AN EFFICIENT SYNTHETIC ROUTE FOR A VERSATILE CILIAPTERIN DERIVATIVE AND THE FIRST CILIAPTERIN D-MANNOSIDE SYNTHESIS

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Abstract –The key precursor, \( N^2-(N,N\text{-dimethylaminomethylene})-1'-O-(4\text{-methoxybenzyl})-3\text{-}[2\text{-}(4\text{-nitrophenyl})\text{ethyl}]\text{ciliapterin (15)} \) was efficiently prepared from D-xylose via an improved route. The first synthesis of \( 2'\text{-}O\text{-}(\alpha\text{-D-mannopyranosyl})\text{ciliapterin (2c)} \) was achieved by treatment of \( 15 \) with \( 2,3,4,6\text{-}O\text{-benzoyl-} \alpha\text{-D-mannopyranosyl bromide in the presence of silver triflate and tetramethylurea, followed by removal of the protecting groups.} \)

INTRODUCTION

Some pterins having a hydroxyalkyl side-chain at C-6, such as biopterin (1a) and ciliapterin (L-threo-biopterin) (2a), have been found as glycosidic forms in certain prokaryotes. As examples of pterin glycosides from green sulfur photosynthetic bacteria, limipterin (1b) and tepidopterin (2b) were isolated from \( \text{Chlorobium limicola f. thiosulfatophilum} \) and \( \text{Chlorobium tepidum} \), respectively. Meanwhile, from cyanobacteria have been found \( 2'\text{-}O\text{-}(\alpha\text{-D-glucopyranosyl})\text{biopterin (1c)} \) (from \( \text{Anacystis nidulans} \), \( \text{Synechococcus sp.} \), and \( \text{Spirulina platensis} \)) and \( 2'\text{-}O\text{-}(\alpha\text{-D-mannopyranosyl})\text{ciliapterin (2c)} \) (from \( \text{Aphanizomenon flos-aquae} \)). Various other glycosides consisting of different pterins and sugar moieties have also been found in nature, although some of them have remained unclear concerning the position and the anomeric structure of the glycosidic linkage.
The physiological functions of these pterin glycosides appear to have been little investigated in contrast to the well-documented parent pterin: e.g., 1a exhibits enzyme cofactor activity in hydroxylation of aromatic amino acid and synthesis of nitric oxide as the form of its tetrahydro derivative. Attempts at preparing natural pterin glycosides have also scarcely been made so far, except for our synthetic studies on limipterin (1b) from 1a and tepidopterin (2b) from D-glucitol via an appropriately protected ciliapterin derivative (15) (Scheme 1). In this scheme, the L-threo configuration of the side chain of 15 was derived from those of C-3 and C-4 of L-xylose prepared from D-glucitol in three steps.

In the present study, we have attempted to improve preparation of the key intermediate (15) via an alternative route involving inversions of C-3 and C-4 configurations of D-xylose. We also describe the first synthesis of a natural pterin glycoside, 2'-O-(α-D-mannopyranosyl)ciliapterin (2c).

![Scheme 1](image-url)

**RESULTS AND DISCUSSION**

D-xylose was converted into 5-deoxy-1,2-O-isopropylidene-α-D-xylofuranose (4) via the 5-O-tosyl derivative (3) according to the reported procedures with a slightly modification (Scheme 2). The transformation of D-xylo configuration of 4 into L-lyxo form was achieved by way of the 3-enofuranose intermediate. Namely, compound 4 was oxidized with oxalyl chloride-DMSO to give the α-D-erythro-pentofuranos-3-uloose derivative (5) (in 98% yield), which was treated with acetic anhydride in pyridine at 80 °C to afford the enol acetate (6) in 74%. Hydrogenation of 6 in the presence of Pd-C exclusively proceeded from less hindered upper side of the furanose ring, providing the 3-O-acetyl-L-lyxofuranose derivative (7).

The 3-O-acetyl group of 7 was then cleaved with sodium methoxide in methanol to yield 5-deoxy-1,2-O-isopropylidene-β-L-lyxofuranose (8). Treatment of 8 with p-methoxybenzyl chloride and sodium hydride in DMF gave the 3-O-PMB derivative (9), which afforded 5-deoxy-3-O-PMB-L-lyxose (10) by hydrolysis in 70% acetic acid. The selective oxidation of 2-hydroxy group of 10 with cupric acetate provided the L-erythro-pentos-2-uloose derivative (11).
The pteridine ring formation of 11 with 2,5,6-triamino-4-hydroxypyrimidine (12) and the subsequent introduction of protecting groups were carried out by employing the reported procedures\(^\text{13}\) (Scheme 3). Namely, condensation of 11 and 12 in an aqueous sodium bicarbonate solution afforded an inseparable mixture of the ciliapterin derivative (13a) and its C-7 substituted isomer (13b) in a ratio of 75:25. These products were separated after the three-step procedures for introduction of N,N-dimethylaminomethylene, acetyl, and 2-(4-nitrophenyl)ethyl (NPE) groups, providing 2'-O-acetyl-\(N^2\)-(N,N-dimethylaminomethylene)-1'-O-PMB-3-NPE-ciliapterin (14a) (52% overall yield from 11) and its C-7 substituted congener (14b) (17%). Methanolation of the isolated 14a in the presence of sodium methoxide yielded the 1'-O-PMB derivative (15), a key precursor for the 2'-O-glycosylation. As an appropriate glycosyl donor for preparation of 2c, 2,3,4,6-tetra-O-benzoyl-\(\alpha\)-D-mannopyranosyl bromide (16)\(^\text{19}\), available from D-mannose in two steps, was utilized. Thus, glycosylation of 15 was attained by the condensation with 16 in the presence of silver triflate and tetramethylurea (TMU) in dichloromethane at room temperature for 3 h, affording the 2'-O-(\(\alpha\)-D-mannopyranosyl)ciliapterin derivative (17) in 77% yield. The \(\alpha\)-anomeric structure of 17 was derived from the results of NOE experiments of the \(^1\)H-NMR spectrum in addition to the characteristic tendency of the predominant \(\alpha\)-glycoside formation of D-mannose. In an NOESY spectrum of 17, no cross peaks with respect to H-1/H-3 and H-1/H-5 of the D-mannopyranosyl moiety were detected, whereas a strong NOE was observed between the 1,3-syn-diaxial H-3 and H-5 protons, indicating an equatorial orientation of H-1, namely an \(\alpha\)-anomeric form, of 17.

Removal of the protecting groups of 17 was carried out as follows: oxidation of 17 with 2,3-dichloro-5,6-dicyano-1,4-benzoquinone (DDQ) afforded 1'-O-free 18 (in 83% yield), which was treated with sodium methoxide in methanol to cleave benzoyl groups, and then with aqueous ammonia to cleave the \(N,N\)-dimethylaminomethylene group. The NPE group was finally removed by treatment with DBU\(^\text{20}\), thus providing 2'-O-(\(\alpha\)-D-mannopyranosyl)ciliapterin (2c) in 85% (overall yield from 17). The precise structure of 2c was established by \(^1\)H-NMR spectrum with the aid of decoupling techniques and
2D COSY measurement (Table 1). It is noteworthy that an extraordinary upfield shift was observed for the H-5 signal (δ 2.51) of the mannopyranosyl moiety compared with that of α-D-mannopyranose (δ 3.49). This suggests that 2c in a DMSO solution exists in such a conformation as H-5 of the sugar moiety locating above the pterin ring where an appreciable shielding effect is exerted. Thus the first synthesis of ciliapterin D-mannnoside (2c) was achieved utilizing an improved synthesis of the key precursor (15) by way of an alternative route from D-xylose in a 1.5 time better overall yield. Extension of this work including applications of these findings in synthesizing other natural pterin glycosides and their analogs is in progress.

EXPERIMENTAL

All reactions were monitored by TLC (Merck Silica gel 60 F254) with an appropriate solvent system [(A) 1:2, (B) 1:1 AcOEt-hexane, (C) AcOEt, and (D) 5:3:1 2-ProOH-AcOEt-H2O]. Column chromatography was performed with Daiso Silica Gel IR-60/210w. Components were detected by exposing the plates to UV light and/or 20% H2SO4-EtOH, with subsequent heating. Optical rotations were measured with a JASCO P-1020 polarimeter in CHCl3. The NMR spectra were measured in CDCl3 with Varian Unity...
The following modification of the literature procedures was made. To a solution of 3 (7.82 g, 22.7 mmol) in dry Et₂O (80 mL) was added lithium aluminum hydride (1.72 g, 45.5 mmol) at 0 °C under argon. The mixture was stirred at rt for 6 h, and then water was added. The precipitates were filtered

**Table 1.** 600 MHz ¹H-NMR Spectral Parameters for ciliapterin derivatives (15, 17, 18, 2c).^a

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<th>Compound</th>
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<th>Chemical shifts / δ (coupling constants / Hz)</th>
<th>Other signals</th>
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<th>H-5 (J₅₆)</th>
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<td>(1.6)</td>
<td>(11.7)</td>
<td>(3.4)</td>
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^a 15, 17, 18 in CDCl₃, 2c in DMSO-d₆.  
^b δ 7.21 [H(o), J₁-o₉ = 8.7 Hz], 6.84 [H(m)], 4.47, 4.41 (CH₃, ²J = 11.2 Hz).  
^c δ 7.28 [H(o), J₁-o₉ = 8.8 Hz], 6.78 [H(m)], 4.54, 4.50 (CH₃, ²J = 11.2 Hz).  
^d Confirmed by D₂O exchange.

Inova AS600 (600 MHz for ¹H, 151 MHz for ¹³C) at 23 °C, unless otherwise stated. The solvent peak was used as an internal standard for chemical shifts: in CDCl₃, δ 7.26 for ¹H, 77.00 for ¹³C; in DMSO-d₆, δ 2.50 for ¹H, 39.70 for ¹³C. The assignments of ¹³C signals were made with the aid of 2D C-H COSY measurements.

**5-Deoxy-1,2-O-isopropylidene-α-D-xylofuranose (4).**

The following modification of the literature procedures was made. To a solution of 3 (7.82 g, 22.7 mmol) in dry Et₂O (80 mL) was added lithium aluminum hydride (1.72 g, 45.5 mmol) at 0 °C under argon. The mixture was stirred at rt for 6 h, and then water was added. The precipitates were filtered
off and washed with CHCl₃. The filtrate was washed with aqueous NaCl, dried (Na₂SO₄), and evaporated in vacuo. The residue was purified by column chromatography with 1:2 AcOEt-hexane to give 4 (3.72 g, 94%) as colorless needles: mp 67–68 °C (from 1:1 AcOEt-hexane) (lit.,¹⁵α mp 66–67 °C, 84% yield); Rᵢ = 0.42 (B).

5-Deoxy-1,2-O-isopropylidene-α-D-erythro-pentofuranose-3-ulose (5).¹⁷
To a solution of oxalyl chloride (5.00 mL, 57.3 mmol) in dry CH₂Cl₂ (30 mL) was added a solution of DMSO (6.50 mL, 91.6 mmol) in dry CH₂Cl₂ (10 mL) at −60 °C. After having been stirred for 20 min, a solution of 4 (4.03 g, 23.1 mmol) in dry CH₂Cl₂ (5.0 mL) was slowly added at −60 °C. Stirring was continued at the same temperature for 8 h and then TEA (18.0 mL, 129 mmol) was added. The mixture was stirred at rt for 30 min, diluted with CHCl₃ and washed with water, dried (Na₂SO₄), and evaporated in vacuo. The residue was purified by column chromatography with 1:2 AcOEt-hexane to give 8 (3.91 g, 98%) [lit., 81%¹⁷b (by use of RuO₄), 83%¹⁷b (by use of PCC)] as a colorless syrup: Rᵢ = 0.33 (B).

3-O-Acetyl-5-deoxy-1,2-O-isopropylidene-α-D-glycero-pent-3-enofuranose (6).
To a solution of 5 (2.00 g, 11.6 mmol) in dry pyridine (15.0 mL) was added acetic anhydride (6.0 mL, 63 mmol). The mixture was stirred at 80 °C for 24 h and then concentrated in vacuo. The residue was diluted with AcOEt and the precipitate was filtered off. The filtrate was evaporated in vacuo and the residue was purified by column chromatography with 1:4 AcOEt-hexane to give 6 (1.84 g, 74%) as a pale yellow syrup: Rᵢ = 0.68 (A); ¹H-NMR δ 1.46, 1.50 (3H each, 2s, CMe₂), 1.77 (3H, d, 3J₂,₅ = 1.3 Hz, H-5), 2.21 (3H, s, AcO-3), 5.38 (1H, dq, 1J₁,₂ = 5.4 Hz, H-2), 5.98 (1H, d, H-1). Anal. Caled for C₁₀H₁₄O₅: C, 56.07; H, 6.59. Found: C, 55.88; H, 6.80.

3-O-Acetyl-5-deoxy-1,2-O-isopropylidene-β-L-lyxofuranose (7).
To a solution of 6 (1.80 g, 8.41 mmol) in AcOEt (25 mL) was added 10% Pd-C (1.50 g, 1.41 mmol). The mixture was stirred at rt for 4 h under atmospheric pressure of hydrogen. The catalyst was filtered off and the filtrate was evaporated in vacuo. The residue was purified by short-path column chromatography with 1:2 AcOEt-hexane to give 7 (1.71 g, 94%) as a colorless syrup: Rᵢ = 0.46 (A); [α]ᵢD²⁴ +65.4° (c 3.33); ¹H-NMR δ 1.35 (3H, d, 4J₄,₅ = 6.6 Hz, H-5), 1.36, 1.57 (3H each, 2s, CMe₂), 2.15 (3H, s, AcO-3), 4.27 (1H, quint, 3J₃,₄ = 6.1 Hz, H-4), 4.78 (1H, dt, 2J₂,₃ = 5.6, 1J₁,₂ = 4.4 Hz, H-2), 4.96 (1H, dd, H-3), 5.72 (1H, d, H-1); ¹³C-NMR δ 15.71 (COCH₃), 20.69 (C-5), 26.70, 26.83 (Me₂C), 72.53 (C-4), 75.71 (C-3), 79.57 (C-2), 104.50 (C-1), 114.37 (Me₂C), 170.38 (COCH₃). Anal. Caled for C₁₅H₁₆O₅: C, 55.55; H, 7.46. Found: C, 55.41; H, 7.62.

5-Deoxy-1,2-O-isopropylidene-β-L-lyxofuranose (8).
Compound 7 (975 mg, 4.51 mmol) was dissolved in dry MeOH (10 mL) and then NaOMe (28% in MeOH, 0.30 mL, 1.5 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 purified by column chromatography with 1:2 AcOEt-hexane to give 8 (715 mg, 91%) as
colorless needles: mp 86–88 °C (from AcOEt-hexane); \( R_f = 0.25 \) (A); [\( \alpha \)]\(_D\)\(^{24}\) +9.16° (c 2.21); \(^1\)H-NMR \( \delta \) 1.35 (3H, d, \( J_{3,5} = 6.4 \) Hz, H-5), 1.38, 1.60 (3H each, 2s, CMe\(_2\)), 2.60 (1H, d, \( J_{3,OH} = 8.1 \) Hz, HO-3), 4.11 (1H, quint, \( J_{3,4} = 6.1 \) Hz, H-4), 4.13 (1H, dt, \( J_{2,3} = 5.9 \) Hz, H-3), 4.63 (1H, dd, \( J_{1,2} = 4.4 \) Hz, H-2), 5.71 (1H, d, H-1); \(^{13}\)C-NMR \( \delta \) 15.18 (C-5), 26.69, 26.83 (Me\(_2\)C), 70.59 (C-4), 78.36 (C-3), 80.09 (C-2), 104.58 (C-1), 114.09 (Me\(_2\)C). Anal. Calcd for C\(_8\)H\(_{14}\)O\(_4\): C, 55.16; H, 8.10. Found: C, 54.99; H, 8.21.

5-Deoxy-1,2-O-isopropylidene-3-O-(4-methoxybenzyl)-\( \beta \)-L-lyxofuranose (9).

Compound 8 (292 mg, 1.68 mmol), \( p \)-methoxybenzyl chloride (0.450 mL, 3.30 mmol), and tetrabutylammonium iodide (180 mg, 0.49 mmol) were dissolved in DMF (10 mL) and, with stirring, sodium hydride (60% in mineral oil, 130 mg, 3.25 mmol) was added at 0 °C under argon. The mixture was stirred at rt for 12 h, diluted with saturated NH\(_4\)Cl (5 mL), and evaporated in vacuo. The residue was dissolved in CHCl\(_3\), washed with water, dried (Na\(_2\)SO\(_4\)), and evaporated in vacuo. The residue was purified by column chromatography to give 15 (465 mg, 94%) as a colorless syrup: [\( \alpha \)]\(_D\)\(^{24}\) +36.2° (c 2.91); \( R_f = 0.50 \) (A); \(^1\)H-NMR \( \delta \) 1.33, 1.61 (3H each, 2s, CMe\(_2\)), 1.41 (3H, d, \( J_{4,5} = 6.8 \) Hz, H-5), 3.81 (3H, s, MeO), 3.92 (1H, dd, \( J_{3,4} = 7.1, J_{2,3} = 5.1 \) Hz, H-3), 4.11 (1H, quint, H-4), 4.55, 4.64 (1H each, 2d, \( J = 11.8 \) Hz, CH\(_2\)O-3), 4.58 (1H, dd, \( J_{1,2} = 4.2 \) Hz, H-2), 5.68 (1H, d, H-1), 6.88, 7.29 (2H each, 2d, \( J_{o,m} = 8.8 \) Hz, C\(_6\)H\(_4\)); \(^{13}\)C-NMR \( \delta \) 16.91 (C-5), 26.01, 26.64 (Me\(_2\)C), 55.24 (MeO), 72.04 (CH\(_3\)O), 76.97 (C-4), 77.04 (C-3), 78.97 (C-2), 104.42 (C-1), 113.41 (Me\(_2\)C), 113.79 [C(m) of PMB], 129.51 [C(o) of PMB], 129.69 [C(ipsa) of PMB], 159.38 [C(p) of PMB]. Anal. Calcd for C\(_{16}\)H\(_{22}\)O\(_5\): C, 65.29; H, 7.53. Found: C, 65.18; H, 7.64.

5-Deoxy-3-O-(4-methoxybenzyl)-\( \alpha, \beta \)-L-lyxofuranoses (10).

A solution of 9 (200 mg, 0.679 mmol) in 70% AcOH (10 mL) was heated at 40 °C for 4 h. After addition of pyridine (0.20 mL), the mixture was evaporated in vacuo. The residue was purified by column chromatography to give an inseparable anomeric mixture (\( \alpha: \beta = 33:67 \)) of 10 (154 mg, 89%) as a colorless syrup: \( R_f = 0.08 \) (A), 0.15 (B); \(^1\)H-NMR for \( \alpha \)-anomer \( \delta \) 1.27 (3H, d, \( J_{4,5} = 6.6 \) Hz, H-3,5), 2.87 (1H, d, \( J_{2,OH} = 5.6 \) Hz, HO-2), 3.07 (1H, d, \( J_{1,OH} = 2.9 \) Hz, HO-1), 3.81 (3H, s, MeO), 4.09 (1H, td, \( J_{2,3} = 5.4, J_{1,2} = 1.5 \) Hz, H-2), 4.11 (1H, d, \( J_{3,4} = 4.9 \) Hz, H-3), 4.38 (1H, qd, H-4), 4.54, 4.56 (1H each, 2d, \( J = 11.2 \) Hz, CH\(_2\)O-3), 5.28 (1H, dd, H-1), 6.89, 7.27 (2H each, 2d, \( J_{o,m} = 8.8 \) Hz, C\(_6\)H\(_4\)). \(^1\)H-NMR for \( \beta \)-anomer \( \delta \) 1.33 (3H, d, \( J_{4,5} = 6.6 \) Hz, H-3,5), 2.89 (1H, d, \( J_{2,OH} = 9.5 \) Hz, HO-2), 3.46 (1H, d, \( J_{1,OH} = 11.2 \) Hz, HO-1), 3.81 (3H, s, MeO), 3.86 (1H, d, \( J_{2,3} = 4.6, J_{3,4} = 3.9 \) Hz, H-3), 4.06 (1H, qd, H-4), 4.10 (1H, dt, \( J_{1,2} = 4.4 \) Hz, H-2), 4.62, 4.64 (1H each, 2d, \( J = 11.2 \) Hz, CH\(_2\)O-3), 5.05 (1H, dd, H-1), 6.89, 7.27 (2H each, 2d, \( J_{o,m} = 8.8 \) Hz, C\(_6\)H\(_4\)). Anal. Calcd for C\(_{13}\)H\(_{18}\)O\(_5\): C, 61.41; H, 7.13. Found: C, 61.20; H, 7.29.

5-Deoxy-3-O-(4-methoxybenzyl)-\( \alpha, \beta \)-L-threreo-pentoses-2-uloses (11). Compound 10 (250 mg, 0.983 mmol) was dissolved in MeOH (6 mL) and water (3 mL). The solution was refluxed and then cupric acetate hydrate (1.00 g, 5.01 mmol) was added. The mixture was refluxed for 1 h and then precipitates were filtered off and washed with AcOEt. The filtrate was concentrated in vacuo and the residue was separated by column chromatography with 1:2 AcOEt-hexane to give 17 (126
mg, 51% yield) as a colorless syrup; $R_f = 0.33–0.24$ (B). From the slower-eluting fraction, compound 10 (51.0 mg, 20%) was recovered.

$N^2$-(N,N-Dimethylaminomethylene)-1’-O-(4-methoxybenzyl)-3-[2-(4-nitrophenyl)ethyl]ciliapterin (15).\(^\text{13}\)

By use of the same procedures described in the literature,\(^\text{13}\) compound 11 was converted into 15 in five steps: $R_f = 0.06$ (C); $^1$H-NMR, see Table 1.

$N^2$-(N,N-Dimethylaminomethylene)-1’-O-(4-methoxybenzyl)-3-[2-(4-nitrophenyl)ethyl]-2’-O-(2,3,4,6-tetra-O-benzoyl-α-D-mannopyranosyl)ciliapterin (17).

To a solution of 15 (35.0 mg, 0.0623 mmol), 2,3,4,6-tetra-O-benzoyl-α-D-mannopyranosyl bromide (16) (123 mg, 0.187 mmol) and TMU (0.008 mL, 0.066 mmol) in dry CH\(_2\)Cl\(_2\) (1.0 mL) was added silver triflate (32.0 mg, 0.125 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\(_2\)SO\(_4\)), and evaporated in vacuo. The residue was purified by column chromatography with 2:1 AcOEt-hexane to give 17 (54.7 mg, 77%) as a pale yellow foam; $R_f = 0.48$ (C); $^1$H-NMR, see Table 1. Anal. Calcd for C\(_{62}\)H\(_{57}\)N\(_7\)O\(_{15}\): C, 65.31; H, 5.04. Found: C, 65.09; H, 5.19.

$N^2$-(N,N-Dimethylaminomethylene)-3-[2-(4-nitrophenyl)ethyl]-2’-O-(2,3,4,6-tetra-O-benzoyl-α-D-mannopyranosyl)ciliapterin (18).

To a solution of 17 (74.9 mg, 0.0657 mmol) in CH\(_2\)Cl\(_2\) (2.0 mL) containing water (0.10 mL) was added DDQ (89.5 mg, 0.394 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\(_2\)SO\(_4\)), and evaporated in vacuo. The residue was purified by column chromatography with 1:99 MeOH-CHCl\(_3\) to give 18 (55.9 mg, 83%) as pale yellow syrup; $R_f = 0.12$ (C); $^1$H-NMR, see Table 1. Anal. Calcd for C\(_{54}\)H\(_{49}\)N\(_7\)O\(_{14}\): C, 63.59; H, 4.84. Found: C, 63.70; H, 5.01.

2’-O-(α-D-Mannopyranosyl)ciliapterin (2c).

Compound 18 (55.9 mg, 0.0548 mmol) was dissolved in dry MeOH (2.0 mL) and then NaOMe (28% in MeOH, 0.030 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 (4.0 mL) and 28% aqueous ammonia solution (4.0 mL) was added. The mixture was stirred at rt for 2 h and evaporated in vacuo. The residue was dissolved in DMF (1.0 mL) and DBU (0.050 mL, 0.33 mmol) was added. The mixture was stirred at rt for 20 h, diluted with water (4.0 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 2c (18.6 mg, 85%) as pale yellow solid; $R_f = 0.25$ (D); $^1$H-NMR, see Table 1; $^{13}$C-NMR (DMSO-$d_6$) $\delta$ 14.90 (C-3\(^\prime\)), 60.66 (C-6\(^\prime\)), 66.34 (C-4\(^\prime\)), 70.93 (C-2\(^\prime\)), 70.98 (C-3\(^\prime\)), 72.82 (C-2\(^\prime\)), 73.73 (C-5\(^\prime\)), 75.09 (C-1\(^\prime\)), 96.98 (C-1\(^\prime\)), 127.45 (C-4a), 148.35 (C-7), 151.40 (C-6), 155.20 (C-8a), 156.51 (C-2), 163.15 (C-4), *for
glycosyl moiety. Anal. Calcd for C_{15}H_{21}N_{5}O_{8}: C, 45.11; H, 5.30. Found: C, 44.85; H, 5.51.

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REFERENCES AND NOTES
7. Although NMR spectral data of ciliapterin D-mannoside (2c) have not been reported in Ref. 6, the hydrolyzed products of 2c were identified to be ciliapterin (2a) and D-mannose. The 2′-O-(α-D-mannopyranosyl) form of 2c was assigned to be the most likely glycosidic linkage on the ground of the structural tendency of other pterin glycosides and the predominant α-glycoside formation of D-mannose.
21. $^1$H NMR of α-D-mannopyranose (DMSO-$d_6$, D$_2$O exchange) δ 3.34 (1H, t, $J_{3,4} = J_{4,5} = 9.5$ Hz, H-4), 3.43 (1H, dd, $J_{6a,6b} = 11.5$, $J_{5,6b} = 5.9$ Hz, H$^b$-6), 3.49 (1H, ddd, $J_{5,6a} = 2.0$ Hz, H-5), 3.51–3.53 (2H, m, H-2,3), 3.60 (1H, dd, H$^a$-6), 4.85 (1H, d, $J_{1,2} = 1.5$ Hz, H-1).