study seems to favor the superoxide($Fe^{III}O_2^-$)[11,12] over the neutral O_2(Fe $\rightleftarrows$ O_2) model,[13,14] since even the O_2 in "base-free" Fe(TPP)O_2 is close to O_2^-.

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Registry No. Fe(TPP)(pip)$_2$, 17845-65-7; Fe(TPP), 16591-56-3; Fe(TPP)O_2, 67887-55-2.

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Conformation of the Progesterone Side Chain: Resolution of the Apparent Conflict between X-ray Data and Force-Field Calculations Using MM2

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Recently, Duax et al.[1] analyzed crystallographic data on 85 20-oxopregnanes and found that in virtually all cases the value for τ (the C(16)–C(17)–C(20)–O(20) torsional angle) lay between 0 and −46° (see Figure 2 in ref 1). They inferred a "conflict between X-ray data and force-field calculations", since such calculations,[2] as well as earlier quantum mechanical calculations,[3] give a minimum energy value for τ of −60°. Duax et al.[1] reject the proposal of Schmit and Rousseau[2] that this deviation comes from crystal packing forces. Moreover they conclude that the 6 kcal/mol barrier to complete rotation around τ calculated by Schmit and Rousseau is unrealistically low and that this barrier "might be better represented by rigid-molecule results", suggesting values in excess of 50 kcal/mol, with the τ = 120° conformation about 25 kcal/mol higher than the τ = −30° conformation.

In this communication we show that the apparent conflict is due to the use of an inadequate force-field model and that the putative discrepancies are not observed when energy refinement using MM2CDC (hereafter referred to as MM2)[4] is employed. As part of our studies of steroid structures, we carried out force-field calculations on a number of corticosteroids, as well as on models for the steroid D ring. For compounds without a 16β substituent the minimum-energy τ values range from −4 to −20°, with generally small deviations from the crystal values (Table I).

As noted by Duax et al.[1] steroids with a 16β substitution have crystal structures with τ = −109° as well as −20°. Our calculations on 16β substituted and unsubstituted steroids (Figure 1) show a number of local minima in the torsional potential in both series, the relative energies of which are determined by steric interactions when the C=O bond is close to elipsing the adjacent C–C bond. The local minima in the potential at τ = 0 and −120° come from the tendency for the C=O to eclipse the C(16)–C(17) and C(13)–C(17) bonds, respectively.

The MM2 force field for aliphatic carbonyl compounds[4] was developed by one of us (S.P.) to model the tendency of aliphatic groups to eclipse carbonyl groups. Additionally, the torsional potentials in MM2 have been confirmed for accuracy by ab initio calculations[5,6] at several levels. These ab initio rotational potentials show a clear preference for carbonyl groups to eclipse methyl (or methylene) groups relative to eclipsing hydrogens.[7] Carbonyl

Table I. Comparison of Selected τ Values from X-ray and MM2 Calculations

molecule	X-ray	MM2
progesterone	−6.6	−4.2
16β-methylprogesterone	−108.0	−110.0
21-hydroxyprogesterone	−11.1	−4.6
cortisone	−28.9	−19.7
cortisol	−30.1	−19.4
9α-fluorocortisol	−26.8	−18.0
9α-chlorocortisol	−28.2	−18.4
9α-bromocortisol	−18.4	−17.7
9α-fluorocortisone	−(28.9)[a]	−20.4
17α-progesterone acetate	−18.9	−8.9
6α-methyl-9α-fluoroprednisolone	−16, −32	−18.0
6α-hydroxyprogesterone	−9.0	−4.0

[a] This structure generated by adding a 9α-fluorine atom to the heavy-atom coordinates of cortisone.

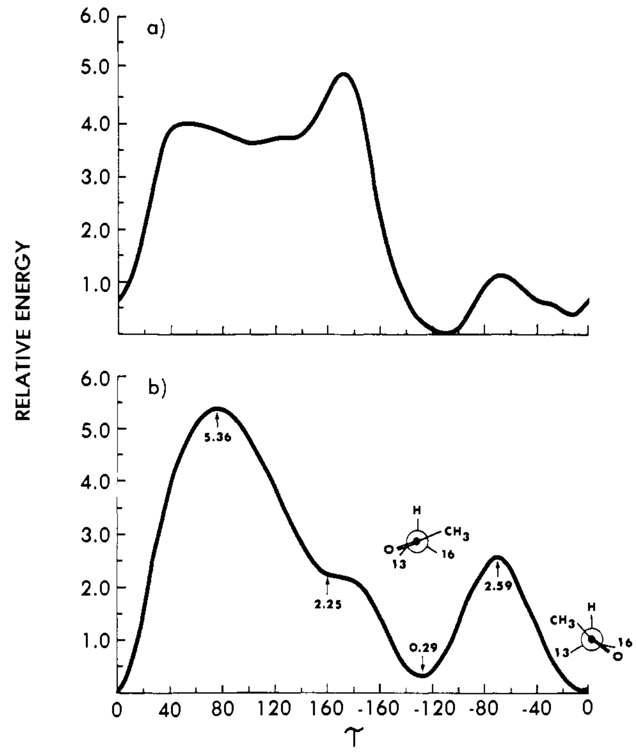

Figure 1. (a) Energy as a function of $f(\tau)$ for 16β-methylprogesterone (II); units of energy in kcal/mol. (b) Energy as a function of $f(\tau)$ for progesterone (I); units of energy in kcal/mol.

(1) Duax, W. L.; Griffin, J. F.; Rohrer, D. C. *J. Am. Chem. Soc.* **1981**, *103*, 6705.
(2) Schmit, J.-P.; Rousseau, G. G. *J. Steroid Biochem.* **1978**, *9*, 909.
(3) Kier, L. B. *J. Med. Chem.* **1968**, *11*, 915.
(4) MM2 is available from the Quantum Chemistry Program Exchange (Allinger, N. L.; Yuh, Y. H. *QCPE*, **1980**, *12*, 395. Profeta, S., Jr. *QCPE Bull.* **1981**, *1*, 57). These programs contain the carbonyl force field discussed herein. The calculated τ values reported in Table I represent those obtained by energy minimization of the crystal-structure coordinates. The functions in Figure 1 were derived by fixing τ to a given value and refining the remaining degrees of freedom.
(5) Profeta, S., Jr.; Allinger, N. L., unpublished data. This work details STO-3G and 6-31G calculations of the rotational potential functions of 2-butanone, 2-pentanone, and 3-pentanone. Similar calculations were also performed on the conformers of 2-methylcyclohexanone. In all cases, both calculation and experiment indicate at least a 1 kcal/mol preference for the C=O/C–C eclipsed conformation over the C=O/C–H.
(6) Allinger, N. L.; Profeta, S., Jr. *J. Comp. Chem.* **1980**, *1*, 181. Allinger, N. L.; Burkert, U.; Profeta, S., Jr. *J. Comp. Chem.* **1980**, *1*, 281. Allinger, N. L.; Van-Catledge, F. A., unpublished data.

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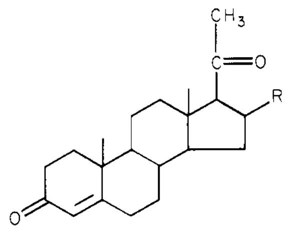
groups gauche to methyl, methylene, or hydrogen functions are calculated as relative maxima. Thus, deviations from τ = 0 and −120° must come principally from steric repulsion.

Eclipsing of the 17α H by the C═O is less favorable despite the tendency of the C═O to eclipse nearby bonds.[5,7] Such a conformation places the C(21) methyl group over the D ring and in repulsive steric contact with C(18) and C(16). In fact, this occurs in only one case analyzed by Duax et al.[1] and is due to a steric effect from a large 17α substituent.

MM2 predicts the observed minimum-energy angle in the crystal for progesterone as well as for 16β-methylprogesterone (II) (τ = −110°). Thus, *state of the art* molecular mechanics is



I, R = H
II, R = CH_3

able to reproduce the low-energy structures for these compounds. The failure of the previous force-field calculations[2] is due, in part, to lack of inclusion of appropriate torsional terms that model the observed tendency of a C═O bond to eclipse neighboring aliphatic bonds. MM2 contains such terms.[4] It is more difficult to explain why extended Hückel theory fails in this connection, but in that study[3] the energy was evaluated as a function of τ at only 60° increments, and no attempt was made to optimize the geometry as a function of τ. Thus, the energy of conformations having τ = 0 and −120° was probably overestimated by steric effects.

The average difference between the X-ray and MM2-calculated values for τ (Table I) is 7°, the same as the mean deviation of τ for the 21 unsubstituted progesterone crystal structures. The calculated values show a more eclipsed C—C—C═O orientation than the X-ray. We cannot rule out either crystal packing forces or inadequacies in the force fields as a source of this systematic discrepancy but note that such differences in dihedral angles would correspond to energy differences of 0.1–0.2 kcal/mol.

The conclusion of Duax et al.[1] that the rigid rotation surface of Schmit and Rousseau[2] is an adequate representation of the energy as a function of τ is almost certainly wrong. In the study of Schmit and Rousseau and in this study a difference in energy of 5–10 kcal between the low-energy regions (τ = 0 to −120°) and the high-energy regions (τ = 60–180°) was observed. The CD results of Wellman and Djerassi[8] suggest that the two local minima found in our study differ in energy by 1.1 kcal/mol in progesterone. Again, the dipole moments at 25° of progesterone[9] (2.7 D) and 16β-methylpregnane-3,20-dione[10] (2.66 D) argue strongly against the suggestion by Duax et al.[1] that the energy difference between the τ = −20° and −110° conformations has been "greatly underestimated". We calculate dipole moments for the minimum-energy conformations of progesterone (τ = −4° and −135°) to be 1.91 and 3.62 D. The corresponding values for τ = −13 and −110° for 16β-methylprogesterone are 1.78 and 3.12 D, respectively. Only by assuming a Boltzmann average of these two conformers can one arrive at dipole moments (2.73 D from 68% τ = −4° and 32% τ = −135° for progesterone and 2.74 D from 66% τ = −110° and 34% τ = −13° in 16β-methylprogesterone)[11] in reasonable agreement with experiment. Thus, our results indicate that in solution both conformers are significantly populated, as is also maintained by Wellman and Djerassi[8] and Allinger et al.[10] Lastly, the existence of the τ = 162° conformation in a compound with a large 17α substituent rules out the suggestion of Duax et al.[1] that its energy is ~30 kcal higher than the minimum.

Given the small 0.3 kcal/mol calculated energy difference between τ ~ 0 and τ ~ −120° conformers for progesterone, how can one explain a 76:0 ratio of τ ~ 0 to τ ~ −120° conformers for progesterone, but a 3:4 ratio of these conformers with a 16β substituent in crystal structures? A possible explanation is the fact that the τ ~ 0° conformation points the C═O away from the molecule and thus allows favorable intermolecular contacts with this group; the τ ~ −120° conformation has the C═O less accessible to intermolecular interactions. For example, the closest intermolecular contact in the crystal of progesterone (τ = 7°) is between C-21 (methyl) and O-3. In the τ ~ 0° conformation, this distance (3.5 Å) gives an attractive interaction. If τ ~ −120°, then the closest contact would be between O-20 and O-3 and could be electrostatically repulsive. Thus, the distribution of both 16-hydrogen and 16-substituted steroids may be skewed toward favoring the τ ~ 0° conformation by intermolecular packing, compared to the relative preferences calculated (Figure 2, ref 1) for the isolated molecules.

In summary, there is no conflict between X-ray structural data and state of the art force-field calculations. The preponderance of τ = −20° conformations for progesterone and the tendency for 16β substitution to stabilize the τ = −120° conformation are found in both crystal and calculated structures. Moreover, both CD and dipole moment results show that the τ = −20 and −120° conformers are sufficiently close in energy to be significantly populated in solution, and presumably of "potential" importance in drug-receptor interactions. The barrier between τ = −20 and −120° is small (2.5 kcal/mol) and that between τ = −180 and −20° is somewhat larger (5 kcal/mol); however, both barriers can be traversed rapidly.

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(7) This is consistent with the experimental and theoretical studies of the conformational preferences of propanal; see: Pickett, H. M.; Scroggin, D. C. *J. Chem. Phys.* **1974**, *61*, 3954. Allinger, N. L.; Hickey, M. J. *J. Mol. Struct.* **1973**, *17*, 233. Profeta, S., Jr.; Allinger, N. L., unpublished studies.

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(11) All calculations were done with a dielectric constant of 1.5. While the comparison of 16β-methylprogesterone to 16β-methylpregnane-3,20-dione is not "exact", we have found, in the absence of an experimental value for the moment of 16β-methylprogesterone, that the corresponding Δ^4-saturated system provides a very close reference value. It should be noted that the MM2 calculated dipole moments of cyclohexanone and cyclohexenone are very similar (2.91 vs. 2.96 D). While these values are both slightly smaller (~5%) than the corresponding experimental values, they are internally consistent and sufficiently similar to make the 16β-methyl systems comparison valid. Furthermore, if the equilibria in the progesterone systems were as one-sided as has been suggested,[1] then the observed dipole moments would be significantly smaller; e.g., for progesterone, for a 90:10 ratio of τ = −10 to τ = −135° (ΔG = 1.31 kcal/mol) the calculated moment is 2.14 D, *far* lower than that observed. Similar arguments pertain to the 16β systems. The dipole moment for progesterone is calculated to be 3.30 D at τ = −100°, 3.61 D at τ = −135°, 1.90 D at τ = −10°, and 2.02 D at τ = −30°. Thus, no single conformation is consistent with both the energy and dipole moment and a Boltzmann average of two conformations is required to explain the data. A referee has expressed skepticism about the validity of the second minimum τ values used in the dipole moment calculations. For progesterone, we see at τ = −135° the repulsive interaction between the C(18) group and O(20) appears to be greater than that between the 16β hydrogen and the C(21) group. Logically the minimum shifts toward −110° in 16β-methylprogesterone from −135° in progesterone in response to the C(21)–16β-methyl repulsion. However, the repulsive interaction between O(20) and 16β methyl amounts to 0.75 kcal, a value sufficient enough to shift the minimum τ value to −13° and absolute minimum to −110°. This fact, however, should not render the τ = −13° conformation "prohibited".

Registry No. I, 57-83-0; II, 1424-09-5; 21-hydroxyprogesterone, 64-85-7; cortisone, 53-06-5; cortisol, 50-23-7; 9α-fluorocortisol, 127-31-1; 9α-chlorocortisol, 10119-05-8; 9α-bromocortisol, 50732-00-8; 9α-fluorocortisone, 79-60-7; 17α-progesterone acetate, 302-23-8; 6α-methyl-9α-fluoroprednisolone, 382-52-5; 6α-hydroxyprogesterone, 604-20-6; 16β-methylpregnane-3,20-dione, 81737-73-7.

Chirality and Structures of Bacteriochlorophylls *d*

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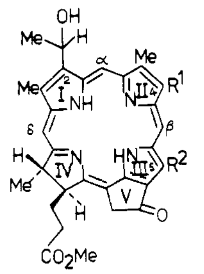
Bacteriochlorophylls (BChl) *c* and *d* comprise two homologous series of chlorophylls found in the antenna and reaction centers of green sulfur bacteria[2] (*Chlorobiacae*). Despite the continuing controversy concerning the structures of the six BChl *c* pigments,[3] assignments of the BChl *d*, which lack the δ methine methyl substituent found in BChl *c*, have been generally accepted. Smith et al.[4] recently deduced that the difference between pairs of BChl *c* lay in the absolute stereochemistry of the chiral 2-(1-hydroxyethyl) substituent; i.e., the bacteriopheophorbide bearing a 4-isobutyl and 5-ethyl was shown to have an *S* absolute stereochemistry,[4] contrary to assignments of *R* for the complete mixture of pigments.[5,6] Here, we show that the 4-isobutyl-5-ethyl- and 4-isobutyl-5-methylbacteriopheophorbides *d* also possess the 2-(*S*)-(1-hydroxyethyl) absolute configuration but that the other bacteriopheophorbides *d* (and presumably BChl *d*) exhibit the expected[5,6] *R* stereochemistry (Table I).

BChl *d* were isolated from *Chlorobium vibrioforme* forma *thiosulfatophilum* (NCIB No. 8327); treatment of the crude chlorophyll extract with methanol and sulfuric acid gave the pheophorbides **1–6** as an intimate mixture, the gross structures of which had been determined earlier by degradative[7] and synthetic work.[8] The stereochemistry of the 2-substituent had subsequently been established through Horeau analysis and degradation as *R*.[5,6] High-pressure liquid chromatography (HPLC)[9] of the intact mixture of methylbacteriopheophorbides *d* (**1–6**) gave the trace shown in Figure 1A, a separation that is superior to that in previous reports.[10,11] Prior separation of the mixture into 5-ethyl (**1**, **3**, **5**) and 5-methyl (**2**, **4**, **6**) series[10] gave the traces shown in Figure 1, parts B and C, respectively. Lengthy HPLC work resulted in accumulation of preparative quantities of all six methylbacteriopheophorbides *d*.

Table I



compd.	R^1	R^2	confign. at 2-position
1	*i*-Bu	Et	*S*
2	*i*-Bu	Me	*S*
3	*n*-Pr	Et	*R*
4	*n*-Pr	Me	*R*
5	Et	Et	*R*
6	Et	Me	*R*

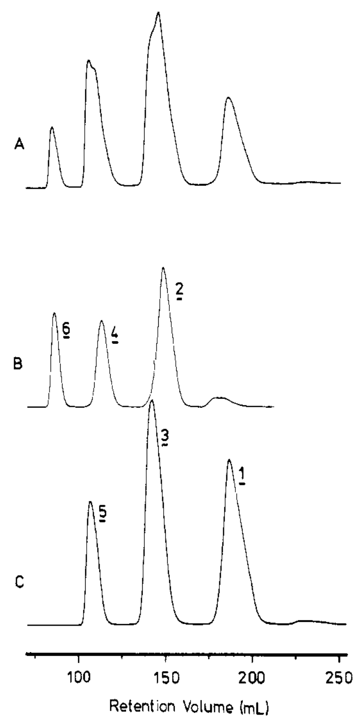


Figure 1. HPLC traces[9] of the methylbacteriopheophorbides *d*: (A) complete mixture; (B) the 5-methyl series, after preliminary separation by chromatography on silica; (C) the 5-ethyl series, after preliminary separation by chromatography on silica. The ratio of the 5-ethyl to the 5-methyl series is 3:1 in our preparation, compared with 10:1 reported by Kemmer et al.[10]

The previous observation that the naturally occurring band 1 of the methyl bacteriopheophorbides *c* (4-isobutyl-5-ethyl) had the unexpected 2*S* configuration[4] suggested that some of the BChl *d* might also have the *S* chirality.[12] The purified individual bands **1–6** were therefore treated with 80% trifluoroacetic acid in water and racemized at the 2-position to give an equal mixture of the

(1) Address inquiries regarding the chemistry to K.M.S. and the crystallography to K.M.B.

(2) Holt, A. S. In "The Chemistry and Biochemistry of Plant Pigments"; Goodwin, T. W., Ed.; Academic Press: New York, 1965; pp 3–28.

(3) For example: Kenner, G. W.; Rimmer, J.; Smith, K. M.; Unsworth, J. F. *Philos. Trans. R. Soc. London, Ser. B* **1976**, *273*, 255–276.

(4) Smith, K. M.; Kehres, L. A.; Tabba, H. D. *J. Am. Chem. Soc.* **1980**, *102*, 7149–7151.

(5) Brockmann, H., Jr.; Tacke-Karimdadian, R. *Liebigs Ann. Chem.* **1979**, 419–430. Risch, N.; Brockmann, H., Jr. *Ibid.* **1976**, 578–583. Tacke, R. Dissertation, T. U. Braunschweig, Germany, 1975.

(6) However, the Horeau method is not quantitative and would not have detected small amounts of the *S* compound. A back calculation from the slightly low rotation obtained[5] for the methyl benzoyllactate degradation product could, with hindsight, be interpreted to indicate contamination with *S* material.

(7) Purdie, J. W.; Holt, A. S. *Can. J. Chem.* **1965**, *43*, 3347–3353.

(8) Archibald, J. L.; Walker, D. M.; Shaw, K. B.; Markovac, A.; MacDonald, S. F. *Can. J. Chem.* **1966**, *44*, 345–362.

(9) A Waters Associates instrument consisting of a RCM-100 radial compression module, a Model 6000A solvent delivery system, and C-18 μBondapak reverse phase columns were used. The detector was a Perkin-Elmer LC 55B variable-wavelength detector set at 660 nm, and solvents were 10–15% water in methanol, run at 2.5 mL/min, generating a back pressure of 2000 psi.

(10) Kemmer, T.; Brockmann, H., Jr.; Risch, N. *Z. Naturforsch., B* **1979**, *34B*, 633–637.

(11) Reductive C-alkylation has shown the earlier partition separations on Celite to be inefficient: Chapman, R. A.; Roomi, M. W.; Norton, J. C.; Krajcarski, D. T.; MacDonald, S. F. *Can. J. Chem.* **1971**, *49*, 3544–3564.

(12) This suggestion resulted from the fact that the methyl benzoyllactates from degradation of both the bacteriopheophorbides *c* and *d* had the same (slightly low) rotation.[5]

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