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Study on phosphorus(III) complex of tetrabenzotriazacorrole: A novel phthalocyanine-like photosensitizer

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Abstract

The photosensitizing properties of a novel phthalocyanine analogue—hydrogen hydroxy-phosphorus(III)-2,9,16,23-tetraisopropoxy tetrabenzotriazacorrole ($H[TBC(O^iPr)_4P(OH)]$) has been studied. Using diphenyl-isobenzofuran as chemical quencher, the photogeneration of singlet oxygen by $H[TBC(O^iPr)_4P(OH)]$ was identified together with its quantitative analysis. It is found that $H[TBC(O^iPr)_4P(OH)]$ is a more efficient singlet oxygen photosensitizer ($\Phi_\Delta = 0.44$, in *N,N*-dimethylformamide) than empty phthalocyanine $H_2Pc(O^iPr)_4$, and can be comparable to zinc phthalocyanine $ZnPc(O^iPr)_4$. $H[TBC(O^iPr)_4P(OH)]$ can also effectively photo-generate the superoxide anion as shown by spin-trapping measurements. From the interpretation of the free energy changes ΔG^* in excited state electron transfer reactions, it is concluded that the superoxide anion is formed by electron transfer from the lowest excited singlet state (not the lowest excited triplet state) of $H[TBC(O^iPr)_4P(OH)]$ to oxygen. The results confirm the presence of both Type I (radical) mechanism and Type II (singlet oxygen) mechanism in the $H[TBC(O^iPr)_4P(OH)]$ photosensitized system.

Keywords: Tetrabenzotriazacorrole; Phthalocyanine; Phosphorus(III) complex; Photosensitization; Singlet oxygen; Superoxide anion

1. Introduction

With few exceptions, phthalocyanines can form stable complexes to many cations by chelating with the four central pyrrole nitrogen atoms of the macrocycle, while the conjugated macrocycle remains unchanged throughout [1]. However, it does not meet with the phosphorus(III). In the recent study [2], we have shown that the phthalocyanine may lose a bridging nitrogen atom when complexated with the trivalent phosphorus, the species formed should be a phthalocyanine analogue—hydrogen hydroxyphosphorus(III)-tetrabenzotriazacorrole ($H[TBCP^{III}(OH)]$). This is another example of the ring contractive reaction of metalloid phthalocyanine besides the previously reported $TBCGe^{IV}(OH)$ [3]. Although the exact mechanism of such marked distortion of the extremely stable phthalocyanine ring is still unclear, the chemistry of TBC complex itself has stimulated considerable interest. These include the facts that the TBC macrocycle will be useful in biological modelling [4], the central phosphorus is the smallest ion found to undergo insertion into a tetrapyrrolic ring, the new complex has lower molecular symmetry and shows unusual electronic-transition spectrum, strong fluorescence, improved solubility and strong resistance to acids [2]. Thus, the TBC complex provides a novel variant to the known chemical and spectroscopic properties of these macrocyclic compounds.

Although the phthalocyanines are being extensively studied as photosensitizers for photodynamic therapy of cancer [5], in view of this recent history for use of phthalocyanines, it is still premature to define the structural requirements for the ideal photochemotherapeutical agent of this class. As a matter of fact, the photosensitizing efficiency of phthalocyanines is mostly affected by the ring structure and the central metal [6]. Against this background, the study of the phthalocyanine analogue–TBC complex is undoubtedly necessary as well as instructive. In this paper, we will deal with the photosensitizing properties of this species. The compound chosen is still hydrogen hydroxy-phosphorus(III)-2,9,16,23-tetraisopropoxytetrabenzotriazacorrole ($H[TBC(O^iPr)_4P(OH)]$) as before [2], the substitution of the iso-propoxy group on the ring can enhance its solubility. During the present investigation, the photosensitizing properties of $ZnPc(O^iPr)_4$ and $H_2Pc(O^iPr)_4$ are studied additionally for comparison.

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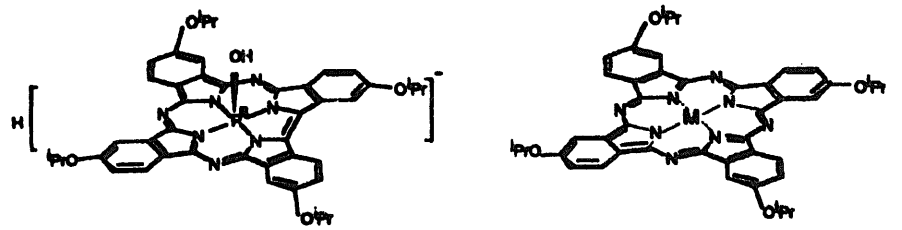

Fig. 1. Chemical structures of (left) H[TBC(O^{i}Pr)$_4$P(OH)] and (right) H$_2$Pc(O^{i}Pr)$_4$ (M = 2H) and ZnPc(O^{i}Pr)$_4$ (M = Zn).

2. Experimental details

2.1. Chemicals

Hydrogen hydroxy-phosphorus(III)-2,9,16,23-tetraisopropoxy tetrabenzotriazacorrole (H[TBC(O^{i}Pr)$_4$P(OH)]) and 2,9,16,23-tetraisopropoxy phthalocyanine (H$_2$Pc(O^{i}Pr)$_4$) were prepared as described before [2], structures shown in Fig. 1.

The synthesis method of 2,9,16,23-tetraisopropoxy zinc phthalocyanine (ZnPc(O^{i}Pr)$_4$) was adopted from that used for H$_2$Pc(O^{i}Pr)$_4$. The mixture of 5 g 4-isopropoxy phthalonitrile and 3.4 g anhydrous zinc acetate was refluxed in 50 ml dimethylaminoethanol for 12 h under an ammonia atmosphere. After cooling, the mixture was diluted with water. The blue residue was collected by suction filtration, washed with 1.0 M hydrochloric acid and then with water thoroughly. The dried product was further purified by silica gel column chromatography with ether/pet. ether (7:3) as eluant, yielding 1.2 g of a purple shining solid, ZnPc(O^{i}Pr)$_4$. Elemental analysis for $C_{44}H_{40}N_8O_4Zn$: found (calcd.) C 65.24 (65.27), H 4.90 (4.94), N 13.76 (13.84). UV-Vis (DMF, nm) 354 (logϵ,4.92), 612 (4.57), 680 (5.27).

Diphenylisobenzofuran (DPBF) and 5,5-dimethyl-1-pyrroline-*N*-oxide (DMPO) were purchased from Sigma Chemical Co.. Actinometer Cr(urea)$_6$Cl$_3$ was prepared according to the literature [7]. 1-iodo-propane was analytically pure and redistilled before use. *N,N*-dimethyl formamide (DMF) was of analytical grade, it was dried over anhydrous MgSO$_4$ and redistilled under vacuum just before use.

2.2. Photolysis procedures

2.2.1. The measurement of photogeneration of singlet oxygen $O_2(^1\Delta_g)$

The photolysis and actinometry were carried out at room temperature in a 1 cm × 1 cm quartz photolysis cell. The cell was modified [8] to permit the irradiation light perpendicular to the optical pathway of measurement, and fitted in a HP8452A diode array spectrophotometer so that the absorption of DPBF could be monitored as a function of irradiation time directly. 414 nm was selected as the analysis wavelength to avoid the interference from photosensitizers and the oxidation products of DPBF. A solution of photosensitizer in DMF, with an absorbance matching that of the probe, was used as reference. Tests were carried out to establish that no photosensitizer was consumed during the irradiation, and that the photobleaching of DPBF can be inhibited by a specific singlet oxygen quencher, sodium azide. Illumination was provided by a 150 W iodine–tungsten lamp with a filter cutting out wavelength shorter than 600 nm ($600<\lambda<750$ nm). The light intensity incident upon the photolysis cell was 1.98×10^{-6} einstein·L^{-1}·s^{-1} measured using Cr(urea)$_6$Cl$_3$ as the actinometer.

The solutions of photosensitizer (6 μM) and DPBF (50 μM) were prepared from the stock photosensitizer DMF solutions and the fresh DPBF–DMF solution by dilution with DMF to the desired concentrations. The preparations were carried out in the dark. Prior to irradiation, oxygen was bubbled through the solution for 10 min, the solution was sealed up and stirred during photolysis to eliminate secondary photolysis and inner filter effects.

In order to determine the light absorbed by the photosensitizer solutions exactly, the emission spectrum of the iodine–tungsten lamp with the filter (i.e. the relative light intensity $i(\lambda)$ vs. the corresponding light wavelength λ) was measured by a grating monochromator (Beijing No.2 Optical Instrument Factory) equipped with a photoelectric multiplier.

2.2.2. The measurement of photogeneration of superoxide anion radical $O_2^{\cdot-}$

Photo-ESR measurements were performed on a Bruker ER200D-SRC spectrometer operating at 9.66 GHz with 1.2 G field modulation. Pure DMPO was dissolved (10 mM) directly in DMF solution of photosensitizer (0.5 mM). The photolysis of oxygenated solution was carried out in a capillary situated in the ESR photolysis cavity, a 150 W iodine–tungsten lamp with a filter cutting off wavelength below 600 nm was used for irradiation. The spectra were recorded after 10 min irradiation.

2.3. Other instruments and methods

Fluorescence measurements were carried out on a PE LS-50B luminescence spectrometer with excitation and emission slit widths of 7.5 nm. Fluorescence lifetimes were measured by a Horiba NAES-1100 time-correlated single-photon-counting spectrofluorometer. Elemental analysis for C, H and N was performed at Beijing Institute of Chemistry, Academia Sinica.

The redox potentials of the photosensitizers in nitrogen saturated DMF solutions were determined by a 79-1 cyclic voltammeter (Jinan No.4 Radio Factory). Saturated calomel electrode (SCE), Pt sheet and Pt wire were used as reference,

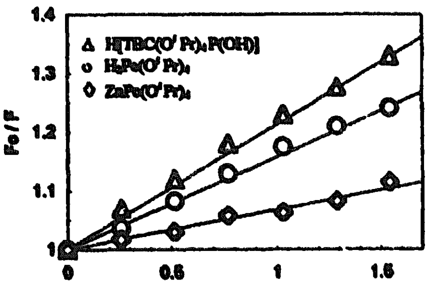

Fig. 2. Fluorescence quenching of photosensitizers (6 μM) by 1-iodopropane in benzene.

auxiliary and working electrode, respectively. Tetra-*n*-butylammonium hexafluorophosphate [TBA]PF_6 was selected as supporting electrolyte.

3. Results and discussion

3.1. Estimation of triplet energy of $H[TBC(O^iPr)_4POH)]$

The energy level of the lowest excited triplet state of photosensitizer is important in mechanistic interpretations of photophysical process and photochemical reaction, however we failed to determine the triplet energies of the phthalocyanine compounds directly from the phosphorescence, because their phosphorescence emission wavelengths are almost beyond 1000 nm and can not be detected on a common luminescence spectrometer.

Alternatively, we have to estimate the triplet energy indirectly from Stern-Volmer quenching constants of fluorescence. Dreeskamp et al. [9] have reported that the fluorescence quenching rate constants k_q ($mol^{-1} \cdot L \cdot s^{-1}$) of polynuclear aromatic hydrocarbons and heterocycles by 1-iodo-propane in benzene solutions decrease exponentially with the energy difference ΔE^{ST} ($kJ \cdot mol^{-1}$), where ΔE^{ST} is given by the difference between the lowest excited singlet state energy E^S and lowest triplet state energy E^T, the correlation can be expressed as

$$\frac{F_0}{F} = 1 + k_q \cdot \tau_f \cdot [Q] \tag{1}$$

$$logk_q = 9.8 - 5.266 \cdot \Delta E^{ST} \tag{2}$$

$$\Delta E^{ST} = E^S - E^T \tag{3}$$

where F_0 and F are the intensities of photosensitizer fluorescence in benzene without and with quencher, respectively, [Q] ($mol \cdot L^{-1}$) is the quencher concentration and τ_f (ns) is the fluorescence lifetime of photosensitizer in benzene without quencher.

A plot of F_0/F vs. [Q] yields $k_q \cdot \tau_f$ as the slope. Using Eq. (2), the ΔE^{ST} value can be determined. With the knowledge of the excited singlet state energy E^S, the triplet energy E^T of the photosensitizers can be estimated by Eq. (3).

Fig. 2 shows the Stern-Volmer quenching curves for $H[TBC(O^iPr)_4P(OH)]$, $ZnPc(O^iPr)_4$ and $H_2Pc(O^iPr)_4$. The results of ΔE^{ST}, E^S and E^T are listed in Table 1 (E^S values have been attained from absorption and fluorescence spectra before [2]), and the triplet energy of $H[TBC(O^iPr)_4P(OH)]$ is around 130 $kJ \cdot mol^{-1}$.

3.2. Photo generation of singlet oxygen $O_2(^1\Delta_g)$ by $H[TBC(O^iPR)_4P(OH)]$ (Type II photosensitization)

Since the triplet energy of $H[TBC(O^iPr)_4P(OH)]$ is higher than that of the singlet oxygen $O_2(^1\Delta_g)$ (94 $kJ \cdot mol^{-1}$) [10], the triplet photosensitized production of $^1O_2$ by $H[TBC(O^iPr)_4P(OH)]$ is possible provided its triplet lifetime is not extremely short. Such possibility has been verified in the present work. The quantum yield (Φ_Δ) for $^1O_2$ photoproduction was measured using DPBF as a chemical trapper for $^1O_2$, and the following equation is obeyed for the photobleaching of DPBF in the photosensitized system [11]

Table 1
Photophysical and thermodynamic properties of photosensitizers

Parameter	$H[TBC(O^iPr)_4P(OH)]$	$ZnPc(O^iPr)_4$	$H_2Pc(O^iPr)_4$
τ_f^a, τ_f^b (ns)	3.1^a, 2.6^b	3.7^a, 2.3^b	4.8^a, 6.7^b
k_q ($\times 10^7 mol^{-1} \cdot L \cdot s^{-1}$)	8.09	2.97	2.35
ΔE^{ST} ($kJ \cdot mol^{-1}$)	52	63	66
E^S ($kJ \cdot mol^{-1}$)	181	173	168
E^T ($kJ \cdot mol^{-1}$)	129	110	102
Φ_Δ	0.44	0.52	0.21
$e_{(D^+/D)}{}^g$ (V)	0.55	0.50	0.71
$e_{(D^+/D)}{}^S$ (V)	−1.33	−1.29	−1.03
$e_{(D^+/D)}{}^T$ (V)	−0.79	−0.64	−0.35
ΔG^g ($kJ \cdot mol^{-1}$)	136	131	152
ΔG^S ($kJ \cdot mol^{-1}$)	−45.4	−41.5	−16.4
ΔG^T ($kJ \cdot mol^{-1}$)	6.76	21.2	49.2

Note: τ_f^a, τ_f^b, fluorescencce lifetimes measured in nitrogen-saturated DMF and benzene solution, respectively; $e_{(D^+/D)}{}^g$, $e_{(D^+/D)}{}^S$, $e_{(D^+/D)}{}^T$ the oxidation potentials (vs. SCE) of photosensitizers in ground state, S_1 state and T_1 state, respectively; ΔG^g, ΔG^S, ΔG^T, the free energy changes of electron transfer from ground state, S_1 state and T_1 state of photosensitizer to oxygen, respectively.

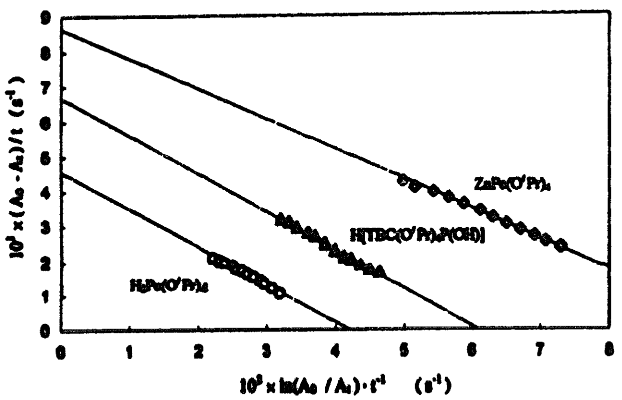

Fig. 3. DPBF consumption as a function of irradiation time in DMF solutions of H[TBC(O^{i}Pr)$_4$P(OH)], ZnPc(O^{i}Pr)$_4$ and H$_2$Pc(O^{i}Pr)$_4$.

$$\frac{[\mathrm{DPBF}]_0 - [\mathrm{DPBF}]_t}{t} = I_a \cdot \Phi_\Delta - \frac{k}{t} \cdot \ln\left(\frac{[\mathrm{DPBF}]_0}{[\mathrm{DPBF}]_t}\right) \qquad (4)$$

where [DPBF]$_0$ and [DPBF]$_t$ are the initial concentration and concentration after irradiation time t(s), k is a constant, I_a is the intensity of light absorbed by the photosensitizer and should be calculated by

$$I_a = I_0 \cdot \frac{\int_{600}^{750} i(\lambda) \cdot (1 - 10^{-A_p}) \mathrm{d}\lambda}{\int_{600}^{750} i(\lambda) \mathrm{d}\lambda} \qquad (5)$$

where I_0 is the intensity of light incident upon the cell, $i(\lambda)$ is the relative intensity of incident light at wavelength λ(nm), and A_p is the absorbance of photosensitizer at the corresponding wavelength λ.

$$\text{Since } [\mathrm{DPBF}] = \frac{A}{\epsilon} \qquad (6)$$

where ϵ is the molar absorptivity of DPBF at 414 nm (19,000 mol^{-1}·L·cm^{-1}), A is the absorbance of DPBF at 414 nm in 1 cm pathlength cell. From Eqs. (4) and (6), we obtain

$$\frac{A_0 - A_t}{t} = \epsilon \cdot I_a \cdot \Phi_\Delta - \frac{k \cdot \epsilon}{t} \cdot \ln\left(\frac{A_0}{A_t}\right) \qquad (7)$$

where A_0 and A_t correspond to [DPBF]$_0$ and [DPBF]$_t$, respectively.

According to Eq. (7), the plot of $A_0 - A_t/t$ vs. $\ln(A_0/A_t)/t$ yields $\epsilon \cdot I_a \cdot \Phi_\Delta$ as the intercept and $k \cdot \epsilon$ as the slope. Fig. 3 presents the plots in three photosensitizer solutions. Since the values of I_a for H[TBC(O^{i}Pr)$_4$P(OH)], ZnPc(O^{i}Pr)$_4$ and H$_2$Pc(O^{i}Pr)$_4$ are attained as 8.02×10^{-7}, 8.76×10^{-7} and 1.14×10^{-6} einstein·L^{-1}·s^{-1}, respectively, and the value of ϵ is known, the Φ_Δ values of these photosensitizers can be calculated.

It is found that the singlet oxygen photosensitizing efficiency of H[TBC(O^{i}Pr)$_4$P(OH)] is much higher than empty phthalocyanine H$_2$Pc(O^{i}Pr)$_4$, and is comparable to zinc phthalocyanine ZnPc(O^{i}Pr)$_4$.

3.3. Photo generation of superoxide anion $O2^{\cdot -}$ by H[TBC(O^{i}PR)$_4$P(OH)] (Type I photosensitization)

Using ESR spin-trapping technique, H[TBC(O^{i}Pr)$_4$P(OH)] photosensitized formation of the superoxide anion was studied . A typical ESR spectrum of the spin adduct DMPO–O$_2^{\cdot -}$, consisting of 12 lines, is given in Fig. 4. The hyperfine coupling constants ($\alpha_N = 13.3$ G, $\alpha_H^\beta = 10.5$ G, $\alpha_H^\gamma = 1.4$ G) are in agreement with the literature [12]. No ESR signal can be observed in the absence of either oxygen, photosensitizer or light, which confirms that the superoxide anion is generated by Type I mechanism.

Generally, the superoxide anion can be produced via electron transfer from either the lowest excited singlet (S$_1$) state or the lowest excited triplet (T$_1$) state of photosensitizer to oxygen in the ground state. In order to decide which of these two excited states (S$_1$, T$_1$ or both) is involved in the photogeneration of superoxide anion, the free energy change ΔG on one-electron transfer from H[TBC(O^{i}Pr)$_4$P(OH)] (in whatever state) to oxygen has been evaluated according to the following equations [13]

$$e_{(\mathrm{D^+/D})}{}^* = e_{(\mathrm{D^+/D})} - E^*/F \qquad (8)$$

$$\Delta G^* = F[e_{(\mathrm{D^+/D})}{}^* - e_{(\mathrm{A/A^-})}] \qquad (9)$$

where E^* is the excited state energy, $e_{(\mathrm{D^+/D})}$ is the oxidation potential of photosensitizer in ground state measured by cyclic voltammeter, $e_{(\mathrm{D^+/D})}{}^*$ is the oxidation potential of photosensitizer in excited state.$e_{(\mathrm{A/A^-})}$ is the reduction potential of oxygen (−0.86 V vs. SCE) [14]. ΔG^* is the free energy change in the excited state electron transfer reaction.

For the S$_1$ state of H[TBC(O^{i}Pr)$_4$P(OH)] as the electron donor, the free energy change ΔG^S of electron transfer is negative. Conversely, when the T$_1$ state acts as the electron donor , the free energy change ΔG^T will be positive. In view of thermodynamic principles, the electron transfer from S$_1$ state to oxygen is feasible, while from T$_1$ state to oxygen is forbidden. This conclusion is consistent with the fluorescence quenching of H[TBC(O^{i}Pr)$_4$P(OH)] by oxygen in DMF solution as shown in Fig. 5. The higher concentration of oxygen in solution, the fluorescence intensity tends to be lower.

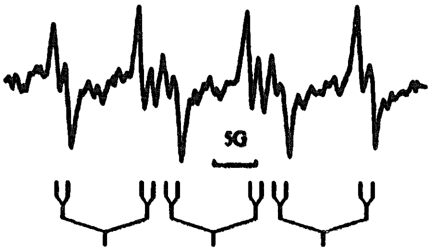

Fig. 4. ESR spectrum of [DMPO–O$_2^{\cdot -}$] spin adduct obtained by irradiating oxygenated DMF solution of H[TBC(O^{i}Pr)$_4$P(OH)] (0.5 mM) and DMPO (10 mM).

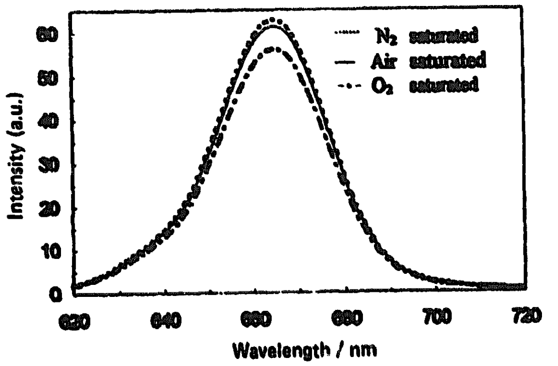

Fig. 5. Fluorescence spectra of $H[TBC(O^iPr)_4P(OH)]$ (5 μM) in DMF solutions saturated by nitrogen, air and oxygen, respectively, $\lambda_{ex} = 600$ nm.

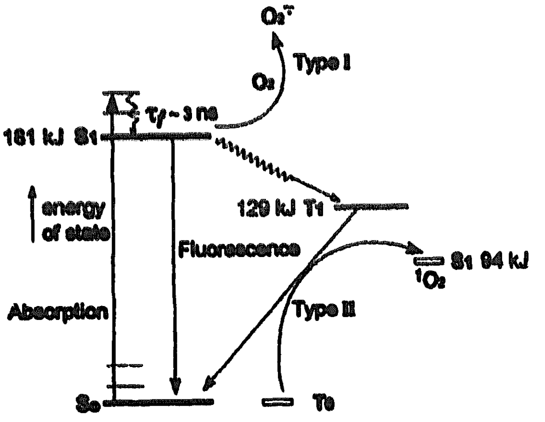

Fig. 6. Generation of excited $H[TBC(O^iPr)_4P(OH)]$ states and reactive dioxygen species.

As to $ZnPc(O^iPr)_4$ and $H_2Pc(O^iPr)_4$, the superoxide anion can also be generated by their photosensitization. And the interpretation of ΔG^* values for these two photosensitizers leads to the same conclusion as for $H[TBC(O^iPr)_4P(OH)]$, i.e. the O_2^- mainly comes from their excited singlet states.

4. Conclusions

The processes involved in the photosensitization of $H[TBC(O^iPr)_4P(OH)]$ can be summarized in an extended Jablonski diagram, Fig. 6. It is likely that Type II photosensitization predominates in DMF solution. The comparative study of $H[TBC(O^iPr)_4P(OH)]$, $ZnPc(O^iPr)_4$ and $H_2Pc(O^iPr)_4$ shows that $H[TBC(O^iPr)_4P(OH)]$ is an efficient singlet oxygen-generating dye, the quantum yields Φ_Δ's sequence is $ZnPc(O^iPr)_4 \cong H[TBC(O^iPr)_4P(OH)] > H_2Pc(O^iPr)_4$.

One of the requirements in tumour phototherapy is to improve the comprehensive properties of the photosensitizers, in which singlet oxygen photosensitizing efficiency is an important criterion for the selection and comparison, since singlet oxygen is the key agent in phototherapy. Taking this into account, further research on the $H[TBC(O^iPr)_4P(OH)]$ photosensitizing properties is worthwhile.

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References

[1] A.L. Thomas, *Phthalocyanine Research and Applications*, CRC Press, Boca Raton, Florida, 1990, p. 2.

[2] J.B. Liu, F.S. Zhang, F.Q. Zhao, Y.W. Tang, X.Q. Song and G.Q. Yao, *J. Photochem. Photobiol. A: Chem., 91* (1995) 99.

[3] M. Fujiki, H. Tabei and K. Isa, *J. Am. Chem. Soc., 108* (1986) 1532.

[4] Y. Murakami, Y. Aoyama and K. Tokunaga, *J. Am. Chem. Soc., 102* (1980) 6736.

[5] R. Bonnett, *Chem. Soc. Rev., 24* (1995) 19.

[6] I. Rosenthal and E. Ben-Hur, in C.C. Leznoff and A.B.P. Lever (eds.), *Phthalocyanines: Properties and Applications*, Vol.1, VCH Publishers, New York, 1989, p. 393.

[7] E.E. Wegner and A.W. Adamson, *J. Am. Chem. Soc, 88* (1966) 394.

[8] Z.X. Guo, G.J. Wang, Y.W. Tang and X.Q. Song, *J. Photochem. Photobiol. A: Chem., 88* (1995) 31.

[9] H. Dreeskamp, E. Koch and M. Zander, *Ber. Bunsenges. Phys. Chem., 78* (1974) 1328.

[10] N.J. Turro, *Modern Molecular Photochemistry*, Benjamin/Cummings Publishing Co., Menlo Park, California, 1978, p. 587.

[11] X.F. Zhang and H.J. Xu, *J. Chem. Soc. Faraday Trans., 89* (1993) 3347.

[12] M. Shopova and T. Gantchev, *J. Photochem. Photobiol. B: Biol., 6* (1990) 49.

[13] X.F. Zhang, H.J. Xu and D.W. Chen, *J. Photochem. Photobiol. B: Biol., 22* (1994) 235.

[14] D.T. Sawyer, *Oxygen Chemistry*, Oxford University Press, New York, 1991, p. 34.