73. *N*-Levulination of Guanosine

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Summary

A procedure have been developed for the synthesis of the *N*-levulinoyl derivative of guanosine.

Progress in the chemical synthesis of polynucleotides has created the need for a protecting group for the amino function of nucleosides that would be stable to all the chemical manipulations leading to the synthesis of an oligonucleotide, and could be removed under conditions which do not permit cleavage of the chain.

In the previous paper [1] we described the successful synthesis of the *N*-levulinoyl derivatives of cytidine, adenosine and guanosine. However in the guanosine case only very severe conditions resulted in the low yield (20%) of *N*-levulination. Since the levulinoyl group showes excellent promise for use in this capacity in nucleotide synthesis, it seemed essential to find an alternative procedure for high yield formation of *N*-levulinoyl guanosine. The present paper represents our achievement toward this goal.

After evaluating several alternatives we found that trisilylguanosine (**1**) [2] could be acylated to compound **2**, in excellent yield, using 4,4-(ethylenedithio)-pentanoyl chloride [3] in pyridine. It should be noted that the 4,4-(ethylenedithio)-pentanoyl group was previously introduced to carbohydrates by *van Boom et al.* [3], as a masked levulinoyl protective group in the synthesis of oligosaccharides.

Table. *Properties of guanosine derivatives*

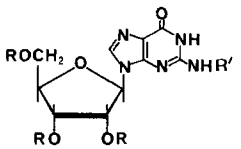
Compound	M.p. [°C]	UV. λ_{max} [nm] (EtOH)	IR. [cm^{-1}] (CH_2Cl_2)		Rf in acetone/ $CHCl_3$ 9:1
			amide	ketone	
2	130–131	281, 256	1700	–	0.82
3	95–98	283, 257	1705	1728	0.77
4	241–245 (dec.)	285, 259	1680	1702	0.06

$$Lev(S) = CH_3-C-(CH_2)_2-CO$$

$$Lev(O) = CH_3-CO-(CH_2)_2-CO$$

The cleavage of the dithioacetal group in compound **2** was achieved by means of $HgCl_2/HgO$ [4] in aqueous acetone, gave, after workup and purification, compound **3** which was in every aspect identical with the authentic sample, prepared and characterized before [1]. Compound **3** was converted into *N*-levulinoylguanosine (**4**) using tetrabutylammonium fluoride in tetrahydrofurane (THF) [5]. Compound **4** was also identical with an authentic sample [1]. The results are summerized in the *Table.*

The *N*-levulinoyl group was readily removed using the standard hydrazine solution (0.5 N hydrazine hydrate in pyridine/acetic acid 4:1) for 15 min [6].



1 R = (*t*-Bu)Me_2Si, R' = H
2 R = (*t*-Bu)Me_2Si, R' = Lev(S)
3 R = (*t*-Bu)Me_2Si, R' = Lev(O)
4 R = H, R' = Lev(O)

Experimental Part

General Procedures. Reagent grade pyridine was distilled first from *p*-toluenesulfonyl chloride, redistilled from calcium hydride and stored over molecular sieves *(type 4A).*

Short Column Chromatography: Short columns of *Merck silica gel 60* (230–400 mesh) were packed in glass columns 2 cm in diameter using 15 g of silica gel per gram of crude mixture; columns were washed first with a low polarity solvent; the desired products were then eluted with a more polar solvent (solvents used are indicated for each preparation described below).

Thin-layer chromatographic data (Rf values) are recorded from *Merck Kieselgel 60 F 254* analytical sheets. Melting points were determined on a *Fisher-Johns* melting point apparatus and are reported uncorrected. All UV. spectra were recorded on a *Cary 118* spectrophotometer. IR. spectra were obtained on a *Beckman IR. 8* spectrophotometer.

Preparation of N-*[4,4-(ethylenedithio)pentanoyl]-2',3',5'-tris(t-butyldimethylsilyl)guanosine* (**2**). To a solution of compound **1** (624 mg, 1 mmol) in pyridine (5 ml) was added over 15 min at 0° 4,4-(ethylenedithio)pentanoyl chloride (2 g, 10 mmol). After 24 h stirring at 25°, TLC. analysis showed the reaction to be complete. The solution was concentrated at reduced pressure. A 5% $NaHCO_3$-solution (20 ml) was added and the resulting solution was extracted with AcOEt (30 ml). The org. phase was washed with 4% HCl-solution (20 ml), water (20 ml), then dried over $MgSO_4$ and evaporated to leave the crude product. Chromatography on silica gel and elution with CH_2Cl_2 removed impurities and the product **2** was eluted with $CHCl_3$/AcOEt 1:1 (80%). Properties of **2** are listed in the *Table.*

Preparation of N-*levulinoyl-2',3',5'-tris(t-butyldimethylsilyl)guanosine* (**3**) *and* N-*levulinoyl guanosine* (**4**). To a stirred solution of **2** (778 mg, 1 mmol) in acetone/water 1:9 (12 ml) was added HgO (0.6 g, 2.3 mmol) and $HgCl_2$ (0.75 g, 2.3 mmol). After 6.5 h at 25°, TLC. analysis showed the reaction to be complete. The mercury salts were filtered off and washed with acetone. The concentrated filtrate was dissolved in $CHCl_3$ (50 ml) and washed with 1M aqueous KBr (2×50 ml) and water (30 ml). The

organic layer was dried ($MgSO_4$) and evaporated. The residue was applied to a column of silica gel and eluted successively with CH_2Cl_2, $CHCl_3$ and AcOEt. Compound **3** was eluted with AcOEt (75%). Properties of **3** are listed in the *Table*.

Compound **3** was converted into *N*-levulinoyl guanosine (**4**) in 65% yield using Bu_4NF in THF [5].

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REFERENCES

[1] *K. K. Ogilvie, M. J. Nemer, G. H. Hakimelahi, Z. A. Proba & M. Lucas,* Tetrahedron Lett. *23*, 2615 (1982).
[2] *K. K. Ogilvie, A. L. Schifman & C. L. Penney,* Can. J. Chem. *57*, 2230 (1978).
[3] *H. J. Koeners, C. H. M. Verdegaal & J. H. van Boom,* J. Roy. Neth. Chem. *Soc.* 3, 118 (1981).
[4] *B. T. Grobel & D. Seebach,* Synthesis *1977*, 357.
[5] *G. Just, G. H. Hakimelahi, A. Ugolini & R. Zamboni,* Synth. Commun. *9*, 113 (1979).
[6] *J. H. van Boom & P. M. J. Burgers,* Tetrahedron Lett. *17*, 4875 (1976).