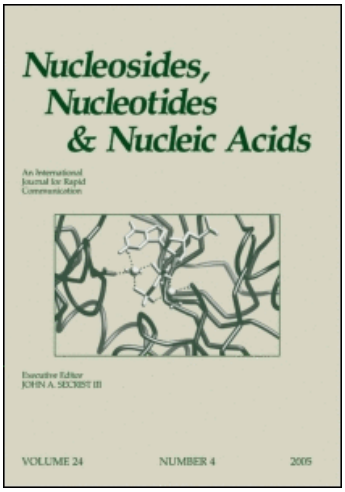

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Nucleosides. LXV. Synthesis of New Pteridine-N_8-Nucleosides

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Nucleosides. LXV.[#] Synthesis of New Pteridine–N_8–Nucleosides[†]

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ABSTRACT

A general synthetic approach to various isoxanthopterin-nucleosides starting from 6-methyl-2-methylthio-4(3H), 7(8H)-pterdinedione (**1**) has been developed. Ribosylation with 1-O-acetyl-2,3,5-tri-O-benzoyl-β-D-ribofuranose via the silyl-method led to **2** and reaction with 1-chloro-2-deoxy-3,5-di-O-p-toluoyl-α-D-ribofuranose using the DBU-method afforded **28**. Protection of the amide function at O^4 by benzylation to **5** and by a Mitsunobu reaction with 2-(4-nitrophenyl)ethanol to **29** gave soluble intermediates which can be oxidized to the corresponding 2-methylsulfonyl derivatives **8** and **30**, respectively. Nucleophilic displacement reactions of the highly reactive 2-methylsulfonyl functions by various amines proceeded under mild conditions to isoxanthopterin-N_8-ribo- (**11–17**) and 2′-deoxyribomucleosides (**31–33**). Debenzylation can be achieve by Pd-catalyzed hydrogenation (**9** to **19**) and cleavage of the npe-protecting group (**31**, **32** to **34**, **35**) works well with DBU by β-elimination.

Key Words: Pteridine nucleosides; Ribosylation reactions; 2-(4-nitrophenyl)ethyl protecting group; Mitsunobu reaction.

[#]See Ref. [1].

[†]In honor and celebration of the 70th birthday of Professor Leroy B. Townsend.

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INTRODUCTION

Pteridine–N_8–nucleosides can be regarded as homologous purine-nucleosides, however, the enlargement of the five-membered imidazole—into the six-membered pyrazine-ring changes the chemistry from a π-excessive into a π-deficient nitrogen-heterocycle. Pteridine–nucleoside syntheses can be performed by various methods including the silyl-approach,[2–4] the sodium salt method[5] and the DBU-catalyzed glycosylation reaction,[6–8] respectively. Since substituted pteridine derivatives[9] are fluorescent this feature is of great interest in labeling experiments especially of oligonucleotides. A lumazine fluorophore has been incorporated into oligonucleotides as a nucleoside analog to study in conjunction with a ruthenium complex energy transfer interactions.[10,11] Isoxanthopterin nucleosides, in particular, show high fluorescene quantum yields in the region of 0.9[9] and can therefore facilitate fluorescence studies involving oligonucleotides.[12] In order to extent our knowledge about pteridine nucleoside fluorophores new types related to isoxanthopterin-N_8-ribo- and 2′-deoxyribofuranosides have been synthesized. Since isoxanthopterin and its derivatives are a very insoluble substances direct glycosylation reactions, in general, have so far been without success. A valuable starting material to achieve the anticipated reactions, however, has been found in the 6-methyl-2-methylthio-4(3H), 7(8H)-pteridinedione **1**[7] which shows the afforded physical and chemical properties to achieve glycosylations in organic solvents followed by chemical modifications of the various substituents.

RESULTS AND DISCUSSION

The ribosylation of 6-methyl-2-methylthio-4(3H), 7(8H)-pteridinedione **1**[7] with 1-O-acetyl-2,3,5-tri-O-benzoyl-β-D-ribofuranose was performed by the Birkofer–Hilbert–Johnson silyl-method[13,14] under BF_3–etherate catalysis in analogy to former experiments[15] to give **2** in 45% yield. Treatment of **2** with methanolic ammonia at room temp. proceeded with debenzoylation of the sugar moiety leading to 6-methyl-2-methylthio-8-β-D-ribofuranosyl-4(3H), 7(8H)-pteridinedione **3** in good yield. In order to improve the solubility of **2** in organic solvents the amide function was protected by benzylation with benzyl bromide in presence of silver carbonate to give a mixture of 4-benzyloxy-2-methylthio-8-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)-7(8H)-pteridinone **4** and the corresponding 3-benzyl isomer **5** in 69% and 20% yield, respectively. 3-Benzyl-6-methyl-2-methylthio-8-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl-4(3H), 7(8H)-pteridinedione **5** is highly reactive towards nucleophiles at the aglycone moiety forming 3-benzyl-6-methylisoxanthopterin-8-ribofuranoside **6** on treatment with methanolic ammonia. The analogous reaction with **4** lead, expectedly, to deprotection of the sugar benzoyl groups but additionally in this case displacement of the 4-benzyloxy function took place yielding 4-amino-6-methyl-2-methylthio-8-β-D-ribofuranosyl-7(8H)-pteridone **7**. In order to get selective substitution in 2-position **4** was oxidized by m-chloroperbenzoic acid to the corresponding 2-methylsulfonyl derivative **8** in 92% yield which showed the expected reactivity due to the fact that the methylsulfonyl function is an excellent leaving group in nucleophilic displacement reactions. Compound **8** turned out to be a valuable intermediate for modifying the pteridine

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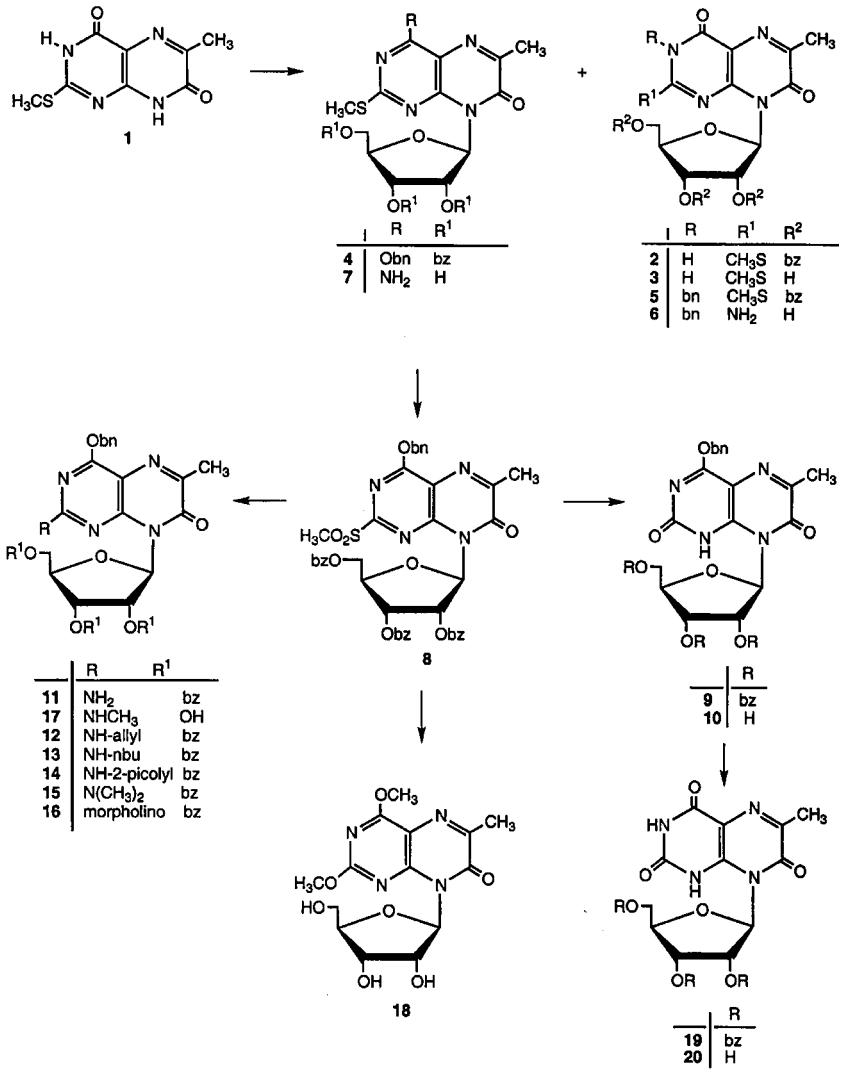
nucleus. Hydrolysis of **8** by 0.1 N NaOH in THF afforded 4-benzyloxy-6-methyl-8-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)-2(1H),7(8H)pteridinedione **9** whereas 1 N NaOH in THF/H_2O was associated with deblocking of the sugar moiety to give **10**. Treatment of **8** with ammonia, allylamine, n-butylamine, 2-picolylamine, dimethylamine and morpholine in organic solvents like, dioxane, THF or AcOEt proceeded well to the 4-benzyloxy-2-subst.amino-8-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)-7-(8H)-pteridones **11–16**. High conc. methanolic methylamine removed simultaneously the benzoyl groups yielding **17** directly. Finally methanolate treatment of **8** afforded in one step 2,4-dimethoxy-6-methyl-8-β-D-ribofuranosylpteridine **18**. The advantage of the benzyl protecting group is seen in its easy removal by Pd/C catalysed hydrogenolysis converting



Scheme 1.

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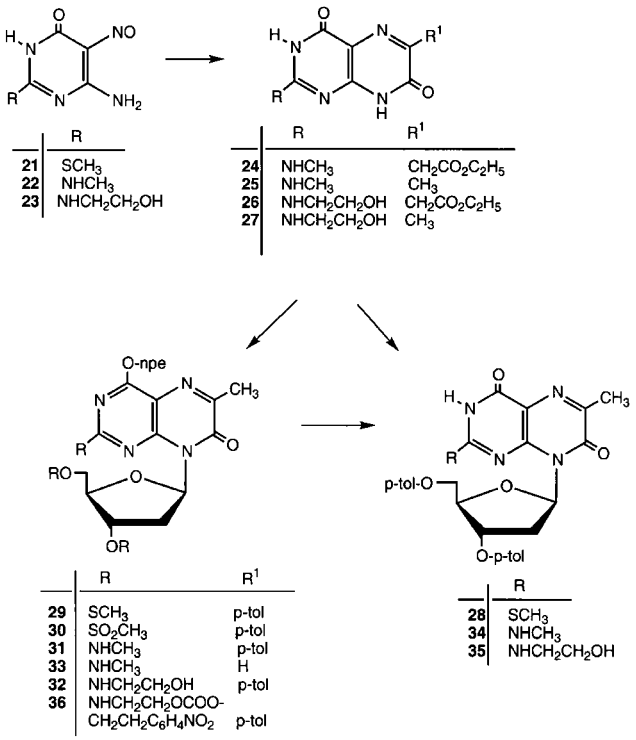
9 into **19** followed by debenzoylation to give 6-methyl-8-β-D-ribofuranosyl-2(1H),4(3H),7(8H)-pteridinetrione **20** (Scheme 1).

In a second series of glycosylation reactions attempts were undertaken to convert N^2,6-dimethyl- **25** and N^2-β-hydroxyethyl-6-methylisoxanthopterin **27** directly with 1-chloro-2-deoxy-3,5-di-O-p-toluoyl-α-D-ribofuranose into their 8-β-D-deoxyribofuranosides. The starting isoxanthopterin derivatives **25** and **27** have been synthesized from 6-amino-2-methylthio-5-nitroso-4(3H)pyrimidone **21** by treatment with methylamine to **22** and ethanolamine to **23**, respectively, followed by reduction to the corresponding 5-amino derivatives, regioselective condensation with ethyl sodium-oxalylacetate to N^2-methyl- **24** and N^2-β-hydroxyethyl-6-ethoxycarbonyl-isoxanthopterin **26** both of which can be hydrolysed and decarboxylated to give **25** and **27**, respectively. The DBU-catalysed glycosylations in acetonitrile, however, were very unsatisfactorily since due to the low solubility of the starting pteridines only low yields of α,β-anomeric mixtures were obtained. From these disappointing results was realized that the anticipated syntheses of new isoxanthopterin-deoxyribofuranoses must start from 6-methyl-2-methylthio-8-(2-deoxy-3,5-di-O-p-toluoyl-β-D-ribofuranosyl)-4(3H),7(H)-pteridinedione **28**[7] which could be prepared in improved yield of 42%. Protection of the amide function by a Mitsunobu reaction forming the O^4-2-(4-nitrophenyl)ethyl derivative **29** and its oxidation to the corresponding 2-methylsulfonyl pteridine-2′-deoxynucleoside **30** was achieved by known procedures[7] in the reported high yields. Displacement



Scheme 2.

reactions of the methylsulfonyl group by dry methylamine and ethanolamine in CH_2Cl_2, respectively, proceeded well to give **31** and **32**. Deacylation of the sugar moiety in **31** works best in MeOH with KCN to form **33**. Removal of the O^4-2-(4-nitrophenyl)ethyl group in **31** and **33** was performed with DBU in a β-elimination process yielding **34** and **35**, respectively. Finally, it was shown that 2-β-hydroxyethylamino-6-methyl-4-2-(4-nitrophenyl)ethoxy-8-(2-deoxy-3,5-di-O-p-toluoyl-β-D-ribofuranosyl)-7(8H)-pteridinone **32** can fully be protected at the β-hydroxy group by treatment with 2-(4-nitrophenyl)ethyl chloroformate forming **36** in 88% yield (Scheme 2).

EXPERIMENTAL

All melting points are uncorrected. Products were dried under high vacuum. TLC: precoated silica gel thin-layer sheets *60 F_{254}* from *Merck*. Column chromatography (CC): silica gel (*Baker*, 30–60 μm); 0.2–0.3 bar. UV/VIS: *Perkin–Elmer Lambda 5*; λ_{max} in nm (log ε). ^{1}H-NMR: *Bruker AC 250*; δ in ppm rel. to $SiMe_4$ or $CDCl_3$, (DMSO–d_6) as internal standard. All solvents used were of anhydrous grade.

6-Methyl-2-methylthio-8-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)-4(3H),7(8H)-pteridinedione (2). A suspension of 6-methyl-2-methylthio-4(3H),7(8H)-pteridinedione (**1**)[7] (13.84 g, 62 mmol) and $(NH_4)_2SO_4$ (0.25 g) in freshly destilled hexamethyldisilazane (HMDS) (150 ml) was heated under reflux and exclusion of moisture for 6 h. The excess of HMDS was distilled off, the yellowish residue kept under high vacuum for 2 h and was then dissolved in CH_2Cl_2 (300 ml). A solution of 1-O-acetyl-2,3,5-tri-O-benzoyl-β-D-ribofuranose (31.28 g, 62 mmol) in CH_2Cl_2 (400 ml) was added, followed by BF_3–etherate (30 ml) and then the mixture stirred at r.t. for 24 h. The reaction solution was poured slowly into a saturated solution of $NaHCO_3$ (400 ml) (fuming!) forming an emulsion. It was stirred for 2 h and then kept in a separation funnel over night. The organic layer was separated, dried over Na_2SO_4, evaporated and coevaporated with EtOH yielding a redish amorphous foam (40.1 g). Purification was achieved by flash silica–gel column chromatography in batches of 5.5 g with toluene/AcOEt 2:1 (600 ml), toluene/AcOEt 1:1 (500 ml), toluene/AcOEt 1:3 (500 ml) and toluene/AcOEt 1:5 (500 ml) collecting 200 ml fractions. The product **2** eluted in fractions 5–8 yielding on evaporation 2.74 g (45%) giving a total of 18.6 g. Recrystallization of 1 g from 100 ml of EtOH gave 0.85 g of yellow-orange crystals of mp. 232–234°C. UV (MeOH): 228 (4.68), [263, (3.96)], 282 (3.82), 298 (3.77), 344 (4.09), [354 (4.05)]. pK_a = 8.70. ^{1}H-NMR ($CDCl_3$): 12.5 (bs, 1H, H–N(3)); 8.12–7.84 (m, 6H, arom.H); 7.60–7.27 (m, 9H, arom.H); 7.19 (d, 1H, H–C(1′)); 6.33 (pt, 1H, H–C(3′)); 6.20 (dd, 1H, H–C(2′)); 4.92–4.64 (m, 3H, H–C(4′;5′;5″)); 2.51 (s, 3H, Me–S); 2.39 (s, 3H, Me(6).

Anal. For $C_{34}H_{28}N_4O_9S$ (668.6) Calcd.: C, 61.08; H, 4.22; N, 8.38. Found: C, 60.98; H, 4.23; N, 8.43.

6-Methyl-2-methylthio-8-β-D-ribofuranosyl-4(3H),7(8H)-pteridinedione (3). In saturated methanolic ammonia (50 ml) compound **2** (1.2 g, 1.8 mmol) was stirred at r.t. for 72 h. It was evaporated to dryness, the residue dissolve in H_2O (150 ml), acidified with AcOH and then extracted with AcOEt (4 × 50 ml). The H_2O layer was

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concentrated till a precipitate starts to separate. The colorless crystals were collected after standing over night, washed with little cold H_2O and dried in high vacuum to give 0.487 g (76%) of mp. 235°C (decomp.). UV (pH 4): 214 (4.43), [244, 399)], 261 (4.09), 351 (4.15). pK_a = 6.55. ^{1}H-NMR (DMSO–d_6): 13.16 (bs, 1H, H–N(3)); 6.73 (d, 1H, H–C(1′)); 4.56 (dd, 1H, H–C(2′)); 4.21 (pt, 1H, H–C(3′)); 3.79–3.37 (m, 3H, H–C(4′;5′;5″)); 2.54 (s, 3H, Me–S); 2.28 (s, 3H, Me(6).

Anal. For $C_{13}H_{16}N_4O_6S$ (356.6) Calcd.: C, 43.82; H, 4.53; N, 15.72. Found: C, 44.03; H, 4.58; N, 15.66.

3-Benzyl-6-methyl-2-methylthio-8-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)-4(3H),7(8H)-pteridinedione (4) and 4-benzyloxy-6-methyl-2-methylthio-8-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)-4(3H),7(8H)-pteridinedione (5). A suspension of **2** (6.05 g (9 mmol) and silver carbonate (6.34 g) in dry benzene (110 ml) was heated with stirring to 80°C for 2 h. After cooling to 60°C benzylbromide (4.5 ml, 36 mmol) was added and the mixture stirred at 60–70°C for 48 h. The reaction solution was filtered hot through a thin layer of SiO_2, washed several times with toluene and the filtrates concentrated to 20 ml for CC (SiO_2, 220 g, 250 ml fractions) with each 500 ml of toluene/AcOEt 50:1, 40:1, 30:1, 20:1, 15:1, 10:1, 8:1, 5:1, 3:1 and 2:1. Fraction 9–13 gave after evaporation 4.71 g (69%) of **5** and fractions 17–20 1.38 g (20%) of **4**.

4: Recrystallization from EtOH (80 ml) gave 0.956 g (14%) of a colorless crystal powder of mp. 108–109°C. UV (MeOH): 227 (4.78), 282 (4.61), 301 (3.91), 343 (4.15), [353 (4.06)]. ^{1}H-NMR ($CDCl_3$): 8.12–7.26 (m, 20H, arom.H); 7.16 (d, 1H, H–C(1′)); 6.35 (pt, 1H, H–C(3′)); 6.19 (dd, 1H, H–C(2′)); 5.50 (d, 1H, CH_2); 5.26 (d, 1H, CH_2); 4.90–4.63 (m, 3H, H–C(4′;5′;5″)); 2.54 (s, 3H, Me–S); 2.34 (s, 3H, Me(6).

Anal. For $C_{41}H_{34}N_4O_9S$ (758.8) Calcd.: C, 64.90; H, 4.52; N, 7.38. Found: C, 64.89; H, 4.45; N, 7.24.

5: Recrystallization from EtOH (180 ml) gave 4.13 g (61%) of a colorless crystal powder of mp. 89–91°C. UV (MeOH): 224 (4.72), 275, (3.84), 282 (3.86), 328 (4.19), 333 (4.20), [346 (4.11)]. ^{1}H-NMR (DMSO–d_6): 7.98–7.79 (m, 5H, arom.H); 7.69–7.32 (m, 15H, arom.H); 7.18 (d, 1H, H–C(1′)); 6.34 (pt, 1H, H–C(3′)); 6.18 (dd, 1H, H–C(2′)); 5.65–5.54 (dd, 2H, CH_2); 4.83–4.52 (m, 3H, H–C(4′;5′;5″)); 2.51 (s, 3H, Me–S); 2.38 (s, 3H, Me(6).

Anal. For $C_{41}H_{34}N_4O_9S$ (758.8) Calcd.: C, 64.90; H, 4.52; N, 7.38. Found: C, 64.97; H, 4.54; N, 7.31.

3-Benzyl-6-methyl-8-β-D-ribofuranosyl-isoxanthopterin (6). A suspension of **4** (1.19 g, 1.5 mmol) in saturated methanolic ammonia (30 ml) was stirred at r.t. for 5 days under exclusion of light to give an orange colored solution. It was evaporated to dryness, the residue treated in toluene/AcOEt 10:1 by ultrasound and then the solid (0.5 g) collected. Recrystallization from H_2O (7 ml) with charcoal gave 0.372 g (59%) of a colorless crystal powder of mp. 154°C (decomp.). UV (MeOH): 217 (4.56), [257 (3.54)], 297 (3.98), 346 (4.11). ^{1}H-NMR (DMSO–d_6): 7.77 (bs, 2H, NH_2); 7.40–7.15 (m, 5H, arom. H); 6.62 (d, 1H, H–C(1′)); 5.28–5.12 (2d, 2H, CH_2); 4.62 (dd, 1H, H–C(2′)); 4.25 (pt, 1H, H–C(3′)); 3.73–3.41 (m, 3H, H–C(4′;5′;5″)); 2.23 (s, 3H, Me(6).

Anal. For $C_{19}H_{21}N_5O_6$ (415.4) Calcd.: C, 54.94; H, 5.10; N, 16.86. Found: C, 54.68; H, 5.41; N, 16.85.

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4-Amino-6-methyl-2-methylthio-8-β-D-ribofuranosyl-7(8H)-pteridinone (7). A suspension of **5** (0.758 g, 1 mmol) in saturated methanolic ammonia (20 ml) was stirred at r.t. for 3 days. The resulting solution was evaporated, the residue was treated with cold MeOH (5 ml), filtered and dried to give 0.247 g (69%). Recrystallization from EtOH gave 0.167 g (47%) of a crystal powder of mp. 242°C (decomp.). UV (MeOH): 216 (4.42), [248 (4.05)], 264 (4.16), [292 (3.65)], 346 (4.13). ^{1}H-NMR (DMSO–d_6): 7.77 (bs, 1H, NH_2); 7.58 (bs, 1H, NH_2); 6.67 (d, 1H, H–C(1′)); 4.59 (dd, 1H, H–C(2′)); 4.23 (pt, 1H, H–C(3′)); 3.78–3.47 (m, 3H, H–C(4′;5′;5″)); 2.32 (s, 3H, Me(6).

Anal. For $C_{13}H_{17}N_5O_5S$ (355.4) Calcd.: C, 43.94; H, 4.82; N, 19.71. Found: C, 44.10; H, 4.88; N, 19.38.

4-Benzyloxy-6-methyl-2-methylsulfonyl-8-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)-7(8H)-pteridinone (8). To a solution of **5** (10.5 g, 13.8 mmol) in CH_2Cl_2 (350 ml) was added in small portions m-chloroperbenzoic acid (8.25 g containing 15% chlorobenzoic acid) with stirring. After 12 h the precipitated m-chlorobenzoic acid was filtered off, the filtrate concentrated to 50 ml and then put onto a silica–gel column (240 g SiO_2, 250 ml fractions) for chromatography with toluene/AcOEt 100:1 (1 l), 50:1 (1 l), 30.1 (600 ml), 20:1 (600 ml), 10:1 (600 ml), 7:1–3:1 (2 l). The product fractions 13–19 were evaporated, the residue coevaporated with CH_2Cl_2 (3 × 100 ml) and then dried in high vacuum to give 10.2 g (92%) of a colorless solid foam. UV (MeOH): [217 (4.64)], 231 (4.76), [276 (3.90)], 282 (3.94), 307 (4.08), [314 (4.05)]. ^{1}H-NMR (DMSO–d_6): 7.98–7.79 (m, 5H, arom.H); 7.69–7.32 (m, 15H, arom.H); 7.18 (d, 1H, H–C(1′)); 6.34 (pt, 1H, H–C(3′)); 6.18 (dd, 1H, H–C(2′)); 5.65–5.54 (dd, 2H, CH_2); 4.83–4.52 (m, 3H, H–C(4′;5′;5″)); 2.51 (s, 3H, Me–S); 2.38 (s, 3H, Me(6)).

Anal. For $C_{41}H_{34}N_4O_{11}S$ (790.8) Calcd.: C, 62.27; H, 4.33; N, 7.09. Found: C, 61.75; H, 4.28; N, 6.91.

4-Benzyloxy-6-methyl-8-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)-2(1H),7(8H)-pteridinedione (9). To a stirred solution of **8** (0.84 g, 1.06 mmol) in THF (24 ml) was added very slowly dropwise in 6 h 0.1 N NaOH (24 ml). The emulsion was concentrated in vacuum to 20 ml, then diluted with H_2O (30 ml) and treated with Lewatit ion-exchange-resin (H^+ form) to pH 4–5. The solution was extracted with AcOEt (4 × 50 ml), the H_2O layer evaporated and the residue purified by CC with a gradient of toluene/AcOEt 50:1–5:1. The product fraction was evaporated to give 0.58 g of a solid foam. Further purification was done on preparative silica–gel plates by repeated chromatography with $CHCl_3$/MeOH 100:1 (3 ×) and $CHCl_3$/MeOH 80:1 (1 ×). The main band was eluted with AcOEt, evaporated and coevaporated with CH_2Cl_2 to give 0.22 g (28%) of mp. 161°C. UV (pH 5): 206 (4.76), 232 (4.61), [274, (3.97)], 327 (4.01), [342 (3.93)]. ^{1}H-NMR ($CDCl_3$): 8.05–7.82 (m, 5H, arom.H); 7.55–7.20 (m, 15H, arom.H); 6.93 (s, 1H, H–C(1′)); 6.52 (bs, 1H, H–C(3′)); 6.33 (bs, 1H, H–C(2′)); 5.50 (dd, 2H, CH_2); 4.90–4.52 (m, 3H, H–C(4′;5′;5″)); 2.50 (s, 3H, Me(6)).

Anal. For $C_{40}H_{32}N_4O_{10}$ (728.7) Calcd.: C, 65.93; H, 4.43; N, 7.69. Found: C, 65.58; H, 4.49; N, 7.42.

4-Benzyloxy-6-methyl-8-β-D-ribofuranosyl-2(1H),7(8H)-pteridinedione (10). To a solution of **8** (1.0 g, 1.26 mmol) in THF (60 mnl) and H_2O (30 ml) was added slowly dropwise with stirring 1 N NaOH (10 ml). After stirring at r.t. for 24 h

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the reaction solution was slightly acidified with AcOH, the THF was distilled off, H_2O (30 ml) was added and then nextracted with AcOEt (4 × 30 ml). The organic layer was separated, washed with H_2O, dried over Na_2SO_4, concentrated to 5 ml, cooled over night and then the crystals collected. After drying in high vacuum 0.205 g (39%) colorless crystals of mp. 140°C were obtained. UV (pH 7): 205 (4.59), [228 (4.07)], 258 (3.76), 280 (3.76), 353 (4.18). pK_a 4.76. ^{1}H-NMR (DMSO–d_6): 12.56 (bs, 1H, H–N); 7.52–7.32 (m, 5H, arom. H); 6.63 (d, 1H, H–C(1′)); 5.51 (2d, 2H, CH_2); 4.64 (dd, 1H, H–C(2′)); 4.24 (pt, 1H, H–C(3′)); 3.77–3.45 (m, 3H, H–C(4′;5′;5″)); 2.28 (s, 3H, Me(6).

Anal. For $C_{19}H_{20}N_4O_7$ (416.4) Calcd.: C, 54.81; H, 4.84; N, 13.46. Found: C, 55.06; H, 4.93; N, 13.16.

2-Amino-4-benzyloxy-6-methyl-8-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)-7(8H)-pteridinone (11). In a solution of dioxane (6 ml), saturated with ammonia, **8** (0.2 g, 0.25 mmol) was stirred for 80 min. It was evaporated, the residue dissolved in AcOEt (30 ml) and wahed with H_2O. The organic phase was dried over Na_2SO_4, evaporated, the residue dissolved in MeOH, treated with charcoal, concentrated to 5 ml and then H_2O dropwise added till a colorless precipitate starts to separate. After standing in thev icebox over night, the crystals were collected, dried in high vacuum to give 0.142 g (78%) of mp. 103°C. UV (MeOH): 207 (4.69), 229 (4.70), 274 (3.87), 282 (3.91), 288 (3.83), 345 (4.16). ^{1}H-NMR (DMSO–d_6): 7.68–7.34 (m, 22H, arom. H, NH_2); 7.13 (bs, 1H, H–C(1′)); 6.36 (pt, 1H, H–C(3′)); 6.24 (bs, 1H, H–C(2′)); 5.48 (2d, 2H, CH_2); 4.73–4.51 (m, 3H, H–C(4′;5′;5″)); 2.28 (s, 3H, Me(6).

Anal. For $C_{40}H_{33}N_5O_9$ (727.7) Calcd.: C, 66.02; H, 4.57; N, 9.62. Found: C, 65.03; H, 4.62; N, 9.39.

2-Allylamino-4-benzyloxy-6-methyl-8-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)-7(8H)-pteridinone (12). A solution of **8** (0.2 g, 0.25 mmol) in THF (10 ml) was treated with allylamine (45 mg, 0.8 mmol) by stirring at r.t. for 30 min. It was evaporated, the residue dissolved in AcOEt (40 ml), washed with H_2O, the organic layer dried over Na_2SO_4 and again evaporated to an oil. It was dissolved in EtOH (15 ml), treated with charcoal, filtered and to the hot solution H_2O (15 ml) added. After cooling the crystals were collected, dried in high vacuum to give 0.108 g (56%) of mp. 161–162°C. UV (MeOH): 209 (4.66), 230 (4.71), [274 (3.82)], 282 (3.86), [293 (3.79)], 356 (4.21). ^{1}H-NMR (DMSO–d_6): 8.01–7.35 (m, 21H, arom. H, NH); 7.18 (bd, 1H, H–C(1′)); 6.41–6.24 (m, 2H, H–C(2′), H–C(3′)); 5.80 (m, 1H, CH_2═C*H*–); 5.50 (d, 2H, CH_2); 5.05 (m, 2H, C*H*$_2$═CH); 4.78–4.58 (m, 3H, H–C(4′;5′;5″)); 3.93 (bs, 2H, CH_2═CHC*H*$_2$); 2.28 (s, 3H, Me(6).

Anal. For $C_{43}H_{37}N_5O_9$ (767.8) Calcd.: C, 67.28; H, 4.86; N, 9.12. Found: C, 67.43; H, 4.86; N, 9.04.

4-Benzyloxy-2-n-butylamino-6-methyl-8-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)-7(8H)-pteridinone (13). Analogous to the preceeding procedure with **8** (0.2 g, 0.25 mmol) and n-butylamine (73 mg, 1 mmol) in 45 min. The same work-up gave 0.114 g (59%) of colorless crystals of mp. 164°C. UV (MeOH): 209 (4.68), 230 (4.71), [274 (3.82)], 282 (3.88), [292 (3.80)], 352 (4.21). ^{1}H-NMR (DMSO–d_6): 8.09–7.36 (m, 21H, arom. H, NH); 7.18 (bd, 1H, H–C(1′)); 6.41–6.24 (m, 2H, H–C(2′), H–C(3′));

5.50 (2d, 2H, CH_2); 4.74–4.52 (m, 3H, H–C(4′;5′;5″)); 3.83 (m, 2H, $HNCH_2$); 2.28 (s, 3H, Me(6); 1.42 (m, 2H, CH_2); 1.05 (Mm, 2H, CH_2); 0.81 (t, 3H, CH_3).

Anal. For $C_{44}H_{41}N_5O_9$ (783.8) Calcd.: C, 67.42; H, 5.23; N, 8.94. Found: C, 67.55; H, 5.27; N, 8.81.

4-Benzyloxy-6-methyl-2-(2-picolylamino)-8-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)-7(8H)-pteridinone (14). Analogous to the preceeding procedure with **8** (0.2 g, 0.25 mmol) in AcOEt (20 ml) and 2-picolylamine (0.1 g, 0.92 mmol) in 45 min. The same work-up gave 0.182 g (89%) of colorless crystals of mp. 144°C. UV (MeOH): 230 (4.72), [262 (4.02)], 282 (3.92), [290 (3.85)], 348 (4.23). ^{1}H-NMR (DMSO–d_6): 8.48 (d, 1H, α-pyridyl); 7.94–7.14 (m, 24H, arom. H, NH); 6.92 (bd, 1H, H–C(1′)); 6.35–6.19 (m, 2H, H–C(2′), H–C(3′)); 5.50 (s, 2H, OCH_2); 5.34 (s, 2H, N–CH_2); 4.70–4.51 (m, 3H, H–C(4′;5′;5″)); 2.23 (s, 3H, Me(6)).

Anal. For $C_{46}H_{28}N_6O_9$ (818.8) Calcd.: C, 67.47; H, 4.68; N, 10.26. Found: C, 67.70; H, 4.66; N, 10.29.

4-Benzyloxy-6-methyl-2-dimethylamino-8-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)-7(8H)-pteridinone (15). A solution of **8** (0.37 g, 0.47 mmol) in toluene (10 ml) was treated at r.t. with a saturated solution of dimethylamine in benzene (1 ml) for 30 min with stirring. It was dilute with toluene (20 ml), washed with H_2O (3 × 10 ml), the organic phasae dried over NA_2SO_4 and then evaporated. The residue was recrystallized from EtOH to give 0.238 g (66%) of colorless crystals of mp. 124°C. UV (MeOH): 212 (4.66), 229 (4.71), 282 (3.82), 295 (3.78), 358 (4.23). ^{1}H-NMR (DMSO–d_6): 8.10–7.79 (m, 6H, arom. H); 7.65–7.55 (m. 3H, arom. H); 7.48–7.30 (m, 11H, arom. H); 7.20 (bs, 1H, H–C(1′)); 6.28 (m, 2H, H–C(2′), H–C(3′)); 5.54 (q, 2H, OCH_2); 4.70–4.56 (m, 3H, H–C(4′;5′;5″)); 3.00 (s, 6H, Me_2N); 2.30 (s, 3H, Me(6)). Anal. For $C_{46}H_{38}N_5O_9$ (756.8) Calcd.: C, 66.67; H, 5.06; N, 9.26. Found: C, 66.92; H, 4.96; N, 9.14.

4-Benzyloxy-6-methyl-2-morpholino-8-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)-7(8H)-pteridinone (16). A solution of **8** (0.2 g, 0.25 mmol) in THF (10 ml) was treated with morpholine (90 mg, 1 mmol) and stirring for 45 min at r.t. It was dilute with AcOEt (40 ml), washed with H_2O (3 × 20 ml), the organic phasae dried over Na_2SO_4 and then evaporated. The residue was recrystallized from EtOH/H_2O to give 0.117 g (63%) of colorless crystals of mp. 167°C. UV (MeOH): 212 (4.70), 229 (4.71), 274 (3.82), 294 (3.81), 354 (4.24). ^{1}H-NMR (DMSO–d_6): 8.00–7.30 (m, 20H, arom. H); 7.19 (s, 1H, H–C(1′)); 6.27–6.15 (m, 2H, H–C(2′), H–C(3′)); 5.58–5.45 (2d, 2H, OCH_2); 4.77–4.52 (m, 3H, H–C(4′;5′;5″)); 3.54 (m, 4H, morpholino); 3.71 (m, 4H, morpholino); 2.30 (s, 3H, Me(6)).

Anal. For $C_{44}H_{39}N_5O_{10}$ (738.8) Calcd.: C, 66.24; H, 4.94; N, 8.78. Found: C, 66.40; H, 4.93; N, 8.43.

4-Benzyloxy-6-methyl-2-methylamino-8-β-D-ribofuranosyl-7(8H)-pteridinone (17). A solution of **8** (0.2 g (0.25 mmol) in MeOH (10 ml) was treated with methanolic methylamine (44%, 1 ml) and stirred at r.t. for 60 h. The resulting precipitate was collected, washed with MeOH and dried in high vacuum to give 48 mg (45%) of a colorless powder of mp. 225°C. UV (MeOH): 211 (4.54), 238 (4.20), [287 (3.76)], 293

(3.78), 352 (4.21). ^{1}H-NMR (DMSO–d_6): 7.46–7.30 (m, 5H, arom. H); 6.64 (bs, 1H, H–C(1′)); 5.51 (d, 1H, CH_2); 5.43 (d, 1H, CH_2); 4.61 (bs, 1H, H–C(2′)); 4.25 (bs, 1H, H–C(3′)); 3.72–3.39 (m, 3H, H–C(4′;5′;5″)); 2.83 (bs, 3H, Me–N); 2.23 (s, 3H, Me(6)).

Anal. For $C_{20}H_{23}N_5O_6$ (429.4) Calcd.: C, 55.94; H, 5.45; N, 16.48. Found: C, 55.94; H, 5.50; N, 16.43.

2,4-Dimethoxy-6-methyl-8-β-D-ribofuranosyl-7(8H)-pteridinone (18). Compound **8** (0.4 g, 5 mmol) was stirred in 0.2 N CH_3ONa solution (10 ml) at r.t. for 2 days. Then H_2O (20 ml) was added and then neutralized with Lewatit ion-exchange-resin (H^+ form). It was evaporated, the residue dissolved in little MeOH and put onto 2 preparative silica–gel plates for chromatography with AcOEt/MeOH 30:1 (2 developments). The main bands have been eluted with AcOEt (100 ml), evaporated and the residue dried in high vaccum to give 32 mg (18%) of a colorless crystal powder of mp. 193°C. UV (pH 7): 236 (3.98), [244 (3.94)], [276 (3.78)], 313 (4.13)], [321 (4.10)], [339 (3.87)]. ^{1}H-NMR (DMSO–d_6): 6.62 (d, 1H, H–C(1′)); 4.63 (dd, 1H, H–C(2′)); 4.26 (pt, 1H, H–C(3′)); 4.03 (s, 3H, MeO); 3.96 (s, 3H, MeO); 3.79–3.37 (m, 3H, H–C(4′;5′;5″)); 2.33 (s, 3H, Me(6)).

Anal. For $C_{18}H_{20}N_4O_7$ (354.3) Calcd.: C, 47.46; H, 5.12; N, 15.81. Found: C, 47.41; H, 5.21; N, 15.75.

6-Methyl-8-(2,3,5-tri-O-benzoyl-β-D-ribofuranosyl)-2(H),4(3H,7(8H)-pteridinetrione (19).[16] A solution of **9** (0.7.3 g, 1 mmol) in AcOEt (30 ml) was hydrogenated with H_2 in presence of Pd/C catalyst (0.25 g, 5%) in a shaking apparatus till the uptake of H_2 is finished after 2 days. The mixture was heated, filtered hot and then the filtrate evaporated to dryness. The residue was purified by CC (15 g, SiO_2) with 200 ml of each toluene/AcOEt 2:1, 1:1 followed by AcOEt/MeOH 25:1, 10:1 and 5:1. The product fraction was evaporated and the residue recrystallized from EtOH (121 ml) to give 0.372 g (58%) of mp.>186°C (decomp.). UV (pH 5): 230 (4.57), 285, (3.95), 353 (3.94); (MeOH): 229 (4.64), 283 (4.02), 290 (4.02), 356 (4.06). pK_a 2.14, 13.12. ^{1}H-NMR (DMSO–d_6): 11.17 (bs, 1H, H-N); 8.02–7.37 (m, 15H, arom. H); 6.75 (s, 1H, H–C(1′)); 6.25 (pt, 1H, H–C(3′)); 6.10 (bs, 1H, H–C(2′)); 4.70–4.52 (m, 3H, H–C(4′;5′;5″)); 2.22 (s, 3H, Me(6)).

Anal. For $C_{38}H_{26}N_4O_{10} \times H_2O$ (656.6) Calcd.: C, 60.37; H, 4.30; N, 8.53. Found: C, 60.34; H, 4.35; N, 8.26.

6-Methyl-8-β-D-ribofuranosyl-2(1H),4(3H),7(8H)-pteridinetrione (20).[16] A solution of **19** (0.24 g, 0.4 mmol) in saturated methanolic ammonia (20 ml) was stirred at r.t. in the dark for 6 days. It was evaporated to dryness, the residue treated with MeOH, the solid collected and dried in high vacuum to give 46 mg (32%) a colorless crystal powder of mp.> 150°C (decomp.). UV (pH 6): 209 (4.38), 291, (3.93), 342 (3.98). pK_a 3.42, 13.11. ^{1}H-NMR (DMSO–d_6): 6.75 (s, 1H, H–C(1′)); 4.62 (dd, 1H, H–C(2′)); 4.25 (pt, 1H, H–C(3′)); 3.76–3.41 (m, 3H, H–C(4′;5′;5″)); 2.13 (s, 3H, Me(6)).

Anal. For [$C_{12}H_{13}N_4O_7$] NH_4 (343.3) Calcd.: C, 41.98; H, 4.99. Found: C, 41.86; H, 4.76.

6-Amino-2-methylamino-5-nitroso-4(3)-pyrimidone (22). A suspension of 6-amino-2-methylthio-5-nitroso-4(3H)pyrimidone (**21**)[17] (8.0 g, 43 mmol) in 20%

aqueous methylamine (120 ml) was stirred at 5–10°C for 6 h. The violet suspension is converted into a dark-red solution. It was evaporated to dryness, the residue was suspended in H_2O (200 ml), slightly acidified by AcOH to pH 4 to get an orange colored precipitate. The solid was collected, washed with cold H_2O and acetone, to give after drying 6.9 g (95%) of orange powder. A sample (0.15 g) was recrystallized from H_2O (200 ml) with charcoal to give 0.104 g (68%) orange glittering crystals of mp.>350°C. UV (pH 12): 208 (4.01), [226 (3.57)], [264 (3.57)], 325 (4.35). ^{1}H-NMR (DMSO–d_6): 11.24 (bs, 1H, H–N(3)); 10.93 (bs, 1H, H–N(6)); 8.52 (bs, 1H, H–N(6)); 7.23 (bs, 1H, MeN–H); 2.85 (d, 3H, MeN).

Calc. For $C_5H_7N_5O_2$ (169.2): C, 35.50, H, 4.18, N, 41.42. Found: C, 35.52, H, 4.29, N, 41.81.

6-Amino-2-(2-hydroxyethyl)amino-5-nitroso-4(3H)-pyrimidone (23). A suspension of **21** (7.0 g, 38 mmol) in a mixture of ethanolamine (40 ml) and H_2O (50 ml) was stirred at r.t. for 24 h. The solution was cooled and then acidified by dropwise addition of AcOH (100ml). The resulting precipitate was collected, washed with H_2O, EtOH and ether to give after drying 6.1 g (81%). Purification was achieved by dissolving in 3 N NaOH (20 ml), charcoal treatment, filtration and dropwise addition of the filtrate into hot AcOH (35 ml) to form an orange-yellow solid 4.6 g (61%). A sample (0.15 g) was recrystallyzed from H_2O (200 ml) to give 0.1 g of orange crystals of mp.> 350°C. UV (pH 12): 207 (4.01), [227 (3.50)], [264 (3.53)], 326 (4.37). ^{1}H-NMR (DMSO–d_6):11.22 (bs, 1H, H–N(3)); 10.60 (bs, 1H, H–N(6)); 8.50 (bs, 1H, H–N(6)); 7.38 (bs, 1H, *H*–NCH_2CH_2); 4.94 (t, 2H, CH_2CH_2O*H*); 3.54–3.37 (m, 4H, CH_2CH_2).

Calc. For $C_6H_9N_5O_3$ (199.2): C, 36.18, H, 4.55, N, 35.17. Found: C, 36.11, H, 4.73, N, 34.88.

6-Ethoxycarbonylmethyl-2-methylamino-4(3H),7(8H)-pteridinedione (24). A 20% ammonium sulfide solution (32 ml) was heated to 70°C and then under stirring **22** (8.0 g, 47 mmol) added. The color of the solution turned yellow and a yellow precipitate separated out. It was heated 30 min at 50°C, then cooled in the icebox and the solid collected after a few hours. The yellow material was washed with cold H_2O and EtOH and dried in high vacuum to give 7.2 g (98%) of 5,6-diamino-2-methylamino-4(3H)-pyrimidone of mp.> 300°C (decomp.). A mixture of 5,6-diamino-2-methylamino-4(3H)-pyrimidone (7.0 g, 45 mmol) and sodium diethyl oxalylacetate (9.7 g, 46 mmol) was heated in AcOH (275 ml) under reflux for 45 min. A yellow precipitate separated from the hot solution. After cooling the solid was collected, washed with H_2O and EtOH and dried to give 10.5 g (83%) of chromatographically pure **24**. A sample (0.25 g) was recrystallized from H_2O with charcoal to give 0.166 g of colorless fine needles of mp.> 350°C. UV (MeOH): 215 (4.50), [234 (3.99)], 292 (4.06), 342 (4.21). ^{1}H-NMR (DMSO–d_6): 12.45 (bs, 1H, H–N(8)); 11.13 (bs, 1H, H–N(3)); 6.82 (bs, 1H, MeN–H); 4.07 (q, 2H, OCH_2CH_3); 3.61 (s, 2H, CH_2COOEt); 2.84 (d, 3H, N–CH_3); 1.16 (t, 3H, OCH_2CH_3).

Calc. For $C_{11}H_{13}N_5O_4$ (279.3): C, 47.31, H, 4.70, N, 25.09. Found: C, 46.94, H, 4.66, N, 24.71.

6-Methyl-2-methylamino-4(3H),7(8)-pteridinedione (25). A solution of **24** (5.1 g, 18 mmol) in 1 N NaOH (200 ml) was heated under reflux for 30 min, then treated with charcoal and the hot filtrate added dropwise into boiling AcOH (100 ml).

The resulting yellow precipitate was collected after cooling, washed with H_2O and dried at 100°C to give 3.5 g (92%) of **25**. Further purification was achieved by reprecipitation. The solid was dissolved in dilute NaOH (150 ml), again treatment with charcoal and the filtrate added nslowly into a boiling mixture of AcOH (50 ml) and H_2O (50 ml) yielding after drying 3.4 g (89%) of a colorless crystal powder of mp.> 350°C. UV (pH 12): 221 (4.59), [247 (4.19)], [279 (3.85)], 342 (4.20). ^{1}H-NMR (DMSO–d_6): 12.23 (bs, 1H, H–N(8)); 11.01 (bs, 1H, H–N(3)); 6.75 (bs, 1H, MeN–*H*); 2.82 (d, 3H, N–CH_3); 2.21 (s, 3H, CH_3–C(6)).

Calc. For $C_8H_9N_5O_2$ (207.2): C, 46.37, H, 4.39, N, 33.81. Found: C, 46.41, H, 4.32, N, 33.51.

6-Ethoxycarbonylmethyl-2-(2-hydroxyethyl)amino-4(3H),7(8H)-pteridinedione (26). To a ice-cold solution of **23** (3.4 g, 17 mmol) in 1 N NaOH (25 ml) was added hydrazine hydrate (0.9 g) and Ni–Al alloy (0.55 g). After a few min a vigorous reaction takes place and a color change from red to yellow is observed. It was stirred at r.t. for 1 h, the Raney-nickel filtered off and then the cooled filtrate neutralized with HCl (20%), EtOH (10 ml) added and stored in the icebox for 2 days. The yellowish precipitate was collected, washed with cold EtOH and ether and dried in a vacuum desiccator to give 3.1 g (98%) of 5,6-diamino-2-(2-hydroxyethyl)amino-4(3H)-pyrimidone of mp.> 200°C. This substance (5.4 g, 29 mmol) was heated with sodium diethyl oxalylacetate (6.3 g, 30 mmol) in conc. AcOH (50 ml) to 80°C for 1 h. The resulting yellow precipitate was collected after cooling, washed with H_2O and EtOH and dried at 100°C to give 5.5 g (61%) of mp.> 350°C. A small sample (0.16 g) was recrystallized from H_2O (45 ml) with charcoal to give sligthly yellowish fine needles. UV (MeOH): 216 (4.49), [234 (3.99)], 292 (4.06), 342 (4.22). ^{1}H-NMR (DMSO–d_6): 12.45 (bs, 1H, H–N(8)); 10.86 (bs, 1H, H–N(3)); 6.99 (bs, 1H, MeN–H); 4.93 (t, 1H, OH); 4.07 (q, 2H, OC*H*$_2$CH$_3$); 3.61 (s, 2H, C*H*$_2$COOEt); 3.55–3.38 (m, 4H, CH_2CH_2); 1.17 (t, 3H, OCH_2CH_3).

Calc. For $C_{12}H_{15}N_5O_5$ (309.3): C, 46.60, H, 4.90, N, 22.65. Found: C, 46.66, H, 4.95, N, 22.71.

6-Methyl-2-(2-hydroxyethyl)amino-4(3H),7(8)-pteridinedione (27). A solution of **26** (3.0 g, 9.7 mmol) in 1 N NaOH (30 ml) was heated under reflux for 30 min. After treatment with charcoal and filtration the hot filtrate was added dropwise with stirring into AcOH (30 ml) forming a yellow precipitate which was collected after cooling, washed with H_2O and EtOH, dried at 100°C to give 2.1 g (91%) of a yellow powder of mp.>350°C. A sample (0.285 g) was recrystallized from H_2O (400 ml) to give 0.176 g of yellowish glittering crystals. UV (pH 12): 222 (4.52), [249 (4.15)], [276 (3.87)], 340 (4.17). ^{1}H-NMR (DMSO–d_6): 12.26 (bs, 1H, H–N(8)); 10.78 (bs, 1H, H–N(3)); 6.91 (bs, 1H, *H*–NCH$_2$CH$_2$); 4.92 (bs, 1H, *H*O–CH$_2$); 3.55–3.35 (m, 4H, CH_2CH_2); 2.21 (d, 3H, CH_3–C(6).

Calc. For $C_9H_{11}N_5O_3 \times 0.5\ H_2O$ (246.2): C, 43.91, H, 4.91, N, 28.45. Found: C, 44.00, H, 4.81, N, 28.69.

6-Methyl-2-methylthio-8-(2-deoxy-3,5-di-O-p-toluoyl-β-D-ribofuranosyl)-4(3H),7(8H)-pteridinedione (28).[7] A suspension of **1** (0.2 g, 0.9 mmol) in dry acetonitrile (30 ml) was treated with DBU (0.4 g, 2.65 mmol) and stirred at r.t. till a

clear solution (30 min) was obtained. The 1-chloro-2-deoxy-3,5-di-O-p-toluoyl-α-D-ribofuranose (0.7 g, 1.8 mmol) was added and the mixture stirred for 30 h. It was neutralized with AcOH in CH_2Cl_2 and then evaporated to dryness. The dark-colored residue was purified by disssolving in CH_2Cl_2 and CC (SiO_2) with toluene/AcOEt 1:1 (150 ml), 1:2 (200 ml) and CH_2Cl_2/MeOH 40:1 (200 ml). The product fraction were evaporated to give 0.265 g (52%) of an α/β-anomeric mixture. Recrystallization from tolene gave 0.213 g (42%) of pure β-anomer as colorless crystals of mp. 196°C. Lit.[7] mp. 196–197°C.

6-Methyl-2-methylamino-8-(2-deoxy-3,5-di-O-p-toluoyl-β-D-ribofuranosyl)-O^4-[2-(4-nitrophenyl)ethyl]-7(8H)-pteridinone (31). Through a solution of **30**[7] (1.0 g, 1.32 mmol) in dry CH_2Cl_2 (90 ml) was bubbled a dry stream of methylamine for 1 h with stirring. It was then evaporated to dryness and coevaporated with CH_2Cl_2. Purification was done by CC with toluene/AcOEt 5:2. The product fraction was again evaporated to give a yellowish solid foam. Recrystallization from EtOH yielded 0.607 g (65%) colorless crystals of mp. 174°C. UV (MeOH): 202 (4.83), [214 (4.61)], 239 (4.72), [272 (4.28)], 349 (4.27). ^{1}H-NMR ($CDCl_3$): 8.20 (d, 2H, *o* to NO_2); 8.05–7.85 (m, 4H, arom. H); 7.50 (d, 2H, *m* to NO_2); 7.36 (m, 1H, H–C(1′)); 7.26–7.13 (2d, 4H, *o* to CH_3); 6.06 (m, 1H, H–C(3′)); 5.72 (bs, 1H, H–NMe); 4.98–4.62 (m, 3H, H–C(4′), H–C(5′, 5″)); 4.62–4.55 (m, 2H, OCH_2); 3.44–3.31 (m, 3H, OCH_2CH_2, H–C(2′)); 3.04 (d, 3H, HN–CH_3); 2.48 (bs, 4H, CH_3–C(6), H–C(2″)); 2.42 (s, 3H, tol–CH_3), 2.36 (s, 3H, tol–CH_3).

Calc. For $C_{37}H_{36}N_6O_9$ (708.7): C, 62.70, H, 5.12, N, 11.86. Found: C, 62.36, H, 5.12, N, 11.54.

6-Methyl-2-(2-hydroxyethyl)amino-8-(2-deoxy-3,5-di-O-p-toluoyl-β-D-ribofuranosyl)-O^4-[2-(4-nitrophenyl)ethyl]-7(8H)-pteridinone (32). To a solution of **30** (2.0 g, 2.64 mmol) in CH_2Cl_2 (60 ml) was added ethanolamine (4.85 g, 80 mmol) and stirred at r.t. for 6 h. It was neutralized with AcOH in CH_2Cl_2, evaporated and the residue purified by CC (SiO_2) with toluene/AcOEt 5:2. Evaporation of the product fraction and coevaporation of the residue with CH_2Cl_2 yielded 1.36 g (70%) of a yellowish solid foam. UV (MeOH): 203 (4.81), [215 (4.59)], 239 (4.70), [283 (4.23)], 348 (4.25). ^{1}H-NMR ($CDCl_3$): 8.16 (d, 2H, *o* to NO_2); 7.91 (d, 4H, arom. H); 7.47 (d, 2H, *m* to NO_2); 7.31 (m, 1H, H–C(1′)); 7.23–7.12 (2d, 4H, *o* to CH_3); 6.03 (m, 1H, H–C(3′)); 4.96 (bs, 1H, H–C(4′)); 4.72 (t, 2H, OCH_2); 4.59 (m, 2H, H–C(5′,5″)); 3.88 (m, 2H, HN–CH_2CH_2); 3.39 (m, 1H, H–C(2′)); 3.30 (t, 2H, OCH_2CH_2); 2.99 (bs, H, CH_2CH_2–*OH*); 2.48 (bs, 4H, CH_3–C(6), H–C(2″)); 2.42 (s, 3H, tol–CH_3), 2.37 (s, 3H, tol–CH_3).

Calc. For $C_{38}H_{38}N_6O_{10}$ (738.8): C, 61.78, H, 5.18, N, 11.38. Found: C, 61.70, H, 5.26, N, 11.33.

6-Methyl-2-methylamino-8-(2-deoxy-β-D-ribofuranosyl-O^4-[2-(4-nitrophenyl)ethyl]-7(8H)-pteridinone (33). A solution of **31** (0.2 g, 0.28 mmol) in MeOH (6 ml) and CH_2Cl_2 (4 ml) was treated with NaCN (65 mg) and stirring at r.t. for 24 h. It was evaporated and then purified by CC with mixtures of CH_2Cl_2/MeOH 100:1 (85 ml), 50:1 (60 ml) and 10:1 (170 ml). The product fraction was evaporated and the resulting foam recrystallized from toluene to give 82 mg (86%) yellowish crystals of mp.> 300°C. UV (MeOH): 210 (4.57), 239 (4.29), 279 (4.19), 350 (4.24). ^{1}H-NMR

(DMSO–d_6): 8.19 (d, 2H, *o* to NO_2); 7.64 (d, 3H, *m* to NO_2, H–NMe); 7.19 (m, 1H, H–C(1′)); 5.17 (m, 1H, HO); 4.83–4.51 (m, 3H, O–CH_2, HO–C(3′)); 4.45 (m, 1H, H–C(3′)); 3.88–3.61 (m, 2H, H–C(5′, 5″)); 3.55 (m, 1H, H–C(4′)); 3.27 (m, 2H, OCH_2CH_2); 2.87 (m, 4H, HN–CH_3, H–C(2′)); 2.27 (bs, 3H, CH_3–C(6)); 2.01 (m, 1H, H.C(2″)).

Calc. For $C_{21}H_{24}N_6O_9$ (472.5): C, 53.39, H, 5.12, N, 17.79. Found: C, 53.00, H, 5.17, N, 17.35.

6-Methyl-2-methylamino-8-(2-deoxy-3,5-di-O-p-toluoyl-β-D-ribofuranosyl)-4(3),7(8H)-pteridinedione (34). In a mixture of dioxane (4 ml) and acetonitrile (5 ml) 31 (0.2 g, 0.28 mmol) was dissolved and then treated with DBU (0.685 g, 4.5 mmol) with stirring for 5 h. It was neutralized with AcOH in CH_2Cl_2, evaporated and the residue put into a silica–gel column for chromatography with CH_2Cl_2 (80 ml) and CH_2Cl_2/MeOH 40:1 (240 ml). The product fraction was evaporated, dried in high vacuum to give 0.147 g (93%) of a yellowish solid of mp. 232–233°C (decomp.). UV (MeOH): 202 (4.68), 217 (4.54), 237 (4.59), 296 (4.07), 347 (4.12). ^{1}H-NMR ($CDCl_3$): 11.69 (bs, 1H, H–N(3), 7.95 (m, 4H, arom. H); 7.46 (m, 2H, H–C(1′), H–NMe); 7.21 (2d, 4H, arom. H); 6.00 (m, H–C(3′)); 4.95–4.41 (m, 3H, H–C(4′), H–C(5′, 5″)); 3.34 (m, 1H, H–C(2′)); 3.07 (d, 3H, HN–CH_3); 2.46 (s, 4H, CH_3–C(6), H–C(2″)); 2.42 (s, 3H, CH_3–tol); 2.37 (s, 3H, CH_3–tol).

Calc. For $C_{28}H_{29}N_5O_7 \times 2\ H_2O$ (571.5): C, 56.74, H, 5.82, N, 12.25. Found: C, 56.73, H, 5.42, N, 12.05.

6-Methyl-2-(2-hydroxyethyl)amino-8-(2-deoxy-3,5-di-O-p-toluoyl-β-D-ribofuranosyl)-4(3),7(8H)-pteridinedione (35). Analogous to the preceding procedure with **32** (0.411 g, 0.56 mmol) in dioxane (5 ml) and acetonitrile (15 ml) with DBU (1.52 g, 10 mmol). Purification by CC with CH_2Cl_2 (200 ml), CH_2Cl_2/MeOH 19:1 (200 ml) and CH_2Cl_2/MeOH 9:1 (200 ml) to give after evaporation and recrystallization from MeOH 0.233 g (71%) of yellowish crystals of mp. 207–208°C. UV (MeOH): 202 (4.77), 222 (4.61), 237 (4.70), 297 (4.14), 345 (4.21). ^{1}H-NMR ($CDCl_3$): 11.02 (bs, 1H, H–N(3), 7.90 (m, 4H, arom. H); 7.35 (m, 5H, arom. H, H–C(1′)); 7.05 (bs, 1H, *H*–NCH_2CH_2); 5.90 (m, H–C(3′)); 4.95 (bs, 1H, CH_2CH_2–O*H*); 4.67–4.45 (m, 3H, H–C(4′), H–C(5′, 5″)); 3.58–3.35 (m, 4H, CH_2CH_2); 3.15 (m, 1H, H–C(2′)); 3.07 (d, 3H, HN–CH_3); 2.38 (s, 4H, CH_3–C(6), H–C(2″)); 232 (s, 3H, CH_3–tol); 2.27 (s, 3H, CH_3–tol).

Calc. For $C_{30}H_{31}N_5O_8$ (589.6): C, 61.11, H, 5.30, N, 11.88. Found: C, 61.11, H, 5.42, N, 11.67.

6-Methyl-2-{2-[2-(4-nitrophenyl)ethoxycarbonyloxy]ethyl}amino-8-(2-deoxy-3,5-di-O-p-toluoyl-β-D-ribofuranosyl)-O^4-[2-(4-nitrophenyl)ethyl]-7(8H)-pteridinone (36). Compound **32** (0.25 g, 0.34 mmol) was coevaporated with dry pyridine (3 × 10 ml) and then dissolved in dry pyridine (4 ml). Under exclusion of moisture 2-(4-nitrophenyl)ethoxy-carbonyl chloride (85 mg, 0.37 mmol) was added and then stirred at 40°C for 30 h. It was evaporated, coevaporated with toluene, the residue dissolved in little CH_2Cl_2 and put on a silica–gel column for chromatography with toluene/AcOEt 7:2. The product fraction was evaporated and coevaporated several times with CH_2Cl_2 to give after drying in high vacuum 0.28 g (88%) of a colorless

solid foam. UV (MeOH): 202 (4.88), 239 (4.73), [270 (4.4)], 346 (4.26). ^{1}H-NMR ($CDCl_3$): 8.16 (m, 4H, *o* to NO_2); 7.92 (d, 4H, arom. H); 7.48 (d, 2H, *m* to NO_2); 7.37 (d, 2H, *m* to NO_2); 7.20 (m, 5H, H–C(1′), *o* to CH_3); 6.01 (m, 2H, H–N, H–C(3′)); 4.94 (bs, 1H, H–C(4′)); 4.74 (t, 2H, OCH_2); 4.58 (m, 2H, H–C(5′,5″)); 4.35 (m. 4H, $OCOOCH_2CH_2$, $HNCH_2CH_2OCO$); 3.76 (m, 2H, $OCOOCH_2CH_2$); 3.38 (m, 1H, H–C(2′)); 3.30 (t, 2H, OCH_2CH_2); 3.06 (bs, H, $HNCH_2CH_2OCO$); 2.50 (bs, 4H, CH_3–C(6), H–C(2″)); 2.44 (s, 3H, tol–CH_3), 2.38 (s, 3H, tol–CH_3).

Calc. For $C_{47}H_{45}N_7O_{14}$ (931.9): C, 60.58, H, 4.87, N, 10.52. Found: C, 60.78, H, 5.01, N, 10.19.

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