Crystal Structure of the Yellow 1:2 Molecular Complex Lumiflavin–Bisnaphthalene-2,3-diol

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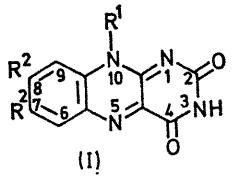
Summary In the first molecular complex of the physiologically active *neutral* form of isoalloxazine studied, lumiflavin–bisnaphthalene-2,3-diol, each flavin is sandwiched between two naphthalenediol molecules with extensive overlap but a moderately large (3·44 Å) spacing, indicating at most weak charge-transfer interaction and in agreement with the yellow colour of the complex, nearly the same as that of the parent lumiflavin.

ISOALLOXAZINE (I) is the aromatic portion of flavin co-factors, the redox-active functional groups in many enzymes including some in the vital mitochondrial electron-transport system. Some of us (Tulane) are studying simple derivatives of (I) which are chosen to be sterically and electronic



(I)

ally as similar as possible to the enzyme co-factor riboflavin (vitamin B_2; R^1 = D-ribityl and R^2 = methyl), so as to deduce the relative hydrogen-bonding effectiveness of the functional groups located in ring positions 1—5, and also to study the ability of isoalloxazine to participate in molecular π-complexes as possible models for its electron-transfer reactions. Two π-complexes of $N(1)$-protonated isoalloxazines[1] have been studied, but since N(1) is protonated only at pH $\leqslant$ O, these cannot be regarded as models of physiological systems.

Several 2:1 complexes of dihydroxynaphthalenes with various simple 10-substituted (yellow) isoalloxazines have been crystallized and with the exception of the one reported here all of the complexes are orange or red, indicating some degree of perturbation of the isoalloxazine π-system. (In fact, an orange complex of composition apparently identical to that of the yellow one reported here is also being studied.)

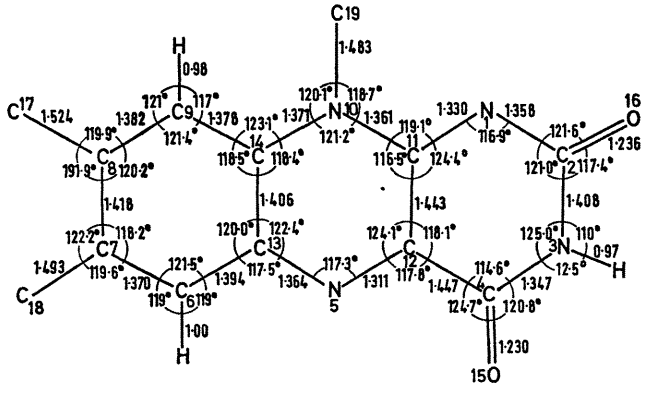


FIGURE 1. *Bond distances and angles in the lumiflavin molecule. Standard deviations are about* 0·008 Å *and* 0·8° *for bonds and angles involving heavy atoms and* 0·04 Å *and* 4° *for those involving hydrogen.*

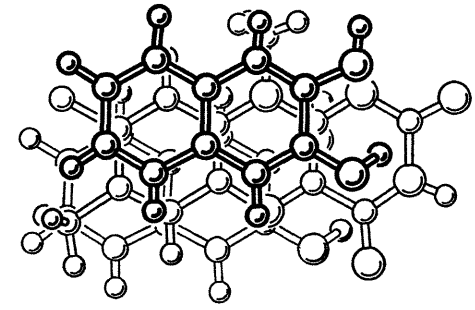


FIGURE 2. *A portion of the structure, showing a lumiflavin molecule sandwiched between two naphthalenediol molecules. The view is in a direction perpendicular to the mean plane of the flavin and nearly parallel to the* b *axis.*

The yellow complex lumiflavin–bisnaphthalene-2,3-diol, $C_{13}H_{12}N_4O_2,2C_{10}H_8O_2$, was crystallized by cooling and evaporation of an acetone–water solution nearly saturated in lumiflavin (I; $R^1 = R^2 = Me$) and containing a large molar excess of naphthalene-2,3-diol. The crystals are monoclinic, space group $P2_1/c$, with $a = 20·78(2)$, $b = 7·24(1)$, $c = 20·15(2)$ Å [λ (Cu-K_α) = 1·5418 Å], $\beta = 116·19 \pm 0·04°$, $D_c = 1·402(3)$; $Z = 4$; $D_m = 1·40 \pm 0·01$. Intensity

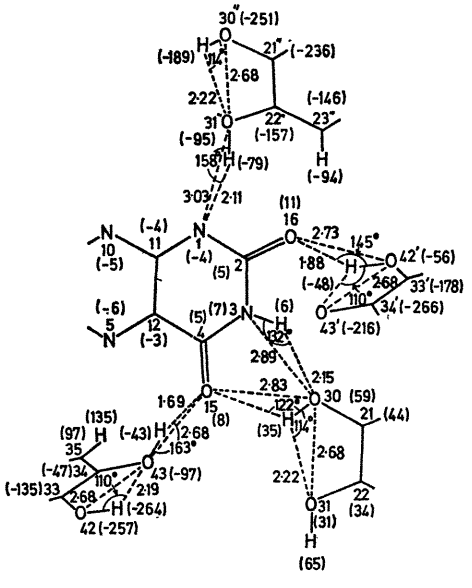


FIGURE 3. *A view perpendicular to the mean flavin plane, showing the pyrimidine end of a lumiflavin molecule surrounded by portions of four naphthalenediol molecules. Figures in parenthesis are displacements (in* Å × 10^2) *from the plane.*

measurements on a Picker 4-circle diffractometer with Ni-filtered Cu-K_α radiation to $2\theta = 100°$ yielded 2826 reflections of which 1753 were greater than 3σ above background. The structure was solved at Cal Tech by use of direct methods of phasing and was refined at Tulane to a current R value of 5·5%. All hydrogen atoms were located unambiguously on difference maps except those of the methyl group at C(8), which appears to be disordered or rotating. The molecular geometry is shown in Figure 1 and in general agrees excellently with bond lengths predicted.[2] The bond lengths also agree reasonably well with comparable ones in 9-bromo-3,7,8,10-tetramethylisoalloxazine.[3] Although the lumiflavin molecule in this structure is sandwiched between translationally related naphthalenediol molecules with extensive overlap of the π-systems (Figure 2), the moderately large spacing of 3·44 Å to *both* upper and lower molecules and the similarity in colour of this complex to lumiflavin itself indicate little perturbation of the isoalloxazine π-system (*i.e.* little charge-transfer bonding).

Of particular interest with reference to protein active-site steric requirements is the test this structure provides for the suggestion of one of us[1a] that the chelate site CO(4)–N(5) will almost always be occupied by a positive group or dipole, and for our other suggestions regarding relative hydrogen-bonding affinities. These suggestions were based entirely on studies of $N(1)$-protonated flavins, but it is seen (Figure 3) that a hydroxide group, OH(43), from a naphthalenediol molecule indeed lies essentially in the chelate site; it is hydrogen bonded to CO(4) and O(43) is 3·44 Å from N(5). A major predictable difference in hydrogen-bonding affinities of the protonated and unprotonated species is the probable high affinity of unprotonated N(1) for hydrogen-bond donors, and in fact such a bond is present. In agreement with the other suggestions, NH(3) serves as a donor and CO(4) is a double acceptor whereas CO(2) is a single acceptor.

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