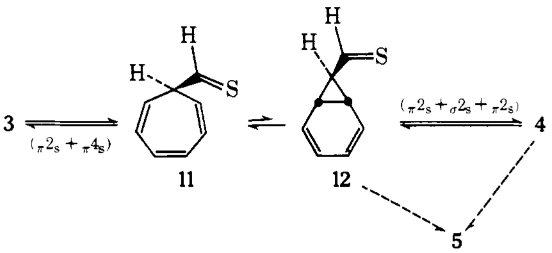
Scheme II



In an effort to gain some mechanistic insight into the thermal transformations described in Scheme I, we searched for possible fleeting intermediates with the use of dienone **6**, a powerful "Diels-Alder" trap.[8] In brief, we find that exposure of the **3**/**4** equilibrium mixture to an equivalent amount of **6** in boiling benzene leads to two 1:1 cycloadducts,[9] **7**[10] (24% yield) (white needles, mp. 233.5–234 °C; ν_{CO} (KBr) 1690 cm^{-1}, ^{1}H NMR (100 MHz; CDCl$_3$) τ 2.5–2.9 (8 H, m, phenyl), 3.4–3.6 (2 H, m, phenyl), 3.7–4.0 (2 H, m), 4.10 (1 H, d d, J = 6.5 Hz, 2.5 Hz), 4.75 (1 H, d d, J = 6.5 Hz, 3.0 Hz), 5.40 (1 H, d d, J = 10.0 Hz, 2.0 Hz), 6.25 (1 H, dq, J = 10.0 Hz, 3.0 Hz), 6.75 (1 H, m), 7.12 (1 H, m), 8.30 (3 H, s), 8.94 (3 H, s); λ_{max} (95% EtOH) 250 nm (ϵ 1070), 266 (1050); *m/e* 396 (P$^+$; 32%) 116 (100%)) and **8** (37% yield) (white crystals, mp 164–164.5 °C; ν_{CO} (KBr) 1760 cm^{-1}. ^{1}H NMR (100 MHz; CDCl$_3$) τ 2.7–3.1 (10 H, m, phenyl), 3.3–3.5 (2 H, m), 3.72 (1 H, d d, J = 9.5 Hz, 5.5 Hz), 3.98 (1 H, d d, J = 9.5 Hz, 5.5 Hz), 4.70 (1 H, d d, J = 9.5 Hz, 7.0 Hz), 4.94 (1 H, d d, J = 9.5 Hz, 7.0 Hz), 6.10 (1 H, d, H$_a$, $J_{a,b}$ = 11.0 Hz), 7.78 (1 H, d t, H$_b$, J = 11.0 Hz, 7.0 Hz), 8.40 (3 H, s), 8.55 (3H, s); λ_{max} (95% EtOH) 242 nm (ϵ 7700), 216 (14,500); *m/e* 396 (P$^+$; 3%), 91 (tropylium; 100%) as well as unreacted **5** (15% recovery). Chemically, the structure assigned to **7** was confirmed by independent preparation of an identical sample (ir, NMR, mixture melting point) upon exposure of authentic **5** to **6** in benzene at 80 °C. Structure **8**[11] derives its support chiefly from the ^{1}H NMR spectrum whose "aliphatic" region consists of two mutually coupled resonances, a doublet (H$_a$) and a doublet of triplets (H$_b$) with the latter being further coupled, equally, to two *nonadjacent*[12] "olefinic" protons. Moreover, the key presence of a tropilidine appendage in **8** was securely established by further cycloaddition. In brief, we find that **8** readily reacts with triazoline dione **9** to produce the 1:1 cycloadduct **10** (white solid, mp 196 °C dec; ν_{CO} (KBr) 1710, 1760 cm^{-1}, ^{1}H NMR (100 MHz; CDCl$_3$) τ 2.5–3.0 (15 H, m, phenyl), 3.90 (1 H, td, J = 6.0 Hz, 2.0 Hz), 4.17 (1 H, td, J = 6.0 Hz, 2.0 Hz), 4.80 (1 H, m), 5.08 (1 H, m), 6.75 (1 H, d, H$_a$, $J_{a,b}$ = 10.0 Hz), 8.2–8.4 (2 H, m), 8.40 (3 H, s), 8.50 (3 H, s), 9.22 (1 H, dt, H$_b$, J = 10.0 Hz, 4.0 Hz); *m/e* 91 (tropylium; 100%)). Significantly, one notes that while the key NMR signals due to H$_a$ and H$_b$ retain their structure on passing from **8** to **10** the H$_b$ signal moves substantially to higher field (~1.4 ppm) due to the newly developed cyclopropane environment in **10**.

The structure of cycloadduct **8** clearly implicates thioaldehyde **11** as an active intermediate in the thermal interconversion between **3** and **4**. The overall process is then best viewed to materialize by the three symmetry-allowed reversible steps depicted in Scheme II. Ultimate conversion to **5** is of course disallowed by orbital symmetry ($_{\sigma}2_s + _{\pi}2_s$ from either **4** or **12**) but the process is no doubt aided by lone pair participation and/or the weak nature of the C–S link.

While the extent of rearrangement is not as extreme as with the iso-π-electronic carbanions where the barbararyl skeleton is known[5] to *irreversibly* rearrange to the [3.2.2] frame there is little doubt that the C$_8$H$_8$S system does gain in overall stability on passing from **3** to **4**. Moreover, judging from the known skeletal stability of the various known barbaralanes[5,13] one finds it tempting to ascribe the observed population imbalance in favor of **4** chiefly to the presence of the heteroatomic lone pair.

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(12) The nonadjacent nature of these olefinic protons was established by spin decoupling.
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Optical Spectroscopic Study of (Chl a·H$_2$O)$_2$ According to the Proposed C_2 Symmetrical Molecular Structure for the P700 Photoactive Aggregate in Photosynthesis

Sir:

It is widely believed that the aggregation of Chl a and H$_2$O lies at the heart of the photosynthesis problem. As early as 1931 the possible role of Chl a–H$_2$O complexes in photosynthesis was postulated.[1] In the succeeding decades, the aggregation properties[2-5] and photoactivity[6,7] of Chl a–H$_2$O aggregates have been extensively investigated by a large number of researchers.[8,9] Latest developments indicate that Chl a exists in two states of hydration.[9-12] Under excess water conditions Chl a crystallizes to yield the dihydrate crystal.[4,9-11] The monohydrate of Chl a can be prepared[9-12] by heating under vacuum up to 80 °C for a prolonged period. At temperatures $\gtrsim$120 °C Chl a·H$_2$O becomes dehydrated leading to at least partial degradation of the chlorophyll.[12]

The balance of the available experimental observations on in vitro and in vivo systems has led to the proposal that the photoactive aggregate P700[13-15] in photosystem I is (Chl a·H$_2$O)$_2$, a C_2 symmetrical dimeric adduct of Chl a monohydrate.[9,16] At low temperatures ($\lesssim$150 K) in homogeneous hydrocarbon solutions, it is possible to prepare[10] in high yield (~85%) the 700 nm-absorbing (A700) dimer (Chl a·H$_2$O)$_2$ whose optical and redox properties resemble closely those of the in vivo P700 aggregate.[9,17] The observation of significant photocurrents in the 700-nm region of the action spectrum of the recently developed reversible Chl a photogalvanic cell suggests that (Chl a·H$_2$O)$_2$ can also be stabilized at room temperatures in a two-dimensional space where the negative configuration entropy change of the Chl a·H$_2$O dimerization process is apparently suppressed to a sufficient extent.[18]

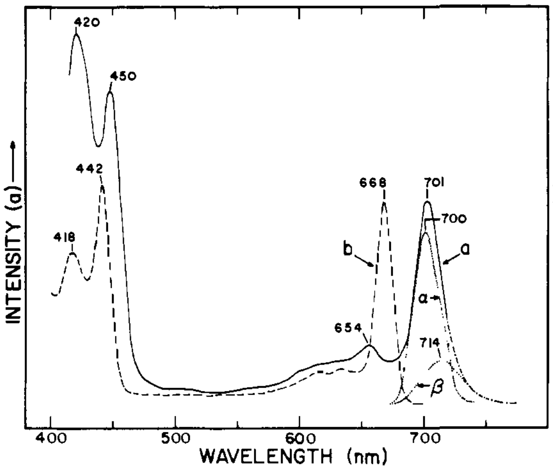

Figure 1. (a) The corrected 121 K excitation spectrum of A700 fluorescence at 720 nm in 1:1 *n*-pentane and methylcyclohexane, The dot–dash line in the long-wavelength region, which is not affected by the A663 and A680 absorption, is taken from a Cary 14 spectrophotometer absorption spectrum of the same sample. The exciton components at 700 nm (α) and 714 (β), shown as dotted lines, have been obtained using the fit parameters given in ref 10. The 700 nm/714 nm intensity peak ratio is 3. (b) The 121 K absorption spectrum of monomeric Chl a in EPA.

In spite of the longstanding interest in the primary processes in photosynthesis, it is only in recent years that detailed molecular structure of the reaction center aggregate has been questioned.[9] No absorption spectra of the "pure" P700 or A700 are available in the literature. Only difference spectra[13-15] have been observed for P700. The A700 absorption spectrum, on the other hand, is complicated by overlapping spectra of Chl $a{\cdot}H_2O$ (A663) and the as yet uncharacterized aggregate A680.[10] Moreover, the Soret region of the P700 absorption is heavily masked by carotenoid absorption,[13] while existing deconvolution procedures are rendered powerless by the spectral complexities of the multiple in vitro aggregates in the blue-band wavelength region.[10]

The present work reports the first corrected excitation spectrum of the A700 fluorescence at 720 nm and provides an analysis of this spectrum in terms of exciton theoretical expressions specialized to the molecular parameters of the proposed C_2 symmetrical structure for (Chl $a{\cdot}H_2O)_2$.

The excitation spectrum of the 121 K A700 fluorescence at 720 nm of a 2.0×10^{-5} M Chl a solution in 1:1 methylcyclohexane and *n*-pentane was measured in the manner described earlier.[10] The spectral data were digitally corrected for the spectral distribution of the excitation source in the manner given in ref 18. The resulting spectrum is shown in Figure 1. For the purpose of comparison the absorption spectrum of monomeric Chl a in EPA (5:5:2 diethyl ether:isopentane: ethanol) is also shown in Figure 1.

It can be shown from standard theory[19,20] that the dipole-dipole exciton coupling energy in (Chl $a{\cdot}H_2O)_2$ may be given by[21,22]

$$\xi = (e^2\mu^2/R_{12}^3)[1 - \sin^2(\theta/2)(3\rho^2 - 1)] \qquad (1)$$

where θ is the angle subtended by the two subunit transition moments $e\vec{\mu}$,

$$0 \leq \rho = R_\perp/R_{12} \leq 1 \qquad (2)$$

and R_{12} and $R_\perp$ are the center-center and perpendicular interplanar distances, respectively (see Figure 2). Solutions to eq 1 represent all possible exciton splittings resulting from parallel translations of the molecular planes in the C_2 dimer. The ordering of exciton components is determined by the geometric factor

$$G = [1 - \sin^2(\theta/2)(3\rho^2 - 1)] \qquad (3)$$

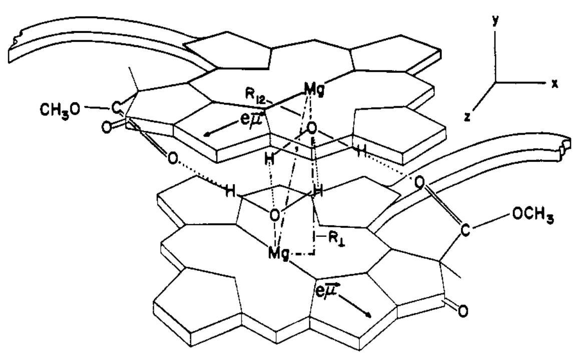

Figure 2. Scaled model representation of the C_2 symmetrical A700 dimer (Chl $a{\cdot}H_2O)_2$ displaying the reciprocal ester CO···H(H)O···Mg linkages. R_{12} and $R_\perp$ are the center–center and perpendicular distances (- · - - • -), respectively. The subunit transition moment $e\vec{\mu}$ is oriented according to the polarization of the 661 nm transition in monomeric chlorophyll a in ether.

The $S_0 \rightarrow S_1$ transition dipole strengths for the two exciton components[19] are given by the ratio

$$D_+/D_- = \cot^2\theta/2 \qquad (4)$$

The interplanar distances $R_\perp$ and R_{12} in Figure 2 may be estimated from interatomic distances determined from x-ray diffraction studies[23] of the polymeric ethyl chlorophyllide a dihydrate aggregate. The expected similarities between the Chl a-H_2O interactions in (Chl $a{\cdot}H_2O)_2$ and (Chl $a{\cdot}2H_2O)_n$ have been described earlier.[11] In Figure 2, the Mg atoms and H_2O oxygens are envisaged to lie in a plane perpendicular to the $C_2(z)$ axis with the (H)HO···Mg bonds directed perpendicular to the chlorin planes. The bonding interactions within (Chl $a{\cdot}H_2O)_2$ are assumed to be equivalent to a symmetrical addition of a Chl a molecule to a monomeric unit of Chl $a2H_2O$.[11b] These interactions are accordingly seen to be equivalent to the subunit interactions in the ethyl chlorophyllide $a{\cdot}2H_2O$ polymeric aggregate,[23] in which the two water oxygens are respectively ~2.4 and ~3.6 Å from the chlorin plane and are separated by ~2.8 Å. The 2.4 Å value corresponds to the water molecule bonded to the Mg atom. For the unique C_2 symmetrical (Chl $a{\cdot}H_2O)_2$ structure given in Figure 2, we obtain $R_\perp = 6.0$ Å, $R_{12} = 6.5$ Å, and $\rho = 0.92$. Using these molecular parameters, we arrive at $G = 0.62$ and $e^2\mu^2 = 12$ debye^2 from eq 3 and 1, respectively.[21]

The computer deconvoluted (Chl $a{\cdot}H_2O)_2$ red A700 absorption band components at 700 and 714 nm (Figure 1) are interpretable in terms of exciton interactions. Using the experimentally observed ratio 3 for the integrated 700 nm/714 nm band intensity ratio in eq 4, we obtain $\theta \simeq 60°$ in agreement with the expected[24] value for the equilibrium configuration of (Chl $a{\cdot}H_2O)_2$. The observed energy difference 280 cm^{-1} between the deconvoluted components (Figure 1) corresponds to $\xi = 140$ cm^{-1}. The sign of the exciton splitting parameter is positive in the region $\theta < 90°$. Consequently the symmetric component with the greater transition dipole intensity for $\theta < 90°$ is on the short wavelength side of the antisymmetric component in agreement with experiment and earlier prediction.[16]

The A700 red absorption in Figure 1 resembles in most details the published P700 difference spectra (for comparison, see Figure 8, ref 10) except for the absence of the variable 680-nm band observed in most P700 difference determinations. It is believed[10] that this 680-nm band is of an adventitious origin. The 654-nm band in Figure 1a is also present in all P700 difference determinations. The findings reported here appear to be relevant to the polarized optical effects of P700 absorbance changes in magnetically oriented chloroplasts observed by Breton and co-workers,[25] who have detected in the 640–660-nm region of the in vivo spectrum a broad band showing a dichroic ratio of 0.35.[26] If this in vivo observation is to be attributed to the P700, it may be interpretable in terms of exciton interations in a higher excited state than the lowest lying S_1 state in (Chl a·$H_2O)_2$. The present analysis has been specialized to the Q_y transition in Chl a.[27] For the Q_x transition the intensity ratio of the two exciton components remains 3 for the same ground-state nuclear configuration except that the long-wavelength component now becomes the dominant transition.[28]

The present work is an initial attempt at an optical characterization of (Chl a·$H_2O)_2$ in terms of a highly specific molecular model. The underlying assumptions implicit in the foregoing analysis are (A) that the A700 excitation spectrum in Figure 1 provides a faithful representation of the A700 absorption spectrum, and (B) that the A700 red band can be deconvoluted into two or less Gaussian exciton components. Assumption (A) seems to be reasonable in view of the comparison of the A700 spectrum in Figure 1 with the corresponding 121 K absorption spectrum displayed in Figure 2 of ref 10. The absence in the A700 excitation spectrum of the 672.5-nm absorption band, attributable to monomeric hydrated chlorophyll,[2,10] indicates that interaggregate excitation transfer is unimportant in the present case. Assumption B is consistent with the fact that no more than two exciton components can arise from a dimeric aggregate. In a dimeric Chl a–H_2O aggregate assuming the geometrical configuration of two neighboring units in the dihydrate polymer (Chl a·$2H_2O)_n$, only a single exciton component is expected[22a] because the transition dipole moments of the two Chl a molecules in this case are parallel to each other, i.e., $\theta = 0$.[23] The assumption of Gaussian behavior for the exciton bands is standard according to the theory of inhomogeneous line width broadening.

The two-component analysis given above has been based on the C_2-symmetrical dimer involving C-10 carbomethoxy C═O···H(H)O···Mg linkages. A somewhat analogous C_2-symmetrical model for (CHl a·$H_2O)_2$ invoking reciprocal C-9 keto C═O···H(H)O···Mg linkages[29] is expected to yield a predominantly one-component red band according to eq 4 because the transition moments of the Chl a molecules in this case are approximately antiparallel, i.e., $\theta = 180°$. We have recently prepared a 700-nm absorbing dimeric aggregate of pyroChl a in which the C-10 carbomethoxy carbonyl group is absent. The red band of this pyroChl a aggregate consists of a single Gaussian.[30]

Note Added in Proof. We note that Katz and co-workers, who, among others, have favored an unsymmetrical reaction center dimer (*Proc. Natl. Acad. Sci. U.S.A.*, **71**, 4897 (1974); *Biochem. Biophys. Comm.*, **71**, 671 (1976)) on the basis of a triplet esr signal that may be attributed to the inactive component P800 of the bacterial reaction center (see ref 9, p 283), now support a model equivalent in stoichiometry and symmetry to our model, and in bonding interactions to the Closs structure[29] (*Proc. Natl. Acad. Sci. U.S.A.*, **73**, 1791 (1976)).

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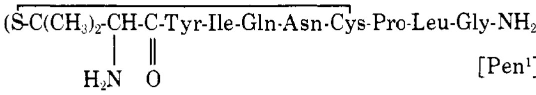
Studies on the Molecular Association of Oxytocin and Related Compounds in Dimethyl Sulfoxide

Sir:

Numerous studies have been made on structural and conformational properties of peptide hormones and related compounds using high resolution NMR spectroscopy[1] and more recently carbon-13 NMR spectroscopy.[2] Among the peptide hormones which have received considerable attention are the neurohypophyseal peptides including oxytocin.[1,3] Most of these studies have been conducted at high peptide concentrations (25–250 mg/ml) where intermolecular interactions and association may affect derived parameters. We have studied the viscosity and proton FT-NMR of oxytocin and [1-penicillamine]oxytocin,



$$(\overline{\text{S-C(CH}_3)_2\text{-}\underset{\text{H}_2\text{N}}{\underset{|}{\text{CH}}}\text{-}\underset{\text{O}}{\underset{\|}{\text{C}}}\text{-Tyr-Ile-Gln-Asn-Cy}}\text{s-Pro-Leu-Gly-NH}_2\text{-}$$

[Pen[1]]oxytocin),

[Pen[1]]oxytocin), an antagonist of oxytocin, and the carbon-13 T_1 values of specific ^{13}C-labeled atoms in oxytocin, as a function of concentration. Comparisons were made of dimethyl sulfoxide (Me_2SO) and aqueous solutions of these peptides. We present evidence which suggests that such interactions or associations may obtain in Me_2SO solutions.