5** (100.0 mg, 0.0.113 mmol) and flame activated 4Å molecular sieves were stirred in dry DCM (10 mL) for 40 minutes at room temperature under argon atmosphere. The mixture was cooled to -60 ºC, and $FeCl_3$ (2.2 mg, 0.013 mmol) was added, and the reaction mixture was allowed to achieve room temperature. After 45 minutes complete consumption of both the starting materials was observed (Checked by TLC).

The reaction was cooled to -60^{o} C, and to this Ph_2SO (50.2 mg, 0.25 mmol), TTBP (42.1 mg, 0.17 mmol) and Tf_2O (0.03 ml, 0.14 mmol) were added one by one. Then the reaction mixture was slowly brought to -40^{o} C, and kept at that temperature for another 1hour, in which phenyl 3,4-di-*O*-benzyl-1-thio-*α*-L-rhamnopyranoside **9** (44.4 mg, 0.10 mmol) in dry DCM (2 mL) was added. The reaction mixture was slowly warmed to room temperature. After full consumption of starting materials (checked by TLC) acceptor **11** (58.0 mg, 0.10 mmol) and NIS (22.5 mg, 0.10 mmol) were then added to the same vessel. After the addition reaction vessel was placed in a 0 °C cold bath and another 10 mol% of $FeCl_3$ (2.2 mg, 0.0078 mmol) was added to it. The reaction was completed within 10 minutes (indicated by TLC). The reaction mixture was quenched by Et_3N and then filtered through celite bed. The bed was washed with DCM (3×15 mL). The combined filtrate and washings was washed subsequently with saturated sodium thiosulphate (2x50 mL), aqueous $NaHCO_3$ (2x50 mL) and water (2x50 mL). The organic layer was dried over anhydrous Na_2SO_4 and concentrated to furnish a syrupy compound. The crude product was purified by flash column chromatography (eluent: toluene/diethyl ether, 9.5:0.5) to afford the desired fully protected pentasaccharide **2** as white foam (188.7 mg, 72%); Spectral data matches with the substrate synthesized previously.

***p*-Methoxyphenyl 3,4-di-*O*-benzyl-*α*-L-rhamnopyranosyl-(1→4)-benzyl-2,3-di-*O*-benzyl-*α*-D-glucopyranosyluronate-(1→2)-3,4-di-*O*-benzyl-*α*-L-rhamnopyranosyl-(1→2)-3,4-di-*O*-benzyl-*α*-L-rhamnopyranosyl-(1→2)-3,4,6-tri-*O*-benzyl-*α*-D-galactopyranoside (26).**

To a solution of **2** (150 mg, 0.075 mmol) in DCM (5 mL) and dry MeOH (5 mL) 1 M methanolic NaOMe (0.2 mL) solution was added, and the reaction mixture was stirred for 3 hours at room temperature. The solvent was removed under pressure, coevaporated with toluene and the crude mixture was diluted with DCM (10 mL), and subsequently washed with brine solution (100 mL).

The combined organic layer was dried over anhydrous Na_2SO_4 concentrated under reduced pressure. The crude material was used for the next step without any purification. To this crude debenzoylated pentasaccharide derivative in DCM-H_2O (2:1, 10 mL), TEMPO (5.9 mg, 0.038 mmol) was added followed by BAIB (72.4 mg, 0.23 mmol) and the two-phase reaction mixture was stirred vigorously at room temperature for 8 hours till TLC (4:1 toluene/acetone) indicated complete conversion of the starting material to a lower running spot. The mixture was diluted with DCM (10 mL) and subsequently washed with saturated sodium thiosulphate solution (100 mL) and saturated $NaHCO_3$ solution (100 mL). The combined organic layer was dried over anhydrous Na_2SO_4 concentrated under reduced pressure and used for further reaction without any purification. Anhydrous K_2CO_3 (12.4 mg, 0.09 mmol) followed by BnBr (10 μL, 0.09 mmol) was added at 0 ^oC under argon to a solution of the foregoing material in dry DMF (10 mL), and the mixture was allowed to warm to room temperature. The organic layer was dried, concentrated, and co-evaporated with toluene. The crude mixture was diluted with DCM (10 mL), and subsequently washed with brine solution (100 mL). The combined organic layer was dried over anhydrous Na_2SO_4 concentrated under reduced pressure. Purification of this crude mass by silica gel Column chromatography (toluene/acetone 9:1) yielded the desired benzyl urinate **26** (124.7 mg, 79% over three steps).; $[\alpha]^{25}_D$ +15.1 (c 1.0, $CHCl_3$).; ^{1}H-NMR (500 MHz, $CDCl_3$): δ 7.34-7.18 (m, 60H, Ar*H*), 6.99-6.97 (d, *J* = 9.2 Hz, 2H, Ar*H*), 6.77-6.74 (d, *J* = 8.8 Hz, 2H, Ar*H*), 5.51 (d, *J* = 3.2 Hz, 1H), 5.09-4.99 (m, 4H), 4.94-4.74 (m, 9H), 4.69-4.53 (m, 10H), 4.50-4.45 (m, 2H), 4.41-4.23 (m, 5H), 4.11-3.81 (m, 11H), 3.78-3.74 (m, 2H), 3.75 (s, 3H, OC*H*$_3$), 3.68 (m, 1H), 3.59-3.49 (m, 4H), 3.37 (m, 1H), 3.26 (app t, *J* = 9.6, 9.2 Hz, 1H), 1.23 (d, *J* = 6.0 Hz, 3H, C*H*$_3$), 1.00 (d, *J* = 6.0 Hz, 3H, C*H*$_3$), 0.86 (d, *J* = 6.0 Hz, 3H, C*H*$_3$).; ^{13}C-NMR (100 MHz, $CDCl_3$): δ 170.0 (*C*=O), 155.1, 151.2, 139.4, 138.9, 138.8, 138.7, 138.6, 138.5,

138.4, 138.3, 138.2, 138.1, 135.0, 128.7, 128.68, 128.6, 128.5, 128.45, 128.4, 128.35, 128.3, 128.2, 128.1, 128.0, 127.7, 127.6, 127.56, 127.52, 127.5, 127.2, 118.1, 114.7, 114.2, 101.4, 99.5, 98.5, 97.9, 95.7, 80.2, 79.9, 78.0, 77.5, 77.4, 75.5, 75.3, 75.2, 75.1, 74.6, 73.5, 73.2, 72.23, 72.2, 72.0, 71.83, 71.8, 70.8, 70.0, 69.0, 68.9, 68.7, 68.3, 68.2, 67.6, 55.8 (O*C*H_3), 18.1 (*C*H_3), 18.0 (*C*H_3), 17.9 (*C*H_3).; Calculated for $C_{121}H_{128}O_{25}K$ (M+K) 2020.8416 found 2020.8438.

_p_-Methoxyphenyl _α_-L-rhamnopyranosyl (1→4)-_α_-D-glucopyranosyluronic acid-(1→2)-_α_-L-rhamnopyranosyl-(1→2)-_α_-L-rhamnopyranosyl-(1→2)-_α_-D-galactopyranoside (1)

A mixture of protected acidic pentasaccharide **35** (75 mg, 0.038 mmol) the resulting mixture and 10% Pd-C (90 mg) was taken in ethyl acetate (1 mL), methanol (3 mL) and H_2O (1 mL), and kept on stirring under H_2 atmosphere for 3 hours. The catalyst was filtered through celite bed, and the bed was washed with methanol (3 x 5 mL). The combined filtrate and washings was concentrated under reduced pressure. It was passed through a 0.45µm Millipore membrane, and lyophilized to afford **1** as white foam (31.3 mg, 92 %).; $[\alpha]^{25}_{D}$ +45.5 (*c* 0.37, H_2O).; ^{1}H-NMR (400 MHz, D_2O): δ 7.08-7.01 (d, *J* = 8.8 Hz, 2H, Ar*H*), 6.95-6.92 (d, *J* = 9.2 Hz, 2H, Ar*H*), 5.60 (d, *J* = 3.6 Hz, 1H, H_1), 5.09 (s, 1H, H_1'), 5.03 (s, 1H, H_1''), 4.99 (d, *J* = 3.6 Hz, 1H, H_1'''), 4.59 (s, 1H, H_1''''), 4.57 (s, 1H, H_5'''), 4.00 (dd, *J* = 10.4, 3.2 Hz, 1H, H_3), 4.05 (m, 1H, H_2'), 4.02-3.99 (m, 3H, H_4, H_5', H_2''), 3.97-3.91 (m, 2H, H_2, H_5), 3.88 (m, 1H, H_3'), 3.83 and 3.81 (each t, *J* = 3.2 Hz, 1H, H_3'', H_3''''), 3.75 (s, 3H, OCH_3), 3.78-3.68 (m, 3H, H_3, H_5''), 3.66 and 3.64 (bs, 2H, H_3''', H_4'''), 3.62-3.53 (m, 4H, H_4'', H_2''', H_2'''', H_5''''), 3.49-3.43 (t, *J* = 10.0 Hz, 2H, H_4', H_4''''), 3.40-3.35 (m, 2H, H_6, H_6), 1.25 (d, *J* = 6.4 Hz, 3H, CH_3''), 1.18 (d, *J* = 6.4 Hz, 3H, CH_3'), 0.91 (d, *J* = 6.0 Hz, 3H, CH_3'''').; ^{13}C-NMR (100 MHz, D_2O): δ 173.0 (*C*=O), 154.6, 150.2, 118.1, 115.2, 101.4, 101.0, 99.9, 97.9, 97.1, 79.2, 78.6, 77.3, 77.0, 71.9, 71.5, 71.34, 71.3, 70.5, 70.3, 70.2,

70.0, 69.6, 69.4, 69.3, 69.1, 68.5, 61.0, 55.9 (O*C*H_3), 16.6 (*C*H_3), 16.5 (*C*H_3), 16.4 (*C*H_3).; Calculated for $C_{37}H_{56}O_{25}Na$ (M+Na) 923.3009 found 923.3005.

ASSOCIATED CONTENT

SUPPORTING INFORMATION

Supporting Information : Syntheses of compounds **10**, **13**, **14** and **17**; ^{1}H- and ^{13}C-NMR spectra of compounds **1**-**8**, **10**, **11**, **13**, **14**, **17**, **19** and **20;** COSY spectra of compounds **1**, **2**, **4**, **6** and **11**; DEPT, HSQC, HMBC and NOSEY spectra of compound **2**. This material is available free of charge via the internet at http://pubs.acs.org.

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Notes

The authors declare no competing financial interest.

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