LASER APPLICATIONS AND OTHER TOPICS IN QUANTUM ELECTRONICS PACS numbers: 42.62.Be; 42.62.Fi; 87.64.Ni
DOI: 10.1070/QE2005v035n08ABEH004086

Deactivation of the S_1 state of a water-soluble cationic porphyrin in a complex with DNA studied by the method of picosecond absorption spectroscopy

I.V. Sazanovich, V.S. Chirvony

***Abstract*. The deactivation of the excited S_1 state of 5,10,15,20-tetrakis(4-N-methylpyridyl)porphyrin (H_2TMPyP4) in a complex with DNA is studied by the methods of kinetic absorption spectroscopy for different concentration ratios r of porphyrin and DNA base pairs. It is found that, when the DNA bound porphyrin is assumed to be in a monomer (non-aggregate) state ($r = 1 : 30$), the S_1 state of porphyrin excited by high-power picosecond pulses is rapidly deactivated, which is manifested as an additional decay component with the decay time 200 – 250 ps. This effect is interpreted as singlet – singlet annihilation of excitation indicating the presence of sites on the DNA surface with a higher local concentration of porphyrin molecules. The decrease in the relative content of DNA in solution down to $r = 1 : 3$ further accelerates the deactivation of the S_1 state of porphyrin and causes the hypsochromic shift of its absorption spectrum due to the formation of stack-like porphyrin aggregates along the DNA surface. It is assumed that the additional component with the decay time 44 ± 7 ps observed in the decay curve of the S_1 state is caused by the annihilation of excitation in porphyrin aggregates.**

Keywords: *cationic porphyrin, DNA, picosecond absorption spectroscopy, aggregation, singlet – singlet annihilation.*

1. Introduction

Much attention devoted to the study of interaction of porphyrins with nucleic acids, in particular, with DNA is caused by a number of promising biomedical applications of porphyrins. The convenient spectral properties of porphyrins such as strong absorption in the visible and near-IR regions, the high quantum yield of the triplet state, rather intense fluorescence in the near-IR region, and paramagnetism of some metal derivatives of porphyrins [1 – 9] make possible to use them as probes and contrast agents in the fluorescence, radiology, and magnetic-resonance diagnostics of oncology diseases and as photosensitisers in the photodynamic phototherapy of cancer [10 – 18]. At present it is known that porphyrins reveal photodynamic activity with respect to virus and bacterial infections [19 – 23], in particular, to the HIV virus [24, 25].

Recently the investigation of the interaction of porphyrins with DNA has received a new impetus due to progress in the field of molecular electronics and nanotechnologies. It was found that natural and synthetic nucleic acids can be used as a carcass for the construction of ordered supramolecular systems with specific properties both in solutions and upon deposition onto solid substrates [26 – 28]. Porphyrins are one of the types of molecules capable of assembling on the DNA template, and due to the development of chemistry of porphyrins it is possible to use supramolecular porphyrin complexes for a variety of applications.

The high efficiency of binding of porphyrins with DNA has been long established, papers [29 – 31] being some of the first in this field. The biological action of porphyrin molecules depends on their aggregate state. It was established that the aggregation of porphyrins results in the changes of their absorption spectra, the fluorescence intensity, paramagnetic properties, and the formation efficiency and lifetime of the triplet state, which in turn determines the ability of porphyrins to generate singlet oxygen (see [32 – 35] and references therein).

The interaction of water-soluble tetracationic metal-free 5,10,15,20-tetrakis(4-N-methylpyridyl)porphyrin (H_2TMPyP4) (Fig. 1) with nucleic acids was studied most thoroughly. This porphyrin attracts attention because it can efficiently generate singlet oxygen, exhibits fluorescence, and contains four positively charged methylpyridyl groups in the periphery of its macrocycle (which provides its efficient binding

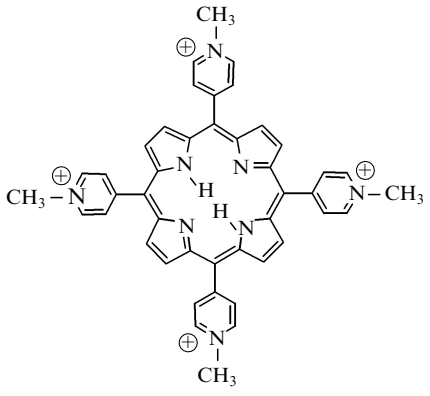

Figure 1. Structural formula of metal-free cationic 5,10,15,20-tetrakis(4-N-methylpyridyl)porphyrin (H_2TMPyP4).

I.V. Sazanovich, V.S. Chirvony Institute of Molecular and Atomic Physics, National Academy of Sciences of Belarus, prosp. F. Skoriny 70, 220072 Minsk, Belarus; e-mail: sazanovich@imaph.bas-net.by

Received 24 February 2005; revision received 27 June 2005
Kvantovaya Elektronika **35** (8) 756 – 760 (2005)
Translated by M.N. Sapozhnikov

with DNA). In addition, it is assumed that H_2TMPyP4 can exert direct photodynamic action on DNA due to transfer of an electron from the guanine base of DNA to porphyrin without the participation of singlet oxygen [36].

It was long assumed that H_2TMPyP4 does not form aggregates on the surface of nucleic acids. The aggregation of this porphyrin in the presence of polynucleotide was first discovered in 1997 upon its binding with polynucleotides of the RNA type and hybrid RNA–DNA polynucleotides [37]. The aggregation of H_2TMPyP4 on the DNA surface was observed one year later [38, 39]. It was found at a high relative concentration of porphyrin $r = 1 : 3$ (r is the ratio of the number of porphyrin molecules to the number of DNA base pairs) and a low ionic strength μ of solution (0.01 M) and was manifested in a decrease in the integrated intensity of porphyrin fluorescence by a factor of 2–2.5 and a decrease in the signal of triplet–triplet absorption of porphyrin approximately by an order of magnitude. However, the authors of papers [38, 39] have failed to observe the decay kinetics of the S_1 state of aggregated porphyrin because of an insufficient time resolution of their equipment.

Thus, although the formation of stack-like H_2TMPyP4 aggregates along the DNA surface for large r was discovered by now, the influence of such interaction with DNA on the deactivation dynamics of the excited states of porphyrin was not studied, which made it impossible, in particular, to predict the possible influence of aggregation on the photodynamic activity and fluorescent properties of H_2TMPyP4 in complexes with nucleic acids at relatively high contents of porphyrin.

This paper is devoted to the study of the deactivation kinetics of the lower excited S_1 singlet state of H_2TMPyP4 bound with DNA at high ($r = 1 : 30$) and low ($r = 1 : 3$) relative concentrations of DNA by the method of kinetic absorption spectroscopy with the picosecond time resolution. The obtained results allow us to determine more accurately the limits of relative porphyrin concentrations at which its aggregation along the DNA surface begins. This is of interest for preparation of the porphyrin–DNA complexes that do not contain porphyrin aggregates and, on the contrary, intended for obtaining large surface aggregates.

2. Experimental

Metal-free catonic H_2TMPyP4, in which iodine was used as a counter ion, was kindly placed at our disposal by Dr. V.L. Malinovskii (A.V. Bogatsky Physico-Chemical Institute, Odessa, Ukraine). DNA from calf thymus was purchased from Sigma Chemical Co. and was used without additional purification. All spectral measurements were performed at room temperature in a phosphate buffer with pH = 6.8 containing 14 mM of KH_2PO_4 and 6 mM of Na_2HPO_4 at the ionic strength of solution $\mu = 0.03$ M. Samples were prepared by mixing the concentrated porphyrin solution with the DNA solution. Concentrations of samples were measured with a spectrophotometer by using the known extinction coefficients of porphyrin and DNA [30].

Samples of porphyrin with DNA had the molar ratios of concentrations of porphyrin and DNA base pairs $r \sim 1 : 30$ and $\sim 1 : 3$. The porphyrin concentration was $\sim 2 \times 10^{-5}$ M. Measurements were performed in silica or glass cells of length 1 or 2 mm. The stationary absorption spectra were measured with a Cary 500 Scan spectrophotometer (Varian). Kinetic absorption measurements were performed by using a modified picosecond absorption spectrometer described in [40].

Samples were excited by the 532-nm second harmonic of a passively mode-locked Nd^{3+} : YAG laser. Probing was performed by using frequency-doubled radiation from an optical parametric oscillator temperature-tunable in the wavelength range from 380 to 1000 nm. Probe radiation was polarised at an angle of 54.7° (the so-called magic angle) with respect to exciting radiation to avoid the manifestation of rotational relaxation of porphyrin molecules in the kinetics of the process [41]. The FWHM of the instrumental function of the picosecond spectrometer was ~ 30 ps. The absorption kinetics of samples was approximated by a sum of two exponentials by the method of least squares. The instrumental function represented by a Gaussian with the FWHM of 27 ps was deconvoluted from the experimental kinetics.

3. Experimental results

Figure 2 presents the stationary absorption spectra of H_2TMPyP4 in a mixture with DNA for $r = 1 : 30$ and $1 : 3$ and the absorption spectrum of free (not bound with DNA) porphyrin in a water buffer. Compared to the absorption spectrum of free porphyrin, the Soret band in the absorption spectrum of the porphyrin–DNA mixture is broadened and exhibits the bathochromic shift from 422 to 440 nm (for $r = 1 : 30$) and 434 nm (for $r = 1 : 3$). Such spectral behaviour is described by the binding of porphyrin with DNA in two ways: by intercalation and groove binding [30]. Of special interest for us is the difference between the absorption spectra of H_2TMPyP4 bound with DNA for different r. One can see from Fig. 2 that, when the relative amount of DNA in solution decreases (i.e., in passing from $r = 1 : 30$ to $r = 1 : 3$), the Soret band shifts to the blue by ~ 6 nm.

Figure 3 shows the bleaching decay kinetics in the Soret band (probed in the wavelength range 420–422 nm) for the H_2TMPyP4–DNA mixture for $r = 1 : 30$ and 1:3. Similar results were obtained by probing in the wavelength range 440–445 nm. Figure 4 shows for comparison the kinetics of bleaching decay for free H_2TMPyP4 is a water buffer. In this case, only a weak decay with the time constant above

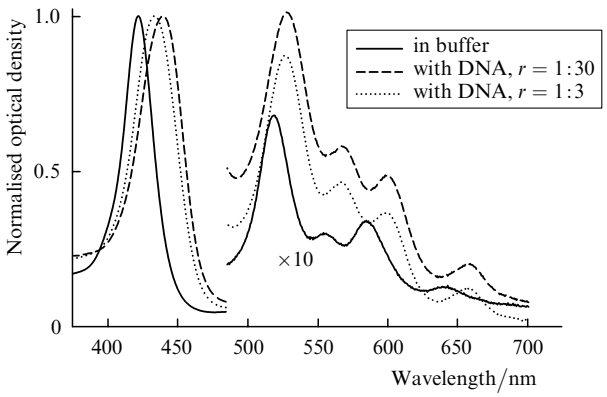

Figure 2. Absorption spectra of free H_2TMPyP4 in water buffer (solid curves) and the H_2TMPyP4–DNA mixture for $r = 1 : 30$ (dashed curves) and $r = 1 : 3$ (dotted curves).

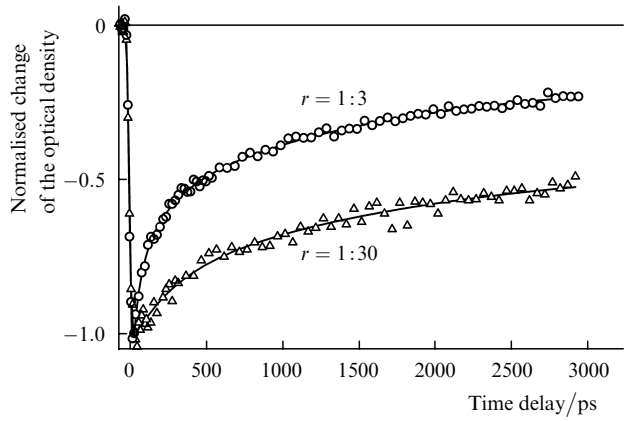

Figure 3. Bleaching decay kinetics for the H_2TMPyP4–DNA mixture for $r = 1:30$ obtained by probing in the wavelength range from 420 to 422 nm. Solid curves are the corresponding exponential approximations.

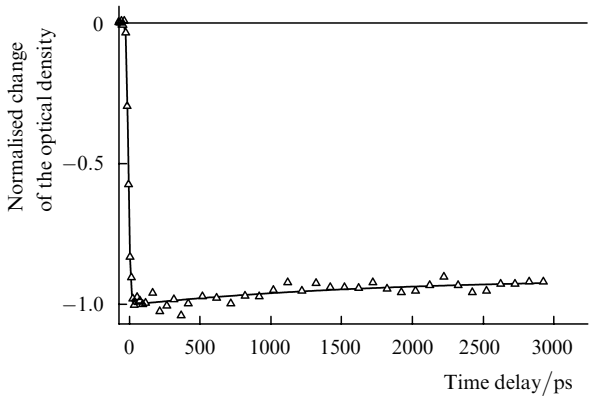

Figure 4. Bleaching decay kinetics for free H_2TMPyP4 in water buffer obtained by probing at 423 nm. The model curve is the sum of a slowly decaying component (with the time constant longer than 1 ns and the weight contribution ~ 10 %) and a constant component (~ 90 %).

1 ns was observed in the time range from 0 to 3 ns, which corresponds to the deactivation of the S_1 state of free porphyrin (with the lifetime equal to 4–5 ns [32, 36, 42–48]) mostly due to intersystem crossing to the T_1 state [36, 44]. At the same time, the bleaching intensity of the H_2TMPyP4–DNA mixture decreased by 50 %–85 % for the first 3 ns.

Analysis of our experimental data showed that the bleaching decay for H_2TMPyP4 bound with DNA for $r = 1:30$ is described by two exponentials with the time constants $\tau_1 = 265 \pm 80$ ps (with the weight contribution 19 % at the probe wavelength of 422 nm or 21 % at 445 nm) and $\tau_2 = 1.9 \pm 0.6$ ns (with the weight contribution 38 % at 422 nm and 65 % at 445 nm) and by the non-decaying component with the weight contribution 42 % (422 nm) or 14 % (445 nm). For $r = 1:3$, the bleaching decay for H_2TMPyP4 bound with DNA occurs more efficiently (Fig. 3). Apart from the two decay components with the time constants compared to those for the case $r = 1:30$ ($\tau_1 \sim 200 - 250$ ps, $\tau_2 \sim 1 - 2$ ns), for $r = 1:3$, another component with a shorter decay time is observed.

As a whole, the decay of photoinduced bleaching of porphyrin bound with DNA for $r = 1:3$ is well described by the sum of three exponentials with a small contribution of a constant component. The time constants of the decay components for $r = 1:3$ are $\tau_1 = 44 \pm 7$ ps (the weight contribution is 31 % for $\lambda = 420$ nm and 32 % for $\lambda = 440$ nm), $\tau_2 = 201 \pm 60$ ps (the weight contribution is 22 % for $\lambda = 420$ nm and 23 % for $\lambda = 440$ nm), and $\tau_3 = 1.5 \pm 0.3$ ns (the weight contribution is 33% for $\lambda = 420$ and 440 nm). The contribution of the constant component is 14 % for $\lambda = 420$ nm and 11 % for $\lambda = 440$ nm.

4. Discussion

The blue shift of the Soret band by 6 nm observed in stationary absorption measurements with decreasing the relative concentration of DNA (Fig. 2) can be reasonably explained by a decrease in the fraction of porphyrin molecules intercalated in DNA due to the formation of porphyrin aggregates along the DNA surface. It was shown in previous studies that the blue shift of the Soret band was greater when H_2TMPyP4 was bound on the DNA surface than upon its intercalation [30]. A similar blue shift of the Soret band of H_2TMPyP4 bound with DNA with decreasing the relative amount of DNA was observed in papers [38, 39] and was explained by the aggregation of porphyrin molecules along the DNA surface. Note that in this case the absorption spectrum of H_2TMPyP4 for $r = 1:3$ does not exhibit any broadening of the absorption bands or formation of new bands, which are typical for porphyrin aggregation. This can be explained by a more ordered structure of porphyrin aggregates formed along the DNA surface, which serves as the structure-forming frame.

Before analysing the experimental results presented in Figs 3 and 4, note that the decay of the photoinduced optical density changes at the time scale studied in our paper is caused completely by the deactivation of the lower excited S_1 state of porphyrin, whereas the lower excited triplet T_1 state of porphyrin bound with DNA in the presence of dissolved oxygen from the air has the lifetime of a few units or tens of microseconds [36] and makes only a constant contribution to the decay kinetics.

The presence of the component with the time constant 200–250 ps in the decay curves obtained for $r = 1:30$ demonstrates a more rapid deactivation of the S_1 state of porphyrin under these conditions that it was assumed earlier based on kinetic fluorescence studies [36, 42]. Indeed, as was shown in these papers, the binding of H_2TMPyP4 near the guanine bases of DNA or poly(dG-dC)$_2$ leads to the quenching of the S_1 state of porphyrin due to the photoinduced electron transfer from guanine to photoexcited porphyrin. In this case, the lifetime of the S_1 state of H_2TMPyP4 measured by the fluorescence method decreased to ~ 2.5 ns.

It is unlikely that the absence of a component with the decay time 200–250 ps in kinetic fluorescence measurements [36, 42] can be explained by an inadequate time resolution of the equipment. The setup for time-correlated single-photon counting used in [42] provided the time resolution better than 50 ps, which together with a sufficiently large number of accumulated counts (~ 10^6) should provide the required resolution of this component.

We assume that our picosecond absorption measurements demonstrate the singlet–singlet ($S_1 - S_1$) annihilation of the electronic excitation energy in porphyrin molecules bound with DNA at comparatively small distances from each other. Although for the ratio $r = 1:30$, approximately 30 base pairs should be located on the average between

porphyrin molecules (which is equivalent to the distance ~ 10 nm [49]), the possibility of a denser localisation of porphyrin on DNA due to fluctuations of binding sites cannot be excluded. Moreover, the manifestation of the initial stage of porphyrin aggregation on DNA already for $r = 1:30$ also cannot be excluded, for example, the formation of porphyrin dimers, which cannot be observed by the methods of circular dichroism and stationary absorption.

The method of kinetic absorption spectroscopy has a higher sensitivity (compared to fluorescence spectroscopy) to the detection of singlet–singlet annihilation in porphyrin molecules bound with DNA because the concentration of excited porphyrin molecules in a sample in this method is considerably higher. The amount of excited porphyrin molecules in our experiments was a few tens of percents of their total amount, which provided the differences in concentrations of excited molecules by a few orders of magnitude compared to [42]. Note that a similar difference of the decay parameters of excited states measured by the method of kinetic spectroscopy with high-power laser excitation from these parameters measured by the method of kinetic fluorescence spectroscopy in the case of the $S_1 - S_1$ annihilation was pointed out in the study of porphyrin aggregates [33].

Therefore, we interpret our picosecond absorption measurements as the evidence that even under conditions, when according to the data in the literature, porphyrin bound with DNA is assumed a monomer (i.e., for $r = 1:30$), the local sites of closely spaced porphyrin molecules are observed. These sites can serve as the centres of growth of large-scale porphyrin aggregates when the amount of DNA in solution is decreased.

It is reasonable to expect that the lifetimes of singlet and triplet excited states of porphyrin will decrease substantially after the formation of stack-like aggregates on the external DNA surface due to the singlet–singlet annihilation of the electronic excitation energy. Therefore, the fast component with $\tau \sim 44$ ps observed in the deactivation kinetics of the H_2TMPyP4–DNA mixture for $r = 1:3$ (Fig. 3) can be assigned to porphyrin molecules that form stack-like aggregates along the DNA chain.

We emphasise that a more rapid deactivation of the S_1 state of porphyrin is caused by a decrease in the DNA amount in solution because for $r = 1:30$, i.e., when the concentration of DNA base pairs exceeds that of porphyrin by 30 times, the component with $\tau = 44 \pm 7$ ps was not observed in the bleaching decay kinetics (Fig. 3). Also, no manifestations of aggregation and quenching of the S_1 state of porphyrin (at the same concentration) in the buffer solution in the absence of DNA was observed, in accordance with the data from the literature. Figure 4 shows that only a weak bleaching decay for porphyrin is observed with the time constant longer than 1 ns, in accordance with the lifetime of the S_1 state equal to ~ 4.5 ns [32, 36, 42–48]. Our assumption that in the presence of DNA for $r = 1:3$ we observe in kinetic measurements the influence of the H_2TMPyP4 aggregation is in good agreement with the results of stationary absorption measurements demonstrating the increase in the relative amount of porphyrin bound on the external DNA surface in passing from $r = 1:30$ to $r = 1:3$.

A short lifetime of the excited S_1 state of aggregated porphyrin (~ 44 ps) inevitably results in a low yield of the long-lived triplet state of porphyrin bound with DNA and in a decrease in the efficiency of electron transfer between guanine and porphyrin. The weight contribution of the constant component to the total bleaching decay kinetics measured in our experiments is 11 %–14 %, which gives a rough estimate of the triplet-state yield (more exactly, its upper limit). These data are in good agreement with the results obtained in [39] by the method of microsecond absorption spectroscopy.

5. Conclusions

The results obtained in this paper for the complex of H_2TMPyP4 with DNA for $r = 1:30$ (when porphyrin bound with DNA is assumed a monomer) suggest the presence of local sites with closely spaced porphyrin molecules on the DNA surface. The fast component with the time constant 44 ± 7 ps observed in the decay kinetics of photoinduced absorption upon aggregation of porphyrin molecules on the DNA surface ($r = 1:3$) corresponds most likely to the deactivation of the excited S_1 state of porphyrin aggregates.

Acknowledgements. The authors thank S.I. Shishporenok for her help in the preparation of samples and measurements of stationary absorption spectra and Prof. P.-Y. Turpin (Pierre and Marie Curie University, Paris, France) for useful discussions of the results of the paper. This work was supported by the Belarusian State Program for Fundamental Research (theme Spectr-06), NATO Public Diplomacy Division (Grant No. PST.CLG.980203) and the Belarusian Republic Foundation for Fundamental Research (Grant Nos F04-274 and B04MC-015).

References

1. Gurinovich G.P., Sevchenko A.N., Solov'ev K.N. *Spektroskopiya chlorofilla i rodstvennykh soedinenii* (Spctroscopy of Chlorophyll and Related Compounds) (Minsk: Nauka i Tekhnika, 1968).
2. Smith K.M. (Ed) *Porphyrins and metalloporphyrins* (Amsterdam: Elsevier, 1975).
3. Dhagarov B.M., Gurinovich G.P., in *Vozbuzhdennye molekuly. Kinteika prevrashchenii* (Excited Molecules: The Kinetics of Transformations), Ed. by Krasnovsky A.A. (Leningrad: Nauka, 1982).
4. *Porfiriny: struktura, svoistva, sintez* (Porphyrins: Structure, Properties, Synthesis), Ed. by Enikolopyan N.S. (Moscow: Nauka, 1985).
5. *Porfiriny: spektroskopiya, elektrokhimiya, primeneniy* (Porphyrins: Spectroscopy, Electrochemistry, Applications), Ed. by Enikolopyan N.S. (Moscow: Nauka, 1987).
6. *Uspekhi khimii porfirinov. Tom 1* (Advances in the Chemistry of Porphyrins, Vol. 1), Ed. by Golubchikov O.A. (St. Petersburg: Research Institute of Chemistry, St. Petersburg State University, 1997).
7. *Uspekhi khimii porfirinov. Tom 2* (Advances in the Chemistry of Porphyrins, Vol. 2), Ed. by Golubchikov O.A. (St. Petersburg: Research Institute of Chemistry, St. Petersburg State University, 1999).
8. *Uspekhi khimii porfirinov. Tom 3* (Advances in the Chemistry of Porphyrins, Vol. 3), Ed. by Golubchikov O.A. (St. Petersburg: Research Institute of Chemistry, St. Petersburg State University, 2001).
9. *Uspekhi khimii porfirinov. Tom 4* (Advances in the Chemistry of Porphyrins, Vol. 4), Ed. by Golubchikov O.A. (St. Petersburg: Research Institute of Chemistry, St. Petersburg State University, 2004).

10. Kvam E., Moan J. *Photochem. Photobiol.*, **52**, 769 (1990).
11. Ali H., van Lier J.E. *Chem. Rev.*, **99**, 2379 (1999).
12. Henderson B.W, Dougherty T.J. *Photochem. Photobiol.*, **55**, 145 (1992).
13. Mironov A.F., in *Uspekhi khimii porfirinov. Tom 1* (Advances in the Chemistry of Porphyrins, Vol. 1), Ed. by Golubchikov O.A. (St. Petersburg: Research Institute of Chemistry, St. Petersburg State University, 1997) p. 357.
14. Reshetnikov A.V., Shvets V.I., Ponomarev G.V., in *Uspekhi khimii porfirinov. Tom 2* (Advances in the Chemistry of Porphyrins, Vol. 2), Ed. by Golubchikov O.A. (St. Petersburg: Research Institute of Chemistry, St. Petersburg State University, 1999) p. 70.
15. Pandey R.K., Zheng G., in *The Porphyrin Handbook. Vol. 6 / Applications: Past, Present and Future*. Kadish K.M., Smith K.M., Guilard R. (Eds). (San Diego: Academic Press, 2000) p. 157.
16. Evstigneeva R.P., in *Uspekhi khimii porfirinov. Tom 3* (Advances in the Chemistry of Porphyrins, Vol. 3), Ed. by Golubchikov O.A. (St. Petersburg: Research Institute of Chemistry, St. Petersburg State University, 2001) p. 150.
17. Mironov A.F., in *Uspekhi khimii porfirinov. Tom 4* (Advances in the Chemistry of Porphyrins, Vol. 4), Ed. by Golubchikov O.A. (St. Petersburg: Research Institute of Chemistry, St. Petersburg State University, 2004) p. 271.
18. Ziolkowski P., in *Uspekhi khimii porfirinov. Tom 4* (Advances in the Chemistry of Porphyrins, Vol. 4), Ed. by Golubchikov O.A. (St. Petersburg: Research Institute of Chemistry, St. Petersburg State University, 2004) p. 293.
19. Kasturi C., Platz M.S. *Photochem. Photobiol.*, **56**, 427 (1992).
20. Merchat M., Spikes J.D., Bertoloni G., Jori G. *J. Photochem. Photobiol. B: Biol.*, **35**, 149 (1996).
21. Li H., Fedorova O.S., Grachev A.N., Trumble W.R., Bohach G.A., Czuchajowski L. *Biochim. Biophys. Acta*, **1354**, 252 (1997).
22. Rovaldi C.R., Pievsky A., Sole N.A., Friden P.M., Rothstein D.M., Spacciapoli P. *Antimicrob. Agents Chemother.*, **44**, 3364 (2000).
23. Valles M.A., in *Uspekhi khimii porfirinov. Tom 3* (Advances in the Chemistry of Porphyrins, Vol. 3), Ed. by Golubchikov O.A. (St. Petersburg: Research Institute of Chemistry, St. Petersburg State University, 2001) p. 179.
24. Grandadam M., Ingrand D., Huraux J.-M., Aveline B., Delgado O., Vever-Bizet C., Brault D. *J. Photochem. Photobiol. B: Biol.*, **31**, 171 (1995).
25. Vever-Bizet C., Boutorine A.S., Delgado O., Brault D., Hélène C. *FEBS Lett.*, **462**, 467 (1999).
26. Winfree E., Liu F., Wenzler L.A., Seeman N.C. *Nature*, **394**, 539 (1998).
27. Braun E., Eichen Y., Sivan U., Ben-Yoseph G. *Nature*, **391**, 775 (1998).
28. Furukawa M., Tanaka H., Kawai T. *J. Chem. Phys.*, **115**, 3419 (2001).
29. Fiel R.J., Howard J.C., Mark E.H., Datta-Gupta N. *Nucleic Acids Res.*, **6**, 3093 (1979).
30. Pasternack R.F., Gibbs E.J., Villafranca J.J. *Biochemistry*, **22**, 2406 (1983).
31. Pasternack R.F., Gibbs E.J., Villafranca J.J. *Biochemistry*, **22**, 5409 (1983).
32. Maiti N.C., Mazumdar S., Periasamy N. *J. Phys. Chem. B*, **102**, 1528 (1998).
33. Khairutdinov R.F., Serpone N. *J. Phys. Chem. B*, **103**, 761 (1999).
34. Kano K., Fukuda K., Wakami H., Nishiyabu R., Pasternack R.F *J. Am. Chem. Soc.*, **122**, 7494 (2000).
35. Micali N., Romeo A., Lauceri R., Purrello R., Mallamace F., Scolaro L.M. *J. Phys. Chem. B*, **104**, 9416 (2000).
36. Chirvony V.S., Galievsky V.A., Kruk N.N., Dzhagarov B.M., Turpin P.-Y. *J. Photochem. Photobiol. B: Biol.*, **40**, 154 (1997).
37. Uno T., Hamasaki K., Tanigawa M., Shimabayashi S. *Inorg. Chem.* **36**, 1676 (1997).
38. Gandini S.C.M., Borissevitch I.E., Perussi J.R., Imasato H., Tabak M. *J. Luminescence*, **78**, 53 (1998).
39. Borissevitch I.E., Gandini S.C.M. *J. Photochem. Photobiol. B: Biol.*, **43**, 112 (1998).
40. Galievsky V.A. *Cand. Sci. Dissertation* (Minsk: Institute of Molecular and Atomic Physics, 2000).
41. Sazanovich I.V., Ganzha V.A., Dzhagarov B.M., Chirvony V.S. *Chem. Phys. Lett.*, **382**, 57 (2003).
42. Sazanovich I.V., Petrov E.P., Chirvony V.S. *Opt. Spektr.*, in print.
43. Vergeldt F.J., Koehorst R.B.M., van Hoek A., Schaafsma T. *J. Phys. Chem.*, **99**, 4397 (1995).
44. Kalyanasundaram K. *Inorg. Chem.*, **23**, 2453 (1984).
45. Liu Y., Koningstein J.A., Yevdokimov Y. *Can. J. Chem.* **69**, 1791 (1991).
46. Kemnitz K., Sakaguchi T. *Chem. Phys. Lett.*, **169**, 497 (1992).
47. Jasuja R., Jameson D.M., Nishijo C.K., Larsen R.W. *J. Phys. Chem. B*, **101**, 1444 (1997).
48. Ito A.S., Azzellini G.C., Silva S.C., Serra O., Szabo A.G. *Biophys. Chem.*, **45**, 79 (1992).
49. Cantor C.R., Schimmel P.R. *Biophysical Chemistry. Part I. The Conformation of Biological Macromolecules* (San Francisco: W.H. Freeman and Company, 1980).