

This article was downloaded by:

On: 26 January 2011

Access details: *Access Details: Free Access*

Publisher *Taylor & Francis*

Informa Ltd Registered in England and Wales Registered Number: 1072954 Registered office: Mortimer House, 37-41 Mortimer Street, London W1T 3JH, UK



Nucleosides, Nucleotides and Nucleic Acids

Publication details, including instructions for authors and subscription information:

<http://www.informaworld.com/smpp/title~content=t713597286>

Expeditious and Stereoselective Synthesis of α -Guanosine

Allen B. Reitz^a; Mary C. Rebarchak^a

^a Medicinal Chemistry Department, The R. W. Johnson Pharmaceutical Research Institute Spring House, Pennsylvania

To cite this Article Reitz, Allen B. and Rebarchak, Mary C.(1992) 'Expeditious and Stereoselective Synthesis of α -Guanosine', *Nucleosides, Nucleotides and Nucleic Acids*, 11: 5, 1115 — 1121

To link to this Article: DOI: 10.1080/07328319208021172

URL: <http://dx.doi.org/10.1080/07328319208021172>

PLEASE SCROLL DOWN FOR ARTICLE

Full terms and conditions of use: <http://www.informaworld.com/terms-and-conditions-of-access.pdf>

This article may be used for research, teaching and private study purposes. Any substantial or systematic reproduction, re-distribution, re-selling, loan or sub-licensing, systematic supply or distribution in any form to anyone is expressly forbidden.

The publisher does not give any warranty express or implied or make any representation that the contents will be complete or accurate or up to date. The accuracy of any instructions, formulae and drug doses should be independently verified with primary sources. The publisher shall not be liable for any loss, actions, claims, proceedings, demand or costs or damages whatsoever or howsoever caused arising directly or indirectly in connection with or arising out of the use of this material.

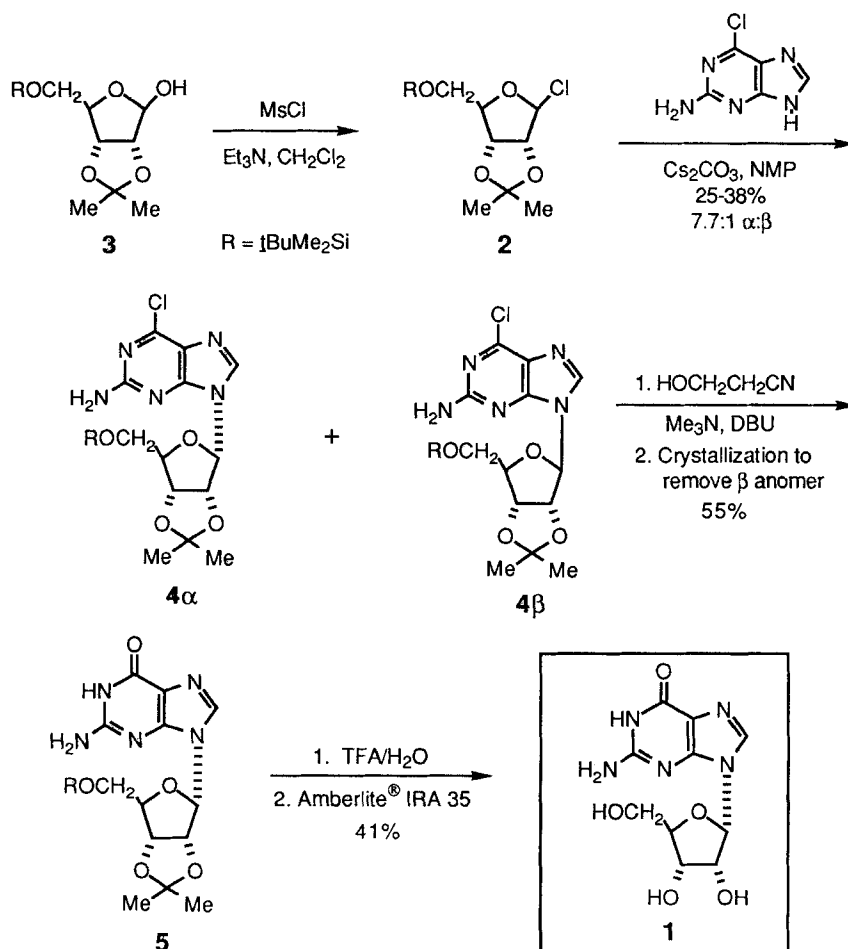
Expedition and Stereoselective Synthesis of α -Guanosine

Allen B. Reitz* and Mary C. Rebarchak

Medicinal Chemistry Department
The R. W. Johnson Pharmaceutical Research Institute
Spring House, Pennsylvania 19477

Abstract: The preparation of the unnatural nucleoside α -guanosine (**1**) has been achieved from readily available, literature precursors in ca. 6-9% overall yield. The key step, construction of the α -anomeric bond between the purine and the sugar, was accomplished by S_N2 displacement of protected β -chlororibose derivative **2** with 2-amino-6-chloropurine. The optimal conditions for this reaction involved cesium carbonate in N-methylpyrrolidinone (α/β ratio: 7.7:1).

α -Nucleosides are of considerable interest as analogs of the essential, natural β -nucleosides. Several α -nucleosides display interesting biological activity and physical properties of their own.¹ Among the α -anomers of key purine and pyrimidine nucleosides, α -guanosine (**1**) has proven to be particularly difficult to prepare. The earliest reported synthesis of α -guanosine entailed a non-stereoselective fusion reaction with a derivatized guanine followed by tedious separation of regio- and stereoisomers.^{2,3} Although this method has only a few chemical steps, the separation required is difficult because the other products of the reaction have similar chromatographic properties. Recently, Imbach and colleagues reported an improved preparation, in which **1** was obtained in eight chemical steps from D-ribose in ca. 7% overall yield.⁴ The main advantage of the Imbach process is that the α -stereochemistry was fixed early in the synthesis by virtue of a 1,2-oxazolidine ring on the ribose moiety. The purine was then constructed from the α -nitrogen at C-1 via a multi-step sequence. One disadvantage to this process is the requirement for a high-pressure reaction vessel.



Scheme I

We report here the synthesis of **1** from easily prepared **2**, as shown in Scheme I. Since **2**, first described by Wilcox and Otoski,⁵ lacks a participating substituent on the 2-hydroxyl, we anticipated that displacement of the chlorine atom would occur stereoselectively to give mainly the desired α -isomer. Indeed, Seela and co-workers have shown that such a displacement leads predominantly to the α product in related reactions.⁶ After manipulation of the purine and deprotection of the ribose, target **1** can be obtained stereochemically pure in 6-9% overall yield for the three chemical steps.

For our synthetic plan to be successful, it was crucial to prepare **2** with a high β : α ratio. In the Wilcox paper,⁵ β -anomer **2** was obtained either by thermal equilibration of the α anomer (90°C neat, 24 h), or by treatment of the anomeric hydroxyl of starting furanose **3**⁷ with CCl_4 and triphenylphosphine. In either case, we encountered difficulties in the work-up of **2**, as reversion to **3** was a pernicious side-reaction. Alternatively, compound **3** (>95% β -anomer by NMR) was treated with mesyl chloride and triethylamine in methylene chloride. After mixing the reaction at 0°C followed by stirring at 23°C for 16 h, and then passing the material through a plug of alumina and Dicalite®, chloride **2** was obtained in a pure state as determined by NMR ($J_{1,2} = 0 \text{ Hz}$ ⁵). This method was consistently reproducible with yields in the 70-80% range. Less than 5% α -anomer ($J_{1,2} = 4.1 \text{ Hz}$ ⁵) was seen in the 400-MHz ^1H NMR spectrum. Separate ^1H NMR experiments performed at 0°C, followed by warming to room temperature, showed that the β -mesylate was initially formed, but then it quickly was displaced by chloride ion to give a mixture that was predominantly α -chloro sugar. The α -chloro sugar subsequently equilibrated at room temperature to a 95:5 mixture of β : α -chloro sugars ($t_{1/2}$ for equilibration ca. 45 min at 0°C).

With **2** in hand, we investigated its condensation with 2-amino-6-chloropurine under a variety of conditions as shown in Table I to give mixtures of **4** α and **4** β . The optimal conditions for obtaining desired **4** α were the use of cesium carbonate as base in N-methylpyrrolidinone (NMP) at 55°C (25-38% yield, entry 1). Under such conditions, which have been reproduced on 1 to 10 g scale several times, the ratio of α : β anomers ranged from 6:1 to 8:1; one experiment conducted at 25°C resulted in a 14:1 ratio. The addition of lithium chloride to the reaction resulted in an α : β ratio of 2.2:1, attributed to equilibration of the furanosyl chlorides prior to displacement with the purine base (entry 2). Inspection of the table reveals that potassium carbonate in NMP gave an α : β ratio of 2.5:1 (entry 3). The yield was low and variable in the potassium carbonate reactions (ca. 20%). The use of sodium hydride in different solvents was unsuccessful (entries 4 and 5). The moderate yields obtained upon reaction of **2** with heterocyclic nucleophiles was also observed by Seela in his work,⁶ and is a consequence of steric hindrance between the incoming nucleophile and the adjacent isopropylidene group. Although use of 2-amino-6-chloropurine would be expected to give the N⁹-regioisomer upon ribosylation,⁸ we also conducted ^{13}C NMR experiments which supported the N⁹ structural assignment by using the method of Kjellberg and Johansson.⁹

Conversion of a 7.7:1 mixture of **4** α and **4** β to compound **5** was accomplished by reaction with 3-hydroxypropionitrile, trimethylamine, and DBU at 0°C.¹⁰ After chromatography and recrystallization, pure **5** (no β anomer detected by 400-MHz ^1H NMR) was obtained as a white solid in 55% yield. The β anomeric portion of the reaction product was removed in the recrystallization step, as it was left behind in the mother liquor. Deprotection of **5** was optimally accomplished with 3 equiv of trifluoroacetic acid in water for 48 h, which, after work up, gave the product as a colorless glass (trifluoroacetate salt). Exposing **5** to large excesses of aqueous TFA or TFA in the presence of alcoholic solvents resulted in significant cleavage of the glycosidic bond along with removal of the protecting groups. The free base of **1** was obtained upon treatment with a weakly basic ion-exchange resin.

Table I: Condensation of 2-amino-6-chloropurine with **2**

Entry	Base	Solvent	Temp.	Results
1	Cs ₂ CO ₃	NMP	55°C	4 α :4 β = 7.7:1; 25-38% yield
2	Cs ₂ CO ₃ ^a	NMP	55°C	4 α :4 β = 2.2:1; ca.20% yield
3	K ₂ CO ₃	NMP	80°C	4 α :4 β = 2.5:1; 23% yield
4	NaH	NMP	80°C	decomposition
5	NaH	DMPU	60°C	decomposition

a: 1.4 mol-equiv. of LiCl added.

In summary, this procedure offers a practical and concise method for obtaining α -guanosine in a brief period of time. Even though the overall yield from starting chlorofuranose **2** was only 6-9%, the short number of chemical steps and the relative ease in carrying out each of them makes this an attractive method for the preparation of α -guanosine (**1**).

Experimental Procedures

General Methods. The ¹H NMR spectra were recorded on a Bruker AM-400WB (400 MHz) spectrometer or a Bruker ACF-300WB (300 MHz) spectrometer; the ¹³C NMR spectra were obtained on a Bruker AM-400WB (100.6 MHz) spectrometer and were proton decoupled. Tetramethylsilane was used as an internal reference. NMR abbreviations: s, singlet; d, doublet; t, triplet; m, multiplet; v, very; b, broad. Melting points were determined in an open capillary tube with a Thomas-Hoover apparatus and are corrected. Elemental analysis were performed primarily by Galbraith Laboratories (Knoxville, TN). Chemical-ionization mass spectra (CI-MS) were recorded on a Finnigan 3300 system with ammonia as the reagent gas. Fast-atom-bombardment mass spectra (MS-FAB) were recorded on a VG 7070E high resolution mass spectrometer using an argon beam at 7 kV and 2 mA of current in a thioglycerol matrix. Solvents were analytical or reagent grade and used as purchased. The NMP and 1,3-dimethyl-3,4,5,6-tetrahydro-2(1a)-pyrimidinone (DMPU) were stored over dry 4A sieves prior to use. When appropriate, glassware was predried with a heat gun and cooled under an inert gas.

β -1-Chloro-1-deoxy-5-Q-(t-butyl)dimethylsilyl-2,3-Q-isopropylidene-D-ribofuranose (2**).** A solution of 3.0 g (9.85 mmol) of 5-Q-(t-butyl)dimethylsilyl-2,3-O-isopropylidene- β -D-ribofuranose (**3**) in triethylamine (2.8 mL, 19.7 mmol) in 10 mL of dry methylene chloride was chilled to 0°C under nitrogen. Methanesulfonyl chloride (1.01 mL, 12.8 mmol) was added dropwise. Triethylamine hydrochloride precipitated immediately. The reaction mixture was allowed to stir at 0°C for 15 min, then warmed to 0°C and stirred for 18 h under positive nitrogen pressure. The few remaining solids were removed by filtering the reaction

mixture through Dicalite®. The filtrate was evaporated in vacuo, the residue was triturated with 2:1 diethyl ether/ethyl acetate, and the triturating solvents passed through a plug of neutral alumina and Dicalite®. The filtrate was evaporated in vacuo to give 2.46 g (77%) of the title compound as a pale yellow liquid. ^1H NMR (400 MHz, CDCl_3) δ 0.09 (s, 6H), 0.91 (s, 9H), 1.34 (s, 3H), 1.47 (s, 3H), 3.77-3.83 (m, 2H), 4.33-4.38 (m, 1H), 4.83-4.86 (dd, J = 5.5 Hz and 1 Hz, 1H), 4.99 (d, J = 5.5 Hz, 1H), 6.13 (s, 1H). There was approximately 4% of the α anomer present as indicated by a doublet at δ 6.15, J = 4.1 Hz.

1-[9'-(2'-Amino-6'-chloropuriny)]-5-*Q*-(*t*-butyl)dimethylsilyl-1-deoxy-2,3-*Q*-isopropylidene-D-ribofuranose (4). A mixture of 2-amino-6-chloropurine (2.43 g, 14.2 mmol) and Cs_2CO_3 (9.84 g, 30.2 mmol) in dry *N*-methylpyrrolidinone (NMP, 20 mL) in a pre-dried flask was heated at 60°C under argon for 5 min. A solution of 4.88 g (15.1 mmol) of freshly prepared chloro sugar **2** in 10 mL of dry NMP was added and the reaction mixture was heated under argon at 60°C for 3 h. The reaction was cooled to room temperature and filtered through Dicalite®, and the filter cake was washed well with fresh NMP. The filtrate and washings were then Kugelrohr (short path) distilled to remove the NMP from the crude product (bp 50-55 °C at 0.15 mm Hg). The residue was purified by flash chromatography using flash silica gel eluting with 2:1 ethyl acetate/hexane to give 1.74 g (25%) of white foam as a 6:1 mixture of α and β anomers of **4**. ^1H NMR (400 MHz, CDCl_3) δ 0.01 (s, 1H, β -anomer), 0.08 (s, 3H, α -anomer), 0.10 (s, 3H, α -anomer), 0.82 (s, 1.5H, β -anomer), 0.92 (s, 9H, α -anomer), 1.27 (s, 3H, α -anomer), 1.35 (s, 0.5H, β -anomer), 1.43 (s, 3H, α -anomer), 1.58 (s, 0.5H, β -anomer), 3.74-3.88 (m, 2.33H, α - and β -anomers), 4.35 (s, 1.17H, α - and β -anomers), 4.78-4.81 (m, 1H, α -anomer), 4.90 (d overlapping m, J = 6 Hz, 1H, 0.17H, α - and β -anomers), 5.08-5.09 (m, 0.17H, β -anomer), 5.32 (bs, 2H, α -anomer), 5.55 (bs, 0.33H, β -anomer), 6.035 (d, J = 1.6 Hz, 0.17H, β -anomer), 6.475 (d, J = 4.3 Hz, 1H, α -anomer), 8.01 (s, 0.17H, β -anomer), 8.04 (s, 1H, α -anomer). ^{13}C NMR ($\text{DMSO}-d_6$, α -anomer) δ 17.8, 23.9, 25.2, 25.3, 25.6, 25.7, 26.8, 64.1, 79.0, 81.7, 82.6, 84.9 (C-1'), 112.6, 122.7, 141.3 (C-8), 149.2, 153.1, 159.6. The chemical shift of the C-8 carbon is consistent with the N-9 structural assignment.⁹ Anal. Calcd for $\text{C}_{19}\text{H}_{30}\text{ClN}_5\text{O}_4\text{Si}$: C, 50.04; H, 6.63; N, 15.35. Found: C, 49.52; H, 6.60; N, 14.93.

5'-*Q*-(*t*-Butyl)dimethylsilyl-2',3'-*Q*-isopropylidene- α -D-guanosine (5). A solution of **4** (1.51 g, 3.31 mmol) and 3-hydroxypropionitrile (1.13 mL, 16.5 mmol) in methylene chloride (12 mL) was chilled to 0°C, at which time 0.74 mL (4.97 mmol) of DBU and 3.3 mL of trimethylamine were added. The flask was sealed and refrigerated for 5 d. The solvents were evaporated in vacuo at 50°C to give a white glass which was purified by flash chromatography with flash silica gel and eluting with 93:7 methylene chloride/methanol. The resulting solid was recrystallized from methylene chloride/methanol to give 1.00 g (69%) of the title compound as a white powder. M.p. 327-330°C. ^1H NMR (400 MHz, $\text{DMSO}-d_6$) δ 0.10 (s, 3H), 0.11 (s, 3H), 0.92 (s, 9H), 1.26 (s, 3H), 1.37 (s, 3H), 3.77 (bs, 2H), 4.34 (bs, 1H), 4.76-4.79 (m, 1H), 4.865 (d, J = 6 Hz), 6.165 (d, J = 4.1 Hz), 6.29 (bs, 2H), 7.65 (s, 1H). UV

(CHCl₃) λ_{max} (ϵ) 234 nm (23,755). $[\alpha]_{\text{D}}^{25} = -40.5$ ($c = 0.65$, DMSO). MS (CI-NH₃) m/e 152, 438 (MH⁺, base pk); IR (KBr) ν 3499 cm⁻¹, 1696, 1628, 1596, 1532, 1369, 1253, 1208, 1163, 1123, 1076.

α -D-Guanosine (1), Trifluoroacetate Salt. A solution of **5** (0.10 g, 0.23 mmol) in H₂O (3 mL) and trifluoroacetic acid (0.05 mL, 0.69 mmol, 3 mol-equiv) was allowed to stir at room temperature for 48 hours. The solution was extracted twice with diethyl ether and the ether extracts discarded. The acidic aqueous solution was evaporated in vacuo at 20°C. The residue was treated with H₂O and evaporated twice to further remove any excess TFA, resulting in a colorless glass of **1** as the trifluoroacetate salt (105 mg). ¹H NMR (300 MHz, DMSO-*d*₆) δ 3.46-3.58 (m, 2H, H-5' and H-5'), 4.07-4.14 (m, 2H, H-4' and H-3'), 4.31 (t, $J = 5.1$ Hz, 1H, H-2'), 4.30-5.80 (vbs, 4H, OH-2', OH-3', OH-5', and OH-TFA), 6.06 (d, $J = 5.2$ Hz, 1H, H-1'), 6.67-6.99 (bs, 2H, NH₂), 8.69 (s, 1H, H-8), 11.10 (bs, 1H, NH-1). MS (FAB-Thio) m/e 152 (base pk), 284 (MH⁺). $[\alpha]_{\text{D}}^{25} = +5.3$ ($c = 0.98$, H₂O).

α -D-Guanosine (1). To a solution of 80 mg (0.20 mmol) of the trifluoroacetate salt of **1** in 2 mL of water was added 1.0 g of freshly washed Amberlite® IRA-35 ion exchange resin. The mixture was stirred for 60 min. The resin was removed by passing the mixture through an 80 mesh screen. The filtrate was collected and evaporated in vacuo to give 23 mg (41%) of white solid, α -D-guanosine free base. ¹H NMR (300 MHz, DMSO-*d*₆) δ 3.43-3.57 (m, 2H, H-5' and H-5'), 4.01 (m, 1H, H-4'), 4.11 (m, 1H, H-3'), 4.24 (m, 1H, H-2'), 4.85 (t, $J = 5.2$ Hz, 1H, OH-5'), 5.30 (d, $J = 5.2$ Hz, 1H, OH-3'), 5.45 (d, $J = 5.2$ Hz, 1H, OH-2'), 6.01 (d, $J = 5.2$ Hz, H-1'), 6.42 (bs, 2H, NH₂), 7.88 (s, 1H, H-8), 10.55 (vbs, 1H, NH-1). MS (FAB-Thio) m/e 284 (MH⁺). UV (MeOH) λ_{max} (ϵ , 11,400), 270 nm (sh, ϵ , 8,100); λ_{min} 222 nm (ϵ , 3,400). The ¹H NMR and UV spectra for **1** are basically identical to that reported.⁴

Acknowledgments. We thank Diane Gauthier for helpful assistance with the NMR spectra and Dr. Bruce E. Maryanoff for useful advice.

References and Notes

- (a) Sharma, B. S.; Sanghvi, Y. S.; Jolley, W. B.; Revankar, G. R.; Robins, R. K. *Intl. J. Immunopharmacol.* **1990**, *12*, 579. (b) Gosselin, G.; Bergogne, M.-C.; De Rudder, J.; De Clercq, E.; Imbach, J.-L. *J. Med. Chem.* **1987**, *30*, 982. (c) Yamaguchi, T.; Saneyoshi, M. *Chem. Pharm. Bull.* **1984**, *32*, 1441. (d) Ekiel, I.; Darzynkiewicz, E.; Shugar, D. *Carbohydr. Res.* **1981**, *92*, 21.
- (a) Imai, K.-I.; Nohara, A.; Honjo, M. *Chem. Pharm. Bull.* **1966**, *14*, 1377. (b) Iwamura, H.; Miyakado, M.; Hashizume, T. *Carbohydr. Res.* **1973**, *27*, 149.

3. See also: Furukawa, Y.; Imai, K.-I.; Honjo, M. *Tetrahedron Lett.* **1968**, 4655.
4. Gosselin, G.; Bergogne, M.-C.; Imbach, J.-L. *Nucleosides Nucleotides* **1990**, 9, 81.
5. Wilcox, C. S.; Otski, R. M. *Tetrahedron Lett.* **1986**, 27, 1011.
6. (a) Seela, F.; Soulimane, T.; Mersmann, K.; Jurgens, T. *Helv. Chim. Acta* **1990**, 73, 1879. (b) Rosemeyer, H.; Seela, F. *Helv. Chim. Acta* **1988**, 71, 1573.
7. Kane, P. D.; Mann, J. *J. Chem. Soc., Perkin Trans. I* **1984**, 657.
8. For examples of the N-9 regioselectivity of 2-amino-6-chloropurine: (a) Harnden, M. R.; Jarvest, R. L.; Bacon, T. H.; Boyd, M. R. *J. Med. Chem.* **1987**, 30, 1636. (b) Haines, D. R.; Tseng, C. K.; Marquez, V. E. *J. Med. Chem.* **1987**, 30, 943. (c) Hanna, N. B.; Ramasamy, K.; Robins, R. K.; Revankar, G. R. *J. Heterocycl. Chem.* **1988**, 25, 1899. (d) Geen, R. G.; Grinter, T. J.; Kinsey, P. M.; Jarvest, R. L. *Tetrahedron* **1990**, 46, 6903. (e) Ref. 1b.
9. Kjellberg, J.; Johansson, N. G. *Tetrahedron Lett.* **1986**, 42, 6541.
10. Ashwell, M.; Bleasdale, C.; Golding, B. T.; O'Neill, I. K. *J. Chem. Soc., Chem. Commun.* **1990**, 955.

Received 9/12/91

Accepted 11/20/91