Carbohydrate Research 345 (2010) 10–22

Contents lists available at ScienceDirect

Carbohydrate Research

journal homepage: www.elsevier.com/locate/carres

Synthesis and antibacterial activity of aminosugar-functionalized intercalating agents

Wei Shi, Sandra L. Marcus, Todd L. Lowary *

Alberta Ingenuity Centre for Carbohydrate Science and Department of Chemistry, The University of Alberta, Gunning-Lemieux Chemistry Centre, Edmonton, AB, Canada T6G 2G2

ARTICLE INFO

Article history:
Received 4 October 2009
Received in revised form 10 October 2009
Accepted 13 October 2009
Available online 20 October 2009

Keywords:
Anthracyclines
Synthesis
Antibacterial

ABSTRACT

A series of previously reported amino sugar-functionalized intercalating agents, **3**–**14**, were evaluated in two antibacterial assays (paper disk diffusion and 96-well microdilution) against *Bacillus atrophaeus*, ATCC 9372 and *Escherichia coli*, ATCC 47076. Although none of the compounds were active against this *E. coli* strain, several showed activity against *B. atrophaeus*. In anticipation of the need for larger amounts of these compounds for future structure–activity relationship studies, improved routes to **11**–**14** were developed.

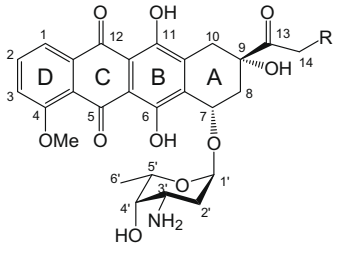
1. Introduction

Since the discovery of the pharmaceutical potential of daunorubicin (**1**, Dauno) and doxorubicin (**2**, Dox), two of the most important members of the anthracycline family of antibiotics (Chart 1),[1,2] significant research effort has been directed toward the identification of semi-synthetic analogs with better pharmaceutical efficacy. This work has led to the discovery of a few other anthracyclines for the clinical treatment of cancer: idarubicin, valrubicin, epirubicin, pirarubicin, and aclarubicin.[3] These anthracycline antibiotics remain in the mainstream of chemotherapeutics against a variety of tumors, either alone or in combination with other drugs.[4]

It is believed that one of the major modes of action of the anthracycline antibiotics arises from their binding to DNA, thus preventing transcription or translation, and, in turn, leading to cell death.[5,6] Indeed, previous structural studies have demonstrated that these molecules bind to DNA by intercalation of the anthraquinone moiety (rings B–D) between adjacent base pairs in the DNA helix, while ring A and the carbohydrate moiety interact with the minor groove.[7,8]

In addition to their anti-cancer potential, the earliest discovered anthracyclines, such as those in the rhodomycin family, also displayed potent antibacterial activity in culture.[9] However, their high toxicity in mice precluded further development as therapeutic drugs.[9] On the other hand, the anthracyclines identified later, such as Dauno, showed less toxicity at the dosage for clinical cancer treatment, but exhibited weaker antibacterial activity.[1a] Because most known naturally occurring anthracyclines are toxic to humans at the dosage required for treating bacterial infections, research on the use of anthracyclines as antibacterial agents has been limited.[10]

In an earlier paper,[11] we described the synthesis of a series of compounds (**3**–**14**, Chart 2) containing an aromatic domain linked to an amino sugar, including those found in both **1** and **2**. These compounds were designed as functional mimics of anthracyclines in that **3**–**14** possess a flat aromatic ring that can intercalate between DNA base pairs attached to a carbohydrate residue, which we anticipated could interact with the minor groove when the aromatic domain was intercalated into DNA. These compounds were, indeed, shown to bind to DNA, albeit less strongly than either **1** or **2**.[11] In this paper, we describe the testing of **3**–**14** for their ability to

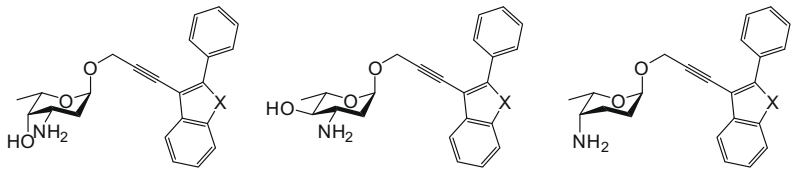


1. Daunorubicin: R = H
2. Doxorubicin: R = OH

Chart 1. Structures of daunorubicin (**1**, Dauno) and doxorubicin (**2**, Dox). The anthraquinone domain is composed of rings B, C, and D.

* Corresponding author. Tel.: +1 780 492 1861.
E-mail address: tlowary@ualberta.ca (T.L. Lowary).

0008-6215/$ - see front matter © 2009 Elsevier Ltd. All rights reserved.
doi:10.1016/j.carres.2009.10.010



3 X = O
4 X = S
5 X = NTs
6 X = NH

7 X = O
8 X = S
9 X = NTs
10 X = NH

11 X = O
12 X = S
13 X = NTs
14 X = NH

Chart 2. Structures of anthracycline analogs **3**–**14**.

act as antibacterial agents against model Gram-negative and Gram-positive organisms, as well as the refinement of the synthetic approaches to some of the target molecules for further biological evaluation, as well as structure–activity relationship (SAR) studies.

2. Results and discussion

2.1. Antibacterial activity of aminosugar analogs 3–14

To evaluate the potential antibacterial activity of **3–14**, one Gram-positive bacteria (*Bacillus atrophaeus*, ATCC 9372) and one Gram-negative bacteria (*Escherichia coli*, ATCC 47076) were selected as the target organisms to screen against. Two different antibacterial assays were applied, a paper disk diffusion assay and a 96-well microdilution assay.[12]

The results from the paper disk assay for **3**–**14** are listed in Table 1. Using this assay, none of these compounds prevented the growth of *E. coli*; therefore, only the results for *B. atrophaeus* are shown. Compared to Dauno and Dox, as well as three positive controls (streptomycin, penicillin, and tetracycline), **3**–**14** all showed much lower antibacterial activity. In addition, in this assay there is no correlation between the antibacterial activity and the DNA-binding affinity for these compounds determined in our earlier study.[11] However, it was found that the water solubility of many of the compounds was low, and we therefore anticipated that the diffusion of these analogs on the agar plate could be slow. To avoid this problem, we performed the more sensitive 96-well microdilution assay,[12] which requires a smaller amount of sample, thus allowing us to work at lower concentrations of each compound.

The MIC values (the minimum inhibition concentration at which 80–100% of bacterial growth is inhibited) resulting from the 96-well microdilution assays are also reported in Table 1. More specific MIC values could be obtained by additional assays at concentrations within this range; however, this was not done due to relatively low antibacterial activity of these compounds. It is only assured that at the top of the concentration range, the compound showed 100% inhibition, while at the lower concentration of the range obvious bacterial growth (>20%) was observed.

There are some obvious discrepancies between the results with each assay. For example, compound **7**, which has low water solubility as determined by the formation of a precipitate of the compound in the paper disk assay, did not show any inhibition in this assay, but it was among the most potent analogs in the microdilution assay. However, regardless of the assay method, compared to Dauno and Dox, which showed stronger DNA-binding affinity than **3**–**14**,[11] the antibacterial activity of the most potent synthetic analogs is about twofold weaker. Considering the slow diffusion of **3**–**14** in aqueous media, we consider the results from the 96-well microdilution assay to be more reliable. In this regard, it is of interest to note that, except for compound **12**, there is a good correlation between DNA-binding affinity and the MIC range; that is, strong DNA binding seems to benefit antibacterial activity.

Table 1
Paper disk diffusion and 96-well microdilution assay results for compounds **3**–**14** against *Bacillus atrophaeus*, ATCC 9372

Compound	Rel. DNA binding (% remaining EtBr)[a] (%)	Disk content of compound (nmol)	Zone diameter (mm)[b]	Range for MIC (μM)
Streptomycin	N/A	14 (10 μg)	19	ND[c]
Penicillin	N/A	19 (6.8 μg, 10 U)	28	ND
Tetracycline	N/A	68 (30 μg)	29	ND
Dauno	N/A	30	19	3.125–6.25
Dox	N/A	30	16	3.125–6.25
3	62.0	30	8	6.25–12.5
4	67.5	30	9	6.25–12.5
5	47.9	30	6	6.25–12.5
6	76.7	30	No inhibition	25–50
7	50.5	30	No inhibition	6.25–12.5
8	73.6	30	7	12.5–25
9	49.7	30	7	6.25–12.5
10	76.5	30	6	25–50
11	68.5	30	8	12.5–25
12	79.9	30	8	12.5–25
13	38.6	30	6	6.25–12.5
14	78.4	30	6	25–50

[a] Taken from Ref. 11. This number represents the percentage of remaining EtBr that is intercalated into DNA after incubation with the compound. The smaller the number, the greater the DNA-binding affinity of the compound.
[b] Mean values. All compounds were measured in duplicate.
[c] ND = not determined.

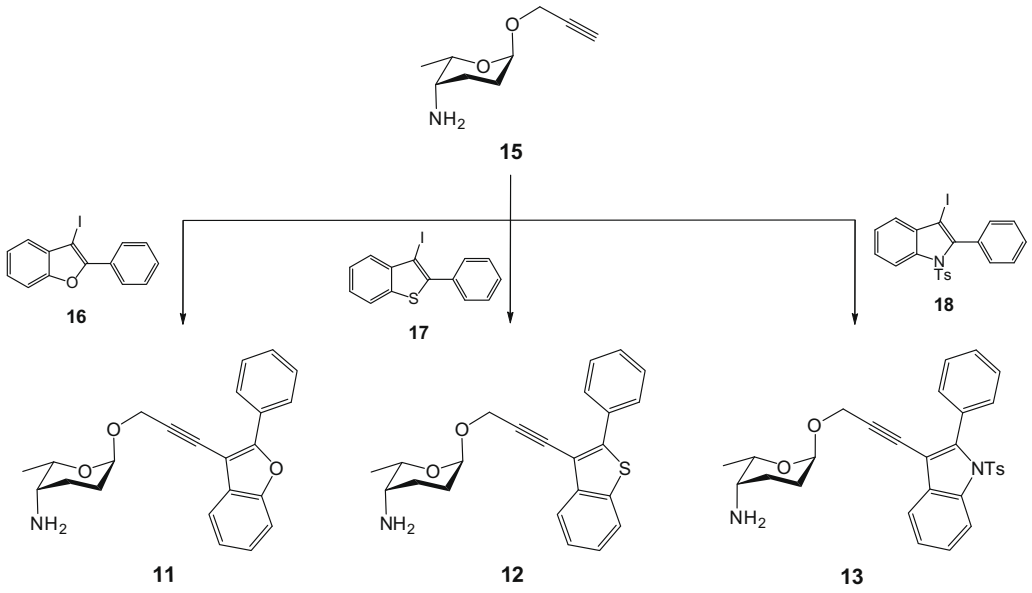


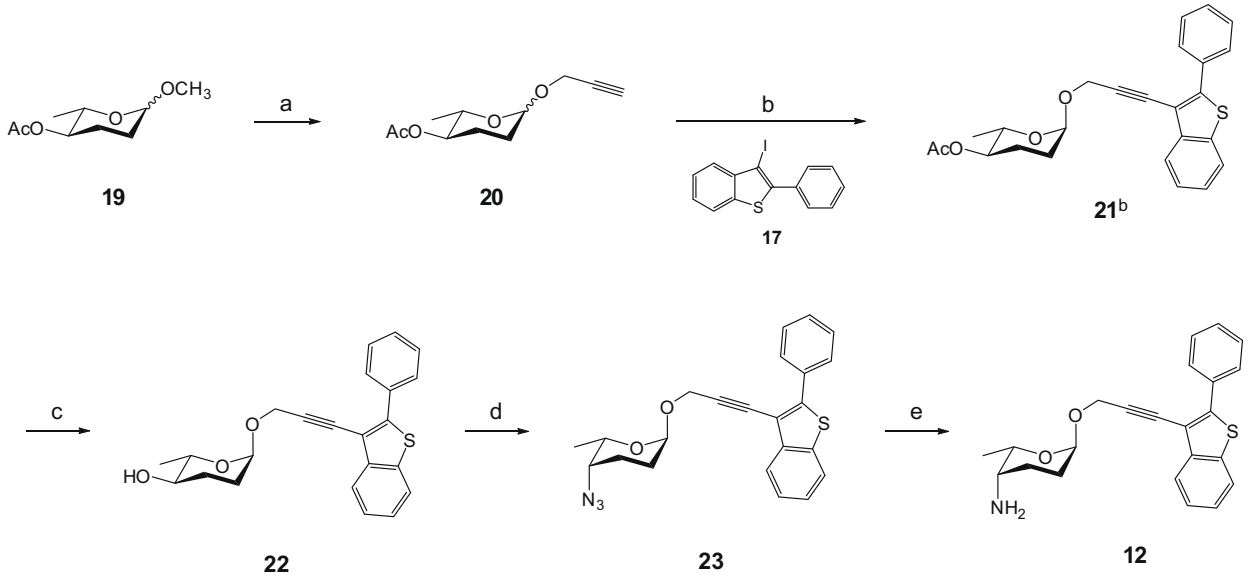
Figure 1. Previous synthesis of compounds **11**–**13** using $Pd(PPh_3)_2Cl_2$ and CuI as catalysts.[11]

2.2. Chemistry

Among analogs **3**–**14**, compound **13**, which consists of a 1-tosyl-2-phenylindol-3-yl group conjugated to a 4-amino-2,3,4,6-tetradeoxy-α-L-hexopyranoside moiety, showed the best DNA-binding affinity[11] and was also among the compounds with the best antibacterial activity against *B. atrophaeus*. However, the yield for its synthesis developed earlier was low (Fig. 1).[11] Direct Sonogashira coupling of the amino propargyl compound **15** with benzofuran or benzothiophene iodide **16** or **17** went smoothly, but almost no coupled product **13** could be observed under similar conditions. Even after the microwave irradiation was applied, the yield was still lower than 20%. Therefore, we decided to explore alternate approaches to **13**, which would enable the preparation of larger amounts of material for further studies.

2.2.1. Model studies

Before exploring an improved route to **13**, we carried out a series of model reactions using the phenylthiophene derivative **17** (Scheme 1), which was more synthetically accessible than the corresponding tosylated phenylindole species, and thus was available in larger amount. We first studied the Sonogashira coupling between **17** and propargyl glycoside **20**, which was synthesized in good yield from previously reported methyl glycoside **19**.[11] In the course of optimizing the reaction conditions (Table 2), removal of oxygen by sparging the solution with argon before the addition of the metal catalysts (entry 2 vs 1) was essential. It has been reported that the presence of oxygen greatly accelerates the homocoupling reaction of the alkyne,[13] and a large amount of this product was detected in the absence of argon. In addition, piperidine was found to be a better solvent than triethylamine (entries



Scheme 1. Reagents and conditions: (a) propargyl alcohol, $BF_3{\cdot}Et_2O$, 95%; (b) 5% $Pd(PPh_3)_2Cl_2$, 10% CuI, piperidine, 72%; (c) K_2CO_3, methanol, 99%; (d) PPh_3, DPPA, DIAD, −20 °C→ rt, 80%; (e) PPh_3, H_2O 87%. [b] The pure β anomers were isolated, and the subsequent transformations were performed separately see Section 2.2.3.

Table 2
Optimization of the Sonogashira reaction of **17** and **20**

	Mol % $Pd(PPh_3)Cl_2$ (%)	Mol % CuI (%)	Base	Sparging	Mol % PPh_3 (%)	Time (h)	Yield (%)
1	2–5	1–5	Et_3N	No	0	16	~5
2	5	1–2	Et_3N	Yes	0	12	40
3	5	1–2	Piperidine	Yes	0	12	74
4	3	1–2	Piperidine	Yes	0	14	65
5	5	10	Piperidine	Yes	0	10	91
6	5	10	Piperidine	Yes	10	27	89
7	5	10	Piperidine	Yes	3	12	93

3–7); the use of pyrrolidine led to partial removal of the acetyl group.[14] High loadings of both palladium (~5%) and copper catalysts (~10 mol %) were also necessary (entry 4 vs 3, 5 vs 4). Although triphenylphosphine has been reported to stabilize the palladium catalyst, especially under thermal conditions,[15] its addition showed no significant effect on the ambient reactions used here (entries 6 and 7 vs 5). Indeed, the use of 10% triphenylphosphine significantly reduced the reaction rate (entry 6 vs 5).

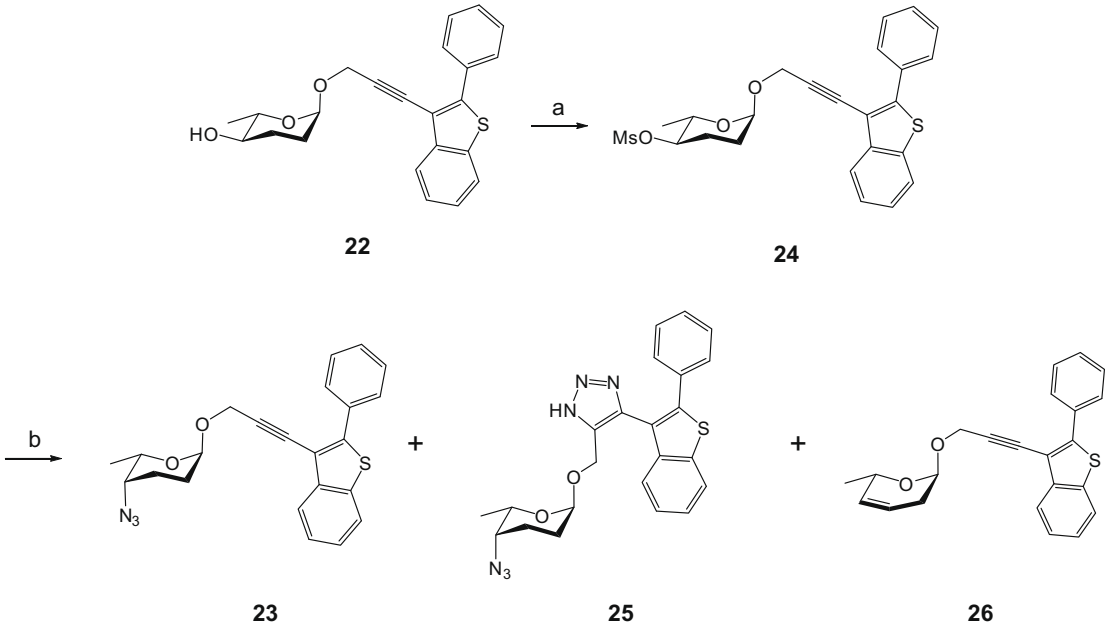
After the optimization of the Sonogashira coupling, removal of the acetyl-protecting group in **21** produced **22** in excellent yield. Next, Mitsunobu reaction with diphenylphosphoryl azide (DPPA) gave an 80% yield of **23**.[16] The last step, the conversion of the azide to an amine in using the Staudinger reaction proceeded smoothly to give **12** in 87% yield.

Prior to the use of DPPA for the synthesis of **23** from **22**, we explored two other approaches, both of which gave unexpected results. Initially, we thought it would be possible to install the azido group into **22** via S_N2 substitution of the corresponding mesylate derivative (Scheme 2). Thus, reaction of **22** with methanesulfonyl chloride and triethylamine in dichloromethane afforded compound **24** in near quantitative yield; however, the following substitution with sodium azide was not very successful. When the substitution reaction was carried out with sodium azide in DMF at 120 °C, the yield of the desired product **23** was only 40%. Two major side products were formed, the 1,2,3-triazole **25** (30% yield), produced from [2+3] cycloaddition between azide and the alkyne, and alkene **26** (19% yield) arising from elimination.

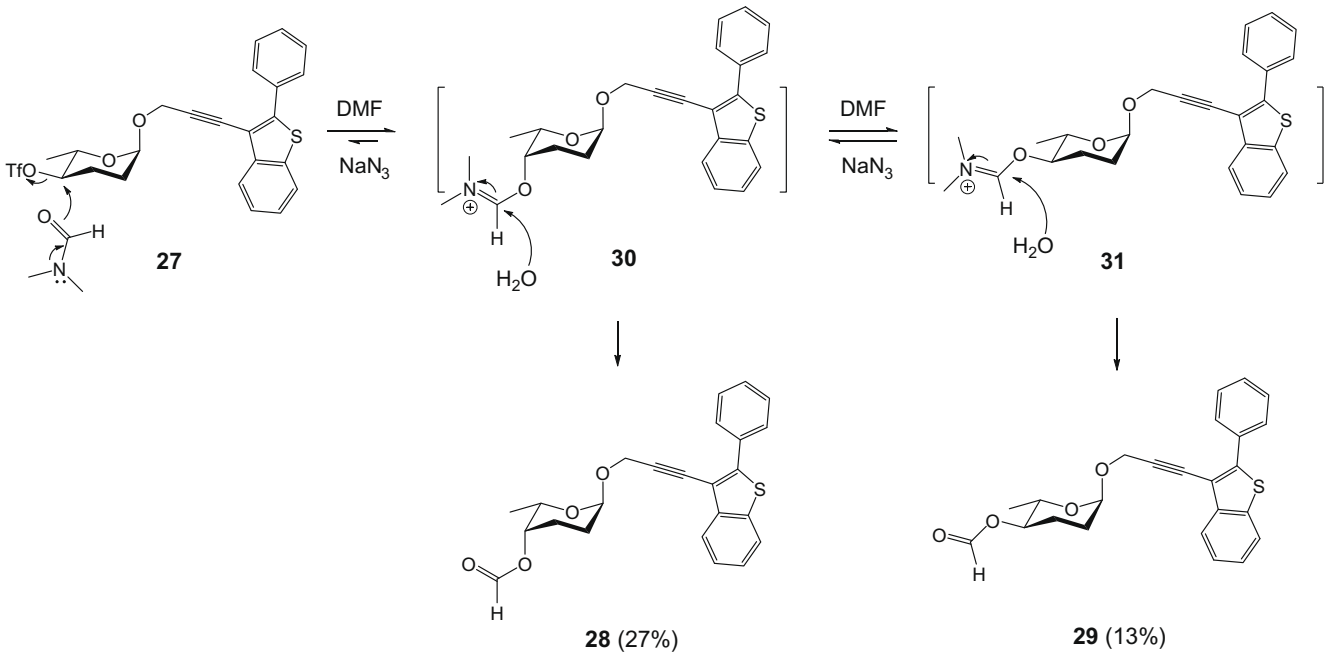
The structure of **25** was supported by mass spectrometry, ^1H NMR and IR spectroscopy. In the IR spectrum, the signal (~2200 cm^{-1}) for the alkyne disappeared, and signals (1733, 1600, 1433, 1215 cm^{-1}) for a 1,2,3-triazole appeared, although they are not very characteristic.[17] In the ^{13}C NMR spectrum, the alkyne signals at ~80 ppm and ~90 ppm also disappeared; however, three signals for quaternary aromatic carbons in **25** were missing, presumably due to their very long relaxation times. Efforts to change the pulse delay led to two of three signals being recovered, but not all three. The alkene compound **26** was confirmed by the shift of H/C-3 and H/C-4 signals from the alkane region to the alkene region in both the ^1H NMR and ^{13}C NMR spectra. Mass analysis also clearly verified the loss of one molecule of methanesulfonic acid. By decreasing the reaction temperature to 80–85 °C, these two byproducts were avoided to some degree, especially the 1,2,3-triazole, but the reaction time was substantially extended, usually requiring more than 2–3 days.

Another approach explored for the preparation of **23** from **22** involved the generation of the more reactive triflate derivative (Scheme 3). Thus, crude **27** after the workup of the triflation reaction of **22** (Tf_2O, pyridine) was treated with sodium azide in DMF at room temperature. As expected, none of triazole or elimination product was formed. However, the yield of **23** was only slightly improved (49%), and two new side products, formate esters **28** and **29**, were generated.

A possible mechanism for the formation of **28** and **29** is shown in Scheme 3. We propose that the formyl group is added via reac-



Scheme 2. Reagents and conditions: (a) MsCl, Et_3N, 0 °C, 99%; (b) NaN_3, DMF, 120 °C, 40% for **23**, 30% for **25**, 19% for **26**.



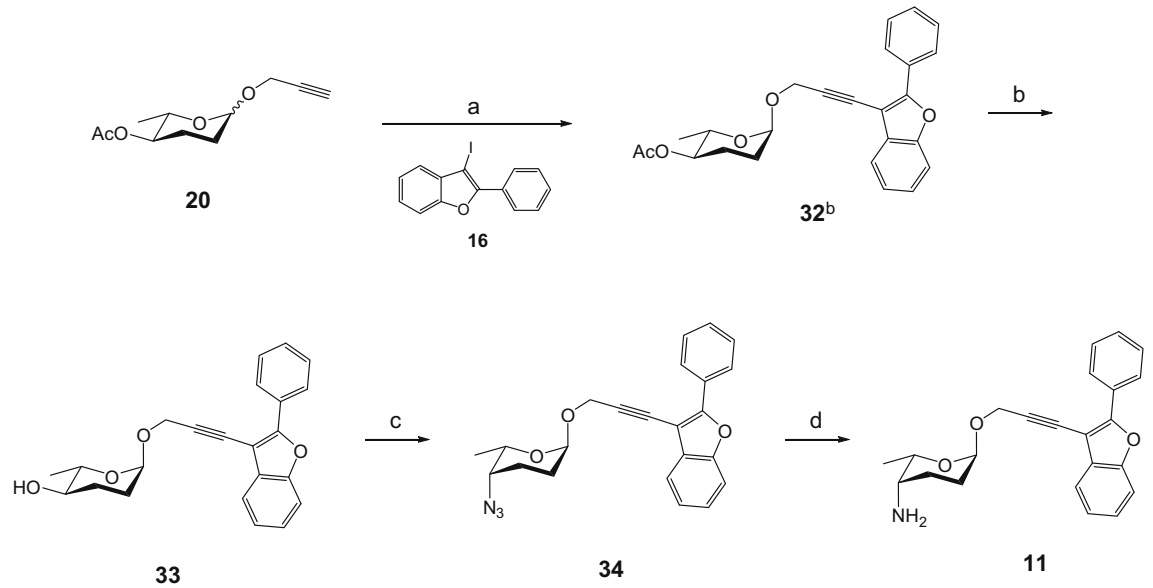
Scheme 3.

tion of triflate intermediate **27** with the solvent DMF. Although DMF is a poor nucleophile, it is present in large excess, and the concentration of sodium azide in the reaction mixture is relatively low due to its poor solubility in DMF at room temperature. The reaction would be further facilitated by the highly deoxygenated carbohydrate ring in **27**, which would enhance the electrophilicity of C-4, compared to the more oxygenated carbohydrate derivatives. We propose that intermediate **30** is first formed. Thereafter, due to the presence of a large amount of DMF, the stereochemistry of C-4 in **30** can be inverted to give **31**, and an equilibrium may be established between them. In theory, **31** should be favored due to the equatorial orientation of the iminium ion at C-4. However, after the aqueous workup, which gave rise to the release of dimethylamine to produce the corresponding formate esters, **29** was produced in a larger amount than **30**, by a ratio of ~2:1. Therefore, it appears that the conversion from **30** to **31** is a slow process.

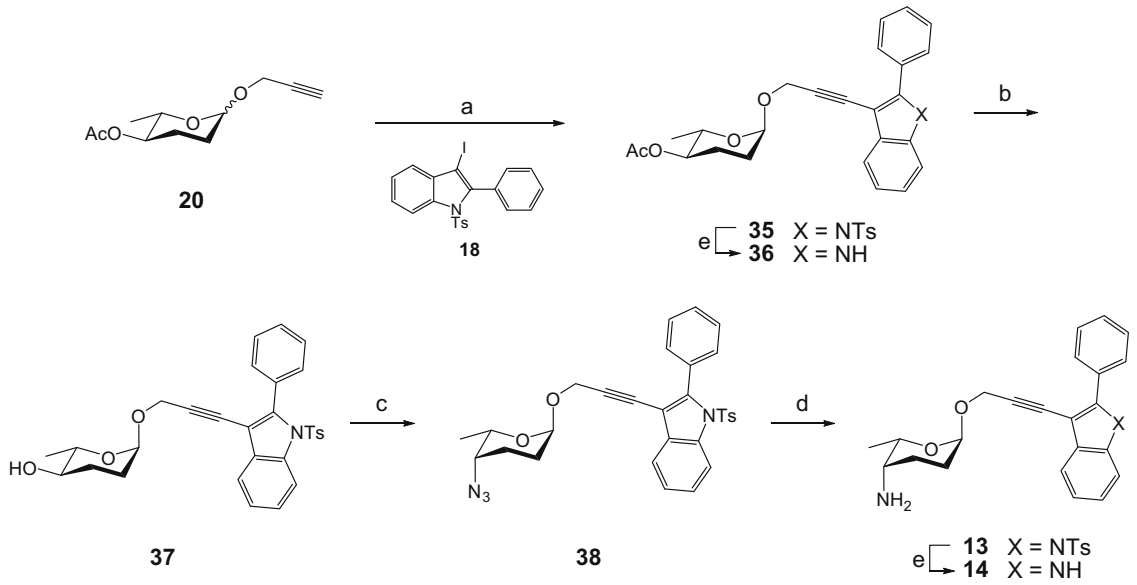
2.2.2. Synthesis of 11, 13 and 14

After succeeding in the synthesis of benzo[*b*]thiophene analogs **12**, we applied the same strategy outlined in Scheme 1 to the benzo[*b*]furan and indole-containing systems, **11** and **13**, respectively. As outlined in Scheme 4, using this route compound **11** was synthesized in 69% overall yield from **16** and **20** with no difficulties (Scheme 4).

The synthesis of the indole derivative **13** via the approach depicted in Scheme 1 turned out to be more challenging than for



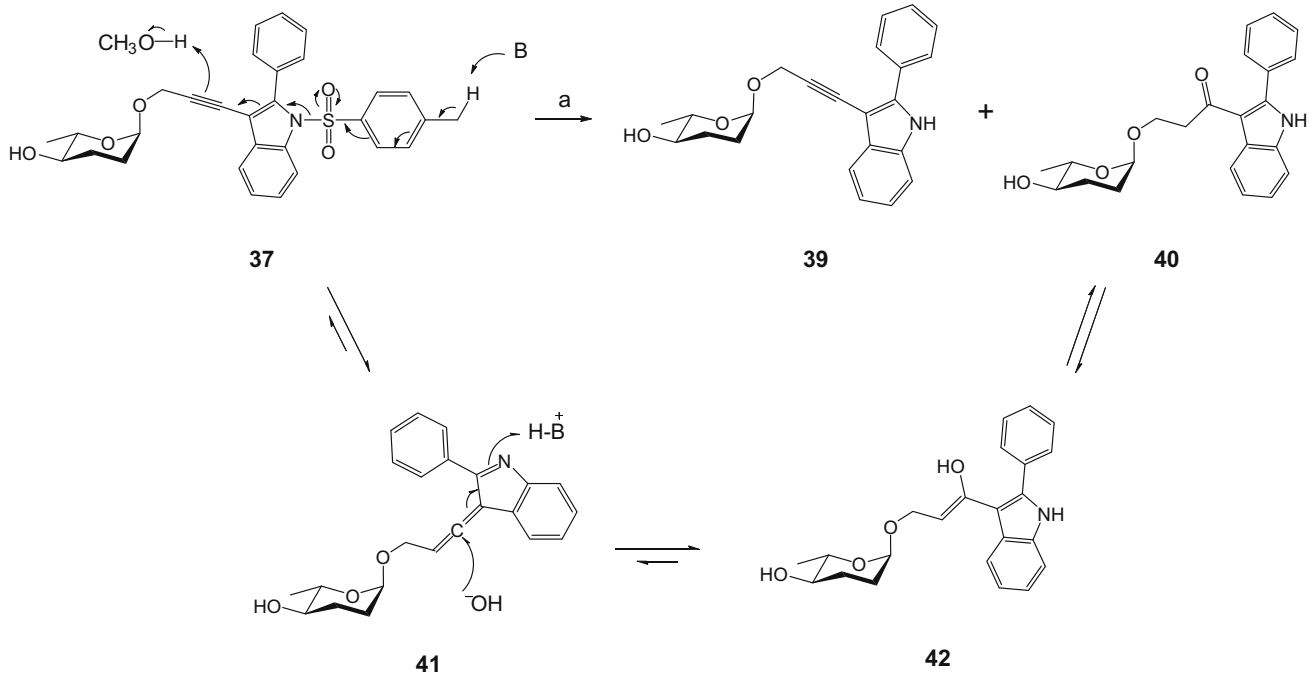
Scheme 4. Reagents and conditions: (a) 5% $Pd(PPh_3)_2Cl_2$, 10% CuI, piperidine, 73%; (b) K_2CO_3, methanol, 99%; (c) PPh_3, DPPA, DIAD, −20 °C→rt, 83%; (d) PPh_3, H_2O 91%. [b] The pure β anomers were isolated, and the subsequent transformations were performed separately, see Section 2.2.3.



Scheme 5. Reagents and conditions: (a) 5% $Pd(PPh_3)_2Cl_2$, 10% CuI, piperidine, 51%; (b) K_2CO_3, methanol, 89%; (c) PPh_3, DPPA, DIAD, −20 °C→rt, 73%; (d) PPh_3, H_2O 86%; (e) *n*-Bu_4NF, THF, rt, 61% for **36** and 75% for **14**. [b] The pure β anomers were isolated, and the subsequent transformations were performed separately see Section 2.2.3.

the thiophene or furan systems. Fortunately, the coupling between propargyl glycoside **20** and tosyl-protected indole iodide **18** was successful, despite giving the product **35** in relatively modest yield (Scheme 5). It was found that the increased loading of the palladium catalyst did not improve the reaction yield significantly. However, it was particularly crucial to protect the reaction from air during the entire process. The rest of the steps for the synthesis of target amine **13** from **35** were straightforward, although the reaction time for the deacylation of **35** needed to be controlled to minimize the concomitant cleavage of the tosyl group under the basic reaction conditions. The target **13**, was obtained from **35** in 56% yield over three steps. Removal of the tosyl group in **13** upon reaction with tetra-*n*-butylammonium fluoride gave the free indole **14** in 75% yield.

In identifying methods for removing the *N*-tosylate group in these indoles, we initially explored heating with potassium hydroxide in methanol at reflux.[18] In a model study, we attempted to deprotect **37** using these conditions; however, only ~20% of the desired free indole product **38**, contaminated with a number of other unknown compounds, was obtained (Scheme 6). The major product, obtained in 65% yield, was ketone **40**. A possible mechanism for the formation of **40** from **37** is illustrated in Scheme 6. After the removal of the tosyl group, protonation of the alkyne could give an allene **41**, which then undergoes a Michael-like



Scheme 6. Reagents and conditions: (a) KOH, CH_3OH, reflux, 65% for **40**.

1,4-conjugate addition of hydroxide anion generating the enol **42**, which tautomerizes to the more stable ketone **40**. Based on this mechanism, the formation of **40** can be prevented by employing an aprotic solvent and a base that is a poor nucleophile. The use of the tetra-*n*-butylammonium fluoride in THF thus satisfies both these criteria. In addition to being successful in cleaving the tosyl group from **13** to yield **14**, these conditions could also be used to prepare compound **36** from **35** (Scheme 5).

2.2.3. Synthesis of -glycoside analogs of 11–14 and related precursors

The synthesis of **11**–**14** as outlined above all began with propargyl glycoside **16**, which was synthesized as a 3.8:1 α/β mixture (see, e.g., Scheme 1). Although separation of this mixture of glycosides was possible, it was difficult and the purification was more conveniently done after the aromatic group was added via Sonogashira coupling. This approach also allowed the synthesis of 12 additional compounds, **43**–**54** (Chart 3) possessing the β stereochemistry at the anomeric center. The preparation of these compounds followed the routes outlined above for the corresponding α anomers, and details can be found in Supplementary data. Access to **43**–**54** will be useful in future SAR studies on this class of compounds.

2.3. Conclusions

In summary, we describe here the testing of compounds **3**–**14** as potential antibacterial agents against both Gram-negative and Gram-positive bacteria. Although none of the compounds showed activity against the Gram-negative organism evaluated (*E. coli*, ATCC 47076) using the preliminary disk diffusion assay, several of the compounds were active against *B. atrophaeus*, ATCC 9372, the model Gram-positive organism we investigated, by using both disk diffusion and microdilution assays. Some of the compounds possess low micromolar MIC values against *B. atrophaeus* thus suggesting that the synthesis of additional analogs of this class of compounds is warranted (e.g., through functionalization of the amino group). To that end, a more efficient synthetic approach to **13** has been developed, in which compared to an earlier route, the overall yield was enhanced from ~8% to ~35% from the same starting materials. The same approach could also be used for an improved synthesis of compounds **11**, **12,** and **14** and allowed the preparation of additional analogs, for example, those with β-glycoside stereochemistry.

3. Experimental

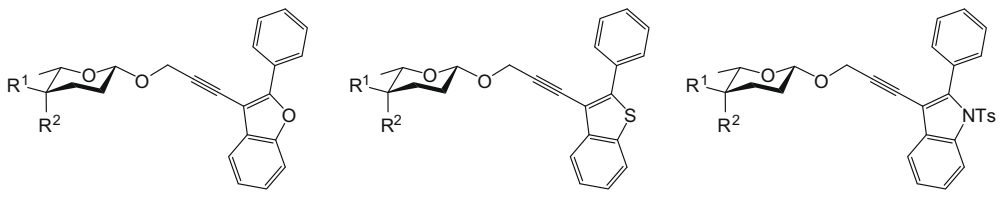
3.1. General methods—chemistry

All reagents were purchased from commercial sources and were used without further purification unless noted. Before use, reaction solvents were purified by successive passage through columns of alumina and copper in a PURESOLV-400 System from Innovative Technology, Inc. under an argon atmosphere. Unless stated otherwise, all reactions were carried out under a positive pressure of argon and were monitored by TLC on silica gel G-25 UV_{254} (0.25 mm, Macherey–Nagel). Spots were detected under UV light and/or by charring with 10% H_2SO_4 in ethanol, or in acidified ethanolic anisaldehyde or vanillin. Solvents were evaporated under reduced pressure and below 40 °C (water bath). Column chromatography was performed on Silica Gel 60 (40–60 μM). The ratio between silica gel and crude product ranged from 100 to 50:1 (w/w). Optical rotations were measured at 22 ± 2 °C. Melting points are uncorrected. ^{1}H NMR spectra were recorded on VARIAN INOVA NMR spectrometers at 400, 500 or 600 MHz, and chemical shifts are referenced to either TMS (0.0, $CDCl_3$) or CD_2HOD (4.78, CD_3OD). ^{13}C NMR spectra were recorded at 100 or 125 MHz, and ^{13}C chemical shifts are referenced to $CDCl_3$ (77.23 ppm, CD_3Cl_3) or CD_3OD (49.00 ppm, CD_3OD). ^{1}H data are reported as though they were first order. The errors between the coupling constants for two coupled protons were less than 0.5 Hz, and the average number was reported. Assignment of NMR resonances was done based on ^{1}H–^{1}H COSY, HMQC, and in some cases HMBC experiments. In the interpretation of the NMR data for methylene protons on the carbohydrate ring, 'a' and 'e' refer to axial and equatorial orientation, respectively. In the cases where no clear assignment of these hydrogens could be made based on all NMR data, assignments were made taking into consideration the anisotropy effect of a ring σ bond, which results in equatorial hydrogens resonating more downfield than axial hydrogens.[19] Electrospray-ionization mass spectra were recorded on samples suspended in mixtures of THF with CH_3OH and added NaCl. Optical rotations were measured on a Perkin–Elmer 241 polarimeter at the sodium D line (589 nm). Optical rotations are in units of deg mL(dm g)$^{-1}$. IR spectra were recorded on the Nicolet Magna 750 FTIR spectrometer. The reported purity values were obtained with a Varian HPLC system, using an evaporative light scattering detector (ELSD) 2000ES from Alltech, and a Varian Microsorb-MV 100-5 C18 column. The eluant consisted of acetonitrile and water, the ratio of which depends on the compound. For all basic amino compounds, 0.1% by volume of trifluoroacetic acid was added to facilitate elution and avoid aggregation. When the purity derived from HPLC analysis is greater than 99.5%, it will be reported as >99%.

3.2. General reaction procedures

3.2.1. General procedure for Sonogashira coupling

To a degassed solution of propargyl glycoside **20** (1.3 equiv), the aromatic iodide (1 equiv), $PdCl_2(PPh_3)_2$ (5 mol %) in piperidine (5–7 mL/mmol) was added CuI (10 mol %). The reaction mixture was stirred under Ar at rt and followed by TLC (10–16 h). Once complete, the reaction was quenched by the addition of a satd aq soln



43 R^1 = OAc, R^2 = H
44 R^1 = OH, R^2 = H
45 R^1 = H, R^2 = N_3
46 R^1 = H, R^2 = NH_2

47 R^1 = OAc, R^2 = H
48 R^1 = OH, R^2 = H
49 R^1 = H, R^2 = N_3
50 R^1 = H, R^2 = NH_2
51 R^1 = OMs, R^2 = H

52 R^1 = OH, R^2 = H
53 R^1 = H, R^2 = N_3
54 R^1 = H, R^2 = NH_2

Chart 3. Structures of the synthesized β anomers.

of NH_4Cl. The aqueous solution was then extracted with Et_2O, and the combined organic layers were washed with brine, dried (Na_2SO_4), filtered, and concentrated to yield the crude product, which was purified by column chromatography.

3.2.2. General procedure for deacetylation

The acetyl-protected compound (1 equiv) was dissolved in CH_3OH (30–40 mL/mmol). K_2CO_3 (30 mol %) was added, and then the reaction mixture was stirred at rt and followed by TLC (~12 h). After the evaporation of the solvent, the residue was diluted with water and extracted with CH_2Cl_2. The combined organic layer was dried (Na_2SO_4), filtered, and concentrated to yield the crude product, which was purified by column chromatography.

3.2.3. General procedure for Mitsunobu reactions

To a solution of the hydroxyl compound (1 equiv) in THF (15–20 mL/mmol) was added PPh_3 (3 equiv) at −20 °C. To this mixture was added a solution of DIAD (2.5 equiv) and DPPA (2.5 equiv) in THF (5–7 mL/mmol) at −20 °C. The reaction mixture was then allowed to warm to rt and followed by TLC (~12 h). The resulting solution was then diluted with Et_2O, washed with brine, dried (Na_2SO_4), filtered, and concentrated to yield the crude product, which was purified by column chromatography.

3.2.4. General procedure for Staudinger reactions

To a solution of the azido compound (1 equiv) in THF (30–40 mL/mmol) and H_2O (>50 equiv) was added PPh_3 (2.5 equiv), and the reaction was stirred under reflux and followed by TLC (~10 h). The solution was cooled to room temperature, and concentrated, and the residue was purified by column chromatography to yield the pure amino compound.

3.2.5. General procedure for tosylate removal

To a solution of the tosylate (1 equiv) in THF (30–40 mL/mmol) was added tetra-*n*-butylammonium fluoride (TBAF) solution in THF (1.0 M, 6 equiv) at rt, and the mixture was stirred at rt and followed by TLC (~18 h). Satd aq $NaHCO_3$ (30 mL) was then added, and the mixture was extracted with CH_2Cl_2. The organic layer was washed with brine, dried over Na_2SO_4, filtered, and concentrated to a residue that was purified by column chromatography.

3.3. 3-(2-Phenylbenzo[*b*]furan-3-yl)prop-2-ynyl 4-amino-2,3,4,6-tetradeoxy-α-L-*threo*-hexopyranoside (11)

This compound was obtained from **34** following the general procedure for Staudinger reactions. The product was purified by column chromatography (1:1 hexanes–EtOAc→10:1 CH_2Cl_2–CH_3OH) in 91% yield. The data matched that reported previously.[11]

3.4. 3-(2-Phenylbenzo[*b*]thiophen-3-yl)prop-2-ynyl 4-amino-2,3,4,6-tetradeoxy-α-L-*threo*-hexopyranoside (12)

This compound was obtained from **23** following the general procedure for Staudinger reactions. The product was purified by column chromatography (1:1 hexanes–EtOAc→10:1 CH_2Cl_2–CH_3OH) in 87% yield. The compound characterization data could be found in Ref. 11.

3.5. 3-(1-Tosyl-2-phenylindol-3-yl)prop-2-ynyl 4-amino-2,3,4,6-tetradeoxy-α-L-*threo*-hexopyranoside (13)

This compound was obtained from **38** following the general procedure for Staudinger reactions. The product was purified by column chromatography (1:1, hexanes–EtOAc→10:1, CH_2Cl_2–CH_3OH) in 86% yield. The data matched that reported previously.[11]

3.6. 3-(2-Phenylindol-3-yl)prop-2-ynyl 4-amino-2,3,4,6-tetradeoxy-α-L-*threo*-hexopyranoside (14)

This compound was obtained from **13** following the general procedure for detosylation. The product was purified by column chromatography (10:1, CH_2Cl_2–CH_3OH) in 75% yield. The data matched that reported previously.[11]

3.7. 2-Propargyl 4-*O*-acetyl-2,3,6-trideoxy-α-L-*erythro*-hexopyranoside (20α) and 2-propargyl 4-*O*-acetyl-2,3,6-trideoxy-β-L-*erythro*-hexopyranoside (20β)

To a flask were added a mixture of **19** (455 mg, 2.42 mmol), crushed activated 4 Å molecular sieves (290 mg) and propargyl alcohol (10 mL). The mixture was stirred for 10 min at rt, cooled to −40 °C, and then $BF_3{\cdot}Et_2O$ (0.768 mL, 6.05 mmol) was added dropwise via a syringe. After adding the $BF_3{\cdot}Et_2O$, the reaction mixture was warmed to −20 °C. Once the starting material was fully consumed (about 5 h), the reaction mixture was neutralized by the addition of K_2CO_3 (500 mg). Next, H_2O (12 mL), satd aq $NaHCO_3$ (12 mL), and CH_2Cl_2 (30 mL) were added. The organic layer was separated, washed with brine, dried (Na_2SO_4), filtered, and concentrated. The crude product was purified by column chromatography (10:1 hexanes–EtOAc) to give pure **20α** as a colorless waxy solid and **20β** as a colorless oil (487 mg, 95%, α:β = 3.8:1). Data for **20α**: R_f 0.65 (6:1 hexanes–EtOAc); IR: ν 2216 (C≡C), 1736 (C=O) cm^{-1}; $[\alpha]_D$ −196.6 (*c* 0.9, CH_2Cl_2); ^1H NMR (400 MHz, $CDCl_3$): δ_H 4.86 (br s, 1H, H-1) 4.40–4.48 (m, 1H, H-4), 4.18 (dd, 1H, *J* = 15.8 Hz, *J* = 2.4 Hz, OC*H*$_2$C≡CH), 4.12 (dd, 1H, *J* = 15.8 Hz, *J* = 2.4 Hz, OC*H*$_2$C≡CH), 3.74 (dq, 1H, $J_{4,5}$ = 9.7 Hz, $J_{5,6}$ = 6.2 Hz, H-5), 2.38 (t, 1H, *J* = 2.4 Hz, OCH$_2$C≡C*H*), 1.98 (s, 3H, O=CCH$_3$), 1.68–1.90 (m, 4H, H-2a, H-2e, H-3a, H-3e), 1.09 (d, 3H, $J_{5,6}$ = 6.2 Hz, H-6); ^{13}C NMR (100 MHz, $CDCl_3$): δ_C 170.1 (C=O), 94.9 (C-1), 79.4 (C≡CH), 74.1 (C≡*C*H), 73.2 (C-4), 66.9 (C-5), 53.9 (OCH_2), 28.8 (C-2), 23.9 (C-3), 21.1 (O=C*C*H$_3$), 17.7 (C-6). ESIMS: *m*/*z* calcd for $[C_{11}H_{16}O_4]Na^+$: 235.0941. Found: 235.0943. Anal. Calcd for $C_{11}H_{16}O_4$: C, 62.25; H, 7.60. Found: C, 62.36; H, 7.78. Data for **20β**: R_f 0.63 (6:1 hexanes–EtOAc); IR: ν 2220 (C≡C), 1737 (C=O) cm^{-1}; $[\alpha]_D$ +56.1 (*c* 1.4, CH_2Cl_2); ^1H NMR (400 MHz, $CDCl_3$): δ_H 4.68 (dd, 1H, $J_{1,2a}$ = 9.0 Hz, $J_{1,2e}$ = 2.3 Hz, H-1), 4.45 (ddd, 1H, $J_{3a,4}$ = 10.4 Hz, $J_{4,5}$ = 8.9 Hz, $J_{3e,4}$ = 4.2 Hz, H-4), 4.34 (d, 2H, *J* = 2.4 Hz, OC*H*$_2$C≡CH), 3.53 (dq, 1H, $J_{4,5}$ = 8.9 Hz, $J_{5,6}$ = 6.2 Hz, H-5), 2.44 (t, 1H, *J* = 2.4 Hz, OCH$_2$C≡C*H*), 2.16 (dddd, 1H, $J_{3a,3e}$ = 13.0 Hz, $J_{2e,3e} = J_{3e,4} = J_{2a,3e}$ = 4.2 Hz, H-3e), 2.05 (s, 3H, O=CCH$_3$), 1.88–1.93 (m, 1H, H-2e), 1.66 (dddd, 1H, $J_{2a,2e} = -J_{2a,3a}$ = 13.0 Hz, $J_{1,2a}$ = 9.0 Hz, $J_{2a,3e}$ = 4.2 Hz, H-2a), 1.50 (dddd, 1H, $J_{3a,3e} = J_{2a,3a}$ = 13.0 Hz, $J_{3a,4}$ = 10.4 Hz, $J_{2e,3a}$ = 4.3 Hz, H-3a), 1.22 (d, 3H, $J_{5,6}$ = 6.2 Hz, H-6); ^{13}C NMR (100 MHz, $CDCl_3$): δ_C 170.2 (C=O), 98.9 (C-1), 79.3 (C≡CH), 74.4 (C≡CH), 73.3 (C-5), 72.7 (C-4), 55.0 (OCH_2), 29.7 (C-2), 26.9 (C-3), 21.1 (O=C*C*H$_3$), 18.1 (C-6). ESIMS *m*/*z* calcd for $[C_{11}H_{16}O_4]Na^+$: 235.0941. Found: 235.0944.

3.8. 3-(2-Phenylbenzo[*b*]thiophen-3-yl)prop-2-ynyl 4-*O*-acetyl-2,3,6-trideoxy-α-L-*erythro*-hexopyranoside (21) and 3-(2-phenylbenzo[*b*]thiophen-3-yl)prop-2-ynyl 4-*O*-acetyl-2,3,6-trideoxy-β-L-*erythro*-hexopyranoside (47)

This compound was synthesized from **20** (237 mg, 1.12 mmol), **17** (283 mg, 0.841 mmol),[11] $PdCl_2(PPh_3)_2$ (28 mg, 5 mol %), and CuI (15 mg, 10 mol %) in 91% combined yield with the β anomer (α:β = 3.8:1) by following general procedure for Sonogashira coupling. The product was purified by column chromatography (10:1 hexanes–EtOAc). Data for **21**: white needle-like solid after recrystallization from hexanes–Et_2O (2:1); mp: 89–91 °C; R_f 0.36 (6:1 hexanes–EtOAc); IR: ν 2216 (C≡C), 1736 (C=O) cm^{-1}; $[\alpha]_D$ −120.2 (*c* 1.4, CH_2Cl_2); ^1H NMR (400 MHz, $CDCl_3$): δ_H 7.96–8.00

(m, 2H, Ar), 7.92–7.96 (m, 1H, Ar), 7.79–7.82 (m, 1H, Ar), 7.36–7.50 (m, 5H, Ar), 5.08 (br s, 1H, H-1), 4.58 (s, 2H, OCH_2C≡C), 4.50–4.57 (m, 1H, H-4), 3.89 (dq, 1H, $J_{4,5}$ = 9.7 Hz, $J_{5,6}$ = 6.3 Hz, H-5), 2.06 (s, 3H, O=CCH_3), 1.80–2.00 (m, 4H, H-2a, H-2e, H-3a, H-3e), 1.17 (d, 3H, $J_{5,6}$ = 6.3 Hz, H-6); ^{13}C NMR (100 MHz, $CDCl_3$): δ_C 170.2 (C=O), 146.9 (Ar), 141.1 (Ar), 137.5 (Ar), 133.7 (Ar), 128.8 (Ar), 128.7 (2, Ar), 128.4 (2, Ar), 125.2 (Ar), 124.9 (Ar), 123.3 (Ar), 122.0 (Ar), 112.9 (Ar), 94.8 (C-1), 90.2 (≡C), 80.4 (≡C), 73.4 (C-4), 67.0 (C-5), 54.8 (OCH_2), 29.0 (C-2), 24.0 (C-3), 21.2 (O=CCH_3), 17.8 (C-6). ESIMS *m*/*z* calcd for $[C_{25}H_{24}O_4S]Na^+$: 443.1288. Found: 443.1289. Anal. Calcd for $C_{25}H_{24}O_4S$: C, 71.40; H, 5.75; S, 7.63. Found: C, 71.34; H, 5.80; S, 7.41. Data for **47**: yellow fluffy solid, R_f 0.32 (6:1 hexanes–EtOAc); IR: *ν* 2217 (C≡C), 1737 (C=O) cm^{-1}; $[\alpha]_D$ +32.7 (*c* 0.9, CH_2Cl_2); ^{1}H NMR (400 MHz, $CDCl_3$, δ_H) 7.96–7.99 (m, 2H, Ar), 7.92–7.95 (m, 1H, Ar), 7.79–7.82 (m, 1H, Ar), 7.36–7.50 (m, 5H, Ar), 4.83 (dd, 1H, $J_{1,2a}$ = 9.1 Hz, $J_{1,2e}$ = 2.2 Hz, H-1), 4.72 (ABq, 1H, *J* = 16.0 Hz, OCH_2C≡C), 4.69 (ABq, 1H, *J* = 16.0 Hz, OCH_2C≡C), 4.48 (ddd, 1H, $J_{3a,4}$ = 10.5 Hz, $J_{4,5}$ = 8.3 Hz, $J_{3e,4}$ = 4.3 Hz, H-4), 3.54 (dq, 1H, $J_{4,5}$ = 8.3 Hz, $J_{5,6}$ = 6.2 Hz, H-5), 2.16 (dddd, 1H, $J_{3a,3e}$ = 12.9 Hz, $J_{2e,3e}$ = 4.3 Hz, $J_{3e,4}$ = 4.3 Hz, $J_{2a,3e}$ = 4.3 Hz, H-3e), 2.06 (s, 3H, O=CCH_3), 1.91–1.97 (m, 1H, H-2e), 1.71 (dddd, 1H, $J_{2a,2e}$ = $J_{2a,3a}$ = 13.1 Hz, $J_{1,2a}$ = 9.1 Hz, $J_{2a,3e}$ = 4.3 Hz, H-2a), 1.50 (dddd, 1H, $J_{3a,3e}$ = $J_{2a,3a}$ = 13.1 Hz, $J_{3a,4}$ = 10.5 Hz, $J_{2e,3a}$ = 4.3 Hz, H-3a), 1.22 (d, 3H, $J_{5,6}$ = 6.2 Hz, H-6); ^{13}C NMR (100 MHz, $CDCl_3$): δ_C 170.2 (C=O), 147.0 (Ar), 141.1 (Ar), 137.5 (Ar), 133.7 (Ar), 128.8 (Ar), 128.7 (2, Ar), 128.4 (2, Ar), 125.3 (Ar), 125.0 (Ar), 123.2 (Ar), 122.1 (Ar), 112.9 (Ar), 98.8 (C-1), 89.9 (≡C), 80.7 (≡C), 73.3 (C-5), 72.8 (C-4), 55.9 (OCH_2), 29.9 (C-2), 27.0 (C-3), 21.2 (O=CCH_3), 18.1 (C-6). ESIMS: *m*/*z* calcd for $[C_{25}H_{24}O_4S]Na^+$: 443.1288. Found: 443.1290. Anal. Calcd for $C_{25}H_{24}O_4S$: C, 71.40; H, 5.75; S, 7.63. Found: C, 71.40; H, 5.73; S, 7.50.

3.9. 3-(2-Phenylbenzo[*b*]thiophen-3-yl)prop-2-ynyl 2,3,6-trideoxy-α-L-*erythro*-hexopyranoside (22)

This compound was synthesized as a colorless waxy solid from **21** (176 mg, 0.418 mmol) and K_2CO_3 (18 mg, 0.13 mmol) in 99% yield by following the general procedure for deacetylation. The product was purified by column chromatography (2:1 hexanes–EtOAc): yellow sirup, R_f 0.34 (2:1 hexanes–EtOAc); IR: *ν* 3415 (O–H), 2215 (C≡C) cm^{-1}; $[\alpha]_D$ −96.3 (*c* 1.8, CH_2Cl_2); ^{1}H NMR (400 MHz, $CDCl_3$): δ_H 7.96–8.00 (m, 2H, Ar), 7.92–7.96 (m, 1H, Ar), 7.79–7.82 (m, 1H, Ar), 7.36–7.49 (m, 5H, Ar), 5.07 (br s, 1H, H-1), 4.57 (s, 2H, OCH_2C≡C), 3.66 (dq, 1H, $J_{4,5}$ = 9.2 Hz, $J_{5,6}$ = 6.3 Hz, H-5), 3.26–3.34 (m, 1H, H-4), 1.75–1.94 (m, 4H, H-2a, H-2e, H-3a, H-3e), 1.41 (br s, 1H, OH), 1.17 (d, 3H, $J_{5,6}$ = 6.3 Hz, H-6); ^{13}C NMR (100 MHz, $CDCl_3$): δ_C 146.9 (Ar), 141.1 (Ar), 137.5 (Ar), 133.7 (Ar), 128.8 (Ar), 128.7 (2, Ar), 128.4 (2, Ar), 125.2 (Ar), 125.0 (Ar), 123.3 (Ar), 122.0 (Ar), 113.0 (Ar), 94.7 (C-1), 90.5 (≡C), 80.3 (≡C), 72.0 (C-4), 70.0 (C-5), 54.7 (OCH_2), 29.5 (C-2), 27.6 (C-3), 17.9 (C-6). ESIMS: *m*/*z* calcd for $[C_{23}H_{22}O_3S]Na^+$: 401.1182. Found: 401.1186. Anal. Calcd for $C_{23}H_{22}O_3S$: C, 72.99; H, 5.86; S, 8.47. Found: C, 72.56; H, 5.85; S, 8.59.

3.10. 3-(2-Phenylbenzo[*b*]thiophen-3-yl)prop-2-ynyl 4-azido-2,3,4,6-tetradeoxy-α-L-*threo*-hexopyranoside (23)

This compound was synthesized as a yellow oil from **22** (49 mg, 0.13 mmol), PPh_3 (102 mg, 0.389 mmol), DIAD (66 mg, 0.33 mmol), and DPPA (89 mg, 0.33 mmol) in 80% yield by following the general procedure for Mitsunobu reactions. The product was purified by column chromatography (20:1 hexanes–EtOAc): yellow oil, R_f 0.33 (15:1 hexanes–EtOAc); IR: *ν* 2214 cm^{-1} (C≡C), 2098 cm^{-1} (N=N=N); $[\alpha]_D$ −67.7 (*c* 2.8, CH_2Cl_2); ^{1}H NMR (400 MHz, $CDCl_3$): δ_H 7.96–8.00 (m, 2H, Ar), 7.92–7.96 (m, 1H, Ar), 7.79–7.82 (m, 1H, Ar), 7.36–7.49 (m, 5H, Ar), 5.14 (br d, 1H, $J_{1,2a}$ = 4.0 Hz, H-1), 4.57 (AB q, 2H, *J* = 16.0 Hz, OCH_2C≡C), 4.07 (dq, 1H, $J_{5,6}$ = 6.5 Hz, $J_{4,5}$ = 1.7 Hz, H-5), 3.47 (br s, 1H, H-4), 2.19 (dddd, 1H, $J_{3a,3e}$ = -$J_{2a,3a}$ = 13.4 Hz, $J_{3a,4}$ = 4.2 or 3.2 Hz, $J_{2e,3a}$ = 3.2 or 4.2 Hz, H-3a), 2.02 (dddd, 1H, $J_{2a,2e}$ = $J_{2a,3a}$ = 13.4 Hz, $J_{1,2a}$ = 4.0 Hz, $J_{2a,3e}$ = 4.0 Hz, H-2a), 1.91–1.97 (m, 1H, H-3e), 1.62–1.68 (m, 1H, H-2e), 1.20 (d, 3H, $J_{5,6}$ = 6.5 Hz, H-6); ^{13}C NMR (100 MHz, $CDCl_3$, δ_C) 147.0 (Ar), 141.1 (Ar), 137.5 (Ar), 133.7 (Ar), 128.8 (Ar), 128.7 (2, Ar), 128.4 (2, Ar), 125.3 (Ar), 125.0 (Ar), 123.2 (Ar), 122.1 (Ar), 121.9 (Ar), 95.7 (C-1), 90.2 (≡C), 80.4 (≡C), 65.7 (C-5), 60.0 (C-4), 55.1 (OCH_2), 23.9 (C-2), 22.9 (C-3), 17.9 (C-6). ESIMS: *m*/*z* calcd for $C_{23}H_{21}N_3O_2S$: 403.1354. Found: 403.1349. Anal. Calcd for $C_{23}H_{21}N_3O_2S$: C, 68.46; H, 5.25; N, 10.41; S, 7.95. Found: C, 68.70; H, 5.10; N, 10.12; S, 7.93.

3.11. 3-(2-Phenylbenzo[*b*]thiophen-3-yl) prop-2-ynyl 4-*O*-methanesulfonyl-2,3,6-trideoxy-α-L-*erythro*-hexopyranoside (24) and 3-(2-phenylbenzo[b]thiophen-3-yl)prop-2-ynyl 4-*O*-methanesulfonyl-2,3,6-trideoxy-β-L-*erythro*-hexopyranoside (51)

The mixture of **22** and its β anomer (42 mg, 0.11 mmol) was dissolved in CH_2Cl_2 (5 mL), and Et_3N (0.047 mL, 0.34 mmol) was added. The solution was cooled to 0 °C and then mesyl chloride (0.013 mL, 0.17 mmol) was added. After stirring for 2 h, the reaction mixture was washed sequentially with 1 N HCl, 1 N NaOH and brine. The solution was dried (Na_2SO_4), and filtered, and the solvent was evaporated to give a yellow sirup that was purified by column chromatography (4:1 hexanes–EtOAc) to give pure **24** and **51**, both as a yellow oils (51 mg, 99% as combined yield, α:β = 3.8:1). Data for **24**: R_f 0.55 (2:1 hexanes–EtOAc); IR: *ν* 2216 (C≡C) cm^{-1}; $[\alpha]_D$ −105.9 (*c* 2.8, CH_2Cl_2); ^{1}H NMR (400 MHz, $CDCl_3$): δ_H 7.96–8.00 (m, 2H, Ar), 7.92–7.95 (m, 1H, Ar), 7.79–7.82 (m, 1H, Ar), 7.36–7.50 (m, 5H, Ar), 5.08 (br d, 1H, $J_{1,2a\ or\ 2e}$ = 2.8 Hz, H-1), 4.58 (s, 2H, OCH_2C≡C), 4.31–4.38 (m, 1H, H-4), 3.92 (dq, 1H, $J_{4,5}$ = 9.4 Hz, $J_{5,6}$ = 6.2 Hz, H-5), 3.03 (s, 3H, SO_2CH_3), 2.10–2.20 (m, 2H, H-3a, H-3e), 1.92–1.98 (m, 1H, H-2e), 1.83–1.91 (m, 1H, H-2a), 1.28 (d, 3H, $J_{5,6}$ = 6.2 Hz, H-6); ^{13}C NMR (100 MHz, $CDCl_3$): δ_C 147.1 (Ar), 141.1 (Ar), 137.5 (Ar), 133.6 (Ar), 128.9 (Ar), 128.7 (2, Ar), 128.4 (2, Ar), 125.3 (Ar), 125.0 (Ar), 123.2 (Ar), 122.1 (Ar), 112.8 (Ar), 94.6 (C-1), 89.9 (≡C), 80.6(4) (C-4), 80.6(2) (≡C), 66.6 (C-5), 55.0 (OCH_2), 38.9 (SO_2CH_3), 29.2 (C-2), 25.5 (C-3), 17.9 (C-6). ESIMS: *m*/*z* calcd for $[C_{24}H_{24}O_5S_2]Na^+$: 479.0957. Found: 479.0953. Anal. Calcd for $C_{24}H_{24}O_5S_2$: C, 63.13; H, 5.30; S, 14.05. Found: C, 63.07; H, 5.40; S, 14.05. Data for **51**: R_f 0.47 (2:1 hexanes–EtOAc); IR: *ν* 2218 (C≡C) cm^{-1}; $[\alpha]_D$ +73.4 (*c* 1.3, CH_2Cl_2); ^{1}H NMR (500 MHz, $CDCl_3$): δ_H 7.95–7.98 (m, 2H, Ar), 7.91–7.94 (m, 1H, Ar), 7.80–7.83 (m, 1H, Ar), 7.37–7.49 (m, 5H, Ar), 4.83 (dd, 1H, $J_{1,2a}$ = 8.7 Hz, $J_{1,2e}$ = 2.3 Hz, H-1), 4.72 (ABq, 1H, *J* = 16.0 Hz, OCH_2C≡C), 4.67 (ABq, 1H, *J* = 16.0 Hz, OCH_2C≡C), 4.30 (ddd, 1H, $J_{3a,4}$ = $J_{4,5}$ = 10.4 Hz, $J_{3e,4}$ = 4.7 Hz, H-4), 3.54 (dq, 1H, $J_{4,5}$ = 8.7 Hz, $J_{5,6}$ = 6.2 Hz, H-5), 3.03 (s, 3H, SO_2CH_3), 2.36–2.40 (m, 1H, H-3e), 1.96–2.01 (m, 1H, H-2e), 1.68–1.82 (m, 2H, H-3a, H-2a), 1.22 (d, 3H, $J_{5,6}$ = 6.2 Hz, H-6); ^{13}C NMR (125 MHz, $CDCl_3$): δ_C 147.2 (Ar), 141.1 (Ar), 137.5 (Ar), 133.7 (Ar), 128.9 (Ar), 128.7 (2, Ar), 128.4 (2, Ar), 125.3 (Ar), 125.0 (Ar), 123.2 (Ar), 122.1 (Ar), 112.8 (Ar), 98.5 (C-1), 89.6 (≡C), 80.9 (≡C), 79.9 (C-5), 73.0 (C-4), 56.1 (OCH_2), 38.8 (SO_2CH_3), 29.9 (C-2), 28.3 (C-3), 18.2 (C-6). ESIMS: *m*/*z* calcd for $[C_{24}H_{24}O_5S_2]Na^+$: 443.1288. Found: 443.1289. Purity: >99%.

3.12. 4-(2-Phenylbenzo[*b*]thiophen-3-yl)-1,2,3-triazol-5-yl-methyl 4-azido-2,3,4,6-tetradeoxy-α-L-*threo*-hexopyranoside (25)

Compound **24** (69 mg, 0.15 mmol) was dissolved in DMF (5 mL), and NaN_3 (99 mg, 1.5 mmol) was added. The reaction mixture was

heated to 120 °C and stirred overnight. After cooling, the reaction mixture was dissolved in water and extracted with EtOAc. The organic layer was dried (Na_2SO_4), and filtered, and the solvent was evaporated to give an orange oil that was purified by column chromatography (10:1 hexanes–EtOAc) to give pure **25** (20 mg, 30%) as a yellow oil: R_f 0.40 (8:1 hexanes–EtOAc); IR: ν 2099 (N=N=N), 1734, 1600, 1433, 1215 (trizole) cm^{-1}; $[\alpha]_D$ −50.1 (*c* 0.5, CH_2Cl_2); ^1H NMR (500 MHz, $CDCl_3$): δ_H 7.86–7.90 (m, 1H, Ar), 7.46–7.50 (m, 1H, Ar), 7.32–7.41 (m, 4H, Ar), 7.27–7.31 (m, 3H, Ar), 4.61 (br d, 1H, $J_{1,2a\ or\ 2e}$ = 3.5 Hz, H-1), 4.37 (ABq, 1H, *J* = 16.2 Hz, OCH_2C≡C), 4.23 (ABq, 1H, *J* = 16.2 Hz, OCH_2C≡C), 3.25 (dq, 1H, $J_{5,6}$ = 6.2 Hz, $J_{4,5}$ = 1.8 Hz, H-5), 3.05 (br s, 1H, H-4), 1.57–1.75 (m, 3H, H-2e, H-3a, H-3e), 1.24–1.30 (m, 1H, H-2a), 0.84 (d, 3H, $J_{5,6}$ = 6.2 Hz, H-6); ^{13}C NMR (125 MHz, $CDCl_3$): δ_C (missing one carbon signal) 143.7 (Ar), 140.6 (Ar), 138.5 (Ar), 133.4 (Ar), 129.2 (Ar), 128.9 (Ar), 128.8 (2, Ar), 128.6 (2, Ar), 125.0 (Ar), 124.9 (Ar), 123.0 (Ar), 122.1 (Ar), 120.6 (Ar), 96.6 (C-1), 65.1 (C-5), 59.7 (C-4), 59.3 (OCH_2), 23.6 (C-2), 22.5 (C-3), 17.7 (C-6). ESIMS: *m*/*z* calcd for $[C_{23}H_{22}\ N_6O_2S]Na^+$: 469.1417. Found: 469.1418.

3.13. 3-(2-Phenylbenzo[*b*]thiophen-3-yl)prop-2-ynyl 4-*O*-formyl-2,3,6-trideoxy-α-L-*threo*-hexopyranoside (28) and 3-(2-phenylbenzo[*b*]thiophen-3-yl)prop-2-ynyl 4-*O*-formyl-2,3,6-trideoxy-α-L-*erythro*-hexopyranoside (29)

To a solution of compound **22** (251 mg, 0.659 mmol) in 19:1 CH_2Cl_2–pyridine (10 mL) at −20 °C was added a solution of Tf_2O (0.25 mL, 1.5 mmol) in CH_2Cl_2 (5 mL). After stirring for 1.5 h while keeping the temperature below −10 °C, TLC showed the starting material was consumed and a new spot (R_f 0.66, 4:1 hexanes–EtOAc) appeared. The reaction mixture was then extracted with ice-cold 1 M HCl and water. The organic layer was dried (Na_2SO_4), filtered, and concentrated to yield an orange liquid, and the resulting product was immediately dissolved in dry DMF (10 mL), and NaN_3 (430 mg, 6.60 mmol) was added. After stirring at rt for 3 h, the reaction was quenched by the addition of water, and the resulting mixture was extracted with EtOAc. The organic solution was dried (Na_2SO_4) and filtered. After evaporation of the solvent, the residue was purified by column chromatography (15:1 hexanes–EtOAc→8:1 hexanes–EtOAc) to yield pure **23** (130 mg, 49%) as a colorless oil (characterization data are listed above), **28** (72 mg, 27%) as a yellow oil, and **29** (35 mg, 13%) as a white waxy solid. Data for **28**: R_f 0.41 (4:1 hexanes–EtOAc); IR ν 2216.1 (C≡C), 1721.5 (C=O) cm^{-1}; $[\alpha]_D$ −77.1 (*c* 2.8, CH_2Cl_2); ^1H NMR (500 MHz, $CDCl_3$): δ_H 8.20 (d, *J* = 0.9 Hz, O=CH), 7.96–8.00 (m, 2H, Ar), 7.92–7.95 (m, 1H, Ar), 7.79–7.82 (m, 1H, Ar), 7.36–7.50 (m, 5H, Ar), 5.18 (br d, 1H, $J_{1,2a}$ = 4.0 Hz, H-1), 4.98 (br s, 1H, H-4), 4.59 (s, 2H, OCH_2C≡C), 4.11 (br q, $J_{5,6}$ = 6.6 Hz, H-5), 2.16 (dddd, 1H, $J_{3a,3e} = J_{2a,3a}$ = 13.8 Hz, $J_{3a,4}$ = 3.5 Hz, $J_{2e,3a}$ = 3.5 Hz, H-3a), 2.02 (dddd, 1H, $J_{2a,2e} = J_{2a,3a}$ = 13.8 Hz, $J_{1,2a}$ = 4.0 Hz, $J_{2a,3e}$ = 4.0 Hz, H-2a), 1.83–1.89 (m, 1H, H-3e), 1.64–1.69 (m, 1H, H-2e), 1.12 (d, 3H, $J_{5,6}$ = 6.6 Hz, H-6); ^{13}C NMR (100 MHz, $CDCl_3$): δ_C 160.6 (C=O), 147.0 (Ar), 141.1 (Ar), 137.5 (Ar), 133.7 (Ar), 128.8 (Ar), 128.7 (2, Ar), 128.4 (2, Ar), 125.2 (Ar), 124.9 (Ar), 123.2 (Ar), 122.1 (Ar), 112.9 (Ar), 95.7 (C-1), 90.2 (≡C), 80.4 (≡C), 69.3 (C-4), 65.1 (C-5), 55.1 (OCH_2), 23.8 (C-2), 22.9 (C-3), 17.1 (C-6). ESIMS: *m*/*z* calcd for $[C_{24}H_{22}O_4S]Na^+$: 429.1131. Found: 429.1133. Anal. Calcd for $C_{24}H_{22}O_4S$: C, 70.91; H, 5.46; S, 7.89. Found: C, 70.77; H, 5.49; S, 7.98. Data for **29**: R_f 0.39 (4:1 hexanes–EtOAc); IR ν 2217.7 (C≡C), 1725.1 (C=O) cm^{-1}; $[\alpha]_D$ −117.9 (*c* 1.5, CH_2Cl_2); ^1H NMR (500 MHz, CD_3OD): δ_H 8.09 (d, *J* = 0.9 Hz, O=CH), 7.96–8.00 (m, 2H, Ar), 7.92–7.95 (m, 1H, Ar), 7.79–7.82 (m, 1H, Ar), 7.36–7.50 (m, 5H, Ar), 5.10 (br d, 1H, $J_{1,2a}$ = 3.7 Hz, H-1), 4.61–4.67 (m, 1H, H-4), 4.59 (s, 2H, OCH_2C≡C), 3.93 (dq, $J_{4,5}$ = 9.3 Hz, $J_{5,6}$ = 6.2 Hz, H-5), 1.83–2.02 (m, 4H, H-3a, H-3e, H-2a, H-2e), 1.19 (d, 3H, $J_{5,6}$ = 6.2 Hz, H-6); ^{13}C NMR (100 MHz, $CDCl_3$): δ_C 160.2 (C=O), 147.0 (Ar), 141.1 (Ar), 137.5 (Ar), 133.7 (Ar), 128.8 (Ar), 128.7 (2, Ar), 128.4 (2, Ar), 125.2 (Ar), 124.9 (Ar), 123.2 (Ar), 122.0 (Ar), 112.9 (Ar), 94.7 (C-1), 90.1 (≡C), 80.5 (≡C), 73.2 (C-4), 66.8 (C-5), 54.9 (OCH_2), 28.9 (C-2), 24.1 (C-3), 17.8 (C-6). ESIMS: *m*/*z* calcd for $[C_{24}H_{22}O_4S]Na^+$: 429.1131. Found: 429.1133. Anal. Calcd for $C_{24}H_{22}O_4S$: C, 70.91; H, 5.46; S, 7.89. Found: C, 70.49; H, 5.40; S, 8.09.

3.14. 3-(2-Phenylbenzo[*b*]furan-3-yl)prop-2-ynyl 4-*O*-acetyl-2,3,6-trideoxy-α-L-*erythro*-hexopyranoside (32) and 3-(2-phenylbenzo[*b*]furan-3-yl)prop-2-ynyl 4-*O*-acetyl-2,3,6-trideoxy-β-L-*erythro*-hexo pyranoside (43)

This compound was synthesized from **20** (220 mg, 1.04 mmol), **16** (256 mg, 0.799 mmol),[11] $PdCl_2(PPh_3)_2$ (28 mg, 5 mol %), and CuI (15 mg, 10 mol %) in 92% combined yield with the β anomer (α:β = 3.8:1) by following general procedure for Sonogashira coupling. The product was purified by column chromatography (10:1 hexanes–EtOAc). Data for **32**: solid yellow paste, R_f 0.46 (4:1 hexanes–EtOAc); IR: ν 2222 (C≡C), 1737 (C=O) cm^{-1}; $[\alpha]_D$ −108.0 (*c* 5.6, CH_2Cl_2); ^1H NMR (500 MHz, $CDCl_3$): δ_H 8.26–8.30 (m, 2H, Ar), 7.66–7.70 (m, 1H, Ar), 7.47–7.53 (m, 3H, Ar), 7.39–7.43 (m, 1H, Ar), 7.28–7.36 (m, 2H, Ar), 5.13 (br s, 1H, H-1), 4.64 (s, 2H, OCH_2C≡C), 4.52–4.58 (m, 1H, H-4), 3.93 (dq, 1H, $J_{4,5}$ = 9.6 Hz, $J_{5,6}$ = 6.3 Hz, H-5), 2.06 (s, 3H, O=CCH_3), 1.83–2.01 (m, 4H, H-2a, H-2e, H-3a, H-3e), 1.20 (d, 3H, $J_{5,6}$ = 6.3 Hz, H-6); ^{13}C NMR (125 MHz, $CDCl_3$): δ_C 170.2 (C=O), 156.8 (Ar), 153.4 (Ar), 129.9(9) (Ar), 129.9(6) (Ar), 129.2 (Ar), 128.6 (2, Ar), 126.0 (2, Ar), 125.3 (Ar), 123.4 (Ar), 120.3 (Ar), 111.2 (Ar), 98.5 (Ar), 95.0 (C-1), 92.6 (≡C), 77.8 (≡C), 73.4 (C-4), 67.1 (C-5), 55.0 (OCH_2), 29.0 (C-2), 24.1 (C-3), 21.2 (O=CCH_3), 17.9 (C-6). ESIMS: *m*/*z* calcd for $[C_{25}H_{24}O_5]Na^+$: 427.1516. Found: 427.1517. Anal. Calcd for $C_{25}H_{24}O_5$: C, 74.24; H, 5.98. Found: C, 74.50; H, 6.33. Data for **43**: white amorphous solid, R_f 0.44 (4:1 hexanes–EtOAc); IR: ν 2219 (C≡C), 1737 (C=O) cm^{-1}; $[\alpha]_D$ +37.9 (*c* 0.6, CH_2Cl_2); ^1H NMR (400 MHz, $CDCl_3$): δ_H 8.26–8.30 (m, 2H, Ar), 7.66–7.69 (m, 1H, Ar), 7.47–7.53 (m, 3H, Ar), 7.40–7.44 (m, 1H, Ar), 7.29–7.37 (m, 2H, Ar), 4.90 (dd, 1H, $J_{1,2a}$ = 9.1 Hz, $J_{1,2e}$ = 2.3 Hz, H-1), 4.77 (s, 2H, OCH_2C≡C), 4.52 (ddd, 1H, $J_{3a,4}$ = 10.5 Hz, $J_{4,5}$ = 9.0 Hz, $J_{3e,4}$ = 4.5 Hz, H-4), 3.60 (dq, 1H, $J_{4,5}$ = 9.0 Hz, $J_{5,6}$ = 6.2 Hz, H-5), 2.16 (dddd, 1H, $J_{3a,3e}$ = 13.4 Hz, $J_{2e,3e} = J_{3e,4} = J_{2a,3e}$ = 4.5 Hz, H-3e), 2.06 (s, 3H, O=CCH_3), 1.96–2.02 (m, 1H, H-2e), 1.75 (dddd, 1H, $J_{2a,2e} = J_{2a,3a}$ = 13.4 Hz, $J_{1,2a}$ = 9.1 Hz, $J_{2a,3e}$ = 4.5 Hz, H-2a), 1.54 (dddd, 1H, $J_{3a,3e} = J_{2a,3a}$ = 13.4 Hz, $J_{3a,4}$ = 10.5 Hz, $J_{2e,3a}$ = 4.4 Hz, H-3a), 1.29 (d, 3H, $J_{5,6}$ = 6.2 Hz, H-6); ^{13}C NMR (100 MHz, $CDCl_3$): δ_C 170.2 (C=O), 156.8 (Ar), 153.4 (Ar), 129.9(9) (Ar), 129.9(8) (Ar), 129.3 (Ar), 128.6 (2, Ar), 126.0 (2, Ar), 125.4 (Ar), 123.4 (Ar), 120.2 (Ar), 111.2 (Ar), 99.0 (C-1), 98.4 (Ar), 92.2 (≡C), 78.1 (≡C), 73.4 (C-5), 72.8 (C-4), 56.1 (OCH_2), 29.9 (C-2), 27.0 (C-3), 21.1 (O=CCH_3), 18.2 (C-6). ESIMS: *m*/*z* calcd for $[C_{25}H_{24}O_5]Na^+$: 427.1516. Found: 427.1517. Purity: 99.3%.

3.15. 3-(2-Phenylbenzo[*b*]furan-3-yl)prop-2-ynyl 2,3,6-trideoxy-α-L-*erythro*-hexopyranoside (33)

This compound was synthesized as a colorless waxy solid from **32** (131 mg, 0.358 mmol) and K_2CO_3 (15 mg, 0.11 mmol) in 99% yield by following the general procedure for deacetylation. The product was purified by column chromatography (2:1 hexanes–EtOAc): R_f 0.21 (3:1 hexanes–EtOAc); IR: ν 3416 (O–H), 2222 (C≡C) cm^{-1}; $[\alpha]_D$ −109.7 (*c* 8.4, CH_2Cl_2); ^1H NMR (500 MHz, $CDCl_3$): δ_H 8.26–8.30 (m, 2H, Ar), 7.66–7.70 (m, 1H, Ar), 7.47–7.53 (m, 3H, Ar), 7.39–7.43 (m, 1H, Ar), 7.28–7.36 (m, 2H, Ar), 5.12 (br s, 1H, H-1), 4.63 (s, 2H, OCH_2C≡C), 3.71 (dq, 1H, $J_{4,5}$ = 9.2 Hz, $J_{5,6}$ = 6.2 Hz, H-5), 3.30–3.36 (m, 1H, H-4), 1.78–1.98 (m, 4H, H-2a, H-2e, H-3a, H-3e), 1.55 (br s, 1H, OH), 1.30 (d, 3H,

$J_{5,6}$ = 6.2 Hz, H-6); ^{13}C NMR (125 MHz, $CDCl_3$): δ_C 156.8 (Ar), 153.4 (Ar), 130.0(0) (Ar), 129.9(8) (Ar), 129.2 (Ar), 128.7 (2, Ar), 126.0 (2, Ar), 125.3 (Ar), 123.4 (Ar), 120.3 (Ar), 111.2 (Ar), 98.5 (Ar), 94.8 (C-1), 92.7 (≡C), 77.7 (≡C), 72.1 (C-4), 70.0 (C-5), 54.8 (OCH_2), 29.5 (C-2), 27.6 (C-3), 17.9 (C-6). ESIMS: *m*/*z* calcd for $[C_{23}H_{22}O_4]Na^+$: 385.1410. Found: 385.1412. Anal. Calcd for $C_{23}H_{22}O_4$: C, 76.22; H, 6.12. Found: C, 75.83; H, 6.16.

3.16. 3-(2-Phenylbenzo[*b*]furan-3-yl)prop-2-ynyl 4-azido-2,3,4,6-tetradeoxy-α-L-*threo*-hexopyranoside (34)

This compound was synthesized as a yellow oil from **33** (156 mg, 0.430 mmol), PPh_3 (338 mg, 1.29 mmol), DIAD (218 mg, 1.08 mmol), and DPPA (297 mg, 1.52 mmol) in 83% yield by following the general procedure for Mitsunobu reactions. The product was purified by column chromatography (20:1 hexanes–EtOAc): R_f 0.18 (20:1 hexanes–EtOAc); IR: *ν* 2221 cm^{-1} (C≡C), 2098 cm^{-1} (N=N=N); $[\alpha]_D$ −72.1 (*c* 1.7, CH_2Cl_2); ^{1}H NMR (500 MHz, $CDCl_3$): δ_H 8.26–8.30 (m, 2H, Ar), 7.66–7.70 (m, 1H, Ar), 7.47–7.53 (m, 3H, Ar), 7.39–7.43 (m, 1H, Ar), 7.28–7.36 (m, 2H, Ar), 5.14 (br d, 1H, $J_{1,2a}$ = 4.0 Hz, H-1), 4.63 (s, 2H, OC*H*$_2$C≡C), 4.13 (dq, 1H, $J_{5,6}$ = 6.5 Hz, $J_{4,5}$ = 1.7 Hz, H-5), 3.49 (br s, 1H, H-4), 2.19 (dddd, 1H, $J_{3a,3e}$ = $J_{2a,3a}$ = 13.4 Hz, $J_{3a,4}$ = 4.2 or 3.2 Hz, $J_{2e,3a}$ = 3.2 or 4.2 Hz, H-3a), 2.02 (dddd, 1H, $J_{2a,2e}$ = $J_{2a,3a}$ = 13.4 Hz, $J_{1,2a}$ = $J_{2a,3e}$ = 4.0 Hz, H-2a), 1.94–2.00 (m, 1H, H-3e), 1.67–1.73 (m, 1H, H-2e), 1.20 (d, 3H, $J_{5,6}$ = 6.5 Hz, H-6); ^{13}C NMR (125 MHz, $CDCl_3$): δ_C 156.8 (Ar), 153.4 (Ar), 129.9(9) (Ar), 129.9(7) (Ar), 129.3 (Ar), 128.7 (2, Ar), 126.0 (2, Ar), 125.4 (Ar), 123.4 (Ar), 120.3 (Ar), 111.2 (Ar), 98.5 (Ar), 95.8 (C-1), 92.6 (≡C), 77.8 (≡C), 65.8 (C-5), 59.9 (C-4), 55.2 (OCH_2), 24.0 (C-2), 23.0 (C-3), 21.2 (O=C*C*H$_3$), 17.9 (C-6). ESIMS: *m*/*z* calcd for $[C_{23}H_{21}N_3O_3]Na^+$: 410.1475. Found: 410.1474. Anal. Calcd for $C_{23}H_{21}N_3O_3$: C, 71.30; H, 5.46; N, 10.85. Found: C, 71.49; H, 5.56; N, 10.57.

3.17. 3-(1-Tosyl-2-phenylindol-3-yl)prop-2-ynyl 4-*O*-acetyl-2,3,6-trideoxy-α-L-*erythro*-hexopyranoside (35)

This compound was synthesized as a yellow sirup from **20** (219 mg, 1.03 mmol) and **18** (488 mg, 1.03 mmol),[11] PPh_3 (11 mg, 4 mol %), $PdCl_2(PPh_3)_2$ (36 mg, 5 mol %), and CuI (20 mg, 10 mol %) in 65% combined yield with its β anomer (α:β = 3.8:1) by following the general procedure for Sonogashira coupling. The α anomer **35** was purified by column chromatography (5:1 hexanes–EtOAc): R_f 0.36 (4:1 hexanes–EtOAc); IR: *ν* 2226 (C≡C), 1736 (C=O) cm^{-1}; $[\alpha]_D$ −86.5 (*c* 1.0, CH_2Cl_2); ^{1}H NMR (500 MHz, $CDCl_3$): δ_H 8.30–8.33 (m, 1H, Ar), 7.53–7.59 (m, 3H, Ar), 7.39–7.48 (m, 4H, Ar), 7.31–7.36 (m, 1H, Ar), 7.24–7.27 (m, 2H, Ar), 7.03–7.07 (d, 2H, *J* = 8.6 Hz, Ar), 4.86 (br s, 1H, H-1), 4.46–4.52 (m, 1H, H-4), 4.36 (s, 2H, OC*H*$_2$C≡C), 3.93 (dq, 1H, $J_{4,5}$ = 9.6 Hz, $J_{5,6}$ = 6.2 Hz, H-5), 2.30 (s, 3H, PhC*H*$_3$), 2.06 (s, 3H, O=CC*H*$_3$), 1.89–1.94 (m, 1H, H-3e), 1.76–1.81 (m, 3H, H-2a, H-2e, H-3a), 1.20 (d, 3H, $J_{5,6}$ = 6.2 Hz, H-6); ^{13}C NMR (125 MHz, $CDCl_3$): δ_C 170.2 (C=O), 144.9 (Ar), 143.8 (Ar), 137.0 (Ar), 134.7 (Ar), 131.2 (2, Ar), 130.7 (Ar), 130.5 (Ar), 129.3 (2, Ar), 129.1 (Ar), 127.3 (2, Ar), 126.8 (2, Ar), 125.7 (Ar), 124.6 (Ar), 120.0 (Ar), 116.5 (Ar), 107.4 (Ar), 94.6 (C-1), 90.2 (≡C), 77.9 (≡C), 73.4 (C-5), 66.9 (C-4), 54.6 (OCH_2), 28.9 (C-3), 24.0 (C-2), 21.5 (PhCH_3), 21.2 (O=CCH_3), 17.8 (C-6). ESIMS: *m*/*z* calcd for $[C_{32}H_{31}NO_6S]Na^+$: 580.1764. Found: 580.1768. Anal. Calcd for $C_{32}H_{31}NO_6S$: C, 68.92; H, 5.60; N, 2.51; S, 5.75. Found: C, 68.53; H, 5.96; N, 2.38; S, 5.64.

3.18. 3-(2-Phenylindol-3-yl)prop-2-ynyl 4-*O*-acetyl-2,3,6-trideoxy-α-L-*erythro*-hexopyranoside (36)

This compound was synthesized as a yellow sirup from **35** (51 mg, 0.13 mmol) and TBAF solution in THF (1.0 M, 0.75 mL, 0.75 mmol) in 61% yield by following the general procedure for detosylation. The product was purified by column chromatography (5:1, hexanes–EtOAc): R_f 0.33 (4:1 hexanes–EtOAc); IR: *ν* 2226 (C≡C), 1734 (C=O) cm^{-1}; $[\alpha]_D$ −99.7 (*c* 1.5, CH_2Cl_2); ^{1}H NMR (500 MHz, $CDCl_3$): δ_H 8.42 (br s, 1H, NH), 7.95–7.99 (m, 2H, Ar), 7.76 (d, 1H, *J* = 7.6 Hz, Ar), 7.49 (t, 2H, *J* = 7.7 Hz, Ar), 7.39 (t, 2H, *J* = 7.7 Hz, Ar), 7.18–7.27 (m, 2H, Ar), 5.13 (br s, 1H, H-1), 4.60 (s, 2H, OC*H*$_2$C≡C), 4.51–4.57 (m, 1H, H-4), 3.92 (dq, 1H, $J_{4,5}$ = 9.6 Hz, $J_{5,6}$ = 6.3 Hz, H-5), 2.06 (s, 3H, O=CCH_3), 1.84–1.98 (m, 4H, H-3a, H-3e, H-2a, H-2e), 1.20 (d, 3H, $J_{5,6}$ = 6.3 Hz, H-6); ^{13}C NMR (125 MHz, $CDCl_3$): δ_C 170.3 (C=O), 139.9 (Ar), 135.2 (Ar), 131.5 (Ar), 130.5 (Ar), 128.9 (2, Ar), 128.4 (Ar), 126.5 (2, Ar), 123.5 (Ar), 120.9 (Ar), 120.1 (Ar), 111.0 (Ar), 95.3 (Ar), 94.7 (C-1), 88.9 (≡C), 80.4 (≡C), 73.5 (C-5), 66.9 (C-4), 55.2 (OCH_2), 29.0 (C-3), 24.1 (C-2), 21.2 (O=C*C*H$_3$), 17.9 (C-6). ESIMS: *m*/*z* calcd for $[C_{25}H_{25}NO_4]Na^+$: 426.1676. Found: 426.1678. Anal. Calcd for $C_{25}H_{25}NO_4$: C, 74.42; H, 6.25; N, 3.47. Found: C, 74.76; H, 5.99; N, 3.46.

3.19. 3-(1-Tosyl-2-phenylindol-3-yl)prop-2-ynyl 2,3,6-trideoxy-α-L-*erythro*-hexopyranoside (37) and 3-(1-tosyl-2-phenylindol-3-yl)prop-2-ynyl 2,3,6-trideoxy-β-L-*erythro*-hexopyranoside (52)

This compound was synthesized as a colorless waxy solid from the mixture of **35** and its β anomer (229 mg, 0.409 mmol) and K_2CO_3 (17 mg, 0.12 mmol) in 89% yield by following the general procedure for deacetylation. The product was purified by column chromatography (2:1 hexanes–EtOAc, α:β = 3.8:1). Data for **36**: R_f 0.40 (2:1 hexanes–EtOAc); IR: *ν* 3403 (O–H), 2222 (C≡C) cm^{-1}; $[\alpha]_D$ −90.2 (*c* 1.7, CH_2Cl_2); ^{1}H NMR (500 MHz, $CDCl_3$): δ_H 8.30–8.33 (m, 1H, Ar), 7.53–7.59 (m, 3H, Ar), 7.39–7.48 (m, 4H, Ar), 7.31–7.36 (m, 1H, Ar), 7.24–7.27 (m, 2H, Ar), 7.03–7.07 (d, 2H, *J* = 8.5 Hz, Ar), 4.85 (br s, 1H, H-1), 4.35 (s, 2H, OC*H*$_2$C≡C), 3.54 (dq, 1H, $J_{4,5}$ = 9.2 Hz, $J_{5,6}$ = 6.2 Hz, H-5), 3.22–3.29 (m, 1H, H-4), 2.30 (s, 3H, PhCH_3), 1.67–1.87 (m, 4H, H-3a, H-3e, H-2a, H-2e), 1.20 (d, 3H, $J_{5,6}$ = 6.2 Hz, H-6); ^{13}C NMR (125 MHz, $CDCl_3$): δ_C 144.9 (Ar), 143.8 (Ar), 137.0 (Ar), 134.6 (Ar), 131.2 (2, Ar), 130.7 (Ar), 130.5 (Ar), 129.4 (2, Ar), 129.1 (Ar), 127.3 (2, Ar), 126.8 (2, Ar), 125.7 (Ar), 124.6 (Ar), 120.1 (Ar), 116.5 (Ar), 107.5 (Ar), 94.6 (C-1), 90.4 (≡C), 77.8 (≡C), 72.0 (C-4), 69.8 (C-5), 54.5 (OCH_2), 29.4 (C-2), 27.5 (C-3), 21.5 (PhCH_3), 17.8 (C-6). ESIMS: *m*/*z* calcd for $[C_{30}H_{29}NO_5S]Na^+$: 538.1659. Found: 538.1655. Purity: 98.9%. Data for **52**: R_f 0.57 (1:1 hexanes–EtOAc); IR: *ν* 3440 (O–H), 2220 (C≡C) cm^{-1}; $[\alpha]_D$ +90.7 (*c* 1.0, CH_2Cl_2); ^{1}H NMR (500 MHz, $CDCl_3$): δ_H 8.30–8.33 (d, 1H, *J* = 8.4 Hz, Ar), 7.52–7.60 (m, 3H, Ar), 7.40–7.50 (m, 4H, Ar), 7.32–7.36 (m, 1H, Ar), 7.25–7.29 (m, 2H, Ar), 7.04–7.08 (d, 2H, *J* = 8.5 Hz, Ar), 4.53 (dd, 1H, $J_{1,2a}$ = 9.3 Hz, $J_{1,2e}$ = 2.2 Hz, H-1), 4.51 (ABq, 1H, *J* = 16.0 Hz, OC*H*$_2$C≡C), 4.47 (ABq, 1H, *J* = 16.0 Hz, OC*H*$_2$C≡C), 3.27 (ddd, 1H, $J_{3a,4}$ = 10.5 Hz, $J_{4,5}$ = 8.8 Hz, $J_{3e,4}$ = 4.2 Hz, H-4), 3.19 (dq, 1H, $J_{4,5}$ = 8.8 Hz, $J_{5,6}$ = 6.1 Hz, H-5), 2.31 (s, 3H, PhCH_3), 2.05 (dddd, 1H, $J_{3a,3e}$ = 13.0 Hz, $J_{2e,3e}$ = $J_{3e,4}$ = $J_{2a,3e}$ = 4.2 Hz, H-3e), 1.78–1.83 (m, 1H, H-2e), 1.52–1.62 (m, 2H, OH, H-2a), 1.42 (dddd, 1H, $J_{2a,3a}$ = 13.5 Hz, $J_{3a,3e}$ = 13.0 Hz, $J_{3a,4}$ = 10.5 Hz, $J_{2e,3a}$ = 4.2 Hz, H-3a), 1.28 (d, 3H, $J_{5,6}$ = 6.1 Hz, H-6); ^{13}C NMR (500 MHz, $CDCl_3$): δ_C 143.3 (Ar), 142.2 (Ar), 135.2 (Ar), 133.0 (Ar), 129.5 (2, Ar), 129.0 (Ar), 128.9 (Ar), 127.7 (2, Ar), 127.5 (Ar), 125.7 (2, Ar), 125.2 (2, Ar), 124.1 (Ar), 123.0 (Ar), 118.4 (Ar), 114.8 (Ar), 105.7 (Ar), 97.1 (C-1), 88.3 (≡C), 76.5 (≡C), 74.1 (C-5), 69.9 (C-4), 54.1 (OCH_2), 29.2 (C-2), 28.8 (C-3), 19.9 (PhCH_3), 16.4 (C-6). ESIMS: *m*/*z* calcd for $[C_{30}H_{29}NO_5S]Na^+$: 538.1659. Found: 538.1658. Purity: 98.5%.

3.20. 3-(1-Tosyl-2-phenylindol-3-yl)prop-2-ynyl 4-azido-2,3,4,6-tetradeoxy-α-L-*threo*-hexopyranoside (38)

This compound was synthesized as a yellow oil from **37** (119 mg, 0.231 mmol), PPh_3 (181 mg, 0.688 mmol), DIAD (116 mg, 0.581

mmol), and DPPA (160 mg, 0.580 mmol) in 73% yield by following the general procedure for Mitsunobu reactions. The product was purified by column chromatography (15:1 hexanes–EtOAc): R_f 0.35 (10:1 hexanes–EtOAc); IR: ν 2223 cm^{-1} (C≡C), 2097 cm^{-1} (N=N=N); $[\alpha]_D$ −69.8 (*c* 3.1, CH_2Cl_2); ^1H NMR (500 MHz, $CDCl_3$): δ_H 8.30–8.33 (m, 1H, Ar), 7.52–7.58 (m, 3H, Ar), 7.38–7.48 (m, 4H, Ar), 7.31–7.36 (m, 1H, Ar), 7.24–7.27 (m, 2H, Ar), 7.03–7.07 (d, 2H, J = 8.5 Hz, Ar), 5.14 (br d, 1H, $J_{1,2a}$ = 2.9 Hz, H-1), 4.35 (s, 2H, $OCH_2C{\equiv}C$), 3.93 (dq, 1H, $J_{5,6}$ = 6.5 Hz, $J_{4,5}$ = 1.7 Hz, H-5), 3.42 (br s, 1H, H-4), 2.30 (s, 1H, $PhCH_3$), 2.05–2.14 (m, 1H, H-3e), 1.86–1.97 (m, 2H, H-3a, H-2e), 1.50–1.56 (m, 1H, H-2a), 1.12 (d, 3H, $J_{5,6}$ = 6.5 Hz, H-6); ^{13}C NMR (125 MHz, $CDCl_3$): δ_C 144.9 (Ar), 143.8 (Ar), 137.0 (Ar), 134.6 (Ar), 131.2 (2, Ar), 130.7 (Ar), 130.5 (Ar), 129.4 (2, Ar), 129.1 (Ar), 127.3 (2, Ar), 126.8 (2, Ar), 125.8 (Ar), 124.6 (Ar), 120.0 (Ar), 116.5 (Ar), 107.4 (Ar), 95.4 (C-1), 90.2 (≡C), 77.9 (≡C), 65.6 (C-5), 59.9 (C-4), 54.8 (OCH_2), 23.8 (C-2), 22.9 (C-3), 21.5 ($PhCH_3$), 17.9 (C-6). ESIMS: *m*/*z* calcd for $[C_{30}H_{28}N_4O_4S]Na^+$: 563.1724. Found: 563.1723. Purity: 98.1%.

3.21. 3-(2-Phenylindol-3-yl)propan-3-onyl 2,3,6-trideoxy-α-L-*erythro*-hexopyranoside (40)

To a solution of N-tosyl-protected indole **37** (107 mg, 0.210 mmol) in CH_3OH (10 mL) was added KOH (60 mg, 1.1 mmol), and the mixture was stirred at 40 °C for 5 h. After cooling to rt, the reaction mixture was quenched by the addition of satd aqueous NH_4Cl (10 mL), and was then extracted with EtOAc. The combined organic layers were washed with brine, dried (Na_2SO_4), filtered, and evaporated under reduced pressure to afford the crude product, which was purified by column chromatography to yield pure **40** (51 mg, 65%) as a yellow oil: R_f 0.33 (1:1 hexanes–EtOAc); IR: ν 3398 (O–H), 1730 (C=O) cm^{-1}; $[\alpha]_D$ −50.2 (*c* 0.9, CH_2Cl_2); ^1H NMR (500 MHz, $CDCl_3$): δ_H 8.51 (br s, 1H, NH), 8.30–8.34 (m, 1H, Ar), 7.58–7.62 (m, 2H, Ar), 7.49–7.53 (m, 3H, Ar), 7.38–7.42 (m, 1H, Ar), 7.28–7.33 (m, 2H, Ar), 4.62 (br d, 1H, $J_{1,2a}$ = 2.8 Hz, H-1), 3.90 (ddd, 1H, J = 9.8 Hz, J = 7.0 Hz, J = 6.0 Hz, OCH_2), 3.90 (dt, 1H, J = 9.8 Hz, J = 6.0 Hz, OCH_2), 3.26 (dq, 1H, $J_{4,5}$ = 9.1 Hz, $J_{5,6}$ = 6.2 Hz, H-5), 3.11 (br s, 1H, H-4), 2.76–2.86 (m, 2H, O=CCH_2), 1.59–1.74 (m, 3H, H-3e, H-2a, H-2e), 1.41–1.52 (m, 1H, H-3a), 1.14 (d, 3H, $J_{5,6}$ = 6.2 Hz, H-6); ^{13}C NMR (125 MHz, $CDCl_3$): δ_C 196.2 (C=O), 143.6 (Ar), 135.2 (Ar), 132.5 (Ar), 129.7 (3, Ar), 128.9 (2, Ar), 127.5 (Ar), 123.7 (Ar), 122.6 (Ar), 122.3 (Ar), 115.6 (Ar), 110.9 (Ar), 96.1 (C-1), 72.0 (C-4), 69.3 (C-5), 62.8 (OCH_2), 41.9 (O=CCH_2), 29.6 (C-3), 27.5 (C-2), 17.9 (C-6). ESIMS: *m*/*z* calcd for $[C_{23}H_{25}NO_4]Na^+$: 402.1676. Found: 402.1676.

3.22. General methods—antibacterial assays

Bacto™ yeast extract, Bacto™ tryptone, Difco™ Nutrient broth, Difco™ Meuller–Hinton broth, and Difco™ granulated agar were purchased from the Becton Dickinson Co. (BD). Penicillin, tetracycline, and streptomycin were purchased from Sigma. Daunomycin and doxorubicin were obtained from IFFECT ChemPhar (Hong Kong) Company Ltd. Petri plates, blank 6-mm paper disks, Puritan cotton tip applicators, applicator sticks, tissue culture treated polystyrene 96-well plates, and plastic inoculating loops were purchased from Fisher Scientific Co. and came as sterile.

3.23. Bacterial strains and growth conditions

Gram-positive bacteria, *B. atrophaeus* (ATCC 9372), and Gram-negative bacteria, *E. coli* (ATCC 47076), were purchased from the American Type Culture Collection (ATCC). Using sterile wooden applicator sticks, *E. coli* was streaked onto a Luria–Burtain agar plate, which was made from a sterile aqueous solution consisting of 15 mg/mL agar, 5 mg/mL Bacto™ yeast extract, 10 mg/mL Bacto™ tryptone, and 10 mg/mL NaCl. Similarly, *B. atrophaeus* was streaked onto a Nutrient Agar plate, which was made from a sterile aqueous solution consisting of 15 mg/mL of agar and 8 mg/mL of Bacto™ Nutrient broth. All the bacterial culturing plates were sealed in a plastic bag and incubated overnight at 37 °C.

3.24. Paper disk diffusion assay

Colonies of *B. atrophaeus* and *E. coli* were inoculated into 4 mL of sterile aq solution containing 8 mg/mL Bacto™ Nutrient broth and 4 mL of sterile aq solution containing 5 mg/mL Bacto™ yeast extract, 10 mg/mL Bacto™ tryptone, and 10 mg/mL NaCl, respectively. The bacterial solutions were incubated for 3–5 h at 35 °C without shaking. Cultures were adjusted with the corresponding blank media solution to an absorbance reading of 0.1 at 625 nm, which means that the bacteria were at the 0.5 McFarland suspension, or 1×10^8 CFU/mL. CFU stands for colony forming unit. The resulting bacterial solutions were streaked on Mueller–Hinton agar (MHA) plates, which were made from a sterile aqueous solution comprising 17 mg/mL agar and 21 mg/mL Difco™ Mueller–Hinton broth. Afterward, paper disks were loaded with the required amount of compounds in 15 μL of the appropriate solvents and firmly placed on the MHA plates with a thin film of bacteria. Next, the plates were sealed in a plastic bag and incubated at 35 °C for ~17 h, and the diameters of the inhibition zones were recorded. Each compound was run in duplicate, and the values shown in the table reflect an average of this data.

3.25. 96-Well plate microdilution assay

Following the above procedure for the paper disk diffusion assay, a *B. atrophaeus* solution that had a concentration of 1×10^6 CFU/mL was prepared. A 50-μL aliquot of the above solution was seeded into each well, thereby giving a test concentration of 5×10^4 CFU/well. Thereafter, another 50 μL of the solution of a test compound was added into each well. To prevent drying, the 96-well plate was then sealed in a plastic bag and incubated at 35 °C without shaking for about 20 h. Finally, the minimum inhibition concentrations, at which no appreciable growth of bacteria was observed by the unaided eye, were recorded. Each sample was run in duplicate, and the values shown in the table reflect an average of this data.

Acknowledgements

This work was supported by the University of Alberta, the Natural Sciences and Engineering Research Council of Canada and the Alberta Ingenuity Centre for Carbohydrate Science. W.S. is the recipient of a Studentship from the Alberta Ingenuity Fund.

Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.carres.2009.10.010.

References

1. (a) Di Marco, A.; Gaetani, M.; Dorigotti, L.; Soldati, M.; Bellini, O. *Nature* **1964**, *201*, 706–707; (b) Cassinelli, F.; Orezzi, P. *Gen. Microbiol.* **1963**, *11*, 167–174.
2. Arcamone, F.; Cassinelli, G.; Fantini, G.; Grein, A.; Orezzi, P.; Pol, C.; Spalla, C. *Biotechnol. Bioenerg.* **1969**, *11*, 1101–1110.
3. (a) Arcamone, F.; Bernardi, L.; Giardino, P.; Patelli, B.; Di Marco, A.; Casazza, A. M.; Pratesi, G.; Reggiani, P. *Cancer Treat. Rep.* **1976**, *60*, 829–834; (b) Israel, M.; Modest, E. J.; Frei, E. *Cancer Res.* **1975**, *35*, 1365–1368; (c) Coukell, A. J.; Faulds, D. *Drugs* **1997**, *53*, 453–482; (d) Umezawa, H.; Takahashi, Y.; Kinoshita, M.; Naganawa, H.; Masuda, T.; Ishizuka, M.; Tatsuta, K.; Takeuchi, T. *J. Antibiot.* **1979**, *32*, 1082–1084; (e) Oki, T.; Matsuzawa, Y.; Yoshimoto, A.; Numata, K.; Kitamura, I. *J. Antibiot.* **1975**, *28*, 830–834; (f) Hori, S.; Shirai, M.; Hirano, S.; Oki,

T.; Inui, T.; Tsukagoshi, S.; Ishizuka, M.; Takeuchi, T.; Umezawa, H. *Gann* **1977**, *68*, 685–690.
4. (a) Weiss, R. B. *Semin. Oncol.* **1992**, *19*, 670–686; (b) Arcamone, F. M. Anthracyclines. In *Anticancer Agents from Natural Products*; Cragg, G. M., Kingston, D. G. I., Newmann, D. J., Eds.; CRC Press: Boca Raton, FL, 2005; pp 299–320.
5. Minotti, G.; Menna, P.; Salatorelli, E.; Cairo, G.; Gianni, L. *Pharmacol. Rev.* **2004**, *56*, 185–229.
6. Gewirtz, D. A. *Biochem. Pharmacol.* **1999**, *57*, 727–741.
7. Qu, X.; Wan, C.; Becker, H. C.; Zhong, D.; Zewail, A. H. *Proc. Natl. Acad. Sci. U.S.A.* **2001**, *98*, 14212–14217.
8. Wang, A. H. J.; Ughetto, G.; Quigley, G. J.; Rich, A. *Biochemistry* **1987**, *26*, 1152–1163.
9. Lown, J. W. *Chem. Soc. Rev.* **1993**, *22*, 165–176.
10. (a) Pani, B.; Monti-Bragadin, C.; Samer, L. *Cell. Mol. Life Sci.* **1975**, *31*, 787–788; (b) Dornberger, K.; Eckardt, K.; Ihn, W.; Tresselt, D.; Schumann, G. *J. Basic Microbiol.* **1989**, *29*, 241–245; (c) Kim, B. S.; Moon, S. S.; Hwang, B. K. *J. Agric. Food Chem.* **2000**, *48*, 1875–1881.
11. Shi, W.; Coleman, R. S.; Lowary, T. L. *Org. Biomol. Chem.* **2009**, 3709–3722.
12. National Committee for Clinical Laboratory Standards. Performance standards for antimicrobial disk and dilution susceptibility tests for bacteria isolated from animals. Approved standard M31-A. National Committee for Clinical Laboratory Standards: Wayne, Pa. 1999.
13. Elangovan, A.; Wang, Y. H.; Ho, T. I. *Org. Lett.* **2003**, *5*, 1841–1844.
14. Alami, M.; Ferri, F.; Linstrumelle, G. *Tetrahedron Lett.* **1993**, *34*, 6403–6406.
15. Erdelyi, M.; Gogoll, A. *J. Org. Chem.* **2001**, *66*, 4165–4169.
16. (a) Lal, B.; Pramanik, B. N.; Manhas, M. S.; Bose, A. K. *Tetrahedron Lett.* **1977**, *23*, 1977–1980; (b) Lautens, M.; Schmid, G. A.; Chau, A. *J. Org. Chem.* **2002**, *67*, 8043–8053.
17. Sengupta, S.; Duan, H.; Lu, W.; Petersen, J. L.; Shi, X. *Org. Lett.* **2008**, *10*, 1493–1496.
18. Garg, N. K.; Sarpong, R.; Stoltz, B. M. *J. Am. Chem. Soc.* **2002**, *124*, 13179–13184.
19. Silverstein, R. M.; Webster, F. X. *Spectrometric Identification of Organic Compounds*, 6th ed.; John Wiley & Sons: New York, 1998; Chapter 4.