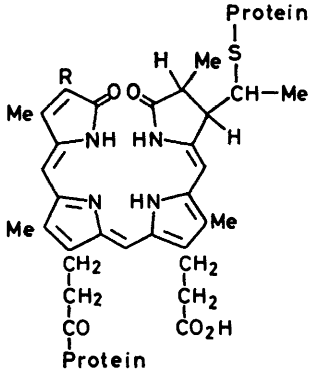
Crystal and Molecular Structure of a 4,5-Dimethyoxbilindione derived from Etiobiliverdin-IVγ: a Possible Model Compound for the Phytochrome Chromophore

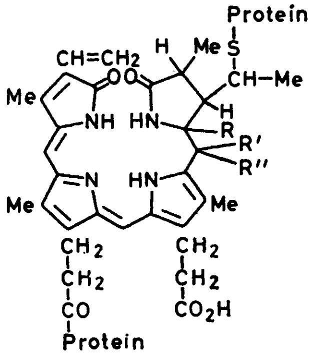
By **David L. Cullen,*** Department of Chemistry, Connecticut College, New London, Connecticut 06320, U.S.A.
Nicole Van Opdenbosch and **Edgar F. Meyer, jun.**, Department of Biochemistry and Biophysics, Texas Agricultural Experiment Station, Texas A and M University, College Station, Texas 77843, U.S.A.
Kevin M. Smith and **Fahimeh Eivazi**, Department of Chemistry, University of California, Davis, California 95616, U.S.A.

The structure of a synthetic bile pigment, 3,8,12,17-tetraethyl-4,5-dimethoxy-2,7,13,18-tetramethyl-4,5-dihydrobilin-1,19(21*H*,24*H*)-dione, $C_{33}H_{44}N_4O_4$, has been determined using three-dimensional diffractometer data. It crystallizes in the triclinic space group $P\bar{1}$ with a = 12.755 (2), b = 12.944(s), c = 12.615(2)A, α = 112.27°(2), β = 113.76°(2), γ = 75.45°(2), Z = 2. There is one-half of an n-hexane molecule of crystallization present per formula unit. The structure was solved by direct methods and refined by full-matrix least squares to yield a final R factor of 0.105. The compound, which could be considered as a model for the far-red form (P_{fr}) of the phytochrome chromphore, belongs to the class of bile pigments known trivially as bilipurpurins. Three of the four pyrrole rings form a conjugated system which is not strictly planar, but is quasi-helical in nature. The fourth ring which is joined to the others by an sp^3 carbon atom, is considerably out of the plane of the other three. Bond lengths and angles show a large degree of bond fixation. There is intermolecular hydrogen bonding so that the molecules form hydrogen-bonded dimers.

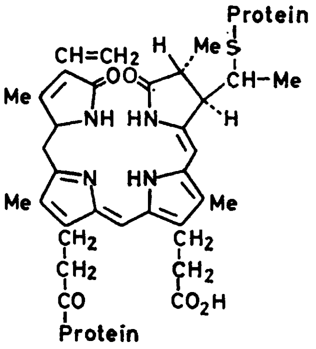
BILE pigments have been the subject of a great deal of interest in recent times. Part of this interest has been devoted to studies of the chemistry and biochemistry of mammalian bile pigments, which are produced as a result of catabolism of waste heme proteins,[1] and a deeper understanding of the factors influencing medical conditions such as jaundice and hyperbilirubinemia has resulted.[2] However, another series of bile pigments exists in plants and algae which serves important photosynthetic and growth-related purposes. Such pigments are the biliproteins phycocyanin, phycoerythrobilin, and phytochrome, the last of these being the pigment responsible for the photoregulation of growth and development in plants. The chromophore in the biliproteins closely resembles that in mammalian bile pigments, with the important difference that the former are covalently attached to a protein residue.[1] In this paper we report the crystal and molecular structure of a bilindione, 3,8,12,17-tetraethyl-4,5-dimethoxy-2,7,13,18-tetramethyl-4,5-dihydrobilin-1,19(21*H*,24*H*)-dione (I) (hereafter referred to as dimethoxybilindione), which is a model for the chromophore proposed [3] to be present in the phytochrome P_{fr} form (fr refers to ' far red '). Both the dimethoxybilindione and phytochrome P_{fr} are saturated at C(4) and C(5) and are substituted with R groups. Unlike phytochrome P_{fr}, however, the dimethoxybilindione is unsaturated in ring A. Other proposals regarding the nature of the P_{fr} chromophore have been advanced.[4] Admittedly, in the protein conformational changes can occur, but model compounds such as (I) should be capable of providing valuable information about bile pigments in general and about the biliprotein chromophores in particular. Dimethoxybilindione can also serve as a model compound for the violinoid compounds found in mammalian bile pigments.[5]



Phycocyanin R = Et
Phytochrome P_r R = CH=CH2

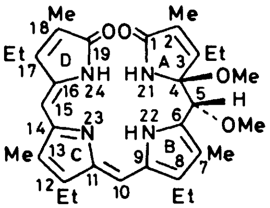


Phytochrome P_{fr}
R, R', R'', undefined



Phycoerythrin

Dimethoxybilindiones similar to (I) were first obtained by treatment of bilitrienes with bromine in methanol,[6] but more recently they have been readily accessible by oxidation of symmetrical bilitrienes, such as etiobiliverdin IVγ,[7] with thallium(III) acetate in methanol[8] or by anodic oxidation in methanol.[9] The last two methods have been proposed to involve formation of bilitriene cation-radicals, followed by addition of methanol.



(I)

EXPERIMENTAL

Brown crystals of dimethoxybilindione were grown from an n-hexane–methylene chloride solution. The crystal chosen for intensity measurements, which was irregularly shaped, was mounted in a glass capillary[10] at an arbitrary orientation that had c approximately parallel to the spindle axis.

Crystal data and some of the experimental conditions are given in Table 1. The cell dimensions were refined by least squares, minimizing the differences between observed and calculated 2θ values for 30 reflections measured at + and − settings of the 2θ angle. Cu-K_α radiation (λ 1.541 78 Å), monochromatized by pyrolitic graphite, was used in the determination of cell dimensions and in the collection of intensity data. The choice of the centric space group $P\bar{1}$ was confirmed by the successful solution of the structure.

TABLE 1

Crystal data for $C_{33}H_{44}N_4O_4\cdot\frac{1}{2}C_6H_{14}$ [a]

a = 12.755(2) Å	Mol. wt. = 603.8
b = 12.944(2) Å	Z = 2
c = 12.615(2) Å	d_{calc} = 1.14 g cm^{-3}
α = 112.27(2)°	d_{meas} [b] = 1.09 g cm^{-3}
β = 113.76(2)°	μ = 6.05 cm^{-1} (Cu-K_α radiation)
γ = 75.45(2)°	$F(000)$ = 648
U = 1 752(1)Å^3	
Systematic absences	None
Space group	$P\bar{1}$
Crystal size	0.55 × 0.30 × 0.18 mm
Scan range	3°
Scan rate	2° min^{-1}
sin θ/λ limit	0.50 (50° in θ)
Independent reflections collected	3 604
' Observed ' reflections	2 298 ($I > 2.5\ \sigma_I$)
Coincidence factor [11]	$\tau = 1.7 \times 10^{-7}$ counts^{-1}

[a] In this and subsequent Tables, the estimated standard deviation of least significant figures is shown in parentheses. Data for cell constants measured at 23 °C. [b] Measured by flotation in aqueous zinc nitrate solution. [c] Machine limit.

Intensity data were collected on a Datex-Syntex diffractometer by the θ–2θ scan method, monitoring the intensity scale by remeasuring a group of five standard reflections periodically. The intensities of these reflections remained constant within statistical limits until near the end of data collection, when they dropped markedly. Intensities of all reflections were corrected by a factor calculated from the weighted mean of the check reflections. The maximum factor was 1.22. Backgrounds at either end of the scan range were collected for half the scan time. The standard deviation in the intensities, σ_I, was determined from counting statistics. Absorption corrections were not applied. Structure factors were calculated in the usual way, assuming an ideally imperfect monochromator.

Determination and Refinement of the Structure.—The structure was solved by an automatic multisolution technique which is part of the SHELX-76 system of programs.[12] 2^{12} Sign permutations were expanded by the Σ_2 formula. 18 Atoms, mostly in the pyrrolic skeleton, were identified. The other 26 non-hydrogen atoms were found from a series of difference syntheses. These included three peaks near a centre of symmetry, which were identified as belonging to an n-hexane molecule of crystallization centred on the symmetry element.

Least-squares refinement using full-matrix least squares was carried out Initially isotropic temperature factors were used but in later refinement cycles, all non-hydrogen atoms, except those belonging to the solvent molecule, were assumed to have anisotropic thermal motion. The function minimized was $\Sigma w(F_o - F_c)^2$. After many trials, it was clear that a unit weighting scheme (w 1.0) was best for this data set.

The electron density peaks corresponding to the atoms of the solvent molecule were very diffuse. Several different ways of describing a model for this solvent molecule were tried. There was no evidence of alternate positions for the atoms, which would serve as the basis for a disorder model. Attempts to use partial occupancy factors were not successful, the values refining back to near unity. In the end the positional parameters were refined, but the thermal parameters were set to a high value (B 25 Å^2) and not refined.

After several cycles of refinement, ΔF syntheses were calculated in an attempt to locate hydrogen atoms. Most could be found, the exceptions being those on the solvent molecule and on some of the ethyl groups. However, since many of the bond parameters were chemically unreasonable, hydrogen atom positions were calculated (C–H = 0.95 Å; staggered configuration for the methyl and ethyl hydrogen atoms) for all but the three imino hydrogen atoms and the hydrogen atoms on the solvent molecule. The contributions from these calculated hydrogen atoms positions were added to the structure factor calculation, but the hydrogen atom parameters were not refined. For the imino-hydrogen atoms, the observed positions from the difference Fourier map were refined, but not the isotropic temperature factor. For all hydrogen atoms, B_H was set to 5.0 Å^2.

After the final cycle, the shifts on most refined parameters were well below 1 σ. The exceptions were some of the thermal parameters on C(24) and C(27) which have highly anisotropic thermal motion. For these the shift/error ratio was slightly over 1.0. Since it was obvious that these parameters were oscillating from cycle to cycle, further refinement was deemed unnecessary.

The final conventional R, defined as $\Sigma||F_o| - |F_c||/\Sigma|F_o|$, was 0.105 while R_w defined as $\Sigma w||F_o| - |F_c||^2/\Sigma wF_o^2$ was 0.095. A structure factor calculation using all 3 604 reflections gave R 0.156. The error in an observation of unit weight, defined as $[\Sigma w||F_o| - |F_c||^2/(N_o - N_v)]^{\frac{1}{2}}$ was 1.78 for N_o 2 298 reflections and N_v 388 variables. A final difference-Fourier synthesis showed a maximum electron density of 0.21 e A^{-3}. This was located near C(27), one of the terminal carbon atoms of an ethyl group. Neither this nor any of the other peaks on this final difference synthesis were considered to have any physical significance.

TABLE 2

Positional parameters derived from least-squares refinement

Atom	*x*	*y*	*z*
O(1)	−0.036 7(5)	0.646 9(5)	1.061 9(6)
O(2)	0.381 8(6)	0.717 9(6)	0.787 4(5)
O(3)	0.083 5(5)	0.556 0(6)	0.745 2(6)
O(4)	0.292 6(5)	0.622 7(5)	1.037 7(5)
N(21)	0.064 0(6)	0.567 0(6)	0.929 2(6)
N(22)	0.291 8(5)	0.391 4(5)	0.763 3(5)
N(23)	0.333 9(6)	0.339 7(5)	0.547 4(5)
N(24)	0.364 8(6)	0.560 6(5)	0.620 6(6)
C(1)	0.010 8(7)	0.653 4(8)	0.998 4(8)
C(2)	0.028 2(8)	0.757 7(7)	0.990 4(8)
C(3)	0.085 1(8)	0.728 3(7)	0.914 5(8)
C(4)	0.118 5(8)	0.603 6(7)	0.873 2(7)
C(5)	0.250 9(7)	0.569 4(7)	0.912 1(7)
C(6)	0.281 8(7)	0.444 4(6)	0.874 3(6)
C(7)	0.304 4(7)	0.365 9(7)	0.930 9(7)
C(8)	0.321 3(8)	0.263 4(7)	0.848 6(8)
C(9)	0.315 9(7)	0.278 9(7)	0.742 4(7)
C(10)	0.328 9(8)	0.202 8(7)	0.631 9(7)
C(11)	0.332 4(8)	0.230 1(7)	0.539 0(7)
C(12)	0.334 0(9)	0.156 8(7)	0.418 3(8)
C(13)	0.332 2(9)	0.222 8(7)	0.358 3(7)
C(14)	0.336 4(7)	0.334 1(7)	0.440 8(7)
C(15)	0.342 0(7)	0.431 7(7)	0.418 5(7)
C(16)	0.353 1(7)	0.535 1(7)	0.498 7(7)
C(17)	0.354 6(8)	0.641 5(7)	0.486 2(7)
C(18)	0.360 4(8)	0.722 7(7)	0.589 8(8)
C(19)	0.371 2(8)	0.671 7(8)	0.679 4(8)
C(20)	−0.018 3(9)	0.867 5(8)	1.056 3(10)
C(21)	0.109 5(15)	0.803 1(16)	0.846 1(26)
C(22)	0.179 1(13)	0.841 0(10)	0.926 4(10)
C(23)	−0.035 7(12)	0.570 8(12)	0.682 9(11)
C(24)	0.411 1(10)	0.630 7(10)	1.084 4(10)
C(25)	0.310 9(9)	0.385 0(8)	1.057 0(8)
C(26)	0.327 0(14)	0.141 3(17)	0.868 1(12)
C(27)	0.421 1(13)	0.124 2(9)	0.897 1(11)
C(28)	0.349 7(14)	0.010 8(15)	0.362 2(11)
C(29)	0.261 7(17)	0.012 0(10)	0.339 5(13)
C(30)	0.326 0(11)	0.188 5(8)	0.228 6(9)
C(31)	0.347 4(9)	0.652 5(8)	0.369 8(9)
C(32)	0.230 3(11)	0.679 1(11)	0.294 1(10)
C(33)	0.359 0(10)	0.846 5(9)	0.621 7(18)
C(34)	−0.017(2)	0.267(2)	0.506(2)
C(35)	0.024(2)	0.172(2)	0.579(2)
C(36)	0.022(2)	0.051(2)	0.566(2)
H(N21)	0.064(5)	0.493(5)	0.926(5)
H(N22)	0.278(5)	0.428(5)	0.716(5)
H(N24)	0.363(5)	0.505(5)	0.639(5)

No evidence of secondary extinction was found. Corrections for anomalous dispersion were applied to all non-hydrogen atoms.[13] Scattering factors were taken from ref. 14. Final positional parameters are in Table 2. Thermal parameters, final calculated positions of the hydrogen atoms, root-mean-square components of thermal displacement along the principal axes of the thermal ellipsoids, and the observed and calculated structure factors are in Supplementary Publication No. SUP 23217 (25 pp.).*

The structure was solved using programs on an Amdahl 470 V/6 computer. Some other calculations were done on this computer using programs among those previously listed,[15] but most of the calculations were performed on a PDP 11/40 using the Enraf-Nonius structure determination package.[16] Use was also made of the PDP 11/40-Vector General graphics system.[17]

RESULTS AND DISCUSSION

An ORTEP [18] drawing of the structure of dimethoxybilindione is shown in Figure 1. Also shown in Figure 1 is the numbering system used and the nomenclature for the various types of carbon atoms. A stereoview of the molecule is shown in Figure 2. The structure confirms the original formulation.[8,9] The two methoxy-groups are located on the same side of the molecule on C(4) and

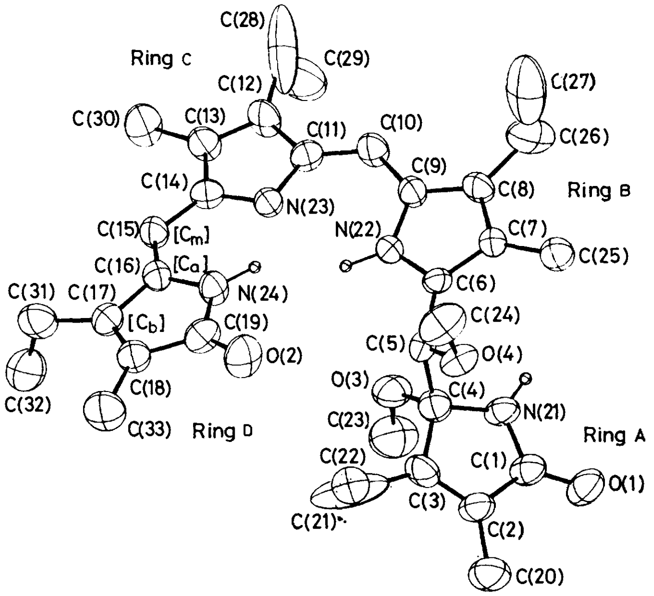


FIGURE 1 ORTEP [18] drawings of the structure of dimethoxybilindione (I). Numbering scheme is shown. Also given is the nomenclature for different types of carbon atoms. The thermal ellipsoids are drawn for 30% probability. The three hydrogen atoms attached to pyrrolic nitrogen atoms are shown. Thermal ellipsoids for hydrogen atoms are not drawn to scale

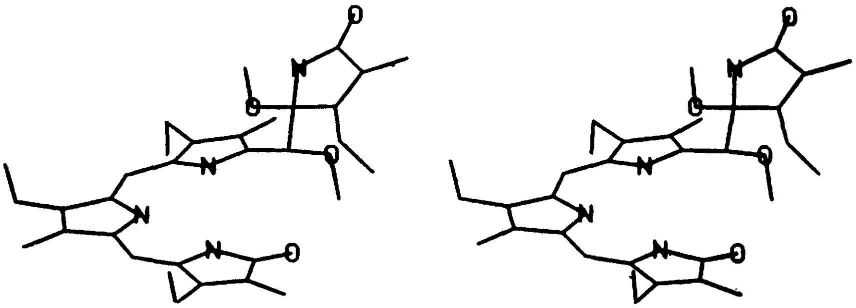


FIGURE 2 Stereoview of the structure of dimethoxybilindione (I)

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* See Notice to Authors No. 7 in *J. Chem. Soc., Perkin Trans. 2*, 1981, Index Issue.

C(5), as shown in (I), but owing to a rotation of the C(4)–C(5) bond, the methoxys are *trans* to each other. This observation allows us to deduce that in the synthesis of the dimethoxybilindione, the initial product is the monomethoxy-compound (II) which suffers bond rotation of ring A to give (III) before addition of the second molecule of methanol across the C(4)–N(21) double bond to give the *trans*-dimethoxy-product (I) (Scheme). It is of interest to note that most of the models for the $P_r \longrightarrow P_{fr}$ transformation involve changes in the vicinity of ring A.[4] Dimethoxybilindione, a model for the P_{fr} chromophore proposed by Grombein *et al.*,[3] indicates that conformational changes can easily occur in this region. As is found for most open-chain polypyrroles, adjacent rings in the conjugated part of the molecule (rings B—D) are in a synperiplanar conformation with the *Z*-configuration. Thus, that part of the molecule resembles three-quarters of a porphyrin ring.

Table 3 lists least-squares planes of interest. Individual pyrrole rings are planar. Rings B—D are relatively coplanar with interplanar angles of 5.3 and 12.7° between adjacent pyrrole rings. This gives a somewhat helical character to that part of the molecule. This is observed for several bilitrienes and bilitriene complexes. However, in these the analogous interplanar angles are generally larger. Steric effects have been postulated for the non-planarity of these tetrapyrrolic compounds. On the other hand in the etiotripyrrinone aldehyde compound which was prepared from dimethoxybilindione, the tripyrrolic skeleton is virtually flat.

Because C(4) and C(5) are sp^3 carbon atoms, ring A is considerably out of the plane of the other three rings. The nitrogen atom is roughly pointed towards the outside of the molecule. The interplanar angles between the plane of ring A and the planes of rings B—D range from 32 to 37°. This is considerably less than found in other structures with an sp^3 carbon atom joining rings. For example, in a model dipyrrole compound, which like dimethoxybilindione has saturated C(4) and C(5) atoms, the interplanar angle is 99.6°.[19]

The imino-hydrogen atoms were found, showing that they were localized as shown in (I). Apparently no tautomerism exists. It is clear from this evidence and from a consideration of the bond lengths and angles in Table 4 that there is a considerable degree of bond fix-

TABLE 3

Least-squares planes [a]

A Angles (°) between least-squares planes

	Plane 2	Plane 3	Plane 4	Plane 5
Plane 1	32.3	32.1	37.4	27.6
Plane 2		12.7	14.1	16.2
Plane 3			5.3	5.8
Plane 4				10.7

B Equation of planes [b]

Plane 1 Ring A: N(21), C(1)—C(4) $7.437x - 0.495y + 6.385z = 6.138$

Plane 2 Ring B: N(22, C(6)—C(9) $11.787x + 2.622y - 0.412z = 4.144$

Plane 3 Ring C: N(23), C(11)—C(14) $11.550x - 0.164y + 0.070z = 3.845$

Plane 4 Ring D: N(24), C(16)—C(19) $11.933x + 0.001y - 1.064z = 3.695$

Plane 5 N(22)—N(24), C(5)—C(19), O(2) $10.128x - 2.250y + 2.514z = 3.993$

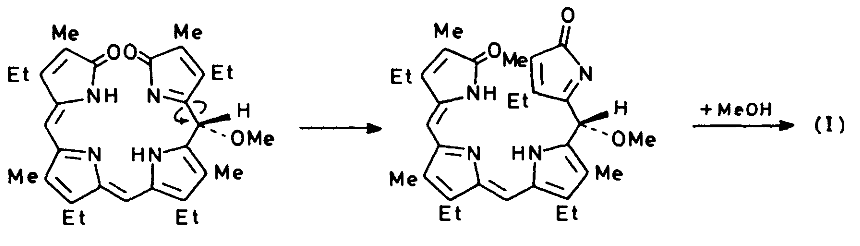
[a] Deviations from least-squares planes are given in SUP 23217. [b] x,y,z are in triclinic fractional co-ordinates.

TABLE 4

Bond lengths (Å) and angles (°) [a]

N(21)–C(1)	1.353(9)	C(1)–N(21)–C(4)	112.6(7)
N(21)–C(4)	1.422(9)	C(6)–N(22)–C(9)	110.0(6)
N(22)–C(6)	1.349(8)	C(11)–N(23)–C(14)	105.6(6)
N(22)–C(9)	1.354(8)	C(16)–N(24)–C(19)	110.5(7)
N(23)–C(11)	1.386(9)	O(1)–C(1)–N(21)	126.5(8)
N(23)–C(14)	1.331(8)	O(1)–C(1)–C(2)	125.8(8)
N(24)–C(16)	1.397(9)	N(21)–C(1)–C(2)	107.4(8)
N(24)–C(19)	1.351(9)	C(1)–C(2)–C(3)	106.8(8)
C(1)–O(1)	1.218(9)	C(1)–C(2)–C(20)	121.2(8)
C(1)–C(2)	1.466(11)	C(3)–C(2)–C(20)	131.9(9)
C(2)–C(3)	1.317(11)	C(2)–C(3)–C(4)	112.1(8)
C(2)–C(20)	1.464(11)	C(2)–C(3)–C(21)	116.8(11)
C(3)–C(4)	1.501(11)	C(4)–C(3)–C(21)	130.7(11)
C(3)–C(21)	1.668(36)	C(3)–C(4)–N(21)	100.9(7)
C(4)–O(3)	1.411(9)	C(3)–C(4)–C(5)	113.5(7)
C(4)–C(5)	1.552(10)	C(3)–C(4)–O(3)	113.2(7)
C(5)–O(4)	1.396(8)	N(21)–C(4)–C(5)	113.4(7)
C(5)–C(6)	1.499(9)	N(21)–C(4)–O(3)	114.4(7)
C(6)–C(7)	1.363(9)	C(5)–C(4)–O(3)	102.0(6)
C(7)–C(8)	1.370(9)	C(4)–C(5)–C(6)	112.4(6)
C(7)–C(25)	1.486(10)	C(4)–C(5)–O(4)	105.0(6)
C(8)–C(9)	1.402(10)	O(4)–C(5)–C(6)	114.4(6)
C(8)–C(26)	1.668(26)	C(5)–C(6)–N(22)	118.7(6)
C(9)–C(10)	1.415(10)	C(5)–C(6)–C(7)	133.0(7)
C(10)–C(11)	1.366(9)	N(22)–C(6)–C(7)	108.2(6)
C(11)–C(12)	1.462(10)	C(6)–C(7)–C(8)	106.7(7)
C(12)–C(13)	1.331(10)	C(6)–C(7)–C(25)	127.8(7)
C(12)–C(28)	1.739(29)	C(8)–C(7)–C(25)	125.5(7)
C(13)–C(14)	1.421(9)	C(7)–C(8)–C(9)	109.3(7)
C(13)–C(30)	1.498(10)	C(7)–C(8)–C(26)	123.4(9)
C(14)–C(15)	1.419(9)	C(9)–C(8)–C(26)	126.6(9)
C(15)–C(16)	1.338(9)	C(8)–C(9)–N(22)	104.6(6)
C(16)–C(17)	1.449(10)	C(8)–C(9)–C(10)	132.5(7)
C(17)–C(18)	1.319(10)	N(22)–C(9)–C(10)	122.9(7)
C(17)–C(31)	1.493(11)	C(9)–C(10)–C(11)	126.4(7)
C(18)–C(19)	1.459(10)	C(10)–C(11)–N(23)	122.6(7)
C(18)–C(33)	1.494(11)	C(10)–C(11)–C(12)	129.1(7)
C(19)–O(2)	1.228(9)	N(23)–C(11)–C(12)	108.3(7)
C(21)–C(22)	1.083(38)	C(11)–C(12)–C(13)	106.8(7)
C(23)–O(3)	1.404(12)	C(11)–C(12)–C(28)	129.1(9)
C(24)–O(4)	1.398(10)	C(13)–C(12)–C(28)	123.8(9)
C(26)–C(27)	1.094(29)	C(12)–C(13)–C(14)	106.9(7)
C(28)–C(29)	1.038(35)	C(12)–C(13)–C(30)	127.4(8)
C(31)–C(32)	1.451(13)	C(14)–C(13)–C(30)	125.8(8)
N(22)–N(23)	2.789(8)	C(13)–C(14)–N(23)	112.1(7)
N(22)–N(24)	3.778(9)	C(13)–C(14)–C(15)	126.7(8)
N(23)–N(24)	2.731(9)	N(23)–C(14)–C(15)	121.2(7)
C(34)–C(35)	1.666(25)	C(14)–C(15)–C(16)	126.5(7)
C(35)–C(36)	1.515(26)	C(15)–C(16)–N(24)	123.3(7)
C(36)–C(36) [b]	1.661(36)	C(15)–C(16)–C(17)	131.9(7)
		N(24)–C(16)–C(17)	104.8(7)
		C(16)–C(17)–C(18)	109.9(7)
		C(16)–C(17)–C(31)	130.5(8)
		C(18)–C(17)–C(31)	127.2(8)
		C(17)–C(18)–C(19)	107.8(7)
		C(17)–C(18)–C(33)	121.8(8)
		C(19)–C(18)–C(33)	130.5(8)
		O(2)–C(19)–N(24)	124.9(8)
		O(2)–C(19)–C(18)	128.2(8)
		N(24)–C(19)–C(18)	106.9(7)
		C(3)–C(21)–C(22)	93.2(29)
		C(4)–O(3)–C(23)	115.3(7)
		C(5)–O(4)–C(24)	113.5(6)
		C(8)–C(26)–C(27)	94.9(22)
		C(12)–C(28)–C(29)	89.4(24)
		C(17)–C(31)–C(32)	113.1(9)
		C(34)–C(35) C(36)	145(2)
		C(35)–C(36)–C(36) [b]	125(3)

[a] Some non-bonded distances of interest are also given. [b] This atom related to given reference atom by the symmetry operation $x' = -x, y' = -y, z' = 1 - z$.



(II) (III)

ation, particularly in rings C and D. It is quite apparent that the resonance form (I) overwhelmingly predominates in these two rings. As observed in the structure of every bile pigment and bile pigment derivative thus far reported, ring D, the pyrrolenone ring, is in the lactam form. This is apparent from the observed bond parameters. The observation that the lactam is the predominant form has also been verified by *X*-ray photoelectron spectroscopy.[20]

Other evidence besides the location of the hydrogen atoms indicates that ring C is a pyrrolenine ring, *i.e.* lacking an imino-hydrogen atom. The C_a-N-C_a angles average 111.3° for rings A, B, and D, while for ring C, this angle is 105.8°. These values are consistent with those tabulated for pyrrole rings respectively bearing and lacking an imino-hydrogen atom.[21] The C(14)-N(23) distance is much shorter than the C(11)-N(23) distance, indicating a greater degree of double bond character in the former. Other bond parameters in ring C are those expected for a pyrrolenine ring.

The bond angles involving C(10) and C(15) are large (126°) but not as large as in oxodipyrromethanes,[22-24] where there is imino-hydrogen-hydrogen steric repulsion. This also indicates N(23) does not bear a hydrogen atom.

The large degree of bond fixation in rings C and D also manifests itself in the bond lengths involving the methine carbon atom C(15). C(15)-C(16) is essentially a double bond. C(14)-C(15) is a single bond, although with some double bond character. By comparison the distances involving C(10) show that there is a good deal more delocalization involving these bonds.

In the etiotripyrrinone aldehyde,[15] which is obtained chemically from dimethoxybilindione,[8,9] the two C_a-C_b distances in the ring analogous to ring B were significantly different. The same tendency is noted in this case, although the difference is not as large. It corresponds to a difference of *ca.* 4 σ. Other than this, ring B shows bond parameters expected for pyrrole rings.

In general the bond parameters observed in rings B—D agree with those found in the etiotripyrrinone aldehyde,[15] 5-nitro-octaethylbiliverdin,[25] and biliverdin dimethyl ester.[26] The same localization of the imino-hydrogen atoms was found in these compounds. The bond parameters in ring A, which is also in the lactam form, agree with those found in the analogous ring in the model dipyrrole compound previously mentioned.[19]

While the pyrrolic skeleton shows only normal thermal motion and the atomic positions are well defined, the atoms on three of the ethyl groups show a very high degree of anisotropic thermal motion. As a result the bond lengths and angles in these groups differ markedly from expected values. A great deal of effort was expended in looking for a disorder model for these ethyl groups. However, difference syntheses showed no evidence of a second site for these atoms. No model tried was more satisfactory than the one finally used. Since both atoms in each of these groups have high thermal motion, corrections using a ' riding ' model were not possible. However, corrections for thermal shortening treating the atoms independently gave terminal C-C bond lengths of *ca.* 1.5 Å. A similar pattern as observed here, though not as extreme, was found for one ethyl group in an octaethylporphyrin monoacid cation.[27] The bond parameters in the ill defined n-hexane molecule of crystallization are also very poor. The methyl groups are well behaved.

There is quite clearly an intramolecular hydrogen bond N(24)-H(24) · · · N(23). The N(22)-N(23) and N(23)-N(24) distances are similar, but the N(23)-H distances are considerably different, 2.05 Å for H(N24) · · · N(23) and 2.30 for H(N22) · · · N(23) respectively, indicating

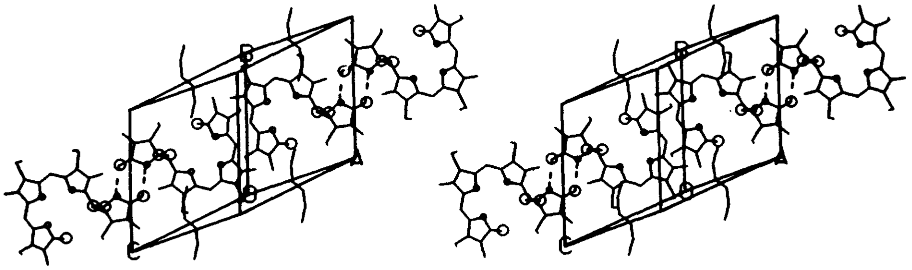


FIGURE 3 Stereoview of the packing in the unit cell. The n-hexane molecules of crystallization are shown. Large circles denote oxygen atoms, small circles denote nitrogen atoms. Hydrogen bonds are indicated by dashed lines. Hydrogen atoms have been omitted

the former hydrogen atom is involved in hydrogen bonding, whereas the latter is not.

A stereoview of the packing is shown in Figure 3. There is an intermolecular hydrogen bond, O(1) · · · H(N21)′–N(21)′. The O(1)–N(21)′ contact distance is 2.92 Å, while the O(1)–H(N21)′ distance is 2.00 Å. The prime superscripts refer to the molecule generated by the symmetry operations $x' = -x$, $y' = 1 - y$, $z' = 2 - z$. As a result of these contacts between the lactam oxygen atom and the imino-hydrogen atoms, both of which are located on ring A, the molecules form dimers joined together by two hydrogen bonds. Such dimers have been observed before in other bile pigment structures, most notably in biliverdin dimethyl ester.[26]

The molecules pack so that the relatively planar portions of the molecules form layers. The planes of ring D related by a centre of symmetry are 3.5 Å apart. Other than the hydrogen bonded contacts, there are only two intermolecular contacts $\leqslant$3.5 Å between non-hydrogen atoms. These are C(32)–O(1)″ (3.48 Å) and C(32)–O(4)″ (3.41 Å) where the double prime superscripts refer to the molecule generated by a unit translation in the negative direction along *c*. It is interesting to note that the ethyl group to which C(32) belongs is the only one which did not show a high degree of anisotropy, but other than this, these contacts probably have little effect on the structure.

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REFERENCES

[1] M. F. Hudson and K. M. Smith, *Chem. Soc. Rev.*, 1975, **4**, 363.

[2] R. Schmid, *Gastroenterology*, 1978, **74**, 1307.

[3] S. Grombein, W. Rüdiger, and H. Zimmerman, *Hoppe-Seyler's Z. Physiol. Chem.*, 1975, **346**, 1709.

[4] J. C. Lagarias and H. Rapoport, *J. Am. Chem. Soc.*, 1980, **102**, 4821, and references therein.

[5] D. A. Lightner in 'The Porphyrins,' ed. D. Dolphin, Academic Press, New York, 1979, vol. VI, pp. 521—584, and references therein.

[6] W. Siedel and W. Fröwis, *Hoppe-Seyler's Z. Physiol. Chem.*, 1941, **267**, 37; W. Siedel and E. Grams, *ibid.*, p. 49; H. Von Dobeneck, U. Sommer, E. Brunner, E. Lippacher, and F. Schnierle, *Liebig's Ann. Chem.*, 1973, 1934.

[7] K. M. Smith, *J. Chem. Soc., Perkin Trans. 1*, 1972, 1471.

[8] F. Eivazi, M. F. Hudson, and K. M. Smith, *Tetrahedron*, 1977, **33**, 2959.

[9] F. Eivazi and K. M. Smith, *J. Chem. Soc., Perkin Trans. 1*, 1979, 544.

[10] E. F. Meyer, jun., *J. Appl. Crystallogr.*, 1973, **6**, 45.

[11] E. Sletten, J. Sletten, and L. H. Jensen, *Acta Crystallogr.*, 1969, **B25**, 1330.

[12] The SHELX System for Crystallographic Calculations, G. M. Sheldrick, University of Cambridge, 1976.

[13] D. T. Cromer, 'International Tables of *X*-Ray Crystallography,' The Kynoch Press, Birmingham, 1974, vol. IV, pp. 149—150.

[14] D. T. Cromer and J. T. Waber, ref. 13, pp. 99—101.

[15] D. L. Cullen, E. F. Meyer, jun., F. Eivazi, and K. M. Smith, *J. Chem. Soc., Perkin Trans. 2*, 1978, 259.

[16] B. A. Frenz, in 'Computing in Crystallography,' Delft University Press, Delft, 1978, pp. 64—71.

[17] C. N. Morimoto and E. F. Meyer, jun., in 'Crystallographic Computing Techniques,' ed. F. R. Ahmed, Munksgaard, Copenhagen, 1976, pp. 488—496; G. M. Cole, E. F. Meyer, jun., S. M. Swanson, and W. G. White, *Am. Chem. Soc., Symposium Series 112*, 1979, pp. 189—204.

[18] C. K. Johnson, ORTEP, A Fortran Thermal Ellipsoid Plot Program for Crystal Structure Illustrations. Rep. ORNL-3794 Revised, Oak Ridge National Laboratory, Oak Ridge.

[19] W. S. Sheldrick, W. Becker, and J. Engel, *Acta Crystallogr.*, 1977, **B33**, 3669.

[20] H. Falk, S. Gergely, K. Grubmayr, and O. Hofer, *Liebig's Ann. Chem.*, 1977, 565.

[21] J. L. Hoard, in 'Porphyrins and Metalloporphyrins,' ed. K. M. Smith, Elsevier, Amsterdam, 1975, pp. 317—380, see especially p. 337.

[22] D. L. Cullen, P. S. Black, E. F. Meyer, jun., D. A. Lightner, G. B. Quistad, and C. S. Pak, *Tetrahedron*, 1977, **33**, 477.

[23] D. L. Cullen, G. Pepe, E. F. Meyer, jun., H. Falk, and K. Grubmayr, *J. Chem. Soc., Perkin Trans. 2*, 1979, 999.

[24] W. S. Sheldrick, A. Borkenstein, M. Blacha-Puller, and A. Gossauer, *Acta Crystallogr.*, 1977, **B33**, 3625.

[25] J. V. Bonfiglio, R. Bonnett, M. B. Hursthouse, K. M. Abdul Malik, and S. C. Naithani, *J. Chem. Soc., Chem. Commun.*, 1977, 829.

[26] W. S. Sheldrick, *J. Chem. Soc., Perkin Trans. 2*, 1976, 1457.

[27] C. P. Hrung, M. Tsutsui, D. L. Cullen, E. F. Meyer, jun., and C. N. Morimoto, *J. Am. Chem. Soc.*, 1978, **100**, 6068.