

First Steps in Photophysics: I. Fluorescence Yield and Radiative Rate Coefficient of 9,10-Bis(phenylethynyl)anthracene in Paraffins

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ABSTRACT: The fluorescence quantum yield of 9,10-bis(phenylethynyl)anthracene is almost unity in every examined solvent. Using different hydrocarbons, one can make a convenient and sufficiently accurate experimental test for determination of the extent of the refractive index correction needed in fluorescence quantum yield determination on a given fluorometer. By comparison of the measurements in *n*-pentane – *cis*-decaline or *n*-hexane – toluene solvent pairs the requirement of the n^2 correction is confirmed for most of the fluorometers; however for one of the examined equipment the necessary correction proved to be slightly lower. By excited state's lifetime measurements the refractive index dependence of the fluorescence rate coefficient was reexamined. At 25 °C for BPEA the

relationship is in agreement with Bakhshiev's prediction: the experimentally determined exponent of n in the rate coefficient deriving equation is around 1.32 using different paraffins as solvents. The negative temperature coefficient of the radiative rate in part originates from the temperature dependence of the refractive index, while also a small intrinsic contribution has been found.

Keywords: transition dipole moment, refractive index,

INTRODUCTION

Starting from measuring a corrected fluorescence spectrum the first and most essential steps in a photophysical study are the determination of the fluorescence quantum yield (Φ_f) and radiative lifetime (τ). Commonly the decay is single exponential, so the radiative rate coefficient (k_f) is simply calculated by:

$$k_f = \Phi_f / \tau \quad (1)$$

The easiest method to determine the fluorescence quantum yield of a molecule is the comparison of its emission intensity with the intensity of a standard of known quantum yield under identical experimental conditions. Probably the standard most often referred to is quinine sulfate (in air saturated 0.1 or 1 n sulphuric acid).¹ One main advantage of this compound is that its fluorescence quantum yield has proved to be practically independent of the excitation wavelength. So the excitation wavelength can be chosen in a favorable way for the molecule under examination. The determination of the quantum yield by this method is based on the comparison of the integrated emission intensity of the corrected spectrum of the sample to that of the standard (*ref*). To avoid inner filter

effects the optical density in the range of overlapping of the absorption and the fluorescence spectrum is typically chosen to be below 0.05. It is a practical approach that the optical densities of the sample and reference standard are matched at the excitation wavelength, than:²

$$\Phi_f = \Phi_f(\text{ref}) \frac{I}{I_{\text{ref}}} \frac{n^2}{n_{\text{ref}}^2}, \quad (2)$$

where n is the refractive index of the solvent at the mean emission frequency if available (otherwise often used the values measured at the D line of Na: 589.3 nm) and I is integrated fluorescence intensity for both the sample and for the reference. Although from geometrical – optical considerations several authors accept as a straightforward fact the necessity of the n^2 correction²⁻⁴ there has been a long discussion on circumstances where the refractive index dependence is smaller than that expected using equation 2.⁵⁻⁹

The radiative rate coefficient k_f (reciprocal of the natural lifetime) depends on the solvent parameters, first of all on refractive index. When the influence of the relaxation processes are negligible the k_f is directly connected to the integral of the corresponding absorption band via the Strickler – Berg equation.^{2,10} The transition dipole moments of the absorption and emission are expected to be similar, especially in apolar solvents, and when the structural relaxation of the excited molecule is small. The value of transition moment might be an invariant descriptor of the process; however the proper calculation of that data from the experimentally measurable quantities remains controversial.

The knowledge of k_f is an essential parameter for derivation the transition dipole moment:^{11,12}

$$|M|_f = \sqrt{\frac{3\varepsilon_0 hc^3}{16\pi^3} \frac{1}{n^3 f^2(n)} \langle v_f^{-3} \rangle k_f}, \quad (3)$$

where the average of v_f^{-3} over the fluorescence spectrum is $\langle v_f^{-3} \rangle = \frac{\int F(v)v^{-3} dv}{\int F(v)dv}$, with

$F(v)$ the emission line shape and the integration is carried out over the entire emission band. Reduction of the physical constants to a number leads to Equation 4, when k_f is given in $[s^{-1}]$, and $\langle v_f^{-3} \rangle$ in $[cm^3]$. (The same final multiplier is given in the paper of Lewis and Maroncelli as in the Equation 7b below.)¹³

$$\frac{|M|_f}{[\text{Debye}]} = 1.78566 \times 10^3 \sqrt{\frac{k_f \langle v_f^{-3} \rangle}{n^3 f^2(n)}} \quad (4)$$

$$k_f = \frac{8\pi h n^3}{c^3} \frac{2\pi^2 f^2(n)}{3\varepsilon_0 h^2} |M|_f^2 \frac{\int F(v)dv}{\int F(v)v^{-3}dv} = \frac{16\pi^3}{3} \frac{n^3 f^2(n)}{\varepsilon_0 h c^3} |M|_f^2 \langle v_f^{-3} \rangle^{-1} \quad (5)$$

In the Equations 4 and 5 $f^2(n)$ function is the local field correction factor and the formula depends on the cavity model. According to the photochemical literature $f(n)$ is defined here in a less common manner as the ratio of the local electric field associated with a single photon, experienced by the fluorescent molecule in the host medium, to the macroscopic electric field associated with a single photon of the same energy in vacuum. Several different models were proposed for the derivation of function $f^2(n)$ in the literature: Förster¹⁴ proposed $f^2(n) = n^{-1}$, Birks¹¹ $f^2(n) = 1$, Chako¹⁵ applied the expression (6a) on the basis of Lorentz local field model, while Bakhshiev¹⁶ suggested the function (6b) which derived applying Böttcher's reaction field theory¹⁷ for an empty cavity. This model was deduced and applied by Toptygin¹⁸ in his detailed paper, which

discusses extensively most of the essential questions to be under further experimental investigation in this study.

$$f^2(n) = (n^2 + 2)^2 / (9n^2) \quad (6a)$$

$$f^2(n) = 9n^2 / (2n^2 + 1)^2 \quad (6b)$$

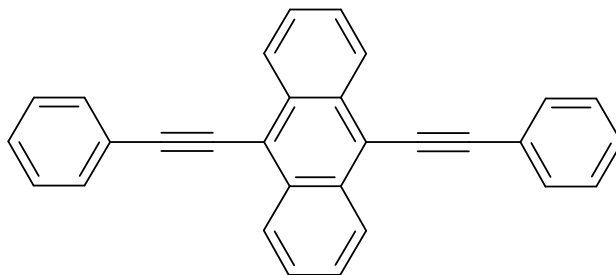
The transition dipole moment can be calculated from the absorption spectrum { $\epsilon(\nu)$ } as follows:¹¹⁻¹³

$$|M|_{\text{abs}} = \sqrt{\frac{3000 \ln 10 \epsilon_0 hc}{2\pi^2 N_A} \frac{1}{n f^2(n)} \int \epsilon(\nu) \frac{d\nu}{\nu}} \quad (7a)$$

$$\frac{|M|_{\text{abs}}}{[\text{Debye}]} = 9.58433 \times 10^{-2} \sqrt{\frac{1}{n f^2(n)} \int \frac{\epsilon(\nu) \nu^{-1} d\nu}{\text{mol}^{-1} \text{cm}^2}} \quad (7b)$$

In photophysical kinetic studies, when the temperature dependence of excited state processes is examined, it is often observed that in contrast to internal conversion or intersystem crossing parameter, the radiative rate has a negative temperature coefficient¹⁹⁻²⁰ (see below). Since the refractive index increases with decreasing temperature this phenomenon is more or less straightforward, however the quantitative understanding of this influence is an issue worthy of further investigation.

CHART 1



9,10-Bis(phenylethynyl)anthracene (BPEA)

The main objective of this experimental study is to show that this commercially available compound is a useful and an easy to handle tool for checking the necessity of the n^2 correction in fluorescence quantum yield determination for a given measuring set-up. Furthermore the influence of the refractive index of the solvent on the fluorescence rate constant of BPEA will be examined by fluorescence lifetime measurements in different type of experiments and compared to the theoretical models stated in eq. 6a and 6b.

EXPERIMENTAL SECTION

The absorption spectra were recorded on a thermoregulated Unicam UV500 spectrophotometer with a resolution typically of 0.5 nm. When necessary, correction was made for density change due to variation in temperature. The corrected fluorescence spectra were obtained on a Jobin-Yvon Fluoromax photon counting equipment (with 0.5 nm resolution), on a FluoroLog-3 (HORIBA Jobin-Yvon), on a Hitachi F-4500 as well as on a quantum corrected Shimadzu RF-5000PC spectrofluorometer (with 1.5 nm resolution). The excitation wavelength was around 420 nm unless otherwise indicated.

In all measurements (except some absorption ones) freeze-pump-thaw degassed samples were used in a sealed 1x1x4 cm Suprasil quartz cells. The calibration of the wavelength scale was made just prior to the measurements with a low pressure mercury arc comparing the measured data with the values of the NIST data base.²¹ Every day both the excitation and emission wavelength scale was checked with the corresponding spectra of the diluted *N*-methyl-2,3-naphthalimide in *n*-hexane having a narrow, well measurable 0-0 absorption and emission bands at 351.3 and 352.4 nm, respectively.²² The room

temperature (25 °C) fluorescence quantum yields were determined relative to that of quinine sulfate ($\Phi_f = 0.546$ in $1n$ H₂SO₄).²³ The excitation wavelengths for these quantum yield determinations were around 310 nm, where both BPEA and quinine sulphate have a reasonable absorption coefficient (unless otherwise stated in the text). The optical density of the samples were around 0.2-0.3 at the absorption maximum to minimize the error coming from the sample and standard preparation, and the inner filter effect was corrected by comparison of the apparent fluorescence spectra and those were measured at infinite dilution (i.e. at ten times smaller concentration). The difference was around 4.5 % and almost exactly the same for different paraffins when the absorbance at the maximum (and around the excitation wavelength) was equal.

The fluorescence decay times were measured by an Edinburgh Instruments FLS 920 time-resolved spectrofluorimeter, using an EPL-375 semiconductor laser for excitation and a Hamamatsu R3809U-50 MCP-PMT for detection.²⁴ Few measurements were done by a picosecond single photon counting system (where the excitation wavelength was 296 nm)¹⁹ to inspect the reliability the time resolved data. The timescale of the two setup was cross calibrated by the diluted solution of anthracene in cyclohexane;²⁵⁻²⁶ and that lifetime in question proved to be 5.19 ± 0.04 ns.

9,10-Bis(phenylethynyl)anthracene (BPEA), 9-methylanthracene (9MeA) and 9,10-diphenylanthracene (DPhA) were received from Aldrich and were purified on a silica column using *n*-hexane – dichloromethane eluent and recrystallized from toluene. The absorption spectra of BPEA samples in *n*-pentane or *n*-hexane under vacuum show small changes over a month; consequently fresh samples were prepared for every experimental run. The *n*-hexane (*hex*) and acetonitrile (MeCN) were of Merck Uvasol quality. Other

solvents as *n*-pentane (*pent*), *n*-heptane (*hept*), *n*-hexadecane (HD), cyclopentane (CP), cyclohexane (CH), *cis*-decalin (CD), toluene (*tol*), diethyl ether (DEE), tetrahydrofuran (THF), butyronitrile (PrCN) were received from Merck, and purified on a combined activated charcoal – alumina column before being used. The *n*-butane (*but*, Fluka puriss) and propane (Messer, P 2.5) was frozen directly into the quartz Fischer-cuvette (containing the necessary amount of dried BPEA) under vacuum. At 25 °C the ~2.5 bar (*n*-butane) and ~9 bar (propane) pressure in the cell caused no difficulties.

For triplet yield determination the energy transfer method was used where excited anthracene was the energy donor, and triplet yield standard,²⁷ and BPEA the acceptor, similar to our previous experiments on 9-ciano-10-(dimethylamino)anthracene.²⁸ This set-up is adequate when the expected triplet energy as well as the triplet yield is small. Equal molar absorbance at 355 nm (excitation wavelength of a frequency-tripled Continuum Surelight Nd:YAG laser) was established in a sample containing only BPEA and in another cell where both BPEA and anthracene was solved at approximately equal absorbance ratio. The triplet-triplet absorption spectrum of BPEA was determined in advance to choose the appropriate wavelengths, i.e. 480 nm and 490 nm for paraffins and acetonitrile, respectively (see Figure S1). Here, at the foot of the ground state absorption the transient absorbance signal ratio for the two samples can be easily determined. The pseudo first order energy transfer occurs typically on the 3-4 μ s range while the anthracene itself decays on a much slower timescale (400-600 μ s). The triplet yields of anthracene donor were taken to be 0.71 in *n*-hexane (and HD),^{27,28} while 0.66 in acetonitrile²⁷ respectively.

The temperature dependence of refractive index was determined with a thermoregulated Abbe refractometer (Modell G, Carl Zeiss Jena) at 589 nm of Na D-line: $n(T) = 1.38607 - 5.46592 \times 10^{-4} \times T$ (T in °C; from -31 to 62 °C) and $n(T) = 1.44264 - 4.09985 \times 10^{-4} \times T$ (T in °C; between 20-95 °C) for *n*-hexane and HD, respectively. The literature relationship for *n*-hexane differs moderately: $n(T) = 1.39174 - 7.04556 \times 10^{-4} \times T$ (T in °C; at 546 nm between 20-100 °C)²⁹.

RESULTS

Absorption and fluorescence properties.

For this study, a molecule is required with following characteristics: a high solvent independent fluorescent yield, ideally near unity; photochemically stable, and with photophysics as simple as possible. For minimization of solute solvent interaction a centrosymmetric aromatic hydrocarbon was chosen (BPEA). The solvents were selected mainly apolar solvents as aliphatic and cycloaliphatic hydrocarbons. The aim behind it was that the influence of the polarizability of the solvent (i.e. refractive index) is not masked by “dipole induced” effects. The absorption and emission spectra of BPEA show approximate mirror symmetry with small Stokes-shift both in *n*-pentane and *cis*-decalin (CD, Figure 1), representing of hydrocarbon solvents having small and large refractive indices, respectively. A moderate red shift of both the absorption and emission spectra with increasing refractive index of the solvent was observed, while the magnitude of the Stokes-shift did not show a significant change. The two absorption spectra of BPEA have four crossing points at optimal dilutions in this solvent couple. The shape of the absorption and even more the emission spectra show the typical feature of rigid aromatic

hydrocarbons: well observable vibronic structure, where the 0-0 vibronic bands are the most intense.

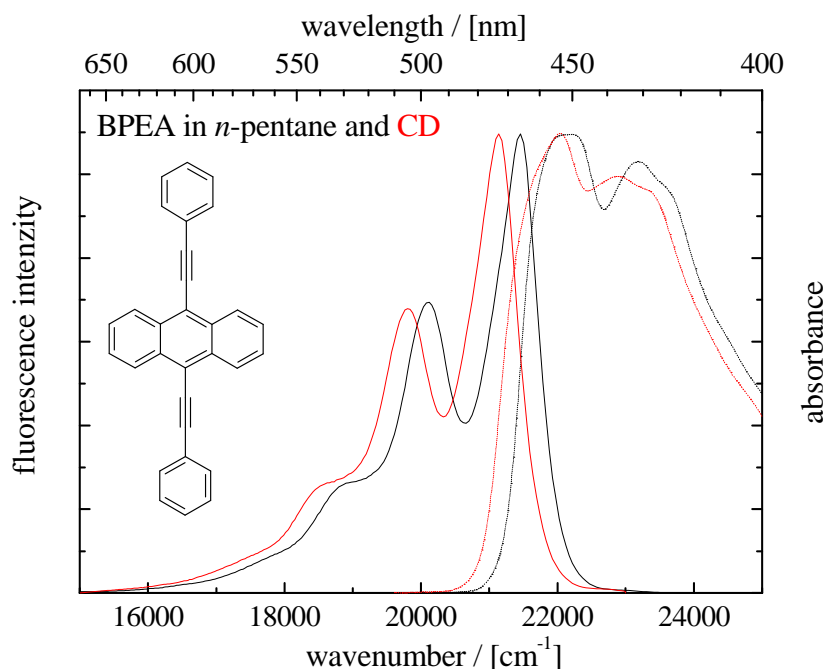


Figure 1. The normalized absorption (dotted line) and fluorescence spectra (full line) of BPEA in *n*-pentane (black) and *cis*-decalin (CD, red).

The fluorescence quantum yields of BPEA (Φ_f) were determined in ten aprotic solvents with different polarity measured relative to quinine sulphate in 1 *n* H₂SO₄ (see Table 1). All the yields are near unity (after n^2 correction, see later). Note that obtained deviations from unity are within the measurement uncertainties. It follows that the competing deactivation channels as internal conversion (IC), intersystem crossing (ISC) or decomposition have much lower rate than that of fluorescence. As a consequence, even relatively big changes in the reaction rates of these channels have only a small effect on Φ_f and τ . It is to be expected that the fluorescence yield will not change in apolar solvents

(aliphatic hydrocarbons). Here the differences in solvent-relaxation of BPEA in the ground and in the excited are small compared to polar solvents.

Table 1. Fluorescence yields of 9,10-Bis(phenylethynyl)anthracene (BPEA) and 9,10-diphenylanthracene (DPhA) measured by Shimadzu RF-5000 fluorometer^a and FluoroMax-2^b at 25 °C in different solvents.

BPEA ^a (exc.: 309-312 nm)	<i>n</i>	$\Phi_f(n^2 \text{ corrected})$	$n^2 \text{ corr.}$
<i>n</i> -pentane (<i>pent</i>)	1.3575	1.01	1.017
<i>n</i> -hexane (<i>hex</i>)	1.3720	0.97, 1.03 1.01*	1.039
<i>n</i> -hexadecane (HD)	1.4340	0.96	1.135
cyclopentane (CP)	1.4065	1.05*	1.092
<i>cis</i> -decalin (CD)	1.4810	0.98	1.210
toluene (<i>tol</i>)	1.4960	1.00, 1.02*	1.235
diethyl ether (DEE)	1.3530	1.04*	1.010
tetrahydrofuran (THF)	1.4050	1.04	1.089
butyronitrile (PrCN)	1.3725	0.97*	1.057
acetonitrile (MeCN)	1.3442	0.99, 0.98	0.997
DPhA ^b (exc.: 367 nm)			
<i>n</i> -pentane (<i>pent</i>)	1.3575	0.84	1.017
<i>n</i> -hexane (<i>hex</i>)	1.3720	0.84	1.039
<i>n</i> -hexadecane (HD)	1.4340	0.86	1.135
<i>cis</i> -decalin (CD)	1.4810	0.85	1.210
toluene (<i>tol</i>)	1.4960	0.86	1.235
<i>In sulphuric acid / water</i>	1.3462		1.000

)* excited on absorption tail

Change of the fluorescence spectrum with refractive index of the solvent.

The subsequent increase of the refractive index of the paraffin solvents resulted in a regular bathochromic shift of the fluorescence spectrum of BPEA as demonstrated on

Figure 2. The $\langle v_f^{-3} \rangle^{-1}$ value (cf. eq. 3) is a power function of the refractive index *n* of the

solvent – see inlet of Figure 2. The relation was found to be: $\ln \langle v_f^{-3} / \text{cm}^3 \rangle^{-1} =$

$$(29.901 \pm 0.007) - (0.521 \pm 0.020) \times \ln(n)$$

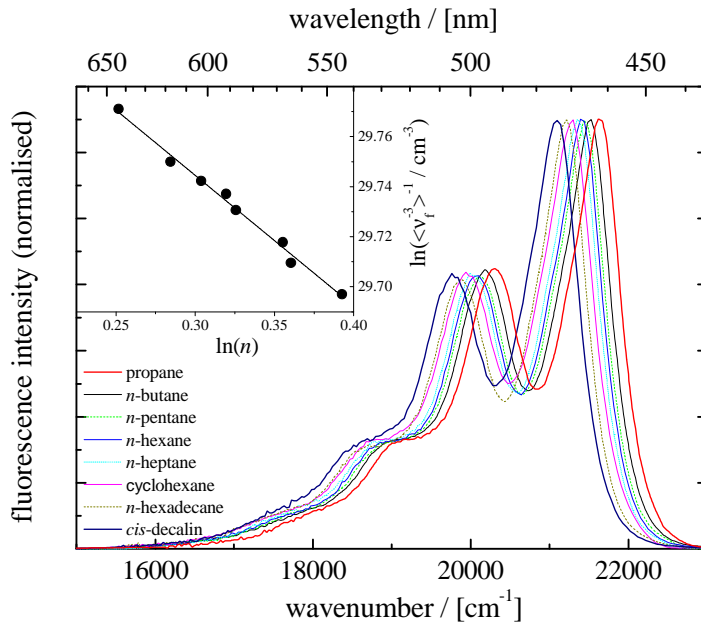


Figure 2. The fluorescence spectra of BPEA in different paraffin's. In the inset the $\langle v_f^{-3} \rangle^{-1}$ values of these spectra are shown as a function of refractive index.

To further investigate the influences of the refractive index on the fluorescence spectrum of BPEA, it was measured as a function of temperature in *n*-hexane and *n*-hexadecane.

The $\ln \langle v_f^{-3} \rangle^{-1}$ values of the spectra again showed a linear dependence on $\ln(n)$, with \ln

$\langle v_f^{-3} / \text{cm}^3 \rangle^{-1} = (29.940 \pm 0.003) - (0.653 \pm 0.009) \times \ln(n)$ for *n*-hexane (Figure 3) and \ln

$\langle v_f^{-3} / \text{cm}^3 \rangle^{-1} = (29.928 \pm 0.005) - (0.618 \pm 0.015) \times \ln(n)$ in HD (Figure 2S). The

fluorescence yield (near unity at room temperature) does not depend significantly on the solvent temperature, except at high temperature in HD, where between 120 and 200 °C an approximately 7 ± 4 % decrease was observed.

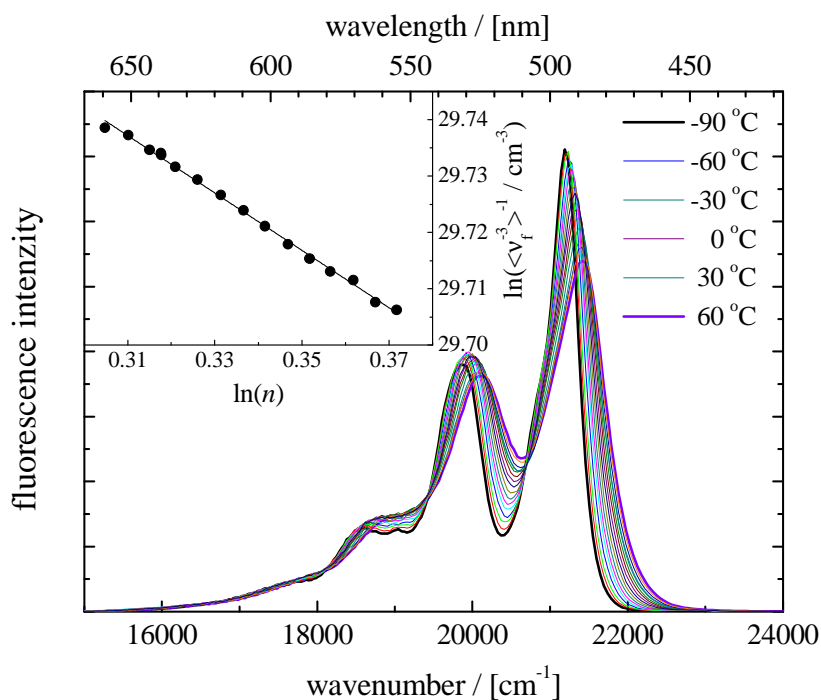


Figure 3. The fluorescence spectra of BPEA in *n*-hexane as the function of temperature. In the inset the $\ln\langle\nu_f^{-3}\rangle^{-1}$ values of these spectra is shown as the function logarithm of the temperature dependent refractive index, $\ln(n(T))$.

It was intended to widen the extent of this study to other results found in the literature published in the field. Hirayama and Phillips³⁰ examined the refractive index dependence of the radiative rate by measurements of 9-methylanthracene (9MeA) in cyclohexane at 25 °C and in the gas phase at 171 °C. The emission spectra of the 9MeA were remeasured (see Figure 4), using the same sample preparation and temperature choices they stated, to derive the $\langle\nu_f^{-3}\rangle^{-1}$ data. The value of $\langle\nu_f^{-3}\rangle^{-1}$ for 9MeA in gas phase at 171 °C proved to be 1.107 bigger than the corresponding value in cyclohexane at 25 °C.

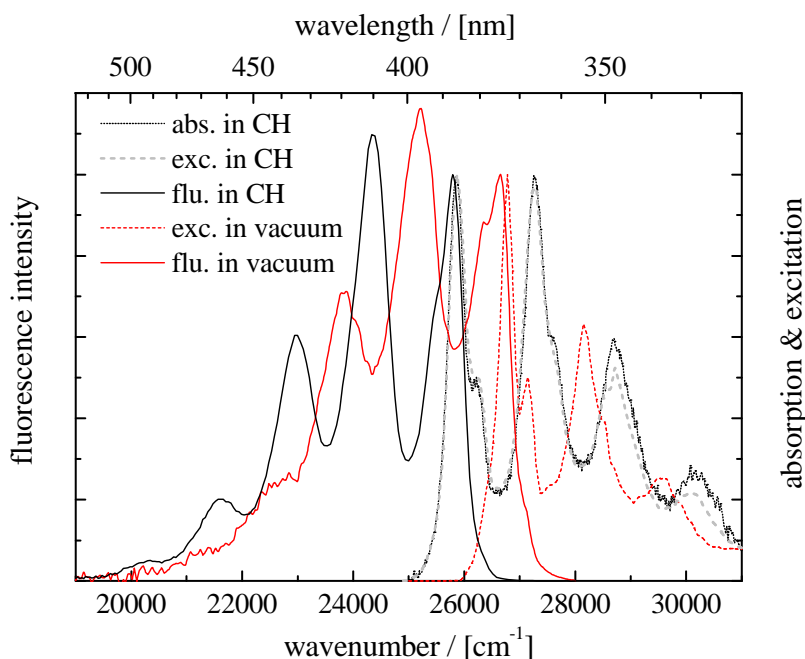


Figure 4. Absorption, fluorescence and excitation spectra of 9-methylanthracene in cyclohexane (CH) at 25 °C and in gas phase (vacuum) at 171 °C.

The necessity of n^2 correction for fluorescence quantum yield determination.

It was found that the value of the fluorescence quantum yield of BPEA is close to 1.0 independent of the solvent used (see Table 1). This allows us to establish a simple experiment to check on the necessity of the n^2 correction for the fluorescence quantum yield determination of a given experimental set-up. For this, the fluorescence yield of BPEA was compared in two hydrocarbon solvents under the same experimental conditions. The two solvents, *n*-pentane ($n = 1.3575$ at 25°C) and *cis*-decalin ($n = 1.4810$ at 25°C), were chosen to have a large enough variation in the refractive index relative to each other. The *cis*-decalin / *n*-pentane ratio of n^2 ratio calculates to 1.190. The method to compare the ratio of the fluorescence yields seems more sensitive since some of the experimental error sources would be minimized or excluded. An example is the effect of the inner filter correction. Although this correction (approx. 4.5 %) was carried out for

every measurement, as is described in the experimental section, the difference in the required corrections in the case of these solvent pairs was very small. The comparison of one single compound in an (apolar) solvent pair has the further advantage that the excitation wavelength can be chosen to be in a more optimal range compared with an experiment use quinine sulphate as standard. Moreover the four crossing points of the two absorption curves leads to possibility of four parallel measurements where the slope directions are changing. Averaging the fluorescence yield ratios measured using neighboring excitation wavelengths will lead surely to more accurate final data. The 453-458 nm range is somehow the best for this type of measurement in case of BPEA (Figure 1): (i) here the absorption spectra run approx. parallel, (ii) the red edge of the spectrum is disturbed less by the absorption of the possible decomposition products. (It was observed that sometimes (*n*-pentane, *n*-hexane) the spectra change little over a timescale of a month, even in absence of irradiation, probably due to a slow decomposition of BPEA.)

From data given in Table 2 one can conclude that for the FluoroLog-3 and Shimadzu RF-5000 fluorometers with similar measurement set-up circumstances (narrow slits) the n^2 correction is needed for good accuracy, however in case of Hitachi F-4500 the necessary correction was significantly smaller, although the samples used and the experimental parameters were the same. The influence of the geometry of the light path on the fluorescence intensity ratio of a BPEA in *n*-hexane, and on a BPEA in toluene sample pair was also examined. The decrease of the surface area of the excitation spot (actually it had the form of a line) does not at all influence the correction factor. Moreover, as Table 2 indicates, the decrease of the diameter of the detection light pass has only a very small effect on the fluorescence intensity ratio.

Table 2. Comparison of fluorescence yield of BPEA in *n*-pentane / CD and *n*-hexane / toluene solvent pairs (3 parallels) at various excitation wavelength using three different fluorometers.

BPEA in <i>n</i> -pentane and CD				
Excitation	FluoroLog-3	Shimadzu RF-5000	Hitachi F-4500	$n^2(\text{CD})/n^2(\text{pent})$
434.8 nm	1.206	1.186	1.115	
444.1 nm	1.185	1.240 / 1.193	1.135	
451.8 nm	1.226	1.194		
456.0 nm	1.198	1.190		
Average	1.20±0.03	1.195±0.02	1.12±0.02	1.190
BPEA in <i>n</i> -hexane and toluene				
	Shimadzu RF-5000	Shimadzu RF-5000 (restricted detection path)	Hitachi F-4500	$n^2(\text{tol})/n^2(\text{hex})$
434.3			1.14	
446.2	1.19	1.15	1.14	
454.0	1.17	1.19	1.15	
average	1.18±0.01	1.17±0.2	1.14±0.01	1.188

Inner filter correction: toluene 1.044, *n*-hexane 1.041

FluoroLog-3 (HORIBA Jobin-Yvon)

Again to compare values found in the literature with results obtained by using the experimental set-ups of this study it was needed to choose another sample molecule. Here a series of quantum yield determinations was done with 9,10-diphenylanthracene (DPhA) in different hydrocarbons on Jobin-Yvon FluoroMax-2 equipment at 25 °C. As it can be seen from Table 1 the Φ_f was found after n^2 correction to be around 0.85 in every solvent, independently of their refractive index (the overall error of the measurement is bigger than the scatter, and can be approximated to be ± 0.04 at 2σ level). Hamai and Hirayama in their very thorough work³¹ found a $\Phi_f = 0.90$ with 4% uncertainty.

The triplet yield of BPEA.

The near unity fluorescence quantum yield of BPEA implies a very low yield of triplet formation. However, other similar aromatic hydrocarbons such as perylene^{19,20} display a small but measurable triplet formation yield (i.e. $\Phi_{ISC} = 0.03$ in almost all solvents). Thus, it is possible that for BPEA, even a small triplet yield could affect the current interpretation of results.

Three room temperature data were determined as described in the Experimental Section in three different solvents, the triplet yields proved to be 0.011 ± 0.006 , 0.014 ± 0.006 and 0.009 ± 0.006 in *n*-hexane, *n*-hexadecane, and acetonitrile, respectively. The temperature dependence of the triplet yield was determined comparing the transient absorption signal ratio measured at the transient absorption maximum at the given temperature (from -21 to 45 C° in *n*-hexane and from 22 to 205 C° in *n*-hexadecane) and at the reference room temperature, where the quantum yield is known. The triplet yield practically does not change over the examined temperature range examined. The observed small increase of the value determined was within the scatter and the uncertainty of the determinations (i.e. 10% increase of Φ_{ISC} at 135 C° in HD compared to value at room temperature). Although the transient absorbance at 205 °C in HD was almost triple to that at room temperature, the transient absorbance did not return to its initial value when the sample was cooled back to the starting condition at room temperature. This indicates that at high temperature a small amount of photoproduct is formed producing the excess time resolved absorbance.

Refractive index dependence of the fluorescence lifetime.

The decay of the time resolved fluorescence intensity of BPEA was found to be strictly single exponential in every examined solvent. As an example two typical results are displayed in Figure 5.

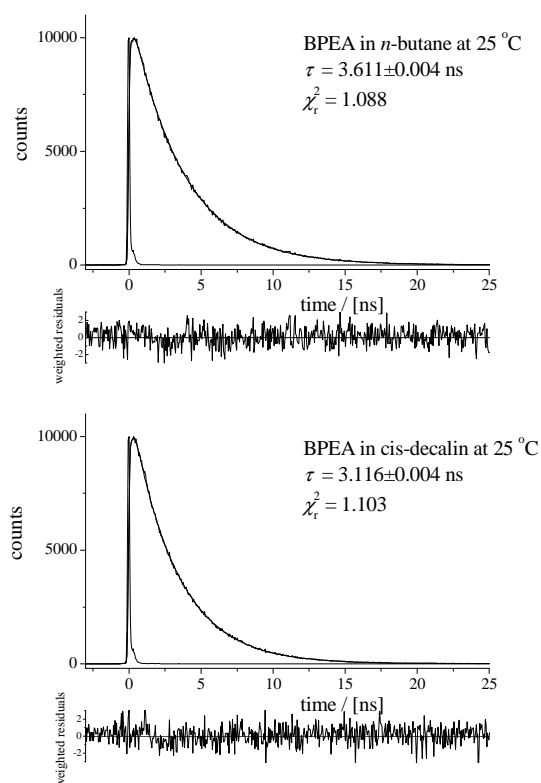


Figure 5. Time resolved fluorescence intensity decay of BPEA in *n*-butane and *cis*-decalin at 25 °C (excitation wavelength 378 nm, detection: 510±5 nm).

The effect of refractive index on the lifetime proved to be more than two magnitudes greater than the uncertainties of the measurements (see Table S1). The reproducibility of the lifetime data was double-checked by rerunning some of the measurements on a different single photon counting set-up. The results matched each other within the experimental uncertainties (Figure 6).

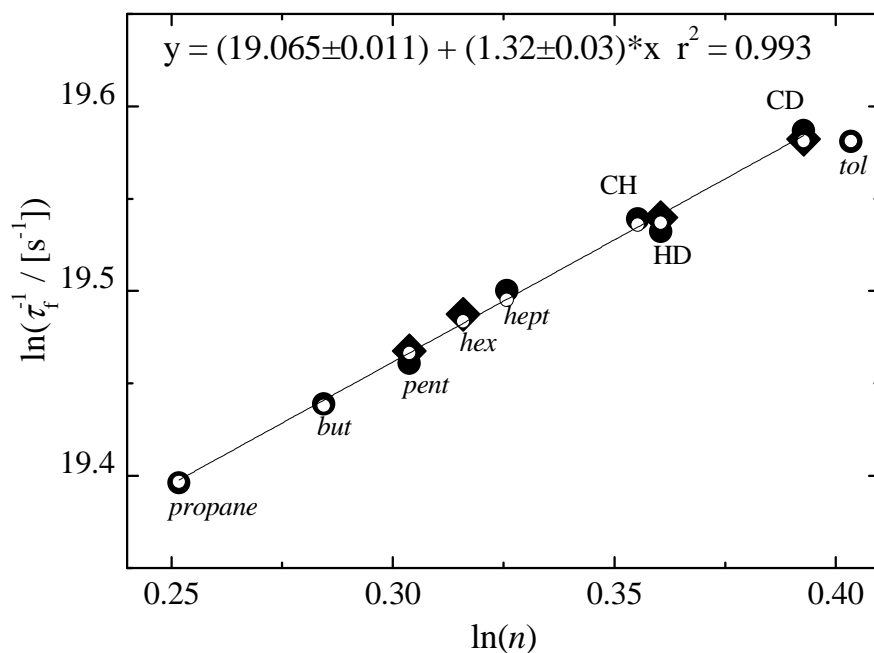


Figure 6. The reciprocal lifetime of singlet excited BPEA determined at 25 °C as a function of refractive index of the paraffin solvents (and toluene) on ln-ln scale. The full circles and diamonds are measured using a diode and a dye laser pumped set-up respectively, while the small empty circles indicate the calculated values using the individual $\langle v_f^{-3} \rangle$ data and a solvent independent $\Phi_f = 0.99$ approximation, see below).

From the linear dependence between $\ln(\tau^{-1})$ and $\ln(n)$ displayed in Figure 6 follows an important conclusion: the reciprocal lifetime (and hence the fluorescence rate coefficient k_f of BPEA in apolar hydrocarbons is found to be a power function of the refractive index, i.e. $k_f \sim n^x$ with $x = 1.32 \pm 0.03$. It should be noted the result cannot be generalized for other molecules (the refractive index dependence of $\langle v_f^{-3} \rangle$ -s are different) and will probably be found only in the case of one single dominant deexcitation channel (i.e.

fluorescence), over all other channels, as intersystem crossing, internal conversion and so on. This was one main reason BPEA was chosen for this study.

The temperature dependence of the fluorescence lifetime of BPEA was measured in *n*-hexane and *n*-hexadecane as well. From an Arrhenius plot approximately -0.49 and -0.69 kJ mol⁻¹ negative activation energies can be derived in *hex* and HD, respectively. (The expression of activation energy is used here as an empirical measure of the temperature dependence of the corresponding rate parameter, as it is usually done in the literature of chemical kinetics.³² In simple reactions the activation energy can be associated with a potential barrier, especially if the barrier is high. In complex processes, such as ours, however, there is no single barrier and the Arrhenius or empirical activation energy can be negative. Yet, this quantity is often used for comparison and interpretation of reactions in related systems.³³)

In *n*-hexane (see the black dots in Figure 7) the dependence of τ^{-1} on *n* can be fitted by the following function (neglecting the points over room temperature): $\ln(\tau^{-1}/s^{-1}) = (18.865 \pm 0.008) + (2.01 \pm 0.03) \times \ln(n)$. In HD the picture is more complicated as can be seen in Figure 7: at higher temperature (at lower *n*) the increase of lifetime is regularly smaller than that is expected from the linear relationship. Nevertheless from the low temperature points the $\ln(\tau^{-1}/s^{-1}) = (18.763 \pm 0.018) + (2.16 \pm 0.05) \times \ln(n)$ relationship can be calculated.

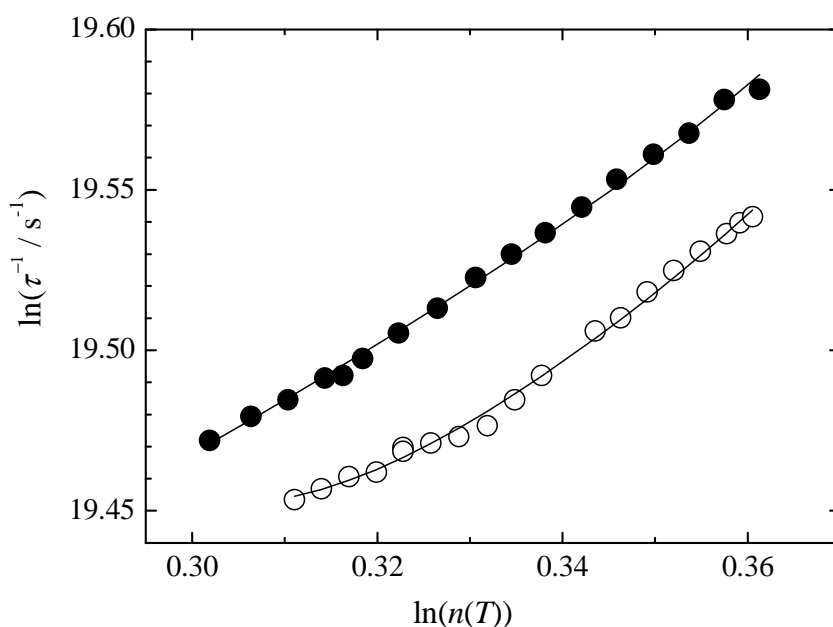


Figure 7. The reciprocal lifetime (τ^{-1}) of singlet BPEA in *n*-hexane (full round) and in HD (empty circle) as the function of refractive index altered via change of temperature (the derivation of calculated curves see in the text).

DISCUSSION

The BPEA molecule proved to be an almost ideal compound for this type of study: it has near unity fluorescence yield in all solvents examined (Table 1). However, the experimentally determined triplet formation yields around 0.01 forced us to decrease the room temperature Φ_{f} value to 0.99. This effect causes some inconvenience, but one can expect that even a considerable change in the almost negligible rate of intersystem crossing will not influence much the further considerations.

While the absorption spectrum of BPEA changes with the change of the refractive index of the solvent, at optimal dilution four “crossing points” can be found for the

spectrum with identical absorbance in all different solvents used (see Figure 1). Choosing them as excitation wavelengths the crossing points are ideal choices for relative fluorescence yield measurements considering that directions of the corresponding slopes of the absorption spectra are changing in couples. (Averaging the fluorescence yield ratios will lead more accurate final data, decreasing the error comes from the occurring bias in wavelength calibration of the excitation monochromator and the UV-VIS spectrometer used for sample preparation.) This also makes the BPEA a good probe molecule to investigate the magnitude of the refractive index correction in the quantum yield determination at a given measuring set-up.

The ratio of n^2 factor for *n*-pentane – CD or *n*-hexane – toluene solvent couples is around 1.19, while using *n*-pentane and toluene the value is even higher: 1.214. Although toluene often shows, (as for example in solvatochromic experiments) dissimilar properties compared with what would be expected from its dielectric parameters, it proved to be a proper choice here for a solvent characterized by a large refractive index. As explained in the results section, by examining the ratio of the normalized integrated fluorescence spectra of BPEA in these solvent couples the measurement errors could be minimized. They were found to be more than magnitude smaller than the effect expected from the refractive index change. For example the error resulting from the inner filter effect correction showed to have almost no influence on the final data. The choose of the “crossing points” as excitation wavelengths further increases the reliability of the value of fluorescence yield ratio. In the same way, the restriction of the excitation and emission light pass does not change significantly the fluorescence yield ratio. From the results shown in Table 2, one can conclude that the fluorometers checked in this study require

the n^2 correction; the only exception was the Hitachi F-4500 for which the necessary refractive index correction proved to be slightly smaller.

The fluorescence quantum yield of 9,10-diphenylanthracene in different hydrocarbons show similar character: after the n^2 correction Φ_f becomes independent of the solvent properties with a final 0.85 ± 0.04 value. The agreement with the value of 0.90 ± 0.04 published by Hamai and Hirayama³³ is acceptable.

The solvent independent 0.99 fluorescence yield of BPEA has given us a reliable tool for examination of the refractive index dependence of the fluorescence rate coefficient k_f . The 21 % increase of the lifetime of BPEA from CD to propane has ± 0.5 % statistical uncertainty (2σ) and the influence of other errors was probably also small, considering that the data were measured under very similar circumstances. The spectral shift data are also easily measurable; consequently the 1.32 ± 0.03 slope parameter of Figure 6 is well established. Eq. 5 with Bakhshiev¹⁶ cavity field correction results practically the same datum while other models lead to quite different numbers (see Table 3).

Table 3. The fluorescence rate coefficient dependence on the refractive index: $\ln(k_f/s^{-1}) = \text{intercept} + \text{slope} \times \ln(n)$.

Slope	Chako (eq6a)	Bakhshiev (eq 6b)	experim.	$\langle V_f^{-3} \rangle^{-1}$	$f^2(n)$ (eq 6a)	$f^2(n)$ (eq 6b)
BPEA n	~ 2.4	1.31	1.32	-0.52	> -0.12 ^c	-1.17
BPEA $n(T)$ <i>hex</i>	~ 2.3	1.17	2.01	-0.65	> -0.07 ^c	-1.18
BPEA $n(T)$ HD	~ 2.3	1.19	2.16	-0.62	> -0.06 ^c	-1.19
9MeA $n(T)$ ^a	2.38	1.76	1.90 ± 0.10 ^a	-0.29	-0.33	-0.95
DCNA $n(p)$ ^b	~ 2.6	1.20	1.39	-0.54	< 0.13 ^c	-1.26
DMeA $n(p)$ ^b	~ 2.7	1.31	1.69	-0.43	< 0.13 ^c	-1.26
DMeOA $n(p)$ ^b	~ 2.5	1.10	0.93	-0.64	< 0.13 ^c	-1.26
DPhA $n(p)$ ^b	~ 2.5	1.15	1.63	-0.59	< 0.13 ^c	-1.26

)^a Hirayama and Phillips³⁰ published 1.94,)^b Hirayama³³, 9,10-dicyanoanthracene (DCNA), 9,10-dimethylantracene (DMeA), 9,10-dimethoxyanthracene (DMeOA), 9,10-diphenylantracene (DPhA),)^c small changing value

The small empty circles in Figure 6 represents the calculated lifetime values (Eq. 5) using for every solvent our spectral datum of BPEA and a value of $|M|_f = 7.60 \pm 0.02$ Debye for the transition dipole moment.

Calculating the transient dipole moment from the absorption spectrum in the different solvents shows that there is almost no influence of the refractive index: the $n \cdot f^2(n)$ factor at the submultiple of Eq. 7a is 1.0244 and 1.0047 for *n*-hexane and toluene respectively, using them $|M|_{\text{abs}} = 7.11$ and 7.02 Debye. In the Strickler–Berg approximation¹⁰ the equality of $|M|_f$ and $|M|_{\text{abs}}$ is assumed. If this assumption fits, depends on the structural change of the molecule between the ground and excited state. For BPEA the ethyne bonds become longer, while the aryl to ethyne bonds turn out to be shorter in the excited state. This structural change may result in the observed variation of transition dipole moments.

Toptygin¹⁸ proposed a modified field correction function $f^2(n)$ for an ellipsoidal cavity

$$f^2(n) = n^2 / \{(1 - L_\mu) n^2 + L_\mu\}^2, \quad (6c)$$

where the L_μ parameter can be calculated from the length of the ellipsoid semiaxes.¹⁸ For BPEA the cavity isn't spherical at all and the transition moment is parallel to the longest dimension of the molecule: $L_\mu = L_x \sim 0.056$. Using eq 6c the slope parameter for the cavity field correction $f^2(n)$ would be -1.89 instead of -1.17 shown in Table 3. However, from the present experiments one may conclude, that this correction for BPEA in

paraffines is not required. (This outcome for apolar solvents in common was predicted by Toptygin.¹⁸)

The fluorescence lifetime of BPEA in toluene (3.134 ns) and acetonitrile (3.666 ns) seems to be 1.4 % and 3 % bigger than what is calculated from the linear fit in Figure 6. Here one should note that in case of the aromatic hydrocarbon the larger spectral shift causes the effect (see the perfect position of the corresponding calculated empty circles on the figure). In the case of MeCN neither the ISC nor the IC channel were found to be responsible for the outcome: the experimentally determined ISC yield is much lower than what could compensate the difference, while the IC is effective at much higher temperature (see below). The small (2 %) increase of transition dipole moment of the fluorescence is understandable in the following way: the increasing polarity of the solvent affects slightly the geometry of the excited state as well as the wave-functions, resulting in a slight change of both the values of the permanent dipole moment and $|M|_f$.

The temperature dependence of the radiative rate coefficient used to be associated with the change in refractive index. However, here the power dependence is bigger (around 2.1) than that was observed in the room temperature measurements (1.32), and that is expected from the theory (Eq. 5 using 6b, see Table 3). Moreover, the assumption of the influence of a temperature dependent ISC or IC reaction with small positive activation energy even would increase more the necessary power parameter in the $k_f \sim n^x$ relation. Furthermore the IC process appears only at higher temperature as can be deduced from the measurements made in HD and shown in Figure 7. The measurements shown on Figure 7 were fitted by a complex model: k_f was calculated by Eq. 5 and 6b using the experimentally determined spectral correction function and the value of $|M|_f$

deduced from the 25 °C measurements. Then the activation energy parameter was optimized. For k_{ISC} was put in the room temperature value, assuming that the corresponding rate is independent from temperature. Finally for temperature dependence of internal conversion an Arrhenius type expression was fitted in HD and used as fixed value in *hex*. The k_{IC} can be described as follows: $\lg k_{IC} = (11.4 \pm 0.6) - (42 \pm 7 \text{ kJ mol}^{-1}) / (\ln 10 \times RT)$. For the activation energy of $|M|_f^2$ we derived -0.16 ± 0.01 and $-0.15 \pm 0.02 \text{ kJ mol}^{-1}$ (approx. 13 cm^{-1}) values in *n*-hexane and HD, respectively. A small negative temperature coefficient for $|M|_f^2$ should be expected. First of all, the overlap integral of the first excited state and ground state must be smaller for the transitions originate from a low frequency vibronically excited state (which are populated more at higher temperature) when the displacement of the PE curves is very small. Furthermore, with increase of temperature even the volume of the molecule will slightly increase, resulting smaller overlap integral as well. The $-0.16 \text{ kJ mol}^{-1}$ activation energy of $|M|_f^2$ is responsible for the additional effect of temperature on the radiative rate coefficient.

Hirayama and Phillips³⁰ have reported a study comparing the fluorescence properties of 9MeA in cyclohexane at room temperature (25 °C, $n = 1.4262$) and in the gas phase at 171 °C ($n = 1.000$). The large change in the refractive index between the two media made the comparison of radiative rates of the two samples significant. (The fluorescence lifetime was published to be $\tau = 6.3 \text{ ns}$ and 4.6 ns respectively, while $\Phi_f = 0.26$ and 0.36 in vacuum and in CH. This led to a ratio of radiative rates is 1.90).³⁰ From the similar shape of absorption spectra under both conditions the authors assumed that the influence of the spectral change on k_f must be small; and so a relation of the radiative rate coefficient to the refractive index of $k_f \sim n^{1.94}$ was deduced. They concluded that the n^2

multiplicator is an acceptable approximation in case of rigid apolar molecules. From our measurements the change of fluorescence spectra results an -0.29 exponent of n {i.e. $\Delta \ln(\langle \nu_f^{-3} \rangle^{-1}) / \Delta \ln(n)$ }, while the effective field correction of Eq. 6b lead to $f^2(n) \sim n^{-0.95}$ and as an outcome $k_f \sim n^{1.76}$. This result is not strongly differing from the value published by Hirayama and Phillips ($k_f \sim n^{1.94}$) considering that it resulted from a small data set (one solvent and one gas phase measurement). One may also recognize that the difference can be explained by a small negative temperature coefficient of the transition dipole, too.

Lampert et al.³⁴ reported a study on the temperature dependence of the fluorescence lifetimes of three BPEA derivatives. With the approximation that fluorescence yield is independent from the experimental conditions (i.e. temperature) they conclude that $k_f \sim n^2$ in xylene, which relation is just nearby of the experimental observation shown here (Table 3). Similarly, they find a semiquantitative $k_f \sim n^2$ relation using DPhA as probe molecule in different aliphatic and aromatic hydrocarbon solvents at 25 °C (after the correction of the experimentally determined Φ_f values).³⁴ However, the interpretation of this observation is a bit problematic considering the large uncertainties of the data. Similarly $k_f \sim n^2$ relation was found³⁶ for the emission of green fluorescent protein, while it was demonstrated that the radiative rate coefficient does not depend on viscosity at al.³⁷ For the interpretation of the result of fluorescence lifetime imaging microscopy the influence of environment on the radiative rate^{18,34-38} has come to a significant scientific issue again.

In another study Hirayama et al.³⁵ examined how the lifetime of five singlet excited anthracene derivatives depend on the refractive index of methylcyclohexane varying with change of applied hydrostatic pressure. Among others they examined the

properties of 9,10-dicyanoanthracene (DCNA: $\Phi_f = 0.9^{28}$), 9,10-dimethylantracene (DMeA: $\Phi_f = 0.93^{28}$), 9,10-dimethoxyanthracene (DMeOA: $\Phi_f = 0.87^{28}$), 9,10-diphenylantracene (DPhA: $\Phi_f = 0.90^{31}$ or 0.85 in this work) derivatives. As it can be noted from Table 3 for DCNA and DMeOA the deviation of calculated values from the experimental data is acceptable. For the other two compounds probably the non-radiative processes have enough strong pressure dependence to cause the observable discrepancy.

CONCLUSION

The BPEA proved to be an easy to use commercial compound to check the requirement of n^2 correction in fluorescence quantum yield determination. For that purpose the *n*-pentane / toluene couple is a proper choice as hydrocarbon solvents. The combined fluorescence and transient absorption measurements indicate that the room temperature fluorescence yield of the BPEA is around 0.99 ± 0.01 independently of the solvent examined. This observation implies that BPEA is an excellent standard for fluorescence quantum yield determinations in the 400-465 absorption and 465-550 nm fluorescence range. By 25 °C fluorescence decay measurements in different apolar solvents, it was shown that the radiative rate dependence of BPEA on the refractive index can be described by a power function of $k_f \sim n^{1.32}$. The exponent of 1.32 of this relationship is almost perfectly agrees with the prediction of Eq. 5 using the experimentally determined spectral shape adjustment and Bakhshiev's spherical empty cavity field correction. A transition dipole moment of the fluorescence of 7.60 Debye was derived in paraffin solvents. The temperature dependence of the radiative rate coefficient shows a more complex picture, the influence of the refractive index induce a negative activation energy

for the k_f parameter, however further processes influence the picture. At temperature higher than 120 °C in HD the internal conversion channel starts to affect the lifetime and fluorescence yield. At lower temperature the increase of k_f with decreasing temperature is proved to be more pronounced than expected from the refractive index variation, which effect may be explained by a small, intrinsic negative temperature coefficient of the transition dipole moment itself.

ASSOCIATED CONTENT

Supporting Information

Figure S1. Triplet-triplet absorption spectra of BPEA;

Figure S2. The temperature dependence of the fluorescence spectra of BPEA in *n*-hexadecane;

Table S1. Selected photophysical parameters of BPEA.

This information is available free of charge via the Internet at <http://pubs.acs.org/>

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