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CONVENIENT SYNTHESIS OF 8-AMINO-2'-DEOXYADENOSINE.

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ABSTRACT

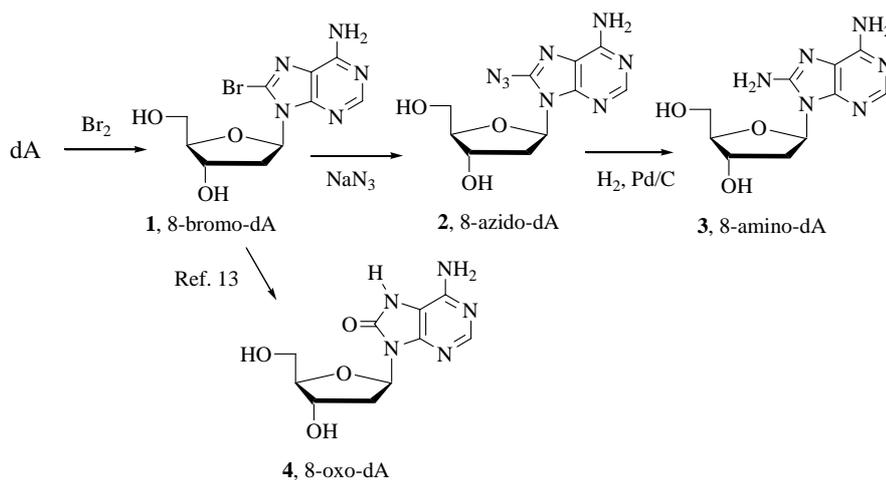
We studied the behaviour of 8-azido-2'-deoxyadenine and 8-bromo-2'-deoxyadenosine in aqueous solutions of ammonia and primary and secondary amines. Unexpectedly, 8-Azido-2'-deoxyadenosine is converted to 8-amino-2'-deoxyadenosine in excellent yields. The use of this reaction for the preparation of 8-aminoadenine derivatives needed for the preparation of oligonucleotides carrying 8-aminoadenine is discussed.

INTRODUCTION

Purine nucleosides carrying azido groups at the nucleobase are important intermediates for the preparation of photoreactive nucleotides used in the study of protein-nucleic acid interactions.¹ Moreover, they are useful intermediates in the synthesis of modified nucleosides like purine nucleosides carrying amino groups.^{2,3} For this purpose, a nucleoside carrying a halogen (Cl or Br) is reacted with sodium or lithium azide yielding the azidopurine, which is reduced by catalytic hydrogenation³ or reducing metals.^{2,4}

Oligonucleotides carrying 8-aminoadenine form very stable triple helices⁵⁻⁸ and parallel-stranded structures.^{8,9} The introduction of the amino group at position 8 increases the stability of Hoogsteen structures owing to the combined effect of the gain of one Hoogsteen purine-pyrimidine H-bond and the tendency of the amino group to be integrated into the ‘spine of hydration’ located in the minor-major groove of the triplex structure.⁶ 8-Amino-2'-deoxyadenine (**3**) was prepared using the following three-step protocol: bromination of dA, nucleophilic displacement to form the 8-azidonucleoside (**2**) and catalytic hydrogenation of 8-azido-2'-deoxyadenine (**2**) (SCHEME I).³ Moreover, 8-azido-2'-deoxyadenosine (**2**) has been incorporated into oligonucleotides.^{10,11} 8-Azido-2'-deoxyadenosine (**2**) is partially decomposed during the ammonia treatment used for the removal of protecting groups of oligonucleotides (conc. NH₃, room temperature and 55 °C). Fàbrega et al. described the formation of one single side product, 8-amino-2'-deoxyadenosine (**3**)¹⁰ and Liu et al. reported two possible side compounds: 8-amino-2'-deoxyadenosine (**3**) and 8-oxo-2'-deoxyadenosine (**4**).¹¹ These two products are also formed during the ammonia treatment of oligonucleotides carrying 8-bromo-2'-deoxyadenine.¹²

SCHEME 1: Synthesis of the 8-substituted 2'-deoxyadenosine derivatives used in this study.



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We analysed the behaviour of 8-azido-2'-deoxyadenosine (**2**) in ammonia solutions and in solutions of primary and secondary amines. We confirmed that only 8-amino-2'-deoxyadenosine (**3**) is formed and that the reaction of compound **2** with volatile primary amines is useful for the preparation of 8-amino-2'-deoxyadenine without hydrogenation.

RESULTS AND DISCUSSION

8-Bromo-2'-deoxyadenosine (**1**, SCHEME I) was prepared by bromination of dA¹³ (73% yield). Reaction of **1** with sodium azide^{3,14} gave 8-azido-2'-deoxyadenosine (**2**) in 75% yield. Hydrogenation³ of compound **2** using Pd/ activated charcoal as catalyst gave 8-amino-2'-deoxyadenosine (**3**) in 90% yield. Finally, starting from compound **1**, 8-oxo-2'-deoxyadenosine (**4**) was prepared as described elsewhere.¹³ Reverse-phase HPLC using diode-array detector allowed the separation and rapid identification of compounds **1-4**. The elution order from more polar to less polar was as follows: 8-amino-dA (**3**), 8-oxo-dA (**4**), 8-azido-dA (**2**) and 8-bromo-dA (**1**).

Aliquots of 8-azido-dA (**2**) and 8-bromo-dA (**1**) were treated with 3 ml of 30% aqueous ammonia and 5M methanolic ammonia at 55°C. The reaction of 8-azido-dA (**2**) with aqueous ammonia gave only 8-amino-dA (**3**). A 45% conversion was observed after 20 hours, 62% after 3 days and 90% after 6 days (TABLE 1). The product was isolated and characterized by ¹H and ¹³C-RMN. Compound **2** decomposed slowly in methanolic ammonia (<50% after 6 days, TABLE 1). On the other hand, treatment of 8-bromo-dA (**1**) with 30% aqueous ammonia and 5M methanolic ammonia at 55°C gave small amounts of a mixture of 8-amino-dA (**3**) and 8-oxo-dA (**4**) (20% decomposition with aqueous ammonia after 16 hours at 55°C, TABLE 1).

These results were in agreement with the behaviour of compounds **1** and **2** in oligonucleotides,¹⁰⁻¹² except that 8-oxo-dA (**4**) was not formed in the treatment of compound **2** with aqueous ammonia. Unexpectedly, the formation of 8-amino-dA (**3**)

from compound **2** was clean and faster than the formation of compound **3** from compound **1**. To gain more information on this reaction, compound **3** was also treated with an aqueous solution of several amines.

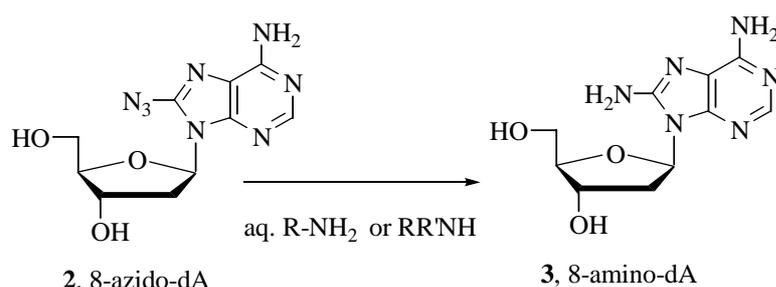
Table 1: Composition of the reaction mixtures obtained after the treatment of compounds **1** and **2** with amine solutions at 55°C.

Compound	Treatment	time	Composition of the reaction mixture (%)				
			2	3	1	4	5
2	30% aq.NH ₃	16 h	55	45	--	--	--
		3 days	38	62	--	--	--
		6 days	10	90	--	--	--
2	5M NH ₃ /MeOH	16 h	96	4	--	--	--
		3 days	65	35	--	--	--
		6 days	53	47	--	--	--
1	30% aqNH ₃	5 h	--	--	80	7	13
1	5M NH ₃ /MeOH	16 h	--	--	80 [#]	9 [#]	7 [#]
2	40% aqCH ₃ NH ₂	5 h	0	100	--	--	--
2	30% aq dimethylamine	16 h	30	70	--	--	--
2	1M aq piperidine	16 h	0	100	--	--	--
2	1M aq 1,6- diaminohexane	5 h	0	100	--	--	--
1	1M aq 1,6- diaminohexane	16 h	--	--	0	20	80

[#]In addition 4% of a product eluting at 9.8 min (UV max 260 nm) was observed. Most probably the product was 8-methoxy-2'-deoxyadenosine.

Aliquots of 8-azido-dA (**2**) were treated with solutions of amines (SCHEME II) at 55°C. The reaction of 8-azido-dA (**2**) with 40% aqueous methylamine gave complete conversion to 8-amino-dA (**3**) in less than 5 hours. Reaction with a 30% aqueous solution of dimethylamine gave 70% conversion to 8-amino-dA after 16 hours. Treatment of compound **2** with 1M aqueous solutions of piperidine and 1,6-diaminohexane gave also gave 8-amino-dA (**3**) at 55°C after 16 and 5 hours respectively. The identity of the product was confirmed by HPLC analysis and by isolation of the products resulting from the reactions and analysis of their ^1H and ^{13}C -NMR spectra.

SCHEME 2: Treatment of 8-azido-2'-deoxyadenosine with amines.

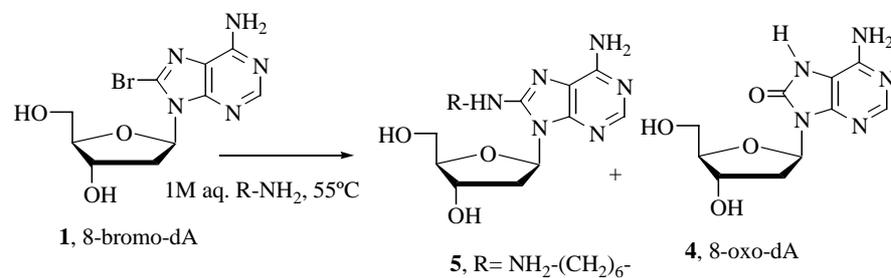


$\text{R-NH}_2 = 40\% \text{ aq. CH}_3\text{NH}_2, 1\text{M aq. NH}_2\text{-(CH}_2\text{)}_6\text{-NH}_2$

$\text{RR'-NH}_2 = 40\% \text{ aq. (CH}_3\text{)}_2\text{NH}, 1\text{M aq. piperidine}$

On the other hand, treatment of 8-bromo-dA (**1**) with hexane-1,6-diamine gave the expected mixture of the products resulting from the attack of both nucleophiles present in the mixture: 8-oxo-dA and 8-(6-aminohexyl)amino-dA (SCHEME 3), in agreement with the results obtained during the preparation of 8-N-aminosubstituted dA derivatives.^{3,15}

SCHEME 3: Products obtained from the treatment of 8-bromo-2'-deoxyadenosine with a primary amine.



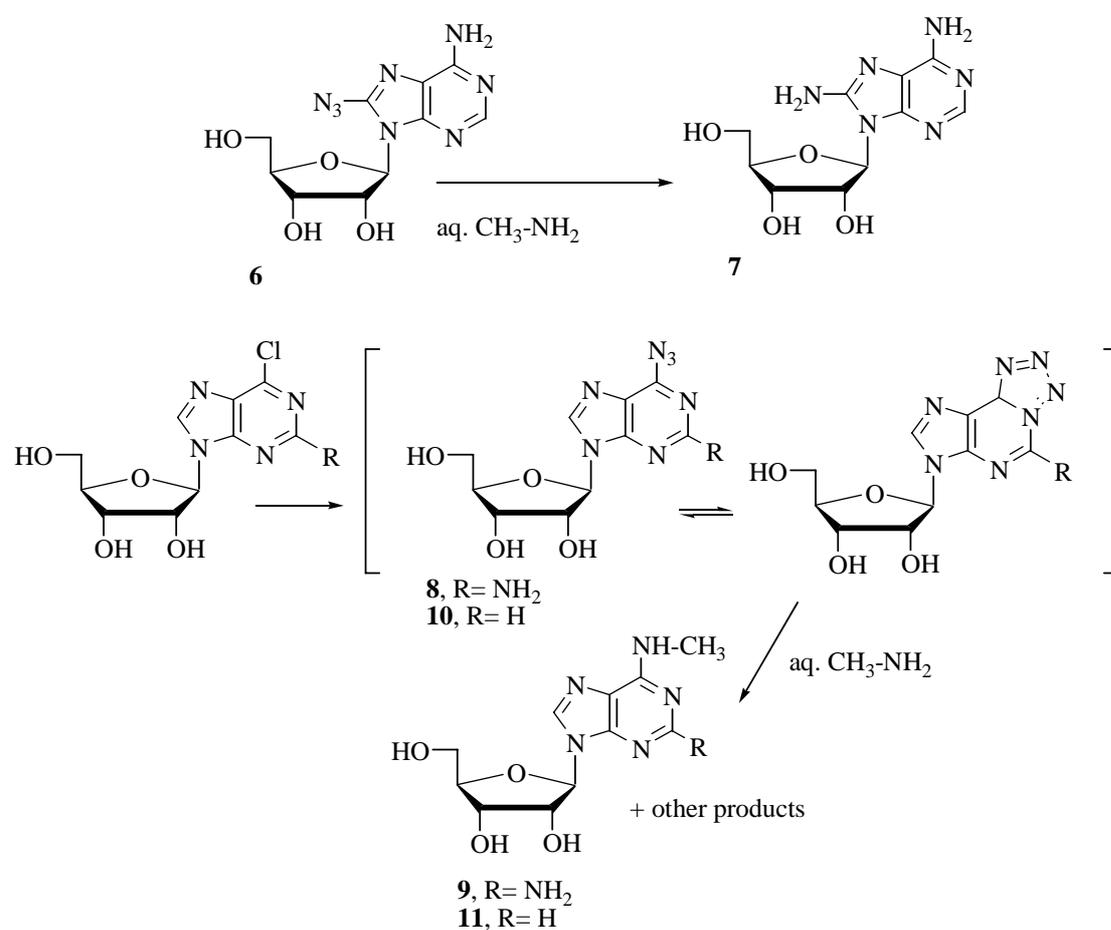
These data reveal a distinct behaviour of compounds **1** and **2** on aqueous amine solutions. The bromo derivative suffers a nucleophilic displacement by the amine and water, while the azido derivative is converted to the amino derivative (**3**) regardless of the amine present in the reaction and without interference with water. The reaction in the presence of primary amines is faster than in the presence of secondary amines, which is faster than in the presence of ammonia. This suggests the involvement of the amine in the thermal decomposition of the azido group to the nitrene intermediate, which later abstracts hydrogen from the environment to give the 8-aminopurine.¹⁶

The scope of this reaction was assayed with several purine ribonucleosides carrying azido groups. As expected the treatment of ribonucleoside 8-azidoadenosine (**6**) with 40% aqueous methylamine gave 8-aminoadenosine (**7**) (SCHEME 4). However, treatment of 6-azido-2-aminopurine riboside (**8**) and 6-azidopurine riboside (**10**) resulted in the addition of methylamine at position 6 of the purine with the subsequent displacement of the azido group (compounds **7** and **9**) together with some decomposition products. In these cases, the azido group is in the tetrazolo tautomeric form (absence of an IR band in the region of 2000-2200 cm⁻¹ and presence of an intense band in the 1700-1500 cm⁻¹ region)^{17,18} and the nitrene is not formed. The reaction observed with compound **2** is only possible when the azido group is at position 8.

Finally, we tested the reaction of compound **2** with amines for preparative purposes. We selected the inexpensive 40% aqueous methylamine solution because it allowed the fast formation of the desired compound **3** was very fast, and the amine was volatile. Several reactions on a 1-5 gram scale gave the desired compound in quantitative yields. The

product resulting from the evaporation of methylamine was pure enough to perform the protection of the amino groups with the dimethylformamide^{19,20} groups needed for the preparation of oligonucleotides carrying 8-amino-2'-deoxyadenosine. The DMT-protected phosphoramidite derivative as well as oligonucleotides carrying 8-amino-2'-deoxyadenosine were prepared as described elsewhere.^{5,6}

SCHEME 4: Products obtained from the treatment of the ribonucleosides carrying azido groups with methylamine.



EXPERIMENTAL SECTION

General Methods. Solvents, including those of HPLC grade, were from SDS and E. Merck. Reagents were from Aldrich and Fluka and were used without further

purification. Analytical TLC was run on aluminium sheets coated with silica gel 60 F₂₅₄ from Merck. Silica gel column chromatography was performed with Chromatogel 60 A C.C. (40-60 microns, 230-400 mesh, SDS). 8-Bromo-2'-deoxyadenosine (**1**),¹³ 8-azido-2'-deoxyadenosine (**2**),^{3,14} 8-amino-2'-deoxyadenosine (**3**)³ and 8-oxo-2'-deoxyadenosine (**4**)¹³ were prepared as described. 8-Bromoadenosine, 2-amino-6-chloro-9-β-D-ribofuranosylpurine, and 6-chloro-9-β-D-ribofuranosylpurine were obtained from Pharma-Waldhof GmbH (Düsseldorf, Germany). N⁶-methyladenosine (**11**) was purchased from Sigma.

Instrumental. ¹H-NMR (250 MHz) and ¹³C-NMR (63 MHz) spectra were recorded on a Brüker AM-250. HPLC chromatography was performed on an HPLC Shimadzu equipped with a diode array detector.

Treatment of 8-amino-2'-deoxyadenosine and 8-bromo-2'-deoxyadenosine with ammonia and amine aqueous solutions. 50 mg aliquots of 8-amino-2'-deoxyadenosine and 8-bromo-2'-deoxyadenosine were placed in screw-cap tubes and 3 ml of the appropriate ammonia or amine solutions was added. The mixtures were heated to 55°C for a period of time between 5 hours and 6 days, allowed to cool to room temperature and evaporated to dryness. The residues were analysed by HPLC. HPLC conditions were as follows: Solution A, 0.1 M aqueous triethylammonium acetate pH 6.5 / acetonitrile (95:5); solution B, 0.1 M aqueous triethylammonium acetate pH 6.5 / acetonitrile (3: 7); Column PRP-1 (Hamilton, 10 μm) 250x 10 mm; flow rate, 3 ml/ min and; 20 min linear gradient from 0% B to 50% B. Retention time for 8-amino-dA: 7.4 min; for 8-oxo-dA: 8.1 min; for 8-azido-dA: 11.3 min and for 8-bromo-dA: 11.4 min. UV maximum for 8-amino-dA: 276 nm; for 8-oxo-dA 272 nm; for 8-azido-dA 284 nm and for 8-bromo-dA 267 nm. Reaction products were also identified by comparison of the chemical shift of the C-8 at ¹³C-NMR spectra with published spectra.^{10,13}

8-Azidoadenosine (6). Compound **6** was prepared as described¹⁴ with minor modifications. 250 mg (0.72 mmol) of 8-bromoadenosine was treated with 148 mg (2.17 mmol) of sodium azide in 5 ml of dimethylformamide (DMF) at 60°C for 16 h.

The solution was cooled and concentrated to dryness. The resulting product was crystallized from water-methanol (MeOH) giving 130 mg (59% yield) of a white solid. UV (max, pH 6.5) 284 nm. IR (KBr, cm^{-1}): intense bands at 2154 and 2043. $^1\text{H-NMR}$ (DMSO- d_6): 8.05 (s, 1H, H-2), 7.31 (wide s, 2H, amino), 5.60 (d, 1H, OH), 5.38 (m, 2H, OH and H-1'), 5.15 (d, 1H, OH), 4.84 (m, 1H, H-2'), 4.18 (m, 1H, H-3'), 3.9 (m, 1H, H-4'), 3.58 (m, 2H, H-5'). $^{13}\text{C-NMR}$ (DMSO- d_6): 154.7 (C6), 156.8 (C2), 149.6 (C4), 144.6 (C8), 117.5 (C5), 87.8 (C1'), 86.5 (C4'), 71.5 (C3'), 71.0 (C2'), 62.3 (C5'). MS (electrospray): found 309.8 ($\text{M}+\text{H}^+$) calculated for $\text{C}_{10}\text{H}_{12}\text{N}_8\text{O}_4$ 308.3.

6-Azido-2-amino-9- β -D-ribofuranosylpurine (**8**). 300 mg (1 mmol) of 8-chloro-2-amino-9- β -D-ribofuranosylpurine was treated with 211 mg (3 mmol) of sodium azide in 7 ml of DMF at 60°C for 16 h. The solution was cooled and concentrated to dryness. The resulting product was crystallized from water-methanol (MeOH) giving 200 mg (65% yield) of a white solid. UV (max, pH 6.5) 270, 300 nm. IR (KBr, cm^{-1}): absence of bands in the region 2200-2000, intense band at 1694. $^1\text{H-NMR}$ (DMSO- d_6): 8.4 (m, 3H, H-8 and amino), 5.91 (d, 1H, H-1'), 5.45 (d, 1H, OH), 5.20 (d, 1H, OH), 5.02 (t, 1H, OH), 4.5 (m, 1H, H-2'), 4.18 (m, 1H, H-3'), 3.9 (m, 1H, H-4'), 3.6 (m, 2H, H-5'). $^{13}\text{C-NMR}$ (DMSO- d_6): 146.3 (C6), 145.1 (C2), 144.1 (C4), 138.5 (C8), 112.4 (C5), 87.3 (C1'), 85.7 (C4'), 74.3 (C3'), 70.2 (C2'), 61.4 (C5'). MS (electrospray): found 309.0 ($\text{M}+\text{H}^+$) calculated for $\text{C}_{10}\text{H}_{11}\text{N}_8\text{O}_4$ 307.2.

6-Azido-9- β -D-ribofuranosylpurine (**10**). 500 mg (1.75 mmol) of 8-chloro-9- β -D-ribofuranosylpurine was treated with 357 mg (5.2 mmol) of sodium azide in 10 ml of DMF at 60°C for 16 h. The solution was cooled and concentrated to dryness. The resulting product was crystallized from water-methanol (MeOH) giving 110 mg (21% yield) of a white solid. UV (max, pH 6.5) 290 nm. IR (KBr, cm^{-1}): absence of bands in the region 2200-2000, intense band at 1645. $^1\text{H-NMR}$ (DMSO- d_6): 10.1 (s 1H), 8.9 (s, 1H), 6.11 (d, 1H, H-1'), 5.62 (d, 1H, OH), 5.29 (d, 1H, OH), 5.1 (t, 1H, OH), 4.53 (m, 1H, H-2'), 4.15 (m, 1H, H-3'), 3.98 (m, 1H, H-4'), 3.64 (m, 2H, H-5'). $^{13}\text{C-NMR}$

(DMSO- d_6): 145.6 (C6), 142.8 (C2), 142.1 (C4), 136.3 (C8), 121 (C5), 88.5 (C1'), 85.9 (C4'), 74.8 (C3'), 70.2 (C2'), 61.1 (C5'). MS (electrospray): found 293.9 (M+H⁺) calculated for C₁₀H₁₁N₇O₄ 293.2.

Treatment of ribonucleosides carrying azido groups with methylamine.

50 mg aliquots of azido-ribonucleosides (**6**, **8**, **10**) were placed in screw-cap tubes and 3 ml of 40% *aqueous* methylamine was added. The mixtures were heated to 60°C for a period of time between 16 and 48 h, allowed to cool to room temperature and evaporated to dryness. The residues were analysed by HPLC as described above. Retention time (UV maxima) for compound **6**: 9.6 min (284 nm); for compound **7**: 6.4 min (272 nm); for compound **8**: 7.5 min (270, 300nm); for compound **9**: 2.7 min (242 nm); for compound **10**: 8.2 min (290 nm) and for compound **11**: 3.8 (250 nm).

The reaction with compound **6** was completed after 16 h, giving one single product that was characterized as 8-aminoadenosine (**7**)¹⁴. ¹H-NMR (DMSO- d_6): 7.89 (s, 1H, H-2), 6.54 (m, 2H, amino), 5.82 (d, 1H, H-1'), 4.64 (m, 1H, H-2'), 4.12 (m, 1H, H-3'), 3.9 (m, 1H, H-4'), 3.58 (m, 2H, H-5'). ¹³C-NMR (DMSO- d_6): 152.6 (C6), 151.8 (C2), 149.5 (C4), 148.5 (C8), 117.4 (C5), 86.8 (C1'), 86.0 (C4'), 71.1 (C3' and C2'), 62.0 (C5'). MS (electrospray): found 283.4 (M+H⁺) calculated for C₁₀H₁₄N₆O₄ 282.2.

The reaction with compound **8** was completed after 48 h, giving one major product that was characterized as 2-amino-N⁶-methyladenosine (**9**)²¹ together with some minor products that were not characterized. ¹H-NMR (DMSO- d_6): 7.9 (s, 1H, H-8), 5.49 (d, 1H, H-1'), 4.32 (m, 1H, H-2'), 4.12 (m, 1H, H-3'), 3.86 (m, 1H, H-4'), 3.58 (m, 2H, H-5'), 2.7 (s, 3H, CH₃). ¹³C-NMR (DMSO- d_6): 156.5 (C6), 155.6 (C2), 150.7 (C4), 133.7 (C8), 112.7 (C5), 87.6 (C1'), 84.4 (C4'), 74.6 (C3'), 69.6 (C2'), 60.6 (C5'), 27.9 (CH₃). MS (electrospray): found 297.5 (M+H⁺) calculated for C₁₁H₁₆N₆O₄ 296.3.

The reaction with compound **10** was completed after 16 h, giving several products. One of the products was characterized as N⁶-methyladenosine (**11**) by comparison with the commercially available product and mass spectrometry (MS (electrospray): found 284.5 (M+H⁺) calculated for C₁₁H₁₅N₅O₄ 281.2).

Synthesis of 8-amino-N,N-bis(dimethylaminomethyliden)-2'-deoxyadenosine

In a screw-cap tube, 4.62 g of 8-azido-2'-deoxyadenosine^{3,14} was dissolved with 20 ml of 40% aqueous methylamine solution and 2 ml of dioxane. The solution was heated overnight to 55°C. It was then cooled to room temperature and concentrated to dryness, yielding an oil that was used in the following step without purification. Purity assessed by HPLC (< 95%).

The product described above (approx. 17.5 mmol) was dissolved in 250 ml of DMF and treated with 12.1 ml of dimethyl acetal of the DMF. The solution was stirred overnight at room temperature and then evaporated to dryness. The resulting product was purified by silica gel chromatography (0-20% methanol in dichloromethane), yielding 5.4 g of the desired compound (82% yield). Physical and spectral data were as described elsewhere.^{5,6}

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REFERENCES

1. Sylvers L.A.; Wower, J. Nucleic acid-incorporated azidonucleotides: probes for studying the interaction of RNA or DNA with proteins and other nucleic acids *Bioconjugate Chem.* 1993, 4, 411-418.
2. Sproat, B.S.; Iribaren, A.M.; Güimil García, R.; Beijer, B. New synthetic routes to synthons suitable for 2'-O-allyloligoribonucleotide assembly. *Nucleic Acids Res.* 1991, 19, 733-738.
3. Long, R.A.; Robins, R.K.; Townsed, L.B. Purine nucleosides. XV. The synthesis of 8-amino- and 8-substituted aminopurine nucleosides. *J. Org. Chem.* 1967, 32, 2751-2756.

4. Bartra, M.; Urpí, F.; Vilarrasa, J. New synthetic tricks. Two useful reagents for the reduction of azides to amines. *Tetrahedron Lett.* 1987, 28, 5941-5944.
5. Kawai, K.; Saito, I.; Sugiyama, H. Stabilization of Hoogsteen base pairing by introduction of amino group at the C8 position of adenine. *Tetrahedron Lett.* 1998, 39, 5221-5224.
6. Güimil García, R.; Ferrer, E.; Macias, M.J.; Eritja, R.; Orozco, M. Theoretical calculations, synthesis and base-pairing properties of oligonucleotides containing 8-amino-2'-deoxyadenosine. *Nucleic Acids Res.* 1999, 27, 1991-1998.
7. Aviñó, A.; Frieden, M.; Morales, J.C.; de la Torre, B.G.; Güimil García, R.; Azorín, F.; Gelpí, J.L.; Orozco, M.; González, C.; Eritja, R. Properties of triple helices formed by parallel-stranded hairpins containing 8-aminopurines. *Nucleic Acids Res.* 2002, 30, 2609-2619.
8. Aviñó, A.; Morales, J.C.; Frieden, M.; de la Torre, B.G.; Güimil García, R.; Cubero, E.; Luque, F.J.; Orozco, M.; Azorín, F.; Eritja, R. Parallel-stranded hairpins containing 8-aminopurines. Novel efficient probes for triple-helix formation. *Bioorg. Med. Chem. Lett.* 2001, 11, 1761-1763.
9. Cubero, E.; Aviñó, A.; de la Torre, B.G.; Frieden, M.; Eritja, R.; Luque, F.J.; Gonzalez, C.; Orozco, M. Hoogsteen-based parallel-stranded duplexes of DNA. The effect of 8-amino derivatives. *J. Am. Chem. Soc.* 2002, 124, 3133-3142.
10. Fàbrega, C.; Güimil García, R.; Díaz, A.R.; Eritja, R. Studies on the synthesis of oligonucleotides containing photoreactive nucleosides: 2-azido-2'-deoxyinosine and 8-azido-2'-deoxyadenosine. *Biol. Chem.* 1998, 379, 527-533.
11. Liu, J.; Fan, Q.R.; Sodeoka, M.; Lane, W.S.; Verdine, G.L. DNA bending by an amino acid residue in the C-terminal half of the Rel homology region. *Chem. Biol.* 1994, 1, 47-55.
12. Liu, J.; Verdine, G.L. Synthesis of photoactive DNA: incorporation of 8-bromo-2'-deoxyadenosine into synthetic oligodeoxynucleotides. *Tetrahedron Lett.* 1992, 33, 4265-4268.
13. Wood, M.L.; Esteve, A.; Morningstar, M.L.; Kuziemko, G.M.; Essigmann, J.M. Genetic effects of oxidative DNA damage: comparative mutagenesis of 7,8-dihydro-8-

oxoguanine and 7,8-dihydro-8-oxoadenine in *Escherichia coli*. *Nucleic Acids Res.* 1992, 20, 6023-6032.

14. Holmes, R.E.; Robins, R.K. Purine nucleosides. IX. The synthesis of 9-β-D-ribofuranosyl uric acid and other related 8-substituted purine ribonucleosides. *J. Am. Chem. Soc.* 1965, 87, 1772-1776.

15. Sarfati, S.R.; Naname, A. Synthesis of fluoresceinylated 2'-deoxyadenosine. Mono, di and triphosphate derivatives. *Tetrahedron Lett.* 1990, 31, 2581-2584.

16. Sheradsky, T. Azides as synthetic starting materials. In *The chemistry of the azido group*; Patai, S. Ed.; Interscience Publishers, London 1971; pp 339-340.

17. Johnson, J.A.; Thomas, H.J.; Schaeffer, H.J. Synthesis of potential anticancer agents. XIII. Ribosides of 6-substituted purines. *J. Am. Chem. Soc.* 1958, 80, 699-702.

18. Mathé, C.; Lioux, T.; Gosselin, G. Synthesis and biological evaluation of β-D-pentofuranonucleoside derivatives of 2-azidoadenine. Abstracts of papers, 15th International Round Table on Nucleosides, Nucleotides and Nucleic Acids, Leuven, Belgium, Sept 10-14, 2002.

19. MacBride, L.J.; Kierzek, R.; Beaucage, S.L.; Caruthers, M.H. Amidine protecting groups for oligonucleotide synthesis. *J. Am. Chem. Soc.* 1986, 108, 2040-2048.

20. Vu, H.; Mc Collum, A.; Jacobson, K.; Thiesen, P. Fast oligonucleotide deprotection phosphoramidite chemistry for DNA synthesis. *Tetrahedron Lett.* 1990, 31, 7269-7272.

21. Saladino, R.; Mincione, E.; Crestini, C.; Mezzetti, M. Transformations of thiopyrimidine and thiopurine nucleosides following oxidation with dimethyldioxirane. *Tetrahedron* 1996, 52, 6759-6780.