Solubility Profiles for the Xanthines in Dioxane – Water Mixtures

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The solubility of caffeine, theophylline, and theobromine were determined in dioxane–water mixtures, and the solubility curves that were determined showed a multiplicity of peak solubility values. These peak solubility values occurred at about the same solvent composition for the subject compounds studied. A property common to these solvent compositions having the same value is the dielectric constant. These peaks are descriptively referred to as dielectric requirements irrespective of the mechanism operative in causing the formation of these unusual solubility curves. These solubility curves are referred to as solubility profiles.

IN AN EARLIER publication (1), it was suggested that dioxane–water mixtures could be used to study the dielectric constant effects upon the enhanced solubility of pharmaceutical solutes. These cosolvent mixtures could be formulated so as to produce any dielectric constant value between 2 to 78. Although this dielectric constant span or spectrum is admittedly rather wide, the area of pharmaceutical utility extends over at least half this range. It has been shown (2) that the relationship of dielectric constants and solubility parameters is essentially linear in the pharmaceutical range, *i.e.*, alcohols, glycols, and water, and implied the usefulness of the former as a polarity spectrum. The usefulness of this polarity spectrum in relation to solubility has been confirmed over a limited range for several solutes in sucrose vehicles (3). Martin (4, 5) has shown that the solubility parameter concept can be meaningful and possibly extended to solutions of moderate polarity.

In describing the occurrence of peak solubility in cosolvents for a given solute, the dielectric requirement has been defined as the dielectric constant of maximum solubility. In the case of salicylic acid (6), it was found that a dielectric requirement (DR) of 15 was quite constant for 30 binary mixtures, the individual solvents bracketing $\epsilon = 15$. Thus, it would seem that maximum solubility would occur at a dielectric constant of about 15 for a given binary mixture, irrespective of the cosolvents used to formulate a given value of the dielectric constant. It should also be indicated that although these DR's varied only about 3 units, the magnitude of the solubility or the solubility curves varied significantly depending upon the system chosen. It was also noted that another peak existed at a dielectric requirement of 25 for two binary mixtures bracketing that value of the dielectric constant. Several other binary mixtures showed shouldering at $\epsilon = 25$. This was the first indication that a given solute could have two solubility peaks over a dielectric constant range. This may not be too surprising in view of the possible existence of different species as the polarity of the solvent system changes.

Consequently, it may be possible that a given solute could show more than one solubility maximum or a multiplicity of peaks as a function of the dielectric constant. From some preliminary studies on theobromine, there was some indication that multiple solubility peaks could be observed, and it was felt that the xanthines would lend themselves well to this study because of self-polymerization (7) as a function of polarity. These present studies were conducted with a view toward establishing the possible multiplicity of peaks for the xanthine drugs.

MATERIALS AND METHODS

Materials.—Caffeine (Eastman-Kodak 355 White Label), theobromine (Eastman-Kodak 1690 White Label), and theophylline anhydrous (Ruger Chemical Co. 16628) were the solutes used. Distilled water and *p*-dioxane (Fisher certified D-111) were the solvents used.

Equipment.—A Beckman DK-2 was used for spectrophotometric analysis of solutes. An Ainsworth type 12 balance was used for the gravimetric analysis of solutes. A Sargent water bath was used with attendant Thermonitor unit for solution equilibration and temperature control.

Methods.—Solvents containing dioxane–water in 2.5% v/v increments were added to 22-ml. screw-capped Teflon-lined vials and excess solute added. Sample vials were tied down to a rotating disk assembly connected to a variable-speed motor. A variac attached to the motor was set at a value so that the vial contents were actively agitated. The bath temperature was maintained at 25 ± 0.2° by means of a Thermonitor unit. Equilibration of samples was found to be about 24 hr. For each sample vial, two samples were withdrawn through

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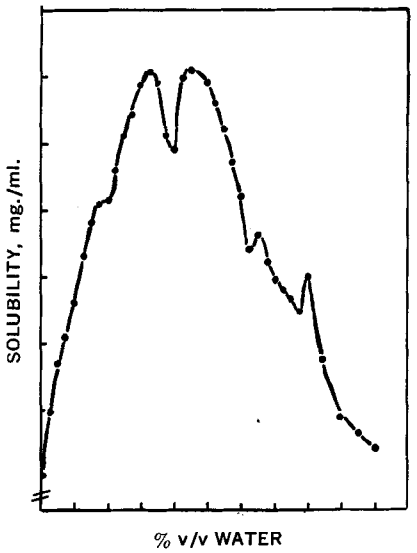

Fig. 1.—The solubility of caffeine at 25°C. in mg./ml. is plotted as a function of composition (v/v) for dioxane–water mixtures.

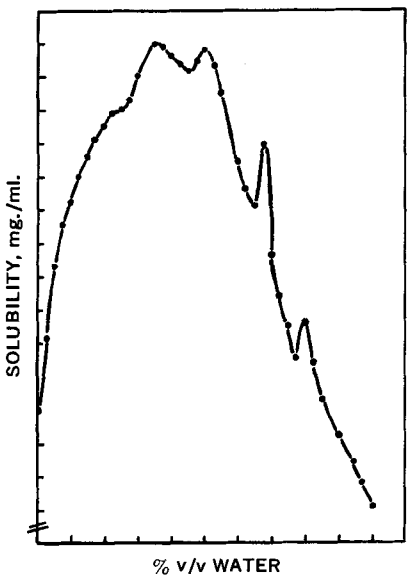

Fig. 2.—The solubility of theophylline at 25°C. in mg./ml. is plotted as a function of composition (v/v) for dioxane–water mixtures.

a pipet wrapped in glass wool, a 1-ml. sample for spectrophotometric analysis and a 5-ml. sample for gravimetric analysis.

Theophylline and theobromine were stored in a vacuum desiccator before use at about 40° and 5 mm. Hg, and caffeine was stored at 80° for 24 hr. before use. This was done to insure that the solutes would be anhydrous before introduction into the sample vials in order to preclude variance in the hydrated state of commercially supplied materials. Each of the xanthine solutes were added to dioxane–water vehicles with a 2.5% increment variation of solvent composition. These solubility runs were done three times, each run being subjected to both spectrophotometric and gravimetric analysis. Internal averaging for each run was performed, and the results reported are for the three-run average.

For the spectrophotometric analysis, the sample was diluted and assayed immediately. The solubility was determined from the absorbance and previously determined Beer's law plots at about 270 mμ for these substances. The gravimetric sample was placed in a tared vial put into a vacuum desiccator filled with a bed of calcium chloride. Vacuum down to about 5 mm. of Hg was placed on this system and allowed to stand for 3–4 days. After about 3 days, the samples rich in dioxane were dry, whereas those samples rich in water were partially wet. These wet samples were heated at 60° for 8–20 hr. depending upon the volume of liquid remaining. The dry samples were weighed, replaced in the vacuum desiccator for 24–48 hr., and this procedure was continued until constant weight had been achieved. Constant weight was assumed when two consecutive weighings did not differ by more than 0.3 mg. for caffeine and theophylline or 0.1 mg. for theobromine.

All samples were stored in a vacuum desiccator and weighed periodically for weight change over a 2-week period. Sample weights were compared with previous results, and it was found that relatively insignificant changes occurred.

RESULTS AND DISCUSSION

The observed solubilities of caffeine, theophylline, and theobromine are shown in Figs. 1–3 as a function of the composition of dioxane–water mixtures. In each case, four solubility peaks are obvious, and another possible peak occurs at a dielectric constant value of about 14. These shoulders in the solubility curves were redetermined several more times and do exist and are not due to experimental error. The solubility shown was the averaging of three solubility runs, including the internal averaging of the spectrophotometric and gravimetric procedures. Each individual analytical method showed the same solubility curve; however, the

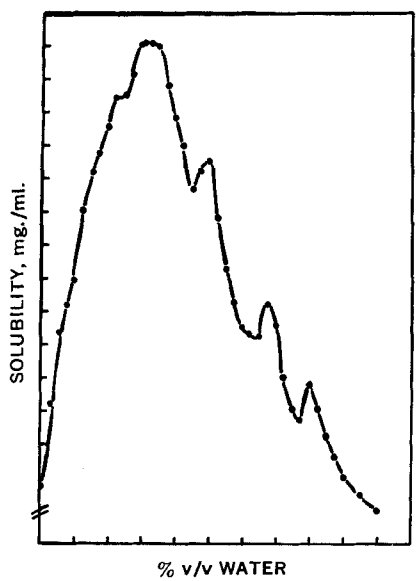

Fig. 3.—The solubility of theobromine at 25°C. in mg./ml. is plotted as a function of composition (v/v) for dioxane–water mixtures.

TABLE I.—SUMMARY OF THE DIELECTRIC REQUIREMENTS OBSERVED FOR THE XANTHINE DRUGS

	Theobromine DR	Theophylline DR	Caffeine DR
Shoulder	14	14	11
Peak 1	22	20	20
Peak 2	34	34	30
Peak 3	50	50	50
Peak 4	61	61	61

TABLE II.—SUMMARY OF THE SOLUBILITY OF XANTHINE AT 25°C. IN PURE WATER COMPARED TO LITERATURE VALUES

	Solubility (mg./ml.)[a]	Solubility (mg./ml.)[b]
Caffeine	21.8	23.2
Theophylline	8.3	8.0
Theobromine	0.5	0.61

[a] "Merck Index" (8). [b] Present work.

TABLE III.—SUMMARY OF THE RATIOS OF THE SOLUBILITY FOR THE SHOULDER AND PEAK VALUES COMPARED TO THE SOLUBILITY IN PURE WATER

	Caffeine	Theophylline	Theobromine
Ratio[a]	44	17	1
Ratio[b]	39	13	1
Shoulder	34	18	1
Peak 1	44	18	1
Peak 2	41	15	1
Peak 3	32	12	1
Peak 4	42	14	1
Av.	39	15	1

[a] Literature values. [b] Present work.

gravimetric procedure ran slightly above (6–10%) the spectrophotometric procedure. It is felt that the gravimetric procedure is somewhat more accurate due to larger sample withdrawal and minimal procedural errors. In averaging results, no new peaks or valleys were created, and the reported results reflect the constancy of these solubility curves.

Table I summarizes the dielectric requirement for each solute in the solvent system studied. It should be noted that the dielectric requirements are quite close to one another, the maximum variation being about 3 dielectric constant units.

Since the DR's for the xanthine molecules were relatively constant, it was felt that these solubility curves might be parallel to one another, indicating the proportionality of the magnitude of solubility. Consequently, the ratio of the solubilities at the peak and shoulder values were compared to the ratio of solubility in pure water. In Table II, the solubilities for the xanthines in pure water are compared with the literature values (8). The agreement is quite good for caffeine and theophylline, whereas the value for the solubility of theobromine is larger than the reported value. However, the literature value for theobromine is approximately 1 Gm./2000 ml. of water, and this work indicates a solubility value of 1 Gm./1630 ml. of water.

In Table III, the ratio of solubility for these materials at their DR's (including shoulder) are shown relative to the solubility of theobromine which has been defined as unity at each point. From this table, it can be seen that although variations do occur, the magnitudes of solubility (or solubility curves) are approximately proportional to one another. Again, considering the solubility of theobromine as unity, the ratios in pure dioxane are theobromine, 1; theophylline, 10; caffeine, 23. It should be noted in this respect that the solubility of theobromine and theophylline are higher in dioxane relative to pure water, whereas the solubility of caffeine is just slightly lower. It might be inferred from these values that the higher ratio for theophylline and the lower ratio for caffeine at the shoulder values may be due to the high (75% v/v) dioxane concentrations for these systems. In other words, the magnitude of the solubility is more dependent upon the dioxane than the water at this point. It is further possible that the proportionality of the solubility curves depends upon the relative concentration of one solvent component over the other as well as the respective solubility in each pure component. Since the ratios of solubility at the DR's values are far closer to the ratios in pure water, this may imply the importance of the aqueous solvation of these solutes irrespective of the actual water concentration.

The solubility peaks displayed by these xanthine molecules might be explained by the existence of n-mer states of the solute as the polarity changed. It has been shown (7) that caffeine exists as a monomer in nonpolar solvents and as various polymers in water. Theophylline and theobromine are structurally similar to caffeine and may be expected to exhibit similar behavior. However, Higuchi (7) found that with theophylline, at the concentrations and solvents used, no self-polymerization occurred and that theobromine would not lend itself to studies of this type because of limited solubility. For caffeine, the tetrameric species and other n-mer states are found in water which has a dielectric constant of 78. The appearance of peaks at values intermediate between the stated extremes may indicate n-mers of the solute with n assuming values between the monomeric and tetrameric states. It is further possible that the polymeric forms exist to varying degrees at a given point or all along the solubility profile. Although the above mechanism may be possible in these systems, it is suspect, unless it can be shown that all these xanthine solutes used in this study are able to undergo self-polymerization. A study of the colligative properties at various cuts along the solubility profile would seem logical.

A stronger possibility exists, however, and that would be the formation of solvated or hydrated states of the solute. As the solvent composition changed above the solid phase of these systems, the nature of the excess solute in equilibrium may also have changed. Thus, the initial increase in solubility could have been due to the formation of a more soluble water complex or hydrate of the xanthine. The shoulder might indicate saturation of the system with respect to that species and conversion of the solid state to hydrate. The subsequent rise in solubility might be due to the formation of a hydrate of higher order, etc. In order to aid in the delineation of the mechanism, future studies on the nature of the solid phase with respect to the extent of hydration will be carried out by thermogravimetric procedures or other applicable methods.

At the present time, the factors involved in the above points cannot be delineated to any greater extent. Regardless of the mechanistic aspect, the xanthine drugs do exhibit multiple solubility peaks as a function of the dielectric constant of the solvent system studied. The multiplicity of peaks is not limited to the above solutes. The dielectric solubility profiles for several antipyretic drugs such as acetanilid, *p*-methyl acetanilid, and *p*-ethoxy acetanilid (phenacetin) have been determined and are the subject of another communication. The aforementioned solutes also illustrate a multiplicity of dielectric requirements.

It may be advantageous to consider these dielectric requirements as being more representative of actual behavior relative to the solubility parameter concept. It is the authors' understanding that the solubility parameter concept apparently predicts only one value for solubility maximum. This occurs when the solubility parameter of the solvent mixture is greater or lesser than the value for the solute, the solubility is decreased, and therefore a solubility curve is observed. However, it should be noted that only one DR or solubility parameter may exist in which the observed solubility curve may supercede in magnitude all other subservient solubility curves. It can then be said that an optimum dielectric requirement or solubility parameter value may exist relative to the magnitude of solubility.

Occurrence of solubility peaks at the same value of the dielectric constant in solvent pairs other than dioxane–water should aid in lending some validity to the dielectric constant approach. Studies of this type will be reported in future communications.

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Some Neuropharmacological Properties of the Ephedrine Isomers

By GEORGE LANCIAULT and HAROLD H. WOLF

The central nervous system stimulating activity of the ephedrine isomers was compared with that of racemic amphetamine. Central nervous system alterations induced by acute administration of the drugs were evaluated by employing several standard techniques, including low-frequency electroshock and chemoshock threshold determinations, hexobarbital sleep-time alteration, and a behavioral rating scale. Evidence was obtained to show that the ephedrines vary in their ability to produce central stimulation. It was found that D(−)ephedrine and L(+)ephedrine were considerably more potent than D(−)pseudoephedrine and L(+)pseudoephedrine.

Previous work with the ephedrine isomers involving the cardiovascular system has demonstrated that these compounds vary markedly in their effects on this system (1, 2). D(−)Pseudoephedrine is reported to lack the ability to produce a typical ephedrine pressor response, causing instead a depression of blood pressure. This isomer also has been observed to produce vasodilation in vascular beds of dogs, in contrast to D(−)ephedrine which produces vasoconstriction. Furthermore, renal and vertebral arterial blood flow in the dog decreases when L(+)ephedrine or L(+)pseudoephedrine are given, whereas with D(−)ephedrine the flow increases.

Although there are many literature references relative to D(−)ephedrine and its effects on the central nervous system (3–6), reports of work with the other isomers are scanty and inconclusive. However, some quantitative differences in the central activity of these compounds have been reported. For example, Trevan (3) has demonstrated that the isomers vary considerably in their ability to act as analeptics in anesthetized

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