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Aggregation of antiacetylcholine drugs in aqueous solution: micellar properties of some diphenylmethane derivatives

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Light scattering methods have been used to examine the aggregation in aqueous solution of a series of antiacetylcholine drugs based on the diphenylmethane nucleus. The drugs investigated included adiphenine hydrochloride, piperidolate hydrochloride, benztropine mesylate, orphenadrine hydrochloride, chlorphenoxamine hydrochloride, lachesine hydrochloride, poldine methylsulphate, pipenzolate bromide, clidinium bromide, benzilonium bromide and ambutonium bromide. A micellar pattern of association was established for all compounds and critical micellar concentrations and aggregation numbers have been determined.

A large number of drugs from many pharmacological groups of compounds are now known to aggregate in aqueous solution (see reviews by Florence, 1968 and Felmeister, 1972). In previous studies, it was established that some antihistamines based on the diphenylmethane nucleus associated in aqueous solution by a typically micellar process. The micellar properties (Attwood 1972; Attwood & Udeala, 1974, 1975a) and surface activity (Attwood & Udeala, 1975b, c) of these compounds have been reported. Many antiacetylcholine drugs also contain a diphenylmethane nucleus and this present investigation is concerned with the possible aggregation of these drugs in aqueous solution.

MATERIALS AND METHODS

Materials. The following drugs were sufficiently well characterized and purified by the manufacturers to be used without further purification; adiphenine hydrochloride (Ciba); poldine methylsulphate B.P. (Beecham Research); lachesine chloride B.P.C. (Vestric); chlorphenoxamine hydrochloride (Evans Medical); piperidolate hydrochloride and pipenzolate bromide (M.C.P. Pharmaceuticals); orphenadrine hydrochloride B.P. (Brocades, Gt Britain); benztropine mesylate B.P. (Merck Sharp and Dohme); clidinium bromide (Roche); ambutonium bromide (Wyeth) and benzilonium bromide (Parke-Davis).

Light scattering measurements. Measurements were made at 303K with a Fica 42000 photogoniodiffusometer (A.R.L. Ltd), using a wavelength of 546 nm. Aqueous solutions were clarified by ultrafiltration through 0·1 μm Millipore filters until the ratio of the light scattering at angles of 30 and 150° did not exceed 1·10. The refractive index increments of the micellar species (dn/dm_2) were measured at 546 nm using a differential refractometer.

RESULTS

The light scattering results are presented in Figs 1 and 2 as plots of the scattering at 90°, S_{90}, as a function of the molal concentration, m. Critical micelle concentrations (cmc) were determined from the discontinuities in the S_{90}, *vs* m plots. For all compounds, a theoretical line, calculated for monomeric scattering, was a good representation of the experimental scattering in the pre-cmc region. The micellar aggregation number, N, and the effective thermodynamic micellar charge, p, were evaluated using equations proposed by Anacker & Westwell (1964). In a solution containing no added electrolyte

$$p = [2B\,cmc + (8B\,cmc)^{\frac{1}{2}}]\,A^{-1}(2\text{-}A)^{-1} \quad .. \quad (1)$$

$$N = p(p + 1)\,A\,(2\,B\,cmc + pA^2)^{-1} \quad .. \quad (2)$$

A and B are the intercept and slope respectively of plots of $Km_2/\Delta R_{90}$ against the molal concentration of micelles, m_2. ΔR_{90} is the Rayleigh ratio of the solution in excess of that of a solution at the cmc; $K = 2\pi^2 n_0^2 (dn/dm_2)^2 V^0/L\lambda^4$; n_0 is the refractive index of the solvent; V^0 is the volume of solution containing 1 kg of water; L is the Avogadro number and λ is the wavelength of the incident light. The

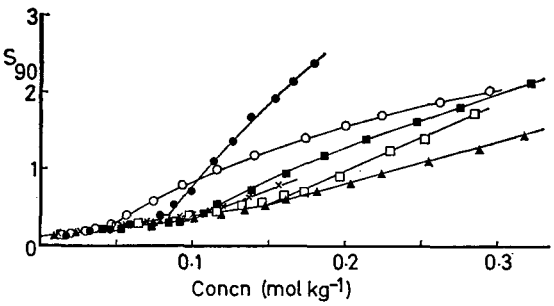


FIG. 1. Variation of the scattering ratio, S_{90}, with concentration for aqueous solutions of ○, benztropine mesylate; ●, piperidolate HCl; ×, pipenzolate Br; □, poldine methylsulphate; ▲, ambutonium Br; ■, orphenadrine HCl.

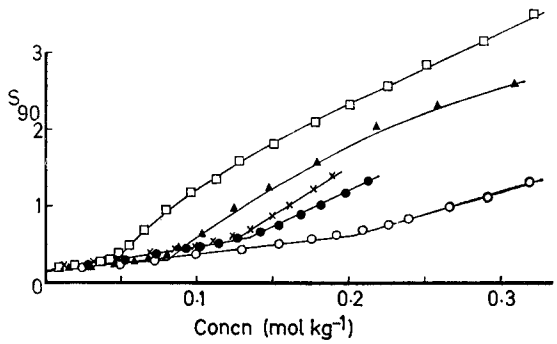


FIG. 2. Variation of the scattering ratio, S_{90}, with concentration for aqueous solutions of ○, lachesine Cl; ●, benzilonium Br; ×, clidinium Br; □, chlorphenoxamine HCl; ▲, adiphenine HCl.

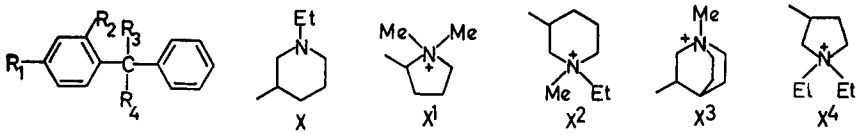
intensity of scattering from some compounds was not sufficiently high to allow the calculation of reliable values of p and in such cases N was equated with A^{-1}. The light scattering results are summarized in Table 1.

DISCUSSION

The effect of the nature of the hydrophobic group on the pattern of association of hydrophobic solutes has been discussed by Mukerjee (1974). Surfactants with aromatic hydrophobic groups are generally thought to associate by a non-micellar process in which aggregate growth occurs by a continuous stepwise addition of monomers. Several antihistamines have been shown to conform to this type of association process (Attwood & Udeala, 1975d). The S_{90}-m plots for such compounds show a continuous increase in scattering intensity with increased concentration with no apparent cmc. The scattering curves of the antiacetylcholine drugs examined here are clearly not of this type. All these compounds showed well-defined points of inflection, typical of those noted at the cmc's of conventional surfactants. No curvature of the S_{90}-m curves in the pre -cmc regions could be detected and the concentration—dependence of the scattering intensity in this region conformed to that calculated for monomers. Similarly, any curvature at higher concentrations was, as is normal with ionic surfactants, concave to the concentration axis and not convex, as is a feature of continually associating systems. Similar, typically micellar, scattering curves were noted for those antihistamines with hydrophobic groups also based on diphenylmethane, (Attwood, 1972). One of the characteristics of molecules which aggregate by continuous self association is the possession of a flat or nearly flat aromatic hydrophobic group which allows a face-to-face stacking of monomers within the aggregate. Diphenylmethane is, however, non-planar and such monomeric stacking would clearly be hindered by rotation around the central C atom. This may be the reason why the diphenylmethane antiacetylcholine and antihistamine drugs, although essentially aromatic in character, aggregate by a micellization process which is more commonly associated with the flexible chain aliphatic compounds.

The effect on the micellar properties of modifications of the chemical structure of the hydrophobic and hydrophilic portions of the molecule may be

Table 1. *Micellar properties of antiacetylcholine drugs at 303k.*



Compound	R_1	R_2	R_3	R_4	dn/dm_2 kg mol^{-1}	cmc mol kg^{-1}	N	p	α
Adiphenine HCl	H	H	H	$COO[CH_2]_2$ N Et_2	0·0630	0·082	10	2·3	0·23
Piperidolate HCl	H	H	H	COOX	0·0666	0·082	12	2·0	0·17
Benztropine mesylate	H	H	H	Tropine	0·0642	0·041	7	1·5	0·21
Orphenadrine HCl	H	CH_3	H	$O[CH_2]_2$·N Me_2	0·0593	0·096	7	1·6	0·23
Chlorphenoxamine HCl	Cl	H	CH_3	$O(CH_2)_2$·N Me_2	0·0619	0·045	13	2·4	0·19
Lachesine Cl	H	H	OH	$COO[CH_2]_2$·N Me_2 Et	0·0655	0·204	2	—	—
Poldine methylsulphate	H	H	OH	$COO-CH_2X^1$	0·0671	0·150	3	—	—
Pipenzolate Br	H	H	OH	$COOX^2$	0·0733	0·105	3	—	—
Clidinium Br	H	H	OH	$COOX^3$	0·0722	0·123	4	—	—
Benzilonium Br	H	H	OH	$COOX^4$	0·0722	0·143	4	—	—
Ambutonium Br	H	H	$CONH_2$	$[CH_2]_2$ N Me_2 Et.	0·0697	0·136	2	—	—

assessed from the data presented in Table 1. In this respect the drugs investigated here present an interesting series of model surfactants. The most pronounced effect on the micellar properties results from the -OH or -$CONH_2$ groups in the R_3 position. As expected, these groups confer a greater hydrophilicity, as evidenced by the higher cmc's and lower aggregation numbers of compounds with these substituents. The magnitude of the effect may be assessed by considering pairs of compounds with otherwise similar structures, for example, piperidolate compared with pipenzolate and adiphenine with lachesine. A comparison of the properties of lachesine, poldine, pipenzolate, clidinium and benzilonium illustrates the well-known effect on the hydrophobicity of increasing the number of CH_2 groups in the polar chain (R_4). There is a decrease in cmc (increased hydrophobicity) with increase in the number of CH_2 groups, in the order lachesine ($6CH_2$) > poldine ($7CH_2$) > pipenzolate, clidinium and benzilonium ($8CH_2$). It is of interest to compare orphenadrine and chlorphenoxamine with the antihistamine, diphenhydramine ($R_1 = R_2 = R_3 = H$) which has a cmc of 0·132 mol kg^{-1} and forms trimers in aqueous solution (Attwood 1972). These three compounds have identical R_4 substituents and a comparison of their properties illustrates the influence of substituents on the diphenylmethane nucleus. The gradual increase in N and decrease in cmc in the order diphenhydramine—orphenadrine—chlorphenoxamine illustrates the increasing hydrophobicity resulting from the ring substituted methyl group of orphenadrine and the substitution of Cl and CH_3 in the R_1 and R_2 positions respectively in chlorphenoxamine.

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