Ground and Excited State Dipole Moments of LAURDAN Determined from Solvatochromic and Thermochromic Shifts of Absorption and Fluorescence Spectra

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The electric dipole moments in the ground μ_g and excited μ_e state of the fluorescent probe LAURDAN are determined from solvatochromic and thermochromic shifts to be $\mu_g = 3.46$ D, $\mu_e = 10.6$ D and $\mu_g = 3.76$ D, $\mu_e = 10.24$ D, respectively. These values concern the free LAURDAN molecule. The values of the dipole moments of LAURDAN (= 6-decanoyl-2-dimethylamine-naphthalene) exceed distinctly those of PRODAN (= 6-propionyl-2-dimethylamine-naphthalene).

Key words: Solvatochromic and Thermochromic Shifts; Dipole Moments in the Ground and Excited States; LAURDAN-Fluorescent Probe.

1. Introductions

LAURDAN (6-dodecanoyl-2-dimethylamine-naphthalene), a fluorescent probe, is often used to study the polarity of lipid interfaces [1–6]. In highly polar solvents LAURDAN shows a considerable red shift of its fluorescence spectrum due to dipolar relaxation processes. This effect is attributed to the high dipole moment of the LAURDAN molecule in its first single excited state.

Figure 1 shows the structural formula of LAURDAN ($n = 10$) and PRODAN ($n = 1$). Due to the lack in the literature of the dipole moments of LAURDAN in the ground, μ_g, and excited, μ_e, state it is commonly assumed that they are comparable to those determined experimentally for PRODAN, which of course may be incorrect.

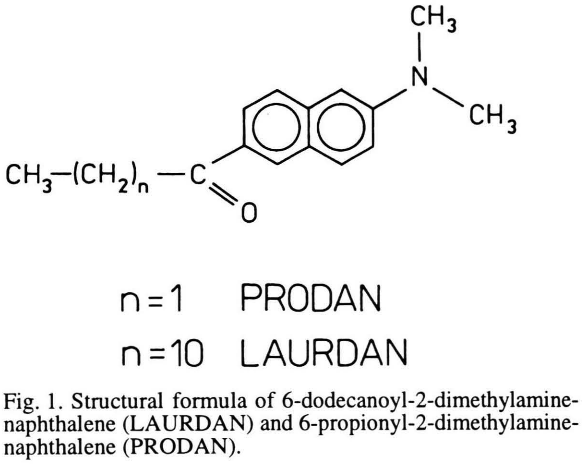

Fig. 1. Structural formula of 6-dodecanoyl-2-dimethylamine-naphthalene (LAURDAN) and 6-propionyl-2-dimethylamine-naphthalene (PRODAN).

The change in the dipole moment $\Delta\mu = \mu_e - \mu_g$ caused for PRODAN by the optical excitation was studied experimentally by several investigators [7–12] who obtained very different values. The value of $\mu_e = 20$ D determined by Weber and Farris [7] for PRODAN is incorrect, as proved by Balter et al. [9]. In our previous works [11, 12] both μ_g and μ_e have been simultaneously determined for PRODAN by the "solvent perturbation" method [13, 14] which makes use of solvatochromic and thermochromic effects. It has been found that the μ_e/μ_g ratio determined by the thermochromic method amounts to 2.7 and is smaller than that determined by the solvatochromic method, for which $\mu_e/\mu_g = 3.0$. The thermochrmoic method is more accurate since the spread in experimental data is smaller. This method strongly restricts specific interactions between luminescent and solvent molecules as well as interactions between the solvent molecules.

In the present work both mentioned methods have been used to determine the dipole moments μ_g and μ_e of LAURDAN.

2. Basic Equations for the Analysis of Electric Dipole Moments

The following equations are based on the quantum-mechanical perturbation theory [13, 14] of the absorp-

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tion ($\tilde{\nu}_A$) and fluorescence ($\tilde{\nu}_F$) band shifts (in wavenumbers) in solvents of different permittivities ε and refractive indexes n for a spherical molecule [15]:

$$\tilde{\nu}_A - \tilde{\nu}_F = m_1 f(\varepsilon, n) + \text{const}\,, \qquad (1)$$

$$\tilde{\nu}_A + \tilde{\nu}_F = -\, m_2\, [f(\varepsilon, n) + 2\, g\,(n)] + \text{const}\,, \qquad (2)$$

where

$$m_1 = \frac{2(\boldsymbol{\mu}_e - \boldsymbol{\mu}_g)^2}{hca^3} = \frac{2(\mu_e^2 + \mu_g^2 - 2\mu_g\, \mu_e \cos\varphi)}{hca^3}, \qquad (3)$$

$$m_2 = \frac{2(\mu_e^2 - \mu_g^2)}{hca^3}, \qquad (4)$$

$$f(\varepsilon, n) = \frac{\dfrac{\varepsilon - 1}{2\varepsilon + 1} - \dfrac{n^2 - 1}{2n^2 + 1}}{\left(1 - \dfrac{2\alpha}{a^3}\dfrac{\varepsilon - 1}{2\varepsilon + 1}\right)\left(1 - \dfrac{2\alpha}{a^3}\dfrac{n^2 - 1}{2n^2 + 1}\right)^2} \qquad (5)$$

$$g\,(n) = \frac{\dfrac{n^2 - 1}{2n^2 + 1}\left(1 - \dfrac{\alpha}{a^3}\dfrac{n^2 - 1}{2n^2 + 1}\right)}{\left(1 - \dfrac{2\alpha}{a^3}\dfrac{n^2 - 1}{2n^2 + 1}\right)^2}\,. \qquad (6)$$

μ_g and μ_e are the dipole moments in the ground and excited state, respectively, α is the polarizability and a the Onsager interaction radius of the solute, h the Planck constant, and c is the velocity of light in the vacuum.

In the case of isotropic polarizability of molecules, the condition $\alpha/a^3 = 1/2$ is frequently satisfied and (5) and (6) are then considerably simplified:

$$f(\varepsilon, n) = \frac{2n^2 + 1}{n^2 + 2}\left(\frac{\varepsilon - 1}{\varepsilon + 2} - \frac{n^2 - 1}{n^2 + 2}\right), \qquad (7)$$

$$g\,(n) = \frac{3}{2}\,\frac{n^4 - 1}{(n^2 + 2)^2}\,, \qquad (8)$$

where $f(\varepsilon, n)$ is the solvent polarity parameter.

If the ground and excited state dipole moments are parallel, based on (3) and (4) the following equations are obtained [13, 16]:

$$\mu_g = \frac{m_2 - m_1}{2}\left(\frac{hca^3}{2m_1}\right)^{1/2}, \qquad (9)$$

$$\mu_e = \frac{m_1 + m_2}{2}\left(\frac{hca^3}{2m_1}\right)^{1/2}, \qquad (10)$$

or

$$\mu_e = \frac{m_1 + m_2}{m_2 - m_1}\,\mu_g\,, \quad (m_2 > m_1). \qquad (11)$$

Generally, the dipole moments μ_g and μ_e are not parallel but make and angle φ. The use of (3) and (4) leads then to

$$\mu_e = \left(\mu_g^2 + \frac{1}{2}\, m_2\, hca^3\right)^{1/2}, \qquad (12)$$

$$\cos\varphi = \frac{1}{2\,\mu_g\,\mu_e}\left[(\mu_g^2 + \mu_e^2) - \frac{m_1}{m_2}(\mu_e^2 - \mu_g^2)\right]. \qquad (13)$$

The parameters m_1 and m_2 can be determined from the absorption and fluorescence band shifts given by (1) and (2). From the discussion of (13) it follows that for $m_1/m_2 = 1/2$ and $\mu_e/\mu_g = 3$ is $\varphi = 0$, and the dipole moments $\boldsymbol{\mu}_g$ and $\boldsymbol{\mu}_e$ are parallel.

3. Experimental

3.1 Methods

The absorption and fluorescence spectra of LAURDAN in ethyl acetate between 246 and 383 K were measured with the apparatus described in [12] using home constructed high pressure cells. The determination of permittivities ε and refractive indexes n at different temperatures were determined from the empirical formulas and are listed for ethyl acetate in [12, 15, 17].

3.2 Solvatochromic Shifts of Absorption and Fluorescence Spectra of LAURDAN in Different Solvents

Figure 2 shows selected spectra of LAURDAN in different solvents. In unpolar *n*-hexane, the absorption and fluorescence maxima are $\tilde{\nu}_A = 29142$ cm^{-1} and $\tilde{\nu}_F = 25610$ cm^{-1}, respectively. However, in the strongly polar acetonitrile $\tilde{\nu}_A = 28239$ cm^{-1} and $\tilde{\nu}_F = 22025$ cm^{-1}. Passing from the unpolar *n*-hexane ($f(\varepsilon, n) = 0.0043$) to the strongly polar acetonitrile ($f(\varepsilon, n) = 0.8636$) the fluorescence spectrum shifts very strongly ($\Delta\nu_F = 3585$ cm^{-1}) compared to the absorption spectrum ($\Delta\nu_A = 903$ cm^{-1}). Figures 3 and 4 show the spectra shifts $\tilde{\nu}_A - \tilde{\nu}_F$ and $\tilde{\nu}_A + \tilde{\nu}_F$ of LAURDAN plotted for 14 solvents versus the solvent polarity functions $f(\varepsilon, n)$ and $f(\varepsilon, n) + 2g\,(n)$, respectively. A linear regression was carried out and a fit

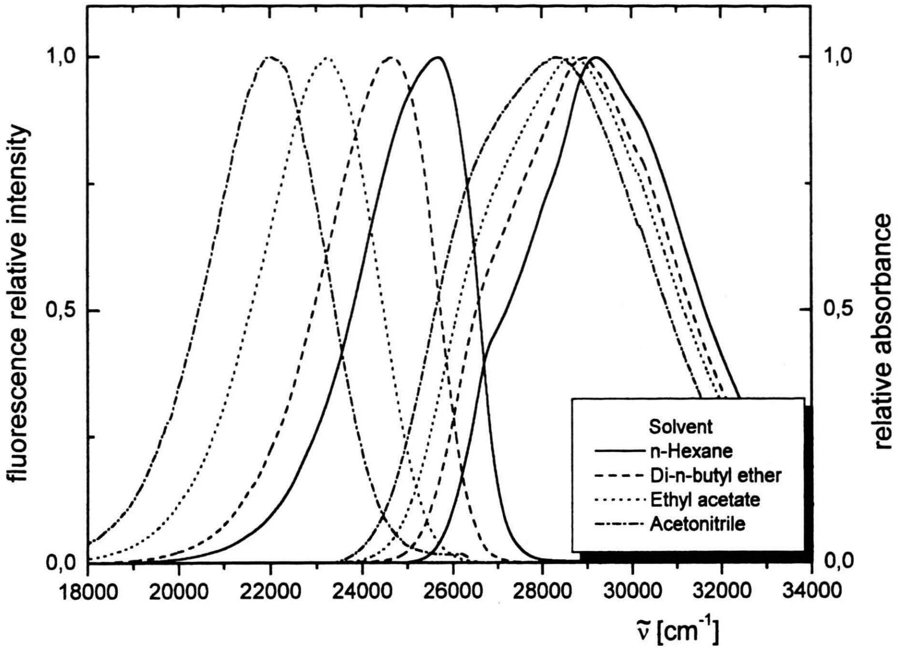

Fig. 2. Absorption and fluorescence spectra of LAURDAN in different solvents.

Table 1. Dipole moments determined from solvatochromic shifts and thermochromic shifts for LAURDAN. The conversion factor for the dipole moment: $[\mu]_{SI}/\mathrm{Cm} = 3.33564 \times 10^{-30}\ [\mu]_{cgs}/\mathrm{D}$, where D is the symbol of Debye and $1\ \mathrm{D} = 10^{-18}$ esu cm.

Onsager radius a [Å]	Solvatochromic shifts						Thermochromic shifts					
	m_1	m_2	μ_g	μ_e	$\Delta\mu$	μ_e/μ_g	m_1	m_2	μ_g	μ_e	$\Delta\mu$	μ_e/μ_g
	[cm⁻¹]		in Debye [D]				[cm⁻¹]		in Debye [D]			
5.5	2640	5410	3.46	10.06	6.6	2.91	2540	5490	3.76	10.24	6.48	2.72

to these data was obtained *. The slopes of the fitted lines presented in Figs. 3 and 4 were found to be $m_1 = 2640$ cm^{-1} and $m_2 = 5410$ cm^{-1}, repectively, and they are given in Table 1.

3.3 Thermochromic Shifts of Absorption and Fluorescence Spectra of LAURDAN in Ethyl Acetate

Figure 5 illustrates the LAURDAN absorption and fluorescence spectra in ethyl acetate between 246 and 383 K. A blue shift of the spectra is observed with increase in temperature. With increase in temperature the dielectric permittivity, ε decreases [12, 17], which leads to the decrease of solvent polarity parameter $f(\varepsilon, n)$. Since for LAURDAN, similarly as for PRODAN, a strong fluorescence quenching takes place with increase in temperature (Fig. 6), it was not possible to exceed the temperature 373 K.

Figures 7 and 8 show the thermochromic spectra shifts $\tilde{\nu}_A - \tilde{\nu}_F$ and $\tilde{\nu}_A + \tilde{\nu}_F$ versus the solvent polarity functions $f(\varepsilon, n)$ and $f(\varepsilon, n) + 2\,g(n)$ as well as on temperature T (according to (14) and (15) in our previous paper [12]), respectively. The slopes of the fitted lines presented in Figs. 7 and 8 were found to be $m_1 = 2540$ cm^{-1} and $m_2 = 5490$ cm^{-1}, respectively, and they are given in Table 1.

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* It should be remarked that in this case the sequence of experimental points in Fig. 4 is not identical to that in Fig. 3, which is caused by the function $g(n)$, i.e., a large irregularity of the refractive index n when passing from one to another solvent.

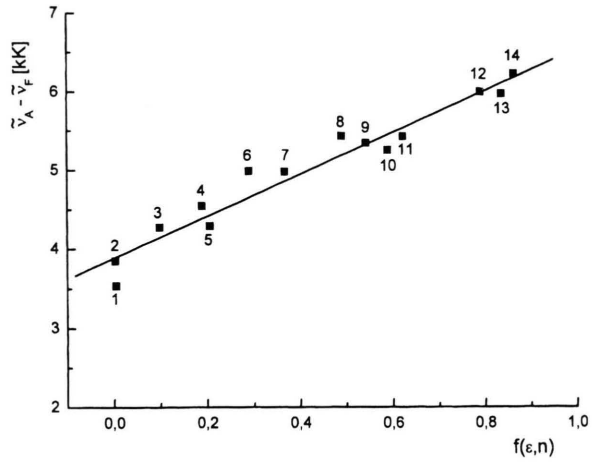

Fig. 3. Plots of $\tilde{\nu}_A - \tilde{\nu}_F$ versus $f(\varepsilon, n)$ for LAURDAN in different solvents: 1 = *n*-heptane, 2 = *n*-hexane, 3 = triethylamine, 4 = trichloroethylene, 5 = di-*n*-butyl ether, 6 = anisole, 7 = diethylether, 8 = ethyl acetate, 9 = tetrahydrofuran, 10 = 1,2-dichloromethane, 11 = 1,2-dichloroethane, 12 = acetone, 13 = N,N-dimethylformamide, 14 = acetonitrile.

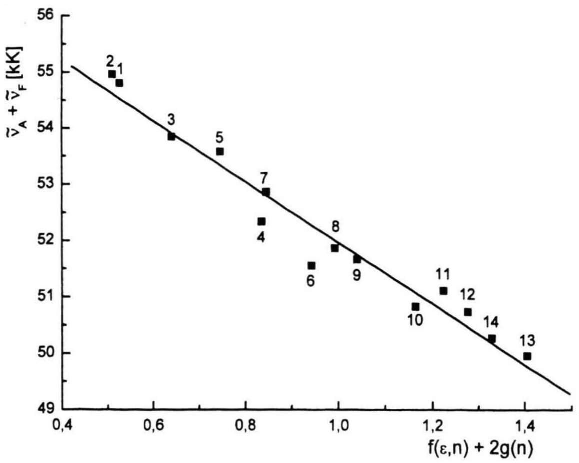

Fig. 4. Plots $\tilde{\nu}_A + \tilde{\nu}_F$ versus $f(\varepsilon, n) + 2\,g(n)$ for LAURDAN in the same solvents as in Figure 3.

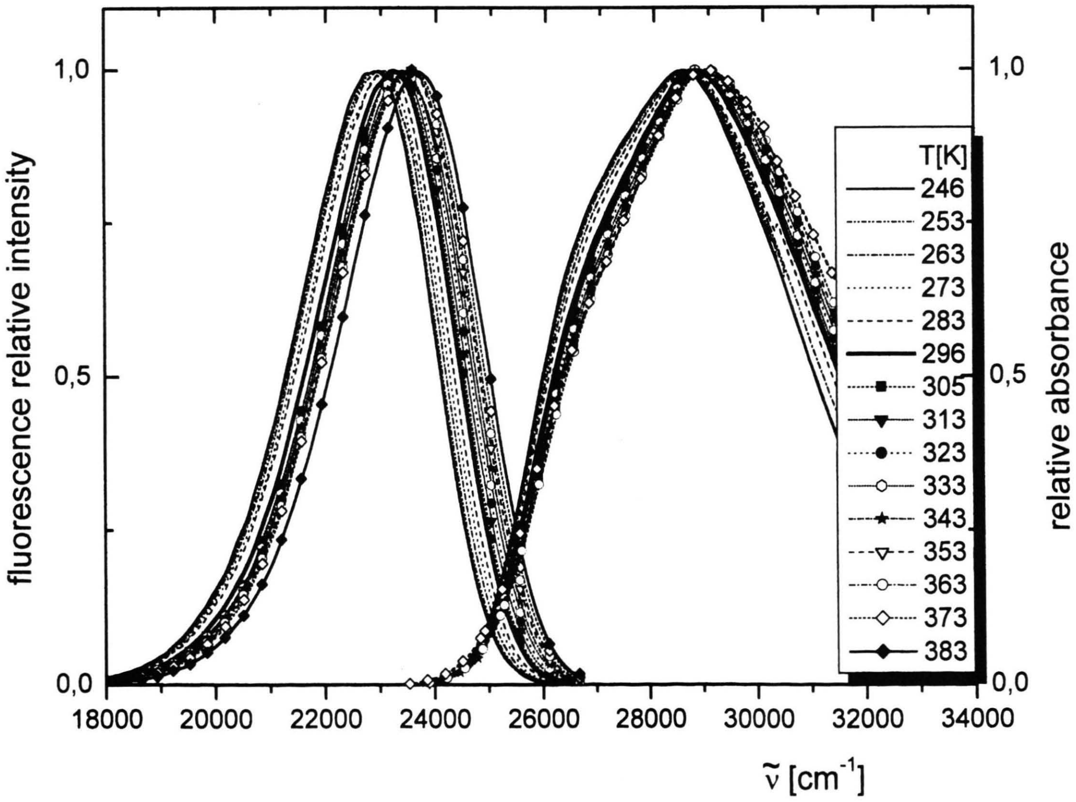

Fig. 5. Absorption and fluorescence spectra shifts for LAURDAN in ethyl acetate at different temperaures *T*.

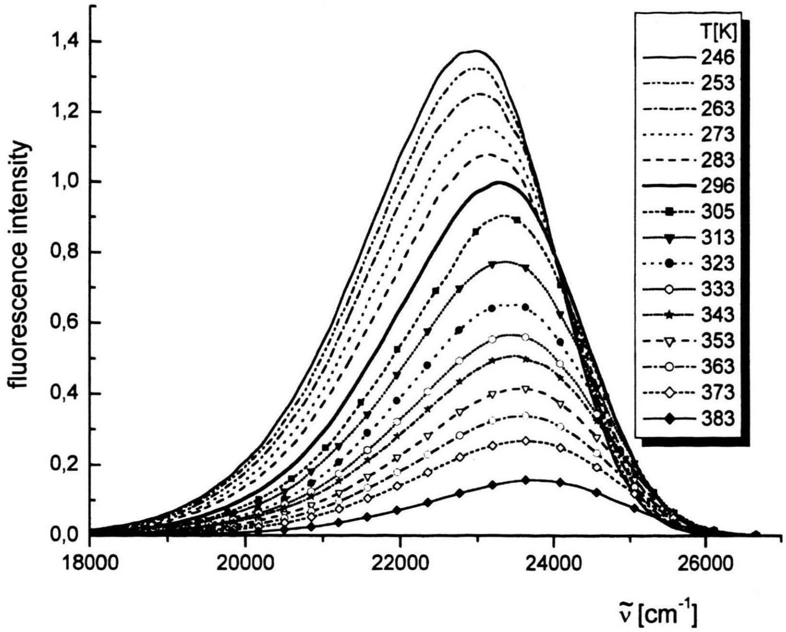

Fig. 6. Influence of temperature on the fluorescence intensity for LAURDAN in ethyl acetate from 246 to 383 K.

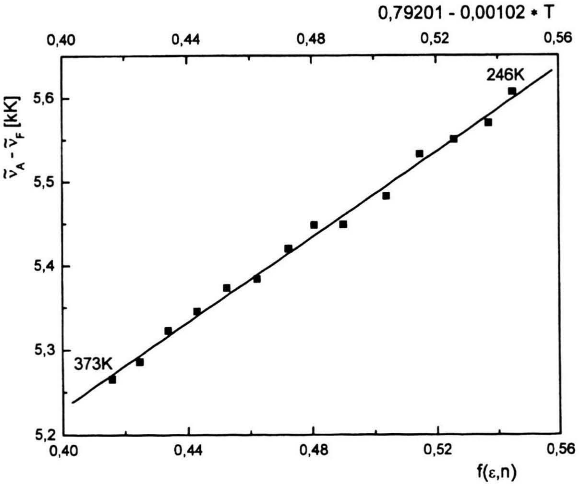

Fig. 7. Plots of $\tilde{\nu}_A - \tilde{\nu}_F$ versus $f(\varepsilon, n)$ for LAURDAN in ethyl acetate at different temperatures T.

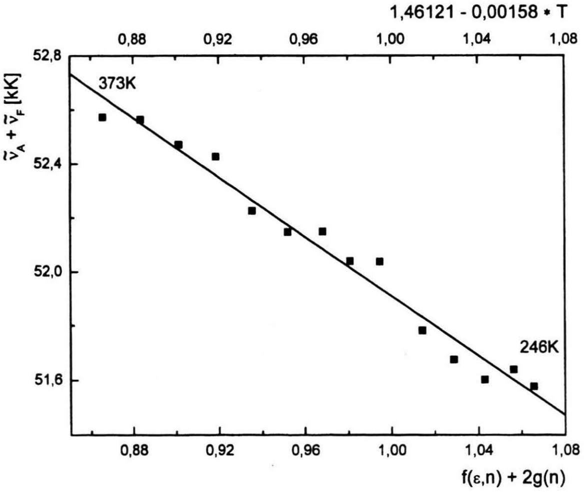

Fig. 8. Plots $\tilde{\nu}_A + \tilde{\nu}_F$ versus $f(\varepsilon, n) + 2\,g(n)$ for LAURDAN in ethyl acetate at different temperaures T.

4. Ground and Excited State Dipole Moments of LAURDAN

According to (3) and (4) or (9) and (10) the values of the dipole moments μ_g and μ_e depend not only on the parameters m_1 and m_2 but also on the Onsager interaction radius a. For LAURDAN one can assume $a = 5.5$ Å, the same as calculated for MANA** (Fig. 9) by Sández et al. [18] with the help of Discover software from Biosym, which allows molecules to be constructed and optimized by molecular dynamics, and to calculate interatomic distances. They used an a value of one-half the average

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** ω-[6-(N-methyl-N-alkylamine)naphthoyl]alcanoic acid.

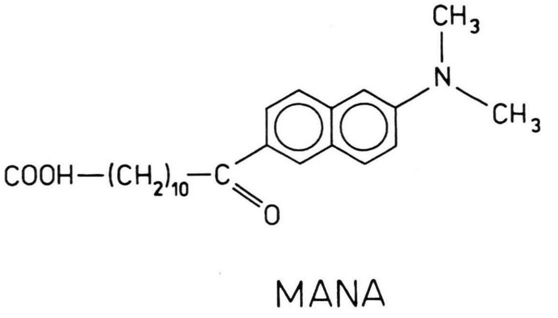

Fig. 9. Stuctural formula of ω-[6-(N-methyl-N-alkylamine)-naphthoyl]alcanoic acid (MANA).

molecular size in the three dimensions. Comparing the structural formulae (Figs. 1 and 9) it is seen that both chemical compounds LAURDAN and MANA differ only in the substituents CH_3 and COOH, respectively.

The values of μ_g and μ_e for LAURDAN determined using both methods are given in Table 1. It is seen that the ratio $\mu_e/\mu_g = 2.91$ determined from the solvatochromic method is somewhat bigger than that $\mu_e/\mu_g = 2.72$ obtained from the thermochromic method. A similar behaviour was observed for PRODAN [12]. The ratio μ_e/μ_g is for both molecules (LAURDAN, PRODAN) independent of the Onsage radius, which is in accordance with (11). This indicates that $\boldsymbol{\mu}_g$ and $\boldsymbol{\mu}_e$ are parallel to one to another.

5. Concluding Remarks

The electric dipole moments of LAURDAN (μ_g = 3.76 D, μ_e = 10.24 D for a = 5.5 Å) are much bigger than those of PRODAN (μ_g = 2.45 D, μ_e = 6.65 D for a = 4.2 Å). These values concern free molecules. The thermochromic method allows for more accurate determination of the above quantities.

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