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Acid–base properties and solubility of pindolol, diazepam and chlordiazepoxide in SDS micelles

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Abstract

The effect of sodium dodecyl sulphate (SDS) on the acid-base properties and on the solubility of a β-blocker (pindolol) and of two benzodiazepines (diazepam and chlordiazepoxide) has been assessed. The study was performed by potentiometric and spectrophotometric determinations of the acidity constants and by spectrophotometric evaluation of the solubilities of the pharmaceutical drugs in aqueous solution and in solutions to which was added SDS with concentrations below and above the critical micelle concentration (cmc), at 25°C and at an ionic strength 0.1 M (NaCl). The effect of the organized assemblies on the pK_a values was quantified by the application of two theoretical models that differ in the inclusion of ionic exchange between positively charged species in solution. These models have allowed the determination of the binding constants for drug/micelle and yielded values in good agreement with those obtained by the solubility method, and in addition provide a more detailed picture of the effect of drug charge on its partition. The results can be taken to evidence different interaction modes of the drugs with the SDS micelles.

Keywords: Benzodiazepines; β-Blockers; SDS micelles; Acidity constants; Solubility

1. Introduction

Micellar media can dissolve substances with low solubility in water by incorporation of ions and molecules on the surface and/or within the organized assemblies, and thus modify acid-base or redox properties of the solutes. These aspects can be of interest in connection with the increasing use of aqueous micellar systems as solvents in analytical chemistry (Khaledi and Rodgers, 1990) and as biomembranar models (He et al., 1989).

Quantification of the effect of micelles in acid–base and solubilization properties of pharmaceutical drugs allows for the determination of binding constants drug/micelle. These constants can be determined if there is at least one molecular property that changes when the drug is transferred from water to the micellar medium. As this variation is related with changes in the microenviron-

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ment of the molecule undergoing solubilization, it can provide a detailed picture for partitioning of the neutral form of the drug in micellar pseudo-phase and of the interactions of the charged form with micelle surface.

In this work we report the behavior of a β-blocker (pindolol) and of two benzodiazepines (chlordiazepoxide and diazepam) in sodium dodecyl sulphate solutions (SDS) with concentrations below and above the cmc, at 25°C and at an ionic strength 0.1 M NaCl. The characterization of the several solution equilibria for this system drug/SDS was performed by potentiometry and spectrophotometry. Two models have been used to quantify micellar effects on equilibrium constants (Berezin et al., 1973; Pramauro and Pelizzetti, 1981) and the PIE model (Quina and Chaimovich, 1979). Their differences lies mainly in an assumption used by PIE model, that the counterions of SDS (Na^+) exchange with other cationic species in solution. We also used solubility data to differentiate the contribution of the distribution of the undissociated species of the drugs in the micellar pseudo-phase from their apparent increased ionization, to the enhanced solubility in micellar media. Application of these models have allowed the determination of binding constants and has been extremely helpful in choosing a good model for the description of the micelle role on the drug/micelle interactions.

2. Material and methods

2.1. Reagents and solutions

All compounds were used as received: pindolol from Sigma; sodium dodecyl sulphate (SDS) from Aldrich; diazepam and chlordiazepoxide were a gift from Hoffman-La Roche, and HCl (Titrisol) and all other chemical were from Merck (grade pro analysis). Solutions were prepared with double deionized water (conductivity less than 0.1 μS cm^{-1}. Solutions of the drugs were typically less than 10^{-4} M, thus assuring that no self-micellization took place, as their cmc are in the milimolar range. (Attwood et al., 1993)

2.2. Potentiometric determination of acidity constants

All potentiometric measurements were carried out with a Crison 2002 pH meter and 2031 buret controlled by a personal computer which was also used for data manipulation. The electrode assembly was made up of an Orion 900029/4 AgCl/Ag reference electrode and a Russel SWL glass electrode. System calibration was performed by Gran method (Gran, 1952) in terms of hydrogen ion concentration, using strong acid/strong base titration {HCl (0.001 M)/NaOH ($\approx$0.02M)} with solutions whose ionic strength was adjusted to 0.1 M with NaCl. Titrations were always carried out under a nitrogen atmosphere at 25°C in a double-walled glass cell.

Acidity constants for pindolol were obtained by titrating 20.00 ml of acidified solutions (1 mM HCl) of the β-blocker (0.8 mM), either in pure water or in aqueous solutions of methanol (10, 20, 30, 40 e 50% v/v methanol/water) or SDS (0.5, 1, 2, 2.5, 3, 4, 7, 10, 20 mM), with NaOH ($\approx$20 mM). All titrations were performed at 25°C under nitrogen, and for all solutions the ionic strength was adjusted to 0.1 M with NaCl.

System calibration was always performed before and after each determination by titrating HCl with NaOH, both for aqueous and micellar media; in the latter experiments the concentration of SDS was that of the titrated solution. The characteristics of the glass electrode were similar below and above critical micelle concentration (cmc), except that the response time was longer at high concentrations of SDS. Calculations were performed with data obtained from at least six independent titrations, each with more than 30 points, and the experimental titration data were analyzed using the computer program Superquad (Gans et al., 1985). The errors reported in this work were calculated by the method of Albert and Serjeant (Albert and Serjent, 1971), in which the errors are calculated as the maximum difference between the logarithm of the average of the antilogarithms of the calculated pK_a values and their individual values.

2.3. Spectrophotometric determination of acidity constants

All absorption spectra were recorded with a Hitachi U-2000 dual-beam spectrophotometer using quartz cells with 1 cm path length that were thermostated at 25°C. Acidity constants of the β-blocker and the benzodiazepines were obtained from UV data of solutions (aqueous and micellar media) of pindolol ($2.6 \times 10^{-5} - 5 \times 10^{-5}$ M), chlordiazepoxide and diazepam ($5 \times 10^{-5} - 1 \times 10^{-4}$ M), for which the ionic strength was adjusted to 0.1 M with NaCl. The SDS concentrations used were the same of the potentiometric studies, viz. 0.5, 1, 2, 2.5, 3, 4, 7, 10, 20 mM. Aliquots of strong base or strong acid were added to 20 ml of the stock solution to adjust pH ($-\log [H^+]$) to the desired value. The calculations were performed with the program SQUAD (Leggett and MacBryde, 1975) by using data from at least two independent experiments, each with more than 6 solutions, and in the range from 200 to 350 nm, at 2 nm intervals for pindolol and from 200 to 500 nm at 5 nm intervals for diazepam and chlordiazepoxide.

2.4. Solubility studies

Saturated solutions were prepared by dispersing excess drug in 25 ml of water and each one of the following aqueous solutions of SDS (5×10^{-4}, 1×10^{-3}, 2×10^{-3}, 3×10^{-3}, 4×10^{-3}, 7×10^{-3}, 1×10^{-2} e 2×10^{-2} M); in all solutions the ionic strength was adjusted to 0.1 M with NaCl. For each drug two set of solutions were prepared: one with pH $\approx$ p$K_a - 3$ and the other with pH $\approx$ p$K_a + 2$, to insure that practically all drugs are protonated/deprotonated. The dissolution was assisted with a vortex mixer and after 48 h at 25°C, clear saturated solutions were obtained by filtration through filter paper (Lida 0.45 μm). The concentration of each drug was determined spectrophotometrically at the wavelength of maximum absorbance, after the appropriate dilution with a solution with the same composition, but in which the drug was absent. In each case, the corresponding solvent system diluted in the same way as the measured filtrate was used to correct any absorbance of the surfactant and as a solvent to prepare working standard solutions of each drug studied for the construction of a calibration curve.

3. Results and discussion

3.1. Acidity constants in aqueous solution and below the cmc of SDS

The acidity constants of pindolol, diazepam and chlordiazepoxide, both in aqueous solution and in micellar media are included in Table 1. The pK_a values in aqueous solution for diazepam, chlordiazepoxide and pindolol are identical to those reported in the literature (Laxer et al., 1981; Pfendt et al., 1990; de Castro et al., 1993). The values for the pK_a of pindolol were determined by spectrophotometry and by titrimetry in water/methanol using published methods (de Castro et al., 1993). These latter results reveal that the pK_a of pindolol decreases with an increase in methanol content and the following equation was obtained: $pK_a^s = 9.52 - 0.011 \times (\%\text{methanol})$; $r^2 = 0.997$.

In SDS solutions below the cmc (1.4 mM) (Jones, 1995), it must be pointed out that the behavior of the drugs studied is markedly different, probably arising from different interactions of the drugs with the monomers, what hinders determination of acidity constants in this region. For chlordiazepoxide no determinations could be made below the cmc of SDS, as a new precipitate is formed, probably arising from an interaction with the monomer of SDS in acidic media. The spectra of chlordiazepoxide (5×10^{-5} M) in water and in SDS (4.0×10^{-4}, 1.4×10^{-3}, 2.0×10^{-3}, 8.0×10^{-3} M) in the acidic region show a decrease in absorbance as the concentration of SDS increases till the cmc; and for values above the cmc the absorbance remains unchanged and approximately identical to that in aqueous solution. In Fig. 1 are depicted the spectra of chlordiazepoxide at pH 3 and 9: in acidic media below the cmc the chlordiazepoxide exists in protonated form and an electrostatic interaction with the negatively charged monomers of SDS is likely to occur, and a reduction in absorbance can be due to the disappearance of free chlordiazepoxide. On

the other hand, alkaline solutions show only a slight decrease in absorbance, as the neutral form of chlordiazepoxide is not expected to interact strongly with the negatively charged monomer of SDS. For pindolol, as a precipitate is also formed in acidic media we propose that similar interactions must take place between the protonated from of pindolol and the negatively charged monomer of SDS (see below solubility section).

However, for diazepam, there is no problems with solubility and pK_a values were determined for an extended range of SDS concentrations. The pK_a values are practically constant below the cmc, raise abruptly near the cmc and stabilize to a value ≈ 4 in the presence of SDS micelles.

3.2. Acidity constants in the presence of micellar media

Analysis of the data in Table 1 shows that in SDS micellar media the pK_a values for the three substances are higher than in aqueous solution. This behavior has been observed for numerous indicators, for which at least one form is cationic, that interact with micelles of anionic surfactants, and the shift in the apparent pK_a is a measure of the strength of this interaction (Rychlovský and Nemcová, 1988; Khaledi and Rodgers, 1990; Pal and Jana, 1996). The maximum of the observed shifts were 0.47 for pindolol, 0.43 for diazepam and 1.3 for chlordiazepoxide. The shifts for pindolol and for chlordiazepoxide are in the range reported for other β-blockers and benzodiazepines (de Castro et al., 1998); but the shift for diazepam is much smaller than expected.

3.2.1. Diazepam

The dependence of pK_a values for diazepam with SDS concentration is also different from that of the other drugs studied: for SDS concentrations below 8.0×10^{-4} M they are identical to that in aqueous solution, but start to raise abruptly above this concentration and above 1.0×10^{-3} M are practically independent of SDS concentration. As has been observed for the pK_a dependence of several indicators with concentration of anionic surfactants, this abrupt change occurs near the cmc of the surfactant, as is also found for the other drugs studied. This observation suggests that the cmc of SDS in the presence of diazepam is reduced from 1.4×10^{-3} M to about 9.0×10^{-4} M. Probably, diazepam is incorporated in the SDS framework with formation of mixed micelles, for which the cmc is different from that of SDS. Additional support for this hypothesis can be gathered by noting the ba-

Table 1
Acidity constants (pK_{app}) of pindolol, diazepam and chlorodiazepoxide, in aqueous solution and in SDS, obtained by potentiometry or spectrophotometry at 25 °C and $I = 0.1$ M in NaCl

SDS (M)	Pindolol		Diazepam	Clorodiazepoxide
	Potentiometry	Spectrophotometry	Spectrophotometry	Spectrophotometry
0	9.50 ± 0.02	9.46 ± 0.03	3.58 ± 0.06	4.82 ± 0.03
4.0×10^{-4}	–	–	3.53 ± 0.02	–
8.0×10^{-4}	–	–	3.57 ± 0.04	–
9.0×10^{-4}	–	–	3.72 ± 0.04	–
1.5×10^{-3}	–	–	3.93 ± 0.02	5.37 ± 0.08
2.0×10^{-3}	9.48 ± 0.03	–	3.96 ± 0.02	5.59 ± 0.01
2.5×10^{-3}	–	–	–	–
3.0×10^{-3}	9.63 ± 0.04	9.60 ± 0.02	–	5.88 ± 0.01
4.0×10^{-3}	9.69 ± 0.04	–	3.98 ± 0.01	6.01 ± 0.01
7.0×10^{-3}	9.85 ± 0.03	9.84 ± 0.02	–	–
1.0×10^{-2}	9.96 ± 0.05	–	4.01 ± 0.05	6.02 ± 0.01
2.0×10^{-2}	9.97 ± 0.03	9.93 ± 0.03	–	6.06 ± 0.01
5.0×10^{-2}	–	–	–	6.14 ± 0.03

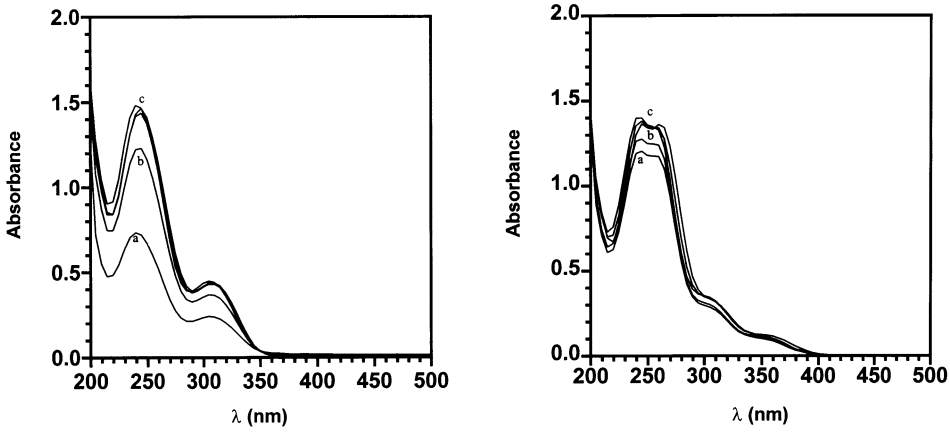

Fig. 1. Absorption spectra of chlordiazepoxide (4.985×10^{-5} M) in 0.1 M NaCl aqueous solution and different SDS concentrations: (A) pH 3; (B) pH 9; both with a = 4.0×10^{-4} M; b = 1.4×10^{-3} M; c > 2.0×10^{-3} M and H_2O.

tochromic shift observed for λ_{max} when diazepam changes from aqueous to micellar solutions of SDS (Pramauro and Pelizzetti, 1996). Our results are in disagreement with those in the literature (Cirugeda and Soriano, 1989) that report no changes in the cmc of SDS in the presence of diazepam and no changes in the apparent pK_a of the drug. We believe that their conclusions are due to experimental limitations: their pK_a values are affected by a reported error of ± 0.1 and the methods used to determine the cmc, measurement of surface tension, is known to be insensible to small changes in this quantity (Mukerjee and Mysels, 1971).

3.2.2. Pindolol and chlordiazepoxide

For these drugs no changes in the wavelengths of maximum absorption were detected upon addition of SDS at the concentrations used, thus suggesting that incorporation of the drugs into the micelles is unlikely and that the interactions with the SDS molecules must take place at the micellar surface. Two models were used to quantify the effect of the organized assemblies on the acidity constants of these drugs in SDS micelles—that of Berezin (Pramauro and Pelizzetti, 1981) and the pseudophase ion exchange (PIE) (Quina and Chaimovich, 1979). The main difference between these two models lies in the latter incorporates ionic exchange between the counter-ion of the surfactant, in the present case Na^+, and other protonated species in solution, H^+ and HB^+ (the protonated form of pindolol or chlordiazepoxide). Within the framework of the PIE model, the equilibrium of these drugs in anionic micellar media (for which the monomer is represented by Y^+S^-) is described by the following set of equilibria that characterize their dissociation and the partition coefficients of the protonated and neutral species between the micelles and the solution:

$$HB_f^+ \rightleftharpoons H_f^+ + B_f \qquad K_a = [B]_f[H^+]_f/ = [HB^+]_f \tag{1}$$

$$B_f \rightleftharpoons B_b \qquad K_B^m = [B]_b/[B]_f C_D \tag{2}$$

$$HB_f^+ + Y_b \rightleftharpoons Y_f + HB_b^+$$

$$K_{HB^+/Y}^m = [HB^+]_b[Y]_f/[HB^+]_f[Y]_b \tag{3}$$

$$H_f^+ + Y_b \rightleftharpoons H_b^+ + Y_f$$

$$K_{H^+/Y} = [H^+]_b[Y]_f/[H^+]_f[Y]_b \tag{4}$$

$$B_b + H_b^+ \rightleftharpoons HB_b^+$$

$$K_{a,b} = [HB^+]_b C_D/[B]_b[H^+]_b \tag{5}$$

in which the subscript f stands for free species in solution and b for micelle bound species, and C_D is the micellized detergent concentration, (equal to the difference between the total detergent concentration, C_T, and the critical micellar concentration, cmc, i.e. $C_D = C_T - \text{cmc}$). The model developed by Berezin and co-workers (Pramauro and Pelizzetti, 1981) uses only equilibria 1, 2 and

5, thus excluding ion exchange between the counter-ion and other positively charged species.

As both drugs are weak acids in solution, both models assume that the dissociation constant for equilibrium $HB^+ \rightleftharpoons H^+ + B$, in the presence of surfactants (pK_{app}) is given by

$$K_{app} = \frac{([B]_f + [B]_b)}{([HB^+]_f + [HB^+]_b)} [H^+]_f \quad (6)$$

and the value of pK_{app} is the intermicellar pH ($pH = -\log [H^+]_f$) when $[B]_f + [B]_b = [HB^+]_f + [HB^+]_b$ (Quina and Chaimovich, 1979).

In the Berezin model, the apparent dissociation constant for an acid HB^+ is related to the dissociation constant in water (K_a) by the following expression (Pramauro and Pelizzetti, 1981)

$$K_{app} = [H^+]_f = K_a \frac{(1 + K_B^m C_D)}{(1 + K_{HB+}^m C_D)} \quad (7)$$

where and are the binding constants to the micellar pseudo-phase, for the protonated and neutral forms of the substrate.

Taking into account ion exchange, Eqs. (3) and (4), the expression derived for pK_{app} within the framework of the PIE model is:

$$K_{app} = [H^+]_f = K_a \frac{(1 + K_B^m C_D)}{(1 + K_{HB+/Y}^m (Y_b / Y_f)} \quad (8)$$

where $Y_b = (1 - \alpha)\ C_D - W$ and $Y_f = \alpha C_D + \text{cmc} + W + [BY]_T$, with $W = [H^+]_b + [HB^+]_b$. On applying both models we have used the optimum value of $\alpha = 0.75$ for the ionization degree of the anionic micelles (Quina and Chaimovich, 1979; He et al., 1989) and a value of 1.4 mM for the cmc of SDS in 0.1 M NaCl (Jones, 1995).

Application of Berezin model to our data, but with Eq. (7) rewritten as

$$\frac{\left(1 - \frac{K_{app}}{K_a}\right)}{C_D} = K_{HB+}^m \frac{K_{app}}{K_a} - K_B^m \quad (9)$$

provides a good test to assess the applicability of this simple model. For chlordiazepoxide, plots from Eq. (9) yields a straight line, with slope 1.2×10^{-4} and intercept (K_{B+}^m) 5.18×10^{-2} ($R^2 = 0.975$). The data for pindolol could not be fitted by Eq. (9), thus indicating that this simple model is inadequate to describe the acid-base properties in micellar media As it is clear from our data, the linearized form of Eq. (9) is a useful screening test to determine if this model can be used to describe the effect of micelles on acid-base properties, but as it gives a high weight to the low concentrations points we have fitted Eq. (7) directly to our data and the results are presented in Table 2.

We have also applied the PIE model to our data and obtained good fits for chlordiazepoxide and pindolol and the results are presented in Table 2. In accordance with the results outlined above, as the data for chlordiazepoxide are fitted to an equation with more parameters a better fit is always to be expected; however, as the calculated

Table 2
Binding constants to micelles of protonated and non-protonated forms of the pindolol and chlorodiazepoxide, with and without ionic exchange of the counterions of SDS with other cationic species in solution

Drug	Without exchange				With exchange			
	K_B^m [a]	K_{HB+}^m [a]	R^2		K_B^m [a]	$K_{HB+/Y}^m$ [a]	W [c]	R^2
Pindolol	3	242	0.929		103	208	9×10^{-5}	0.992
Chlorodiazepoxide	518	12283	0.975		226	1760	-15×10^{-5}	0.981
	K_B^m [b]	R^2	K_{HB+}^m [b]	R^2				
Pindolol	103	0.991	158	0.992				
Chlorodiazepoxide	600	0.985	–	–				

[a] Values determined from pK_a shifts.
[b] Values determined from solubility measurements.
[c] $W = [H^+]_b + [HB^+]_b$, see text.

Table 3
Solubility (M) of the pindolol and chlorodiazepoxide in SDS solutions

SDS (M)	Pindolol		Chlorodiazepoxide
	S (pH = 5)	S (pH = 11)	S (pH = 9)
0	2.44×10^{-3}	1.31×10^{-4}	–
5×10^{-4}	4.92×10^{-3}	1.69×10^{-4}	3.16×10^{-4}
1×10^{-3}	7.12×10^{-3}	1.94×10^{-4}	–
2×10^{-3}	8.82×10^{-3}	2.11×10^{-4}	–
3×10^{-3}	9.97×10^{-3}	2.29×10^{-4}	–
4×10^{-3}	1.26×10^{-2}	2.59×10^{-4}	5.18×10^{-4}
7×10^{-3}	1.52×10^{-2}	3.24×10^{-4}	9.67×10^{-4}
1×10^{-2}	1.92×10^{-2}	4.05×10^{-4}	1.14×10^{-4}
2×10^{-2}	1.98×10^{-2}	6.66×10^{-4}	2.08×10^{-4}

amount of Na^+ exchanged with the other cationic species in solution is negative, the applicability of this model is precluded on chemical grounds. In fact, as Na^+ exchanges with and, the concentration of exchanged Na^+, W, must be equal to the concentration of bound H^+ and HB^+, $W=[H^+]_b+[HB^+]_b$, and this quantity was found to be negative when the PIE model is applied to chlordiazepoxide, an impossible result as bound concentrations must be non-negative.

3.3. Solubility data

Changes in solubility of weak acids in micellar media can be due to (1) partitioning of the neutral form of the acid in the micellar pseudo-phase or (2) interactions with the micellar surface, that increase their pK_{app} values (Gerakis et al., 1993). To distinguish between these processes, we have determined the solubility of all drugs in (1) aqueous solution, both in acidic (S_w,H^+) and alkaline (S_w,OH^-) conditions, and in (2) micellar solutions, again in acidic (S_m,H^+) and alkaline (S_m,OH^-) conditions. An increase in the ratio (S_m,OH^-)/(S_w,OH^-) is taken as to suggest an increase in partition of the neutral molecules in the micellar pseudo-phase, whereas an increase in the ratio (S_m,H^+)/(S_w,H^+) an increase in apparent ionization. Data for the solubilities of pindolol and chlordiazepoxide in different media are presented in Table 3, no data is presented for diazepam as it is hydrolyzed before the 48 h used to incubate the solutions (de Castro et al., 1993). Some remarks have to be made concerning the solubility of chlordiazepoxide in acidic media in all attempts to determine solubilities, a yellow precipitate was formed thus precluding its determination. On the basis of the large binding constants chlordiazepoxide/micelle, on the formation of an identical precipitate at very low concentrations of SDS and on the decrease on the absorbance bands of chlordiazepoxide, we propose this precipitate to be a complex between the positively charged form of the drug with SDS.

The binding constant of any substance to a micelle is defined by the general expression $Kmm=(S_m-S_w)/S_wC_D$ (Berezin et al., 1973; Pramauro and Pelizzetti, 1996), and this equation applied to the drugs studied yields $S_m/S_w=1+K_B^mC_D$ for neutral molecules, and $S_m/S_w=1+K_{HB+}^mC_D$ for the protonated form. These latter expressions are valid only for concentrations near the cmc, as the values of S_m/S_w level off at high concentrations of surfactant. For the neutral form of chlordiazepoxide (alkaline media) this model yielded $K_B^m=600$, in good agreement with the value obtained from the Berezin model.

Application of these equations to pindolol requires an additional approximation, as this drug interact with the monomer of SDS (see above) and the value of solubility in water changes with the concentration of SDS, thus making the ratio $S_m/S_w\neq1$, when C_D is zero. We have used instead the value of the solubility at the cmc (S_{cmc}) for S_w and obtained good fits to our data (Fig. 2), and the values obtained for K_B^m (103) and K_{HB+}^m (158) are in good agreement with those from the PIE model.

4. Concluding remarks

Diazepam in micellar media behaves differently from the other drugs as the pK_a shifts are much smaller than expected for benzodiazepines (de Castro et al., 1998), what could be taken to presuppose weaker binding to the micelle surface. However, by noting that the observed batochromic shift on going from water to micellar media parallels a similar shift on going from

water to non polar solvents, it can be argued that in both cases a low-polar environment exists for the drug, thus suggesting that some diazepam must lie inside the micelle, thus not affecting the concentration of free H^+ and explaining the small shifts in pK_a.

For pindolol and chlordiazepoxide, it is clear that the binding constants for neutral species are always smaller than those of positively charged (protonated) species and, as observed for others β-blockers and benzodiazepines, the neutral form of pindolol binds more weakly than the corresponding form of chlordiazepoxide. These observations correlate with the known higher hydrophobic properties of benzodiazepines and provide support for stronger hydrophobic interactions of the non-protonated form of these molecules with the lipophilic side-chains of SDS. Furthermore, as the binding of protonated benzodiazepines is also much stronger than for protonated β-blockers, and as no physically realistic model can invoke ionic exchange in the binding of benzodiazepines we suggest that these drugs, as a consequence of their larger lipophilicity, must penetrate the micelles to a larger extent than the β-blockers and be less prone to exchange with the main solution.

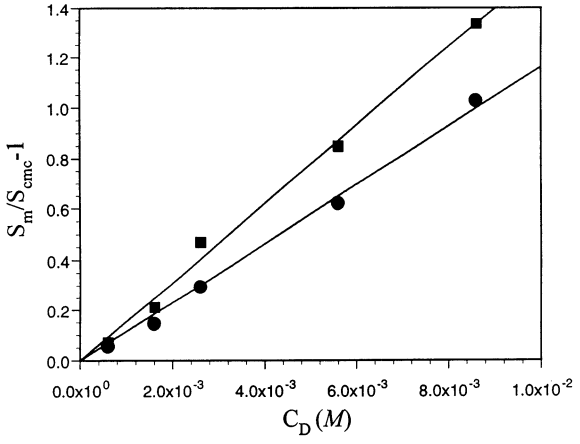

Fig. 2. Graphical representation of $S_m/S_{cmc} - 1$ vs C_D for pindolol.

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References

Albert, A., Serjent, E.P., 1971. The Determination of Ionization Constants, 2nd edition. Chapman and Hall, London.

Attwood, D., Blundell, R., Mosquera, V., Garcia, M., 1993. Association and surface properties of amphiphilic benzodiazepine and benzothiozepine drugs in aqueous solution. J. Coll. Interface. Sci. 161, 19–23.

Berezin, I.V., Martinek, K., Yatsimirskii, A.K., 1973. Physicochemical foundations of micellar catalysis. Russian Chem. Rev. 42, 787–802.

de Castro, B., Gameiro, P., Lima, J.L.F.C., 1993. Determination of the pK_a values of sparingly soluble substances in water revisited: application to some benzodiazepines. Anal. Chim. Acta 281, 53–62.

de Castro, B., Gameiro, P., Guimarães, C., Lima, J.L.F.C., Reis, S., 1998. Acid/base properties of β-blockers and benzodiazepines in sodium dodecyl sulphate micelles. A spectrophotometric and potentiometric study. J. Pharm. Sci. 87, 356–359.

Cirugeda, M.G., Soriano, F.R., 1989. Micellar enhancement of benzodiazepine fluorescence. Analyst 114, 77–82.

Gans, P., Sabatini, A., Vacca, A., 1985. Superquad: an improved general program for computation of formation constants from potentiometric data. J. Chem. Soc. Dalton Trans., 1195–1200.

Gerakis, A.M., Koupparis, M.A., Efstathiou, C.E., 1993. Micellar acid-base potentiometric titrations. J. Pharm. Biomed. Anal. 11, 33–41.

Gran, G., 1952. Determination of the equivalence point in potentiometric titrations. Analyst 77, 661–671.

He, Z.M., O'Connor, P.J., Romsted, L.S., Zanette, D., 1989. Specific counterion effect on indicator equilibria in micellar solutions of decyl phosphate and lauryl sulfate surfactants. J. Phys. Chem. 93, 4219–4226.

Jones, M.N., 1995. Micelles, Monolayers and Biomenbranes, Chapman, Wiley-Liss, New York, pp. 68.

Khaledi, M.G., Rodgers, A.H., 1990. Micellar-mediated shifts of ionization constants of amino acids and peptides. Anal. Chim. Acta 239, 121–128.

Laxer, M., Capomacchia, A.C., Hardee, G.E., 1981. Intramolecular hydrogen-bonding in beta adrenergic blocking agents. Talanta 28, 973–976.

Leggett, D.J., MacBryde, W.A.E., 1975. General computer program for the computation of stability constants from absorbance data. Anal. Chem. 47, 1065–1070.

Mukerjee, P., Mysels, K.J., 1971. Critical Micelle Concentrations of Aqueous Surfactant Systems. National Bureau of Standards, Washington.

Pal, T., Jana, N.R., 1996. Polarity dependent positional shift of probe in a micellar environment. Langmuir 12, 3114–3121.

Pfendt, L.B., Sladic, D.M., Janjié, T.J., Popović, G.V., 1990. Study of heterogeneous equilibria in saturated aqueous solutions of some 7-chloro-1,4-benzodiazepines. Analyst 115, 383–385.

Pramauro, E., Pelizzetti, E., 1981. Effect of micellar systems on the equilibrium of chemical reactions. Anal. Chim. Acta 126, 253–257.

Pramauro, E., Pelizzetti, E., 1996. In: Weber, S.G. (Ed.), Comprehensive Analytical Chemistry. Wilson & Wilson's XXXI. Surfactants in analytical chemistry. Applications of organized amphiphilic media. Elsevier, Amsterdam, pp. 203–215.

Quina, F.H., Chaimovich, H., 1979. Ion Exchange in micellar solutions. 1. Conceptual framework for ion exchange in micellar solutions. J. Phys. Chem. 83, 1844–1850.

Rychlovský, P., Nemcová, I., 1988. The effect of surfactants on the dissociation constants of phenothiazine derivatives. Alkalimetric determination of diethazine and chlorpromazine. Talanta 35, 211–214.