J. Pharm. Pharmacol. 1980, 32: 761–765 Received May 9, 1980

Self-association of analgesics in aqueous solution: association models for codeine, oxycodone, ethylmorphine and pethidine

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Light scattering, vapour pressure osmometry, conductivity and surface tension techniques have been used to examine aqueous solutions of several narcotic analgesics for evidence of association. Contrary to a previous report, no significant association could be detected in solutions of morphine sulphate and codeine phosphate. Other drugs which showed no evidence of aggregation in water included morphine hydrochloride, ethylmorphine hydrochloride, oxycodone hydrochloride and dihydrocodeine tartrate. Self-association of ethylmorphine hydrochloride, oxycodone hydrochloride and codeine phosphate was observed in the presence of 0·5 mol dm^{-3} electrolyte, the pattern of association conforming to that of a stepwise association process with all association constants of equal value. The association of pethidine hydrochloride in 0·5 mol dm^{-3} sodium chloride could be represented by an association scheme in which association constants K_N increased sequentially with aggregation number N according to the relation $K_N = K(N - 1)/N$.

We have previously shown (Attwood & Tolley 1980) that the self association of two analgesics, dextropropoxyphene hydrochloride and methadone hydrochloride conforms to a micellar pattern. Association commenced at clearly defined critical micelle concentrations (cmc) and agreement between cmcs determined by several varied techniques was reasonable. In this paper we report an investigation of the solution properties of a series of narcotic analgesics, both in the presence and absence of electrolyte. Perrin & Ishag (1971) have examined solutions of salts of morphine, codeine and hydromorphine using conductivity and optical rotatory dispersion (ORD) techniques and have presented evidence of association in the absence of added electrolyte. Cmc values are quoted based on apparent inflections in graphs of conductivity and ORD as a function of concentration. There is an inconsistency in their paper which leads to some uncertainty as to the intended cmc values. Inflection points indicated on the conductivity graphs are an order of magnitude lower than tabulated values. However, agreement between inflections in the ORD data and the tabulated values (0·32 g/100 g of morphine sulphate is equivalent to $4{\cdot}35 \times 10^{-3}$ mol dm^{-3}) suggests an error in the concentration axis of the conductivity plot. On this assumption the cmc values were taken to be $4{\cdot}35 \times 10^{-3}$ and $8{\cdot}61 \times 10^{-3}$ mol dm^{-3} for morphine sulphate and codeine phosphate respectively. The surface activity of morphine and codeine salts has been reported by Sliwa (1961).

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MATERIALS AND METHODS

Materials

The following drugs were used as received, morphine sulphate B.P; morphine hydrochloride B.P; ethylmorphine hydrochloride Eur. P; codeine phosphate B.P. (Macfarlan Smith); dihydrocodeine tartrate B.P. (Allen & Hanburys); and pethidine hydrochloride B.P. (Roche Products). Oxycodone hydrochloride was prepared from oxycodone base (Boots Company). The product was recrystallized from absolute ethanol and dried over phosphorous pentoxide under vacuum. [Found: C, 60·5; H, 6·7; N, 3·7. Calculated for $C_{18}H_{21}NO_4$ HCl: C, 61·5; H, 6·3; N, 4·0].

Sodium chloride and sodium tartrate were of Analar grade, sodium dihydrogen phosphate was reagent grade.

Water for surface tension and conductivity measurements was distilled from alkaline permanganate in an all-glass still.

Methods

Light scattering, surface tension, vapour pressure osmometry and conductivity measurements were performed at 303 K as described in the previous paper. Refractive index increments were morphine hydrochloride, 0·064; morphine sulphate 0·127; ethylmorphine hydrochloride, 0·066; oxycodone

hydrochloride, 0·063; codeine phosphate, 0·066, dihydrocodeine tartrate, 0·074 and pethidine hydrochloride 0·048 kg mol^{-1}.

RESULTS

Fig. 1 shows plots of the light scattering ratio, S_{90}, as a function of the solution concentration, m. With the exception of pethidine hydrochloride, the light scattering data for all the drugs in water could be described by the respective theoretical scattering lines, derived assuming ideality, for unassociated monomers. In view of the previously reported cmc

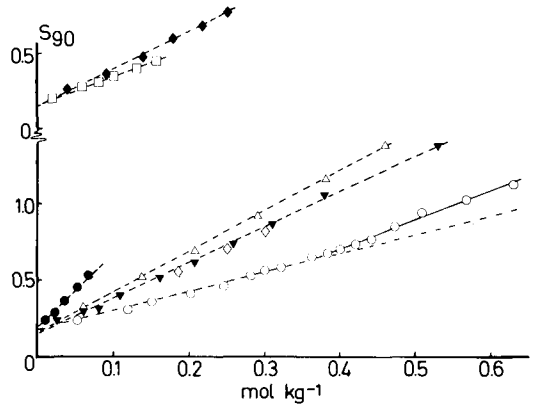

FIG. 1. Variation of the light scattering ratio, S_{90}, (ordinate) with concentration (abscissa: mol kg^{-1}) for ●, morphine sulphate; □, morphine hydrochloride; ◆, ethylmorphine hydrochloride; ▼, codeine phosphate; ◇, oxycodone hydrochloride; △, dihydrocodeine tartrate and ○, pethidine hydrochloride in water (– – –) theoretical light scattering from unassociated monomers.

values (Perrin & Ishag 1971) for morphine sulphate and codeine phosphate, this unexpected result was examined in greater detail using a variety of techniques. With refractive index increments of the magnitude of those of the drugs under investigation and at the concentration ranges examined, the light scattering technique is sufficiently sensitive to detect the formation of even small aggregates, indeed the deviation from the monomer line for pethidine hydrochloride may be described in terms of a monomer-dimer equilibrium.

Conductivity data plotted both as conductivity against concentration and, as in Fig. 2, as molar conductivity against (concentration)$^{\frac{1}{2}}$ showed no detectable inflection points for any of the drugs even though, in the case of morphine sulphate and codeine phosphate, measurements were made over a concentration range which included the reported cmcs.

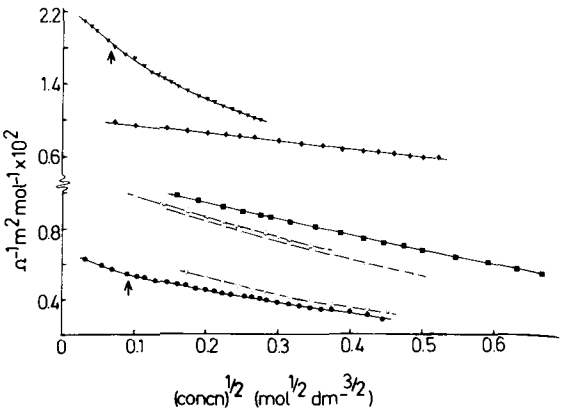

FIG. 2. Molar conductivity, Λ, (ordinate: Ω^{-1} m^2 mol^{-1} × 10^2) of aqueous solutions of ▼, morphine sulphate: ◇, morphine hydrochloride; □, ethyl morphine hydrochloride; ●, codeine phosphate; ◆, oxycodone hydrochloride; ▽, dihydrocodeine tartrate and ■, pethidine hydrochloride. Abscissa: (concentration)$^{\frac{1}{2}}$ (mol$^{\frac{1}{2}}$ dm$^{-3/2}$). Arrows indicate apparent cmc values reported by Perrin & Ishag (1971).

Similarly, no inflection attributable to a cmc was noted in the plots of surface tension against log m for morphine sulphate and codeine phosphate in water (see Fig. 3).

Vapour pressure measurements on solutions of morphine sulphate and codeine phosphate are presented in Fig. 4 as plots of V/c against c where V is the bridge output voltage and c is the weight concentration of the solution. Molecular weights of

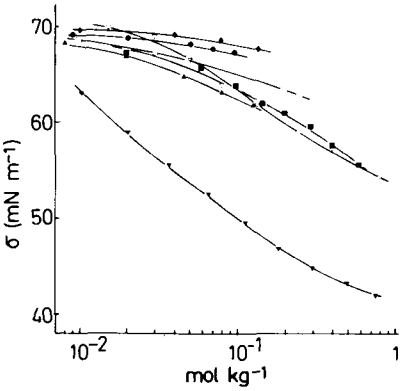

FIG. 3. Surface tension, σ (ordinate: mN m^{-1}) as a function of log molal concentration for aqueous solutions of ●, morphine sulphate; ◆, morphine hydrochloride and ▽, codeine phosphate in water and ▲, ethylmorphine hydrochloride; □, oxycodone hydrochloride; ▼, pethidine hydrochloride in 0·5 mol dm^{-3} sodium chloride; ■, codeine phosphate in 0·5 mol dm^{-3} sodium phosphate. Abscissa: concentration (mol kg^{-1}).

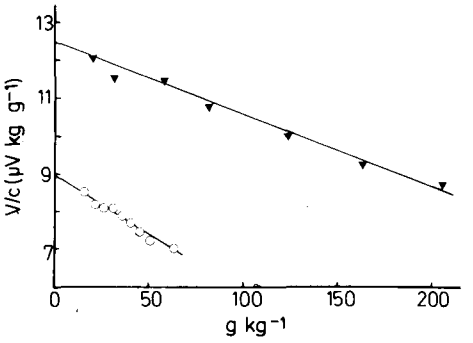

FIG. 4. V/c (ordinate: μV kg g^{-1}) against concentration (abscissa: g kg^{-1}) for ○, morphine sulphate and ▼, codeine phosphate.

847 and 396 were calculated from the values of $(V/c)_{c=0}$ for morphine sulphate and codeine phosphate respectively assuming these electrolytes to be completely ionized at infinite dilution. The lack of inflection in the vapour pressure plots and the agreement between the extrapolated molecular weights and those of the respective monomeric species further supports the conclusion that these drugs do not associate to any significant extent in aqueous solution.

In the presence of 0·5 mol dm^{-3} sodium chloride, the light scattering plots for oxycodone hydrochloride, ethylmorphine hydrochloride and pethidine hydrochloride deviate markedly from the respective monomer lines indicating appreciable association. A similar tendency was noted for codeine phosphate in 0·5 mol dm^{-3} sodium dihydrogen phosphate (Fig. 5). No discontinuities in the concentration dependence of S_{90} attributable to a cmc were apparent, suggesting a non-micellar pattern of association. This conclusion is supported by a similar lack of inflection in the surface tension plots for these compounds (Fig. 3). No appreciable association was noted for dihydrocodeine tartrate in 0·5 mol dm^{-3} electrolyte. The morphine salts were too insoluble in electrolyte to allow measurement.

Association models

The analysis of the light scattering data in terms of stepwise association models requires the knowledge of the variation of monomer concentration, $[b_1]$, with total solution concentration. This relationship may be determined by graphical integration of the light scattering data according to (Steiner 1952)

$$\ln x = \int_0^c [(M/M_{app}) - 1]\, d\ln c$$

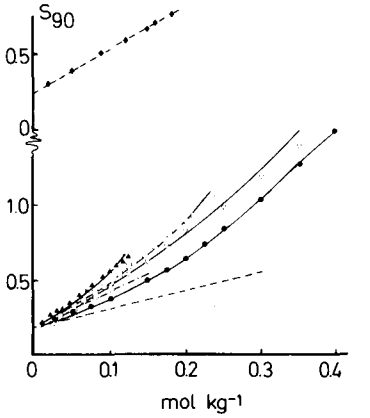

FIG. 5. Variation of the light scattering ratio, S_{90}, (ordinate) with concentration (abscissa: mol kg^{-1}) for ▲, ethylmorphine hydrochloride; □, oxycodone hydrochloride; and ●, pethidine hydrochloride in 0·5 mol dm^{-3} sodium chloride; ▽, codeine phosphate in 0·5 mol dm^{-3} sodium phosphate and ◆, dihydrocodeine tartrate in 0·5 mol dm^{-3} sodium tartrate. (——) theoretical scattering calculated using stepwise association models given in Table 1. (— · — · —) theoretical scattering from unassociated monomers for phenanthrene derivatives. (— — —) theoretical scattering from unassociated monomers of pethidine hydrochloride and dihydrocodeine tartrate.

where x is the weight fraction of monomers and M is the monomer molecular weight. The apparent weight-average molecular weight, M_{app}, was calculated from the light scattering intensity assuming ideality. Fig. 6 shows the variation of monomer concentration ($[b_1] = xc/M$) with total solution concentration, so obtained.

The equilibrium constant, β_N, for the self association reaction

$$N\ b_1 \rightleftharpoons b_N$$

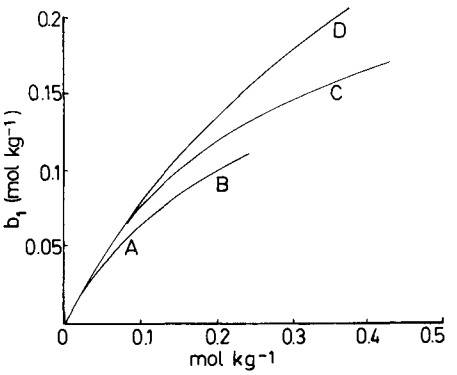

FIG. 6. Variation of monomer concentration $[b_1]$ (ordinate: mol kg^{-1}) with total solution concentration (abscissa: mol kg^{-1}) for A, ethylmorphine hydrochloride; B, oxycodone hydrochloride; C, pethidine hydrochloride and D, codeine phosphate in 0·5 mol dm^{-3} electrolyte.

in which multimer, b_N, is formed from N monomers, b_1, is expressed as

$$\beta_N = [b_N]/[b_1]^N \quad .. \quad .. \quad (1)$$

Equation (1) is not directly applicable to charged aggregates such as are present here. The equilibrium constant, β_N, derived from this equation incorporates charge effects which are assumed to remain constant with increase in aggregate size. This assumption is most likely to be valid for systems containing added electrolyte (Mukerjee 1972).

The stepwise association constant for the reaction

$$b_{N-1} + b_1 = b_N$$

is defined as K_N and hence

$$\beta_N = \prod_2^N K_N \quad .. \quad .. \quad (2)$$

where $\prod_2^N K_N = K_2K_3 \ldots K_N$

Several models of self-association were considered in which explicit relationships between all stepwise association constants were assumed, the relationships being expressed using a generalized parameter K. Both cooperative and anticooperative models were considered.

Model 1. This model assumes the equality of all K values i.e. $K_2 = K_3 = K_N = K$, leading to the relation (Ghosh & Mukerjee 1970)

$$([b_1]/m)^{\frac{1}{2}} = 1 - K[b_1] \quad .. \quad (3)$$

Model 2. Stepwise association constants increase sequentially with N according to $K_N = K(N-1)/N$ i.e. $K_2 = K/2$, $K_3 = 2K/3$ etc. This model leads to the relation (Ghosh & Mukerjee 1970)

$$(m/[b_1]) = 1 + Km \quad .. \quad .. \quad (4)$$

Model 3. Stepwise association constants decrease sequentially with N according to $K_N = K/N$ i.e. $K_2 = K/2$, $K_3 = K/3$ etc. This model which has not previously been examined may be treated as follows. The total equivalent concentration of all species, B, is given by

$$B = \sum_1^N N[b_N] = [b_1] + 2[b_2] + 3[b_3] + \ldots N[b_N] \quad (5)$$

From equations 1 and 2

$$B = [b_1] + 2K_2[b_1]^2 + 3K_2K_3[b_1]^3 + \ldots N\prod_2^N K_N[b_1]^N \quad (6)$$

substituting $K_N = K/N$ gives

$$B = [b_1]\left(1 + X + \frac{X^2}{2!} + \frac{X^3}{3!} + \ldots \frac{X^N}{N!}\right) \quad .. \quad (7)$$

where $X = K[b_1]$

thus $B = Xe^X/K \quad .. \quad .. \quad .. \quad .. \quad (8)$

Similarly a quantity G may be defined

$$G = \sum_1^N N^2[b_N] = [b_1] + 4[b_2] + 9[b_3] + \ldots N^2[b_N] \quad (9)$$

$$= [b_1](1 + 2X + 3/2X^2 + 2/3X^3 \ldots) \quad .. \quad (10)$$

$$= Xe^X(1 + X)/K$$

The weight-average degree of association including monomers, N_W, is thus

$$N_W = G/B = 1 + K[b_1] \quad .. \quad (11)$$

Figs 7, 8 and 9 show the experimental light scattering data plotted according to equations (3), (4) and (11). The best fit of data for ethylmorphine hydrochloride, oxycodone hydrochloride and codeine phosphate is achieved using model 1; whilst pethidine hydrochloride is best fitted with Model 2. K values calculated from the slopes of these plots are given in Table 1. Values of the association constants were used to recalculate the light scattering plots in the following way.

The weight-average degree of association for a system conforming to Model 1 is (Mukerjee 1974)

$$N_W = (1 + K[b_1])/(1 - K[b_1]) \quad .. \quad (12)$$

For an associating system conforming to Model 2, summation of the series for B and G leads to (Attwood et al 1980)

$$N_W = 1/(1 - K[b_1]) \quad .. \quad .. \quad (13)$$

The scattering intensity was calculated as a function of concentration from the values of N_W for the appropriate association model assuming ideality. Fig. 5 shows a satisfactory fit of experimental data

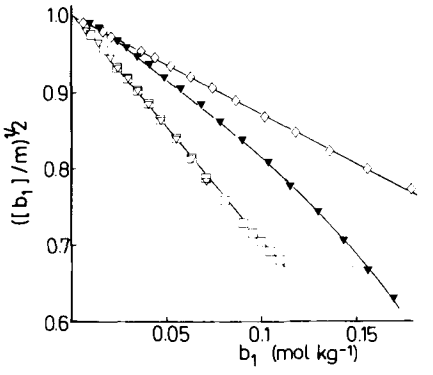

FIG. 7. Data plotted according to eqn 3 (Model 1) for ▽, ethylmorphine hydrochloride; □, oxycodone hydrochloride; ▼, pethidine hydrochloride; and ◇, codeine phosphate in 0·5 mol dm^{-3} electrolyte. Ordinate: $([b_1]/m)^{\frac{1}{2}}$. Abscissa: $[b_1]$ (mol kg^{-1}).

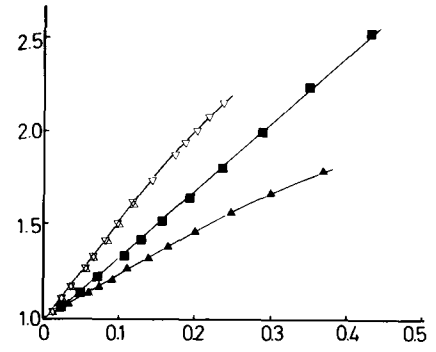

FIG. 8. Data plotted according to eqn 4 (Model 2) for △, ethylmorphine hydrochloride; ▽, oxycodone hydrochloride; ■, pethidine hydrochloride and ▲, codeine phosphate in 0·5 mol dm^{-3} electrolyte. Ordinate: m/[b_1]. Abscissa: m(mol kg^{-1}).

for all systems using the association models and equilibrium constants of Table 1.

DISCUSSION

There is a pronounced difference between the order of magnitude of the association constants of the analgesics investigated here and those of other drugs which associate in a nonmicellar manner. For example, the tricyclic drugs pavatrine hydrochloride (Attwood et al 1980), and propantheline and methantheline bromide (Attwood 1976) have K values that exceed those of the analgesics by a factor of 10 to 10^2. The large planar hydrophobic regions of these tricyclic drugs allow strong stacking interactions and can result in the formation of large aggregates. In contrast, the phenanthrene analgesics are non-planar bulky molecules which might be

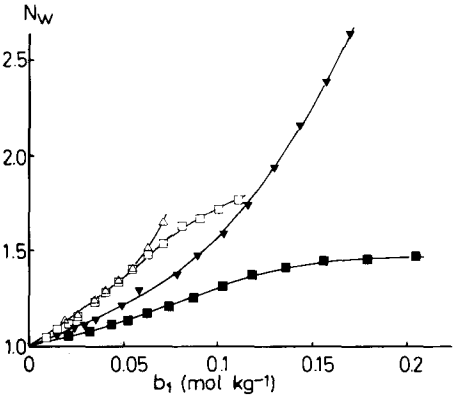

FIG. 9. Data plotted according to eqn 11 (Model 3) for △, ethylmorphine hydrochloride; □, oxycodone hydrochloride; ▼, pethidine hydrochloride and ■, codeine phosphate in 0·5 mol dm^{-3} electrolyte.

Table 1. Association models of analgesics in presence of 0·5 mol dm^{-3} electrolyte.

	Association Model	K dm^3 mol^{-1}
Codeine phosphate	$K_N = K$	1·26
Ethylmorphine HCl	,,	3·08
Oxycodone HCl	,,	2·99
Pethidine HCl	$K_N = K(N-1)/N$	3·58

expected to undergo only weak hydrophobic interaction. These analgesics more closely resemble the nucleotides (Ts'o et al 1963) both in their mode of association and in the order of magnitude of the association constants.

The more cooperative pattern of association of pethidine compared with the phenanthrene derivatives is a consequence of its less complex molecular structure which more readily allows the formation of larger aggregates. The higher association constants of ethylmorphine hydrochloride compared with codeine phosphate (methylmorphine) reflect the greater hydrophobicity of the ethoxy as opposed to the methoxy substituent.

Similarly a comparison of codeine phosphate with dihydrocodeine tartrate, which fails to aggregate even in electrolyte, illustrates the hydrophobic nature of the double bond at the 7,8 position of the codeine molecule. In making such comparisons, however, no account has been taken of the possible effects of the differing counterions on the extent of association.

Acknowledgements

The authors gratefully acknowledge the award of a research studentship to J.A.T. by the Science Research Council. We wish to thank Glaxo, Boots Co. and Roche Products for generous gifts of drugs.

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