New stereoselective reaction of methylglyoxal with 2-aminopyridine and adenine derivatives: Formation of imino acid-nucleic base derivatives in water under mild conditions

Christel Routaboul,a Lionel Dumas,a Isabelle Gautier-Luneau,b Jacques Vergne,c Marie-Christine Maurela and Jean-Luc Décout*a

a Laboratoire de Chimie Bio-organique, Département de Pharmacochimie Moléculaire, UMR 5063 CNRS/Université Joseph Fourier-Grenoble I, Domaine de la Merci, F-38706 La Tronche Cedex, France.
E-mail: Jean-Luc.Decout@ujf-grenoble.fr; Fax: 33 4 76 04 10 07; Tel: 33 4 76 63 74 57
b LEDSS 2, UMR 5616 CNRS/Université Joseph Fourier-Grenoble I, BP53, F-38041 Grenoble Cedex 9, France; Fax: 33 4 76 51 42 51; Tel: 33 4 76 51 44 67
c Laboratoire Biochimie de l’Évolution et Adaptabilité moléculaire, Institut Jacques Monod, Tour 43, 2 place Jussieu, F-75251 Paris Cedex 05, France; Fax: 33 1 44 27 40 21; Tel: 33 1 44 27 59 94

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A remarkable stereoselective reaction of methylglyoxal with 2-aminopyridine, the nucleic base adenine and adenine nucleosides leads in good yield to heterocycles of a new family in water under mild conditions and should be of interest in the understanding of the biological effects of methylglyoxal which is toxic, mutagenic and involved in diabetic complications.

Methylglyoxal (MG) 1 (pyruvic aldehyde, 2-oxopropanal, Scheme 1) is an interesting bifunctional reagent in organic synthesis. For instance, self-condensation1 or condensation with other aldehydes2b by a C–C bond leads to dicarbonyl intermediates interesting in synthesis and MG can be used in the preparation of various heterocycles.2c

MG can be formed in vivo by slow glucose degradation under physiological conditions3 and it appears to be involved in the development of diabetic complications, in mutagenesis and apoptosis.3,4 Recently, it was reported that MG may function as a signal molecule during the regulation of cell death.4c Reactions with cysteine, lysine and arginine residues in proteins3,4 and with guanine in DNA and RNA have been reported.4a,b Very little has been published on the reaction with the nucleic base adenine4a,b however formation of cyclic4a or acyclic4b monoadducts with 2-aminopyridine has been described under different conditions. On the basis of these results, a reaction of glyoxal or MG with adenine derivatives at 100 °C in propan-2-ol containing HCl or tungstosilicic acid has been reported to detect DNA.4a Treatment of the uncharacterized products induces chemiluminescence.

We report here a new stereoselective reaction of MG with 2-aminopyridine (AP), adenine and adenine nucleosides that occurs in water under mild conditions and leads in good yields to heterocycles of a new family.

The reaction of MG with AP was first investigated under argon at 50 °C using the commercial 40% acidic aqueous solution containing different impurities (pH 5, 0.75 M AP, 8 equiv. MG). After 12 h, the reaction of AP was complete and led essentially to two compounds absorbing in the UV region essentially to two compounds absorbing in the UV region (Fig. 1, Scheme 2). This ring results from condensation of two MG molecules by a C–C bond leading to the isomers 3 and 4 detected in a 60:40 ratio and isolated after further chromatography and crystallisation respectively in 32 and 26% yields. Surprisingly, the new ring bears a carboxylate group, two methyl groups and two hydroxy functions located on three successive asymmetric carbon atoms. In both isomers, the methyl groups are trans. The adjacent hydroxy functions are cis in the major isomer 3 and trans in the minor isomer 4 (X-ray studies revealed the presence of racemic mixtures).

The reaction conducted at pH 8 or 4 (acidification with acetic or sulfuric acid) with a dilute aqueous solution of MG freshly prepared5 gave the same products (HPLC, NMR and mass spectra). The same reaction also occurs at 50 °C but more slowly than at 50 °C.

Scheme 1 Structures of methylglyoxal and adenine.

Scheme 2 Structures of the methylglyoxal adducts formed from 2-aminopyridine, adenine, adenosine and 2’-deoxyadenosine.
The reaction was conducted with adenine 2 and an aqueous commercial solution of MG under argon at 50 °C (pH 4.0, 0.82 M adenine, 7 equiv. MG). After complete reaction (18 h), two compounds absorbing in the UV were detected and isolated in a 70:30 ratio after chromatography in 46 and 20% yields, respectively. The X-ray structure of the major product was obtained and confirmed the expected structure 5′d corresponding to the major AP adduct 3 (Fig. 2, Scheme 2). The minor product could not be crystallized but its characteristics indicate clearly the structure 6 related to that of the corresponding minor AP isomer (Scheme 2). The same reaction was observed with a dilute aqueous solution of MG freshly prepared, at pH 4 (addition of aqueous H₂SO₄) or under neutral conditions.

The reaction was performed at pH 4 with adenine or 2′-deoxyadenosine and the commercial concentrated solution of MG. For each nucleoside, two isomers were selectively formed and isolated. They were characterised as compounds 7 and 9 (major adducts: 47 and 20% yields) and 8 and 10 (minor adducts: 8 and 7% yields), respectively, by comparison of their spectral characteristics with those of the corresponding AP and adenine adducts (Scheme 2). The low yields obtained in minor adducts can be explained by difficulties in the purification procedure. Analysis of the 1H and 13C NMR spectra of the adenine adducts (Scheme 2). The minor yields obtained in the adducts: 8 and 7% yields), respectively, by comparison of their signals of equal intensity that indicates the presence of the two nucleoside adducts showed the splitting of some peaks in two fold excess of MG, phosphate buffer). Formation of similar adducts was observed with 9-propyladenine and the cytosine base (1H, 13C NMR, LRMS for each purified adduct) and with polyA.11

In conclusion, a new stereoselective reaction of methylglyoxal with 2-aminopyridine and adenine derivatives was evidenced in water under mild conditions. This reaction which leads to heterocycles of a new family in good yield should present interest in organic synthesis and by its mechanism which remains to be elucidated. The condensation of two MG molecules by a C−C bond to form a new ring is also remarkable by its stereoselectivity in regard to the number of reactive sites.

The reactions with adenine nucleosides, polyA and cytosine evidenced under physiological conditions show the capability of MG to react with different bases than guanine and in a different way to that previously described.4,5a,b,c These reactions should be interesting in the understanding of some of the biological effects of methylglyoxal. The new imino acid adenine derivatives 5 and 6 possess interesting catalytic activity in the model hydrolysis of p-nitrophenylacetate12 and could have been intermediates in prebiotic chemistry.13

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Notes and references


7 Crystal data: C₁₁H₁₂N₂O₄M₀, m = 238.24, monoclinic, a = 6.66(3), b = 11.32(3), c = 14.50(3) Å, β = 108.6(5), β = 293 K, space group P2₁/n, Z = 4, μ(Mo-Kα) = 0.112 mm−1, 3464 reflections measured of 3349 unique reflections, 2593 were obtained (F > 3σ(F)) and used in the full matrix least-squares refinement of 154 refined parameters. R(F) = 0.037, Rw(F) = 0.051, goodness of fit S = 1.88. CCDC 1706969–170700. See http://www.rsc.org/suppdata/cc/b2/b201901a/ for electronic files in cif or other electronic format.

8 Crystal data: C₁₁H₁₂N₂O₄M₀, m = 238.24, monoclinic, a = 7.809(2), b = 13.97(4), c = 9.99(1) Å, β = 106.0(4), β = 293 K, space group P2₁/n, Z = 4, μ(Mo-Kα) = 0.112 mm−1, 3457 reflections measured, 2035 unique, R(F) = 0.038 (for 1497 F > 3σ(F) and 154 refined parameters), R(Fw) = 0.052, S = 1.99.

9 The solution of aqueous MG substantially free of impurities was freshly prepared by acidic hydrolysis of methylglyoxal dimethyl acetal and then distillation: M. W. Kellum, B. Oray and S. Norton, J. Anal. Biochem., 1978, 85, 586; C. Rae, J. S. Berners-Price, B. T. Bulliman and P. W. Kuchel, Eur. J. Biochem., 1990, 193, 83.

10 Crystal data: C₁₁H₁₂N₂O₄M₀, m = 315.29, orthorhombic, a = 7.42(6), b = 12.45(8), c = 13.74(7) Å, β = 1455.5(2), β = 293 K, space group P2₁2₁2₁, Z = 4, μ(Mo-Kα) = 0.118 mm−1, 9486 reflections measured, 2109 unique (Rint = 0.035, R(F) = 0.039 for 1174 F > 3σ(F) and 197 refined parameters), R(Fw) = 0.036, S = 1.56.

11 7 mM polyA, 13 mM MG, pH 7, argon atmosphere to prevent oxidation and acidification, 10 h at 37 °C; nuclease P1 and calf intestinal alkaline phosphatase digestion; HPLC analysis with a diode array detector (retention times and absorption spectra of two hydrolysis products detected identical to those of adducts 7 and 8).

12 The catalytic activities in the p-nitrophenylacetate hydrolysis measured for 2-aminopyridine adducts 3, 4, adenine, adenine adducts 5, 6 with respect to histidine were respectively 0, 0.05, 1.1, 1.2 at 20 °C, pH 7.7 (sodium phosphate buffer).