Inter-relationships between solubilities, distribution coefficients and melting points of some substituted benzoic and phenylacetic acids

N. A. ARMSTRONG, K. C. JAMES* AND C. K. WONG†

Welsh School of Pharmacy, University of Wales Institute of Science and Technology, King Edward VII Avenue, Cardiff, CF1 3NU, U.K.

Ten 4-hydroxy and 4-alkoxy benzoic and phenylalkanoic acids have been investigated. Solubilities in aqueous buffer at pH 1·2 were determined, together with distribution co-efficients between the buffer and either octanol or isopropyl myristate. When plotted against the total number of carbon atoms in the side chains, log octanol/water distribution co-efficients gave two parallel straight lines, one for the substituted benzoic acids, and the other for the substituted phenylalkanoic acids. The slopes approximated to 0·5, the generally accepted value for methylene. Similar plots could be obtained with isopropyl myristate, provided the hydroxy acid results were ignored, and also when log aqueous solubilities were plotted against carbon number, although there was considerable scatter. The differences between the distribution coefficient results were explained in terms of solute-solvent inter-actions, and the scatter attributed to variations in the heats of fusion of the solutes. Yalkowsky's equation (1977), linking aqueous solubilities and melting points with distribution coefficients, was applied to the results, and found to be of limited predictive value.

The release of a drug from its delivery system is often the most important factor affecting bioavailability. For many solid orally administered dosage forms, the drug dissolution rate in vitro is directly related to the absorption rate of the drug in vivo, and it is reasonable to assume that where drugs are presented in the form of a solution in a lipophilic solvent, as in soft gelatin capsules, the in vitro release from the solvent will give a similar indication of bioavailability. One would anticipate that the rate of release would be influenced by the distribution coefficient of the drug between the lipophilic solvent and the digestive fluids. As a preliminary to an attempt to link in vitro release from lipophilic solvents with in vivo bioavailability, distribution coefficients of 10 aroma-tic acids have been determined. We describe the determination and evaluation of these results.

MATERIALS AND METHODS

Materials

Details of the ten substituted aromatic acids used are in Table 1. Those claimed to be at least 99% pure were used without further refinement. 4-Ethoxy-

* Correspondence.
† Present address: Department of Pharmacy, University of Singapore, Singapore 3, Republic of Singapore.

phenylacetic acid and 4-methoxy-phenylacetic acid were recrystallized from absolute ethanol, a sufficient amount of active charcoal being added for decolor-ization. Further recrystallization from benzene yielded white crystals, which were dried at 35 °C under reduced pressure. 4-Methoxyphenylbutyric acid was recrystallized alternately from absolute ethanol and light petroleum, 40–60 °C, to obtain white crystals, which were dried at 35 °C under reduced pressure.

n-Octanol was a specially pure grade from BDH Ltd. It was washed with M sodium carbonate solu-tion and distilled water. After preliminary drying with calcium chloride, fractional distillation was carried out in all glass apparatus, and only the middle fraction of the distillate (194 ± 0·5 °C) was collected. The distillate was then passed through a column of alumina. The eluant showed an absorb-ance of less than 0·2 at 260 nm, against spectroscopic ethanol.

Isopropyl myristate of at least 95% purity (Fluka Ltd), was extracted twice with an equal volume of M sodium carbonate solution. The ester layer was filtered and washed several times with distilled water until the washes were neutral. The liquid was then dried by passing through a column of calcium chloride. The product was distilled at about 260 Pa and the middle fraction collected at 145 ± 1 °C.

Table 1. Physical properties of substituted aromatic acids.

Comp.	Substituted aromatic acids	Source	M.p. °C	Heat of fusion ΔH^f ($\pm$ s.e.m.)* (K J mol^{-1})	Solub. in pH 1·2 buffer S_{mf}($\pm$ s.e.m.* ($\times 10^5$ mol fraction)	Partition coefficient $\pm$ s.e.m.* n-octanol– water (P_{oct})	Isopropyl myristate–water (P_{Ipm})
Substituted benzoic acids							
1	4-Hydroxy-	BDH Ltd	214·9	30·9 (0·59)	151·5 (0·8)	25·58 (0·09)	0·545 (0·005)
2	4-Methoxy-	BDH Ltd	184·6	28·4 (0·13)	4·32 (0·02)	75·96 (0·44)	8·72 (0·10)
3	4-Ethoxy-	Aldrich	199·6	29·4 (0·29)	1·49 (0·02)	242·9 (1·27)	28·14 (0·26)
4	4-Butoxy-	Aldrich	147·7	19·1 (0·08)	0·14 (0·001)	3726·1 (24·6)	403·7 (8·12)
Substituted phenylacetic acids							
5	4-Hydroxy-	Aldrich	150·4	28·4 (0·08)	1382·1 (11·8)	5·65 (0·06)	0·127 (0·01)
6	4-Methoxy-	Aldrich	84·9	21·8 (0·08)	121·6 (0·7)	29·90 (0·24)	2·95 (0·08)
7	4-Ethoxy-	Aldrich	87·0	23·0 (0·08)	42·11 (0·10)	89·66 (0·81)	9·60 (0·12)
Substituted phenylpropionic acids							
8	4-Hydroxy-	Koch Light	129·3	28·9 (0·13)	524·3 (0·9)	14·35 (0·01)	0·439 (0·03)
9	4-Methoxy-	Aldrich	103·7	28·5 (0·13)	16·72 (0·13)	79·04 (0·66)	10·44 (0·06)
Substituted phenylbutyric acids							
10	4-Methoxy-	Aldrich	57·7	25·3 (0·05)	18·14 (0·16)	214·0 (0·79)	27·28 (0·42)

* $P' = 0.01$.

This was then passed alternately through columns of silica gel and alumina until a saturated solution in aqueous buffer gave no measurable absorbance in the ultraviolet region.

Melting points and heats of fusion

Melting points were determined on a Perkin-Elmer DSC-1B differential scanning calorimeter, and located by extrapolating the initial segment of the endotherm to the baseline. Five replicates were taken for each compound. Mean values are shown in Table 1. In all cases, thermograms gave narrow and sharp endothermic peaks, indicating a high degree of purity. Heats of fusion were determined from the melting point peak areas, using a Programmable Computing Integrator type CRS 404/30 (Infotronic UK Ltd.), connected to the DSC output. The instrument was standardized using tin.

Solubility determinations

Solubilities were determined in pH 1·2 buffer solution, prepared by dissolving 2 g sodium chloride and 7 ml 35·4% HCl in sufficient distilled water to produce 1 litre. Preliminary experiments indicated that all compounds obeyed Beer's Law at their u.v. absorption maxima.

A quantity of material, in excess of its solubility, was placed in a 50 ml conical flask with about 20 ml of buffer solution, and shaken mechanically in a water bath at 37 $\pm$ 0·2 °C. Triplicate runs were carried out for each compound, and sampling

continued until equilibrium was reached, saturation being achieved in less then 96 h. Excess solute was removed by filtering through a 0·45 μm membrane filter, and samples were diluted and assayed spectrophotometrically. Mole fraction solubilities are shown in Table 1.

Determination of distribution coefficients

Two systems were investigated, n-octanol–water and isopropyl myristate–water. A pH 1·2 buffer was used for the aqueous phase, because all the aromatic acids were known to be almost totally unionized at this pH. Liquid phases were equilibrated with respect to each other before use. Solutions were initially made in the non-aqueous phase, and 10 ml of this added to 10 ml of buffer, and vigorously shaken for at least an hour. The mixture was then placed in a water bath at 37 $\pm$ 0·2 °C for a minimum of 48 h. Using a glass syringe fitted with 1·0 mm diameter PTFE tubing, samples were withdrawn from the aqueous phase, diluted and analysed spectrophotometrically. Concentrations in the non-aqueous phase were calculated by difference. Results are given in Table 1.

DISCUSSION

Distribution coefficients of the substituted aromatic acids between octanol and the aqueous phase (P_{oct}) were determined over a range of solute concentrations, since it has been reported (for example, Beckett & Moffat 1969) that benzoic and phenylacetic acid

derivatives associate in some organic solvents. However no evidence of association was found in this study, the values of the distribution coefficients being independent of concentration, as were the distribution coefficients of the isopropyl myristate-water systems (P_{ipm}).

Two parallel rectilinear plots (Fig. 1) were obtained when log P_{oct} was plotted against total carbon number in the two side chains. Regression data for the general equation log P_{oct} = An + B are given
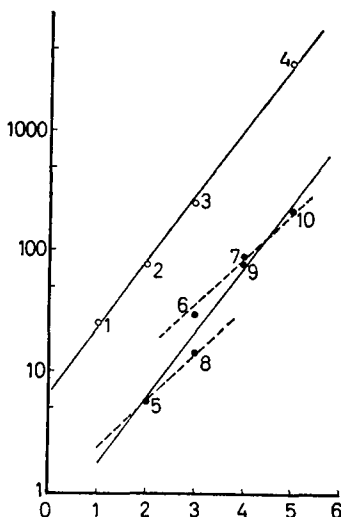

FIG. 1. Relationship between the n-octanol-water partition coefficients (ordinate) and the total carbon number(s) (abscissa) on the side chains of substituted aromatic acids. Solute code as in Table 1. ○: substituted benzoic acids. ●: substituted phenylalkanoic acids. —: all solutes. - - - -: hydroxy phenylalkanoic and alkoxy-phenylalkanoic acids treated individually.

in Table 2, the slopes for octanol being in good agreement with the generally accepted hydrophobic substituent constant for methylene of 0·50 (Tute 1971). In contrast, the isopropyl myristate-water distribution coefficient (P_{ipm}) only gave acceptable rectilinear plots if the phenolic acids were ignored (Fig. 2 and Table 2).

It is not unusual for the first member of a homologous series to behave differently from the remainder, particularly when it is a different type of compound, as is the case here. However if this is the reason why the results for the hydroxy compounds deviate from the overall trends in the isopropyl myristate-water systems, it is difficult to reconcile this suggestion with the octanol-water results, since the distribution coefficients for the hydroxy compounds in this latter system appear to fit the overall regression lines. It is possible that the hydroxy com-

Table 2. Regression data for the equation Log P = A n + B.

Non-aqueous solvent	A	B	No of comps in series (N)	Corr. coeff. (r)	s.d.
Benzoic acid series (Compounds 1–4)					
octanol	+0·544 (0·021)	+0·816	4	0·998	0·063
Phenyl aliphatic acid series (Compounds 5–10)					
octanol	+0·541 (0·055)	−0·299	6	0·980	0·129
Benzoic acid series (Compounds 1–4) isopropyl					
myristate	+0·684 (0·095)	−0·698	4	0·981	0·281
Phenyl aliphatic acid series (Compounds 5–10) isopropyl					
myristate	+0·807 (0·144)	−2·384	6	0·942	0·337
Benzoic acid series (Compounds 2–4 isopropyl					
myristate	+0·559 (0·017)	−0·197	3	1·000	0·037
Phenylaliphatic acid series (Compounds 6, 7, 9, 10) Isopropyl					
myristate	+0·483 (0·027)	−0·955	4	0·997	0·038

The figures in parentheses represent the standard error of the coefficient ($P' = 0.05$).

pounds do behave differently from the alkoxy compounds in octanol-water, but the difference is not sufficiently conspicuous to be detected.

Close scrutiny of Fig. 1 does suggest that the phenylaliphatic acid results could be resolved into two straight lines, one for the phenols and one for the ethers, as indicated in the figure, but there are insufficient data points to confirm this treatment.

Numerous investigators have studied the effects of the methylene group on the solubilities of homologous series (see, for example, Davis et al 1974).
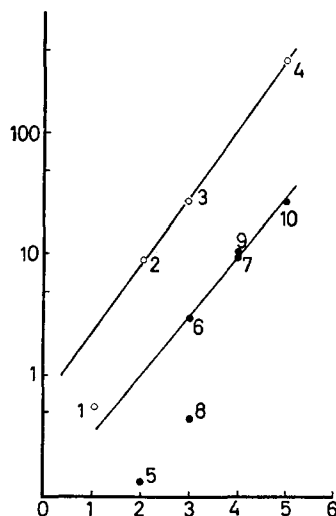

FIG. 2. Relationship between the isopropyl myristate/aqueous partition coefficients (ordinate) and the total carbon number(s) (abscissa) on the side chains of the substituted aromatic acids. Solute code as in Table 1. ○: substituted benzoic acids. ●: substituted phenyl-alkanoic acids.

For liquid solutes, solubilities of homologues, being linearly related to free energy, are proportional to carbon number. For solid solutes, dissolution involves not only mixing, but also liquefaction of the crystal lattice. The energy involved in liquefaction has been shown, for some systems to vary irregularly as the homologous series is ascended (James & Roberts 1968), so that a similar approach to that used with liquid solute systems is often impracticable. However, several homologous series of solids have been shown to give rectilinear plots of log aqueous solubility (X_2^{aq}) against carbon number (James 1976). The success of these correlations was attributed to the large entropy of mixing involved on dissolution of the liquid solute in water, in comparison with the energy changes required to liquefy the solid solute. The aqueous solubilities reported here could be resolved into two such rectilinear plots, one for the benzoic acid series and the other for the phenyl aliphatic acid series. However, there is considerable scatter about the regression lines, and it is suggested that this is due to irregular changes in heats of fusion in moving from homologue to homologue, sufficient to give scatter, but not enough to significantly change the overall shape of the plot. Excess free energy of solution (ΔG^E), the free energy in excess of that required for ideal solution, eliminates this variation. It is defined by equation (1) where X_2 is the observed solubility, and X_2^l the ideal solubility, calculated from equation (2)

$$\Delta G^E = -RT \ln \frac{X_2^l}{X_2} \qquad .. \qquad .. \qquad (1)$$

$$\ln X_2^l = \frac{\Delta Hf}{R} \left[\frac{Tm - T}{TmT} \right] \qquad .. \qquad .. \qquad (2)$$

ΔHf is the heat of fusion and Tm, the melting point.

Plots of ΔG^E against carbon number are shown in Fig. 3. There are two parallel straight lines, encompassing the same combinations as observed before, but the points are still highly scattered about the lines. If the phenolic acids are ignored as before, two different rectilinear plots are obtained as indicated in Fig. 3. These plots are approximately parallel, their slopes being 3·13 and 4·55. There are not enough data points to speculate on whether these plots are more realistic than the previous pair, or on how significant the possible parallelism is, but one important point in their favour is that their mean slope is 3·8, close to the mean $\Delta G^E_{CH_2}$ 3·6 kJ mol^{-1} recorded by Davis et al (1974).

Hansch et al (1968) proposed equation 3 (where a and b are constants) as a means of predicting
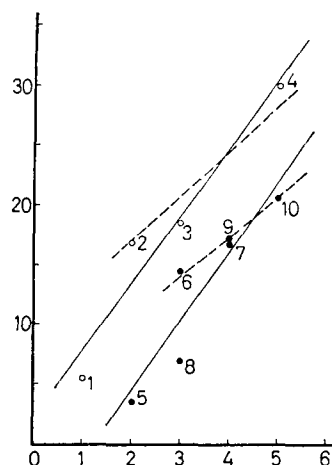

FIG. 3. Relationship between the excess free energies of solution in water (ordinate: kJ mol^{-1}) and the total carbon number(s) (abscissa) on the side chains of the substituted aromatic acids. Solute code as in Table 1. $\bigcirc$: substituted benzoic acids. $\bullet$: substituted phenyl-alkanoic acids. —: all solutes. - - -: ether acids only.

aqueous solubilities of liquids (X_2^{aq}) using distribution coefficients calculated from hydrophobic substituent constants (π values).

$$\text{Log } X_2^{aq} = a - b \log P_{oct} \qquad .. \qquad (3)$$

Equation 3 can also be used to perform the reverse function, namely to predict octanol–water distribution coefficients, which are usually more difficult to measure than solubilities, from aqueous solubilities.

Regression data between log P and ΔG^E are shown in Table 3. The relationship for P_{ipm} is good, but that for P_{oct} can only be described as fair. However, if only the ether acids are considered, the relationship for the octanol–water system is significantly improved, whilst that for isopropyl myristate–water is hardly changed.

Table 3. Regression data for the equation log P = aΔG_E + b.

Non-aqueous solvent	a	b	No of comps in series (N)	Corr. coeff. (r)	s.d.	F
All compounds (1–10)						
Octanol	+0·094 (0·011)	+0·472	10	0·953	0·251	79·3
Ether acids (compounds 2–4, 6–7, 9–10)						
Octanol	+0·130 (0·011)	−0·287	7	0·983	0·136	141·4
All compounds (1–10)						
Isopropyl myristate	+0·129 (0·006)	−1·196	10	0·991	0·150	421·1
Ether acids (compounds 2–4, 6–7, 9–10)						
Isopropyl myristate	+0·131 (0·012)	−1·238	7	0·981	0·144	127·2

The figures in parentheses represent the standard error of the coefficient ($P' = 0·05$).

Table 4. Coefficients of the equation $\log P = a + b \log X_2^{aq} + cT_m$

Non-aqueous solvent	a	b	c	No comps in series (N)	Corr. coeff. (r)	s.d.	F
All compounds 1–10)							
Octanol	0·011	−0·634 (0·063)	−0·003 (0·001)	10	0·967	0·226	50·7
Ether acids (compounds 2–4, 6–7, 9–10)							
Octanol	−0·497	−0·872 (0·072)	−0·007 (0·001)	7	0·988	0·128	81·1
All compounds 1–10 Isopropyl myristate	−1·452	−0·852 (0·030)	−0·006 (0·001)	10	0·996	0·107	415·5
Ether acids (Compounds 2–4, 6–7, 9–10) Isopropyl myristate	−1·460	−0·883 (0·058)	−0·007 (0·001)	7	0·992	0·103	127·9

The figures in parentheses represent the standard error of the coefficient ($P' = 0\cdot05$).

All the solutes considered here contain a carboxy group together with either ether oxygen or a hydroxyl group, whilst the solvents contain either a hydroxyl or an ester group. Solute–solvent interaction by hydrogen bonding could therefore be expected, particularly as the distribution coefficients indicate that solute–solute association does not occur. The results given above suggest that in isopropyl myristate, the mode of interaction is the same with hydroxy acids and ether acids, but with water and octanol, the hydroxy acids interact with the solvent in a different manner from the ether acids. This situation predicts that plots of $\log P_{oct}$ against n and of $\log P_{ipm}$ against ΔG^E will be rectilinear, and plots of $\log P_{ipm}$ against n and of $\log P_{oct}$ against ΔG^E will form two series, representing ether acids and hydroxy acids.

Whilst data reported in Table 3 provide a means of predicting distribution coefficients, their usefulness is largely limited to liquid solutes, because excess free energies of solution of solid solutes are themselves difficult parameters to measure. Recently Yalkowsky (1977) introduced equation 4 which utilizes the melting point of the solute and eliminates the need to determine heats of fusion

$$\log P = a + b \log X \text{ aq.} + cT_m \quad .. \quad (4)$$

Application of our data to the Yalkowsky treatment using multiple regression analysis yielded the coefficients shown in Table 4, which gives the same overall picture as before, namely that the isopropyl

myristate system gives a good correlation embracing all ten compounds, but with octanol, a better result is obtained if the hydroxy acids are omitted.

Using the solubilities and distribution coefficients of a large number of compounds, Yalkowsky calculated that the values of the constants b and c were $-1\cdot0$ and $-0\cdot01$ respectively. In this study, in those cases where reliable correlations are obtained, coefficients for $\log X_2^{aq}$ and T_M agree well with Yalkowsky's prediction. Our results therefore indicate that equation 4 can be used for estimating the distribution coefficients of closely related compounds but the regression constant varies from one solvent system to another, and would have to be calculated from experimental results.

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