

Laser photochemistry of oxygen. Application to studies of the absorption spectra of dissolved oxygen molecules

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Abstract. This paper summarizes the data of our lab on the rates of photooxygenation of singlet oxygen traps upon direct laser excitation of oxygen in air-saturated organic solvents and water. Methods of application of these data to calculation of absorbance (A) and molar absorption coefficients (ϵ) in the maxima of the main oxygen absorption bands (1273, 765 and 1070 nm) are discussed. The most accurate results were obtained from comparing the photooxygenation rates upon porphyrin-photosensitized and direct excitation of oxygen molecules. It is shown that ϵ_{1273} is not sensitive to the presence of heavy atom (bromine) in solvent molecules and markedly decreases on going from non-polar solvents to water being proportional to the radiative rate constants obtained from the quantum yields of singlet oxygen phosphorescence at 1274 nm. The coefficient ϵ_{765} markedly increases in the presence of bromine. In solvents lacking heavy atoms the 1.5-2-fold increase of ϵ_{765} was observed on going from non-polar solvents to water. Simultaneously, the ratios $\epsilon_{1273}/\epsilon_{765}$ are changed from (7-10)/1 in non-polar solvents to 1.5/1 in water. The value of ϵ_{1070} obtained in carbon tetrachloride is shown to be about two orders smaller than ϵ_{1273} in the same solvent. The results are important for both analyses of oxygen photonics and dosimetry of laser radiation in biomedical experiments. © 2017 Journal of Biomedical Photonics & Engineering.

Keywords: Molecular oxygen; singlet oxygen; photosensitizers; direct oxygen excitation.

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1 Introduction

It has been known since the seminal papers of R. Mulliken that oxygen molecules have triplet ground state and two relatively low-lying singlet levels [1,2]. Fig. 1 shows energy diagram of the major electronic transitions in monomeric oxygen molecules in rarified gas as follows from measurements of the absorption spectra in Earth atmosphere and in the gas phase at atmospheric pressure [3-6]. These transitions are highly forbidden therefore the absorption bands of monomeric oxygen molecules are so weak that they normally cannot be measured using conventional spectrophotometers under ambient conditions.

Reliable absorption spectra were obtained in the gas phase and solutions under high oxygen pressure of 50-140 atm and in liquid oxygen. However, under these conditions dimerization of oxygen molecule occurs, therefore the absorption spectra correspond mainly to dimols (O₂)₂. In the dimol spectra, the IR maxima analogous to those of monomols are shifted toward shorter wavelengths, and a series of additional maxima appear in visible region. Relative intensities of the spectral peaks of dimols are strongly different from those in the gas phase at atmospheric pressure [6-11] (Fig. 2).

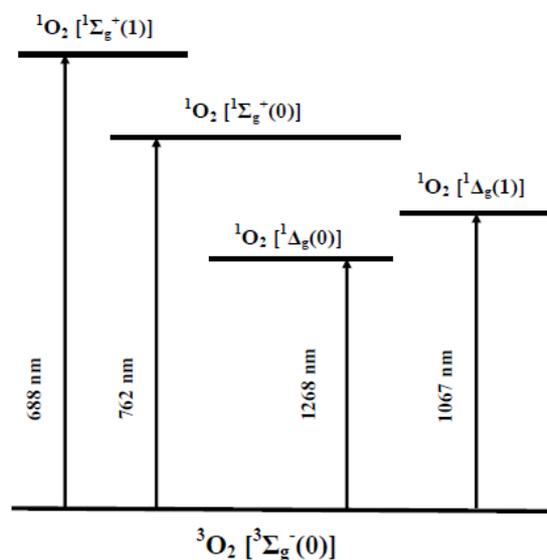


Fig. 1 The main electronic transitions in monomeric oxygen molecules in the rarefied gas as reported in refs [3-6]. The vibrational sublevels are shown in brackets.

It was reported in the classic papers by Evans [7] and Matheson's group [12,13] that excitation of oxygen dimols dissolved in organic solvents at oxygen pressure 130-140 atm. leads to formation of the $^1\Delta_g$ singlet oxygen, which causes oxygenation of added organic

singlet oxygen traps, aminoacids, proteins and other molecules.

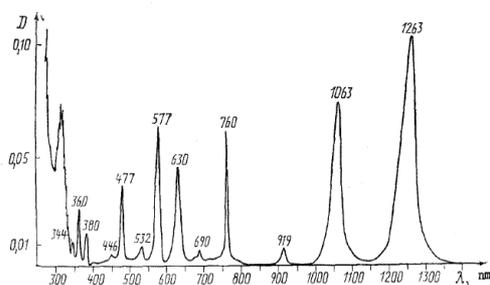


Fig. 2 Absorption spectrum of oxygen dimols in compressed oxygen at 150 atm. in the 5 cm optical path [10,11].

Based on these experiments, Ambartzumian and his collaborators advanced an idea that direct laser excitation of intrinsic dimol and monomol oxygen molecules in living cells might be a reason for biological action of laser radiation in visible and infrared [14,15]. It was also suggested that laser excitation of oxygen might be applied to destruction of cancer cells in vitro and in animal and human organisms.

These papers caused heated discussions. Apparently, biological action caused by laser excitation of oxygen molecules should be directly proportional to the rate of $^1\text{O}_2$ production by laser radiation, which in turn, depends on laser power (I_{las}) and the fraction of laser light absorbed by oxygen molecules ($1-10^{-A_{ox}}$), where A_{ox} is oxygen absorbance ($A_{ox} \ll 0.01$). Hence:

Biological effect ~

$$\sim I_{las} (1-10^{-A_{ox}}) = I_{las} \times 2.3 \times A_{ox}. \quad (1)$$

Although biological and therapeutic action of red and IR lasers has been reported many times ([15-19] and refs therein) there is no crucial evidence showing that this action under ambient conditions and moderate (therapeutic) laser power is always due to oxygen excitation. Thus, reliable information on absorption coefficients of oxygen molecules under normal conditions is of great importance for both understanding of oxygen photonics in natural environment and mechanistic analyses of laser action and dosimetry of IR laser radiation in biomedical experiments.

Several years ago our group started work on mimicking biological effects of laser radiation in solutions of singlet ($^1\Delta_g$) oxygen traps in organic solvents and water at normal pressure and temperature. It was established that monomeric oxygen molecules dissolved in these solvents work as photosensitizers of oxygenation reactions. Kinetic analyses of the reaction rates allowed us to develop methods for calculation of absorbance (optical density) for oxygen absorption bands under natural conditions. The present paper summarizes major results of our group [20-28].

2 Experimental procedure

For sample irradiation, diode lasers LAMI Gelios ("Surgical innovation technologies", Moscow) were applied with the maxima of the emission bands at 1267-1270, 1064-1070 and 760-762 nm and the half bandwidth 4-12 nm. Laser light was focused into a fiber light guide with of 1 mm diameter. The eliciting light power, was controlled by the power meter Ophir ORION-TH with the sensor head 20C-SH (Israel). Solutions were irradiated in the 10 mm rectangular quartz cell under normal pressure and room temperature. Volume of the solutions was 1.5 ml. Diameter of the irradiated spot was 5-8 mm.

Tetracene, (Aldrich Chemistry, >98%), 1,3-diphenylisobenzofuran (DPIBF) and uric acid (Acros Organics) were used as singlet oxygen traps. Tetraphenylporphyrin (TPP) (Aldrich, Chemistry) was employed in certain experiments as photosensitizers of singlet oxygen production. Absorption spectra were measured using a Hitachi U-3400 (Japan) and SF-56 (LOMO Spekt, St. Petersburg) spectrophotometers. The rate of $^1\text{O}_2$ formation was calculated from the rate of photobleaching of the main absorption maxima of the traps (474 nm for tetracene, 410-414 nm for DPIBF, 530 nm for rubrene and 290 nm for uric acid) under irradiation by IR lasers.

It was shown that irradiation of air-saturated solutions of the traps in organic solvents and water by IR lasers of moderate power (0.03-2 W) with the emission maxima at 1270, 1070 and 765 nm causes oxygenation of the traps, which is accompanied by bleaching of their main absorption bands. The oxygenation rates were measured as follows. At first, the absorbance (optical density) (A_0) in the main absorption maximum of a trap was recorded. In most experiments, solutions with $A_0 = 0.8-1.5$ in the 1 cm cell quartz were employed. Then, the solutions were irradiated during 1-30 min, shaken, and the absorbance (A_f) was measured again. The photoreaction rates (V_{lasr} , $\Delta A/\text{min}$) were calculated from measuring the illumination time (t) needed to decrease the absorbance so that $\Delta A = A_0 - A_f = 0.05 - 0.1$. As a result, not more than 15% overall trap content was oxidized during irradiation. The oxygenation rates were then normalized to power of laser radiation in einstein/s (see details in refs [20-28]).

2.1 Mechanistic analyses

The photooxygenation action spectra were measured using tunable lasers. These spectra indicate that oxygenation of the traps occurred due to direct laser excitation of oxygen molecules. In CCl_4 , C_6F_6 and ethanol the main IR absorption maximum was found at 1273 nm. The width (FWHM) of this band was 17-18 nm (106 cm^{-1}) [20,21,23] (Fig. 3). This maximum is shifted by 5 nm to longer wavelengths compared to the absorption maxima of monomeric oxygen in rarified gas (Fig. 1). More recently these results were confirmed by

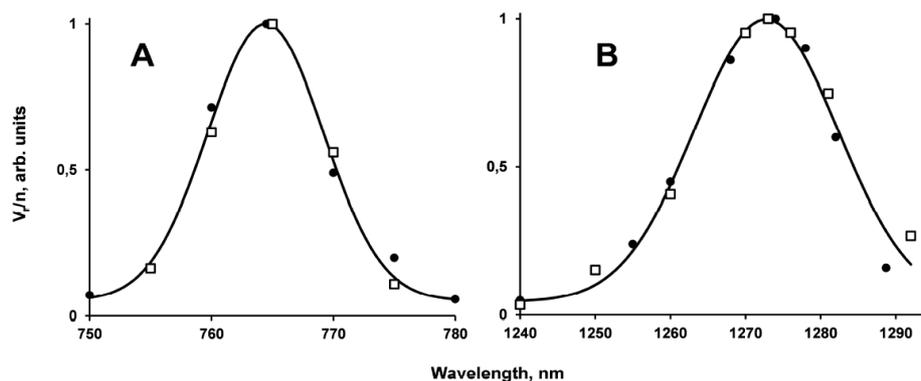
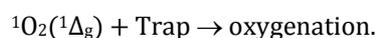
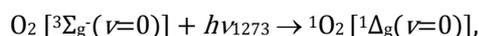
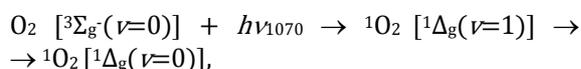
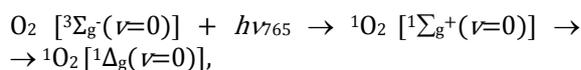


Fig. 3 Action spectra of oxygenation of the singlet oxygen traps under direct excitation of oxygen molecules by IR laser radiation. A: spectra obtained using tunable titan-sapphire laser in solutions of tetracene (•) [24] and 1,3-diphenylisobenzofuran (□) in CCl_4 [26]. B: spectra obtained using tunable forsterite laser in solutions of tetracene in CCl_4 (•) [21] and 1,3-diphenylisobenzofuran in ethanol (□) [23]. V_r is the rate of trap photooxygenation, n is irradiation intensity in number of photons per second.

Courtade's group in France who observed the oxygen excitation band at 1273 nm in CCl_4 , acetone, ethanol and D_2O with FWHM ~ 15 nm [29,30].

The action spectra in the dark red region were reported for the first time in papers of our group [24,26] (Fig. 3). In CCl_4 , C_6F_6 and ethanol the maximum was found at 765 nm, at slightly longer wavelengths than in the rarefied gas (Fig. 1). The band width was estimated to be 8-9 nm (~ 130 cm^{-1}). Recently Ogilby's group in Denmark confirmed main results of our studies using luminescence technique and extended measurements to several additional solvents. In the luminescence experiments of this group the excitation maxima varied within 763-772 nm and FWHM -within 6-17 nm. In agreement with our data in CCl_4 they observed the excitation maximum at 765 nm with the bandwidth of 7 nm (120 cm^{-1}) [31]. Apparently, parameters of the action spectra characterize the major absorption bands of dissolved oxygen under ambient conditions.

In organic solvents the oxygenation of the traps was inhibited by singlet oxygen quencher – β -carotene, in water and alcohols - by sodium azide. The photoreaction was not observed after purging with argon or nitrogen. After saturation with pure oxygen the photoreaction rates increased 5-fold compared to air-saturated solutions [20-24]. Thus, evidence was obtained that under ambient conditions, monomeric oxygen molecules work as photosensitizers of oxygenation reactions similarly to oxygen dimols appearing at high oxygen pressure. The mechanisms of the photoreactions correspond to the following scheme:



According to this scheme, photoexcitation of oxygen at 1273 nm causes direct population of the [$^1\Delta_g(v=0)$] (v is a vibrational sublevel) state. Photoexcitation at 1070 nm causes direct population of the [$^1\Delta_g(v=1)$] state and photoexcitation at 765 nm causes population of the singlet level [$^1\Sigma_g^+(v=0)$]. It is known that in the solution-phase [$^1\Delta_g(v=1)$] and [$^1\Sigma_g^+(v=0)$] rapidly (for nano- or picoseconds) lose extra energy and populate the [$^1\Delta_g(v=0)$] state due to collisions of oxygen and solvent molecules ([19-21]). Thus, no matter what level of oxygen is initially populated, chemical activity is determined by the [$^1\Delta_g(v=0)$] state of O_2 .

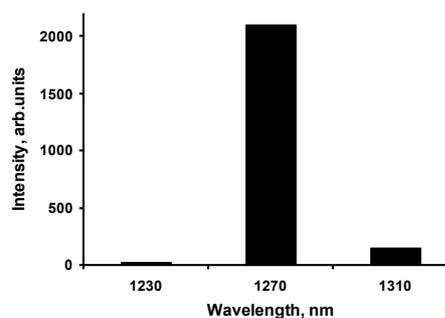


Fig. 4 Spectrum of $^1\text{O}_2$ phosphorescence in air-saturated pigment-free C_6F_6 upon excitation of dissolved oxygen by monochromatic light of cw diode laser at 766 nm. The spectrum was measured using three changeable interference filters transmitting light at 1230, 1270 and 1310 nm respectively. The luminescence data were obtained in collaboration with A.S. Benditkis.

Quite recently we assembled a very sensitive spectrometer, in which phosphorescence of $^1\text{O}_2$ at 1270 nm is detected using cooled S-1 PMT (FEU-112) through three changeable interference filters with the transmission maxima at 1230, 1270 and 1310 nm. In

agreement with the above scheme and the data of Ogilby's group [31] we observed singlet oxygen phosphorescence at 1270 nm in air-saturated CCl_4 and C_6F_6 under excitation by cw laser at 765 nm (900 mW) without pigments-photosensitizers. First results are shown in Fig. 4. No light emission was observed upon excitation at 740 nm. Addition of acetone (50%) caused the decrease of luminescence intensity by more than two orders. Under oxygen purging the luminescence intensity increased (more detailed paper is in preparation).

2.2 Principles of data analysis

Based on this mechanistic scheme, the following equation was proposed for the rate (V_{lasr} , M/s) of the trap photooxygenation under laser excitation of oxygen [27-28]:

$$V_{\text{lasr}} = 2.3 A_{\text{max}} I_{\text{las}} \alpha k_{\text{ox}} [\text{Trap}] / (k_{\Delta} + k_{\text{ox}} [\text{Trap}]), \quad (2)$$

where A_{max} is absorbance (optical density) in the maxima of the oxygen absorption spectrum, I_{las} is the incident photon flux of IR laser radiation in $\text{einstein} \times \text{L}^{-1} \text{s}^{-1}$, k_{ox} is the rate constant of oxygenation of the traps by singlet oxygen; $[\text{Trap}]$ is the molar concentration of the trap; k_{Δ} is the rate constant of the $^1\text{O}_2$ deactivation in the trap-free solutions, i.e. $k_{\Delta} = 1/\tau_{\Delta}$ where τ_{Δ} is the lifetime of singlet oxygen in solvents lacking the singlet oxygen traps.

The coefficient α corresponds to degree of overlapping of the action spectrum with the spectrum of laser radiation. It was estimated from the photooxygenation action spectra (Fig. 3) and laser emission spectra using the "Origin" program:

$$\alpha = \int A(\nu) I(\nu) d\nu / \int I(\nu) d\nu, \quad (3)$$

where $A(\nu)$ is a function characterizing normalized excitation spectrum of oxygen (Fig. 2) in energy units, in which A varies from 0 to 1; and $I(\nu)$ is a function characterizing the normalized spectrum of laser radiation, where I varies from 0 to 1. Apparently, if the wavelength of laser light exactly corresponds to the maximum of the oxygen absorption band, α is equal to 1. Usually, the emission maxima of diode lasers, were shifted to shorter wavelengths compared to the oxygen absorption maxima, therefore α varied from 0.2 to 0.9 (see [27,28] for details).

Eq. 2 allows for accurate determination of the oxygen absorption coefficients corresponding to the maxima of oxygen absorption bands using experimental values for the oxygenation rates under laser irradiation:

$$A_{\text{max}} = V_{\text{lasr}} (k_{\Delta} + k_{\text{ox}} [\text{Trap}]) / 2.3 \alpha I_{\text{las}} k_{\text{ox}} [\text{Trap}]. \quad (4)$$

Knowing A_{max} , one can calculate molar absorption coefficients of oxygen $\varepsilon_{\text{max}} = A_{\text{max}} / [O_2]$, where $[O_2]$ is the molar concentration of oxygen in solutions. The

absorption cross section $\sigma_{\text{max}} = 2300 \varepsilon_{\text{max}} / N_{\text{av}}$ where N_{av} is Avogadro number.

2.3 Absorbance at 1273 nm

The first paper, in which A_{1273} was estimated using Eq. 4 and air-saturated carbon tetrachloride as a solvent, was published in 2004 [25]. The data were obtained from measurement of the oxygenation rates of tetracene upon excitation of dissolved oxygen at 1273 nm by tunable forsterite laser. The authors arrived to the values: $A_{1273} \approx 7.2 \times 10^{-6}$, $\varepsilon_{1273} \approx 0.003 \text{ M}^{-1} \text{cm}^{-1}$, $\sigma_{1273} \approx 10^{-23} \text{ cm}^2$, which served us as a reference during several years. Because of technical limitations higher precision could not be reached at that time. Then, accurate measurements of the relative rates of DPIBF oxygenation in different solvents were carried out. The results allowed analysis of the solvent dependence of the oxygen absorption coefficients. The first paper on this subject was published in 2005 [22]. Later, more detailed papers followed [23,25]. In particular, it was established that in accord with the Einstein law, the molar absorption coefficients corresponding to the oxygen absorption band at 1273 nm in series of organic solvents were proportional to the radiative rate constants (k_r) for this electronic transition obtained from measurement of the quantum yield of singlet oxygen phosphorescence (n is the refractive index) [22-25]:

$$\varepsilon_{1273} \sim k_r / n^2. \quad (5)$$

However, in the papers mentioned above the proportionality was not revealed in water and alcohols. We proposed that this effect might be due to problems in kinetic analyses of the data [26,27]. Indeed, in the first papers we assumed that the rate constant (k_{ox}) for the reaction of the trap with $^1\text{O}_2$ and molar absorption coefficients of traps ($\varepsilon_{\text{trap}}$), which were used for determination of the trap concentrations, did not depend on solvents. In order to solve this problem an experimental procedure was developed, which excluded $\varepsilon_{\text{trap}}$ and k_{ox} from calculations. To reach this goal, the rates of oxygenation of the traps were compared upon direct and photosensitized oxygen excitation at equal trap concentrations. In this case, the following equation was obtained [26-28]:

$$A_{\text{max}} = (V_{\text{lasr}} / V_{\text{psr}}) (I_{\text{psex}} / \alpha I_{\text{las}}) \Phi_{\Delta} (1 - 10^{-A_{\text{ps}}}) / 2.3, \quad (6)$$

where V_{psr} is the rate of photosensitized oxygenation of the traps; I_{las} and I_{psex} are the incident photon flux in $\text{einstein L}^{-1} \text{s}^{-1}$ for light applied to excitation of oxygen and photosensitizer respectively; Φ_{Δ} is the yield of singlet oxygen generation by pigments photosensitizers (TPP and TPPS were employed in our experiments). For calculations with Eq. 6, the photooxygenation rates were measured in the $\Delta A/t$ units, where ΔA is a change of absorbance in the maxima of the absorption spectra of the traps and t is time (minutes) of laser irradiation. Instead, one has to know accurate values of Φ_{Δ} for

Table 1 Values of the oxygen absorption coefficients in several solvents calculated according to Eqs. 4 and 6, using the rates of oxygenation of the singlet oxygen traps under ambient conditions.

	$A_{1273} \times 10^5$ (1 cm)		$A_{765} \times 10^5$	[O ₂], mM [32]	$\epsilon_{1273} \times 10^3$, M ⁻¹ cm ⁻¹	$\epsilon_{765} \times 10^3$, M ⁻¹ cm ⁻¹	$\epsilon_{765}/$ ϵ_{1273}	Permit- tivity
	Eq. 4*	Eq. 6**	(1 cm) Eq. 6**					
CCl ₄	-	1.33	0.19	2.6	5.1	0.7	0.14	2.24
C ₆ F ₆	2.25	2.25	0.34	4.4	5.1	0.77	0.15	2.05
C ₆ H ₆	1.2	1.20	0.13	1.9	6.3	0.68	0.11	2.27
Toluene	1.2	1.18	0.12	1.8	6.5	0.67	0.10	2.38
n-heptane	-	0.92	0.15	2.8	3.3	0.53	0.16	1.94
1-Bromo- hexane	-	0.96	0.36	3.0	3.2	1.2	0.38	5.4
Acetone	0.70	0.66	0.20	2.4	2.8	0.83	0.30	20,7
Ethanol	-	0.48	0.14	1.65	2.9	0.85	0.29	24.5
H ₂ O	-	0.043	0.032	0.29	1.5	1.1	0.75	80.1

*The data of this column were obtained using Eq. 4 from comparison of the rates of DPBIF oxygenation in organic solvents and carbon tetrachloride. The absorption coefficients in CCl₄ were measured using Eq. 6 (see [28] and refs therein). **The numbers in these columns (new data of our experiments) were obtained using Eq. 6 from comparison of the oxygenation rates upon direct and photosensitized oxygen excitation in each solvent. Maximum deviation from the average values indicated in this table did not exceed 10%.

photosensitizers. Our experiments show that for monomeric TPP and TPPS in most solvents used in our work, $\Phi_{\Delta} = 0.73 \pm 0.05$ ([26-28] and refs therein), only in 1-bromohexane we obtained $\Phi_{\Delta} = 0.92 \pm 0.02$ (unpublished).

This method was applied for the first time in 2012 [26], in which we averaged current and previous results of our group for four solvents: CCl₄, acetone, ethanol and water. The ways to estimate coefficients α were not found at that time therefore it was suggested that $\alpha \approx 1$ in all experiments. It was obtained using this assumption, DPBIF and Eq. 6 that absorbance of oxygen at 1270 nm in CCl₄ is 1.5 times higher than we reported previously [21]. Besides, the relative values of the molar absorption coefficients for this band were proved to be proportional to the radiative rate constants (k_r) of singlet oxygen also in water and ethanol. Deviation from the proportionality in water and alcohol reported in the preceding papers [22,23,25] was shown to be due to the strong increase of k_{ox} for DPBIF in these media compared to organic solvents having no OH-groups (see [26,28] for details).

Papers of 2003-2012 solved principal experimental problems arising upon measurement of the absorption coefficients for dissolved oxygen. As a result, estimation of the numerical values for these coefficients in different environment was carried out [24-26]. As knowledge of the oxygen absorption coefficients is of fundamental importance for oxygen photonics, subsequent experiments were aimed at increasing the accuracy of our measurements.

For this goal, diode lasers having relatively narrow spectral band (4-6 nm) were employed. Coefficients α were calculated from the action spectra of trap oxygenation (Fig. 2) and spectra of laser radiation (in energy units) using Eq. 2 (see [27,28] for details). Two methods of analysis were applied. According to one of

them, A_{1273} was carefully measured in CCl₄ using Eq. 6 and relative rates of oxygenation of three traps – DPBIF, tetracene and rubrene upon photosensitized and direct oxygen excitation [28]. It was established that for all traps the relative oxygenation rates upon photosensitized and direct oxygen excitation normalized to power of exciting radiation and the fraction of light absorbed by porphyrins were $\sim 10^4$:

$$(V_{psr} / V_{lasr}) / (I_{psex} / \alpha I_{las}) \approx 10^4. \quad (7)$$

Hence, if 100% exciting light is absorbed by porphyrins, the efficiency of photosensitized oxygen excitation is 10^4 times higher than the efficiency of direct oxygen excitation. In real experiments porphyrin absorbed not more than 10% exciting light, therefore relative efficiency of photosensitized oxygen excitation was by one order smaller, therefore the ratio corresponding to Eq. 7 is about 10^3 [28].

In addition, it was found that A_{1273} in CCl₄ is almost two times greater than the value reported in ref. [21] (Table 1). The difference was shown to be mainly due to the fact that the real value of the molar absorption coefficient for tetracene was almost two times smaller than the literature value applied in the previous work [21] (see [28] for details). The obtained A_{1273} in CCl₄ was then used for correction of numerous results of our previous measurements of A_{1273} in other organic solvents, which were estimated using Eq. 4 and CCl₄ as the reference solvent [28] (Table 1). As mentioned above, these results were obtained upon direct laser excitation of oxygen from relative rates of DPBIF oxygenation in CCl₄ and other organic solvents having no OH groups in their molecules.

According to the second method, the rates of DPBIF oxygenation were compared upon direct and photosensitized oxygen excitation in each solvent. The

Table 2 Correlation between obtained ε_{1273} and the radiative rate constants (k_r, s^{-1}) in several solvents.

Parameters	CCl ₄	C ₆ F ₆	n-heptane	Benzene	Toluene	Acetone	Ethanol	H ₂ O
$\varepsilon_{1273} \times 10^3, M^{-1}cm^{-1}$	5.1	5.1	3.3	6.2	6.7	2.8	2.9	1.5
$\varepsilon_{1273}, \text{rel. units}$	1	1.0	0.65	1.2	1.3	0.55	0.57	0.29
$k_r/n^2, \text{rel. units}$ [23,33]	1	1.0	0.62	1.2	1.2	0.53	0.54	0.25

Table 3 The radiative rate constants (k_r) and the phosphorescence quantum yields (Φ_r) for triplet-singlet transitions in oxygen molecules dissolved in carbon tetrachloride estimated from the experimentally obtained ε_{1273} and ε_{765} (Table 1) using Eq. 8.

Transitions	ν, cm^{-1}	$\Delta\nu, cm^{-1}$	$\varepsilon_{max} \times 10^3, M^{-1}cm^{-1}$	k_r, s^{-1}	$k_r, s^{-1}{}^a$	Φ_r
${}^1\Delta_g(0) \leftarrow {}^3\Sigma_g^-(0)$	7855	~103	5.1	0.60	1.17; 0.25	0.018
${}^1\Sigma_g^+(0) \leftarrow {}^3\Sigma_g^-(0)$	13072	~130	0.71	0.22	0.40; 1.55	2.9×10^{-7}

^aThis column shows the experimentally measured k_r , reported in papers cited in Ref. [26];

^b $\Phi_r = k_r \tau$ where τ is the lifetime of singlet oxygen equal to ~ 30 ms for the ${}^1\Delta_g$ state [37] and 130 ns for the ${}^1\Sigma_g^+$ state in the reagent grade CCl₄ ([38,39] and refs therein), n is 1.46 in CCl₄.

most recent results of our studies are shown in Table 1. As seen from the Table, both methods yield similar values of the absorption coefficients. Correctness of the coefficients ε_{1273} presented in Table 1 is supported by apparent correlation of their relative values with the relative values of k_r/n^2 for the ${}^1\Delta_g$ -state of singlet oxygen in different solvents (Table 2).

Thus, the data of Table 2 indicate that one and the same mechanism is responsible for the solvent dependence of both k_r , calculated from the quantum yield of singlet oxygen phosphorescence at 1270 nm and ε_{1273} obtained by methods of laser photochemistry. Both coefficients are about 1000 times greater than those in rarefied gas (see [21] for refs). It follows from Minaev's theoretical considerations that due to spin-orbit coupling in O₂ molecules and perturbations caused by their collisions with solvents, the $a^1\Delta_g \leftarrow X^3\Sigma_g^-$ transition which is purely magnetic in the rarefied gas becomes allowed as electric dipole in solutions [34,35].

Hence, one can estimate the absolute value of k_r from the obtained ε_{1273} using the equation for the electric dipole transitions (so called, Kravetz equation). The simplest form of this equation was provided by Strickler and Berg [36]:

$$k_r = n^2 2.88 \times 10^{-9} \nu^2 \Delta\nu \varepsilon_{ox} g_1 / g_2, \quad (8)$$

where n is the solvent refractive index, ν is the transition frequency in cm^{-1} , $\Delta\nu$ is the half-width of the oxygen absorption band (FWHM), ε_{ox} is the molar absorption coefficient of oxygen in $M^{-1}cm^{-1}$ and g_1 and g_2 are multiplicity of the initial and final states, $g_1/g_2 = 3/1$ (see also [26] and refs therein). Table 3 shows the results of application of Eq. 8 to the parameters of oxygen obtained in CCl₄ (Table 1) and compares these results with the experimentally measured values obtained by different groups. Taking into account an

approximate character of Eq. 8 and scattering of the experimental numbers, correlation between the calculated and measured k_r is satisfactory.

2.4 Absorbance at 765 nm

The first paper, in which A_{765} was estimated in air-saturated carbon tetrachloride was published in 2007 [24]. Tetracene was used as the singlet oxygen trap and the tunable titan-sapphire laser was applied for oxygen excitation. The maximum of the photooxygenation action spectrum was observed at 765 nm (Fig. 3); A_{765} was roughly estimated to be 3.5 times smaller than A_{1273} and $\varepsilon_{765} \approx 10^{-3} M^{-1}cm^{-1}$ was obtained [24]. Later, similar measurements were performed using DPIBF as a singlet oxygen trap. Values of A_{765} were estimated in carbon tetrachloride, acetone, ethanol and water. It was established that $\varepsilon_{765} \approx 10^{-3} M^{-1}cm^{-1}$ in these solvents [26]. After further improvement of the measurement procedure and data analyses with accounting for coefficients α (see previous section) it was found that ε_{1273} is almost two times greater than that obtained previously and the ratio ($\varepsilon_{1273}/\varepsilon_{765}$) was equal to ~7 in CCl₄. A slight increase of ε_{765} with the increase of solvent polarity was also reported [27,28].

The most recent studies were performed based on Eq. 6 and comparison of the rates of DPIBF oxygenation upon direct and photosensitized oxygen excitation. In these experiments we used a new diode laser with the emission maximum at exactly 765 nm and 2.5 nm bandwidth. Several additional solvents were studied. Some results are presented in Table 1 and Fig. 5. It is seen that the ratio ($\varepsilon_{765}/\varepsilon_{1273}$) varies within 0.1-0.15 in non polar solvents, increases with the increase of solvent polarity to about 0.3 in acetone and ethanol and to 0.75 in water. Exclusion is 1-bromohexane, which despite low polarity, shows relatively high ε_{765} . This

effect can be attributed to the presence of heavy atom (bromine), which is known to enhance the ${}^1\Sigma_g^+(v=0) \rightarrow {}^3\Sigma_g^-(v=0)$ transition [9,31]. Based on these values of k_r , the quantum yield of 1O_2 phosphorescence at 765 nm can be estimated. Table 3 indicate that even in CCl_4 the phosphorescence yield of this emission is very low. In other organic solvents and water the lifetime of ${}^1\Sigma_g^+$ is known to be much smaller. For instance, it was estimated that $\tau_\Sigma \approx 30$ ps in ethanol and 6.5 ps in water [38,39]. Therefore, the quantum yield of phosphorescence in these solvents should be by ~4 orders smaller than in CCl_4 , being about 10^{-11} .

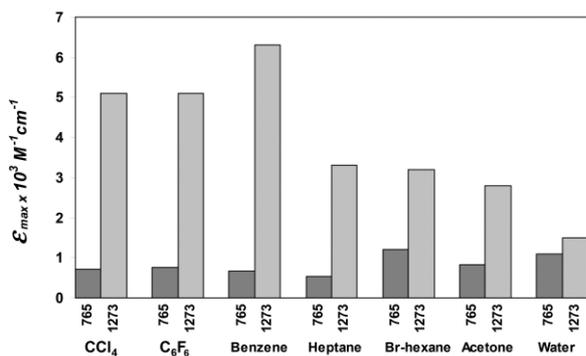


Fig. 5 Molar absorption coefficients corresponding to the main absorption maxima of molecular oxygen in several solvents.

2.5 Absorbance at 1070 nm

The absorption coefficient for the band at 1073 nm corresponding to the transition ${}^1\Delta_g(v=1) \leftarrow {}^3\Sigma_g^-(v=0)$ was reported in Refs. [26,28], in which oxygenation of tetracene and DPIBF solutions were studied under irradiation by the Nd-Yag laser (1064 nm) or by powerful diode lasers (1060 nm). As a result, we arrived to the ratio $A_{1273}/A_{1073} = 60 \pm 15$. Fig. 6 illustrates relative intensities of the major oxygen absorption bands in CCl_4 .

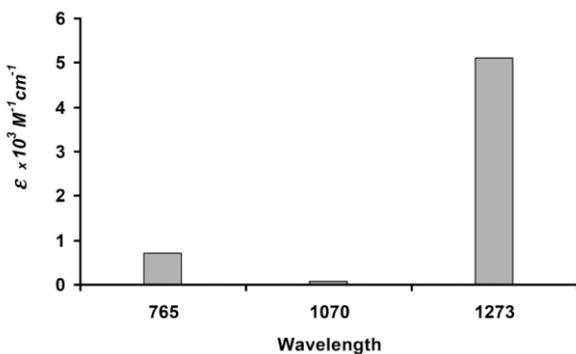


Fig. 6 Relative values of the molar absorption coefficients in the maxima of the absorption spectrum of oxygen dissolved in CCl_4 under normal conditions.

Comparison with Fig. 2 shows that the ratio A_{1073}/A_{1273} obtained in our experiments is 30-40 times smaller than in the absorption spectra of oxygen dimols (O_2)₂ (see

also [9,11,26]). Judging by the ratio A_{1073}/A_{1273} , the concentration of oxygen dimols is very low under normal conditions being less than 3%. However, the ratio A_{1073}/A_{1273} is a mirror image of the intensities of the major bands in the phosphorescence spectrum of monomeric singlet oxygen at 1274 and 1590 nm I_{1590}/I_{1274} (see refs in [26,28]).

3 Conclusions

Thus, application of the methods of laser photochemistry allowed measurement of the absorption coefficients for major absorption bands of monomeric oxygen molecules dissolved in organic solvents and water under ambient conditions. This information has never been reported before our studies. Knowledge of these coefficients is obviously of basic importance for photonics of triplet and singlet oxygen because it provides information on the electronic structure of oxygen molecules and their interaction with environment. Recently, major results of our studies were confirmed by other researchers [29-31]. Although at this state of research numerical values reported by different groups sometimes do not quite coincide (only in acetone identical values were obtained by all authors) the reported values are still of the same order of magnitude. The difference does not exceed the factor of 3. As mentioned in the present paper and previous papers of our group, the values reported by our group also passed through certain evolution due to improvement of measurement techniques and data processing. As mentioned above, to obtain correct numbers one should take into account many parameters, determination of which is not always simple.

Biomedical importance of this research is also apparent. Our experiments demonstrate that direct laser excitation of oxygen molecules is a real natural process, which can be detected using modern laser and spectroscopic techniques. However, the effectiveness of this process is very low, because the absorption coefficients of oxygen have a very small value. The rate of photosensitized singlet oxygen production is 10^3 - 10^4 times greater than the rate of direct oxygen excitation. Therefore, it is difficult to expect that under moderate laser power allowed for photodynamic and laser therapy direct excitation of free oxygen dissolved in cell structures causes appreciable destruction of living tissues. It was shown recently that reasonable rates of cell damage can be observed under laser power of $200 W/cm^2$ that greatly exceeds the excitation power allowed for medical applications (usually $< 200 mW/cm^2$) [40]. However, low-power laser radiation might influence enzyme-bound oxygen molecules whose concentration is much higher. The data are reported that singlet oxygen produces signaling effect, triggers expression of antistress genes and apoptosis and stimulates immune system response [41-44]. In conclusion it is worth noting that according to our results, the lasers with the wavelength 765 nm are probably more appropriate for oxygen excitation in biomedical systems than lasers emitting at 1273 nm.

Indeed, both lasers have rather similar efficiency. However, the dark red light penetrates deeper and causes less heating (see [28] for more details).

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