

80. Synthesis of Biopterin from Neopterin? The Formation of Pyrrolo[1,2-*f*]pteridins upon Side-Chain Activation of Neopterin

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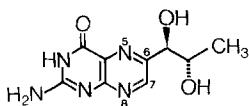
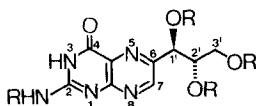
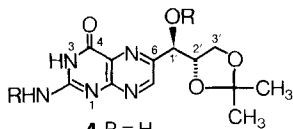
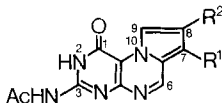
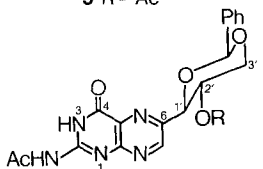
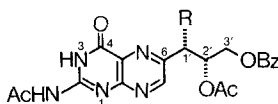
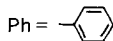
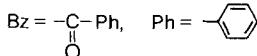
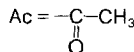
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The formation of derivatives of L-neopterin (**2**) is investigated with respect to a possible transformation into L-biopterin (**1**). High-yield syntheses of isopropylidene and benzylidene derivatives **5** and **7**, respectively, are described. Activation of the primary C-atom in the side chain of neopterin leads to formation of pyrrolo[1,2-*f*]pteridins *via* oxonium-ion intermediates.

Introduction. – (6*R*)-5,6,7,8-Tetrahydro-L-biopterin has been recognized to be the cofactor for phenylalanine, tyrosine, and tryptophan hydroxylases [1] and thus influences the biosynthesis of neurotransmitting catecholamines. Decrease of this tetrahydrobiopterin causes neurological disorders like atypical phenylketonuria [2] and *Parkinson's* disease [3], but it may also be clinically relevant in the treatment of these diseases [4] [5]. Its precursor L-biopterin (**1**) is synthetically available, *e.g.* from 2,5,6-triamino-4-hydroxypyrimidine and 5-deoxy-L-arabinose [6] [7]; the latter has to be prepared in several steps from L-rhamnose [8] or D-glucose [9]. While the biopterin synthesis furnishes only fair yields, the analogous preparation of L-neopterin (**2**) (starting with 2,5,6-triamino-4-hydroxypyrimidine and commercially available L-arabinose) proceeds in a much cleaner way and gives high yields [10] [11]. Thus, reduction of the terminal OH group in the side chain of L-neopterin (**2**) would constitute an interesting synthesis of L-biopterin (**1**). Some results on the selective modification of L-neopterin are discussed in the following.

Results. – Since acylation (tosylation, acetylation, pivaloylation) of the primary OH function of L-neopterin (**2**) or transesterification [12] of the peracetate **3** [13] were not selective enough, we turned our attention to the formation of cyclic acetals of **2**. Reaction of L-neopterin with acetone/conc. H₂SO₄ 50:1 or 2,2-dimethoxypropane/*p*-toluenesulfonic acid (TsOH) afforded 2',3'-*O*-isopropylideneneopterin (**4**) in 86 or 93% yield, respectively. This excellent regioselectivity is expected for an erythrose system [14]. The synthesis of the *N*²,*N*²-dimethyl analogue of **4** has been communicated recently in a review on the side-chain chemistry of pteridines [15].

O-Isopropylidene derivatives in terminal positions of carbohydrates are known to be converted to primary bromides by action of HBr/AcOH [16] [17]. Treatment of **4** with HBr/AcOH did not proceed satisfactorily, presumably due to the poor solubility of this compound. This problem was overcome by the use of diacetate **5**, readily obtained by standard acetylation of crude **4**. Bromination with HBr/AcOH led to *N*-(7-bromo-1,2-dihydro-1-oxopyrrolo[1,2-*f*]pteridin-3-yl)acetamide (**6**) as the major reaction product (judged by TLC), isolated in 22% yield.

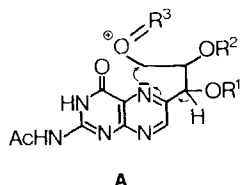
**1****2** R = H**3** R = Ac**4** R = H**5** R = Ac**6** R¹ = Br, R² = H**10** R¹ = OBz, R² = OAc**12** R¹ = OAc, R² = H**7** R = Ac**8** R = CH(OCH₃)Ph**9** R = Br**11** R = H

Next, the formation of an *O*-benzylidene derivative of L-neopterin (**2**) was investigated. Standard reaction conditions (α, α -dimethoxytoluene, TsOH, DMF [18]) gave an unseparable mixture of numerous benzylidene derivatives (¹H-NMR). Omission of solvent increased the regioselectivity of the reaction drastically to yield a dioxane-type acetal in a heterogeneous reaction. This product was best isolated after acetylation and chromatography as the diacetyl derivative **7** in excellent yield (90%). The presence of a six-membered-ring benzylidene acetal is evidenced by the coupling of the diaxial H–C(1') and H–C(2') ($J(1',2') = 9.6$ Hz) and the characteristic downfield shift of H–C(2') after acetylation. Compound **8** was isolated as a minor by-product (the formation of mixed acetals is known in acetal-exchange reactions [19]). *N*-Bromosuccinimide (NBS) opening [20] of the benzylidene derivative **7** in CCl₄ in the presence of a radical initiator (benzoyl peroxide) furnished the 1'-bromo compound **9** as main product (80%) along with small amounts (6%) of pyrrolo[1,2-*f*]pteridin **10**. Brominating-benzylidene-ring opening at secondary C-atoms (here C(1') of **7**) is known [21] to occur with inversion of configuration, and the configuration of the L-monapterin derivative **9** is deduced from its positive optical rotation (*cf.* [7] for an analogous 1'-chloro compound). The 1'-position of the Br-atom in **9** was corroborated by hydrogenation to the 1'-deoxy derivative **11**. The selectivity of this reaction was best using Pd/C in dioxane, whereas employment of Pt₂O furnished complex reaction mixtures.

Another pyrrolo-pteridin, namely **12**, was obtained as by-product in the acetylation of L-neopterin (**2**) with AcOH/conc. H₂SO₄ or with Ac₂O/AcOH according to [13]. The non-acetylated form of **12** has already been isolated after treatment of neopterin with polyphosphoric acid [22].

All three pyrrolo-pteridins **6**, **10**, and **12** exhibit similar UV spectra. Characteristically, H–C(9) appears as a broad signal in the ¹H-NMR spectra due to the proximity of the amide C=O group. Consistently, the IR absorption of this C=O is at lower energy compared to the neopterin derivatives. For the vicinal ¹H-NMR coupling constants in the pyrrole ring, $J(8,9) = 2.8$ – 2.9 Hz is found, as opposed to an allylic coupling constant of $J(7,9) = 1.5$ Hz in similar systems [23], thus supporting the structures assigned.

Discussion. – It is seen from the isolation of bromides **6** and **9** as main products that C(1') is the preferred site of attack in the brominations of isopropylidene-neopterin **5** and benzylidene-neopterin **7**, respectively. Only of minor importance seems to be an attack at the primary C-atom C(3'). Both, the reaction of isopropylidene compounds in HBr/AcOH and the NBS opening of benzylidene acetals are postulated to proceed *via* intermediate oxonium ions [16] [24]. This activation of the terminal C-atom C(3') of the neopterin side chain obviously occurs, however, it initiates the pyrrole-ring formation as depicted in **A**.



The formation of oxonium ions is also likely in the acid-catalyzed acetylation of neopterin (**2** → **3** + **12**), and this initiation of ring formation would be in keeping with the previously described [22] preparation of a pyrrolo-pteridin in the presence of polyphosphoric acid. The aromatic pyrrole ring would then result from elimination of a substituent at C(8) or C(9). Preservation of AcO–C(8) in **10** may be rationalized by another bromination at C(9) and subsequent elimination.

These results and considerations demonstrate that no high-yield synthesis of biopterin from neopterin can be expected due to pyrrolo-pteridin formation upon activation of the side-chain terminus of neopterin.

The skillful technical assistance of Mr. R. Keller is gratefully acknowledged. We also wish to thank our colleagues from the Central Research Department for determination of physical and analytical data: Dr. W. Arnold (NMR), Dr. L. Chopard (IR, UV), Dr. A. Dirscherl (MA), Dr. M. Grosjean (IR, UV), Mr. W. Meister (MS), and Dr. W. Vetter (MS).

Experimental Part

General. Solvents and reagents were obtained from Fluka (puriss. p.a.). Evaporation: Büchi-rotary evaporator, at 30–40°/in vacuo. TLC: precoated silica gel 60F-254 plates (Merck), detection by UV light (254 and 350 nm) and spraying with a 10% soln. of conc. H₂SO₄ in MeOH followed by heating. MPLC (medium pressure liquid chromatography): Lobar columns, LiChroprep Si 60 (40–63 µm, Merck) at 2–5 bar (Labomatic MD 80/100 pump). M.p. (uncorrected): Büchi-510 apparatus. [α]_D: Perkin Elmer 241 polarimeter, 1 dm cell. UV: Uvikon 810. IR (cm⁻¹): Nicolet-7199-FT-IR spectrophotometer. ¹H-NMR: Bruker AS-250 (250 MHz), Bruker HX-270 (270 MHz), or Bruker WM 400 (400 MHz), chemical shifts in ppm relative to tetramethylsilane as internal standard, coupling constants *J* in Hz.

2',3'-O-Isopropylidene-L-neopterin (= 2-Amino-6-(1'-hydroxy-2',3'-isopropylidenedioxypropyl)pteridin-4-(3H)-one; **4**). To a stirred suspension of **2** (1.0 g, 3.95 mmol) [10] [11] in 2,2-dimethoxypropane (200 ml) was added TsOH (750 mg, 3.95 mmol). After stirring for 24 h at r.t., the mixture was evaporated to ½ the volume. Pyridine (2 ml) was added, and the solvents were evaporated. The residue was suspended in hot H₂O (50 ml), cooled, and filtered. The filter cake was suspended in MeOH (50 ml) and filtered when still warm to give **4** (1.174 g, 93%) as colourless crystals, m.p. > 250°. ¹H-NMR (270 MHz, (D₆)DMSO): 11.59 (br., NH(3)); 8.72 (s, H–C(7)); 6.98 (br., NH₂–C(2)); 5.89 (d, *J* = 4.5, OH–C(1')); 4.60 (dd, *J*(1',2') = 5.5, H–C(1')); 4.41 (ddd ≈ dt, H–C(2')); 4.04 (dd, *J*(2',3a') = 6.3, *J*(3a',3b') = 8.3, H_B–C(3')); 3.93 (dd, *J*(2',3b') = 5.7, H_B–C(3')); 1.24 (s, CH₃); 1.21 (s, CH₃). EI-MS: 278 (8, *M*⁺–CH₃), 193 (100, *M*⁺–C₅H₈O₂), 101 (38, C₅H₉O₂⁺). Anal. calc. for C₁₂H₁₅N₅O₄ · 0.6 H₂O (304.09): C 47.40, H 5.36, N 23.06; found: C 47.54, H 5.32, N 22.78.

2-N,1'-O-Diacetyl-2',3'-O-isopropylidene-L-neopterin (= 1'-(2-Acetamido-3,4-dihydro-4-oxopteridin-6-yl)-2',3'-(isopropylidenedioxy)propyl Acetate; **5**). After evaporation of the solvents, the residue **4** (see above; from 1 g

of **2**) was suspended in pyridine (100 ml) and Ac_2O (50 ml). The clear soln. obtained after stirring for 22 h was poured into ice/ H_2O , extracted with CH_2Cl_2 , the extract dried (Na_2SO_4), and evaporated. MPLC (acetone/hexane 2:1) gave 1.46 g (98%) of crystalline **5**, m.p. 201–204°. $^1\text{H-NMR}$ (250 MHz, $(\text{D}_6)\text{DMSO}$): 12.31 (br., NH(3)); 11.9 (br., NH-C(2)); 8.96 (s, H-C(7)); 5.77 (d, $J(1',2') = 6.1$, H-C(1')); 4.60 (ddd $\approx$ dt, H-C(2')); 4.12 (dd, $J(2',3'a) = 6.5$, $J(3'a,3'b) = 8.9$, H_a -C(3')); 4.01 (dd, $J(2',3'b) = 5.0$, H_b -C(3')); 2.22 (s, NAc); 2.10 (s, OAc); 1.23 (s, 2CH_3). EI-MS: 362 (10, $M^{++} - \text{CH}_3$), 277 (22, $M^{++} - \text{C}_5\text{H}_8\text{O}_2$), 235 (62, $M^{++} - \text{C}_5\text{H}_8\text{O}_2 - \text{C}_2\text{H}_2\text{O}$), 101 (23, $\text{C}_5\text{H}_8\text{O}_2^+$), 43 (100, $\text{C}_2\text{H}_3\text{O}$). Anal. calc. for $\text{C}_{16}\text{H}_{19}\text{N}_5\text{O}_6$ (377.357): C 50.93, H 5.08; found: C 50.95, H 5.18.

N-(7-Bromo-1,2-dihydro-1-oxopyrrolo[1,2-f]pteridin-3-yl)acetamide (**6**). To a freshly prepared sat. soln. of dry HBr in glacial AcOH (2 ml), **5** (400 mg, 1.06 mmol) was added stirring rigorously. After 20 min at 0°, the soln. was diluted with CH_2Cl_2 (20 ml) and poured onto ice/ NaHCO_3 soln. The org. soln. was washed twice with H_2O , dried (Na_2SO_4), and evaporated. Treatment of the residue (327 mg) with hot acetone (10 ml) gave crude crystals (140 mg) that were recrystallized from MeON to yield dark-yellow crystals of pure **6** (75 mg, 22%), m.p. > 270°. UV (EtOH): 392 (3070), 348 (5330), 333 (5820), 295 (29560), 283 (31130), 244 (11300), 202 (17120). $^1\text{H-NMR}$ (270 MHz, $(\text{D}_6)\text{DMSO}$): 12.4 (br., NH(2)); 11.97 (s, NH-C(3)); 8.98 (s, H-C(6)); 8.88 (d, $J = 2.8$, H-C(9)); 7.18 (d, H-C(8)); 2.21 (s, NAc). EI-MS: 323/321 (45/50, M^{++}), 281/279 (96/100, $M^{++} - \text{C}_2\text{H}_2\text{O}$), 200 (22, $M^{++} - \text{C}_2\text{H}_2\text{O} - \text{Br}$), 43 (86, $\text{C}_2\text{H}_3\text{O}$).

2-N,2'-O-Diacetyl-1',3'-O-benzylidene-L-neopterin (= [4-(2-Acetamido-3,4-dihydro-4-oxopteridin-6-yl)-2-phenyl-1,3-dioxan-5-yl] Acetate; **7**) and 2-N-Acetyl-1',3'-O-benzylidene-2'-O-(α -methoxybenzyl)-L-neopterin (= N-{3,4-Dihydro-6-[5-(α -methoxybenzyloxy)-2-phenyl-1,3-dioxan-4-yl]-4-oxopteridin-2-yl}acetamide; **8**). To a suspension of **2** (1.0 g, 3.95 mmol) [10][11] in α,α -dimethoxytoluene (35 ml) was added TsOH (750 mg, 3.95 mmol). After stirring for 48 h at r.t., Et_3N (10 ml) was added to the slurry, which was then evaporated at 60°. The residue was stirred with pyridine (50 ml) and Ac_2O (25 ml) for 4.5 h at r.t. The resulting soln. was poured into ice/ NaHCO_3 soln. and extracted with CH_2Cl_2 . Drying (Na_2SO_4), evaporation, and MPLC (acetone/hexane 1:1, 0.1% Et_3N) gave **8** (150 mg, 7.5%) followed by **7** (1.512 g, 90%). **7**: M.p. 250–251° (dec.; after precipitation from acetone), $[\alpha]_D^{20} = -26.7^\circ$ ($c = 0.09$, dioxane). $^1\text{H-NMR}$ (400 MHz, $\text{CDCl}_3/(\text{D}_6)\text{DMSO}$ 3:1): 12.52 (s, NH(3)); 12.03 (s, NH-C(2)); 9.07 (s, H-C(7)); 7.56–7.53 (m, 2 arom. H); 7.40–7.38 (m, 3 arom. H); 5.78 (s, PhCH); 5.22 (ddd $\approx$ dt, H-C(2')); 5.15 (d, $J(1',2') = 9.6$, H-C(1')); 4.45 (dd, $J(2',3'a) = 5.2$, $J(3'a,3'b) = 10.0$, H_a -C(3')); 2.28 (s, NAc); 1.99 (s, OAc). EI-MS: 365 (1, $M^{++} - \text{C}_2\text{H}_4\text{O}_2$), 259 (54, 365 – $\text{C}_7\text{H}_6\text{O}$), 217 (30, 259 – $\text{C}_2\text{H}_2\text{O}$), 234 (50, $M^{++} - \text{C}_{11}\text{H}_{11}\text{O}_3$), 192 (34, 234 – $\text{C}_2\text{H}_2\text{O}$), 107 (24, $\text{C}_7\text{H}_7\text{O}$), 105 (22, $\text{C}_7\text{H}_5\text{O}$), 43 (100, $\text{C}_2\text{H}_3\text{O}$). Anal. calc. for $\text{C}_{20}\text{H}_{19}\text{N}_5\text{O}_6$ (425.40): C 56.47, H 4.50, N 16.46; found: C 57.12, H 4.64, N 16.66.

8: $^1\text{H-NMR}$ (400 MHz, $\text{CDCl}_3/(\text{D}_6)\text{DMSO}$ 3:1): 12.50 (br., NH(3)); 11.97 (br., NH-C(2)); 8.95 (s, H-C(7)); 7.57–7.30 (m, 10 arom. H); 5.72 (s, PhCH); 5.39 (s, PhC(OCH₃)H); 5.09 (d, $J(1',2') = 9.2$, H-C(1')); 4.49 (dd, $J(2',3'a) = 5.2$, $J(3'a,3'b) = 10.8$, H_a -C(3')); 4.38 (ddd, H-C(2')); 3.93 (dd $\approx$ t, $J(2',3'b) = 10.0$, H_b -C(3')); 3.14 (s, CH_3O); 2.29 (s, NAc).

2-N-Acetyl-6-[(1',2',3'-2'-acetoxy-3'-(benzoyloxy)-1'-bromopropyl]pterin (= [3-(2-Acetamido-3,4-dihydro-4-oxopteridin-6-yl)-2-acetoxy-3-bromopropyl] Benzoate; **9**) and [3-Acetamido-8-acetoxy-1,2-dihydro-1-oxopyrrolo[1,2-f]pteridin-7-yl] Benzoate (**10**). To a warm soln. of **7** (500 mg, 1.175 mmol) in CCl_4 (200 ml; dried over activated (300°) molecular sieves, 4 Å) containing small amounts of BaCO_3 and benzoyl peroxide was added NBS (314 mg, 1.77 mmol). After refluxing for 4.5 h under Ar, the mixture was cooled, poured onto ice/ H_2O and extracted with CH_2Cl_2 . Drying (Na_2SO_4) and evaporation followed by MPLC (acetone/hexane 1:1; sample applied in CH_2Cl_2 soln.) yielded **9** (477 mg, 80%) followed by **10** (30 mg, 6%). **9**: Colourless amorphous solid, $[\alpha]_D^{20} = +28.7^\circ$ ($c = 0.3$, dioxane). UV (EtOH): 337 (8750), 289 (15860), 229 (23500), 201 (26930). $^1\text{H-NMR}$ (270 MHz, CDCl_3): 12.85 (br., NH(3)); 11.10 (br., NH-C(2)); 9.14 (s, H-C(7)); 7.96 (m, 2 arom. H); 7.58 (m, 1 arom. H); 7.44 (m, 2 arom. H); 5.87 (ddd $\approx$ q, H-C(2')); 5.55 (d, $J(1',2') = 6.0$, H-C(1')); 4.59 (dd, $J(2,3'a) = 4.7$, $J(3'a,3'b) = 12.0$, H_a -C(3')); 4.48 (dd, $J(2',3'b) = 5.7$, H_b -C(3')); 2.18 (s, 2CH_3). EI-MS: 424 (0.4, $M^{++} - \text{Br}$), 364 (1, 424 – $\text{C}_2\text{H}_4\text{O}_2$), 322/324 (1/1, $M^{++} - \text{C}_6\text{H}_5 - \text{COOH} - \text{C}_2\text{H}_3\text{O}_2$), 259 (36, 322/324 – Br), 217 (50, 259 – $\text{C}_2\text{H}_2\text{O}$), 122 (44, $\text{C}_6\text{H}_5\text{COOH}$), 105 (100, 122 – OH), 77 (62, 105 – CO), 43 (73, $\text{C}_2\text{H}_3\text{O}$). Anal. calc. for $\text{C}_{20}\text{H}_{18}\text{BrN}_5\text{O}_6 \cdot 0.4\text{C}_6\text{H}_4$ (538.77): C 49.29, H 4.42, Br 14.83, N 13.00; found: C 49.50, H 4.45, Br 14.21, N 13.05.

10: Yellow crystals, m.p. > 260°. $^1\text{H-NMR}$ (270 MHz, $(\text{D}_6)\text{DMSO}$): 12.47 (br., NH(2)); 11.92 (br., NH-C(3)); 9.14 (br., H-C(9)); 9.08 (s, H-C(6)); 8.15 (m, 2 arom. H); 7.79 (m, 1 arom. H); 7.64 (m, 2 arom. H); 2.40 (s, OAc); 2.21 (s, NAc). EI-MS: 421 (1, M^{++}), 379 (16, $M^{++} - \text{C}_2\text{H}_2\text{O}$), 105 (100, $\text{C}_6\text{H}_5\text{CO}^+$).

2-N-Acetyl-6-[(2'R)-2'-acetoxy-3'-(benzoyloxy)propyl]pterin (= [3-(2-Acetamido-3,4-dihydro-4-oxopteridin-6-yl)-2-acetoxypropyl] Benzoate; **11**). To a pre-hydrogenated suspension of 5% Pd/C (40 mg) in dioxane (2 ml) was added a soln. of **9** (100 mg, 0.198 mmol) in dioxane (3 ml). After hydrogenation for 5 h at r.t., the mixture was filtered over a pad of Speedex, evaporated, and chromatographed (MPLC, acetone/hexane 3:2). The main fraction was precipitated from acetone/ Et_2O to yield **11** (20 mg, 24%) as amorphous solid. $^1\text{H-NMR}$ (270 MHz, CDCl_3): 12.85 (br., NH(3)); 11.08 (br., NH-C(2)); 8.86 (s, H-C(7)); 8.03 (m, 2 arom. H); 7.59 (m, 1 arom. H); 7.46 (m, 2

arom. H); 5.66 (*ddt*, H–C(2'')); 4.69 (*dd*, $J(2',3a') = 5.2$, $J(3a',3b') = 12.0$, H_a–C(3'')); 4.64 (*dd*, $J(2',3b') = 3.4$, H_b–C(3'')); 3.44 (*d*, $J(1',2') = 6.3$, 2 H–C(1'')); 2.52 (*s*, NAc); 2.03 (*s*, OAc). EI-MS: 365 (4, $M^{++} - C_2H_4O_2$), 260 (22, 365 – C₆H₅CO⁺), 244 (50, 365 – C₆H₅COO⁺), 218 (18, 260 – C₂H₂O), 202 (18, 244 – C₂H₂O), 122 (12, C₆H₅COOH), 105 (100, C₆H₅CO⁺), 77 (38, C₆H₅), 60 (6, C₂H₄O₂), 43 (40, C₂H₃O).

2-N-Acetyl-1',2',3'-tri-O-acetyl-L-neopterin (= 1-(2-Acetamido-3,4-dihydro-4-oxopteridin-6-yl)propan-1,2,3-triyl Triacetate; **3**) and [3-Acetamido-1,2-dihydro-1-oxopyrrolo[1,2-f]pteridin-7-yl] Acetate (**12**). To a suspension of **2** (200 mg, 0.79 mmol) [10] [11] in Ac₂O (10 ml) was added conc. H₂SO₄ (0.4 ml) dropwise. After stirring for 16 h at r.t., a yellow soln. was obtained. Upon addition of Et₃N (1 ml), the mixture was poured into ice/H₂O and extracted with CH₂Cl₂. The combined org. soln. was dried (MgSO₄), evaporated, and coevaporated with toluene repeatedly. The resulting yellow foam was crystallized from MeOH to give **12** (27 mg, 11%). The mother liquor was evaporated *in vacuo* and purified by MPLC (acetone/hexane 3:2) yielding **3** (254 mg, 85%). **3**: Foam, $[\alpha]_D^{20} = -77.5^\circ$ ($c = 0.2$, CHCl₃) ([13]: $[\alpha]_D^{23} = -76.5^\circ$ ($c = 2.7$, CHCl₃)). UV (EtOH): 331 (7770), 283 (16200), 201 (14960). IR (KBr): 3427w, 3189m, 3024w, 2985w, 1748s, 1694s, 1626s, 1565s, 1496m, 1456s, 1970s, 1224s, 1128w, 1048m, 954w, 827w, 784w, 733w, 603w. ¹H-NMR (270 MHz, (D₆)DMSO): 12.33 (br., NH(3)); 12.02 (br., NH–C(2)); 8.99 (*s*, H–C(7)); 6.03 (*d*, $J(1',2') = 5.7$, H–C(1'')); 5.54 (*ddd*, H–C(2'')); 4.31 (*dd*, $J(2',3a') = 4.5$, $J(3'a,3'b) = 12.0$, H_a–C(3'')); 4.27 (*dd*, $J(2',3b') = 6.4$, H_b–C(3'')); 2.22 (*s*, NAc); 2.16 (*s*, OAc); 1.96 (*s*, OAc); 1.95 (*s*, OAc). EI-MS: 379 (3, $M^{++} - C_2H_2O$), 361 (2, $M^{++} - C_2H_4O_2$), 319 (20, 361 – C₂H₂O), 277 (12, 319 – C₂H₂O), 235 (45, 277 – C₂H₃O), 43 (100, C₂H₃O⁺). Anal. calc. for C₁₇H₁₉N₅O₈ (421.37): C 48.46, H 4.55, N 16.62; found: C 48.31, H 4.79, N 16.34.

12: Yellow crystals, m.p. > 260°. UV (EtOH): 392 (2600), 348 (5080), 333 (5810), 294 (29310), 283 (30880), 243 (12080), 206 (12080), 206 (19610). IR (KBr): 3430 (br.), 3144m, 2925m, 2854 (sh), 2790 (sh), 1778 (sh), 1761m, 1672s, 1629s, 1587s, 1537m, 1482m, 1446m, 1369s, 1341w, 1304s, 1218s, 1190s, 1112m, 1048m, 1009w, 952w, 884m, 866 (sh), 785m, 758w, 720w. ¹H-NMR (270 MHz, (D₆)DMSO): 12.45 (br., NH(2)); 11.90 (br., NH–C(3)); 9.02 (*s*, H–C(6)); 8.80 (br., H–C(9)); 6.93 (*d*, $J(8,9) = 2.9$, H–C(8)); 2.38 (*s*, Ac); 2.20 (*s*, Ac). EI-MS: 301 (16, M^{++}), 259 (76, $M^{++} - C_2H_2O$), 217 (100, 259 – C₂H₂O), 43 (42, C₂H₃O). Anal. calc. for C₁₃H₁₁N₅O₄ · 0.5 CH₃OH (317.28): C 51.11, H 4.13, N 22.07; found: C 51.48, H 3.84, N 21.81.

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