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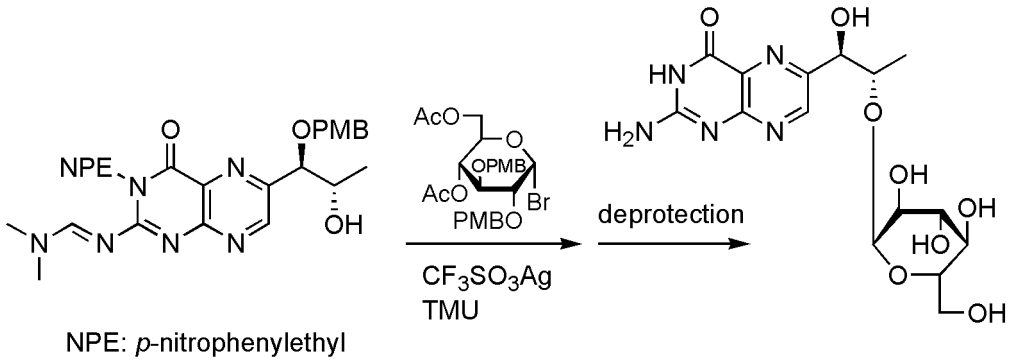

NPE: *p*-nitrophenylethyl

Synthetic Studies on Pterin Glycosides: The First Synthesis of 2'-*O*-(α-D-Glucopyranosyl)biopterin

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Abstract

L-Rhamnose was led, in a 14-step-sequence, to N^2-(*N*,*N*-dimethylaminomethylene)-1'-*O*-(4-methoxybenzyl)-3-[2-(4-nitrophenyl)ethyl]biopterin (**23**), an appropriately protected precursor for 2'-*O*-glycosylation, while 4,6-di-*O*-acetyl-2,3-di-*O*-(4-methoxybenzyl)-α-D-glucopyranosy bromide (**32**), a novel glycosyl donor, was efficiently prepared from D-glucose in 8 steps. The first synthesis of 2'-*O*-(α-D-glucopyranosyl)biopterin (**2a**) was achieved by treatment of the key intermediate **23** with **32** in the presence of silver triflate and tetramethylurea, followed by successive removal of the protecting groups.

Key words: Pterin glycoside; Pteridine; Glycosylation; Protecting group; Total synthesis

1. Introduction

A variety of pterin derivatives having a hydroxyalkyl side-chain at C–6, a representative example being biopterin (**1**), have been found in nature. Some of them isolated from certain prokaryotes possessed a glycosidic form having a sugar attached to the side-chain (Figure 1); for example, 2'-*O*-(α-D-glucopyranosyl)biopterin (**2a**)[1-4] and limipterin [2'-*O*-(2-acetamido-2-deoxy-β-D-glucopyranosyl)biopterin] (**3**)[5] were isolated from cyanobacteria and a green sulfur photosynthetic bacterium, respectively. Various other glycosides consisting of different pterins (such as ciliapterin,[6] neopterin,[7] and 6-hydroxymethylpterin[8]) and sugar moieties (such as D-ribose, D-mannose, D-galactose, and D-glucronic acid) have also been isolated from cyanobacteria, anaerobic photosynthetic bacteria, and chemoautotrophic archaebacteria, although the glycosidic linkages of some derivatives remain unclear. Among these pterin glycosides, biopterin α-D-glucoside (**2a**) is the most noteworthy because of its abundant occurrence in various kinds of cyanobacteria, *Anacystis nidulans*,[1] *Oscillatoria* sp.,[2] *Synechococcus* sp.,[3] and *Spirulina platensis*,[4] but there has been no report for synthesis of **2a** since its first discovery in 1958.

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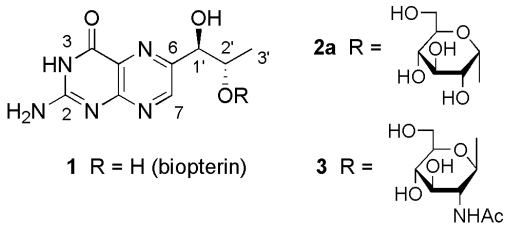
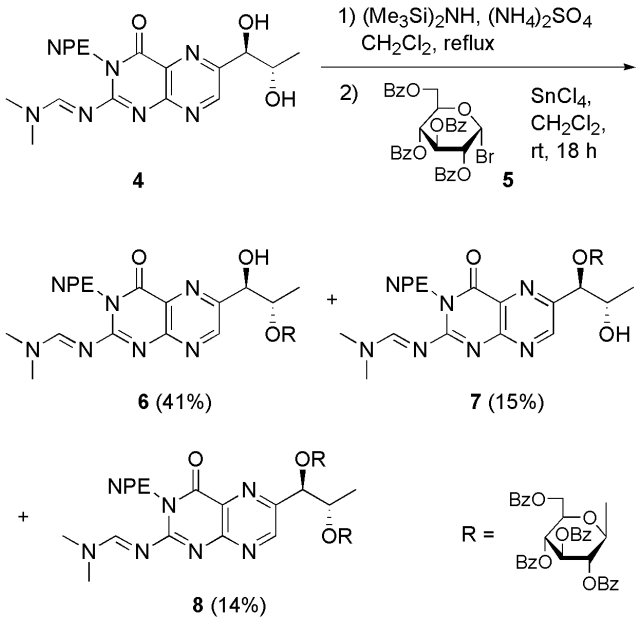


Figure 1. Structures of biopterin and its glycosides

The physiological function of the parent pterins has been studied in detail: for example, **1** plays, in the form of its tetrahydro derivative, an important role as an enzyme cofactor in aromatic amino acid hydroxylation[9] and nitric oxide synthesis.[10] By contrast, the functional roles of pterin glycosides have remained obscure, although some inhibitory activities against tyrosinase[11] and photostabilization of photosynthetic pigments[4,12] were reported for **2a**. Despite a considerable interest from the viewpoint of their biological activities and functions, as well as structural proof of hitherto reported natural products, attempts at preparation of pterin glycosides have so far scarcely been made, except for our synthetic studies on limipterin (**3**) and ciliapterin glycosides.[13,14]

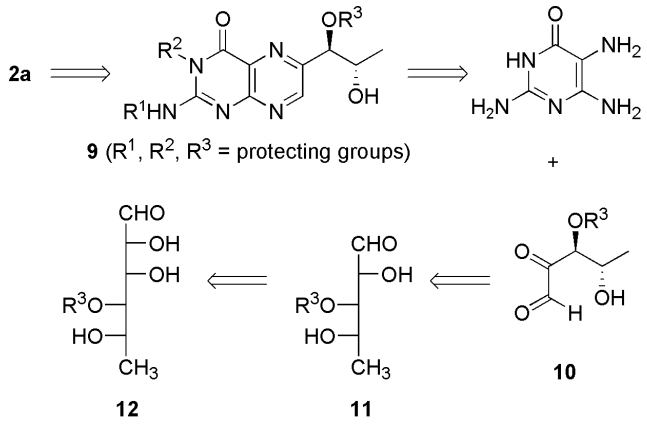
We reported in a previous paper[13] that the glycosylation of a biopterin derivative whose two hydroxy groups were unprotected did not yield the 2'-*O*-(D-glucopyranosyl)biopterins with high selectivity: for example, treatment of N^2-(*N*,*N*-dimethylaminomethylene)-3-[2-(4-nitrophenyl)ethyl]biopterin (**4**) with tetera-*O*-benzoyl-α-D-glucopyranosyl bromide (**5**)[15] (3 mol equiv.) in the presence of tin (IV) chloride afforded 2'-*O*-(β-D-glucopyranosyl)biopterin (**6**) (41% yield), together with 1'-*O*-glycosyl isomer **7** (15%) and 1',2'-di-*O*-glycosyl derivative **8** (14%) (Scheme 1). These results prompted us to develop a more efficient protocol for selective 2'-*O*-monoglycosylation. In addition, we undertook preparation of an effective glycosyl donor leading to preponderant production of pterin α-glycosides instead of β-glycosides. We give herein a full account of the first, efficient synthesis of the representative, natural pterin glycoside, 2'-*O*-(α-D-glucopyranosyl)biopterin (**2a**).[16]



Scheme 1.

2. Results and discussion

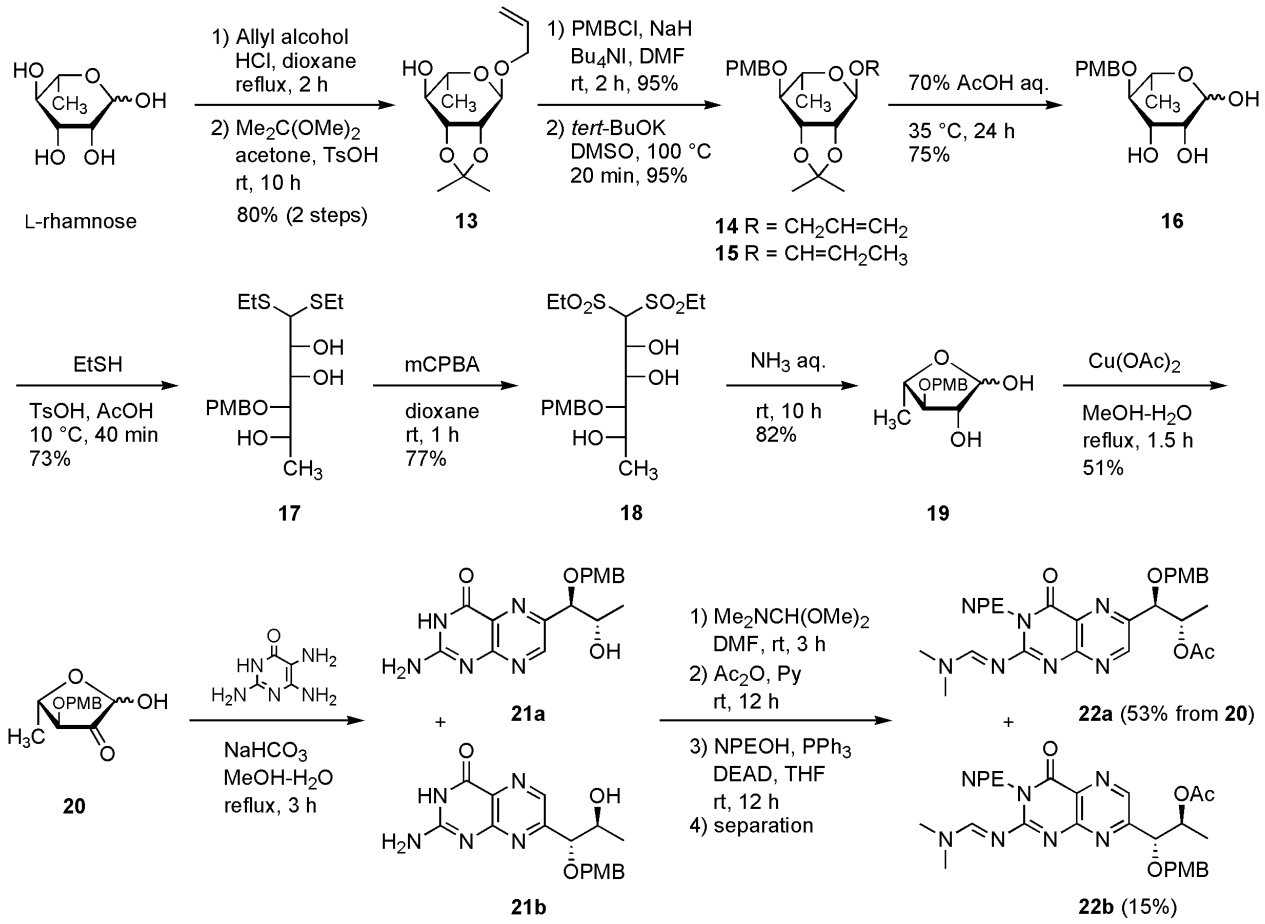
A retrosynthetic analysis for **2a** is outlined in Scheme 2. The biopterin derivative **9**, whose pyrimidine ring moiety and 1'-hydroxy group of the side chain are protected, can be perceived as the key precursor to accomplish complete 2'-*O*-glycosylation, while the pteridine ring formation of **9** would be achieved by condensation of 2,5,6-triamino-4-hydroxypyrimidine with the pentos-2-ulose **10**, which would be derived from the 4-*O*-protected L-rhamnose **12** via the 3-*O*-protected 5-deoxy-L-arabinose **11**.[17] A rational consideration of the available conditions to remove the protecting groups of the glycoside derived from **9** led us to employ *p*-methoxybenzyl (PMB) group for protection of 1'-hydroxy, *N,N*-dimethylaminomethylene group for 2-amino, and 2-(4-nitrophenyl)ethyl (NPE) group for N-3 of the ring.[18]



Scheme 2.

L-Rhamnose, which served as the starting material to obtain the key intermediate 5-deoxy-3-*O*-PMB-L-arabinose (**19**), was subjected to glycosidation with allyl alcohol in the presence of hydrochloric acid, followed by acetalization with 2,2-dimethoxypropane, providing allyl 2,3-*O*-isopropylidene-α-L-rhamnopyranoside (**13**)[19] (80%) along with the corresponding β-anomer (8%) (Scheme 3). Treatment of **13** with *p*-methoxybenzyl chloride and sodium hydride in DMF gave the 4-*O*-PMB derivative **14**, which was then converted into the 1-propenyl glycoside **15** with potassium *tert*-butoxide in DMSO. Hydrolysis of **15** in 70% acetic acid[20] afforded 4-*O*-PMB-L-rhamnopyranose (**16**).

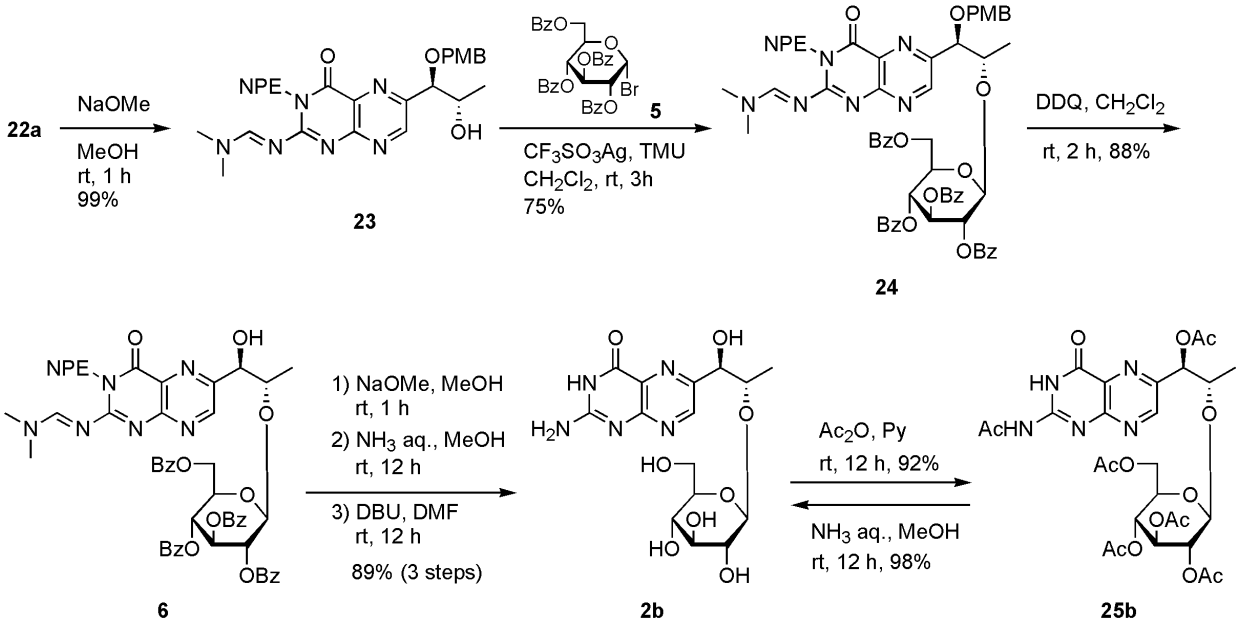
The cleavage of C-1 of **16** was accomplished by application of the Hough and Taylor's procedures[21] with a slight modification. Namely, treatment of **16** with ethanethiol in the presence of *p*-toluenesulfonic acid in acetic acid gave the dithioacetal **17**, which was then oxidized with *m*-chloroperbenzoic acid (*m*CPBA) to the corresponding sulfone **18**. Degradation of **18** with dilute aqueous ammonia afforded 5-deoxy-3-*O*-PMB-L-arbinofuranose (**19**). The selective oxidation for 2-hydroxy group of **19** with cupric acetate[22] provided the L-*erythro*-pentos-3-ulose derivative **20**.



Scheme 3.

The pteridine ring formation of **20** with 2,5,6-triamino-4-hydroxypyrimidine sulfate was carried out in aqueous sodium bicarbonate solution to give an inseparable mixture of the biopterin derivative **21a** and its C-7 substituted isomer **21b** in a ratio of 78:22. These products were separated by column chromatography after having been subjected to the three-step-procedures for introduction of *N,N*-dimethylaminomethylene, acetyl, and NPE groups, thus providing 2'-*O*-acetyl-N^2-(*N,N*-dimethylaminomethylene)-1'-*O*-PMB-3-NPE-biopterin (**22a**) (53% overall yield from **20**) and its C-7 substituted congener **22b** (17%).[14]

Methanolysis of 2'-*O*-acetyl-1'-*O*-PMB-biopterin derivative **22a** in the presence of sodium methoxide provided the 1'-*O*-PMB derivative **23**, a versatile precursor for 2'-O-monoglycosylation (Scheme 4). Glycosylation of **23** was then examined by use of tetra-*O*-benzoyl-α-D-glucopyranosyl bromide (**5**) as a glycosyl donor in the presence of various activators. While treatment of **23** with **5** in dichloromethane at room temperature in the presence of tetrabutylammonium bromide and *N*-ethyldiisopropylamine[23] did not proceed, the same reaction in the presence of tin (IV) chloride[13] as an activator resulted in the formation of diol **4**, instead of glycosylation, by cleavage of PMB group. Efficient glycosylation of **23**, however, was attained by the condensation with 3.0 mol equiv. of **5** in the presence of silver triflate (2.0 mol equiv.) and tetramethylurea (TMU)[24] (1.0 mol equiv.) in dichloromethane at room temperature for 3 h, giving the 2'-*O*-(β-D-glucopyranosyl)biopterin derivative **24** as a sole product in 75% yield.

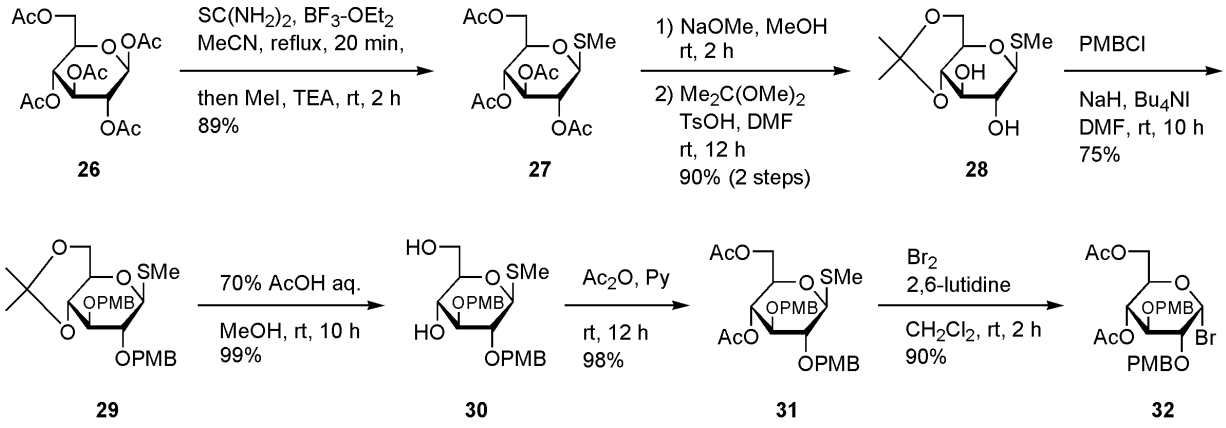


Scheme 4.

Removal of the protecting groups of **24** was performed by the following 4-step-procedures: first, cleavage of PMB by use of 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) to afford **6** in 88%, then the three successive treatment with sodium methoxide (to cleave benzoyl groups), aqueous ammonia (to

cleave the *N*,*N*-dimethylaminomethylene group), and DBU (to cleave the NPE group)[18] furnished 2'-*O*-(β-D-glucopyranosyl)biopterin (**2b**), the anomeric isomer of the natural product, in 89% overall yield from **6**. Structure of **2b** was unambiguously established as the corresponding hexaacetyl derivative **25b** obtained by usual acetylation. Treatment of **25b** with aqueous ammonia regenerated **2b** quantitatively. The precise ^{1}H NMR parameters of **25b** and **2b** are summarized in Tables 1 and 2.

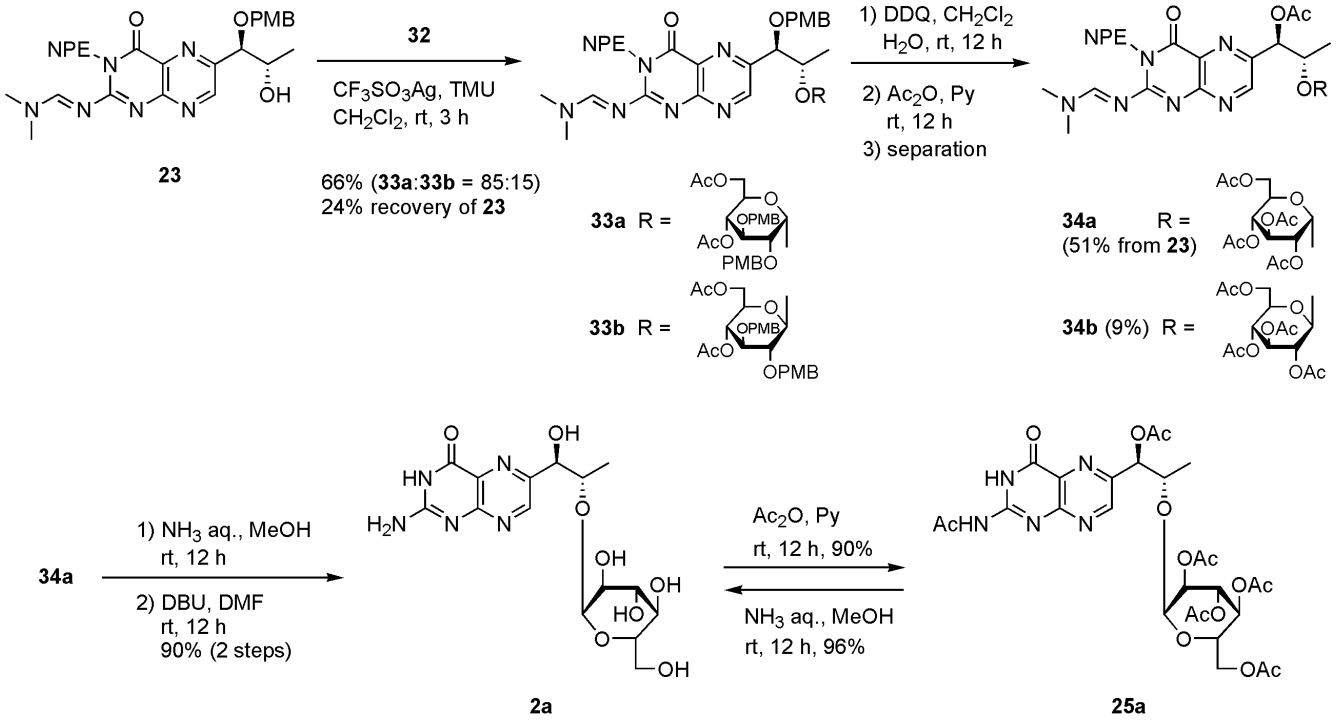
This successful synthesis of biopterin 2'-*O*-β-D-glucoside (**2b**) led us to execute preparation of the natural product, biopterin 2'-*O*-α-D-glucoside (**2a**). The stereoselective formation of the β-glycoside **24** from **23** was mainly caused by participation of the 2-*O*-benzoyl group of the glycosyl donor **5** through the formation of an acyloxonium ion intermediate.[25] Accordingly, in order to avoid such a neighboring group participation, we sought to introduce an ether substituent for protection of 2-OH of a glycosyl donor. Taking into consideration the available combination of protecting groups employed for the synthetic pathway, PMB and acetyl groups were respectively chosen for protection of 2,3-OH and 4,6-OH. We thus undertook the preparation of methyl 4,6-di-*O*-acetyl-2,3-di-*O*-PMB-1-thio-β-D-glucopyranose (**31**) and its α-D-glucopyranosyl bromide derivative **32**, the potential glycosyl donors for the pterin α-glycosides, starting with penta-*O*-acetyl-β-D-glucopyranose (**26**)[26] which is readily available from D-glucose (Scheme 5).



Scheme 5.

Treatment of **26** with thiourea and boron trifluoride etherate, followed by the action of methyl iodide and triethylamine, gave rise to the methyl 1-thio-β-D-glucopyranose derivative **27**.[27] Methanolysis of **27** in the presence of sodium methoxide and the subsequent acetalization with 2,2-dimethoxypropane in the presence of *p*-toluenesulfonic acid provided the 4,6-*O*-isopropylidene derivative **28**. Treatment of **28** with *p*-methoxybenzyl chloride and sodium hydride in DMF gave the 2,3-di-*O*-PMB derivative **29**. Hydrolysis of **29** in 70% acetic acid provided methyl 2,3-di-*O*-PMB-1-thio-β-D-glucopyranoside (**30**), which was then acetylated to give the desired 4,6-di-*O*-acetyl derivative **31**. Then the thioglycoside **31** was transformed to the correponding D-glucopyranosyl bromide **32** by the action of bromine in dichloromethane in the presence of 2,6-lutidine.

Glycosylation of the 1'-*O*-PMB-biopterin derivative **23** with glycosyl donors (**31**, **32**) was examined under various conditions in the presence of activators (Scheme 6). Treatment of **23** with the thioglycoside **31** in dichloromethane at room temperature in the presence of methyl triflate[28] or *N*-iodosuccinimide-silver triflate[29] as activators resulted in the formation of unidentified, decomposed compounds instead of the desired glycoside.[30] Glycosylation of **23** with 4.0 mol equiv. of the glycosyl bromide **32** in dichloromethane in the presence of tetrabutylammonium bromide and *N*-ethyldiisopropylamine did not proceed, whereas the same reaction in the presence of silver triflate (2.0 mol equiv.) and tetramethylurea (TMU) (1.0 mol equiv.) afforded an inseparable anomeric mixture (85:15) of the 2'-*O*-(α-D-glucopyranosyl)biopterin derivative **33a** and its β-anomer **33b** in 66% yield, along with the recovery of **23** (24%). Separation of these isomers was achieved by removal of PMB groups and the subsequent acetylation. Thus, the mixture of **33a,b** was treated with DDQ in dichloromethane, followed by acetylation with acetic anhydride in pyridine, afforded the 2'-*O*-(2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl)biopterin derivative **34a** in 51% (total yield from **23**) and its β-anomer **34b** in 9%. The α-anomeric structure of **34a** was derived from its $J_{1,2}$ value (3.9 Hz) of ^{1}H-NMR, while the larger $J_{1,2}$ value (8.1 Hz) confirmed the β-form of **34b** (Table 1).



Scheme 6.

Removal of the protecting groups of **34a** was accomplished in the following manner: **34a** was treated with aqueous ammonia to cleave the *N*,*N*-dimethylaminomethylene and acetyl groups and then with DBU to cleave NPE group, furnishing the desired 2'-*O*-(α-D-glucopyranosyl)biopterin (**2a**) in 90% overall yield. For the purpose of structural confirmation and further purification, as in the case of the

β-isomer **2b**, the α-isomer **2a** was converted into the hexaacetyl derivative **25a**, which regenerated **2a** upon ammonolysis. The precise parameters obtained on ^{1}H and ^{13}C NMR spectra for **2a** and **25a** are listed in Tables 1 and 2. The spectral data of the synthetic α-glucoside **2a** were found to be essentially identical with those reported for natural product[4,11] (Table 2).

The considerable difference between the $J_{1',2'}$ values of **2a** (7.1 Hz) and **2b** (4.9 Hz) indicates that these isomers are likely to exist in different rotamors along C-1'–C-2' of the pterin side-chain. Accordingly we have calculated the most favorable conformations of **2a,b** using semi-empirical (MOPAC PM3)[31] methods. As depicted in Figure 2, the optimized structure (**A**) for **2a** has *anti* H–C-1'–C-2'–H conformation, whereas that for **2b** (**B**) has the *gauche* conformation. Moreover, an extraordinary upfield shift observed for the H-5 signal (δ 2.40) of the D-glucopyranosyl moiety of **2a** in comparison with the relatively normal value (δ 3.43) of the corresponding β-glucoside (**2b**) could be explained in terms of such a conformation as H-5 of the sugar moiety locating above the pterin ring where an appreciable shielding effect is exerted, as visualized in its optimized structure (**A**).

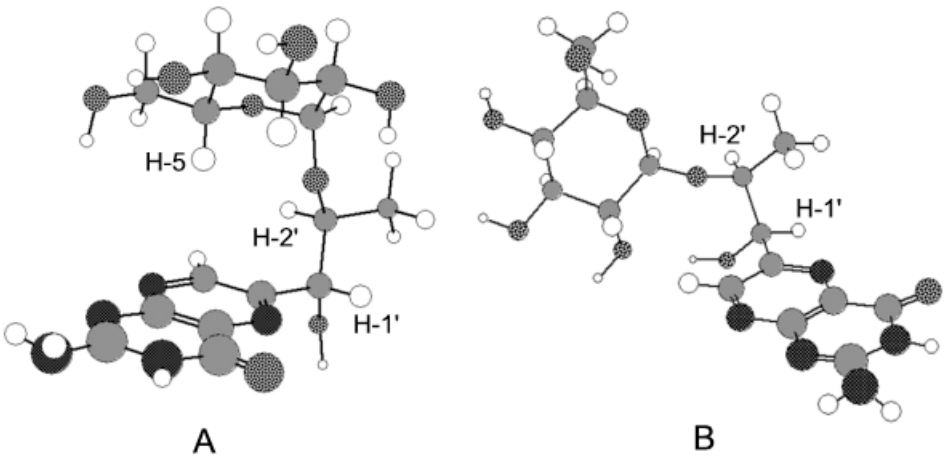


Figure 2. The optimized structures **A** (for **2a**) and **B** (for **2b**) based on MOPAC PM3 methods.

3. Conclusion

We have developed a novel, effective way for selective preparation of both pterin 2'-O-β- and 2'-O-α-glycosides. By use of the key intermediate 1'-*O*-PMB-biopterin derivative **23** and the novel glycosyl donor **32** the first synthesis of biopterin α-D-glucoside (**2a**) was achieved. This synthetic strategy has proved a useful method applicable to a series of other natural pterin glycosides and their analogs.

4. Experimental

4.1. General procedures

All reactions were monitored by TLC (Merck Silica gel 60 F_{254}) with an appropriate solvent system. Column chromatography was performed with Daiso Silica Gel IR-60/210w. Components were detected by exposing the plates to UV light and/or 20% H_2SO_4-EtOH, with subsequent heating. The NMR spectra were measured in $CDCl_3$ with Varian Unity Inova AS600 (600 MHz for ^1H, 151 MHz for ^{13}C) or Mercury300 (300 MHz for ^1H) at 23 °C. Chemical shifts are reported as δ values relative to $CHCl_3$ (7.26 ppm) for ^1H and $CDCl_3$ (77.00 ppm) for ^{13}C as an internal standard, unless otherwise stated. Optical rotations were measured with a JASCO P-1020 polarimeter in $CHCl_3$.

4.2. Allyl 2,3-*O*-isopropylidene-α-L-rhamnopyranoside (13)[19] and its β-anomer

The following modification of the literature procedures was made. To a solution of L-rhamnose monohydrate (3.56 g, 19.5 mmol) in allyl alcohol (28 mL) was added 4 M HCl in dioxane (6.0 mL, 24 mmol). The mixture was refluxed for 2 h, neutralized with TEA (10 mL), and concentrated in vacuo. The residue was dissolved in toluene (20 mL) and evaporated in vacuo to remove allyl alcohol three times. The residual syrup was dissolved in dry acetone (12 mL) and 2,2-dimethoxypropane (9.6 mL, 78 mmol) and then *p*-toluenesulfonic acid monohydrate (30 mg, 0.16 mmol) was added. The mixture was stirred at rt for 10 h and then TEA (5 mL) was added. The mixture was concentrated in vacuo and the residue was purified by column chromatography with 1:4 AcOEt-hexane to give **13** (3.83 g, 80%) (lit.[19] 68% yield on acetalization with acetone alone) and its β-anomer (372 mg, 7.8%).

13: Colorless syrup; R_f = 0.67 (1:1 AcOEt-hexane); ^1H-NMR (600 MHz, $CDCl_3$) δ 1.29 (3H, d, $J_{5,6}$ = 6.3 Hz, H_3-6), 1.35, 1.52 (3H each, 2s, CMe_2), 2.45 (1H, br s, HO-4), 3.40 (1H, dd, $J_{4,5}$ = 9.4, $J_{3,4}$ = 7.2 Hz, H-4), 3.68 (1H, dq, H-5), 4.00 (1H, ddt, $J_{1'a,1'b}$ = 12.7, $J_{1'b,2'}$ = 6.4, $J_{1'b,3'Z}$ = $J_{1'b,3'E}$ = 1.3 Hz, H^b-1'of allyl), 4.09 (1H, dd, $J_{2,3}$ = 5.7 Hz, H-3), 4.16 (1H, d, $J_{1,2}$ = 0.7 Hz, H-2), 4.19 (1H, ddt, $J_{1a',2'}$ = 5.3, $J_{1'a,3'E}$ = $J_{1'a,3'Z}$ = 1.6 Hz, H^a-1' of allyl), 5.00 (1H, br s, H-1), 5.21 (1H, dq, $J_{2',3'E}$ = 10.5, $J_{3'E,3'Z}$ = 2.0 Hz, H^E-3' of allyl), 5.30 (1H, dq, $J_{2',3'Z}$ = 17.1 Hz, H^Z-3' of allyl), 5.90 (1H, dddd, H-2' of allyl).

β-Anomer of **13**: Pale yellow prisms; mp 52–53 °C (from AcOEt-hexane); $[\alpha]_D^{27}$ +80.1° (*c* = 2.50, $CHCl_3$); R_f = 0.38 (1:1 AcOEt-hexane); ^1H-NMR (600 MHz, $CDCl_3$) δ 1.34 (3H, d, $J_{5,6}$ = 6.1 Hz, H_3-6), 1.39, 1.57 (3H each, 2s, CMe_2), 2.25 (1H, br s, HO-4), 3.29 (1H, dq, $J_{4,5}$ = 9.8 Hz, H-5), 3.53 (1H, dd, $J_{3,4}$ = 7.3 Hz, H-4), 4.02 (1H, dd, $J_{2,3}$ = 5.6 Hz, H-3), 4.18 (1H, ddt, $J_{1'a,1'b}$ = 12.9, $J_{1'b,2'}$ = 6.8, $J_{1'b,3'Z}$ = $J_{1'b,3'E}$ = 1.2 Hz, H^b-1' of allyl), 4.24 (1H, dd, $J_{1,2}$ = 2.2 Hz, H-2), 4.43 (1H, ddt, $J_{1'a,2'}$ = 4.9, $J_{1'a,3'E}$ = $J_{1'a,3'Z}$ = 1.6 Hz, H^a-1' of allyl), 4.78 (1H, d, H-1), 5.23 (1H, dddd, $J_{2',3'E}$ = 10.5, $J_{3'E,3'Z}$ = 2.0 Hz, H^E-3' of allyl), 5.31 (1H, dddd, $J_{2',3'Z}$ = 17.3 Hz, H^Z-3' of allyl), 5.94 (1H, dddd, H-2' of allyl). Anal. Calcd for $C_{12}H_{20}O_5$: C, 59.00; H, 8.25. Found: C, 58.89; H, 8.45.

4.3. Allyl 2,3-*O*-isopropylidene-4-*O*-(4-methoxybenzyl)-α-L-rhamnopyranoside (14)

To a solution of **13** (620 mg, 2.54 mmol) and *p*-methoxybenzyl chloride (0.69 mL, 5.09 mmol) in dry

DMF (6.0 mL) was added tetrabutylammonium iodide (281 mg, 0.726 mmol) and then sodium hydride (60% in oil, 305 mg, 7.26 mmol) at 0 °C. The mixture was stirred at rt for 2 h and then saturated NH_4Cl was added slowly at 0 °C. The mixture was diluted with aqueous $NaHCO_3$ and evaporated in vacuo. The residue was dissolved in $CHCl_3$, washed with water, dried ($MgSO_4$), and evaporated in vacuo. The residue was purified by column chromatography with 1:9 AcOEt-hexane to give **14** (880 mg, 95%) as a colorless syrup: R_f = 0.24 (1:9 AcOEt-hexane); ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.26 (3H, d, $J_{5,6}$ = 6.3 Hz, H_3-6), 1.38, 1.52 (3H each, 2s, CMe_2), 3.20 (1H, dd, $J_{4,5}$ = 9.9, $J_{3,4}$ = 7.1 Hz, H-4), 3.69 (1H, dq, H-5), 3.81 (3H, s, MeO), 3.98 (1H, ddt, $J_{1'a,1'b}$ = 13.1, $J_{1b',2'}$ = 6.3, $J_{1'b,3'E} = J_{1b',3'Z}$ = 1.3 Hz, H^b-1' of allyl), 4.16 (1H, ddt, $J_{1'a,2'}$ = 5.3, $J_{1'a,3'E} = J_{1a',3'Z}$ = 1.5 Hz, H^a-1' of allyl), 4.16 (1H, dd, $J_{2,3}$ = 5.8, $J_{1,2}$ = 0.7 Hz, H-2), 4.27 (1H, dd, H-3), 4.56, 4.84 (1H each, 2d, ^2J = 11.2 Hz, CH_2O-4), 5.01 (1H, d, H-1), 5.20 (1H, dq, $J_{2',3'E}$ = 10.4, $J_{3'E,3'Z}$ = 1.8 Hz, H^E-3' of allyl), 5.29 (1H, dq, $J_{2',3'Z}$ = 17.2 Hz, H^Z-3' of allyl), 5.89 (1H, dddd, H-2' of allyl), 6.89, 7.29 (2H each, 2d, $J_{o,m}$ = 8.8 Hz, C_6H_4). Anal. Calcd for $C_{20}H_{28}O_6$: C, 65.91; H, 7.74. Found: C, 66.11; H, 7.59.

4.4. (*Z*)-1-Propenyl 2,3-*O*-isopropylidene-4-*O*-(4-methoxybenzyl)-α-L-rhamnopyranoside (15)

Compound **14** (880 mg, 2.41 mmol) was dissolved in dry DMSO (10 mL) and potassium *tert*-butoxide (770 mg, 6.85 mmol) was added in small portions. The mixture was stirred at 100 °C for 20 min and then saturated NH_4Cl was added at 0 °C. The mixture was diluted with aqueous $NaHCO_3$ and evaporated in vacuo. The residue was dissolved in water and extracted with $CHCl_3$ three times. The combined organic layers were dried ($MgSO_4$) and evaporated in vacuo. The residue was purified by column chromatography with 1:9 AcOEt-hexane to give **15** (837 mg, 95%) as a colorless syrup: R_f = 0.33 (1:9 AcOEt-hexane); ^{1}H NMR (300 MHz, $CDCl_3$) δ 1.24 (3H, d, $J_{5,6}$ = 6.3 Hz, H_3-6), 1.39, 1.53 (3H each, 2s, CMe_2), 1.56 (3H, dd, $J_{2',3'}$ = 6.9, $J_{1',3'}$ = 1.8 Hz, H-3' of propenyl), 3.21 (1H, dd, $J_{4,5}$ = 9.9, $J_{3,4}$ = 6.9 Hz, H-4), 3.68 (1H, dq, H-5), 3.80 (3H, s, MeO), 4.25 (1H, dd, $J_{2,3}$ = 5.8, $J_{1,2}$ = 0.7 Hz, H-2), 4.32 (1H, dd, H-3), 4.57 (1H, quint, $J_{1',2'}$ = 6.3 Hz, H-2' of propenyl), 4.57, 4.84 (1H each, 2d, ^2J = 11.2 Hz, CH_2O-4), 5.18 (1H, d, H-1), 6.14 (1H, dq, H-1' of propenyl), 6.88, 7.29 (2H each, 2d, $J_{o,m}$ = 8.7 Hz, C_6H_4). Anal. Calcd for $C_{20}H_{28}O_6$: C, 65.91; H, 7.74. Found: C, 66.16; H, 7.63.

4.5. 4-*O*-(4-Methoxybenzyl)-α,β-L-rhamnoprranoses (16)

Compound **15** (830 mg, 2.28 mol) was dissolved in 70% aqueous AcOH (10 mL) and the mixture was stirred at 35 °C for 24 h. The mixture was concentrated in vacuo and the residue was purified by column chromatography with 1:1 AcOEt-hexane to give an inseparable anomeric mixture (α:β = ca. 1:1) of **16** (515 mg, 79%) as a colorless foam: R_f = 0.21 (AcOEt); ^{1}H NMR [300 MHz, $CDCl_3$ (D_2O exchange)] δ 1.19*, 1.24 (3H each, 2d, $J_{5,6}$ = 6.3*, 5.6 Hz, H_3-6 of α*,β), 3.25 (2H, m, H-4,5 of β), 3.32 (1H, t, $J_{3,4} = J_{4,5}$ =9.1 Hz, H-4 of α), 3.56 (1H, dd, $J_{3,4}$ = 8.7, $J_{2,3}$ = 3.5 Hz, H-3 of β), 3.65*, 3.68 (3H, 2s, MeO of α*,β), 3.83-3.95 (4H, m, H-2,3,5 of α and H-2 of β), 4.465, 4.47, 4.68, 4.69 (1H each, 4d, ^2J =

10.7 Hz, CH_2O-4 of α,β), 4.62, 5.11* (1H, 2d, $J_{1,2}$ = 1.0, 1.2* Hz, H-1 of α*,β), 6.77, 6.75, 7.18, 7.19 (2H each, 4d, $J_{o,m}$ = 8.7 Hz, C_6H_4 of α,β). Anal. Calcd for $C_{14}H_{20}O_6$: C,59.14; H, 7.09. Found: C, 59.02; H, 7.26.

4.6. 4-*O*-(4-Methoxybenzyl)-L-rhamnose diethyl dithioacetal (17)

To a solution of **16** (95.4 mg, 0.336 mmol) in ethanethiol (3.6 mL) and AcOH (1.2 mL), *p*-toluenesulfonic acid monohydrate (6.4 mg, 0.034 mmol) was added at 0 °C. The mixture was stirred at ca. 10 °C for 40 min, diluted with saturated $NaHCO_3$, and concentrated *i*n vacuo. The residue was dissolved in $CHCl_3$, washed with water, dried ($MgSO_4$), and evaporated in vacuo. The residue was purified by column chromatography with 1:2 AcOEt-hexane to give **17** (95.4 mg, 73%) as a colorless prisms: mp 47–48 °C (from AcOEt-hexane); $[\alpha]_D^{26}$ –20.5° (*c* = 2.95, $CHCl_3$); R_f = 0.41 (1:1 AcOEt-hexane). ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.27, 1.28 (3H each, 2t, ^3J = 7.4 Hz, CH_3CH$_2$S), 1.29 (3H, d, $J_{5,6}$ = 6.6 Hz, H_3-6), 2.48 (3H, br s, HO-2,3,5), 2.61, 2.72 (2H each, 2q, CH_2S), 3.63 (1H, dd, $J_{4,5}$ = 4.4, $J_{3,4}$ = 1.5 Hz, H-4), 3.80 (3H, s, MeO), 3.86 (1H, dd, $J_{2,3}$ = 8.8, $J_{1,2}$ = 2.4 Hz, H-2), 4.07 (1H, dd, H-3), 4.14 (1H, qd, H-5), 4.25 (1H, d, H-1), 4.59, 4.67 (1H each, 2d, ^2J = 11.2 Hz, CH_2O-4), 6.88, 7.28 (2H each, 2d, $J_{o,m}$ = 8.8 Hz, C_6H_4). ^{13}C NMR (151 MHz, $CDCl_3$) δ 14.60 (*C*H$_3$CH$_2$S), 14.71 (*C*H$_3$CH$_2$S), 19.22 (C-6), 25.73 (CH_2S), 26.16 (CH_2S), 54.56 (C-1), 55.27 (MeO), 68.59 (C-5), 70.75 (C-3), 72.90 (CH_2O), 72.91 (C-2), 79.16 (C-4), 113.90 (C(*m*) of PMB), 129.87 (C(*o*) of PMB), 129.96 (C(*ipso*) of PMB), 159.49 (C(*p*) of PMB). Anal. Calcd for $C_{18}H_{30}O_5S_2$: C, 55.36; H, 7.42. Found: C, 55.48; H, 7.48.

4.7. 1,6-Dideoxy-1,1-bis(ethylsulfonyl)-4-*O*-(4-methoxybenzyl)-L-mannitol (18)

To a solution of **17** (199 mg, 0.510 mmol) in dry dioxane (4 mL) was added *m*CPBA (572 mg, 2.55 mmol). The mixture was stirred at rt for 1 h and then evaporated in vacuo. The residue was diluted with $CHCl_3$, washed with cold saturated $NaHCO_3$, dried ($MgSO_4$), and evaporated in vacuo. The residue was purified by column chromatography with 1:1 AcOEt-hexane to give **18** (179 mg, 77%) as colorless needles: mp 115–116 °C (from AcOEt-hexane); $[\alpha]_D^{27}$ –5.43° (*c* = 2.30, $CHCl_3$); R_f = 0.20 (1:1 AcOEt-hexane). ^{1}H NMR (600 MHz $CDCl_3$): δ = 1.31 (3H, d, $J_{5,6}$ = 6.4 Hz, H_3-6), 1.42, 1.44 (3H each, 2t, ^3J = 7.6 Hz, CH_3CH$_2$S), 2.37 (3H, br s, HO-2,3,5), 3.38, 3.57 (1H each, 2dq, ^2J = 14.0 Hz, CH_2S), 3.39, 3.62 (1H each, 2dq, ^2J = 14.0 Hz, CH'$_2$S), 3.53 (1H, dd, $J_{4,5}$ = 5.1, $J_{3,4}$ = 1.7 Hz, H-4), 3.81 (3H, s, MeO), 4.19 (1H, qd, H-5), 4.32 (1H, dd, $J_{2,3}$ = 9.5 Hz, H-3), 4.53, 4.68 (1H each, 2d, ^2J = 11.2 Hz, CH_2O-4), 4.66 (1H, dd, $J_{1,2}$ = 1.0 Hz, H-2), 4.83 (1H, d, H-1), 6.90, 7.30 (2H each, 2d, $J_{o,m}$ = 8.6 Hz, C_6H_4). ^{13}C NMR (151 MHz, $CDCl_3$): δ = 5.33 (*C*H$_3$CH$_2$S), 5.59 (*C*H$_3$CH$_2$S), 19.60 (C-6), 48.34 (CH$_3$*C*H$_2$S), 51.18 (CH$_3$*C*H$_2$S), 55.30 (MeO), 68.19 (C-5), 69.92 (C-3), 70.22 (C-2), 72.80 (CH_2O), 77.70 (C-1), 77.96 (C-4), 114.10 (C(*m*) of PMB), 129.22 (C(*o*) of PMB), 130.29 (C(*ipso*) of PMB), 159.71 (C(*p*) of PMB). Anal. Calcd for $C_{18}H_{30}O_9S_2$: C, 47.56; H, 6.65. Found: C, 47.68; H, 6.72.

4.8. 5-Deoxy-3-*O*-(4-methoxybenzyl)-α,β-L-arabinofuranoses (19)[14]

Compound **18** (153 mg, 0.337 mmol) was dissolved in 10% aqueous ammonia (3 mL). The mixture was stirred at rt for 10 h and then concentrated in vacuo. The residue was purified by column chromatography with 1:2 AcOEt-hexane to give an inseparable mixture (40:60) of α- and β-anomers of **19** (70.1 mg, 82%) as a colorless syrup: R_f = 0.15 (1:1 AcOEt-hexane), 0.54 (AcOEt). ^{1}H NMR spectra were in accord with previously published data.[14]

4.9. 5-Deoxy-3-*O*-(4-methoxybenzyl)-α,β-L-*erythro*-pentos-2-uloses (20)[14]

Compound **19** (282 mg, 1.11 mmol) was dissolved in MeOH (6 mL) and water (3 mL). The solution was refluxed and then cupric acetate hydrate (1.44 g, 7.23 mmol) was added. The mixture was refluxed for 1 h and then precipitates were filtered off and washed with ethyl acetate. The filtrate was evaporated in vacuo and the residue was separated by column chromatography with 1:3 AcOEt-hexane to give **20** (142 mg, 51% yield, lit,[14] 46%) as a colorless syrup: R_f = 0.25–0.33 (1:1 AcOEt-hexane). From the slower-eluting fraction, compound **17** (55.2 mg, 20%) was recovered.

4.10. N^2-(*N,N*-Dimethylaminomethylene)-1'-*O*-(4-methoxybenzyl)-3-[2-(4-nitrophenyl)ethyl]-biopterin (23)[14]

By use of the same procedures described in the literature,[14] compound **20** was converted into **23** in five steps: R_f = 0.60 (1:9 MeOH-$CHCl_3$).

4.11. N^2-(*N,N*-Dimethylaminomethylene)-1'-*O*-(4-methoxybenzyl)-3-[2-(4-nitrophenyl)ethyl]-2'-*O*-(2,3,4,6-tetra-*O*-benzoyl-β-D-glucopyranosyl)biopterin (24)

To a solution of **23** (56.0 mg, 0.100 mmol), glycosyl bromide **5** (200 mg, 0.303 mmol) and TMU (0.012 mL, 0.10 mmol) in dry CH_2Cl_2 (1.0 mL) was added silver triflate (56.0 mg, 0.218 mmol). The mixture was stirred at rt for 3 h in the dark, diluted with $CHCl_3$, and filtered through Celite. The filtrate was washed with aqueous $NaHCO_3$, dried (Na_2SO_4), and evaporated in vacuo. The residue was purified by column chromatography with 2:1 AcOEt-hexane to give **24** (85.6 mg, 75%) as a pale yellow foam: R_f = 0.38 (AcOEt); ^{1}H NMR (600 MHz, $CDCl_3$), see Table 1. Anal. Calcd for $C_{62}H_{57}N_7O_{15}$: C, 65.31; H, 5.04. Found: C, 65.18; H, 4.96.

4.12. N^2-(*N,N*-Dimethylaminomethylene)-3-[2-(4-nitrophenyl)ethyl]-2'-*O*-(2,3,4,6-terta-*O*-benzoyl-β-D-glucopyranosyl)biopterin (6)[13]

To a solution of **22** (54.4 mg, 0.0477 mmol) in CH_2Cl_2 (1.0 mL) containing water (0.10 mL) was added DDQ (16.2 mg, 0.0716 mmol). The mixture was stirred at rt for 2 h and then evaporated in vacuo. The residue was purified by column chromatography with 1:99 MeOH-$CHCl_3$ to give **6** (40.6 mg, 83%) as a pale yellow syrup: R_f = 0.63 (1:9 MeOH-$CHCl_3$); $[\alpha]_D^{20}$ +37.1° (*c* 2.17, $CHCl_3$). ^{1}H NMR spectra were in accord with previously published data.[13] Anal. Calcd for $C_{54}H_{49}N_7O_{14}$: C, 63.59; H, 4.84. Found: C, 63.44; H, 4.99.

4.13. 2'-*O*-(β-D-Glucopyranosyl)biopterin (2b)

4.13.1. From 6. Compound **6** (54.1 mg, 0.0530 mmol) was dissolved in MeOH (2.0 mL) and a 28% methanolic NaOMe (0.03 mL, 0.15 mmol) was added at 0 °C. The mixture was stirred at rt for 1 h and neutralized with Amberlite IR-120(H^+). The resin was filtered off and the filtrate was evaporated in vacuo. The residue was dissolved in MeOH (3.0 mL) and 28% aqueous ammonia solution (3.0 mL) was added. The mixture was stirred at rt for 12 h and evaporated in vacuo. The residue was dissolved in DMF (1.0 mL) and DBU (0.050 mL, 0.32 mmol) was added. The mixture was stirred at rt for 12 h, diluted with water (3.0 mL), and neutralized with Amberlite FPC3500(H^+). The resin was filtered off and the filtrate was evaporated in vacuo. The residue was washed with $CHCl_3$ and dried under reduced pressure to give **2b** (18.7 mg, 89% from **6**) as a yellow powder.

4.13.2. From 25b. Compound **25b** (26.0 mg, 0.0399 mmol) was dissolved in MeOH (2.0 mL) and 28% aqueous ammonia solution (1.0 mL) was added. The mixture was stirred at rt for 12 h and evaporated in vacuo. The residue was washed with $CHCl_3$ to give **2b** (15.6 mg, 98%): R_f = 0.23 (5:3:1 i-PrOH-AcOEt-H_2O); ^{1}H NMR (600 MHz, D_2O) and ^{13}C NMR (151 MHz, D_2O and DMSO-d_6), see Table 2. Anal. Calcd for $C_{15}H_{21}N_5O_8$: C, 45.11; H, 5.30. Found: C, 44.89; H, 5.53.

4.14. Di-N^2:1'-*O*-acetyl-2'-*O*-(2,3,4,6-tetra-*O*-acetyl-β-D-glucopyranosyl)biopterin (25b)

Compound **1b** (18.7 mg, 0.0468 mmol) was dissolved in pyridine (2.0 mL) and then acetic anhydride (1.0 mL) was added at 0 °C. The mixture was stirred at rt for 12 h and evaporated in vacuo. The residue was purified by column chromatography with AcOEt to give **25b** (28.0 mg, 92%) as a pale yellow syrup: R_f = 0.52 (1:9 MeOH:$CHCl_3$); ^{1}H NMR (600 MHz, $CDCl_3$), see Table 1. Anal. Calcd for $C_{27}H_{33}N_5O_{14}$: C, 49.77; H, 5.10. Found: C, 49.92; H, 5.02.

4.15. Methyl 2,3,4,6-tetra-*O*-acetyl-1-thio-β-D-glucopyranoside (27)[27]

The following modification of the literature procedures was made.[27] To a solution of **26** (5.41 g, 13.9 mmol) in dry acetonitrile (28 mL), were added thiourea (1.17 g, 15.4 mmol) and BF_3-etherate (3.7 mL, 29.1 mmol). The mixture was refluxed for 20 min and then TEA (23.2 mL, 166 mmol) and methyl

iodide (8.6 mL, 139 mmol) were slowly added at 0 °C. The reaction mixture was stirred at rt for 2 h and concentrated in vacuo. The residue was dissolved in $CHCl_3$ and then the mixture was washed with brine, dried (Na_2SO_4), and evaporated in vacuo. The residue was purified by column chromatography with 1:5 AcOEt-hexane to give **27** [4.66 g, 89% (lit.[27] 57% yield)] as colorless crystals: mp 89-90 °C (from AcOEt-hexane); R_f = 0.26 (1:2 AcOEt-hexane); ^{1}H NMR (600 MHz, $CDCl_3$) δ 2.01, 2.03, 2.07, 2.09 (3H each, 4s, AcO-2,3,4,6), 2.17 (3H, s, SMe), 3.73 (1H, ddd, $J_{4,5}$ = 10.0, $J_{5,6a}$ = 4.9, $J_{5,6b}$ = 2.4 Hz, H-5), 4.15 (1H, dd, $J_{6a,6b}$ = 12.5 Hz, H^b-6), 4.25 (1H, dd, H^a-6), 4.39 (1H, d, $J_{1,2}$ = 10.0 Hz, H-1), 5.07 (1H, dd, $J_{3,4}$ = 9.5 Hz, H-4), 5.08 (1H, dd, $J_{2,3}$ = 9.5 Hz, H-2), 5.23 (1H, t, H-3).

4.16. Methyl 4,6-*O*-isopropylidene-1-thio-β-D-glucopyranoside (28)

Compound **27** (607 mg, 1.60 mmol) was dissolved in dry MeOH (3.5 mL) and then a 28% methanoic sodium methoxide (0.87 mL, 4.33 mmol) was added at 0 °C. The mixture was stirred at rt for 2 h and neutralized with Amberlite IR-120(H^+). The resin was filtered off and the filtrate was evaporated in vacuo. The residue was dissolved in dry DMF (5.0 mL) and then 2,2-dimethoxypropane (0.79 mL, 6.42 mmol) and *p*-toluenesulfonic acid monohydrate (24 mg, 0.13 mmol) were added. The mixture was stirred at rt for 12 h and then pyridine (0.5 mL) was added. The mixture was concentrated in vacuo and the residue was purified by column chromatography with 1:2 AcOEt-hexane to give **28** (364 mg, 90%) as a colorless syrup: R_f = 0.16 (1:1 AcOEt-hexane); ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.43, 1.50 (3H each, 2s, Me_2C), 2.21 (3H, s, SMe), 3.00, 3.25 (1H each, 2br s, HO-2,3), 3.32 (1H, ddd, $J_{5,6b}$ = 10.3, $J_{4,5}$ = 9.6, $J_{5,6a}$ = 5.4 Hz, H-5), 3.47 (1H, dd, $J_{1,2}$ = 9.8, $J_{2,3}$ = 8.3 Hz, H-2), 3.57 (1H, t, $J_{3,4}$ = 9.3 Hz, H-4), 3.67 (1H, dd, H-3), 3.75 (1H, t, $J_{6a,6b}$ = 10.9 Hz, H^b-6), 3.93 (1H, dd, H^a-6), 4.32 (1H, d, H-1). Anal. Calcd for $C_{10}H_{18}O_5S$: C, 47.98; H, 7.25. Found: C, 48.11; H, 7.02.

4.17. Methyl 4,6-*O*-isopropylidene-2,3-di-*O*-(4-methoxybenzyl)-1-thio-β-D-glucopyranoside (29)

Compound **28** (778 mg, 3.11 mmol), *p*-methoxybenzyl chloride (2.10 mL, 15.5 mmol) and tetrabutylammonium iodide (344 mg, 0.93 mmol) were dissolved in dry DMF (30 mL) and then sodium hydride (60% in oil, 621 mg, 15.5 mmol) was added with stirring at 0 °C. The mixture was stirred at rt for 10 h, diluted with saturated NH_4Cl, and evaporated in vacuo. The residue was dissolved in $CHCl_3$, washed with water, dried (Na_2SO_4), and evaporated in vacuo. The residue was purified by column chromatography with 1:6 AcOEt-hexane to give **29** (1.25 g, 82% yield) as colorless prisms: mp 80–81 °C (from AcOEt-hexane); $[\alpha]_D^{26}$ +4.25° (*c* = 1.44, $CHCl_3$); R_f = 0.34 (1:4 AcOEt-hexane); ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.43, 1.51 (3H each, 2s, Me_2C), 2.20 (3H, s, SMe), 3.26 (1H, dt, $J_{5,6b}$ = 10.2, $J_{4,5}$ = 9.7, $J_{5,6a}$ = 5.4 Hz, H-5), 3.38 (1H, dd, $J_{1,2}$ = 9.8, $J_{2,3}$ = 8.5 Hz, H-2), 3.60 (1H, t, $J_{3,4}$ = 9.1 Hz, H-3), 3.69 (1H, t, H-4), 3.75 (1H, t, $J_{6a,6b}$ = 11.0 Hz, H^b-6), 3.80 (6H, s, MeO), 3.93 (1H, dd, H^a-6), 4.37 (1H, d, H-1), 4.69, 4.72*, 4.74*, 4.80 (1H each, 4d, ^2J = 11.0, 10.0* Hz, CH_2O-2,3), 6.855, 6.86 (1H each, 2d, $J_{o,m}$ = 8.6 Hz, *m* of PMB), 7.28, 7.30 (2H each, 2d, *o* of PMB). Anal. Calcd for $C_{26}H_{34}O_7S$: C, 63.65; H, 6.99.

Found: C, 63.81; H, 7.01.

4.18. Methyl 2,3-di-*O*-(4-methoxybenzyl)-1-thio-β-D-glucopyranoside (30)

To a solution of **29** (240 mg, 0.489 mmol) in MeOH (1.5 mL) was added 70% aqueous AcOH (3.0 mL). The mixture was stirred at rt for 10 h and evaporated in vacuo. The residue was purified by column chromatography with 1:1 AcOEt-hexane to give **30** (213 mg, 97% yield) as colorless needles: mp 45-46 °C (from AcOEt-hexane); $[\alpha]_D^{26}$ -16.2° (*c* = 1.81, $CHCl_3$); R_f = 0.14 (1:1 AcOEt-hexane); ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.97 (2H, br s, HO-4,6), 2.24 (3H, s, SMe), 3.33 (1H, ddd, $J_{4,5}$ = 9.4, $J_{5,6b}$ = 5.2, $J_{5,6a}$ = 3.4 Hz, H-5), 3.38 (1H, t, $J_{1,2}$ = 9.5, $J_{2,3}$ = 8.8 Hz, H-2), 3.45 (1H, t, $J_{3,4}$ = 8.9 Hz, H-3), 3.53 (1H, t, H-4), 3.74 (1H, dd, $J_{6a,6b}$ = 11.9 Hz, H^b-6), 3.80 (6H, s, MeO), 3.87 (1H, dd, H^a-6), 4.39 (1H, d, H-1), 4.64, 4.70*, 4.86*, 4.91 (1H each, 4d, ^2J = 11.3, 9.8* Hz, CH_2O-2,3), 6.88, 6.89 (2H each, 2d, $J_{o,m}$ = 8.5 Hz, *m* of PMB), 7.26, 7.36 (2H each, 2d, *o* of PMB). Anal. Calcd for $C_{23}H_{30}O_7S$: C, 61.31; H, 6.71. Found: C, 61.10; H, 6.93.

4.19. Methyl 4,6-di-*O*-acetyl-2,3-di-*O*-(4-methoxybenzyl)-1-thio-β-D-glucopyranoside (31)

Compound **30** (295 mg, 0.655 mmol) was dissolved in pyridine (3.0 mL) and then acetic anhydride (0.62 mL, 6.56 mmol) was added at 0 °C. The mixture was stirred at rt for 12 h and evaporated in vacuo. The residue was purified by column chromatography with 1:3 AcOEt-hexane to give **31** (342 mg, 98% yield) as colorless needles: mp 82–83 °C (from AcOEt-hexane); $[\alpha]_D^{26}$ -0.69° (*c* = 1.21, $CHCl_3$); R_f = 0.22 (1:2 AcOEt-hexane); ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.94, 2.06 (3H each, 2s, AcO-4,6), 2.22 (1H, s, SMe), 3.48 (1H, t, $J_{1,2}$ = 9.8, $J_{2,3}$ = 8.8 Hz, H-2), 3.54 (1H, ddd, $J_{4,5}$ = 9.8, $J_{5,6a}$ = 5.2, $J_{5,6b}$ = 2.4 Hz, H-5), 3.61 (1H, t, $J_{3,4}$ = 9.5 Hz, H-3), 3.80 (6H, s, MeO), 4.08 (1H, dd, $J_{6a,6b}$ = 12.2 Hz, H^b-6), 4.20 (1H, dd, H^a-6), 4.35 (1H, d, H-1), 4.59, 4.67*, 4.77, 4.81* (1H each, 4d, ^2J = 11.0, 10.1* Hz, CH_2O-2,3), 5.02 (1H, t, H-4), 6.855, 6.86 (2H each, 2d, $J_{o,m}$ = 8.8 Hz, *m* of PMB), 7.18, 7.30 (2H each, 2d, *o* of PMB). Anal. Calcd for $C_{27}H_{34}O_9S$: C, 60.66; H, 6.41. Found: C, 60.58; H, 6.55.

4.20. 4,6-Di-*O*-acetyl-2,3-di-*O*-(4-methoxybenzyl)-α-D-glucopyranosyl bromide (32)

Compound **31** (650 mg, 1.22 mmol) and 2,6-lutidine (0.40 mL, 3.41 mmol) were dissolved in dry CH_2Cl_2 (4.0 mL) and then bromine (0.15 mL, 2.92 mmol) was added with stirring at 0 °C. The mixture was stirred at rt for 2 h and then cyclohexene (0.30 mL, 2.96 mmol) was added. The mixture was concentrated in vacuo and the residue was dissolved in AcOEt. The insoluble matter was filtered off and the filtrate was evaporated in vacuo. The residue was purified by column chromatography with 1:4 AcOEt-hexane to give **32** (623 mg, 90%) as a colorless syrup: R_f = 0.42 (1:2 AcOEt-hexane); ^{1}H NMR (600 MHz, $CDCl_3$) δ 1.95, 2.05 (3H each, 2s, AcO-4,6), 3.53 (1H, dd, $J_{2,3}$ = 9.2, $J_{1,2}$ = 3.9 Hz, H-2), 3.80, 3.81 (3H each, 2s, MeO), 3.92 (1H, t, $J_{3,4}$ = 9.5 Hz, H-3), 4.02 (1H, dd, $J_{6a,6b}$ = 12.5, $J_{5,6b}$ = 2.1 Hz, H^b-6),

4.14 (1H, ddd, $J_{4,5}$ = 10.4, $J_{5,6a}$ = 4.6 Hz, H-5), 4.26 (1H, dd, H^a-6), 4.58*, 4.62, 4.67, 4.80* (1H each, 4d, ^2J = 11.6, 11.3* Hz, CH_2O-2,3), 5.05 (1H, dd, H-4), 6.26 (1H, d, H-1), 6.86, 6.88 (2H each, 2d, $J_{o,m}$ = 8.6 Hz, *m* of PMB), 7.19, 7.29 (2H each, 2d, *o* of PMB). Anal. Calcd for $C_{26}H_{31}BrO_9$: C, 55.03; H, 5.51. Found: C, 54.91; H, 5.74.

4.21. N^2-(*N,N*-Dimethylaminomethylene)-1'-*O*-(4-methoxybenzyl)-3-[2-(4-nitrophenyl)ethyl]-2'-*O*-[4,6-di-*O*-acetyl-2,3-di-*O*-(4-metoxybenzyl)-α-D-glucopyranosyl]biopterin (33a) and its β-anomer 33b

To a solution of the biopterin derivative (**23**) (30.0 mg, 0.0534 mmol), the D-glucopyranosyl bromide (**32**) (122 mg, 0.21 mmol) and TMU (0.0064 mL, 0.054 mmol) in dry CH_2Cl_2 (2.0 mL), was added silver triflate (28.0 mg, 0.109 mmol). The mixture was stirred at rt for 3 h and the suspension was filtered through Celite. The residue was washed with $CHCl_3$ and the filtrate was treated with saturated $NaHCO_3$. The mixture was extracted with $CHCl_3$ three times. The combined organic layers were dried (Na_2SO_4) and evaporated in vacuo. The residue was purified by column chromatography with 1:1 AcOEt-hexane to give an inseparable anomeric mixture (85:15) of **33a** and **33b** (37.0 mg, 66%) as a pale yellow syrup: R_f = 0.50 (AcOEt). From the slower-eluting fraction, compound **23** (7.2 mg, 24% recovery) was recovered: R_f = 0.12 (AcOEt).

33a: ^{1}H NMR (600 MHz, $CDCl_3$), see Table 1.

4.22. 1'-*O*-Acetyl-N^2-(*N,N*-dimethylaminomethylene)-3-[2-(4-nitrophenyl)ethyl]-2'-*O*-(2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl)biopterin (34a) and its β-anomer 34b

To a solution of **33a**,**b** (59.0 mg, 0.0563 mmol) in CH_2Cl_2 (2.0 mL) containing water (0.2 mL) was added DDQ (154 mg, 0.68 mmol). The mixture was stirred at rt for 2 h and then diluted with $CHCl_3$. The mixture was washed with aqueous $NaHCO_3$, dried (Na_2SO_4), and evaporated in vacuo. The residue was dissolved in dry pyridine (2.0 mL) and then acetic anhydride (0.27 mL, 2.82 mmol) was added at 0 °C. The mixture was stirred at rt for 12 h and then evaporated in vacuo. The residue was purified by column chromatography with 2:1 AcOEt-hexane to give **34a** (35.2 mg, 51% from **23**) and **34b** (6.2 mg, 9%).

34a: Pale yellow syrup; R_f = 0.27 (AcOEt); ^{1}H NMR (600 MHz, $CDCl_3$), see Table 1. Anal. Calcd for $C_{36}H_{43}N_7O_{15}$: C, 53.13; H, 5.33. Found: C, 53.01; H, 5.49.

34b: Pale yellow syrup; R_f = 0.30 (AcOEt); ^{1}H NMR (600 MHz, $CDCl_3$), see Table 1.

4.23. 2'-*O*-(α-D-Glucopyranosyl)biopterin (2a)

4.23.1. From 34a. Compound **34a** (30.2 mg, 0.0371 mmol) was dissolved in MeOH (2.0 mL) and 28% aqueous ammonia solution (2.0 mL) was added. The mixture was stirred at room temperature for 12 h

and then evaporated in vacuo. The residue was dissolved in DMF (2.0 mL) and DBU (0.027 mL, 0.18 mmol) was added. The mixture was stirred at rt for 12 h and neutralized with Amberlite FPC3500(H^+). The resin was filtered off and the filtrate was evaporated in vacuo. The residue was washed with $CHCl_3$ and dried under reduced pressure to give **2a** (13.4 mg, 90%).

4.23.2. From 25a. Compound **25a** (18.0 mg, 0.0276 mmol) was dissolved in MeOH (1.0 mL) and 28% aqueous ammonia solution (1.0 mL) was added. The mixture was stirred at rt for 12 h and then evaporated in vacuo. The residue was washed with $CHCl_3$ and dried under reduced pressure to give **2a** (10.4 mg, 94%) as a pale yellow solid: R_f = 0.11 (5:3:1 2-PrOH-AcOEt-H_2O); ^{1}H NMR (600 MHz, D_2O) and ^{13}C NMR (151 MHz, D_2O and DMSO-d_6), see Table 2. Anal. Calcd for $C_{15}H_{21}N_5O_8$: C, 45.11; H, 5.30. Found: C, 45.01; H, 5.50.

4.24. Di-*N*^{2}:1'-*O*-Acetyl-2'-*O*-(2,3,4,6-tetra-*O*-acetyl-α-D-glucopyranosyl)biopterin (25a)

Compound **2a** (13.4 mg, 0.0336 mmol) was dissolved in dry DMF (1.0 mL) and dry pyridine (1.0 mL) and then acetic anhydride (0.17 mL, 1.84 mmol) was added at 0 °C. The mixture was stirred at rt for 12 h and then evaporated in vacuo. The residue was purified by column chromatography with 4:1 AcOEt-hexane to give **25a** (19.6 mg, 90%) as a yellow syrup: R_f = 0.29 (AcOEt); ^{1}H NMR (600 MHz, $CDCl_3$), see Table 1. Anal. Calcd for $C_{27}H_{33}N_5O_{14}$: C, 49.77; H, 5.10. Found: C, 49.99; H, 5.29.

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