168. Synthesis of 2'-Deoxyribofuranosides of 8-Aza-7-deazaguanine and Related Pyrazolo[3,4-*d*]pyrimidines

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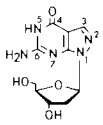
The *N*(1)- and *N*(2)-(2'-deoxyribofuranosides) **1** and **2**, respectively, of 8-aza-7-deazaguanine were prepared *via* phase-transfer glycosylation in the presence or absence of Bu_4NHSO_4 as catalyst of 6-amino-4-methoxy-1*H*-pyrazolo[3,4-*d*]pyrimidine (**7c**) with 2-deoxy-3,5-di-*O*-(*p*-toluoyl)-α-D-erythro-pentofuranosyl chloride (**10**). On a similar route, but without catalyst and employing THF as organic phase, the 6-amino-4-chloronucleosides **11b** and **12b** were synthesized from **7a** and converted into the *N*(1)-and *N*(2)-substituted 4-thioxo analogues **17a** and **18a**, respectively. The ratio of *N*(1)- to *N*(2)-glycosylation was 2:1 for **7c** and 1:1 for **7a**, *viz.* depending on the nucleobase structure. The rate of the H^+-catalyzed N-glycosyl hydrolysis was strongly decreased for the *N*(2)-(β-D-2'-deoxyribofuranosides) as compared to the *N*(1)-compounds. However, the *N*(1)-nucleoside **1**, which is an isostere of 2'-deoxyguanosine, is sufficiently stable to be employed later in solid-phase oligonucleotide synthesis.

Introduction. – Pyrazolo[3,4-*d*]pyrimidines as well as their ribofuranosides exhibit extraordinary therapeutic activities against severe diseases [1] [2]. A classical example of these biologically active purine analogues is allopurinol (= 1*H*-pyrazolo[3,4-*d*]pyrimidin-4(5*H*)-one) which is the compound of choice in the treatment of gout [3]. Due to the closely related structures of purines and pyrazolo[3,4-*d*]pyrimidines – only C(8) and N(7) are exchanged (purine numbering) – it is expected that incorporation of the corresponding 2'-deoxyribofuranosides in DNA does not affect *Watson-Crick* base pairing, but can influence stacking interactions between the bases. Recently, we were able to show that allopurinol 2'-deoxyribofuranoside can successfully replace 2'-deoxyinosine in alternating oligomers with the result of a stabilized duplex structure [4]. Due to these findings, we now investigate the synthesis of still unknown pyrazolo[3,4-*d*]pyrimidine 2'-deoxyribofuranosides, which are related to 2'-deoxyguanosine (**3**) and to the pyrrolo-[2,3-*d*]pyrimidine nucleoside **4** and can be used for the site-directed modification of DNA fragments.

In the course of the synthesis of pyrazolo[3,4-*d*]pyrimidine ribofuranosides, it became apparent that ribosylation of the silylated bases gave rise to the formation of regioisomers and even two-fold glycosylated heterocycles [5]. In order to direct the electrophilic attack into the five-membered-ring system, we have generated the nucleobase anion preferentially under phase-transfer conditions [6]. By this means, allopurinol 2'-deoxyribofuranoside and 4-aminopyrazolo[3,4-*d*]pyrimidine 2'-deoxyribofuranoside have been recently synthesized [7]. We now report on the synthesis of the 2'-deoxyribofuranosides **1** and **2** of the nucleobase 8-aza-7-deazaguanine (**9a**) [8], of the 4-thioxo analogues (**17a**) and **18a**, and of the related methoxy derivatives **15** and **16**. Furthermore, we investigated the stability of *N*(1)- and *N*(2)-(2'-deoxyribofuranosides) under acidic conditions.



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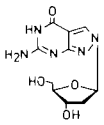

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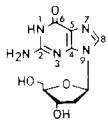

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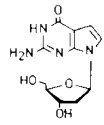

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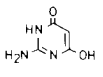

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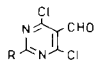
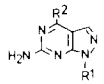
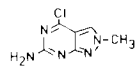


6a R = NH_2
b R = N=CHN$(CH_3)_2$



7	R^1	R^2
a	H	Cl
b	CH_3	Cl
c	H	CH_3O



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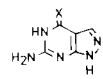


9a X = O
b X = S

Results and Discussion. – A synthesis of 6-amino-1*H*-pyrazolo[3,4-*d*]pyrimidin-4(5*H*)-one (**9a**), the aglycone of the nucleosides **1** and **2** has been already described [9]. This synthesis was based on the anellation of the pyrimidine-ring system on an appropriately functionalized pyrazole moiety. We have approached the synthesis of a useful nucleobase intermediate in a different way by starting with a pyrimidine derivative and condensing the pyrazole moiety afterwards.

Synthesis of Nucleobase **9b**. *Vilsmeyer-Haack* formylation of compound **5** yielded the aldehyde **6a**. According to the protocol of *Klötzer* and *Herberz* [10], this reaction results only in low yield (29%). Surprisingly, *Bell et al.* [11] which actually used the same reaction conditions came up with a yield of 51%. We have repeated this protocol and found it difficult to obtain reproducible results. Therefore, a series of experiments was set up employing different ratios of the pyrimidine **5** *vs.* $POCl_3$ and DMF. As a result, a ratio **5**/$POCl_3$/DMF of 1:8:2.2 was found to be superior to the ratio of 1:5:2, as reported earlier. At the same time, the workup procedure was changed. Simple hydrolysis of the reaction product in a large volume of H_2O formed pure **6a** as crystalline precipitate with a yield of 80%.

In a separate experiment, we only allowed the hydrolysis of $POCl_3$ and neutralized the resultant immediately with NH_3. Under these conditions, compound **6b** was isolated which is the amidine of **6a**. It demonstrates that **6b** is a reaction intermediate consuming already 2 equiv. of DMF. However, the amidine group protects compound **6a** very efficiently, but has to be hydrolysed subsequently before obtaining compound **6a**. We feel that an incomplete hydrolysis of the intermediate **6b** was the reason for obtaining low yields as reported earlier [10] [11].

Reaction of compound **6a** with aqueous hydrazine gave compound **7a** in 78% yield. Further treatment with thiourea in MeOH and subsequent hydrolysis with MeONa/MeOH yielded crystalline **9b**. This exhibited the same UV data (λ_{max} 333 nm) as the material obtained by *Robins* on a different route [9]. Moreover, the ^{13}C-NMR data *(Table 1)* confirmed this structure.

Phase-Transfer Methylation of Nucleobase **7a**. In order to test the utility of compound **7a** for further glycosylation studies, we determined the pK_a value of deprotonation in *Teorell-Stenhagen* buffer [12] UV-spectrophotometrically at 290 or 320 nm. According to a value of 10.3, the anion of compound **7a** is formed in aqueous solution, which makes it applicable for reactions in biphasic systems.

Table 1. *^{13}C-NMR Chemical Shifts* ($(D_6)Me_2SO$) *of Pyrazolo[3,4-d]pyrimidines and their 2'-Deoxyribofuranosides*

Compound	Substituents and anomeric configuration	C(3)	C(3a)	C(4)	C(6)	C(7a)	CH_3	CH	C(1')	C(2')	C(3')	C(4')	C(5')
1	6-NH_2, 4-oxo, 1-subst., β-D-conf.	134.9	99.7	157.4	154.6	155.2			83.1	37.9	71.0	87.3	62.4
2	6-NH_2, 4-oxo, 2-subst., β-D-conf.	127.8	102.2	159.3	153.5	160.7			89.8	40.0	70.5	88.1	61.9
7a	6-NH_2, 4-Cl	132.5	105.7	153.1	161.4	157.4							
7b	6-NH_2, 4-Cl, 1-Me	131.6	106.0	153.3	161.3	155.6	33.3						
7c	6-NH_2, 4-CH_3O	131.3	95.4	163.4	161.8	158.4	53.0						
8[a])	6-NH_2, 4-Cl, 2-Me	125.2	108.6	156.5	161.1	163.5	40.8						
9a	6-NH_2, 4-oxo	134.5	99.2	158.0	154.4	156.6							
9b	6-NH_2, 4-thioxo	137.2	110.9	178.7	153.4	151.7							
11a	6-NH_2, 4-CH_3O, 3',5'-di-*O*-Tol,1-subst., β-D-conf.	132.5	96.2	163.5	162.1	158.0	53.2		81.0	34.9	74.9	83.4	64.0
11b[a])	6-NH_2, 4-Cl, 3',5'-di-*O*-Tol, 1-subst., β-D-conf.	133.6	108.7	155.2	161.0	156.6			85.0	35.6	75.5	82.4	64.2
12a	6-NH_2, 4-CH_3O, 3',5'-di-*O*-Tol, 2-subst., β-D-conf.	124.5	97.9	165.0	161.5	163.4	53.2		89.9	36.7	74.3	81.9	64.1
12b[a])	6-NH_2, 4-Cl, 3',5'-di-*O*-Tol, 2-subst., β-D-conf.	123.6	108.2	157.3	160.4	161.7			91.6	38.8	74.9	84.2	60.3
13	6-NH_2, 4-CH_3O, 3',5'-di-*O*-Tol, 1-subst., α-D-conf.	132.4	96.2	163.6	162.2	158.1	53.3		80.1	35.2	74.0	83.1	64.0
14	6-NH_2, 4-CH_3O, 1-subst., α-D-conf.	132.1	96.1	163.7	162.1	157.6	53.3		82.8	37.7	70.7	86.6	61.4
15	6-NH_2, 4-CH_3O, 1-subst., β-D-conf.	132.0	96.1	163.4	161.9	157.6	53.2		83.3	37.8	71.0	87.3	62.4
16	6-NH_2, 4-CH_3O, 2-subst., β-D-conf.	123.5	97.8	165.2	161.5	163.3	53.0		90.1	40.3	70.4	88.2	61.7
17a	6-NH_2, 4-thioxo, 3',5'-di-*O*-Tol, 1-subst., β-D-conf.	137.9	111.9	179.3	154.2	151.4			81.5	35.3	75.2	83.8	64.3
17b	6-NH_2, 4-thioxo, 1-subst., β-D-conf.	137.2	111.5	178.8	153.7	150.8			83.3	37.7	71.0	87.5	62.4
18a	6-NH_2, 4-thioxo, 2-subst., β-D-conf.	130.4	112.4	182.6	152.3	155.9			90.2	40.1	70.5	88.3	61.9

[a]) Measured in $CDCl_3$.

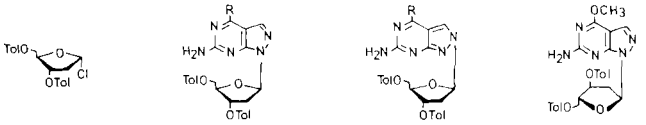
As reported earlier, phase-transfer methylation of pyrazolo[3,4-*d*]pyrimidines can be used as a test for the phase-transfer catalyzed glycosylation in biphasic systems [13]. Therefore, compound **7a** was treated with MeI in the biphasic mixture CH_2Cl_2/50% aqueous NaOH soln. in the presence of benzyltriethylammonium chloride furnishing two products **7b** and **8** which were separated chromatographically and obtained crystalline (ratio 2:1). The UV spectrum of the faster migrating **7b** (306 nm) was similar to that of the non-methylated **7a** (305 nm), whereas the slow migrating **8** showed a strong bathochromic shift (322 nm). Assignment of the position of methylation was made according to the coupling pattern of the non-decoupled ^{13}C-NMR spectra and according to the chemical shift of C(3) (see *Table 1*).

Previous studies in this laboratory [7] [13] as well as in others [14] have demonstrated that N(2)-substitution of pyrazolo[3,4-*d*]pyrimidines by Me, ribofuranosyl, or 2'-deoxyribofuranosyl residues results in an upfield shift of the C(3) signal in the ^{13}C-NMR by *ca.* 8 ppm compared to that of the non-substituted compounds. This occurs in all cases where the predominant species was the HN(1) tautomer. According to the UV and ^{13}C-NMR data (see *Exper. Part* and *Table 1*), the proton at the five-membered ring of **7a** can be localized at N(1); location at N(2) as found for oxoallopurinol [15] would shift the C(3) signal upfield and results in an altered UV spectrum. Consequently, the upfield shift of C(3) of **8** confirms the assigned structure with the Me group at N(2). This assignment is further underlined by the ^3J coupling pattern of C(3) of **8**. Due to the Me group at N(2), a ^3J(C(3),CH_3) of 3.2 Hz is observed, additionally to the large ^1J of 196 Hz. The faster migrating Me isomer **7b** exhibited almost unchanged ^{13}C-NMR data compared to **7a**. Also the non-decoupled ^{13}C-NMR spectrum of **7b** showed a complex coupling pattern for the signal of C(7a), together with sharp signals for C(4) and C(6). These findings are in agreement with the *N*(1)-location of the Me group.

Aqueous NaOH (50%) is the mostly used base in phase-transfer catalysis (PTC), but other inorganic bases such as $Ba(OH)_2$ or KOH either in solution or as solids are applied from time to time [16]. The latter was the most powerful base in the alkylation of benzamide [17]. As a consequence, we employed 30% aqueous KOH solution instead of 50% NaOH solution as inorganic phase for the methylation of **7a**. Under these conditions, the latter was readily soluble in the inorganic medium, which was not the case in 50% NaOH solution. The enhanced solubility of **7a** ensures a sufficient supply of reactive species and makes the 30% KOH solution to the solvent of choice for the phase-transfer methylation or glycosylation of **7a**.

Due to the strong alkaline conditions, highly reactive substituents such as the 4-Cl group of **7a** are expected to be displaced readily by OH ions. This displacement is unlike to take place in the nucleobase *N*(1)- or *N*(2)-anion, the reactivity being decreased very strongly. However, such substituents are displaced in *N*(1)- or *N*(2)-alkylated or *N*(1)- or *N*(2)-glycosylated nucleobases, unless the latter are very rapidly extracted into the organic medium. From earlier investigations on PTC-catalyzed glycosylations of allopurinol derivatives, we found it advantageous to use 4-MeO compounds such as **7c** instead of 4-Cl derivatives [15]. Additionally, it has been observed on other 4-methoxypyrazolo[3,4-*d*]pyrimidines [7] that the displacement of the MeO group on compounds such as **7c** employing OH ions occurs without side reactions, *e.g.* ring opening, and results in the formation of the aglycone such as **9a**. This sequence is a prerequisite of applying **7c** in the synthesis of the nucleoside **1**. Consequently, **7a** was converted into **7c** with NaOMe/MeOH in a yield of 90%.

Phase-Transfer Glycosylation of Nucleobase **7c** *to* **11a**/**12a**/**13**. Due to the pK_a value of **7c** (12.0) which was determined spectrophotometrically at 270 nm in *Teorell-Stenhagen* buffer [12] **7c** was soluble in 30% aq. KOH solution as anion. Employing CH_2Cl_2 as



10 **11a** R = CH_3O **12a** R = CH_3O **13**
b R = Cl **b** R = Cl

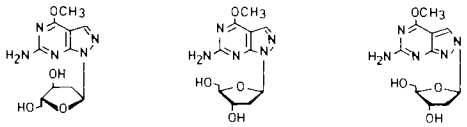
Tol = p-$CH_3C_6H_4CO$

organic phase, Bu_4NHSO_4 as catalyst, and the halogenose **10**, three reaction products were formed which could be separated by chromatography. Two of them, **11a** and **13**, exhibited very similar R_f values and almost identical UV spectra, implying that they were anomers. The third, more polar compound (formed in 15% yield) showed a bathochromically shifted UV spectrum, pointing to the regioisomeric structure **12a**. This was confirmed by ^{13}C-NMR spectroscopy. According to *Table 1*, C(3) of **12a** is shifted upfield compared to the parent **7c** which is an unambiguous indicator for $N(2)$-glycosylation [7] [14]. The site of glycosylation of the fast-migrating compounds **11b** and **13** could not be deduced from the ^{13}C-NMR data, but was later confirmed at the deprotected nucleosides. The anomeric configurations were deduced from the ^{1}H-NMR data.

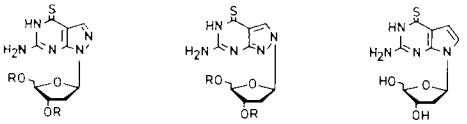
According to the findings of *Nuhn et al.* [18] for regular 2'-deoxyribofuranosides and by us for pyrrolo[2,3-*d*]pyrimidine 2'-deoxyribofuranosides [19] as well as pyrazolo[3,4-*d*]pyrimidine 2'-deoxyribofuranosides [7], the anomeric configuration can be deduced from ^{1}H-NMR shifts of toluoylated derivatives. They exhibit signals for H–C(4') and CH_2(5') with large differences in the series of α-D-anomers, whereas the same signals of β-D-compounds coincide. Applying these rules to the glycosylation products of **7c**, the $N(1)$-glycosylated **11a** and the $N(2)$-glycosylated isomer **12a** have both β-D-configuration, whereas the $N(1)$-glycosylated **13** which is more polar than **11a** has α-D-configuration.

In a forthcoming communication, we will show that an increasing amount of phase-transfer catalyst favours formation of 2'-deoxy-α-D-ribofuranosides [20], due to an *in situ* equilibration of the α-halogenose **10** [21]. Therefore, we omitted $BuNHSO_4$ from the glycosylation mixture (CH_2Cl_2/Bu_4NHSO_4/30% aq. KOH solution). However, applying CH_2Cl_2 as organic phase was not successful. But replacing CH_2Cl_2 by THF gave rise to the formation of only 2 products in a total yield of 60%, *i.e.* the β-D-anomers **11a** (47%) and **12a** (13%). Our findings indicate that the formation of α-D-anomers is directly related to the presence and amount of the phase-transfer catalyst. A still open question is the special action of THF in the biphasic reaction. We think, that an ion pair is formed between K^+ and the nucleobase anion which is almost insoluble in CH_2Cl_2, but can be dissolved in THF due to the complexation of the K^+ ion.

Nucleosides **1** *and* **2** *from* **11a** *and* **12a** via **15** *and* **16**, *respectively*. Deprotection of the anomers **13** and **11a** and of regioisomer **12a** was accomplished upon treatment with NaOMe/MeOH at room temperature yielding, after chromatography, the crystalline methoxynucleosides **14**, **15**, and **16**, respectively. Again, the nucleobase moieties of the $N(1)$-glycosylated anomers **14** and **15** exhibited very similar ^{13}C-NMR data as the aglycone **7c**, whereas the data of the $N(2)$-glycosylation product **16** were different (*Table 1*). These findings and further ^{13}C-NMR data were in agreement with the formerly assigned glycosylation sites, and the ^{1}H-NMR data confirmed the attributed anomeric configurations.



14 **15** **16**



17a R = H **18a** R = H **19**
b R = Tol **b** R = Tol

In the series of $N(1)$-ribofuranosides, the order of the sugar signals in the ^{13}C-NMR spectrum was C(4'), C(1'), C(3'), C(5'), C(2'), whereas the $N(2)$-compounds showed the sequence C(1'), C(4'), C(3'), C(5'), C(2'). This was confirmed by $^{1}H/^{13}C$ coupling experiments and was in agreement with earlier results on compounds lacking the 6-amino function [7].

The anomeric configuration derived from the ^{1}H-NMR data of H–C(4') and CH_2(5') of the protected nucleosides **11a** and **13** was confirmed by the coupling pattern of the anomeric proton of the corresponding free nucleosides **14** and **15**. Indeed, it has been shown [22] that α-D-anomers exhibit a *dd* for H–C(1'), whereas β-D-anomers show a pseudo-*t*. This empirical rule is in agreement with the structures assigned for compounds **14–16**.

Nucleophilic displacement (2N KOH) of the MeO group of **15** and **16** yielded the crystalline nucleosides **1** and **2**, respectively. These reactions proceeded much slower than in the case of the corresponding allopurinol 2'-deoxyribofuranoside. Quantitative data were obtained from measurements carried out in 0.2N KOH at elevated temperature (60°). The reaction was followed spectrophotometrically at 290 nm for **15** and at 315 nm for **16**. In accordance with results obtained from allopurinol 2'-deoxyribofuranoside, MeO displacement for the $N(2)$-compound **16** ($\tau/2 = 9.6$ min; $k = 7.2 \times 10^{-2}\text{sec}^{-1}$) was faster than for the $N(1)$-nucleoside **15** ($\tau/2 = 33$ min; $k = 2.1 \times 10^{-2}\text{sec}^{-1}$).

4-Thioxo Analogues **17a** *and* **18a**. We were also interested in a facile synthesis of the 4-thioxo analogues **17a** and **18a** of the nucleosides **1** and **2**, respectively. It has been shown on other pyrazolo[3,4-*d*]pyrimidine nucleosides [23], that nucleophilic displacement of 4-Cl substituents with thiourea is an efficient method to achieve this goal. As a prerequisite, the precursors **11a** and **12b** had to be synthesized. Thus, phase-transfer glycosylation of the chloro derivative **7a** (see above) with **10** under the same conditions as used for **7c** + **10**→**11a** + **12a** (see above) gave the $N(1)$-isomer **11b**, and the $N(2)$-glycosylation product **12b** in a ratio of *ca.* 1:1 (total yield 78%). In the case of **7c**, a ratio of 2:1 was observed for **11a**/**12a**. This difference in the $N(1)/N(2)$ glycosylation ratio may be due to the following factors: *(i)* Electron-withdrawing substituents such as Cl can affect the nucleophilicity of N(1) and N(2) of the nucleobase in a different manner. As a consequence, the kinetically controlled glycosylation at N(2) may be more influenced as the thermodynamically controlled reaction at N(1). Thermodynamic control of $N(1)$-glycosylation can be deduced from the most favoured tautomeric structure of the nucleobases **7a** or **7c**. As the ^{13}C-NMR spectra *(Table 1)* show, both compounds carry the

proton at N(1) which implies that the same location is valid for other residues such as the 2'-deoxyribofuranosyl group. *(ii)* Since it can be expected that the formation of the *N*(1)- and *N*(2)-isomers occurs with a different rate, side-reactions of the halogenose **10** may become important for the product ratio. That these side-reactions occur in biphasic systems such as the one employed during glycosylation of **7a** or **7c** has recently been shown [20].

If *Zemplen* deprotection was employed for **11b** and **12b**, detoluoylation of the sugar moiety was followed by nucleophilic displacement of the 4-Cl group by CH_3O, furnishing compounds **15** and **16**, respectively. The ease of this reaction sequence made **11b** and **12b** particularly suitable for the conversion into other nucleosides. Treatment of **11b** with thiourea furnished **17b** in 89% yield. This was converted into **17a** by the action of NaOMe in MeOH. Applying the same conditions to **12b** but without isolation of **18b** gave the *N*(2)-nucleoside **18a**.

Stability of **1**, **2**, **17a**, *and* **18a** *towards Hydrolysis.* The stability of the N-glycosylic bond of 2'-deoxynucleosides is a severe problem during oligonucleotide synthesis. Due to a H^+-catalyzed deprotection step neccessary for the detritylation, depurination can occur. In a previous study on 2'-deoxyribofuranosides of allopurinol or 4-aminopyrazolo[3,4-*d*]pyrimidine, we were able to show that these pyrazolo[3,4-*d*]pyrimidine *N*(1)-nucleosides are more stable under acidic conditions than purine *N*(9)-(2'-deoxyribofuranosides) [23].

Table 2. *Kinetic Data of N-Glycosyl Bond Hydrolyses of Nucleobase-Modified 2'-Deoxyribofuranosides*[a])

	1[b])	**17a**[b])	**3**[b])	**2**[c])	**18a**[c])
$k \times 10^2$ [min^{-1}]	14.0	1.7	6.5	9	11
$\tau_{1/2}$ [min]	4.95	42.0	10.6	7.7	6.3

[a]) Measured at 1×10^{-4} M nucleoside concentration. k was calculated according to the equation $k = 1/t \cdot \ln(E_0-E_\infty)/(E_t-E_\infty)$.
[b]) In 0.5N HCl at 25°.
[c]) In 1N HCl at 60°.

In order to test the stability of nucleosides **1**, **2**, **17a**, and **18a**, they were treated with aq. HCl at 25°. Under these conditions, the intact corresponding nucleobases were released from the nucleosides as shown by comparison of the pH-dependend UV spectra of the hydrolysis products with authentic aglycones. Data were obtained from the decrease of the UV absorbance differences between the nucleoside and the corresponding aglycone and are depticted in *Table 2*. The measurements were carried out where the UV difference was most pronounced. For comparison, compounds **3** and **4** were also measured. *Fig. a* shows the cleavage of the 2'-deoxyguanosine analogues. As expected, compound **4** was stable in 0.5N HCl at 25°, whereas the purine nucleoside **3** decayed rapidely. A slightly faster reaction was observed for the isostere **1**. In contrast to this *N*(1)-nucleoside, the regioisomer **2** was unexpectedly stable and underwent a significant hydrolysis only at elevated temperature. Similar results were obtained for the thioxonucleosides **17a**, **18a**, and **19** *(Fig. b)*. This was different from the behaviour of the corresponding allopurinol 2'-deoxyribonucleosides, where the *N*(2)-nucleoside ($k = 4.5 \times 10^{-2}$ min^{-1}) hydrolyzed more rapidly than the *N*(1) regioisomer ($k = 1.6 \times 10^{-2}$ min^{-1}).

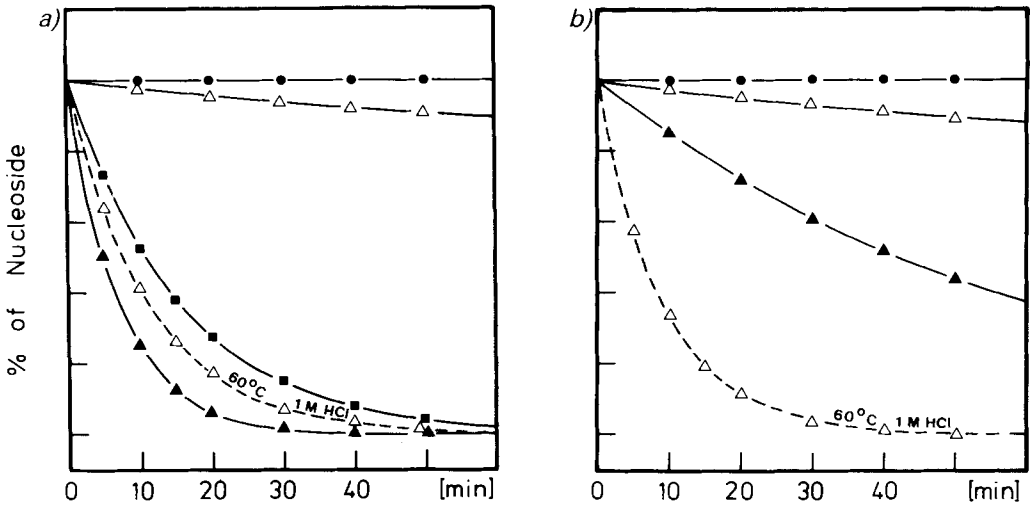


Fig. *Hydrolyses rates of isosteric 2'-deoxyribofuranosides in 0.5* M *HCl at 25°*. Data are taken from the decrease of the UV absorption at the wavelength indicated: *a)* **3** at 260 nm (■——■), **4** at 270 nm (●——●), **1** at 251 nm (▲—— ▲), and **2** at 260 nm (△——△); *b)* **19** at 350 nm (●——●), **17a** at 237 nm (▲——▲), and **18a** at 345 nm (△——△).

Table 3. *p*K_a *Values*[a]) *and UV Data of (2-Deoxy-*D-erythro-*pentofuranosyl)-pyrazolo[3,4-*d*]pyrimidines*

Compound	pK_a	λ_{max} [nm]		
		Mono-cation	Neutral molecule	Mono-anion
1 (*N*(1)-substitution)	0.5[b]), 9.3	(248)[b])	253	265
2 (*N*(2)-substitution)	2.9, 9.4	257	276	255, 284
17a (*N*(1)-substitution)	7.7	–	237, 270, 330	236, 281, 315
18a (*N*(2)-substitution)	1.8, 8.2	232, 260, 330	237, 262, 297, 345	284, 334
Allopurinol *N*(1)-(*β*-D-2'-deoxyriboside)	8.7	–	252	271
Allopurinol *N*(2)-(*β*-D-2'-deoxyriboside)	9.5	–	261	283
9a	2.9, 9.2	248	252	267
9b	1.5, 7.5	233, 259, 325	233, 257, 330	231, 270, 314
Allopurinol	9.3	–	250	261

[a]) Determined in *Teorell-Stenhagen* buffer [12].
[b]) Values could only be approximately determined due to hydrolysis of the *N*-glycosylic bond.

An interpretation of the increased stability of the *N*(2)-(2'-deoxyribofuranosides) **2a** and **18a** over the *N*(1)-compounds **1** and **17a** which was opposite to that in the allopurinol series can be given on the basis of the p*K* values, which were determined spectrophotometrically in *Teorell-Stenhagen* buffer *(Table 3)*. From the p*K* values of the *N*(2)-isomers **2** and **18a**, it can be concluded that these molecules form mono-cations under hydrolysis conditions (0.5N HCl). However, both cations are fairly stable at 25°. Under the same conditions, rapid hydrolysis of the *N*(1)-regioisomers **1** and **17a** took place. The basic p*K* values of **9a** and **9b** imply that also the nucleosides **1** and **17a** might have basic p*K* values around the p*K* values of their aglycones and form cations in 0.5N HCl. However, these values are not accessible, due to the rapid hydrolysis occurring under acidic conditions. Therefore, we conclude that the differences in glycosylic-bond hydro-

lysis of the cations of $N(1)$- and $N(2)$-nucleosides are due to different protonation sites at their nucleobases.

Earlier investigations on guanine $N(7)$- and $N(9)$-(β-D-ribofuranosides) have shown that the $N(7)$-regioisomers hydrolyze much more slowly than the $N(9)$-compounds [25]. The slow hydrolysis of the $N(7)$-compound was explained by protonation of this molecule at the guanidine moiety, whereas guanosine is protonated at $N(7)$ [25]. A similar behaviour, *i.e.* protonation of the $N(1)$-compounds at $N(2)$ and of the $N(2)$-compounds at the guanidine moiety and localization of a positive charge at the five-membered ring as a prerequisite for N-glycosyl hydrolysis [26], would explain the different rates of N-glycosyl hydrolysis as depicted in the *Figure*. Experiments assigning these protonation sites are in progress.

We thank *H. Driller* for providing us with an improved procedure for isolation of compound **1** and Ms. *Sabine Menkhoff* for technical assistance. Financial support by the *Deutsche Forschungsgemeinschaft* and the *Fonds der Chemischen Industrie* is gratefully acknowledged.

Experimental Part

General. See [7b]. Solvent systems: CH_2Cl_2/EtOAc 9:1 (*A*), CH_2Cl_2/EtOAc 4:1 (*B*), CH_2Cl_2/EtOAc 1:1 (*C*), CH_2Cl_2/EtOAc 1:4 (*D*), EtOAc (*E*), cyclohexane/EtOAc 3:2 (*F*), cyclohexane/EtOAc 1:4 (*G*), $CHCl_3$/MeOH 19:1 (*H*), $CHCl_3$/MeOH 9:1 (*I*), $CHCl_3$/MeOH 4:1 (*K*), H_2O/*i*-PrOH 9:1 (*L*), $CHCl_3$/MeOH 3:2 (*M*). ^{1}H-NMR: multiplicities between inverted commas ('') refer to pseudomultiplicities.

2-Amino-4,6-dichloropyrimidine-5-carboxaldehyde (**6a**) was prepared according to the method of *Klötzer* [4] but using other conditions: To an ice-cold soln. of $POCl_3$ (150 ml, 0.61 mol), abs. DMF (35 ml, 0.46 mol) was added dropwise within 20 min. The ice-bath was removed and powdered 2-amino-6-hydroxypyrimidine-4(3*H*)-one (**5**; 25 g, 0.2 mol) was added in small portions under stirring and N_2. After the exothermic reaction had ceased, the mixture was brought to 100°. Heating was continued for 1.5 h. The soln. was cooled, reduced to half of the volume, and given in small portions into water/ice 3:1 (4000 ml). The resultant was warmed gently to 50°. Within 2 h, **6a** precipitated. It was filtered, washed with H_2O, dried, and recrystallized from THF/EtOAc. Pale yellow needles (30.0 g, 80%) which decomposed above 130°. For anal. characterization, a small sample was filtered through a bed of silica gel with EtOAc and crystallized from a reduced volume yielding colorless needles which decomposed above 200°. TLC (silica gel, *F*): R_f 0.5. UV (MeOH): 281 (18100). ^{1}H-NMR ((D$_6$)Me$_2$SO): 8.49 (*s*, NH_2); 10.05 (*s*, CHO). ^{13}C-NMR ((D$_6$)Me$_2$SO): 112.9 (C(5)); 161.7, 163.1 (2*s*, C(2), C(4), C(6)); 184.5 (CHO).

4,6-Dichloro-2-{[(dimethylamino)methylidene]amino}pyrimidine-5-carboxaldehyde (**6b**) was prepared as described for **6a** except that the icewater mixture was neutralized (conc. NH_3) immediately after the formation of a homogenous aq. soln. The precipitate formed was collected, dried, and purified on silica gel (column 20 × 5 cm, *C*). From the main zone, a solid material was isolated which was crystallized from MeOH in pale yellow needles (65% yield), m.p. 140°. TLC (silica gel, *C*): R_f 0.2. UV (MeOH): 335 (45000). ^{1}H-NMR ((D$_6$)Me$_2$SO): 3.14, 3.28 (2*s*, 2 Me); 8.78 (*s*, N–CH = N); 10.15 (*s*, CHO). ^{13}C-NMR ((D$_6$)Me$_2$SO): 35.4, 41.3 (2 Me); 115.8 (C(5)); 160.7 (N–CH = N); 162.1 (C(4), C(6)); 165.2 (C(2)); 185.3 (CHO). Anal. calc. for $C_8H_8Cl_2N_4O$: C 38.99, H 3.26, Cl 28.70, N 22.68; found: C 39.19, H 3.10, Cl 28.94, N 22.89.

*6-Amino-4-chloro-1*H*-pyrazolo[3,4-*d*]pyrimidine* (**7a**). A soln. of **6a** (9.6 g, 50 mmol) in THF/H_2O 3:1 (200 ml) at 50° was treated with hydrazine hydrate (5.1 ml, 100 mmol) in H_2O (50 ml) at r.t. under stirring. Within 10 min, a yellowish precipitate was formed. The crude mixture was poured into ice-cold H_2O (250 ml) yielding a precipitate. Precipitation was completed by evaporation to a reduced volume. The solid material was filtered and recrystallized from DMF/H_2O. Pale yellow needles (6.6 g, 78%) which decomposed above 200°. TLC (silica gel, *I*): R_f 0.4. UV (MeOH): 227 (26500), 305 (6000). ^{1}H-NMR ((D$_6$)Me$_2$SO): 7.14 (*s*, NH_2); 7.94 (*s*, H–C(3)); 13.25 (*s*, H–C(1). ^{13}C-NMR ((D$_6$)Me$_2$SO): 105.7 (C(3a)); 132.5 (C(3)); 153.1 (C(4)); 157.4 (C(7a)); 161.4 (C(6)). Anal. calc. for $C_5H_4ClN_5$: C 35.42, H 2.38, Cl 20.91, N 41.30; found: C 35.37, H 2.42, Cl 21.01, N 41.38.

Phase-Transfer Methylation of **7a**. A suspension of **7a** (500 mg, 2.95 mmol) in CH_2Cl_2 (50 ml) was added to a soln. of benzyltriethylammonium chloride (70 mg, 0.31 mmol) in 50% aq. NaOH soln (20 ml). During agitation with a vibromixer, MeI (0.56 ml, 9 mmol) dissolved in a small volume of CH_2Cl_2 was added in 1 portion and mixing was continued for 2 min at r.t. The org. layer was separated and the aq. phase extracted twice with CH_2Cl_2 (50 ml, each). The combined extracts were filtered, adsorbed on silica gel (5 g), and loaded on the top of a silica-gel column

(20 × 5 cm). Elution with solvent *H* gave 2 main zones. From the fast migrating zone, colorless crystals (220 mg, 41%) of *6-amino-4-chloro-1-methyl-1*H*-pyrazolo[3,4-*d*]pyrimidine* (**7b**) were obtained after crystallization from MeOH, m.p. 242° (dec.). TLC (silica gel, *I*): R_f 0.7. UV (MeOH): 230 (28500), 306 (5100). ^{1}H-NMR ($(D_6)Me_2SO$): 3.80 (*s*, CH_3N); 7.28 (*s*, NH_2); 7.93 (*s*, H–C(3)). ^{13}C-NMR ($(D_6)Me_2SO$): 33.3 (CH_3N); 106.0 (C(3a)); 131.6 (C(3)); 153.3 (C(4)); 155.6 (C(7a)); 161.3 (C(6)). Anal. calc. for $C_6H_6ClN_5$: C 39.25, H 3.29, Cl 19.31, N 38.15; found: C 39.28, H 3.26, Cl 19.14, N 38.25.

The slow-migrating zone afforded pale yellow crystals (90 mg, 17%) of *6-amino-4-chloro-2-methyl-2*H*-pyrazolo[3,4-*d*]pyrimidine* (**8**). After recrystallization from MeOH, m.p. 178° (dec.). TLC (silica gel, *I*): R_f 0.5. UV (MeOH): 225 (25000), 281 (6300), 322 (4500). ^{1}H-NMR ($(D_6)Me_2SO$): 3.99 (*s*, CH_3); 6.87 (*s*, NH_2); 8.41 (*s*, H–C(3)). ^{13}C-NMR ($(D_6)Me_2SO$): 33.3 (CH_3N); 106.0 (C(3a)); 131.6 (C(3)); 153.3 (C(4)); 155.6 (C(7a)); 161.3 (C(6)). Anal. calc. for $C_6H_6ClN_5$: C 39.25, H 3.29, Cl 19.31, N 38.15; found: C 39.42, H 3.29, Cl 19.23, N 38.21.

*6-Amino-4-methoxy-1*H*-pyrazolo[3,4-*d*]pyrimidine* (**7c**). Compound **7a** (6.8 g, 0.04 mol) in 1M MeONa/MeOH (100 ml) was refluxed for 1 h. Upon cooling, Et_2O (100 ml) was added and the precipitate filtered off. The filtrate was neutralized with AcOH and evaporated. The residue was dissolved in MeOH and **7c** precipitated in pale yellow needles by addition of ice-cold H_2O (5.9 g, 90%), m.p. > 220°. TLC (silica gel, *I*): R_f 0.3. UV (MeOH): 244 (5500), 276 (7800). ^{1}H-NMR ($(D_6)Me_2SO$): 3.95 (*s*, CH_3O); 6.61 (*s*, NH_2); 7.78 (*s*, H–C(3)); 12.81 (*s*, NH). Anal. calc. for $C_6H_7N_5O$: C 43.63, H 4.28, N 42.41; found: C 43.52, H 4.28, N 42.50.

*6-Amino-1*H*-pyrazolo[3,4-*d*]pyrimidin-4(5*H*)-thion* (**9b**). A mixture of **7a** (500 mg, 2.9 mmol) and thiourea (250 mg, 3.3 mmol) in MeOH (50 ml) was refluxed for 1 h. Then, 1M MeONa/MeOH (7 ml) was added and refluxing continued for 15 min. After cooling, the soln. was neutralized with 1M aq. HCl (7 ml) yielding a precipitate which was redissolved by addition of conc. NH_3 (20 ml) and H_2O (50 ml). Insoluble material was filtered off and the soln. concentrated to 30 ml yielding a precipitate. This was filtered off and washed with H_2O. Yellowish needles (410 mg, 85%) which did not melt above 300°. TLC (silica gel, *K*): R_f 0.2. UV (MeOH): 235 (18200), 268 (7000), 333 (19500). UV (pH 1): 258, 326. UV (pH 11): 277, 328 [9]. ^{1}H-NMR ($(D_6)Me_2SO$): 6.79 (*s*, NH_2); 7.89 (*s*, H–C(3)); 11.9 (*s*, H–C(5)); 13.01 (*s*, NH).

Glycosylation of **7c** *with 2-Deoxy-3,5-di-*O*-(*p*-toluoyl)-β-*D*-*erythro*-pentofuranosyl Chloride* (**10**). Compound **7c** (500 mg, 3.0 mmol) was dissolved in 30% aq. KOH soln. (4 ml) by gentle warming. It was cooled to r.t. and covered with a soln. of **10** (1.5 g, 3.85 mmol) in THF (20 ml). The layers were vigorously mixed with a vibromixer for 2 min at r.t. Thereupon, CH_2Cl_2 (70 ml) was added, the org. layer separated, dried over Na_2SO_4, filtered, and evaporated to dryness. The yellowish oily residue was dissolved in CH_2Cl_2 and chromatographed on silica gel (column 15 × 5 cm) with eluant *A* followed by *B* and then *E*. From the fast migrating main zone, colorless *6-amino-1-(2′-deoxy-3′,5′-di-*O*-(*p*-toluoyl)-β-*D*-erythro-pentofuranosyl)-4-methoxy-1*H*-pyrazolo[3,4-*d*]pyrimidine* (**11a**; 740 mg, 47%) was isolated. Crystallization from MeOH afforded colorless needles, m.p. 157°. TLC (silica gel, *F*): R_f 0.31. UV (MeOH): 225 (36500), 241 (38300), 274 (10900). ^{1}H-NMR ($(D_6)Me_2SO$): 2.35, 2.38 (2*s*, CH_3); 2.68, 3.22 (2*m*, H_a–C(2′), H_b–C(2′)); 3.97 (*s*, CH_3O); 4.43 (*m*, H–C(4′), CH_2(5′)); 5.81 (*m*, H–C(3′)); 6.60 (*'t'*, *J* = 6.2, H–C(1′)); 6.93 (*s*, NH_2); 7.28–7.91 (4*d*, *J* = 8.1, 8 arom. H); 7.94 (*s*, H–C(3)). Anal. calc. for $C_{27}H_{27}N_5O_6$: C 62.66, H 5.29, N 13.53; found: C 62.81, H 5.37, N 13.45.

From the second zone, colorless amorphous *6-amino-2-(2′-deoxy-3′,5′-di-*O*-(*p*-toluoyl)-β-*D*-erythro-pentofuranosyl)-4-methoxy-1*H*-pyrazolo[3,4-*d*]pyrimidine* (**12a**; 210 mg, 13%) was isolated. TLC (silica gel, *G*): R_f 0.3. UV (MeOH): 224 (29000), 240 (34200), 283 (8800), 295 (8600). ^{1}H-NMR ($(D_6)MeSO$): 2.34, 2.37 (2*s*, CH_3); 2.75, 3.12 (2*m*, H_a–C(2′), H_b–C(2′)); 3.96 (*s*, CH_3O); 4.51 (*m*, H–C(4′), CH_2(5′)); 5.83 (*m*, H–C(3′)); 6.43 (*'p'*, *J* = 5.7, H–C(1′)); 6.51 (*s*, NH_2); 7.26–7.91 (4*d*, *J* = 8.1, 8 arom. H); 8.46 (*s*, H–C(3)). Anal. calc. for $C_{27}H_{27}N_5O_6$: C 62.66, H 5.29, N 13.53; found: C 62.70, H 5.39, N 13.52.

Phase-Transfer Glycosylation of **7c** *with* **10** *in the Presence of* Bu_4NHSO_4. A soln. of **7c** (510 mg, 3.1 mmol) in 30% aq. KOH soln. (4 ml) was prepared by gentle warming. The soln. was cooled to r.t. and covered with a soln. of Bu_4NHSO_4 (100 mg, 0.3 mmol) in CH_2Cl_2 (20 ml). The layers were thoroughly mixed with a vibromixer, and a soln. of **10** (1.53 g, 3.9 mmol) in CH_2Cl_2 was added within 1 min. Stirring was continued for another min. The org. layer was separated, the aq. layer extracted twice with CH_2Cl_2, and the combined org. extracts filtered and evaporated. The yellowish oil was dissolved in CH_2Cl_2 and chromatographed on silica gel (column 15 × 5 cm). Solvent *I* eluted a colorless material (590 mg, 37%) which was identical with **11a**. Solvent *B* yielded a colorless solid (100 mg, 6%) which was **13**, and EtOAc eluted a slow migrating material which was identical with **12a** (240 mg, 15%). *6-Amino-1-(2′-deoxy-3′,5′-di-*O*-(*p*-toluoyl)-α-*D*-erythro-pentofuranosyl)-4-methoxy-1*H*-pyrazolo[3,4-*d*]pyrimidine* (**13**): TLC (silica gel, *F*): R_f 0.3. UV (MeOH): 225 (34900), 241 (36300), 274 (11000). ^{1}H-NMR ($(D_6)Me_2SO$): 2.36, 2.38 (2*s*, CH_3); 2.94 (*dt*, *J* = 5.1, 14.0 H_b–C(2′), 3.13 (*'quint.'*, *J* = 7.4, H_a–C(2′)); 4.02 (*s*, CH_3O); 4.48 (*'q'*, *J* = 4.7, H–C(4′)); 5.54 (*m*, H–C(3′)); 6.55 (*dd*, *J* = 5.0, 7.3, H–C(1′)); 6.91 (*s*, NH_2); 7.29–7.92 (4*d*, *J* = 8.1, 8 arom. H); 7.93 (*s*, H–C(3)). Anal. calc. for $C_{27}H_{27}N_5O_6$: C 62.66, H 5.29, N 13.53; found: C 62.50, H 5.36, N 13.45.

*6-Amino-1-(2'-deoxy-β-*D*-*erythro*-pentofuranosyl)-4-methoxy-1*H*-pyrazolo[3,4-*d*]pyrimidine* (**15**). A soln. of **11a** (2.1 g, 4.05 mmol) in 0.01M NaOMe/MeOH (150 ml) was stirred for 2 h at r.t. Then, the soln. was adsorbed onto silica gel (15 g) and chromatographed on silica gel column (20 × 5 cm). Elution with solvent *I* gave a main zone which yielded a colorless solid after evaporation. Crystallization from H_2O gave **15** as colorless needles (790 mg, 69%), m.p. 149–151°. TLC (silica gel, *I*): R_f 0.3. UV (MeOH): 223 (23300), 253 (7200), 277 (8700). ^{1}H-NMR ($(D_6)Me_2SO$): 2.18 (*dd*, *J* = 4.0, 6.6, 13.3, H_b–C(2')); 2.74 (*'quint.'*, *J* = 6.4, H_a–C(2')); 3.35, 3.49 (*m*, CH_2(5')); 3.78 (*m*, H–C(4')); 3,97 (*s*, CH_3O); 4.39 (*m*, H–C(3')); 4.74 (*t*, *J* = 5.7, OH–C(5')); 5.23 (*d*, *J* = 4.3, OH–C(3')); 6.40 (*'t'*, *J* = 6.5, H–C(1')); 6.85 (*s*, NH_2); 7.88 (*s*, H–C(3)). Anal. calc. for $C_{11}H_{15}N_5O_4$: C 46.97, H 5.38, N 24.90; found: C 46.71, H 5.38, N 24.98.

*6-Amino-1-(2'-deoxy-α-*D*-*erythro*-pentofuranosyl)-4-methoxy-1*H*-pyrazolo[3,4-*d*]pyrimidine* (**14**). A soln. of **13** (600 mg, 1.16 mmol) in 0.01M NaOMe/MeOH (40 ml) was treated as **15**. After chromatography (column 20 × 2 cm), colorless **14** (230 mg, 71%) was isolated. After crystallization from H_2O, plates with m.p. 244°. TLC (silica gel, *I*): R_f 0.3. UV (MeOH): 223 (22700), 253 (7400), 277 (8100). ^{1}H-NMR ($(D_6)Me_2SO$): 2.50 (*m*, H_b–C(2')); 2.67 (*'quint.'*, *J* = 6.9, H_a–C(2')); 3.39, 3.47 (2*m*, CH_2(5')); 3.90 (*m*, H–C(3')); 3.98 (*s*, CH_3O); 4.13 (*m*, H–C(4')); 4.73 (*t*, *J* = 5.6, OH–C(5')); 5.60 (*d*, *J* = 7.6, OH–C(3')); 6.34 (*dd*, *J* = 5.0, 7.2, H–C(1')); 6.89 (*s*, NH_2); 7.94 (*s*, H–C(3)). Anal. calc. for $C_{11}H_{15}N_5O_4$: C 46.97, H 5.38, N 24.90; found: C 47.10, H 5.40, N 24.92.

*6-Amino-2-(2'-deoxy-β-*D*-*erythro*-pentofuranosyl)-4-methoxy-2*H*-pyrazolo[3,4-*d*]pyrimidine* (**16**). Treatment of **12a** (1.1 g, 2.12 mmol) in 0.01M NaOMe/MeOH (80 ml) as described for **15**, chromatography on silica gel (column 15 × 4 cm, *K*), and evaporation of the solvent to a reduced volume gave colorless needles (410 mg, 69%), which did not show a definite m.p. TLC (silica gel, *K*): R_f 0.45. UV (MeOH): 221 (29200), 266 (6800), 295 (7400). ^{1}H-NMR ($(D_6)Me_2SO$): 2.29 (*'quint.'*, *J* = 6.1, H_b–C(2')); 2.57 (*'quint.'*, *J* = 6.6, H_a–C(2')); 3.54 (*m*, CH_2(5')); 3.87 (*m*, H–C(4')); 4.38 (*m*, H–C(3')); 5.01 (*t*, *J* = 5.6, OH–C(5')); 5.26 (*d*, *J* = 4.3, OH–C(3')); 6.16 (*'t'*, *J* = 5.9, H–C(1')); 6.44 (*s*, NH_2); 8.45 (*s*, H–C(3)). Anal. calc. for $C_{11}H_{15}N_5O_6$: C 46.97, H 5.38, N 24.80; found: C 46.88, H 5.50, N 24.74.

*6-Amino-1-(2'-deoxy-β-*D*-*erythro*-pentofuranosyl)-1*H*-pyrazolo[3,4-*d*]pyrimidin-4-(5*H*)-one* (**1**). A soln. of **15** (1.0 g, 3.6 mmol) in 2N NaOH (100 ml) was stored for 24 h at r.t. then neutralized with 96% AcOH under cooling, and concentrated. The resulting precipitate was filtered off and recrystallized from H_2O yielding colorless needles (894 mg, 93%) with m.p. 196° (dec.). TLC (silica gel, *K*): R_f 0.45. UV (MeOH): 354 (14200). ^{1}H-NMR ($(D_6)Me_2SO$): 2.16 (*ddd*, *J* = 4.0, 6.6, 12.9, H_b–C(2')); 2.69 (*'quint.'*, *J* = 6.4, H_a–C(2')); 3.41 (*m*, CH_2(5')); 3.78 (*m*, H–C(4')); 4.74 (*t*, *J* = 6.4, OH–C(5')); 5.22 (*d*, *J* = 4.2, OH–C(3')); 6.31 (*'t'*, *J* = 6.4, H–C(1')); 6.69 (*s*, NH_2); 7.83 (*s*, H–C(3)); 10.61 (*s*, H–C(5)). Anal. calc. for $C_{10}H_{13}N_5O_4$: C 44.94, H 4.90, N 26.21; found: C 44.81, H 5.00, N 26.11.

*6-Amino-2-(2'-deoxy-β-*D*-*erythro*-pentofuranosyl)-2*H*-pyrazolo[3,4-*d*]pyrimidin-4(5*H*)-one* (**2**). A suspension of **16** (430 mg, 1.53 mmol) in 2N aq. KOH (20 ml) was stored 2 days at r.t. The soln. was neutralized with 1N aq. AcOH, the resulting precipitate collected and combined with a second crop obtained from a reduced volume. Recrystallization from H_2O yielded colorless needles (270 mg, 66%), which did not melt up to 300°. TLC (silica gel, *M*): R_f 0.26. UV (MeOH): 224 (27400), 240 (sh, 6900), 279 (6600). ^{1}H-NMR ($(D_6)Me_2SO$): 2.25 (*'quint.'*, *J* = 6.0, H_b–C(2')); 2.54 (*'quint.'*, *J* = 6.4, H_a–C(2')); 3.52 (*m*, CH_2(5')); 3.84 (*m*, H–C(4')); 4.35 (*m*, H–C(3')); 4.96 (*t*, *J* = 5.6, OH–C(5')); 5.24 (*d*, *J* = 4.1, OH–C(3')); 6.08 (*'t'*, *J* = 6.0, H–C(1')); (*s*, NH_2); 8.45 (*s*, H–C(3)). Anal. calc. for $C_{10}H_{13}N_5O_4$: C 44.94, H 4.90, N 26.21; found: C 45.00, H 4.92, N 26.27.

Phase-Transfer Glycosylation of **7a** *with* **10**. It was carried out as for **11a**/**12a** without catalyst using **7a** (600 mg, 3.5 mmol) and **10** (1800 mg, 4.6 mmol). After chromatography 3 zones were separated in the indicated order. With solvent *A*, colorless *6-amino-4-chloro-1-(2'deoxy-3',5'-di-*O*-(*p*-toluoyl)-β-*D*-erythro-pentofuranosyl)-1*H*-pyrazolo[3,4-*d*]pyrimidine* (**11b**) (725 mg, 39%) was obtained. Crystallization from MeOH afforded colorless needles, m.p. 125–128°. TLC (*F*): R_f 0.54. UV (MeOH): 233 (55600), 272 (42000), 283 (4300), 305 (6500). ^{1}H-NMR ($CDCl_3$): 2.37, 2.40 (2*s*, CH_3); 2.70 (*m*, H_b–C(2')); 3.21 (*'quint.'*, *J* = 6.6, H_a–C(2')); 4.46 (*m*, H–C(4'), H–C(5')); 4.80 (*m*, H–C(3')); 6.60 (*'t'*, *J* = 6.2, H–C(1')); 7.31, 7.36 (2*d*, *J* = 8.1, 4 arom. H); 7.44 (*s*, NH_2); 7.87, 7.92 (2*d*, *J* = 8.1, 4 arom. H); 8.11 (*s*, H–C(3)). Anal. calc. for $C_{26}H_{24}ClN_5O_5$: C 59.83, H 4.63, Cl 6.79, N 13.42; found: C 59.87, H 4.65, Cl 6.68, N 13.36. Solvent *D* yielded pale yellow amorphous *6-amino-4-chloro-2-(2'-deoxy-3',5'-di-*O*-(*p*-toluoyl)-β-*D*-erythro-pentofuranosyl)-2*H*-pyrazolo[3,4-*d*]pyrimidine* (**12b**) (715 mg, 39%). Crystallization from MeOH afforded pale yellow needles, m.p. 141° (dec.). TLC (*F*): R_f 0.14. UV (MeOH): 230 (41700), 278 (7900), 327 (4900). ^{1}H-NMR ($CDCl_3$): 2.37, 2.40 (2*s*, CH_3); 2.78 (*'quint.'*, *J* = 6.4, H_b–C(2')); 3.05 (*'quint.'*, *J* = 6.4, H_a–C(2')); 4.44, 4.57 (2*m*, H–C(4'), H–C(5')); 5.79 (*m*, H–C(3')); 6.41 (*'t'*, *J* = 5.7, H–C(1')); 7.29–7.92 (4*d*, *J* = 8.1, 8 arom. H); 8.31 (*s*, NH_2); 8.79 (*s*, H–C(3)). Anal. calc. for $C_{26}H_{24}ClN_5O_5$: C 59.83, H 4.63, Cl 6.79, N 13.42; found: C 59.97, H 4.81, Cl 6.63, N 13.25.

*6-Amino-1-(2'-deoxy-3',5'-di-O-(p-toluoyl)-β-*D-erythro-*pentofuranosyl)-1*H-*pyrazolo[3,4-*d*]pyrimidin-4-(5*H*)-thione* (**17b**). A soln. of **11b** (520 mg, 1 mmol) in MeOH (35 ml) was refluxed in the presence of thiourea (150 mg, 2 mmol) for 1 h. On cooling (ice bath), colorless needles (460 mg, 89%) with m.p. 174–176° precipitated. TLC (*H*): R_f 0.31. UV (MeOH): 238 (46900), 272 (sh, 9300), 336 (21700). ^{1}H-NMR ($(D_6)Me_2SO$): 2.37, 2.39 (2*s*, CH_3); 2.69 (*'quint.', J* = 6.2, H_b–C(2')); 3.15 (*'quint.', J* = 6.9, H_a–C(2')); 4.44 (*m*, H–C(4'), H–C(5')); 5.78 (*m*, H–C(3')); 6.47 (*'t', J* = 6.1, H–C(1')); 7.04 (*s*, NH_2); 7.31–7.91 (4*d*, *J* = 8.4, 8 arom. H); 8.01 (*s*, H–C(3)); 12.09 (*s*, H–C(5)). Anal. calc. for $C_{26}H_{25}N_5O_5S$: C 60.10, H 4.85, N 13.48, S 6.17; found: C 60.33, H 4.93, N 13.45, S 6.25.

*6-Amino-1-(2'-deoxy-β-*D-erythro-*pentofuranosyl)-1*H-*pyrazolo[3,4-*d*]pyrimidin-4(5*H*)-thione* (**17a**). A mixture of **17b** (490 mg, 0.94 mmol) in 10 mM MeONa/MeOH (50 ml) was kept for 2 h at r.t. Then, the soln. was neutralized with AcOH and adsorbed onto silica gel (5 g, coevaporation *in vacuo*). The silica gel was suspended in $CHCl_3$ and loaded on the top of a silica-gel column (10 × 4 cm). Elution with solvent *K* and evaporation to a reduced volume yielded colorless needles (180 mg, 67%). M.p. 174–176° (dec.). TLC (*K*): R_f 0.55. UV (MeOH): 238 (15700), 272 (6700), 336 (21200). ^{1}H-NMR ($(D_6)Me_2SO$): 2.18 (*ddd*, *J* = 4.2, 6.6, 13.0, H_b–C(2')); 2.71 (*'quint.', J* = 6.3, H_a–C(2')); 3.41 (*m*, CH_2(5')); 3.77 (*m*, H–C(4')); 4.38 (*m*, H–C(3')); 4.71 (*s*, OH–C(5')); 5.22 (*s*, OH–C(3')); 6.28 (*'t', J* = 6.3, H–C(1')); 6.99 (*s*, NH_2); 7.96 (*s*, H–C(3)); 12.04 (*s*, H–C(5)). Anal. calc. for $C_{10}H_{13}N_5O_3S$: C 42.40, H 4.63, N 24.72, S 11.32; found: C 42.28, H 4.63, N 24.67, S 11.24.

*6-Amino-2-(2'-deoxy-β-*D-erythro-*pentofuranosyl)-2*H-*pyrazolo[3,4-*d*]pyrimidin-4(5*H*)-thione* (**18a**). A soln. of **12b** (630 mg, 1.2 mmol) and thiourea (165 mg, 2.2 mmol) was refluxed for 1 h and then cooled in an ice bath. The precipitate was collected by filtration and was then dissolved in 1M MeONa/MeOH (7 ml). The soln. was allowed to stand for 30 min at r.t. Neutralization with AcOH formed a precipitate. After addition of H_2O (30 ml), the solvent was evaporated. The solid residue was recrystallized form H_2O to give pale yellow needles (820 mg, 62%), m.p. 178–180° (dec.). TLC (*K*): R_f 0.45. UV (MeOH): 240 (27200), 282 (2300), 298 (2000), 346 (8200). ^{1}H-NMR ($(D_6)Me_2SO$): 2.27 (*'quint.', J* = 6.1, H_b–C(2')); 2.53 (*'quint.', J* = 6.6, H_a–C(2')); *ca.* 3.50 (*m*, CH_2(5')); 3.86 (*m*, H–C(4')); 4.36 (*m*, H–C(3')); 5.01 (*s*, OH–C(5')); 5.35 (*s*, OH–C(3')); 6.13 (*'t', J* = 6.0, H–C(1')); 6.49 (*s*, NH_2); 8.69 (*s*, H–C(3)). Anal. calc. for $C_{10}H_{13}N_5O_3S$: C 42.40, H 4.63, N 24.72, S 11.32; found: C 42.50, H 4.63, N 24.62, S 11.25.

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