A spectroscopic/thermodynamic study of the rhodamine B lactone \rightleftharpoons zwitterion equilibrium

DANIEL A. HINCKLEY* and PAUL G. SEYBOLD[†]

Departments of Chemistry and Biological Chemistry, Wright State University, Dayton, OH 45435, U.S.A.

(Received 24 February 1988; accepted 9 March 1988)

Abstract—Temperature dependences of the visible absorption spectra of rhodamine B (RB) in water and 18 alcohol solvents have been examined and utilized to estimate thermodynamic parameters $(\Delta G^{\circ}, \Delta H^{\circ}, \Delta S^{\circ})$ for the RB lactone \rightleftharpoons zwitterion $(L \rightleftharpoons Z)$ equilibria in these solvents. From the $L \rightleftharpoons Z$ equilibrium in the strong hydrogen-bonding solvent trifluoroethanol an intrinsic RB zwitterion molar absorptivity of $\varepsilon = 1.30 \times 10^5$ dm³/mol cm is estimated. The thermodynamic results suggest that the position of the RB $L \rightleftharpoons Z$ equilibrium is aliphatic alcohols can be predicted from simple features of the solvent. The position of the results regarding calculation of the natural radiative lifetime of the fluorescent state from the absorption spectrum are discussed.

INTRODUCTION

Rhodamine dyes have found a wide variety of practical applications, including roles as biological stains, tracing agents, solar collectors, and laser dyes. These dyes can exist in solution in several forms, and for many applications it is crucial that the forms present be properly characterized. For fluorescence studies, for example, each form possesses its own absorption and emission spectra, fluorescence yield, and excitedstate lifetime. In an earlier study [1] we examined the solvatochromism of rhodamine B (RB) in 16 protic and 14 aprotic solvents and the thermochromism of RB in three solvents. It was concluded that the zwitterion (Z) form of RB is absent in aprotic solvents and present in equilibrium with the lactone (L) form in protic solvents. The position of the $L \rightleftharpoons Z$ equilibrium was found to depend on solvent-dye hydrogen bonding (favoring the zwitterion) and solvent dielectric/ polarizability properties. The cation (C) form of RB appears in both protic and aprotic solvents under acidic conditions.

A goal of this research has been to understand the dependence of RB solution behavior on the solvent environment, including solvent hydrogen-bonding ability and other characteristics. Our earlier study of the RB $L \rightleftharpoons Z$ equilibrium [1] encompassed only a limited number of secondary alcohols and just one tertiary alcohol, and it was therefore of interest to examine this equilibrium in additional solvents exhibiting an expanded range of structural variations. Moreover, examination of the temperature variations of the RB spectra in these solvents allows one to derive thermodynamic data for the $L \rightleftharpoons Z$ equilibrium. In our previous analysis of RB spectra, temperature de-

pendences were examined for only three solvents, and an Arrhenius form for the temperature dependence was assumed. In the present study temperature variations of the RB spectra in 19 solvents have been examined, and a more complete temperature analysis has been performed.

In [1], the reference molar absorptivity for the Z form of RB was assumed to be 13.0 $\times 10^4$ dm³/mol·cm. Here a derivation of this value is presented from an evaluation of the temperature-dependent behavior of RB in the strong hydrogenbonding solvent trifluoroethanol (TFE).

EXPERIMENTAL

The experimental procedure outlined in Ref. [1] was followed. The RB was Eastman laser grade, vacuum desiccated before use. All neutral RB solutions in secondary and tertiary alcohols showed the presence of RB cation; 5μ l of triethylamine were added to 10 ml of these solutions to eliminate this interference. Base was not normally added to RB solutions in primary alcohols because no significant differences in RB absorptivity were noted between the neutral and basic solutions. Absorption spectra of RB in all solvents were obtained from 15°C to 55°C at 10°C intervals, with a minimum of four runs for each. Spectra were taken on a Varian 2300 spectrometer.

Suitable density data for several solvents (TFE, *m*-cresol, cyclohexanol) could not be located in the literature. Densities were experimentally determined for these solvents using Weld pycometers.

METHODS

Calculation of the equilibrium constant K

1

For the RB L \rightleftharpoons Z equilibrium K was defined as

$$K = \frac{[Z]}{[L]}.$$
 (1)

The fraction of Z present at each temperature was calculated from the visible absorption spectrum using

$$[Z] = \frac{[\varepsilon_x(T)][\rho(15^{\circ}C)]}{[13.0 \times 10^4 \,\mathrm{dm^3/mol \cdot cm}][\rho(T)]}$$
(2)

^{*}ACS-Petroleum Research Fund Scholar, 1982–83. Present address: Program in Marine Science, University of South Carolina, Columbia, SC 29208, U.S.A.

[†]Author to whom correspondence should be addressed.



where $\varepsilon_x(T)$ is the observed molar absorptivity at temperature T, $\rho(15^{\circ}C)$ and $\rho(T)$ are the densities of the solvent at 15°C and temperature T, respectively, and $13.0 \times 10^4 \text{ dm}^3/\text{mol} \cdot \text{cm}$ is the standard molar absorptivity ε_0 determined for 100% Z (see below). The fraction of RB present as lactone was calculated from

$$[L] = 1 - [Z]. \tag{3}$$

Analysis of the data

The standard Gibbs energy change ΔG° for the $L \rightleftharpoons Z$ equilibrium is

$$\Delta G^{\circ} = -\mathbf{R} T \ln K \tag{4}$$

where \mathbf{R} is the gas constant and T is the temperature in kelvins. Substituting into

$$\Delta G^{\circ} = \Delta H^{\circ} - T \Delta S^{\circ} \tag{5}$$

and solving for ln K one obtains

ŀ

$$\ln K = \Delta S^{\circ}/\mathbf{R} - (\Delta H^{\circ}/\mathbf{R}) (1/T).$$
(6)

If ΔH° and ΔS° are approximately constant over the temperature range examined, a plot of $\ln K$ vs 1/T should yield a straight line with slope $-(\Delta H^{\circ}/R)$ and intercept $\Delta S^{\circ}/R$. However, examinations of such plots revealed small to moderate deviations from linearity (cf. Fig. 8 of Ref. [1]).

To obtain a more accurate analysis temperature variations of the equilibrium constant were fit by regression analysis to the polynomial

n
$$K = a + b/T + c/T^2$$
. (7)

This allowed determination of the coefficients, a, b, and c. From Eqn. (4),

$$\Delta G^{\circ} = -\mathbf{R} \, Ta - \mathbf{R} b - \mathbf{R} c / T. \tag{8}$$

Since [2]

$$\Delta S^{\circ} = -\left(\frac{\partial \Delta G^{\circ}}{\partial T}\right)_{P} \tag{9}$$

it follows that

$$\Delta S^{\circ} = \mathbf{R}a - \mathbf{R}c/T^2, \qquad (10)$$

and from Eqn. (5),

$$\Delta H^{\circ} = -\mathbf{R}b - 2\mathbf{R}c/T. \tag{11}$$

Thus, having determined the coefficients a, b, and c for a solvent, ΔH° was calculated using Eqn. (11), ΔS° from Eqn. (10), and ΔG° from Eqn. (4).

Correlation with properties of the solvent

Values of ΔH° and ΔS° for the $L \rightleftharpoons Z$ equilibrium at 25°C and the experimental ln K at 25°C were next modeled using simple solvent structural parameters described previously [1]: $N_{\rm e}$, the number of carbons in the alcohol; C_{α} , the number of carbons bonded to the α carbon, and $T_{\rm m}$, the number of terminal methyl groups. ΔH° , ΔS° , and ln K were also regressed against MCRAE's solvent functions [3] and the solvent dielectric constant, D.

Additional details of the methods used can be found elsewhere [4].

RESULTS

Reference absorptivity of Z

Before an analysis of the data could be performed, it was necessary to establish the intrinsic molar absorptivity ε_0 of the Z form of **RB**, which was to be used as a reference in Eqn. (2). The Z form exhibits its maximum absorbance in the strong hydrogen-bond donating solvent TFE. Analysis of the temperature variations of the RB spectrum in TFE revealed changes in molar absorptivity exceeding those which could be accounted for by volume changes of the solvent. Arrhenius analyses of the data were performed based on ε_0 values ranging from 12.0 to 13.5 $\times 10^4$ dm³/mol·cm. Better correlations were obtained for the higher values of ε_0 . Moreover, values for ΔH° and ΔS° obtained using both $\varepsilon_0 = 13.0 \times 10^4$ and $\varepsilon_0 = 13.5 \times 10^4 \text{ dm}^3/\text{mol}\cdot\text{cm}$ were comparable in magnitude to values found previously for other solvents (Ref. [1], Table 4).

Many workers have compared the alcohols TFE and ethanol because of their special similarities and differences [5–7]. TFE clearly is a better hydrogenbond donor than ethanol, but it is a poorer hydrogenbond acceptor. As a consequence, self-association in TFE is less than in ethanol [7–10]. Based on our previous model of the $L \rightleftharpoons Z$ equilibrium,

$$L + A \rightleftharpoons Z \cdots A$$

where the dots represent hydrogen bonding between Z and the alcohol (donor) solvent A, it follows that both ΔS° and ΔH° for the L \rightleftharpoons Z equilibrium should be more negative in TFE than in ethanol. Table 1 contains thermodynamic values obtained for the L \rightleftharpoons Z equilibrium using different ε_0 reference values. Use of $\varepsilon_0 = 13.5 \times 10^4 \text{ dm}^3/\text{mol} \cdot \text{cm}$ yields more negative ΔH° and ΔS° values in ethanol than in TFE, the reverse of what we expect. Both $\varepsilon_0 = 12.7 \times 10^4$ and $\varepsilon_0 = 13.0 \times 10^4 \text{ dm}^3/\text{mol} \cdot \text{cm}$ yield more negative ΔH° and ΔS° values in TFE than in ethanol, in line with the prediction.

For the above reasons the 100% Z molar absorptivity reference used in our analysis was chosen to be 13.0 $\times 10^4$ dm³/mol·cm. This value is probably accurate to within $\pm 0.3 \times 10^4$ dm³/mol·cm. From Eqns (1)–(3) it is apparent that a small inaccuracy in this value will have a relatively large effect on thermodynamic results calculated for solvents in which [Z] approaches unity, i.e. TFE, but only a relatively small effect for most solvents (cf. ethanol in Table 1).

In their examination of rhodamine spectra, ROSENTHAL *et al.* [11] assumed that **RB** existed as 100% Z in glacial acetic acid, an erroneous assumption since the **RB** cation is the dominant form in that solvent [1]. However, the molar absorptivity of RB in glacial acetic acid of $12.9 \times 10^4 \text{ dm}^3/\text{mol}\cdot\text{cm}$ is remarkably close to the value used here.

Solvent effects

Solvent effects on the RB $L \rightleftharpoons Z$ equilibrium at 25°C are summarized in Table 2 for 25 protic solvents examined in this work and Ref. [1]. In these solvents the position of the $L \rightleftharpoons Z$ equilibrium exhibits a wide range, from 95% Z in TFE to 0.4% Z in *t*-pentanol.

The two most apparent trends in the solvent data are the shifts of the equilibrium from Z toward L with (i) greater aliphatic character of the alcohol and (ii) greater steric hindrance about the hydroxyl group. Both were noted previously [1]. The former is best illustrated by the steady decrease of K with increasing chain length of the normal alcohols. The latter trend is more striking, as seen in the series *n*-butanol > 2-butanol > t-butanol and *n*-pentanol > 2-

Table 1. Thermodynamic values for the $L \rightleftharpoons Z$ equilibrium in TFE and ethanol assuming different intrinsic molar absorptivities for the Z form

100% Z Reference (×10 ⁻⁴)	${ m TFE} \Delta H^{\circ} ({ m kJ/mol})$	$\Delta S^{\circ}(\mathbf{J}/\mathrm{mol}\cdot\mathbf{K})$	Ethanol Δ <i>H</i> °(kJ/mol)	$\Delta S^{\circ}(J/mol \cdot K)$
12.7	- 38.4	-103	- 19.1	- 53.1
13.0	-23.6	- 55.4	-18.0	- 53.2
13.5	-13.4	-25.6	-16.6	- 49.3

	olvent	$\hat{\lambda}_{max}$	ε* (dm³/mol∙cm)	%Z	ĸ	ΔG°
		()	(um / mor vm)	, 62		(0) mory
1.	Methanol	545	11.6×10^{4}	89.2	8.29	-5240
2.	Ethanol	540	9.28×10^{4}	70.6	2.40	-2170
3.	n-Propanol	542	8.57×10^{4}	65.2	1.88	-1560
4.	i-Propanol	539	3.80×10^{4}	28.9	0.407	+2230
5.	n-Butanol	542	7.10×10^{4}	54.1	1.18	-408
6.	<i>i</i> -Butanol	542	7.38×10^{4}	56.2	1.28	- 619
7.	2-Butanol	539	2.17×10^{4}	16.6	0.199	+4000
8.	t-Butanol†	538	1440	1.6	0.016	+10500
9.	n-Pentanol	543	5.87×10^{4}	44.7	0.810	+ 524
10.	<i>i</i> -Pentanol	543	5.98×10^{4}	45.8	0.845	+416
11.	2-Methyl-1-butanol	540	5.33×10^{4}	40.6	0.683	+ 944
12.	2-Pentanol	540	1.01×10^{4}	7.7	0.083	+6160
13.	3-Pentanol	542	7790	5.9	0.063	+6850
14.	3-Methyl-2-butanol	540	9220	7.0	0.076	+6400
15.	t-Pentanol	532	473	0.36	0.004	+14000
16.	<i>n</i> -Hexanol	545	5.80×10^{4}	44.6	0.806	+ 535
17.	2-Ethyl-1-butanol	546	3.80×10^{4}	29.1	0.410	+2210
18.	n-Octanol	543	4.29×10^{4}	32.9	0.489	+1770
19.	n-Decanol	545	3.50×10^{4}	26.9	0.368	+2480
20.	Ethylene glycol	553	11.4×10^{4}	87.7	7.13	-4870
21.	Water	552	10.6×10^{4}	81.5	4.40	-3670
22.	Formamide	554	11.5×10^{4}	88.5	7.67	- 5050
23.	2,2,2-Trifluoroethanol					
	(TFE)	546	12.3×10^4	94.6	17.6	-7110
24.	m-Cresol	560	11.4×10^{4}	87.1	6.73	-4730
25.	Cyclohexanol [†]	544	1.80×10^{4}	13.9	0.161	+4610

Table 2. Solvent influences on the RB L \rightleftharpoons Z equilibrium at 25°C

*Apparent molar absorptivity, \pm 5%.

†t-Butanol and cyclohexanol at 30°C.

pentanol > t-butanol. In Table 3 it is seen that ΔG° for the L \rightleftharpoons Z equilibrium increases roughly 6 kJ/mol for each step in going from primary to secondary to tertiary alcohols. Additional $-CH_{2^{-}}$ groups increase ΔG° by about 1 kJ/mol for primary alcohols, 2 kJ/mol for secondary alcohols, and 3.5 kJ/mol for the tertiary alcohols. As a rule only small differences in %Z and ΔG° are found within isomeric groups of primary and secondary alcohols. An exception is the significant difference in %Z and ΔG° found between *n*-hexanol and 2-ethyl-1-butanol.

Temperature effects

For illustration, temperature dependences of the RB spectra in *n*-pentanol and *i*-pentanol are shown in Figs 1 and 2. In most cases the RB absorption maximum exhibited a small hypsochromic shift with increasing temperature from 15° C to 55° C, e.g. from 544 nm to 543 nm for *n*-pentanol, and from 541 nm to 537 nm for *i*-pentanol.

The standard enthalpy and entropy changes found for the RB L \Rightarrow Z equilibrium in 19 solvents at 25°C are given in Table 4. Most ΔH° values were in the range -23 to -29 kJ/mol, with extremes of -34.7 kJ/mol in 3-methyl-2-butanol and -5.9 kJ/mol in water. ΔS° values commonly fell between -60 and -110 J/mol·K with extreme values of -138 J/mol·K in 3-methyl-2-butanol and -16.4 J/mol·K in water.

For the normal alcohols ΔG° became more positive and ΔS° more negative with increasing chain length (Table 5). ΔH° became more negative with increasing solvent chain length, with the exceptions of *n*-butanol and *n*-pentanol, which have nearly identical ΔH° values. Within each isomer class, branching caused ΔG° to become more positive and ΔH° and ΔS° to become more negative. (The data for *t*-pentanol, a difficult solvent to work with, due in part to the extremely low RB absorbance, appear to be anomalous.)

Correlation of ln K with properties of the solvent

Linear regression of ln K with the aliphatic alcohol solvent parameters C_{α} , N_{c} , and T_{m} yielded

$$\ln K = -0.236(\pm 0.058)N_{\rm c} - 0.459(\pm 0.262)T_{\rm m}$$
$$-1.904(\pm 0.263)C_{\alpha} + 0.568(\pm 0.417) \quad (12)$$
$$n = 18, \qquad r = 0.967, \qquad s = 0.441$$

where r is the correlation coefficient and s is the standard deviation. A plot of calculated [Eqn. (12)] vs experimental values of ln K is shown in Fig. 3. The coefficients of Eqn. (12) are similar to coefficients found earlier {Ref. [1], Eqn. (2)} for a smaller sample of solvents. Neither ΔH° (r = 0.626) nor ΔS° (r = 0.875) correlated very well with the same parameters.

Examination of the dependence of $\ln K$ on the dielectric constant, D, of these aliphatic alcohol solvents showed a correlation (r = 0.701) much smaller

Table 3. Effect of C_{α} on ΔG° for propanols, butanols, and pentanols

Solvent	Ca	$\Delta G^{\circ}(J/mol)$
n-Propanol	1	-1560
i-Propanol	2	+ 2230
n-Butanol	1	-408
i-Butanol	1	-619
2-Butanol	2	4000
t-Butanol	3	10 500
n-Pentanol	1	416
i-Pentanol	1	525
2-Methyl-1-butanol	1	944
2-Pentanol	2	6160
3-Pentanol	2	6400
3-Methyl-2-butanol	2	6850
t-Pentanol	3	14 000



Fig. 1. Absorption spectra of RB in n-pentanol, 15-55°C.

than that found in our previous evaluation (r = 0.921)[1]. Figure 4 reveals the reason for this poor correlation: the number of secondary and tertiary alcohols included in the present sample is greater, and as found earlier, these alcohols do not follow the simple pattern of the primary alcohols. This observation implies that addition of the steric hindrance parameter C_{α} to the analysis should improve the correlation. Linear regression of ln K against both D and C_{α} yields

$$\ln K = -1.859(\pm 0.202)C_{a} + 0.087(\pm 0.021)D$$

+0.344(±0.517) (13)
$$n = 17, \quad r = 0.964, \quad s = 0.345.$$



Fig. 2. Absorption spectra of RB in 2-pentanol, 15-55°C.

Table 4. Standard enthalpy and entropy changes for the $L \rightleftharpoons Z$ equilibrium at 25°C*

Solvent	Δ <i>H</i> ° (kJ/mol)	ΔS° (J/mol·K)
Ethanol	- 18.0	-53.2
n-Propanol	- 19.8	-61.2
i-Propanol	-24.8	-90.6
n-Butanol	-23.7	-78.2
i-Butanol	- 24.8	-81.0
2-Butanol	-28.5	-109.0
t-Butanol [†]	-28.4	-128.0
n-Pentanol	-23.1	- 79.5
<i>i</i> -Pentanol	- 27.0	-91.9
2-Methyl-1-butanol	- 27.2	- 94.4
2-Pentanol	- 28.6	-117.0
3-Pentanol	- 29.6	-122.0
3-Methyl-2-butanol	- 34.7	-138.0
t-Pentanol	(-17.1)‡	(-104.0)
n-Octanol	- 28.1	-100.0
TFE	-23.6	- 55.4
m-Cresol	-22.8	- 60.5
Cyclohexanol [†]	27.0	-104.0
Water	- 5.9	-16.4

* \pm 10% for most solvents; less accurate for TFE (see text).

†At 30°C.

‡Values uncertain due to extremely low RB absorbance.

Table 5. Thermodynamic values for $L \rightleftharpoons Z$ in normal alcohols

Solvent	$\Delta G^{\circ}(\mathrm{J/mol})$	$\Delta H^{\circ}(\mathrm{kJ/mol})$	$\Delta S^{\circ}(J/mol \cdot K)$
Ethanol	-2170	-18.0	-53.2
n-Propanol	-1560	-19.8	-61.2
n-Butanol	408	-23.7	-78.2
n-Pentanol	+ 524	-23.1	- 79.5
n-Octanol	+1770	-28.1	100

When ΔH° and ΔS° were modeled against D alone and D plus C_{α} , no correlations better than r = 0.765were found.



Fig. 3. Rhodamine B $L \rightleftharpoons Z$ in alcohols: Comparison against C_{α} , T_{m} , and N_{c} : \bullet normal alcohols; \bigcirc other primary alcohols; \ominus secondary alcohols; and \oplus tertiary alcohols.



Fig. 4. Relationship between $\ln K$ for the RB L \rightleftharpoons Z equilibrium and the dielectric constant of aliphatic alcohols: ● normal alcohols; \bigcirc other primary alcohols; \ominus secondary alcohols; and \oplus tertiary alcohols.

MCRAE's solvent functions [3] and yielded poor correlations, ranging from 0.682 for ln K to 0.816 for ΔH° . Addition of the steric hindrance parameter C_x greatly improved the correlations:

$$\ln K = 4.33(\pm 16.75) \left(\frac{n_r^2 - 1}{2n_r^2 + 1}\right) + 7.06(\pm 1.78)$$
$$\left(\frac{D - 1}{D + 2} - \frac{n_r^2 - 1}{n_r^2 + 2}\right) - 1.92(\pm 0.17)C_x$$
$$- 3.07(\pm 4.07)$$
(14)

n = 17, r = 0.976, s = 0.376.

Ln K, ΔH° , and ΔS° were regressed against where n_r is the solvent refractive index. ΔS° correlated

well with MCRAE's terms and C_{α} :

$$\Delta S^{\circ} = -3650(\pm 445) \left(\frac{n_{\rm r}^2 - 1}{2n_{\rm r}^2 + 1} \right)$$
$$-199(\pm 45) \left(\frac{D - 1}{D + 2} - \frac{n_{\rm r}^2 - 1}{n_{\rm r}^2 + 2} \right)$$
$$-31.3(\pm 3.9)C_a + 778(\pm 111) \quad (15)$$
$$n = 13, \qquad r = 0.966, \qquad s = 5.27 \text{ J/mol·K.}$$

The fit for ΔH° was

$$\Delta H^{\circ} = -1.0(\pm 0.1) \times 10^{6} \left(\frac{n_{r}^{2} - 1}{2n_{r}^{2} + 1}\right)$$
$$-7.77(\pm 1.20) \times 10^{4} \left(\frac{D - 1}{D + 2}\right)$$
$$-\frac{n_{r}^{2} - 1}{n_{r}^{2} + 2}\right)$$
$$-4.38(\pm 1.05) \times 10^{3} C_{\alpha}$$
$$+ 2.21(\pm 0.30) \times 10^{5} \qquad (16)$$

$$n = 13$$
, $r = 0.941$, $s = 1420 \text{ J/mol.}$

DISCUSSION

Because the highest Z absorption observed for RB occurs in the strong hydrogen-bonding solvent TFE, it is tempting to assume that in TFE RB is present completely in its Z form. Analysis of the temperature dependence of the spectrum of RB in TFE, however, reveals a decrease in absorbance with increased temperature greater than can be accounted for by solvent expansion. In addition, if the assumption is made that an $L \rightleftharpoons Z$ equilibrium exists in TFE, resulting ΔH° and ΔS° values calculated for the equilibrium are consistent with those found in other solvents (Table 4). These observations support the view that an RB $L \rightleftharpoons Z$ equilibrium exists in TFE.

The 100% Z reference molar absorptivity value ε_0 = 13.0 (± 0.3) × 10⁴ dm³/mol·cm was chosen from two considerations. First, the fit of the data for the RB L \Rightarrow Z equilibrium in TFE was best for higher ε_0 values. Second, this choice for ε_0 yielded, as expected, more negative values for ΔH° and ΔS° in TFE relative to those in ethanol (Table 1), whereas still higher ε_0 values yield a contrary result. Nonetheless, we prefer to regard this value as tentative and believe further investigation of this subject would be valuable. Fortunately, modest alterations of this reference value would lead to only small changes in the results obtained here for most solvents.

An important consequence of the $L \rightleftharpoons Z$ equilibrium arises in the calculation of the natural radiative lifetime τ_r of the fluorescent excited state [12, 13]. If our conclusions about the $L \rightleftharpoons Z$ equilibrium are substantially correct, values for the integrated absorption intensity of the visible transition of RB obtained from absorption spectra will be underestimates of the

true values, since only a fraction of the dye in solution is actually present as the zwitterion. As a result, natural radiative lifetimes calculated from the absorption spectra will overestimate τ , for RB. The same condition will hold for other dyes subject to similar equilibria. As an alternative, τ_r can be calculated from the fluorescence quantum yield ϕ_f and the observed fluorescence lifetime τ , as $\tau_r = \tau/\phi_f$. Both τ and ϕ_f for RB are known to decrease markedly with increasing temperature [14-17], but quite recently JOHANSSON and NIEMI [17] have shown that τ_r , values calculated from the ratio $\tau/\phi_{\rm f}$ remain constant. This result was "unexpected" since τ_r values obtained from the absorption spectrum increased significantly over the same temperature range [17]. These observations are readily explained by the temperature dependence of the RB $L \rightleftharpoons Z$ equilibrium observed here. (Still unexplained is the observation [17] that the esterified RB analog octadecyl-RB displayed temperature-dependent changes similar to those of RB. We found only relatively minor temperature-dependent changes in the spectrum of the esterified dye rhodamine 6G (see below) [1].)

Examination of the trends found for ΔH° and ΔS° in different solvents (Tables 4 and 5) suggests that, in addition to factors previously identified, self-association of the alcohol solvent A strongly influences the position of the L \rightleftharpoons Z equilibrium. That is, the observed thermodynamic changes reflect not only the ability of the solvent to form solvent-dye hydrogen bonds, which stabilize the Z form, but also importantly the degree of self-association of the solvent itself. For the equilibrium

$$\mathbf{L} + \mathbf{A} \rightleftharpoons \mathbf{Z} \cdots \mathbf{A} \tag{17}$$

it follows that

Z

$$\mathbf{A}S^{\circ} = S^{\circ}(\mathbf{Z} \cdot \cdot \cdot \mathbf{A}) - S^{\circ}(\mathbf{L}) - S^{\circ}(\mathbf{A}).$$
(18)

If $S^{\circ}(Z \cdots A)$ and $S^{\circ}(L)$ remain sensibly constant from solvent to solvent, as may be expected, the solvent entropy term $S^{\circ}(A)$ will dominate changes in ΔS° . Therefore ΔS° should have its least negative values in solvents which are highly associated, e.g. water and ethanol, and its most negative values in less associated solvents, e.g. higher alcohols and secondary and tertiary alcohols. These are indeed the trends found in Tables 4 and 5, and the Z form is favored in the more associated solvents. A similar argument can be made for variations in ΔH° , since in associated alcohols solvent-solvent hydrogen bonds must on average be broken to form hydrogen bonds to the dye. This latter trend is also apparent in Tables 4 and 5, and to some extent counteracts the trend in ΔS° . However, the variations in ΔH° for these solvents are less severe and the entropy term tends to dominate the equilibrium.

The usual Arrhenius assumption of constant ΔH° and ΔS° over the temperature range examined is not a good assumption for this system. Plots of ln K vs 1/Tdisplay noticeable concave downward curvatures (Ref. [1], Fig. 8), and sizable changes in ΔH° and ΔS° toward more negative values take place with increased temperatures. As a rule relatively greater variations occur in the more associated solvents: e.g., based on Eqns (10) and (11), both $|\Delta S^{\circ}|$ and $|\Delta H^{\circ}|$ for *n*-pentanol increase by about 50%, but those for 2-pentanol increase by less than 30%, over the range 15°C to 55°C [4]. The same arguments used previously rationalize these trends. With increasing temperature the S°(A) term in Eqn. (18) increases significantly due to thermal disruption of the solvent structure, so that ΔS° becomes more negative. Solvents with more self association, i.e. more structure to be disrupted, are more affected by temperature changes.

The observed changes in ΔH° and ΔS° can be compared with changes calculated for the solvent alone over the 40°C temperature range studied. Taking $C_n = 209 \text{ J/mol} \cdot \text{K}$ for *n*-pentanol [18] at 25°C one estimates $\Delta H^{\circ} = 8.4 \text{ kJ}$ (observed, 10.5 kJ) and ΔS° = 27.2 J/mol·K (observed, 34.6 J/mol·K). Thus, by this estimate, solvent changes can account for roughly 80% of the apparent changes in these thermodynamic properties. However, we have observed that both the esterified dye rhodamine 6G [1], a presumed constant standard, and the RB cation (D. A. HINCKLEY, unpublished results) display small, unexplained decreases in absorbance with increased temperature. (These changes may involve reversible photobleaching [19, 20] or other effects at the elevated temperatures.) Similar alterations in the Z absorbance may exaggerate somewhat the changes in ΔH° and ΔS° obtained from Eqns (10) and (11). Therefore, solvent changes may in fact account for an even greater portion of the thermodynamic variations.

The present study of additional protic solvents and thermodynamic parameters has expanded the argument of Ref. [1] that hydrogen-bonding ability and solvent dielectric/polarizability are important factors influencing stabilization of the Z form of RB. Correlations of ln K with structural descriptors, dielectric constants and MCRAE's solvent terms (representing general polar and dispersion forces) improve markedly upon introduction of the steric hindrance parameter C_{α} [Eqns. (12)–(14)]. Moreover, C_{γ} enters these equations with a negative coefficient, consistently near -1.90. This suggests that the ability of the solvent hydroxyl group to form hydrogen bonds to Z, strongly reduced in secondary and tertiary alcohols, is an important factor. However, self-association of the solvent, also influenced by hydrogen bonding, also plays a strong role. Equation (12) allows the prediction of ln K at 25°C in aliphatic alcohol solvents to within about ± 0.4 . ΔH° and ΔS° at 25° can be predicted to within about $\pm 10\%$ by use of Eqns (15) and (16), given the alcohol's dielectric constant, refractive index, and C_x value.

Acknowledgements We thank Dr. RUBIN BATTINO for helpful discussions of this topic. We also thank the donors of the Petroleum Research Fund, administered by the American Chemical Society, for partial support of this work. The Varian 2300 spectrometer was purchased through NSF funding grant NSF # PRM8111214.

REFERENCES

- [1] D. A. HINCKLEY, P. G. SEYBOLD and D. BORRIS, Spectrochim. Acta 42A, 747 (1986).
- [2] P. W. ATKINS, *Physical Chemistry*, 2nd Edn, pp. 165, 175. W. H. Freeman, San Francisco, CA (1982).
- [3] E. G. MCRAE, J. Chem. Phys. 61, 562 (1957).
- [4] D. A. HINCKLEY, M.S. Thesis, Wright State University (1985).
- [5] J. FIGUERAS, J. Am. chem. Soc. 93, 3255 (1971).
- [6] L. M. MUKHERJEE and E. GRUNWALD, J. phys. Chem. 62, 1311 (1958).
- [7] H. C. ECKSTROM, J. E. BERGER and L. R. DAWSON, J. phys. Chem. 64, 1458 (1960).
- [8] W. J. MIDDLETON and R. V. LINDSAY JR., J. Am. chem. Soc. 86, 4948 (1964).
- [9] W. E. STRUVE and P. M. RENTZEPIS, Chem. Phys. Lett. 29, 23 (1974).
- [10] A. DECLEMY, C. RULLIERE and PH. KOTTIS, Chem. Phys. Lett. 133, 448 (1987).
- [11] I. ROSENTHAL, P. PERETZ and K. A. MUSZKAT, J. phys. Chem. 83, 350 (1979).
- [12] S. J. STRICKLER and R. A. BERG, J. Chem. Phys. 37, 814 (1962).
- [13] P. G. SEYBOLD, M. GOUTERMAN and J. CALLIS, Photochem. Photobiol. 9, 229 (1969).
- [14] J. FERGUSON and A. W.-H. MAU, Aust. J. Chem. 26, 1617 (1973).
- [15] T. KARSTENS and K. KOBS, J. phys. Chem. 84, 1871 (1980).
- [16] R. F. KUBIN and A. N. FLETCHER, J. Luminesc. 27, 455 (1982).
- [17] L. B.-A. JOHANSSON and A. NIEMI, J. phys. Chem. 91, 3020 (1987).
- [18] H. C. ZEGERS and G. SOMSEN, J. chem. Thermodyn. 16, 225 (1984).
- [19] M. KOIZUMI and Y. USUI, Molec. Photochem. 4, 57 (1972).
- [20] V. KRUGER and R. MEMMING, Ber. Bunsenges. Phys. Chem. 78, 679 (1974).