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THE REACTION OF GUANOSINE AND DEOXYGUANOSINE WITH GLYCIDALDEHYDE

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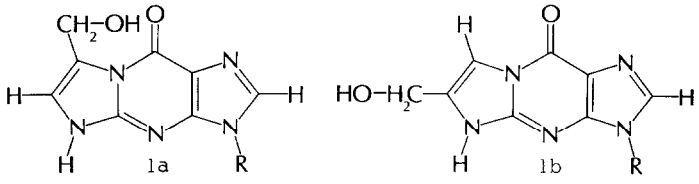
The purine moiety of guanosine is known to react with epoxides and β-propiolactone at the 7-nitrogen (1, 2), with N-acetoxy-N-2-fluorenylacetamide at the 8-carbon (3); and reversibly with formaldehyde (4, 5, 6) at the amino group of the 2-position, and with 1,2-dicarbonyl compounds at both the 1-nitrogen and the amino group of the 2-position (7). This report describes the reaction of glycidaldehyde, $H_2\overset{O}{\overset{/\backslash}{C}}CHCHO$, with guanosine and 2'-deoxyguanosine to give a Δ^4-imidazoline ring fused to the pyrimidine ring of guanosine.

Guanosine was allowed to react with one equivalent of glycidaldehyde at pH 10.0 at room temperature for 0.5 hr. and then acidified to pH 8.0. This yielded 1a or 1b, R = 1-β-D-ribofuranosyl, in 57% yield, recrystallized from $DMSO/H_2O$. The reaction of 2'-deoxyguanosine with glycidaldehyde under the same conditions yielded 1a or 1b, R = 1-(2'-deoxy-β-D ribofuranosyl), in 62% yield, recrystallized from $EtOH/H_2O$ The reaction also takes place in neutral media, but at a much slower rate.



R = 1-β-D-ribofuranosyl
R = 1-(2'-deoxy-β-D-ribofuranosyl)

Spectroscopic studies helped to establish that 2-amino-6-hydroxy purines react via the 1-nitrogen and 2-amino group with glycidaldehyde. The absorption maxima of 1a or 1b (see Table I), in comparison to guanosine and deoxyguanosine, showed a hypochromic shift of the principle purine absorption band, a new higher intensity band at shorter wavelength, and a second new band of comparable intensity at longer wavelength. When 8-bromoguanosine, 7-(2-hydroxyethyl)guanine (1) and guanine were allowed to react with glycidaldehyde in base, similar spectral shifts to those described above were seen. These findings indicated that the 7-, 8- and 9-positions of guanosine are not involved in the reaction. The ultraviolet spectra of adenosine, uridine, cytidine, N_2-dimethylguanine (8), N_1-methylguanine (9), and 2-amino-6-chloropurine in the presence of base and glycidaldehyde showed little or no change. The IR spectra, KBr pellets, of 1a or 1b had a band at 1708 cm^{-1} indicating that a carbonyl group was still present (10).

The NMR spectra of 1a or 1b in DMSO-d_6 (TMS, internal standard, δ = 0.00 ppm) and basified D_2O (TMS, external standard, δ = 0.00 ppm) aided in the structural assignment. In DMSO-d_6 the N_2-amino group of guanosine and deoxyguanosine appeared as a two-proton singlet at 6.47 δ and 6.43 δ, respectively. These peaks disappeared in 1a or 1b, and two new singlets when R = 1-β-D-ribofuranosyl at 7.26 δ (one proton) and 4.90 δ (two protons), and at 7.27 δ (one proton) and 4.89 δ (two protons) when R = 1-(2'deoxy-β-D-ribofuranosyl) appeared. That these protons were not amino or hydroxy protons was confirmed by the spectra of 1a or 1b in basified D_2O. R = 1-β-D-ribofuranosyl: 8.44 δ singlet, 1H (C_8-proton); 7.58 δ singlet, 1H (vinyl proton); 6.29 δ doublet, 1H ($C_{1'}$ proton); and 5.54 δ singlet, 2H (allylic protons). R = 1-(2'-deoxy-β-D-ribofuranosyl): 8.33 δ singlet, 1H (C_8-proton); 7.53 δ singlet, 1H (vinyl proton); 6.75 δ triplet, 1H ($C_{1'}$ proton); and 5.37 δ singlet, 2H (allylic protons). The NMR spectra of the remaining protons of 1a or 1b in D_2O were similar to those of the parent nucleosides.

TABLE I

Compound	Calcd., %	Found, %	0.1N HCl λmax, mμ(ε)	0.1N NaOH λmax, mμ(ε)	R_f in solvents[1] A	B
$C_{13}H_{15}N_5O_6$	C) 46.29 H) 4.48 N) 20.77	C) 46.19 H) 4.52 N) 20.62	300 sh(7,100) 275 (9,800) 225 (24,000)	310 (6,700) 283 (6,800) 236 (25,000)	0.27	0.23
$C_{13}H_{15}N_5O_5$	C) 48.60 H) 4.71 N) 21.80	C) 48.02 H) 4.80 N) 21.88	297 (7,100) 273 (8,100) 224 (24,700)	309 (8,000) 283 (7,400) 235 (28,800)	0.24	0.30

1) Data are for thin layer chromatography on cellulose powder.
Solvent A: 2-propanol-conc. hydrochloric acid-water (13/3.3/3.7 v/v).
Solvent B: ethanol-conc. ammonium hydroxide-water (8.0/0.1/1.8 v/v).

Supporting the analytical data (see Table I) for the assignment of structures 1a or 1b are the U.V. absorption maxima listed in Table I. These suggest that the anionic and cationic forms of 1a or 1b have a more extensive chromophore than simple alkylated guanines (11). The fluorescence spectrum of 1a or 1b,R = 1-β-D-ribofuranosyl, in base gave additional evidence for the new unsaturated site, as it had an excitation maxima at 334 mμ and an emission maxima at 420 mμ, which is at longer wavelengths than those reported for simple purines (12).

Whether the reaction product has the hydroxymethyl group alpha (1a) or beta (1b) to the guanine 1-position cannot be decided with our present evidence. However, the reaction of glycidaldehyde with the guanine moiety probably proceeds by the aldehyde reacting with one nitrogen, and the other nitrogen acting as a nucleophile to open the epoxide and thus form the five-membered ring. The reactivity of the 2-amino group of guanosines with aldehydes is well established (6, 7) and this would indicate that the alpha isomer would form. However, the formation of the beta isomer cannot be excluded, as recent work (13) showed that formaldehyde reacts readily with imino nitrogen groups. The role of the base in the reaction may be to facilitate the dehydration and double bond isomerization, or to enhance the nucleophilicity of the 1-nitrogen by removal of the proton bonded to it, forming the anion. The function of the base in this reaction is probably not the rearrangement of glycidaldehyde (14, 15). This was shown when no 1a was obtained when guanosine was allowed to react with glyceraldehyde, dihydroxyacetone, and methylglyoxal (7), respectively at pH 10.

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