Modification of Guanine with Photolabile *N*-Hydroxypyridine-2(1*H*)-thione: Monomer Synthesis, Oligonucleotide Elaboration, and Photochemical Studies

by **Despoina Vrantza**[a]), **Panagiotis Kaloudis**[a]), **Leondios Leondiadis**[a])[1]), **Thanasis Gimisis***[a]), **Georgios C. Vougioukalakis**[b]), **Michael Orfanopoulos**[b]), **Didier Gasparutto**[c]), **Jean Cadet**[c]), **Susana Encinas**[d]), **Cecilia Paris**[d]), and **Miguel A. Miranda***[d])

[a]) Organic Chemistry Laboratory, Department of Chemistry, University of Athens, 15771 Athens, Greece (phone: +30-2107-274-928; fax: +30-2107-274-761; e-mail: gimisis@chem.uoa.gr)

[b]) Department of Chemistry, University of Crete, Heraklion, Greece

[c]) Laboratoire 'Lésions des Acides Nucléiques', SCIB/LAN, CEA, Grenoble, France

[d]) Instituto de Tecnología Química, Universidad Politécnica de Valencia, Valencia, Spain

Dedicated to the memory of Professor *Hanns Fischer*

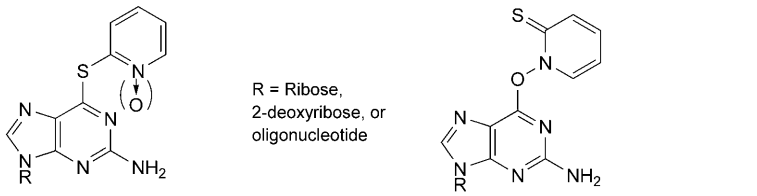
The photochemistry of *N*-hydroxypyridine-2(1*H*)-thione (NHPT), inserted as a photolabile modifier at the 6-position of 2′-deoxyguanosine or guanosine, has been evaluated. In particular, 6-[(1-oxidopyridin-2-yl)sulfanyl]- (**1a**) and 6-[(pyridin-2-yl)sulfanyl]-2′,6-dideoxyguanosine (**2a**), novel photolabile derivatives of the natural nucleosides, were synthesized and characterized. The observed photolysis products of **1a** in organic solvents could only be rationalized by assuming a rapid equilibrium with the corresponding 6-[(2-thioxopyridin-1(2*H*)-yl)oxy] analogue **3a** (*Scheme 5*). Transient spectroscopy of **1a** indicated a strong triplet-excited state suitable for triplet → triplet energy transfer or singlet-oxygen generation. The NHPT function was stable enough for (slightly modified) automated solid-phase oligonucleotide synthesis. The utility of the above compounds is discussed, as well as their potential use in photosensitization of reactive oxygen species in DNA.

1. Introduction. – The oxidation chemistry of the guanine base in DNA is rich [1], and numerous product lesions need to be examined for their mutagenic potential, with implications in the mechanisms involved in many human conditions including aging and cancer [2]. The generation of synthetic lesions for the study of their mutagenicity profile can be accomplished through two different lines of research. In the first approach, stable lesions are independently synthesized and subsequently inserted into oligonucleotides. Alternatively, stable or unstable lesions can be generated in oligonucleotides by inserting a stable modification capable of chemoselectively generating the lesion under study. Light is the ideal 'reagent' for such *in situ* transformations, and light-sensitive *tert*-butyl ketones have already been inserted in various positions of the sugar moiety for the generation of reactive sugar-radical intermediates [3–6]. Purine-base radicals, particularly guanine, possessing the lowest reduction potential [7], as well as pyrimidine triplet-excited states, leading to pyrimidine dimers [8], are also important intermediates in DNA chemistry. To date, the generation of these reactive species in

[1]) Present address: Mass Spectrometry & Dioxin Analysis Laboratory, NCSR 'Demokritos', 15310 Athens, Greece.

isolated monomers or within DNA has been effected in the presence of one-electron oxidants and photosensitizers.

The purpose of this study was to evaluate *N*-hydroxypyridine-2(1*H*)-thione (NHPT) as a photolabile modifier introduced as (1-oxidopyridin-2-yl)sulfanyl moiety at the 6-position of 2′-deoxyguanosine. The synthetically targeted compounds are shown in *Fig. 1*. The amphiphilicity and rich photochemistry of this group [9] could provide a handle for the generation of reactive intermediates such as triplets, radicals, and reactive-oxygen species in the vicinity of the DNA double helix. The photochemistry of both 'pyridine-2-thioneoxycarbonyl' (PTOC) imidate esters [10] and pyridine-*N*-oxides [11] is well-established, and the particular case of photo-deoxygenation has been reported to generate reactive species [12] that could be used in the study of oxidative DNA damage.



1 6-[(1-Oxidopyridin-2-yl)sulfanyl]
2 6-[(Pyridin-2-yl)sulfanyl]
3 6-[(2-Thioxopyridin-1(2*H*)-yl)oxy]

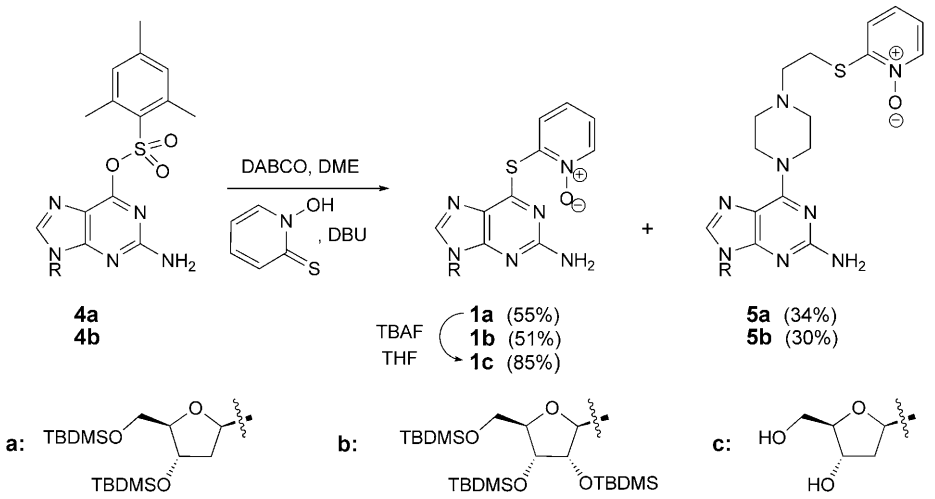
Fig. 1. *Structures of target compounds*

Compounds **1**, **2**, and **3** have not been previously reported. The structure of **3** could be considered reminiscent of PTOC imidate esters, the synthesis and photochemistry of which have been extensively studied by *Newcomb* and co-workers [10][13]. The major difference of the proposed derivative was the aromaticity of the imidate moiety and, as such, these structures were unknown. A small number of heteroaromatic '2-thiopyridine-*N*-oxides' (= pyridine-2-thiol 1-oxides), structurally related to compound **1**, with the S-function *ortho* (or α) to a heterocyclic N-atom, has been reported [14–19]. Although the related products exhibited interesting properties, mainly herbicidal, fungicidal, antimicrobial, and even anticancer actions, the photochemical properties of these compounds have not been studied. In all the above cases, the method of choice for the insertion of the (1-oxidopyridin-2-yl)sulfanyl function α to a heterocyclic N-atom was the reaction of NHPT with the corresponding chloroimidate.

On the other hand, it has been known [20] that 6-arylsulfonyl derivatives of 2′-deoxyguanosine can be used for the functionalization of the 6-position with various nucleophiles. In a recent improvement of this methodology, activation of 6-mesityl-2′-deoxyguanosine derivatives with DABCO (= 1,4-diazabicyclo[2.2.2]octane) allowed for the addition of various O- and N-nucleophiles [21].

2. Results and Discussion. – 2.1. *Monomer Synthesis.* The synthesis of the 6-deoxy-6-[(1-oxidopyridin-2-yl)sulfanyl]- and of the corresponding 2′,6-dideoxy-guanosine deriv-

Scheme 1



DABCO = 1,4-diazabicyclo[2.2.2]octane, DME = 1,2-dimethoxyethane
DBU = 1,8-diazabicyclo[5.4.0]undec-7-ene, TBDMS = ^{t}Bu(Me)$_2$Si

atives **1** is presented in *Scheme 1*[2]). The O^6-[(2,4,6-trimethylphenyl)sulfonyl] derivatives **4** were synthesized according to the above-mentioned methodology for the 2′-deoxy analogue [20][22]. By introducing NHPT through DABCO-induced activation [21], compounds **1a** and **1b** were formed in 55 and 51% yield, respectively, together with the readily separable nucleoside side products **5a** (34%) and **5b** (30%), respectively.

In accordance with the aforementioned literature data, the spectroscopic data of the major reaction products **1a** or **1b** were in agreement with the proposed structure. The longest-wavelength absorption (λ_{max}) in the UV/VIS spectrum was centered at 325 nm. At the same time, there was no ^{13}C-NMR absorption at $\delta(C) > 160$ ppm, indicating the lack of a thiocarbonyl function. The isolated compounds were off-white solids that were relatively stable to visible light, although they slowly developed a grey coloration while stored at low temperature. Product **1a** was readily deprotected in the presence of tetrabutylammonium fluoride (TBAF) in THF, and the water-soluble derivative **1c** was isolated in good yield. It is interesting that the NHPT function, although stable under these conditions, was readily replaced by MeOH when exposed to NH_4F in boiling MeOH.

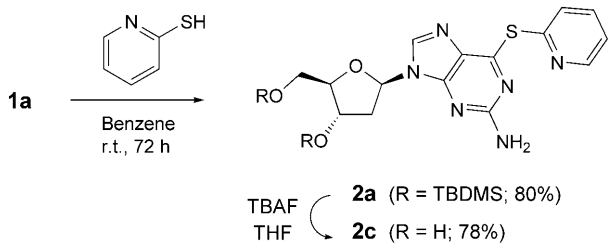
A significant effort went into the characterization of the side product **5a**, as it was possible that it had arisen from O-nucleophilic attack of the ambident NHPT nucleophile. However, ESI-MS as well as ^{1}H- and ^{13}C-NMR spectroscopy indicated that DABCO is covalently bonded in **5a**. Specifically, the $[M+H]^+$ peak at m/z 717.1 and fragments at 373.3 ([base + DABCO + NHPT + H]$^+$) and 246.2 ([base + DABCO]$^+$)

[2]) For systematic compound names, see the *Exper. Part.*

were observed. At the same time, one of the three ethylene groups was differentiated by both ^{1}H- and ^{13}C-NMR, one methylene appearing at high field (δ(C) 28.3). Product **5a** was stable under the reaction conditions and did not transform to **1a** when it was re-subjected to the same reaction conditions. The above evidence led to the conclusion that **5a,b** result from S-nucleophilic attack of NHPT at the α-C-atom of the positively charged DABCO–purine intermediate, followed by an S_N2 addition and opening of the ethylene DABCO bridge. Such S_N2 displacements have not been previously observed when DABCO was used as an activator of the C(6) position of purines [23][24], but have been reported for analogous reactions activated by Me_3N [25]. These findings imply that S-nucleophiles induce this side reaction in the trialkylamine-mediated activation of the (trimethylphenyl)sulfonyl leaving group in related reactions.

In order to avoid the above side reaction in the synthesis of the 6-(pyridin-2-ylsulfanyl) derivatives **2** (*Scheme 2*), these compounds were synthesized directly by treating **1a** with '2-thiopyridine' (=pyridine-2(1*H*)-thione) in benzene. The substitution product **2a** was finally transformed to the water-soluble derivative **4c** by short exposure to TBAF in THF.

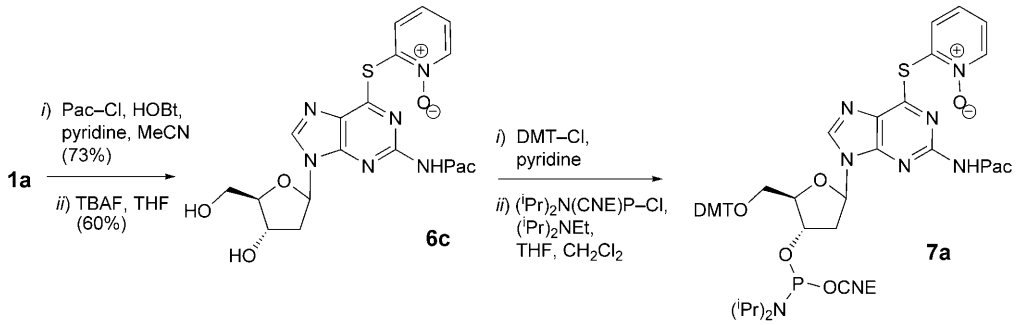
Scheme 2



2.2. *Monomer Stability.* Insertion of the above modified guanosines in synthetic oligonucleotides requires that they are stable under the standard condition used in automated oligonucleotide synthesis on solid support. Compound **1c** was indefinitely stable in D_2O at neutral pH, and its solutions showed no appreciable change (in spectroscopic terms) for at least two months when stored at room temperature. It was also stable at 80° in the dark for at least 24 h, as well as in 80% AcOH at room temperature. However, compound **1c** was less stable in 0.1M I_2 in THF (>90% recovery after 1 h), and similarly sensitive to 25% aq. NH_4OH solution (>90% recovery after 2.5 h). Finally, it decomposed relatively fast under more-basic conditions. Therefore, this type of chemical modification could be inserted in synthetic oligonucleotides, but certain precautions had to be taken, especially with regards to base treatment.

2.3. *Phosphoramidite Synthesis.* For the above reasons, we proceeded in the preparation of the corresponding phosphoramidite (*Scheme 3*) by applying phenoxyacetic (Pac) protection for the guanine 2-amino function [26][27]. Pac Protection of the 2-amino function of **1a**, followed by removal of the ^{t}Bu(Me_2)Si (TBDMS) ethers of the sugar functions gave **6c**. The latter was then 'dimethoxytritylated' at 5′-position and 'phosphitylated' at 3′-position to provide the phosphoramidite **7a**, suitable for oligonucleotide synthesis.

Scheme 3



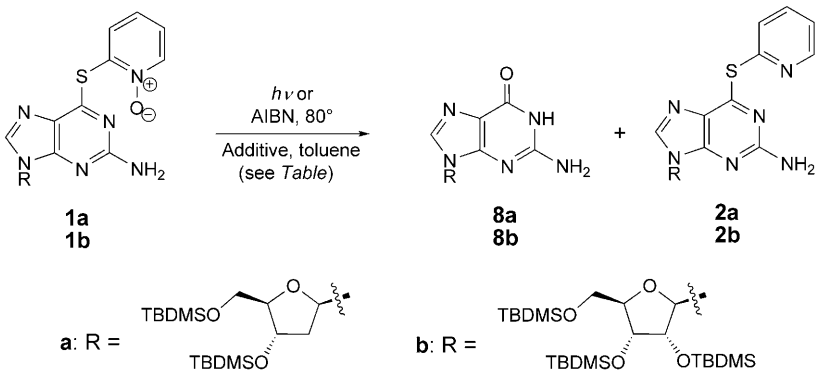
Pac = Phenoxyacetyl, HOBt = 1*H*-benzotriazol, TBAF = Bu_4NF
DMT = dimethoxytrityl, CNE = 2-cyanoethyl

2.4. *Solid-Phase Synthesis of Oligonucleotides.* The synthesis of all oligonucleotides (3-, 5-, 9-, and 15-mers) was performed on a 1-μmol scale using phosphoramidite chemistry on an *Applied Biosystems 392* automated nucleic-acids synthesizer. The Pac-protection system for the nucleobase amino functions was used with regard to the high lability of the modified nucleoside under alkaline deprotection conditions. The synthesis was performed on controlled-pore-glass (CPG) support following the standard protocol, with the exception of three modifications: first, the duration of coupling of the modified nucleoside phosphoramidite was extended to 5 min instead of 30 s for normal nucleosides. Under the latter conditions, an adduct coupling efficiency up to 80–90% was achieved. Moreover, the capping step was carried out with a solution of phenoxyacetic anhydride ($(Pac)_2O$) in THF to prevent possible transacylation reactions. The oxidation step was subsequently performed with a 20-mM I_2 solution to prevent possible oxidizing degradation of the modified nucleoside during solid-phase assembly. Upon completion of the synthesis, the base-labile protecting groups of the oligodeoxyribonucleotides were removed by treatment with concentrated aqueous NH_3 (32%) at room temperature for 4 h. Solvents were removed by evaporation under vacuum. The crude oligomers were then purified by reverse-phase HPLC.

2.5. *Monomer Photolysis in Organic Solvents.* Irradiation of a 10-mM solution of **1a** in anhydrous toluene under Ar atmosphere in the presence of an excess of *tert*-butylthiol (20 equiv.) at 20° led to complete disappearance of the starting material within 20 min of photolysis. There was a single nucleosidic product isolated in 95% yield, which was identified as the TBDMS-protected 2′-deoxyguanosine **8a** (*Scheme 4*). Two other nonnucleosidic side products were identified as 2,2′-dipyridinyl disulfide (=2,2′-disulfanediyldipyridine; **9**), by comparison with a commercial sample, and as '*tert*-butyl-2-pyridinyldisulfide' (=2-[(*tert*-butyl)disulfanyl]pyridine) by comparison with literature data [28].

Similarly, irradiation of **1b** gave a near quantitative yield of protected guanosine **8b** together with the above two side products. When the photolysis of **1a** was performed in the presence of less-efficient hydrogen donors compared to ^{t}BuSH, such as Bu_3SnH or $(Me_3Si)_3SiH$, the same nucleoside product **8a** was observed in slightly lower yield. When no hydrogen donor was used, **8a** was isolated in 60% yield, together with a second nucleoside component in 15% yield. ESI-MS indicated a loss of 16 amu in the new

Scheme 4



AIBN = 2,2'-Azobis(isobutyronitrile), TBDMS = ^{t}Bu(Me)$_2$Si

product when compared with **1a**. The new product was identified as **2a** by comparison with an authentic sample (*Scheme 4*). The same product was also observed (together with **8a**) when other additives, sources of Sn-radical intermediates (*e.g.*, ditin and allyltin compounds) were used (*Table*). In all cases, both products, **8a** and **2a**, were isolated in comparable amounts, with the highest yield of **2a** obtained by initiation at 80° in the presence of allyl(tributyl)tin and AIBN (=2,2′-azobis[2-methylpropanenitrile]).

Table. *Product Distribution upon Photolysis or Radical-Induced Cleavage of* **1a** *in the Presence of Various Additives* (for details, see *Exper. Part*)

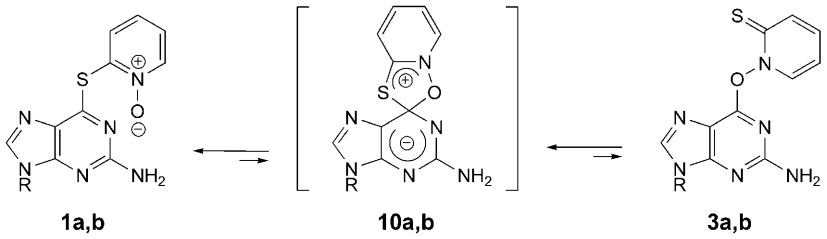
Entry	Additive	Initiation	**8a** [%]	**2a** [%]
1	None	*hv*	60	15
2	^{t}BuSH	*hv*	95	–
3	^{t}BuSH	AIBN[a]), 80°	95	–
4	Bu_3SnH	*hv*	90	–
5	$(Me_3Si)_3SiH$	*hv*	85	5
6	$Me_3SnSnMe_3$	*hv*	47	40
7	$Bu_3SnSnBu_3$	*hv*	45	45
8	CH_2=$CHCH_2SnBu_3$	AIBN, 80°	30	65

[a]) AIBN = 2,2′-Azobis[2-methylpropanenitrile].

The above results are remarkable, since it is the first time that pyridine-*N*-oxides are reported to behave like pyridine-2-thiones quantitatively providing the corresponding reduction product in the presence of hydrogen donors. The observed products can only be rationalized by assuming an equilibrium between 2′,6-dideoxy-6-[(1-oxidopyridin-2-yl)sulfanyl]guanosine (**1a**) and 2′-deoxy-6-[(2-thioxopyridin-1(2*H*)-yl)oxy]guanosine (**3a**), as outlined in *Scheme 5*. This would correspond to a [1,4]-sigmatropic rearrangement of a pyridine-*N*-oxide to a pyridine-2-thione. The reverse [1,4]-sigmatropic rearrangement was reported by *Hay* and *Beckwith* [29], and more recently also by *Hartung et al.* [30] for related *O*-alkyl pyridine-2-thiones to be an irreversible process rendering

photolytically inactive pyridine-*N*-oxides. This is the first report that this rearrangement appears to be reversible and that pyridine-*N*-oxides behave photolytically as pyridine-2-thiones. We propose that this reversibility is due to the resonance stabilization of the *ipso* intermediate **10** by the aromatic purine ring (*Scheme 5*).

Scheme 5. *Proposed Equilibrium between Pyridine*-N-*oxides* **1** *and pyridine-2-thiones* **3**



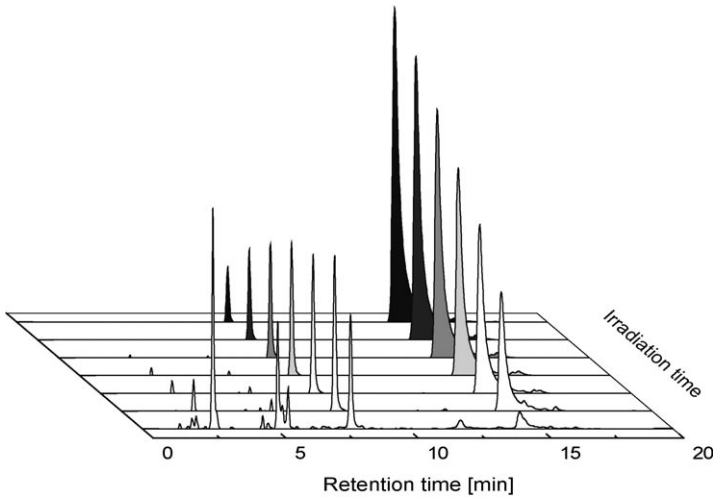
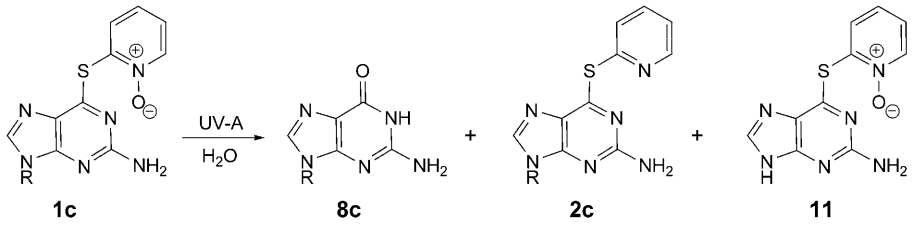


Fig. 2. *HPLC Profile for the photolysis of* **1c** *in* H_2O

2.6. *Photolysis in Aqueous Solutions.* The Photolysis of **1c** in H_2O was followed by HPLC (*Fig. 2*). The starting HPLC signal disappeared at least one order of magnitude more slowly than in the case of **1a** in organic solvents. The only product observed in small conversion was 2′-deoxyguanosine (**8c**; *Scheme 6*). At longer times of continuous irradiation, 2′,6-dideoxy-6-[(pyridin-2-yl)sulfanyl]guanosine (**2c**) appeared, together with a new product characterized by LC/MS ($[M+H^+]$ signal at *m*/*z* 261.3) as the product of *N*-glycosidic bond cleavage of **1c**. This product (**11**) did not appear, when aqueous phosphate buffer (pH 7.2) was used; as expected in this case, the consumption of the starting material was even slower than in the previous experiment. More importantly, there was no ^{18}O incorporation into **8c**, which was isolated and characterized by ESI-MS after 5 h of irradiation of **1c** in $H_2{}^{18}O$ solution. This indicated that, as observed in the absence of irradiation (*vide supra*), no hydrolysis takes place under these conditions. There were no differences observed in the product distribution in the presence or absence of molecular oxygen (O_2).

Scheme 6



As observed in the 3D HPLC plot shown in *Fig. 2*, there is a built up of **8c** coupled with the disappearance of **1c** at lower than 40% conversion. Eventually, **8c** reaches a plateau and starts decomposing concurrently with the further consumption of **1c** and the formation of both **2c** and a significant amount of the acid-catalyzed product of glycosidic bond cleavage (**11**), after extensive continuous photolysis.

Preliminary photolysis experiments were also performed on the prepared oligonucleotides with short UV-C treatment, followed by HPLC and ESI-MS analyses. Again, in the case of oligonucleotides, reversion to the unmodified oligonucleotide was the major event observed. A few other degradation and rearrangement products were also detected. The identification and characterization of these products is currently in progress.

2.7. *Comparison of Photoreaction Efficiency under Different Conditions.* The course of the photolysis was monitored by following the decrease of the UV/VIS absorption band at λ_{max} 325 nm. The data are shown in *Fig. 3*. Approximate values of the photolysis quantum yields ϕ can be estimated from the slopes of the straight lines obtained when the absorbance at 325 nm is plotted against irradiation time, after correction for the relative number of absorbed photons. In agreement with the product studies reported above, the value for H_2O ($\phi = 0.015$) was clearly lower than in MeCN ($\phi = 0.03$). Moreover, the reaction efficiency was duplicated ($\phi = 0.07$) when a thiol was present in the organic medium. A solution of benzophenone in i-PrOH was used as actinometer, as its photoreduction quantum yield is well-established ($\phi = 1.96$) [31].

2.8. *Transient Spectroscopy.* Laser flash photolysis of **1** and **2** was performed upon excitation at 308 nm in MeCN/H_2O 4 : 1 under an anaerobic atmosphere. In the case of compounds **1**, a weak signal was generated, with a maximum at *ca.* 420 nm; the same transient was also formed when **1** was photolyzed in MeOH, MeCN, or benzene. By contrast, compounds **2** gave rise to the same type of transient, albeit with a much higher absorbance. The signal displayed two maxima at *ca.* 390 and 435 nm (*Fig. 4,a*), and its lifetime was 10 μs. In all cases (for **1** and **2**), the transient species were quenched by O_2 (*Fig. 4,b*), with a quenching rate k_q of 2×10^9 s^{-1} M^{-1}.

To assign the signals obtained for **1** and **2**, different model compounds were also photolyzed (*Scheme 7*). Thus, 2,2′-dithiodipyridine (**9**) was irradiated under the same conditions, giving rise to the known pyridin-2-ylsulfanyl radical [9], whose spectroscopic features (λ_{max} 480 nm, no quenching by O_2) were very different from those of the transients obtained for **1** or **2**. Likewise, photolysis of the known '2,2′-dithiodi(pyr-

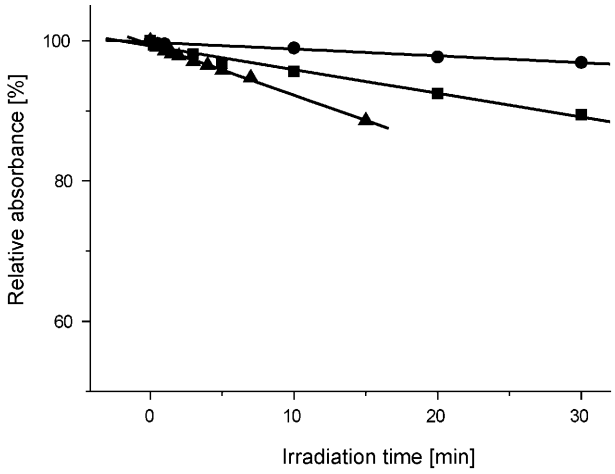


Fig. 3. *Plot of the relative UV/VIS absorbance* (in %) *at 325 nm* vs. *irradiation time for* **1c** *in water* (●), *and for* **1a** *in MeCN in the absence* (■) *and presence* (▲) *of dodecane-1-thiol* (20 equiv.)

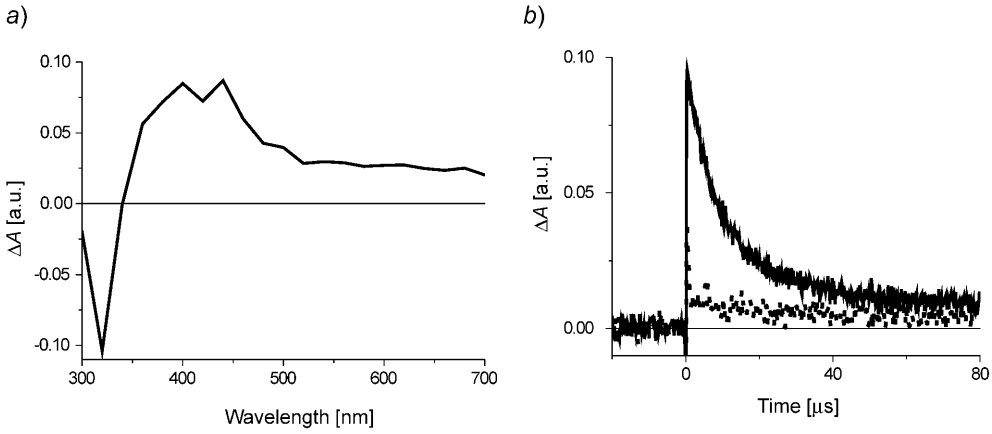


Fig. 4. a) *Change in UV/VIS absorbance of a solution of* **2** *in anaerobic MeCN/H_2O 1 μs after the laser pulse.* A similar transient spectrum, though much weaker, was obtained for **1**. b) *Decay of the UV/VIS signal at 420 nm of* **2** *under anaerobic atmosphere* (N_2) (solid curve) *and under aerobic conditions* (O_2) (dashed curve).

idine-*N*-oxide)' (=2,2′-disulfanediylbis(pyridine) 1,1′-dioxide; **12**) led to the (1-oxidopyridin-2-yl)sulfanyl radical [9], which also showed a significantly different UV/VIS spectrum (λ_{max} 340 nm, no quenching by O_2).

Hence, the transients observed for compounds **1** and **2** were very similar and were tentatively assigned to a triplet-excited state. Confirmation of this assignment was achieved by using well-known triplet quenchers such as biphenyl and naphthalene [32]. Only the latter was found to be an efficient quencher of the transient, though, with a rate constant of 2×10^9 M^{-1} s^{-1}. This value was estimated from the *Stern–Volmer* plot

Scheme 7. *Laser-Flash Photolysis of Model Compounds* **9** *and* **12**. The reactions were performed under anaerobic conditions in $MeCN/H_2O$ 4:1.

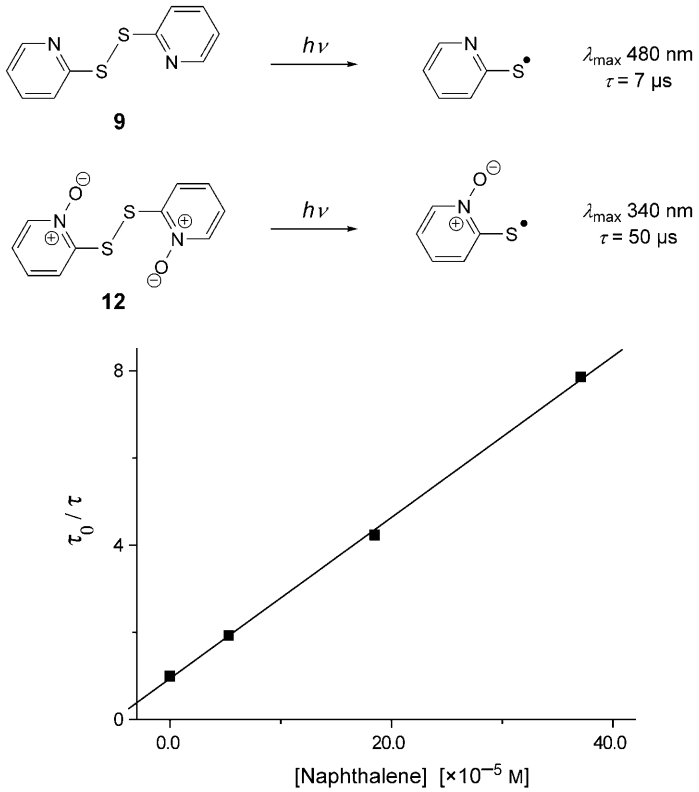


Fig. 5. Stern–Volmer *plot for the triplet-lifetime quenching of* **2** *in MeCN/H_2O 4:1 in the presence of increasing concentrations of naphthalene*

obtained by monitoring the triplet decay at 460 nm in the presence of increasing amounts of naphthalene (*Fig. 5*). As biphenyl was unable to act as a quencher, the triplet energy of **1** and **2** must be in the range of 63–65 kcal/mol.

Thus, the same type of transients are detected for compounds **1** and **2**. However, the observation that the triplet signal is much weaker in the case of **1** can be attributed to the occurrence of other photophysical processes that should be competing with intersystem crossing (ISC) to the triplet state. One such process might well be deoxygenation of the *N*-oxide moiety, with generation of atomic oxygen, as previously reported [12]. This would be in agreement with the formation of the deoxygenated products **2** upon prolonged photolysis of **1**.

It should be noted that no evidence for the 2′-deoxyguanosine neutral radical (the likely precursor of **8**) was obtained in these laser-flash-photolysis experiments. However, this is not surprising, as the quantum yield of the reaction leading to **8** is very low, since the photoactive species **3** is only a minor compound in the equilibrium shown in *Scheme 5*.

3. Conclusions. – We have synthesized and characterized novel photolabile 6-[(1-oxidopyridin-2-yl)sulfanyl]-substituted 6-deoxy- and 2′,6-dideoxyguanosines of type **1**. The observed photolysis products can only be rationalized by assuming a rapid equilibrium of these compounds with the corresponding 6-[(2-thioxopyridin-1(2*H*)-yl)oxy] analogues **3** (*Scheme 5*) in organic solvents. In the presence of hydrogen donors, a radical-chain mechanism can account for the fast formation of the products. In their absence, the photochemistry observed is mainly homolysis of the N–O bond present in both the prevalent pyridine-*N*-oxide and its reversible pyridine-2-thione isomer. Triplet formation was the primary photochemical event observed by transient spectroscopy. A higher absorbance was exhibited by compounds **2** when compared with compounds **1**. The energy of the triplet-excited state is suitable for triplet → triplet energy transfer or singlet-oxygen generation, but not high enough for the generation of thymine dimers within DNA.

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Experimental Part

General. For reactions conducted under inert atmosphere, flasks were flame-dried and purged with anh. Ar gas until cool. Thin-layer (TLC) and column chromatography (CC) were performed on silica gel F_{254} and silica gel *60* (SDS, 230–400 mesh; *ASTM*), resp. UV/VIS Spectra: *Varian Cary-50* apparatus; λ_{max} in nm (ε in M^{-1} cm^{-1}). IR Spectra: *Perkin-Elmer 841* or *1760X* FT-IR spectrometers, in $CHCl_3$ soln.; in cm^{-1}. ^1H- and ^{13}C-NMR Spectra: *Varian Mercury-200*, *Bruker MSL-300* or *AMX-500* spectrometers; δ in ppm rel. to residual solvent signals, *J* in Hz.

Mass Spectrometry. Electrospray-ionization mass-spectrometry (ESI-MS) analyses of the nucleoside derivatives, together with those of the modified oligonucleotides, were conducted on either a *Finnigan AQA* quadrupole, a *ThermoFinnigan LCQ Ion Trap*, or a *Finnigan LTQ-FT* apparatus; values in *m*/*z*. Samples were prepared at a concentration of 10 μM in MeCN/H_2O 1:1 (*v*/*v*) containing either 0.1% of HCOOH (pos. mode) or 1% of Et_3N (neg. mode), and introduced into the ESI source with a syringe pump at a flow rate of 10 μl/min. MALDI Mass spectra were obtained with a *Bruker Biflex* time-of-flight mass spectrometer equipped with a 337-nm N_2 laser and a pulsed-delay source extraction. The matrix was prepared by dissolving 3-hydroxypicolinic acid/picolinic acid 4:1 (*w*/*w*) in 50% aq. MeCN containing a small amount of *Dowex-50W* (*50X8-200*; *Sigma*) cation-exchange resin. Then, 1 μl of an aq. soln. of the sample was added to 1 μl of the dissolved matrix, and the resulting soln. was stirred. The sample was subsequently placed on top of the target plate and allowed to dry by itself. The mass spectra were calibrated with the aid of myoglobin (1 pmol/μl; *m*/*z* 16,952) using the same assay conditions as described for the oligonucleotides.

UV-A Irradiation of Monomers. Typically, monomer-photolysis experiments were performed with a variable-intensity Xe-arc lamp (1000 W; *ORIEL6269*) equipped with a UV-A filter. 10 mM Solns. were prepared and irradiated in *Pyrex* glass vials with a local light intensity of 4.5 mW/cm^2. Reactions in org. solvent (toluene) were followed by TLC and NMR spectroscopy, whereas those performed in aq. solns. were followed by RP-HPLC and LC coupled with ESI-MS.

Determination of Photolysis Quantum Yields. Solutions ($A_{350}=0.1$) of **1a** or **1c** in MeCN and in H_2O, resp., were irradiated at 350 nm with a low-pressure Hg lamp emitting mainly at 350 nm (*Gaussian* distribution) under N_2. The UV/VIS absorption at 325 nm was measured after different irradiation times in

the range 0–30 min. A soln. of benzophenone in i-PrOH was used as actinometer. In this case, the decrease of the C=O absorption at 254 nm was followed after the irradiation times. Conversions were kept below 10% throughout the measurements.

UV-C Irradiation of Precursor-Containing Modified Oligodeoxyribonucleotides. Typically, an aq. soln. (2 ml) of compound containing the precursor moiety (1 AU_{260}/ml) was exposed during 5 min to 16 black-light lamps (λ_{max} 260 nm) of a *Rayonet* photochemical reactor. The resulting mixture was analyzed on a *Hypersil ODS* column (5 μm, 250 mm × 4.6 mm i.d.) with a gradient of MeCN/10 mM triethylacetate ammonium (pH 7) at a flow rate of 1 ml/min (detection at 260 nm). The collected samples were analyzed by ESI-MS (neg. mode).

Nanosecond Laser-Flash Photolysis. For excitation at 308 nm, an excimer laser with a Xe/HCl/Ne mixture was used. The single pulses were *ca.* 17 ns in duration, and the energy was 200 mJ output at the source. For both systems, a pulsed *Oriel-Lo255* Xe lamp was employed as the detecting light source. The laser-flash-photolysis apparatus consisted of the pulsed laser, the Xe lamp, an *Oriel-77200* monochromator, and an *Oriel* photomultiplier-tube (PMT) system made up of a *77348* side-on PMT tube, a *70680* PMT housing, and a *70705* PMT power supply. The output signal from the oscilloscope (*Tektronix TDS-640A*) was transferred to a personal computer. Concentrations of **1** and **2** were *ca.* 2×10^{-5} M, which ensured an absorbance of *ca.* 0.4 in the laser cell at the excitation wavelengths. When different quenchers were added to the soln. of **2**, their concentrations oscillated from equimolar to 50 times for the case of biphenyl, and up to ten times for naphthalene.

*9-{2-Deoxy-3,5-bis-*O*-[(1,1-dimethylethyl)(dimethyl)silyl]-β-*D*-*erythro*-pentofuranosyl}-6-{[(2,4,6-trimethylphenyl)sulfonyl]oxy}-9*H*-purin-2-amine* (**4b**). Prepared according to [22] for the 2′-deoxy analogue. TLC (AcOEt/hexane 15:85): R_f 0.25. ^{1}H-NMR: (200 MHz, $CDCl_3$): −0.22, −0.04 (2*s*, 3 H, MeSi); 0.09, 0.12 (2*s*, 6 H, MeSi); 0.79, 0.92, 0.94 (3*s*, 9 H, ^{t}Bu); 2.30 (*s*, 3 H, Me); 2.75 (*s*, 6 H, 2 Me); 3.75 (*dd*, $J_{4'}$ = 2.6, $J_{5''}$ = 11.4, H–C(5″)); 3.95 (*dd*, $J_{4'}$ = 2.6, $J_{5''}$ = 11.4, H–C(5′)); 4.08 (*dd*, $J_{3'}$ = 5.0, $J_{5'}$ = 2.6, H–C(4′)); 4.25 (*dd*, $J_{2'}$ = 5.0, $J_{4'}$ = 5.0, H–C(3′)); 4.46 (*dd*, $J_{1'}$ = $J_{3'}$ = 5.0, H–C(2′)); 4.79 (br. *s*, 2 H, H–C(2)); 5.88 (*d*, $J_{2'}$ = 5.0, H–C(1′)); 6.97 (*s*, 2 arom. H); 8.06 (*s*, H–C(8)). ^{13}C-NMR (50 MHz, $CDCl_3$): −5.37, −5.06, −4.75, −4,64, 4.38 (MeSi); 17.9, 18.1, 18.5 (^{t}BuSi); 21.1 (Me); 22.8 (Me); 25.6, 25.8, 26.1 (^{t}BuSi); 62.4 (C(5′)); 71.8 (C(3′)); 76.0 (C(2′)); 85.3 (C(4′)); 87.9 (C(1)′); 117.7 (C(5)); 131.6 (2 arom. *m*-C); 132.3 (C(8)); 140.3 (2 arom. *o*-C); 140.4 (arom. *o*-C); 142.5 (CS); 143.8 (arom. *p*-C); 153.1 (C(6)); 155.8 (C(4)); 158.3 (C(2)).

*9-{2-Deoxy-3,5-bis-*O*-[(1,1-dimethylethyl)(dimethyl)silyl]-β-*D*-*erythro*-pentofuranosyl}-6-[(1-oxidopyridin-2-yl)sulfanyl]-9*H*-purin-2-amine* (**1a**) *and 9-[(2*R*,4*S*,5*R*)-4-{[(1,1-dimethylethyl)(dimethyl)silyl]oxy}-5-({[(1,1-dimethylethyl)(dimethyl)silyl]oxy}methyl)tetrahydrofuran-2-yl]-6-(4-{2-[(1-oxidopyridin-2-yl)sulfanyl]ethyl}piperazin-1-yl)-9*H*-purin-2-amine* (**5a**). A soln. of **4a** [22] (905 mg, 1.33 mmol), DABCO (299 mg, 2.67 mmol)[3]), 4-Å molecular sieves (427 mg), and *N*-hydroxypyridine-2(1*H*)-thione (NHPT; 850 mg, 6.68 mmol) in anh. 1,2-dimethoxyethane (DME; 14.5 ml) was stirred for 30 min under Ar gas at r.t. Then, DBU (0.62 ml, 4.14 mmol) was added, and the mixture was stirred at r.t. for an additional 24 h. The molecular sieves were filtered off, washed with AcOEt, and the filtrate was evaporated to dryness. The residue was purified by CC (SiO_2; 1.5–3.5% MeOH in $CHCl_3$) to afford **1a** (438 mg, 55%) as a yellowish solid, together with **5a** (324 mg, 34%) as a colorless solid.

Data of **1a**. TLC (CH_2Cl_2/MeOH 95:5): R_f 0.49. UV ($CHCl_3$): 281 (13,727), 325 (14,500). UV (MeCN): 281 (12,076), 324 (12,803). IR ($CHCl_3$): 1599, 1561, 1471, 1420, 1400, 1362, 1254, 1222, 1109, 1030, 940, 883, 778, 706. ^{1}H-NMR (500 MHz, $CDCl_3$): 0.06, 0.07 (2*s*, 3 H, MeSi); 0.10 (*s*, 6 H, MeSi); 0.89, 0.90 (2*s*, 9 H, ^{t}Bu); 2.30–2.42 (*m*, H–C(2′)); 2.51–2.63 (*m*, H–C(2″)); 3.74 (*dd*, $J_{5''}$ = 11.2, $J_{4'}$ = 3.1, H–C(5′)); 3.80 (*dd*, $J_{5'}$ = 11.2, $J_{4'}$ = 4.2, H–C(5″)); 3.98 (*dd*, $J_{3'}$ = 6.6, $J_{5'}$ = 3.1, H–C(4′)); 4.58 (*ddd*, $J_{4'}$ = 6.6, $J_{2'}$ = 3.3, H–C(3′)); 5.26 (*s*, 2 H, H–C(2)); 6.31 (*dd*, $J_{2'}$ = 6.9, $J_{2''}$ = 6.8, H–C(1′)); 7.15 (*dd*, 2 H, H–C(4,5) of SPy); 7.74 (*dd*, H–C(3) of SPy); 7.98 (*s*, H–C(8)); 8.29 (*dd*, H–C(6) of SPy). ^{13}C-NMR (125 MHz, $CDCl_3$): −5.60, −5.48, −4.89, −4.76 (MeSi); 17.8, 18.2 (^{t}Bu); 25.6, 25.8 (^{t}BuSi);

[3]) Dried by heating over P_2O_5 under vacuum at 56° for 2 h before use.

40.7 (C(2′)); 62.6 (C(5′)); 71.8 (C(3′)); 83.5 (C(1′)); 87.6 (C(4′)); 123.3 (C(5) of SPy); 124.8 (C(4) of SPy); 127.5 (C(5)); 128.3 (C(3) of SPy); 138.9 (C(6) of SPy); 139.7 (C(8)); 145.9 (C(4)); 152.1 (C(2)); 155.3 (SC of SPy); 159.2 (C(6)). HR-ESI-MS (pos.): 605.2749 ($[M+H]^+$, $C_{27}H_{45}N_6O_4SSi_2^+$; calc. 605.2756).

Data of **5a**. M.p. 162–163° (MeOH/H_2O). TLC ($CHCl_3$/MeOH 95:5): R_f 0.31. ^{1}H-NMR (200 MHz, $CDCl_3$): 0.06, 0.09 (2*s*, 6 H, MeSi); 0.90 (*s*, 18 H, ^{t}Bu); 1.66 (br. *s*, 1 H, SH); 2.26–2.37 (*m*, H–C(2′)); 2.48–2.55 (*m*, H–C(2″)); 2.61 (*dd*, *J* = 5.1, 4.6, 4 H, $N(CH_2)_2$); 2.79 (*dd*, *J* = 7.9, 6.3, NCH_2); 3.08 (*dd*, *J* = 7.7, 6.3, SCH_2) 3.67–3.82 (*m*, H–C(5′), H–C(5″)); 3.95 (*dd*, $J_{3'}$ = 6.9, $J_{5'}$ = 4.1, H–C(4′)); 4.24 (br. *s*, 4 H, $N^6(CH_2)_2$); 4.53–4.56 (*m*, H–C(3′)); 4.58 (*s*, 2 H, H–C(2)); 6.30 (*dd*, $J_{2'}$ = 7.1, $J_{2''}$ = 5.9, H–C(1′)); 7.04 (*dd*, *J* = 6.8, 6.4, H–C(5) of SPy); 7.05 (*dd*, *J* = 6.8, 3.0, H–C(3) of SPy); 7.20 (*d*, *J* = 3.0, H–C(4) SPy); 7.70 (*s*, H–C(8)); 8.26 (*d*, *J* = 6.4, H–C(6) of SPy). ^{13}C-NMR (125 MHz, $CDCl_3$): −5.3, −5.1, −4.6, −4.5 (MeSi); 18.2, 18.6, 26.0, 26.2 (^{t}BuSi); 28.3 (SCH_2); 40.8 (C(2′)); 44.9 ($N^6(CH_2)_2$); 53.3 ($N(CH_2)_2$); 56.4 (NCH_2); 63.1 (C(5′)); 72.3 (C(3′)); 83.4 (C(1′)); 87.8 (C(4′)); 115.4 (C(4)); 120.6 (C(5) of SPy); 121.7 (C(3) of SPy); 125.9 (C(4) of SPy); 134.3 (C(8)); 139.0 (C(6) of SPy); 152.6 (C(6)); 153.0 (C(2) of SPy); 154.2 (C(5)); 159.4 (C(2)).

6-[(1-Oxidopyridin-2-yl)sulfanyl]-9-{2,3,5-tris-O-*[(1,1-dimethylethyl)(dimethyl)silyl]-β*-D-*ribofuranosyl}-9*H-*purin-2-amine* (**1b**). Prepared in analogy to **1a**, and purified by CC (SiO_2; 1.5–3.5% MeOH in $CHCl_3$) to yield 60 mg (51%) of **1b**, together with 44 mg (30%) of **5b**, which was not further characterized.

Data of **1b**. Yellowish solid. TLC (SiO2; $CHCl_3$/MeOH 9:1): R_f 0.40. ^{1}H-NMR (200 MHz, $CDCl_3$): −0.02, 0.10, 0.11 (3*s*, 6 H, MeSi); 0.82, 0.92, 0.93 (3*s*, 9 H, *t*-Bu); 3.76 (*dd*, $J_{5''}$ = 11.3, $J_{4'}$ = 2.8, H–C(5′)); 3.96 (*dd*, $J_{5'}$ = 11.3, $J_{4'}$ = 3.5, H–C(5″)); 4.10 (*dd*, $J_{3'}$ = 4.8, $J_{5'}$ = 3.5, H–C(4′)); 4.27 (*dd*, $J_{4'}$ = 4.8, $J_{2'}$ = 4.3, H–C(3′)); 4.48 (*dd*, $J_{1'}$ = 4.7, $J_{3'}$ = 4.3, H–C(2′)); 5.92 (*d*, $J_{2'}$ = 4.7, H–C(1′)); 5.06 (br. *s*, H–C(2)); 7.19 (*dd*, H–C(4,5) of SPy); 7.73 (*dd*, H–C(3) of SPy); 8.09 (*s*, H–C(8)); 8.33 (*dd*, H–C(6) of SPy).

9-(2-Deoxy-β-D-erythro-*pentofuranosyl)-6-[(1-oxidopyridin-2-yl)sulfanyl]-9*H-*purin-2-amine* (**1c**). To a soln. of **2a** (765 mg, 1.26 mmol) in anh. THF (15 ml) were added activated 4-Å molecular sieves (250 mg) and a 1M soln. of TBAF in THF (2.77 ml, 2.77 mmol). The mixture was stirred for 1 h at r.t. under Ar gas, filtered, and washed with MeOH (20 ml). The filtrate was evaporated to dryness, and the residue was purified by CC (SiO_2; CH_2Cl_2/MeOH 91:9) to afford 7 mg (78%) of **1c**. TLC (SiO_2; $CHCl_3$/MeOH 9:1): R_f 0.23 . UV (H_2O): 253 (13,750), 325 (7,002). UV (MeCN): 224 (18,540), 324 (6,750). ^{1}H-NMR (500 MHz, CD_3OD): 2.40 (*ddd*, $J_{2''}$ = 13.5, $J_{3'}$ = 2.9, H–C(2′)); 2.79 (*ddd*, $J_{2'}$ = 13.5, $J_{3'}$ = 1.2, H–C(2″)); 3.75 (*dd*, $J_{5''}$ = 12.1, $J_{4'}$ = 3.8, H–C(5′)); 3.83 (*dd*, $J_{5'}$ = 12.1, $J_{4'}$ = 3.4, H–C(5″)); 4.03 (*dd*, $J_{3'}$ = 6.6, $J_{5'}$ = 3.4, H–C(4′)); 4.58 (*ddd*, $J_{4'}$ = 6.6, $J_{2'}$ = 2.9, H–C(3′)); 6.37 (*dd*, $J_{2'}$ = 6.9, $J_{2''}$ = 6.8, H–C(1′)); 7.45 (*ddd*, *J* = 7.7, 6.3, 1.9, H–C(5) of SPy); 7.50 (*ddd*, *J* = 8.1, 7.7, 1.1, H–C(4) of SPy); 8.00 (*dd*, *J* = 8.1, 1.9, H–C(3) of SPy); 8.24 (*s*, H–C(8)); 8.41 (*dd*, *J* = 6.3, 1.1, H–C(6) of SPy). ^{13}C-NMR (125 MHz, CD_3OD): 40.1 (C(2′)); 62.4 (C(5′)); 71.8 (C(3′)); 85.2 (C(1′)); 88.5 (C(4′)); 124.8 (C(5) of SPy); 126.6 (C(5)); 128.4 (C(4) of SPy); 129.8 (C(3) of SPy); 139.6 (C(6) of SPy); 141.6 (C(8)); 146.3 (C(4)); 152.3 (C(2)); 155.0 (CS of SPy); 160.3 (C(6)). HR-ESI-MS (pos.): 377.1014 ($[M+H]^+$, $C_{15}H_{17}N_6O_4S^+$; calc. 377.1027).

9-{2-Deoxy-3,5-bis-O-*[(1,1-dimethylethyl)(dimethyl)silyl]-β*-D-erythro-*pentofuranosyl}-6-(pyridin-2-ylsulfanyl)-9*H-*purin-2-amine* (**2a**). To a soln. of **1a** (15 mg, 0.025 mmol) in anh. benzene (1 ml) was added pyridine-2(1*H*)-thione (2.8 mg, 0.025 mmol), and the mixture was stirred at r.t. for 72 h. The solvent was evaporated under vacuum, and the residue was purified by CC (SiO_2; 3–5% MeOH in $CHCl_3$) to afford 12 mg (80%) of **2a**. TLC (SiO_2; $CHCl_3$/MeOH 95:5): R_f 0.33. ^{1}H-NMR (200 MHz, $CDCl_3$): 0.07, 0.10 (2*s*, 6 H, MeSi); 0.90, 0.91 (2*s*, 9 H, ^{t}Bu); 2.29–2.41 (*m*, H–C(2′)); 2.52–2.65 (*m*, H–C(2″)); 3.74 (*dd*, $J_{5''}$ = 11.6, $J_{4'}$ = 3.6, H–C(5′)); 3.81 (*dd*, $J_{5'}$ = 11.6, J_4 = 4.5, H–C(5″)); 3.98 (*dd*, $J_{3'}$ = 6.8, $J_{5'}$ = 3.6, H–C(4′)); 4.58 (*ddd*, $J_{4'}$ = 6.8, $J_{2'}$ = 3.6, H–C(3′)); 4.92 (*s*, 2 H, H–C(2)); 6.31 (*dd*, $J_{2'}$ = 7.0, $J_{2''}$ = 6.2, H–C(1′)); 7.22–7.25 (*m*, H–C(5) of SPy); 7.62–7.76 (*m*, H–C(3,4) of SPy); 7.96 (*s*, H–C(8)); 8.59 (*d*, H–C(6) of SPy). ^{13}C-NMR (50 MHz, $CDCl_3$): −5.49, −5.37, −4.79, −4.69 (MeSi); 18.0, 18.4, 25.7, 25.9 (^{t}BuSi); 40.8 (C(2′)); 62.8 (C(5′)); 72.0 (C(3′)); 83.6 (C(1′)); 87.8 (C(4′)); 122.7 (C(5) of SPy); 129.4 (C(3) of SPy); 136.7 (C(4) of SPy); 139.2 (C(4,8)); 150.1 (C(6) of SPy); 151.3 (C(2)); 153.1 (SSC of SPy); 159.0 (C(6)). HR-ESI-MS: 611.2612 ($[M+Na]^+$, $C_{27}H_{44}N_6NaO_3SSi_2^+$; calc. 611.2632).

9-(2-Deoxy-β-D-erythro-*pentofuranosyl)-6-(pyridin-2-ylsulfanyl)-9*H-*purin-2-amine* (**2c**). To a soln. of **2a** (15 mg, 0.025 mmol) in anh. THF (0.5 ml) were added activated 4-Å molecular sieves (50 mg)

and a 1M soln. of TBAF in THF (55 µl, 0.055 mmol). The mixture was stirred for 1 h at r.t. under Ar gas, filtered, and washed with MeOH (2 ml). The filtrate was evaporated to dryness, and the residue was purified by CC (SiO_2; CH_2Cl_2/MeOH 91:9) to afford 7 mg (78%) of **2c**. TLC (SiO_2; CH_2Cl_2/MeOH 9:1): R_f 0.28. ^{1}H-NMR (200 MHz, D_2O): 2.41–2.53 (*m*, H–C(2′)); 2.72–2.86 (*m*, H–C(2″)); 3.69–3.83 (*m*, H–C(5′), H–C(5″)); 4.10 (*d*, H–C(4′)); 4.60 (*s*, H–C(3′)); 6.31 (*dd*, *J*=7.2, 5.8, H–C(1′)); 7.43 (*dd*, *J*=7.0, 4.2, H–C(5) of SPy); 7.69 (*d*, *J*=8.0, H–C(3) of SPy); 7.85 (*dd*, *J*=8.0, 7.0, H–C(4) of SPy); 8.13 (*s*, H–C(8)); 8.47 (*d*, *J*=4.2, H–C(6) of SPy). HR-ESI-MS: 361.1064 ($[M+H]^+$, $C_{15}H_{17}N_6O_3S^+$; calc. 361.1083).

9-[2-Deoxy-3,5-bis-O-*[(1,1-dimethylethyl)(dimethyl)silyl]-β*-D-erythro-*pentofuranosyl]-6-[(1-oxidopyridin-2-yl)sulfanyl]*-N-*[(phenyloxy)acetyl]-9*H-*purin-2-amine* (**6a**). To a soln. of 1-hydroxy-1*H*-benzotriazole (HOBt; 475 mg, 3.4 mmol) and *Tris* buffer (evaporated from anh. MeCN) in anh. MeCN/pyridine 1:1 (6 ml) were added dropwise 2-phenoxyacetyl chloride (446 mg, 3.3 mmol). At the same time, a soln. of **1a** (800 mg, 1.3 mmol) in 6 ml of anh. pyridine was prepared. After 15 min, both solns. were ice-cooled, and the soln. of **1a** was added dropwise to the latter soln. The resulting mixture was stirred for 24 h at r.t., then cooled to 5°, treated with H_2O (10 ml), and stirred for an additional 30 min. Then, $CHCl_3$ (15 ml) was added, the mixture was extracted with sat. aq. $NaHCO_3$ (15 ml), and the org. layer was dried (Na_2SO_4) and evaporated to dryness. The residue was purified by CC (SiO_2; $CHCl_3$/MeOH 99:1) to afford 701 mg (73%) of **6a**. TLC (SiO_2; $CHCl_3$/MeOH 95:5): R_f 0.68 . ^{1}H-NMR (200 MHz, $CDCl_3$): 0.09, 0.12 (2*s*, 6 H, MeSi); 0.91, 0.93 (2*s*, 9 H, *t*Bu); 2.39–2.51 (*m*, H–C(2′)); 2.57–2.70 (*m*, H–C(2″)); 3.77 (*dd*, *J*=11.4, 3.0, H–C(5′)); 3.88 (*dd*, *J*=11.4, 3.8, H–C(5″)); 4.02 (*dd*, H–C(4′)); 4.60–4.65 (*m*, H–C(3′)); 4.69 (*s*, $COCH_2$); 6.26 (*dd*, *J*=6.4, 6.0, H–C(1′)); 6.96–7.39 (*m*, 5 arom H, H–C(4,5) of SPy); 8.28 (*s*, H–C(8)); 8.32 (*d*, H–C(3) of SPy); 8.47 (*d*, H–C(6) of SPy); 8.82 (*s*, H–C(3)).

9-(2-Deoxy-β-D-erythro-*pentofuranosyl)-6-[(1-oxidopyridin-2-yl)sulfanyl]*-N-*[(phenyloxy)acetyl]-9*H-*purin-2-amine* (**6c**). To a soln. of **6a** (520 mg, 0.704 mmol)[4]) in anh. THF (8.5 ml) were added activated 4-Å molecular sieves (150 mg) and a 1M soln. of TBAF in THF (2.11 ml, 2.11 mmol). The mixture was stirred for 1 h at r.t. under Ar gas, filtered, and washed with MeOH (20 ml). The filtrate was evaporated to dryness, and the residue was purified by CC (SiO_2; CH_2Cl_2/MeOH 95:5) to afford 213 mg (60%) of **6c**. TLC (SiO_2; CH_2Cl_2/MeOH 95:5): R_f 0.30. ^{1}H-NMR (200 MHz,$CDCl_3$): 2.51–2.64 (*m*, H–C(2′)); 2.85–2.99 (*m*, H–C(2″)); 3.76–3.79 (*m*, H–C(5′), H–C(5″)); 4.12 (*dd*, *J*=8.8, 4.4, H–C(4′)); 4.70–4.79 (*m*, H–C(3′)); 4.79 (*s*, $COCH_2$); 6.50 (*dd*, *J*=6.9, 5.9, H–C(1′)); 6.91–7.10 (*m*, 3 arom. H); 7.35 (*dd*, 2 arom. H); 7.48 (*ddd*, *J*=7.9, 7.0, 1.7, H–C(5) of SPy); 7.59 (*dd*, *J*=8.0, 7.9, H–C(4) of SPy); 7.95 (*d*, *J*=8.0, H–C(3) of SPy); 8.36 (*s*, H–C(8)); 8.37 (*d*, *J*=7.0, H–C(6) of SPy).

9-(5-O-*[Bis[4-(methyloxy)phenyl](phenyl)methyl]-2-deoxy-β*-D-erythro-*pentofuranosyl)-6-[(1-oxidopyridin-2-yl)sulfanyl]*-N-*[(phenyloxy)acetyl]-9*H-*purin-2-amine* (**7c**). To a soln. of **6c** (100 mg, 0.20 mmol) in anh. pyridine (1.4 ml) was added dimethoxytrityl chloride[5]) (154 mg, 0.46 mmol), and the mixture was stirred at r.t. for 24 h. MeOH (1.2 ml), $CHCl_3$ (7.5 ml), and phosphate buffer (7.5 ml; pH 7.0) were added, and the org. phase was separated and washed again with phosphate buffer. The org. layer was dried (Na_2SO_4), and co-evaporated with toluene (3×2.5 ml). The residue was purified by CC (SiO_2; 2–5% of MeOH in $CHCl_3$) to afford 109 mg (67%) of **7c**. TLC (SiO_2; $CHCl_3$/MeOH 98:2): R_f 0.55. ^{1}H-NMR (200 MHz, $CDCl_3$): 2.59–2.68 (*m*, H–C(2′), H–C(2″)); 3.32–3.44 (*dd*, 2 H, H–C(5)); 3.69 (*s*, 2 MeO); 4.16 (*d*, H–C(4′)); 4.56 (*s*, $COCH_2$); 4.69 (*s*, H–C(3′)); 6.46 (*dd*, H–C(1′)); 6.72 (*d*, 5 arom H); 6.86–7.02 (*m*, 3 arom. H); 7.14–7.36 (*m*, 12 arom. H, H–C(4,5) of SPy); 8.04 (*s*, H–C(8)); 8.25 (*d*, H–C(3) of SPy); 8.38 (*d*, H–C(6) of SPy); 8.94 (*s*, H–C(3)). ^{13}C-NMR (50 MHz, $CDCl_3$): 40.5 (C(2′)); 55.1 (MeO); 63.9 (C(5′)); 67.6 (C–CH_2); 71.8 (C(3′)); 84.2 (C(1′)); 86.3 (C(4′)); 88.0 (*C*–OC(5′)); 113.0 (arom. C); 114.8 (C(5=of SPy); 122.1 (C(4) of SPy); 125.3 (C(3) of SPy); 126.8, 127.0 (arom. C); 127.6 (C(6) of SPy); 127.7, 128.0, 128.9, 129.0, 129.7, 129.9 (arom. C); 135.5 (C(2)); 135.6 (C(4)); 139.1 (C(8)); 142.3 (MeO–*C*), 144.4 (C(6)); 158.3 (C=O). ESI-MS: 813.5 ($[M+H]^+$), 511.3 ($[M-DMT+H]^+$), 395.3 ([base+H]$^+$), 303.3 (DMT^+). HR-ESI-MS (pos.): 835.2455 ($[M+Na]^+$, $C_{44}H_{40}N_6NaO_8S^+$; calc. 835.2526). HR-ESI-MS (neg.): 811.2546 ($[M-H]^-$, $C_{44}H_{39}N_6O_8S^-$; calc. 811.2550).

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[4]) Dried by heating over P_2O_5 under vacuum at 56° for 2 h before use.

[5]) Systematic name: 1,1′-(chlorophenylmethylene)bis[4-methoxybenzene].

*9-(3-*O*-{[Bis(1-methylethyl)amino][(2-cyanoethyl)oxy]phosphanyl}-5-*O*-{bis[4-(methyloxy)phenyl]-(phenyl)methyl}-2-deoxy-β-*D*-erythro-pentofuranosyl)-6-[(1-oxidopyridin-2-yl)sulfanyl]-*N*-[(phenyloxy)-acetyl]-9*H*-purin-2-amine* (**7a**). To a soln. of anh. **7c**[6]) (150 mg, 0.18 mmol) in anh. CH_2Cl_2 (5 ml) under Ar gas was added i-Pr_2NEt (200 µl, 1.14 mmol) and 2-cyanoethyl-*N,N*-diisopropylchlorophosphoramidite (100 µl, 0.43 mmol), and the mixture was stirred for 2 h at r.t. Then, MeOH (1 ml) was added, the org. layer was extracted, dried (Na_2SO_4), and evaporated. The residue was purified by CC (SiO_2; 1–2% of MeOH in CH_2Cl_2/Et_3N 98:2) to afford 130 mg (70%) of **7a**. Yellowish foam. TLC (SiO_2; $CHCl_3$/MeOH/Et_3N 94:5:1): R_f 0.55. ^{1}H-NMR (200 MHz, $CDCl_3$): 1.11–1.42 (*m*, 4 Me of i-Pr); 1.24 (*dd*, 2 CH of i-Pr); 2.71–2.77 (*m*, 2 H, H–C(2)); 3.39–3.67 (*m*, 6 H, H–C(5), CH_2); 3.75 (*s*, 2 MeO); 4.08–4.19 (*m*, 3 H, $COCH_2$, H–C(4′)); 4.50–4.58 (*m*, H–C(3′)); 6.41 (*dd*, H–C(1′)); 6.73–6.87 (*m*, 7 arom. H); 7.01 (*dd*, 1 arom. H); 7.19–7.38 (*m*, 10 arom. H); 7.70 (*dd*, H–C(5) of SPy); 7.85 (*dd*, H–C(4) of SPy); 8.08 (*d*, H–C(3) of SPy); 8.54 (*s*, H–C(8)); 8.58 (*d*, H–C(6) of SPy). ^{31}P-NMR (32.4 MHz, $CDCl_3$/H_3PO_4 (85%)): 150.04 (*s*). ESI-MS (pos.): 1073.3 ($[M-H+Na+K]^+$, 1052.1 ($[M+K]^+$), 1034.9 ($[M+Na]^+$). HR-ESI-MS: 1013.3769 ($[M+H]^+$, $C_{53}H_{58}N_8O_9PS^+$; calc. 1013.3780).

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[6]) Dried by co-evaporation with anh. pyridine (2×).

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