Studies on Flavin Derivatives

II.* X-Ray Structure Investigations of Two Substituted Isoalloxazinium Iodides

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Riboflavin, 7,8-dimethyl-10(D-1'-ribityl)-isoalloxazine, is the prosthetic group of a large number of enzymes with activity in biological redox processes. The physical and chemical properties of flavin derivatives in different oxidation states have in the last few years been intensively investigated.[1,2] Special attention has been paid to studies on light absorption, electron spin resonance, and binding of metal ions. The investigations conducted at this Institute are in the first place concerned with the structural effect exercized on the geometry of the isoalloxazine ring system in different states of oxidation by a number of substituents. This article will describe X-ray structure investigations of two compounds with the isoalloxazinium ring system carrying a positive charge, *viz.* 1,3,7,8,10-pentamethylisoalloxazinium iodide monohydrate ($C_{15}H_{17}N_4O_2I \cdot H_2O$) and 1,10-ethylene-7,8-dimethylisoalloxazinium iodide monohydrate ($C_{14}H_{13}N_4O_2I \cdot H_2O$). The schematic structural formula including the numbering system used for the non-hydrogen atoms of the cations is given in Fig. 1.

X-Ray diffraction data. Preliminary examinations of crystals of the two, red coloured compounds (namned I and II, *cf.* Fig. 1) by Weissenberg methods indicated the symmetries to be orthorhombic (space group $P2_12_12_1$) for compound I and monoclinic ($P2_1/c$) for II. Powder photographs, taken in a Guinier-Hägg type focusing camera with strictly monochromatized $CuK\alpha_1$ radiation ($\lambda = 1.54051$ Å) and with KCl ($a = 6.29228$ Å) as an internal standard, were used for the determination and least-squares refinement of the lattice parameters. The following results were obtained:

I: $a = 12.857 \pm 2$ Å, $b = 18.221 \pm 2$ Å, $c = 7.049 \pm 2$ Å. There are four formula units in the unit cell. The calculated density is 1.730 g/cm³.

II: $a = 11.417 \pm 2$ Å, $b = 10.788 \pm 2$ Å, $c = 13.812 \pm 3$ Å, $\beta = 114.78 \pm .02°$. There are four formula units in the unit cell. The calculated density is 1.781 g/cm³.

The intensity data were collected with a Siemens automatic diffractometer using Ni-filtered CuK radiation and a scintillation counter with pulse height discrimination. The intensities were measured by the $\theta - 2\theta$ technique. Each reflection was counted twice, and background counts were taken at both ends of the scan range. The observed intensities — 1330 independent reflections with $\sigma(I)/I \le 0.10$ for compound I and 1166 with $\sigma(I)/I \le 0.25$ for II — were corrected for absorption ($\mu = 157$ cm⁻¹ (I) and 170 cm⁻¹ (II)) and Lorentz and polarisation effects.
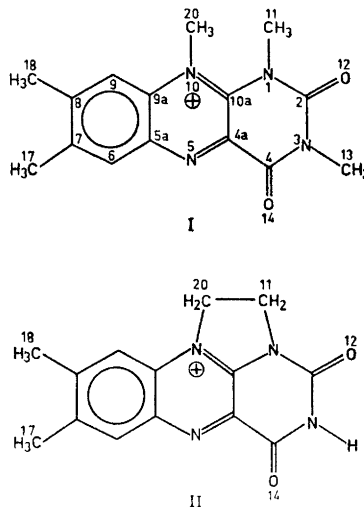

Fig. 1. Schematic structural formula of 1,3,7,8,10-pentamethylisoalloxazinium ion (I) and of 1,10-ethylene-7,8-dimethylisoalloxazinium ion (II).

Determination and refinement of the structures. The structures were determined by the heavy-atom method. The iodine positions were deduced from the Patterson function and the positional parameters of all non-hydrogen light atoms (carbon, nitrogen, and oxygen) were determined from subsequent iterative Fourier calculations. The positional parameters thus
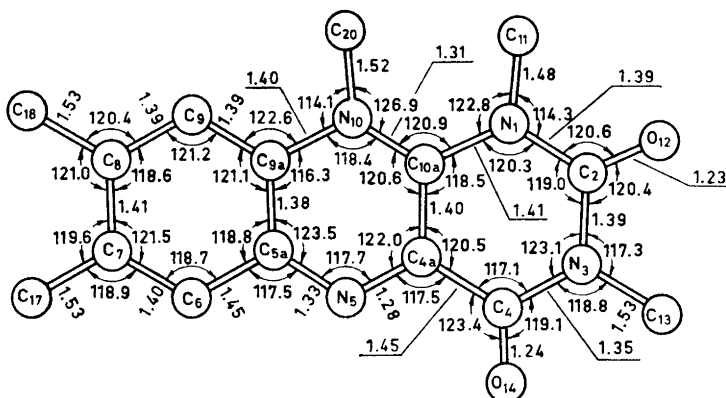
* I: *Chem. Commun.* **1967** 288.

Fig. 2. Bond lengths (in Å) and bond angles within the cation of 1,3,7,8,10-pentamethylisoal-
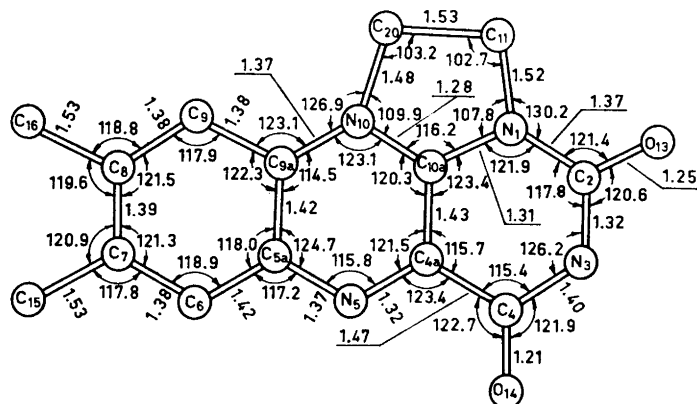loxazinium iodide monohydrate.

Fig. 3. Bond lengths (in Å) and bond angles within the cation of 1,10-ethylene-7,8-dimethyl-
isoalloxazinium iodide monohydrate.

obtained were then refined by the least-squares method using anisotropic temperature factors for the iodine atoms. The light non-hydrogen atoms were refined anisotropically for compound I and isotropically for compound II. The refinements were terminated when all parameter shifts were less than 10 % of their estimated standard deviations. At these stages the discrepancy index, R, defined as $\sum ||kF_o| - |F_c||/\sum |kF_o|$ was 0.069 (I) and 0.067 (II) for the observed reflections.

Discussion of the structures. The bond lengths ($\sigma \approx \pm 0.02$ Å) and bond angles ($\sigma \approx \pm 1.5°$), uncorrected for thermal motion, are given for the two cations in Figs. 2 and 3.

The cations form planar sheets separated by iodide ions located above and below the C(10a)—C(4a) bonds. The isoalloxazinium ring system is essentially planar in both structures. The deviation of any atom from a least-squares plane through all non-hydrogen atoms in the cation of compound II is less than 0.1 Å. However, in compound I the pyrimidine ring is slightly twisted out of this plane. The angle between two least-squares planes through the pyrimidine ring and the twelve atoms forming the remaining two rings in compound I was calculated to be 7.1°. This distortion is possibly due to repulsion effects between the methyl groups at atoms N(1) and N(10).

The distance (3.52 Å) between the water oxygen atom in compound I and the iodide ion indicates a hydrogen bond $O-H\cdots I^-$ in this structure. In compound II two interionic hydrogen bonds $N(3)-H\cdots O(14)$ with a length of 2.88 Å, related by a centre of symmetry are found. In this way, a planar eightmembered ring including the two hydrogen atoms is obtained in this structure. Some $O-O$, $O-N$ or $C-O$ contacts found in the structures may indicate further hydrogen bonds. However, since reliable hydrogen positions could not be obtained for the present heavy-atom derivatives it is not possible to make any definite conclusions about the occurrence of such bonds.

Full details of these investigations including Pariser-Parr-Pople calculations performed on the cation of compound II and discussions of the structures will shortly be presented elsewhere. Also detailed accounts of structure determinations of two reduced compounds, *viz.* 5-hydro-9-bromo-1,3,7,8,10-pentamethyl-1,5-dihydroalloxazine and 5-acetyl-9-bromo-1,3,7,8,10-pentamethyl-1,5-dihydroalloxazine will be published in the near future. Within the present research program the crystal structures of 9-bromo-3,7,8,10-tetramethylisoalloxazine, 5-acetyl-3,7,8,10-tetramethyl-1,5-dihydroalloxazine and the 1:1 complex of lumiflavin-hydroquinone containing one HCl per molecule have also been worked out and will very soon be reported.

1. Dudley, K. H., Ehrenberg, A., Hemmerich, P. and Müller, F. *Helv. Chim. Acta* **47** (1964) 1354.
2. Slater, E. C., Ed., *Flavins and Flavoproteins,* B. B. A. Library, 8, Elsevier, Amsterdam 1966.

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III.* The Crystal and Molecular Structure of 9-Bromo-5-hydro-1,3,7,8,10-pentamethyl-1,5-dihydroalloxazine

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The crystal and molecular structure of 9-bromo-5-hydro-1,3,7,8,10-pentamethyl-1,5-dihydroalloxazine, $C_{15}H_{17}BrN_4O_2$, has been determined by X-ray diffraction methods in order to obtain detailed structural information about a flavin derivative in a reduced state.

The crystalline specimen used was prepared by Maron [2] at this Institute. The crystal structure ($C2/c$, $a=15.680\pm5$ Å, $b=15.440\pm5$ Å, $c=12.845\pm5$ Å, $\beta=103.03\pm0.05°$, $Z=8$) was solved by the heavy-atom method on the basis of 2818 independent, significant reflections registered with $CuK\alpha$ radiation using a Siemens automatic single-crystal diffractometer. The structure was refined by full-matrix least-squares techniques including anisotropic thermal parameters for the non-hydrogen atoms and isotropic parameters for the hydrogens to a final R value of 0.042.

The dimensions of the molecule are given in Figs. 1—3.

The distances within ring III are in full agreement with those characteristic of a benzene ring system. The distances $C(4A)-C(10A)$, $C(2)-O(12)$ and $C(4)--O(14)$ are of lengths indicating double bond character. The ring skeleton is bent along a line through $N(5)$ and $N(10)$ the ring atoms deviating less than 0.02 Å for the pyrimidine ring and less than 0.45 Å for the benzene ring from two least-squares planes intersecting at an angle of 159°. The results support the conclusions drawn by Dudley, Ehrenberg, Hemmerich, and

* For paper II of this series see Ref. 1.