

Reaction of Singlet Oxygen with Vinyl-substituted Porphyrins

A Kinetic Study by Laser Flash Photolysis

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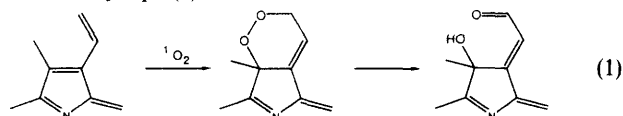
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The reactivity of several vinyl porphyrins towards singlet oxygen ($^1\text{O}_2$) in various solvents has been investigated using time-resolved detection of infrared luminescence at 1270 nm. The porphyrins studied are derived from deuterioporphyrin through 2- or 4-substitution. The singlet-oxygen quenching constant, k_p , is found to be of the order of $10^7 \text{ dm}^3 \text{ mol}^{-1} \text{ s}^{-1}$, a value much higher than that derived from steady-state photolysis measurements reported previously. The quenching rate constant decreases with increasing electron withdrawing effect of substituents on the porphyrin ring. Values of k_p determined for separated isomers and for isomeric mixtures are identical within experimental error. Deuteration of solvents does not affect k_p . Little effect of solvent electronic properties or viscosity can be discerned for aprotic solvents. On the other hand, an accelerating effect of protic solvents such as water, methanol, ethanol and *N*-methylformamide is evidenced. The k_p values for some non-vinyl porphyrins are also reported and compared to literature data. The quenching mechanism is discussed.

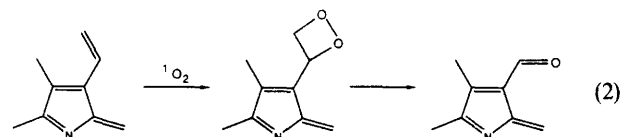
The lowest energy electronically excited state of oxygen, the singlet state $^1\Delta_g$ (hereafter denoted $^1\text{O}_2$), has been the subject of intense research.¹⁻³ This short-lived very reactive species is known to interact with molecules which possess a π -system or a particularly available pair of non-bonding electrons. The reactions of singlet oxygen with numerous substrates such as alkenes,⁴ enol ethers,⁵ phenols,⁶ amines,⁷ sulfides^{6,8} or biomolecules⁹ have been described, and reaction mechanisms are still the subject of intense discussion. Singlet oxygen is more conveniently produced for physico-chemical studies in solution by transfer of energy from a photosensitizer excited at the triplet state towards ground-state oxygen.^{3,10,11} Porphyrins, which are characterized by absorption spectra extending over the whole visible range and also produce singlet oxygen with good yields, are among the most popular photosensitizers. Little attention has been paid, however, to possible back-reaction of porphyrins with singlet oxygen. Moreover, some of the results obtained are contradictory.¹²⁻¹⁴

Damage to biomolecules by singlet oxygen is of great interest with regard to new therapeutic modalities which involve light irradiation of a photosensitizer bound to specific targets. This approach, known as photodynamic therapy (PDT), has been developed with success in the treatment of tumours which preferentially retain some porphyrins.^{15,16} The preparations in clinical use, 'Haematoporphyrin derivative' (HPD) and Photofrin, are mixtures of porphyrins¹⁷ including vinyl-substituted compounds such as protoporphyrin (PP) and hydroxyethylvinyldeuterioporphyrin (HVD) (see Fig. 1). It has been shown that these preparations undergo some photodegradation,¹⁸ but little is known about the reaction mechanism and about the role the photo-product(s) may play.

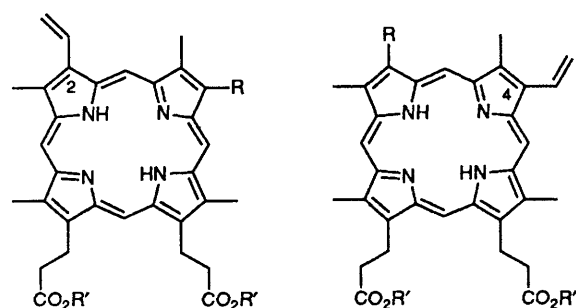
The reaction of PP in the presence of molecular oxygen and light was first reported by Fischer.¹⁹ In organic solutions the major product is a mixture of isomeric hydroxyaldehydes called 'photoprotoporphyrin'.^{12,20} The formation of these chlorin-type molecules has been suggested to arise from $[2 + 4]$ cycloaddition of singlet oxygen to a diene unit constituted by an endocyclic double bond and a vinyl group¹² as indicated by eqn. (1).



Side products such as monoformylmonovinyldeuterioporphyrins and diformyldeuterioporphyrin are also formed,¹² most likely *via* $[2 + 2]$ addition of singlet oxygen to the vinyl group, leading to a dioxetane [see eqn. (2)]. In a previous study we have shown that HVD undergoes similar reactions leading to chlorin-type molecules in organic solution.²¹



Most of the previous studies involved steady-state irradiation providing only indirect information on the reactivity of singlet oxygen with porphyrins. In the present work the reaction rate constants of singlet oxygen quenching by several vinyl and non-vinyl porphyrins have been determined directly using laser flash photolysis with detection of the $^1\text{O}_2$ luminescence in the infrared. The influence of side chains and



	R	R'
PP	CH=CH ₂	H
HVD	CH(OH)CH ₃	H
VD	H	H
EVD	CH ₂ CH ₃	H
PPDME	CH=CH ₂	CH ₃
EVDDME	CH ₂ CH ₃	CH ₃

Fig. 1 Structures of the vinyl porphyrins used in this study. (HP is a dicarboxylic porphyrin with two hydroxyethyl groups, R = CH(OH)CH₃; MPDME and DPDME have respectively: two ethyl groups, R = CH₂CH₃ and two hydrogen, R = H)

solvent on the rate constants is investigated and discussed with regard to possible reaction mechanisms.

Experimental

Materials

Protoporphyrin disodium salt was purchased from Sigma (St. Louis, MO, USA); the free-base form (PP) was obtained by solvent extraction in slightly acidic conditions. Haematoxylin (HP) purchased as the dihydrochloride form from Sigma was purified according to the method described by Vever-Bizet *et al.*²² It was submitted to a partial dehydration at the 2- or the 4-position using a modified version of the procedure of Bonnett *et al.*²³ to yield HVD (see Fig. 1). Ethylvinyldeuterioporphyrin IX (EVD) was obtained in two steps from HVD. The vinyl group of HVD was hydrogenated to an ethyl group in formic acid at room temperature using palladium as catalyst. This reduction was followed by dehydration of the hydroxyethyl group. Vinyldeuterioporphyrin IX (VD) was synthesized from deuterioporphyrin dimethyl ester according to Smith.²⁴ Protoporphyrin dimethyl ester (PPDME) and ethylvinyldeuterioporphyrin dimethyl ester (EVDDME) were prepared by treatment of PP and EVD with methanol containing 5% by volume of concentrated sulfuric acid for 24 h at room temperature. Deuterioporphyrin dimethyl ester (DPDME) and mesoporphyrin dimethyl ester (MPDME) were purchased from Aldrich (Milwaukee, WI, USA). All the porphyrins were chromatographed on silica gel²² before use. Separation of isomers of HVD and EVD was achieved by a preparative high-performance liquid chromatography method. Full details of the preparative methods as well as the chemical characterization and identification of isomers will be described elsewhere. All compounds were at least 97% pure.

Organic solvents from Merck (Darmstadt, Germany) were of analytical or spectroscopic grades except ethyl methyl ketone which was of HPLC grade. Deuteriated *N,N*-dimethylacetamide (99.7 atom%), deuteriated acetone (99.8 atom%) and deuteriated *N,N*-dimethylformamide (99.7 atom%) were purchased from CEA (Saclay, France) and deuterium oxide (99.8 atom%) from Aldrich. All solvents were used as supplied. Water was doubly distilled in quartz.

Laser Flash Photolysis

In all the experiments described the porphyrin under study serves as a photosensitizer for singlet oxygen production. The excitation light was provided by a Quantel Nd/YAG laser whose frequency was doubled to yield 6 ns pulses of 532 nm light. The laser energy was monitored after deflection of *ca.* 10% of the excitation light on to an RjP-735 pyroelectric energy meter (Laser Precision Corp.). Energies lower than 10 mJ per pulse were employed. The laser pulses were passed through BG 39 and KG 5 Schott filters to remove small amounts of the fundamental line at 1060 nm. The unfocused beam (8 mm diameter) was incident upon aerated solutions contained in a Suprasil quartz semimicro cell of 2 mm optical path (Hellma). One face of the cell parallel to the laser beam was silver-plated in order to increase collection of the singlet-oxygen luminescence. Samples were renewed after each shot.

The method for time-resolved detection of the 1270 nm luminescence of singlet oxygen is based on that reported by Rodgers and Snowden.²⁵ Infrared radiation emitted from the sample was detected by a 3 mm² Judson J16 germanium photodiode closely coupled to the cell at right angles and separated from it by an interference filter.^{9,26} The current of the diode was passed through a load resistance of between 0.1

and 2 kΩ. The resulting voltage signal was amplified (Judson amplifier) and fed to a Tektronix 7912 AD digitizer interfaced with a Hewlett-Packard 9816 computer. Signals were analysed according to standard procedure assuming the decays were mono-exponential. In all experiments single shots were used. The experimental error on the rate constants can be estimated to be *ca.* 5%.

High-performance Liquid Chromatography

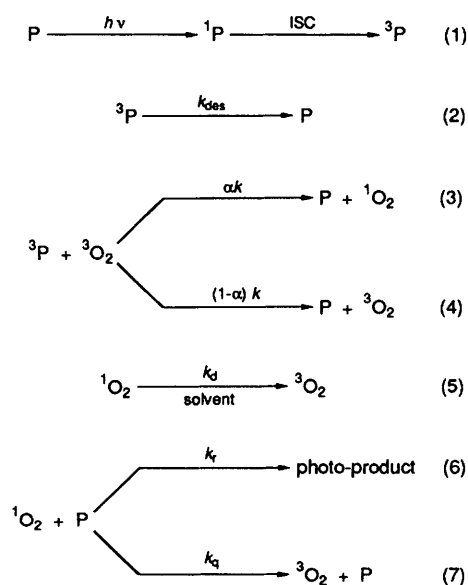
The HPLC system has already been described in detail elsewhere.¹⁷ The absorbance detector was set at 400 nm. The separations were carried out on a 5 µm silica column (25 cm × 4.6 mm i.d., Partisil 5, Whatman) protected with a 7 cm guard column packed with HC Pellosil (Whatman). The mobile phase was ethyl methyl ketone containing water (3%) and sulfuric acid (1.5×10^{-3} mol dm⁻³). The sample was eluted at 1 cm³ min⁻¹.

Results

Determination of Quenching Rate Constants

The photosensitized production of singlet oxygen through energy transfer from porphyrins excited at the triplet state to ground-state oxygen is a well documented process^{10,11,26} which can be described by steps 1–4 in Scheme 1. The singlet oxygen formed can either deactivate *via* (i) emission of a characteristic luminescence in the infrared at 1270 nm, (ii) interactions with the solvent or with the porphyrin (k_q), or (iii) chemical reaction with the porphyrin (k_r) to give photo-products (see scheme 1). The singlet oxygen luminescence has proved to be a very convenient way to monitor the concentration of this species in real time. As shown in Fig. 2, the decay of the 1270 nm luminescence was found to be mono-exponential. A first initial spike which is not taken into account, is due, in part, to the fluorescence of the solution which largely exceeds the signal from singlet oxygen. The observed rate constant was found to depend linearly on the porphyrin concentration in agreement with eqn. (3):

$$k_{\text{obsd}} = k_d + k_p[P] \quad (3)$$



Scheme 1

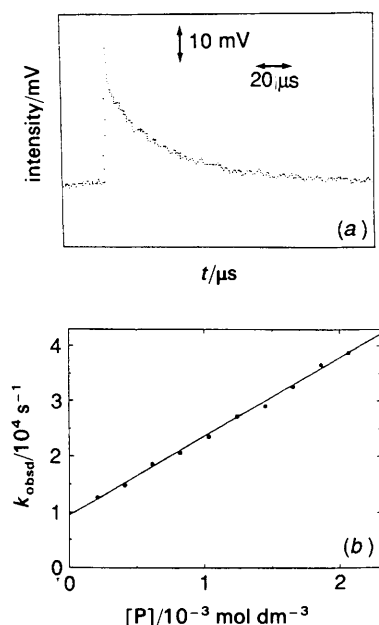


Fig. 2 (a) Example of a mono-exponential decay of the 1270 nm luminescence of singlet oxygen subsequent to a single pulse excitation of a vinyl porphyrin in solution. (b) Plot of the experimentally determined rate constant k_{obsd} (see text) as a function of porphyrin concentration. The $^1\text{O}_2$ sensitizer in this experiment was HVD in deuteriated DMA. The linear correlation shown is to equation: $k_{\text{obsd}} = 9400 + (1.43 \times 10^7) [\text{HVD}]$. The correlation coefficient (R) is 0.998

where k_d includes the radiative and non-radiative solvent-dependent deactivation rate constants²⁷ and k_p is the overall constant for interactions of singlet oxygen with porphyrin. Fig. 2 shows a typical plot from which k_d and k_p can be derived. Owing to the initial spike the lower limit of lifetime ($1/k_d$) which can be computed with reasonable accuracy is ca. 5 μs .

The values of k_p obtained in *N,N*-dimethylacetamide (DMA) for various vinyl porphyrins are reported in Table 1. This solvent possesses good solvating properties for all the compounds used making possible the investigation over a sufficiently large concentration range. In this solvent the $^1\text{O}_2$ lifetime was found to be $15 \pm 1 \mu\text{s}$. In some cases experiments were performed using deuteriated DMA where $\tau_d = 105 \pm 2 \mu\text{s}$. In this solvent k_p for HVD and EVD were found to be 1.43×10^7 and $2.19 \times 10^7 \text{ dm}^3 \text{ mol}^{-1} \text{ s}^{-1}$, respectively, in close agreement with values obtained in non-deuteriated DMA.

Table 1 Singlet oxygen quenching constants k_p for various vinyl-substituted porphyrins in DMA

porphyrin ^a	R	R'	$k_p/10^7 \text{ dm}^3 \text{ mol}^{-1} \text{ s}^{-1} \text{ }^b$
PP	$\text{CH}=\text{CH}_2$	H	1.05
HVD	$\text{CH}(\text{OH})\text{CH}_3$	H	1.36
VD	H	H	1.70
EVD	CH_2CH_3	H	2.27
PPDME	$\text{CH}=\text{CH}_2$	CH_3	1.19
EVDDME	CH_2CH_3	CH_3	2.23

^a PP = protoporphyrin; HVD = hydroxyethylvinyldeuterioporphyrin; VD = vinyldeuterioporphyrin; EVD = ethylvinyldeuterioporphyrin; EVDDME = ethylvinyldeuterioporphyrin dimethyl ester; PPDME = protoporphyrin dimethyl ester.

^b Experimental error is ca. $\pm 5\%$. Each k_p value is the average of at least five experiments.

Influence of the Isomeric Structure

Except for protoporphyrin, the vinyl porphyrins in this study exist as two isomers. The vinyl chain can be bound to the 2- or the 4-position (see Fig. 1). Since Barrett²⁸ showed that the chemical oxidation of a vinyl group is faster if this group is in the 2-position than if it is in the 4-position on the porphyrin nucleus, we also studied the reactivity of singlet oxygen towards the purified isomers. In deuteriated DMA, k_p values for HVD isomers are 1.42×10^7 and $1.41 \times 10^7 \text{ dm}^3 \text{ mol}^{-1} \text{ s}^{-1}$, those for EVD isomers are 2.12×10^7 and $2.08 \times 10^7 \text{ dm}^3 \text{ mol}^{-1} \text{ s}^{-1}$. Isomers in both cases thus appear to react at the same rate. Moreover, this rate is identical to that measured for the isomeric mixture. The same results are observed in other solvents, such as deuteriated and non-deuteriated acetone and ethyl acetate (data not shown). Since the photo-product formation rate constant k_f represents only a part of the total quenching constant k_p ^{12,14} the latter value does not necessarily reflect the chemical reactivity of the vinyl chain. An HPLC analysis of the product formed by a preparative-scale irradiation of isomeric mixtures of vinyl porphyrins indicates that the ratio of the isomeric chlorins is 1 : 1 within experimental error. Thus, in contrast to chemical oxidation, a vinyl group displays the same reactivity towards singlet oxygen regardless of its position on the macrocyclic ring. In all the following, experiments were carried out on isomeric mixtures.

Influence of Side Chains

As shown in Table 1, the rate of singlet oxygen quenching by porphyrins, k_p , increases in the order: PP, PPDME < HVD < VD < EVD, EVDDME. Although the differences between the k_p values are small (k_p is approximately doubled from PP to EVD), they largely exceed the uncertainty of 5%. However, esterification of the propionic chains does not seem to affect k_p . The substituent effect evidenced here is characterized by a decrease in the quenching rate constant with increasing electron-withdrawing ability of side chains directly attached to the conjugated macrocyclic ring.

Influence of Solvents

The k_p values for quenching of $^1\text{O}_2$ luminescence by hydroxyethylvinyldeuterioporphyrin and protoporphyrin dimethyl ester are presented in Table 2. These porphyrins were selected for their high solubility in various solvents. The media used are *N,N*-dimethylformamide (DMF), DMA, acetone, ethyl acetate, chloroform and dichloromethane. Also listed in Table 2 are k_p values for two non-vinyl porphyrins: DPDME and MPDME. Deuteriated solvents were employed in that case.

In Table 2, the $^1\text{O}_2$ lifetimes (τ_d) determined for each non-deuteriated solvent are presented together with published data ($\tau_{d,\text{lit}}$) when available. The variations of singlet-oxygen lifetimes with solvents are in good agreement with those previously reported.

From results in Table 2, note first that in any case, k_p for HVD is higher than k_p for PPDME. The substituent effects previously described seem to be qualitatively solvent independent. Secondly, k_p values vary slightly according to the nature of solvent and decrease in the order DMA > DMF > $\text{CH}_3\text{CO}_2\text{C}_2\text{H}_5$ > CH_3COCH_3 (factor of 1.5) for HVD and in the order DMF \geq DMA > $\text{CH}_3\text{CO}_2\text{C}_2\text{H}_5$ > CH_3COCH_3 > CHCl_3 > CH_2Cl_2 (factor of more than four) for PPDME.

The most obvious property of the solvent that might influence the rate would be the relative permittivity (or one of the other empirical measures of solvent polarity). That no such

Table 2 Singlet-oxygen quenching constants k_p for vinyl and non-vinyl porphyrins in several aprotic solvents

solvent	relative permittivity ^a (25 °C)	τ_{Δ} ^b /μs	$\tau_{\Delta, \text{lit}}$ /μs	$k_p/10^7 \text{ dm}^3 \text{ mol}^{-1} \text{ s}^{-1}$ ^c		$k_p/10^6 \text{ dm}^3 \text{ mol}^{-1} \text{ s}^{-1}$ ^d	
				HVD	PPDME	DPDME	MPDME
DMF	36.71	18.9	7.0 ^e	1.29	1.23	—	1.00
DMA	37.78	14.6	—	1.36	1.19	1.06	1.38
CH ₃ COCH ₃	20.70	53.9	50.5 ^f	0.92	0.63	0.69	—
CH ₃ CO ₂ C ₂ H ₅	6.02	45.0	47.6 ^e	0.97	0.67	—	—
CHCl ₃	4.81 (20 °C)	167.0	160.0 ^g	—	0.39	1.70	—
CH ₂ Cl ₂	8.93 (20 °C)	80.8	82.9 ^f	—	0.30	—	—

^a Ref. 29. ^b Each τ_{Δ} is the mean of data determined from experiments with HVD and with PPDME. ^c Each k_p is the mean of at least five experiments. ^d k_p values for DPDME and MPDME have been determined in deuteriated solvents. ^e Ref. 2. ^f Ref. 30. ^g Ref. 31.

correlation exists is immediately evident from Table 2; for instance, the values of k_p for HVD and for PPDME in acetone and in ethyl acetate where relative permittivities are 20.70 and 6.02, respectively, are only very slightly different from one another. Furthermore, singlet-oxygen quenching by vinyl porphyrins does not show any dependence on solvent viscosity. This result is not surprising since k_p is much lower than the diffusion limit.

In order to rule out the possible influence of deuteration of solvents on quenching, we measured k_p for HVD in deuteriated and non-deuteriated acetone mixtures. The rate constants for singlet-oxygen decay k_d are plotted against mixture composition in Fig. 3(a) where it is seen that excellent linear behaviour holds for that system. k_d was also shown to be a linear function of composition in binary mixtures of carbon disulfide–benzene and of H₂O–D₂O²⁵ and in binary mixtures of acetone–deuteriated acetone and of benzene–deuteriated acetone.³⁰ From Fig. 3(b) where k_p values are plotted vs. mixture composition, it can be observed that these rate constants are identical within experimental error whatever the composition mixture (error bars displayed represent $\pm 5\%$). Such a result is consistent with the absence of difference mentioned above between k_p values for HVD and for EVD determined in DMA and in deuteriated DMA. Thus, if

deuteration of solvents increases significantly the singlet oxygen lifetime (from $54 \pm 3 \mu\text{s}$ in acetone to $599 \pm 30 \mu\text{s}$ in deuteriated acetone and from $15 \pm 1 \mu\text{s}$ in DMA to $105 \pm 2 \mu\text{s}$ in deuteriated DMA), it does not affect its reactivity towards vinyl porphyrins. We also verified that the use of deuterium oxide instead of pure water did not influence k_p values.

In Fig. 4 are summarized the results obtained in mixtures containing hydroxylic solvents. In Fig. 4(a) k_p values for HVD are plotted against the percentage in volume of deuterium oxide, methanol or ethanol in acetone. Singlet-oxygen quenching by HVD appears to be faster in the presence of hydroxylic solvents. Water is found to be more efficient than methanol and much more efficient than ethanol. The same conclusions can be drawn from Fig. 4(b) where k_p values for PPDME are plotted against the percentage in volume of methanol or ethanol in DMF. Whatever the nature of the second solvent of the binary mixture, the accelerating effect of hydroxylic solvents is observed.

A similar effect appears when *N*-methylformamide (NMF), a protic solvent is used. In NMF, where $\tau_{\Delta} = 18 \pm 2 \mu\text{s}$, the quenching rate constant for HVD is found to be $6.09 \times 10^7 \text{ dm}^3 \text{ mol}^{-1} \text{ s}^{-1}$, which is *ca.* five times the value determined

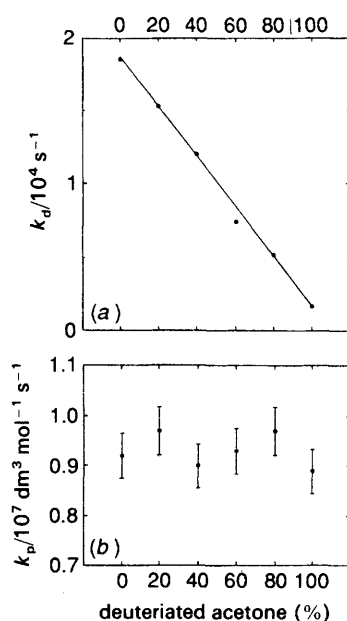


Fig. 3 Influence of solvent deuteration. Plot of (a) the first-order decay constant of singlet oxygen, k_d ; (b) the singlet oxygen quenching constant for HVD, k_p . Both values were plotted vs. proportion (v/v) of deuteriated acetone in acetone. The linear correlation shown is to equation: $k_d = (18640) - (169)(\% \text{CD}_3\text{COCD}_3)$. The correlation coefficient (R) is 0.999

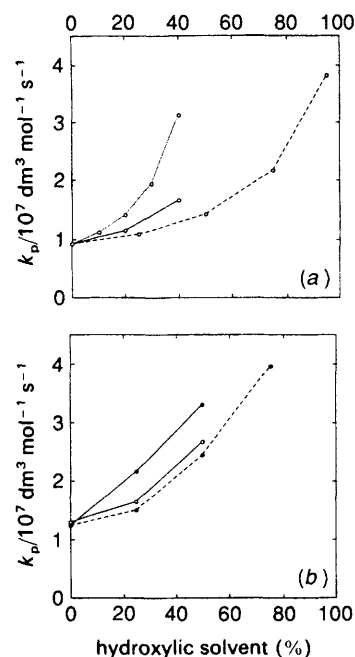


Fig. 4 Influence of the presence of hydroxylic solvents on the rate of singlet-oxygen quenching by vinyl porphyrin (a) in acetone, (b) in DMF. Plots of the rate constant, k_p , for (○) HVD and for (●) PPDME vs. the proportion (v/v) of (·····) deuterium oxide, (—) methanol or (---) ethanol

in DMA. Likewise in a mixture of DMF and NMF (50 : 50), k_p for PPDME is doubled. Even if the low solubility of vinyl porphyrins prevented us from studying their reactivity in other hydrogen-bond donor solvents, it can be concluded that the accelerating effect is specific not only to hydroxylic solvents but more generally to protic solvents.

Discussion

In all the foregoing, the term 'quenching of singlet oxygen' has been used to refer both to 'chemical' and 'physical' quenching. In the former case, singlet oxygen is removed from the system by reaction with the vinyl porphyrins leading to the formation of new products. In the latter case, interaction with the porphyrins converts singlet oxygen to its triplet ground state with neither oxygen consumption nor product formation.

A number of different mechanisms have been postulated for the quenching of singlet oxygen. Transfer of electronic energy of singlet oxygen to high vibrational energy levels (mostly C—H or O—H oscillators) of solvent molecules has been suggested to be the factor limiting the $^1\text{O}_2$ lifetime in solution. The rate constant for the decay of singlet oxygen was found to be an additive function of individual quenching properties associated with various oscillators present in the solvent. Such a physical mechanism cannot be important for porphyrins since the k_p values are two to three orders of magnitude higher than can be estimated from the contributions of oscillators present in these molecules.

Aside from this strictly physical quenching process, a mechanism that may be physical and/or chemical in character has been proposed³² to operate generally in $^1\text{O}_2$ reactions with organic substrates. This mechanism outlined in Scheme 2 proceeds *via* the intermediacy of a reversibly formed exciplex, *i.e.* a complex between the electronically excited oxygen and a substrate molecule P in its ground state. Once formed, the exciplex can revert to reactants, collapse to ground states through spin inversion (physical quenching) or yield products (chemical quenching). Owing to the electrophilic character of singlet oxygen, intermediates with partial charge-transfer can be formed on the way to the last processes. In fact, a correlation was observed between the logarithm of the rate constants of singlet-oxygen quenching by a series of amines and their ionization potentials.³³ Complete electron transfer giving rise to a substrate radical cation and superoxide ion ($\text{O}_2^{\cdot-}$) pair has been evidenced in the case of amines with oxidation potentials lower than 0.5 V *vs.* SCE.⁷ Oxidation potentials for DPDME, MPDME and PPDME are, respectively, 0.76, 0.78 and 0.83 V *vs.* SCE.³⁴ Thus, complete electron transfer is not expected to be the major process involved in the singlet-oxygen quenching by vinyl porphyrins. Indeed, steady-state irradiation experiments carried out by Cox and Whitten¹² with potential scavengers of superoxide or with electron acceptors showed that formation of superoxide was a minor process. Also, the very small effect of solvent polarity on the rate constant for the interaction of vinyl porphyrins with $^1\text{O}_2$ is not consistent with a total charge-transfer inter-

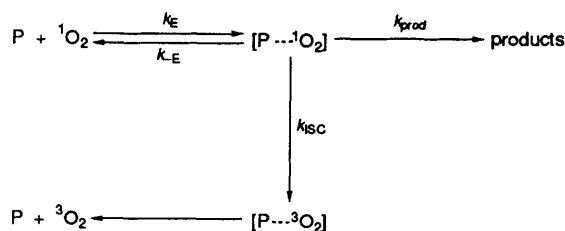
mediate. Moreover, as indicated by the above-mentioned potentials, vinyl chains would be expected to slightly reduce singlet-oxygen quenching. The reverse is actually observed. The k_p values for vinyl porphyrins are 10 to 15 times greater than k_p values for non-vinyl porphyrins, thus indicating the particular role of these unsaturated chains (see Table 2).

As shown in eqn. (1) and (2), interaction of singlet oxygen with vinyl porphyrins may involve [2 + 2] addition giving rise to dioxetane or [2 + 4] cycloaddition on the diene formed by the vinyl chain and the pyrrolic endocyclic double bond, leading to an endoperoxide intermediate. Such reactions have been well documented with various substrates. Reactions of singlet oxygen with enol ethers leading to dioxetanes⁵ and with furans giving rise to endoperoxides^{4,35} have been suggested to proceed through the rapid reversible formation of an exciplex. Rate constants well below that for diffusion, activation enthalpies close to zero and highly negative activation entropy values were indicative of such a mechanism. [2 + 2] additions were found to be sensitive to solvent polarity, suggesting partial charge-transfer in the reaction intermediates. The absence of solvent polarity effects on the overall reaction rate of vinyl porphyrins with singlet oxygen indicates that the preferred path involves the initial formation of an exciplex of very low polarity which should have the form of an endoperoxide. Accordingly, the main products are chlorin-type molecules rather than formyl products which should derive from [2 + 2] addition. This might also suggest that the vinyl and endocyclic double bonds adopt preferentially the *s-cis*- rather than the *s-trans*-conformation. It is known, for example,³⁶ that cyclohepta-1,3-diene and cycloocta-1,3-diene, which have their double bonds twisted from the *cis*-coplanar conformation behave like mono-alkenes and are less reactive towards singlet oxygen than cyclopentadiene and cyclohexadienes.

Note that deuteration of solvents does not affect k_p values. This means that the energy levels of singlet oxygen in the initially formed exciplex are perturbed enough to make unlikely the transfer of electronic energy to C—H or O—H oscillators of the solvent.

The accelerating effects of protic solvents on singlet-oxygen quenching also provide interesting information concerning the mechanism. [2 + 4] cycloaddition of singlet oxygen with naphthalene compounds was found to be similarly sensitive to the presence of protic solvents.³⁷ On the contrary, the quenching ability of amines decreases with increasing hydrogen bond donating ability of solvents.³⁸ A similar but smaller effect was evidenced for the ene reaction of tetramethylethylene.³⁸ In the latter cases the decreasing effect is explained by the formation of a hydrogen bond between the substrates and the solvent which lowers the effective concentration of free quencher. In the case under study, protic solvents may act by increasing the electrophilicity of oxygen by hydrogen bonding, thus enhancing the reactivity of $^1\text{O}_2$ towards vinyl porphyrins. Another possible explanation could be that interaction of the exciplex or subsequent intermediates with protic solvent molecules would increase the probability of yielding products or ground states.

The substituent effect observed and characterized by a decrease in the quenching rate constant with increasing electron-withdrawing ability of ring substituents is not surprising. In fact, since the early data relating to addition of singlet oxygen to conjugated systems *via* endoperoxide, the introduction of electron-donating groups into the diene system is known to lead to increased reactivity.² These substituent effects that seemed to stress the parallel between singlet oxygenation and the normal Diels–Alder reactions were first attributed to the resulting increase in the π -donating ability of the substrate. Such an explanation



Scheme 2

appeared later to be, at best, a gross oversimplification⁵ since activation enthalpies for several reactions of singlet oxygen with molecules which possess a π -system were reported to be near zero.^{3,5,36} The relative electron-donating abilities of the various substrates are now considered to affect activation entropy values by influencing for example, solvation characteristics.^{5,36}

From the kinetic results presented here, it should be noted that in all cases, quenching of singlet oxygen by vinyl porphyrins occurs with a rate constant in the order of $10^7 \text{ dm}^3 \text{ mol}^{-1} \text{ s}^{-1}$. These values are higher than that reported by Cox and Whitten¹² for the photooxidation of PPDME in dichloromethane ($k_p = 8.5 \times 10^5 \text{ dm}^3 \text{ mol}^{-1} \text{ s}^{-1}$, for comparison see in Table 2, our k_p value determined in dichloromethane). It must be emphasized that Cox and Whitten obtained their rate constant from experiments using 1,4-diazabicyclo[2.2.2]octane (DABCO) or cholesterol as quenchers of the formation of photo-product. In such systems, the determination of k_p relies on the value of the rate constant of singlet oxygen reaction with the quencher. Since these constants were also measured by indirect methods and, in addition, in solvents different from the one used for porphyrin (dichloromethane-methanol-pyridine (94 : 3 : 3/v : v : v) for DABCO³⁹ and pyridine for cholesterol⁴⁰), uncertainties accumulate. The k_p values obtained in the present study from $^1\text{O}_2$ luminescence decay measurements thus appear to be much more reliable.

According to Scheme 2, depending on the relative values of k_{-E} and $k_{\text{prod}} + k_{\text{ISC}}$, equilibrium conditions are attained ($k_{-E} \gg k_{\text{prod}} + k_{\text{ISC}}$) or the exciplex is formed essentially irreversibly ($k_{-E} \ll k_{\text{prod}} + k_{\text{ISC}}$).³² In the former hypothesis, the observed rate constant k_p would be equal to $K_e(k_{\text{prod}} + k_{\text{ISC}})$, where $K_e = k_E/k_{-E}$. In the latter hypothesis, for which a mono-exponential decay can be predicted by theoretical analysis, $k_p = k_E$, and a diffusion-controlled rate would be expected. In fact, we found a rate constant several orders of magnitude less than diffusion controlled. As the exciplex is likely to be formed from the porphyrin with the vinyl chain in the *cis*-conformation (see above) such a low value might result from orientation factors.

With k_p values of the order of $10^7 \text{ dm}^3 \text{ mol}^{-1} \text{ s}^{-1}$, vinyl porphyrins are quite effective quenchers of singlet oxygen. Since such porphyrins, particularly PP and HVD, are components of Photofrin, the preparation used clinically in tumour photochemotherapy, it would be worth examining carefully the photoreaction of this drug.

Non-vinyl porphyrins appear to be less efficient singlet-oxygen quenchers by at least one order of magnitude. This finding is in agreement with previous data of Krasnovsky¹⁴ who reported a k_p value of $2.5 \times 10^6 \text{ dm}^3 \text{ mol}^{-1} \text{ s}^{-1}$ for MPDME in carbon tetrachloride. A value higher than $10^7 \text{ dm}^3 \text{ mol}^{-1} \text{ s}^{-1}$ was reported previously for the same porphyrin.¹³ But the latter experiments involved higher laser energies and the rate constant values were extrapolated using only a few data points leading to considerable uncertainties, as quoted by the authors. In our experiments great care was taken to check the linearity of the observed phenomenon *vs.* laser energy. Thus, considering our results and Krasnovsky's data, depending on the solvent, a reaction rate value of $(1-3) \times 10^6 \text{ dm}^3 \text{ mol}^{-1} \text{ s}^{-1}$ should be observed for the quenching of singlet oxygen by mesoporphyrin, a common photosensitizer used for physico-chemical studies in solution.

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