Dissolution and Partitioning Behavior of Hydrophobic Ion-Paired Compounds

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Purpose. This study was conducted to determine the effects of counterion hydrophobicity on organic/aqueous partition coefficients for hydrophobic ion paired (HIP) complexes. Furthermore, the coupled dissolution and reverse ion-exchange kinetics for dissolution of HIP complexes into aqueous electrolyte solutions were measured and mathematically modeled.

Methods. HIP complexes of model drugs tacrine and *l*-phenylephrine were formed using linear sodium alkylsulfates and bis (2-ethylhexyl sodium sulfosuccinate). Equilibrium partition coefficients between chloroform and aqueous solutions for the complexes and the kinetics of dissolution of the complexes in buffered aqueous solutions were measured.

Results. The chloroform/aqueous partition coefficients for *l*-phenylephrine/bis (2-ethylhexyl sodium sulfosuccinate) complexes decrease with increasing molar surface tension increment of salts added to the aqueous solution. The logarithm of the partition coefficient for a homologous series of alkyl sulfate complexes decreases as the hydrophilic-lipophilic balance number increases. Dissolution of HIP complexes in deionized water shows first order kinetics, whereas dissolution in aqueous electrolyte solutions shows biphasic kinetics. A kinetic model explains these dissolution rates.

Conclusions. Solubility and dissolution rates for HIP complexes depend on the hydrophobic-lipophilic balance number of the organic counter ion as well as on the electrolyte composition of aqueous solutions. Reverse ion-exchange kinetics are sufficiently slow to allow HIP complexes to be considered simple prodrugs.

KEY WORDS: hydrophobic ion pairs; prodrug; dissolution; solubility.

INTRODUCTION

Hydrophobic ion pairing (HIP) is a technique whereby ionic pharmaceutical compounds and proteins can be directly solubilized in organic solvents (1–6). HIP consists of exchanging small, hydrophilic counterions on the molecule for large hydrophobic organic ions, producing complexes whose solubility in low-dielectric organic solvents is increased by orders of magnitude compared with that of the parent molecule. This technique makes it possible to obtain true homogeneous solutions of ionic compounds in neat organic solvents, with the only requirement being an accessible charge on the molecule. It should be emphasized that HIP is a general process, because nearly any solvent may be used. HIP can be used to increase bioavailability of ionic drugs (7–13).

In contrast to their enhanced solubilities in organic solvents, HIP complexes exhibit reduced solubilities in aqueous media. However, when HIP compounds are dissolved in aqueous electrolyte solutions, reverse ion exchange can occur as small hydrophilic counterions are substituted for the hydrophobic organic ions, resulting in the reformation of the parent drug, with a concomitant increase in drug solubility. HIP complexes may thus be thought of as a simple prodrug that may exhibit sustained-release profiles whose kinetics are determined by the rates of dissolution of the HIP complex and subsequent reverse ion exchange. Little is known about the factors that influence the solubility of HIP complexes or their dissolution and reverse ion-pairing behavior (1–3,14). In this study, we examine the solubility and reverse ion-exchange kinetics for two model drugs, *l*-phenylephrine hydrochloride and tacrine hydrochloride.

EXPERIMENTAL APPROACH

Materials

Sodium chloride, sodium nitrate, dibasic sodium phosphate, sodium sulfate, and chloroform were purchased from Fisher Scientific. Sodium octylsulfate, sodium dodecylsulfate, sodium tetradecylsulfate, sodium octadecysulfate, and tacrine hydrochloride were from Aldrich and *l*-phenylephrine hydrochloride and sodium bis (2-ethylhexyl) sulfosuccinate (AOT) were from Sigma. All chemicals were used as received.

Phosphate buffered saline solutions (PBS) contained 200 mL deionized water, 1600 mg NaCl, 40 mg KCl, 48 mg KH_2PO_4, 288 mg Na_2HPO_4 at a pH of 7.4.

Measurement of Water/Chloroform Partition Coefficients

To examine the effect of anions on partitioning, stock solutions containing 2-mg/mL (9.8 mM) *l*-phenylephrine hydrochloride in aqueous 0.2-M salt ($NaNO_3$, NaCl, Na_2HPO_4, NaH_2PO_4, or Na_2SO_4) were prepared. Separate aqueous stock solutions of AOT were prepared, also at 9.8 mM. Equal volumes of the two solutions were combined, resulting in spontaneous formation of the ion-paired complexes. The pH of solutions containing *l*-phenylephrine/AOT complexes with no added salt was 6.7. The pH did not change upon addition of 0.2 M $NaNO_3$, NaCl, or Na_2SO_4. pH values for *l*-phenylephrine/AOT complex solutions containing 0.2 M Na_2HPO_4 or NaH_2PO_4 were 9.6 and 5.2, respectively. To examine the effect of changing pH, a solution containing *l*-phenylephrine/AOT complex and a mixture of NaH_2PO_4 and Na_2HPO_4 (0.2 M total phosphate) was prepared to obtain a pH of 6.7.

To examine the effect of organic counterion hydrophobicity on partitioning, separate 9.8 mM aqueous stock solutions of hydrophobic organic ions (sodium octylsulfate, sodium dodecylsulfate, sodium tetradecylsulfate, sodium octadecylsulfate, or AOT) were prepared and added to equal

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volumes of 9.8 mM solutions of *l*-phenylephrine hydrochloride in DI water.

An equal volume of chloroform was added to each of the solutions containing HIP complexes. The mixture was then vigorously shaken for 3 min and placed on a rotating mixer at 15 Hz for 1 h (14). The solutions were then centrifuged in a Sorval RC 5C plus centrifuge at 2300 rpm for 30 min. An aliquot of the aqueous supernatant was taken and drug concentration measured using UV spectrophotometry at 274 nm ($\varepsilon = 6.18 \times 10^{-4}$ moles/dm^3/cm). Standard curves constructed with either free *l*-phenylephrine hydrochloride or *l*-phenylephrine /AOT complex showed that the extinction coefficient for *l*-phenylephrine in aqueous solutions is insensitive to pH or added salts (data not shown). The drug concentration in the organic phase was obtained through a mass balance.

A similar experiment was conducted to measure the partition coefficient for *l*-phenylephrine hydrochloride in the absence of added salts and organic counterions. 1 mg/mL *l*-phenylephrine hydrochloride in water was added to an equal volume of chloroform. pH of the *l*-phenylephrine hydrochloride solution was 5.4. Mixing, centrifuging, and analysis were then conducted as above.

Precipitation Ion-Pairing

Solid hydrophobic ion-paired tacrine complexes were formed by a process termed precipitation ion pairing. We prepared a stock solution of 13mM tacrine hydrochloride in de-ionized water and a 13 mM stock solution of one of the three linear hydrophobic organic ions (sodium dodecylsulfate, sodium tetradecylsulfate, and sodium octadecylsulfate). Solutions were warmed to facilitate dissolution into water. After mixing equal volumes of the drug and hydrophobic organic ion solution vigorously for 5 min, the majority of ion-paired complex precipitated out of solution. Samples were centrifuged 25 min at 2300 rpm. The solid and liquid phases were separated, and the solid phase, vacuum dried. After drying, the complexes were ground in a mortar and pestle to a fine powder and washed with 150 mL of deionized water on a Millipore suction filter system to remove tacrine hydrochloride and sodium chloride from the solid complex. The washed ion-paired complex was dried under vacuum. The resulting solid did not appear to be crystalline. NMR analysis of the resulting powder indicated that high purity levels desired were achieved using this procedure (data not shown).

Dissolution Profile Measurement

Dissolution rates for each of the tacrine-HIP complexes were measured as follows. A 20 mg sample of the tacrine-HIP complex was placed in a stirred, 100 mL Erlenmeyer flask containing 50 mL of PBS or deionized water. 1 mL samples were collected at designated times and filtered through a 0.2 μm pore size, PSU syringe filters (Whatman). Tacrine concentration in each sample was measured using UV spectrophotometry at 324 nm ($\varepsilon = 8.26 \times 10^{-5}$ moles/dm^3/cm).

RESULTS AND DISCUSSION

Measurement of Water/Chloroform Partition Coefficients

The hydrophobicity of HIP complexes can be quantified by measuring the partitioning behavior of the complex between water and chloroform. We define the partition coefficient P_{Chlor} as:

$$P_{Chlor} = [l-phenylephrine]_{Chloroform}/[l-phenylephrine]_{aqueous} \quad (1)$$

In the absence of added salts and organic counterions, the partition coefficient for *l*-phenylephrine HCl greatly favors the aqueous phase (log P_{Chlor} = -2.7). Formation of the hydrophobic ion pair greatly increases the hydrophobicity. We measured log P_{Chlor} for the AOT/ *l*-phenylephrine complex in the absence of added salts to be 0.6, representing an increase in partition coefficient of three orders of magnitude compared to the *l*-phenylephrine hydrochloride starting material.

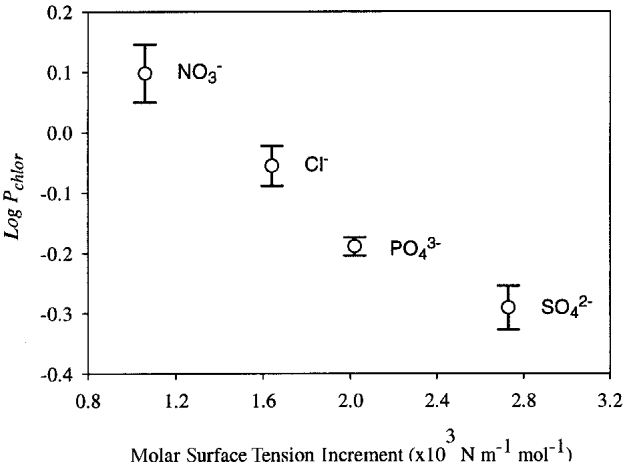

Fig. 1. Effect of the molar surface tension increment (15) of added sodium salts (0.2M) on the chloroform-aqueous partition coefficient of the ion-paired *l*-phenylephrine-AOT complex. The pH of each solution was 6.7. The point marked as $P0_4^{3-}$ was experimentally determined using a mixture of Na_2HPO_4 and NaH_2PO_4 to yield a pH of 6.7, and a PO_4^{3-} concentration of 0.2 M.

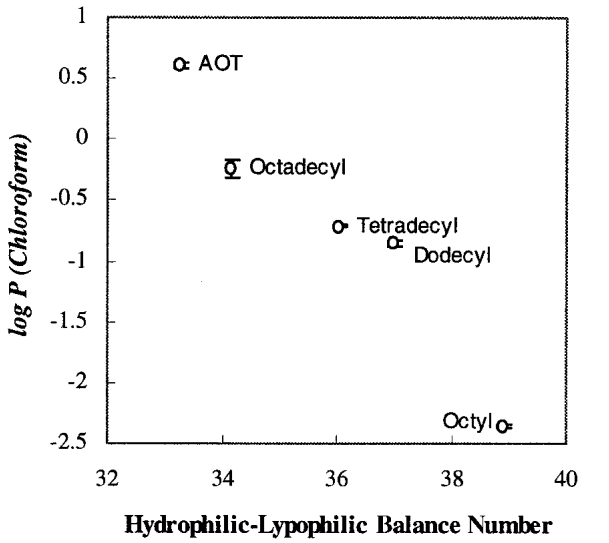

Fig. 2. Effect of hydrophobic organic ion on the chloroform/deionized water partition coefficient of the *l*-phenylephrine HIP complex. The HLB value for AOT has been estimated based on equivalent hydrophilic group number behavior of a sulfosuccinate as a sulfate using the Davies method (17). The pH of each solution was 6.7.

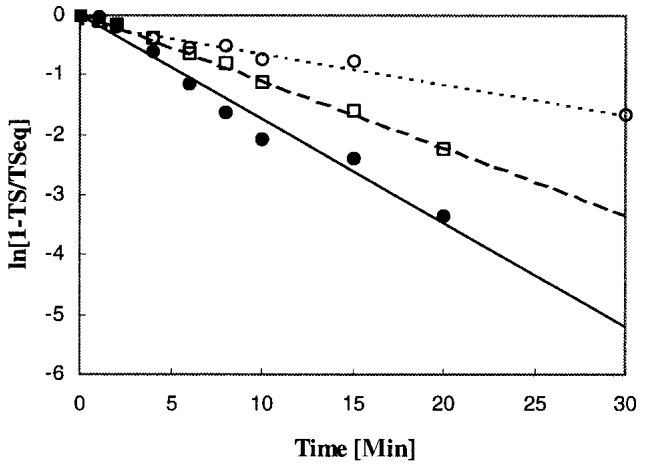

Fig. 3. Dissolution profiles for ion-paired tacrine complexes in deionized water. Closed circles are tacrine dodecylsulfate, open squares are tacrine tetradecylsulfate, and open circles are tacrine octadecylsulfate. Dissolved tacrine HIP complex concentrations, normalized by the equilibrium solubility in deionized water (TS_{eq}) are plotted on a log scale vs. time. The resulting slope, k_1, is the apparent 1st order rate constant for HIP complex dissolution. Values for k_1 are presented in Table I.

Addition of salts to the aqueous phase reduces the partition coefficient by reducing the amount of HIP complex found at equilibrium. Fig. 1 illustrates the strong influence that the presence of various sodium salts (0.2 M) in aqueous solutions has on the partitioning coefficient of *l*-phenylephrine. Melander and Horvath (15) demonstrated that the lyotropic series correlates with the molar surface tension increments of various salts. Counterions with lower molar surface tension increments, such as NO_3^- and Cl^-, exhibit larger partition coefficients. This is to be expected, because the free energy of dissolving a hydrophobic complex can be approximated as the work of forming a cavity of sufficient size to incorporate the complex (15,16). This work increases as the surface tension increases. Thus, as surface tension increases, equilibrium in the aqueous phase shifts to favor the smaller less hydrophobic *l*-phenylephrine salts (e.g., *l*-phenylephrine hydrochloride, *l*-phenylephrine sulfate) over the more hydrophobic HIP complex. This increases the aqueous solubility of *l*-phenylephrine compared with the solubility in the chloroform phase, where essentially only the *l*-phenylephrine-AOT complex is soluble.

Solution pH also weakly affects the partition coefficient of the *l*-phenylephrine-AOT complex. 0.2 M phosphate solutions containing Na_2HPO_4 (pH 9.6), NaH_2PO_4 (pH 5.2), or Na_2HPO_4/NaH_2PO_4 mixtures (pH 6.7) had log P_{Chlor} values of –.31, –.11, and –.19, respectively.

Fig. 2 shows the effect of the organic ion's hydrophobicity on the partitioning behavior of various HIP complexes. Hydrophilic-lipophilic balance numbers (HLB) quantify the hydrophobicity of each organic ion (17). We found that log P_{Chlor} values of the *l*-phenylephrine ion-paired complex decrease monotonically as the HLB number increases for the homologous alkyl sulfate series. The decrease in log P_{Chlor} is roughly linear as a function of HLB ($r^2 = 0.91$). Thus, partitioning behavior between an aqueous electrolyte solution and an organic solvent for a given drug complex may be modulated both by the electrolyte composition of the aqueous phase and the hydrophobicity of the organic ion.

Kinetic Behavior

In aqueous solutions containing hydrophilic electrolytes, HIP complexes may undergo "reverse" ion pairing, wherein the hydrophobic anion on the HIP drug complex exchanges with a hydrophilic anion. If a pure, solid HIP complex is added to an aqueous electrolyte solution, the HIP complex will first dissolve and then subsequently undergo reverse ion pairing, yielding a biphasic dissolution profile. We measured the dissolution profiles for several drug-HIP complexes (tacrine paired with dodecylsulfate, tetradecylsulfate, or octadecylsulfate) in both de-ionized water and PBS. In deionized water, only the dissolution of the HIP complex is possible; in PBS, both dissolution and subsequent reverse ion pairing occurs.

Fig. 3 shows the dissolution profile of the various tacrine-HIP complexes in de-ionized water. The dissolution behavior is well modeled by a single, first order rate constant, k_1 [Eq. (2)]. The rate constant observed in this experiment can be influenced by a number of factors, e.g., the amount of sample, the sample-water surface area, and the mixing conditions. Although mixing conditions were held constant, the specific surface area was presumably different for each preparation. Therefore, k_1, which incorporates an effective specific surface area, cannot be considered an intrinsic property of the complex, in contrast to the ion-exchange rate constant provided later. However, the relative dissolution rates can be compared and as expected, dissolution rates are slower for more hydrophobic complexes.

$$\frac{d[TS]}{dt} = -k_1[TS - TS_{eq}] \qquad (2)$$

Table I. Solubilities and Rate Constants for Dissolution and Reverse Ion Exchange of Hydrophobic Ion-Paired Tacrine Complexes

Tacrine Alkylsulfate Complex	Rate Constants				Solubility Limits		
	k_1 min^{-1}	k_{2f} M^{-1} min^{-1}	k_{2r} M^{-1} min^{-1}	r^2	TS in DI-water μg/mL	Tacrine (total) in PBS μg/mL	*TS* in PBS[b] μg/mL
Dodecyl	0.17	0.027	n.d.[a]	.99	20.7	26.3	25.5
Tetradecyl	0.11	0.018	21.5	.99	4.2	28.7	12.8
Octadecyl	0.05	0.002	31.8	.95	0.9	6.5	3.9

[a] Rate constant cannot be determined; values are too large to measure accurately.
[b] Computed

In PBS solutions, dissolution is biphasic. The solid tacrine HIP complex (*TS*) rapidly dissolves in the solution up to the complex's solubility limit. Once in solution, anions (e.g., Cl^-) exchange with the hydrophobic organic ions, with a bimolecular rate constant k_{2f}. Reformation of the HIP complex also occurs, with a corresponding bimolecular rate constant k_{2r}, as diagrammed below in Scheme A.

$$\underset{\text{Phase Change}}{[TS]_{solid} \xrightarrow{k_1} [TS]_{aq}} \qquad \underset{\text{Ion Exchange}}{[TS]_{aq} + [NaCl]_{aq} \underset{k_{2r}}{\overset{k_{2f}}{\leftrightarrow}} [TCl]_{aq} + [NaS]_{aq}}$$

Scheme A. Dissolution and reverse ion exchange of HIP complexes. *TS* is the tacrine HIP complex and *S* is the hydrophobic organic ion.

The measurement technique employed in this study does not distinguish between the tacrine-hydrochloride (*TCl*) and tacrine HIP (*TS*) compounds in solutions. Therefore, to determine the value of k_{2f} and k_{2r} for the experiments where both dissolution and reverse ion exchange occur, the following series of equations must be solved.

$$\frac{d[T_{measured}]}{dt} = \frac{d[TS]}{dt} + \frac{d[TCl]}{dt}$$

$$\frac{d[TS]}{dt} = k_1[TS - TS_{equ}] - k_{2f}[TS][NaCl] + k_{2r}[TCl][NaS] \qquad \text{(3 abc)}$$

$$\frac{d[TCl]}{dt} = k_{2f}[TS][NaCl] - k_{2r}[TCl][NaS]$$

where $T_{measured}$ is the total tacrine concentration measured in solution ($= TS + TCl$). To estimate the parameters needed for this model, we first fit k_1 to data presented in Fig. 3. Results of the fit are given in Table I. The solubility limit of the tacrine HIP complex (TS_{eq}) is affected by electrolyte concentrations in the aqueous solution. We, therefore, estimate TS_{eq} by fitting the first few data points for dissolution into

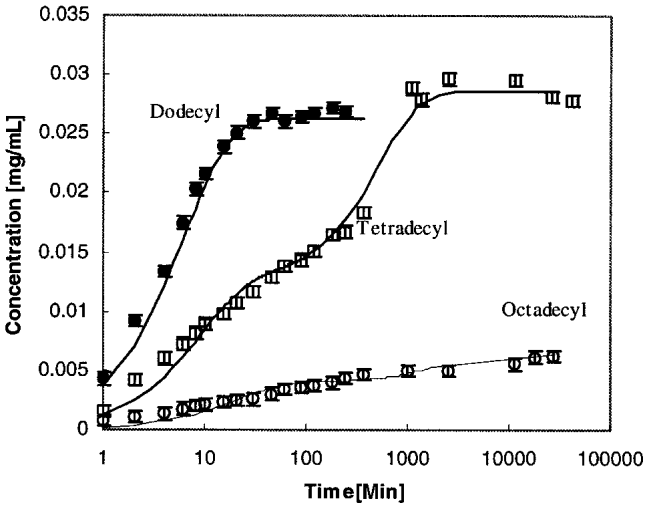

Fig. 4. Dissolution profiles for ion-paired tacrine complexes in hosphate buffered saline solutions with release profiles predicted from the model. The estimated standard errors for the model predictions are 1.1, 1.0, and 0.5 μg/mL and the standard deviation for the experimental measurements never exceeded 1.6, 1.6, or 1.2 μg/mL for tacrine dodecylsulfate, tacrine tetradecylsulfate, and tacrine octadecylsulfate respectively.

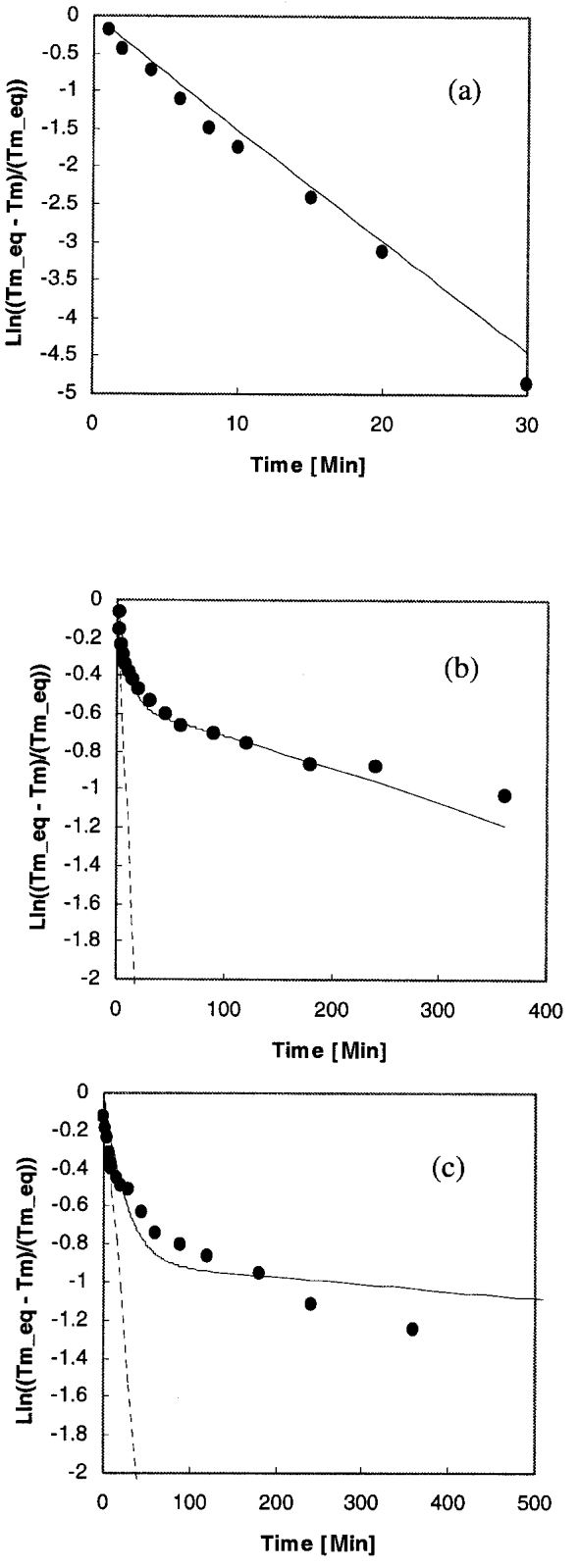

Fig. 5. Plots of the ln (cumulative % release) vs. time for ion-paired tacrine complexes in hosphate buffered saline solutions with predicted release profiles (solid lines indicate full model that includes reverse ion pairing; dashed lines indicate a simple model with a single dissolution rate). Tacrine dodecylsulfate (a) does not demonstrate a biphasic profile, whereas both tacrine tetradecylsulfate (b) and octadecylsulfate (c) do.

PBS to equation 3b, neglecting the last two terms on the right hand side. Neglect of these terms is warranted because at these early time points very low concentrations of *TS* or *TCl* are present. Finally k_{2f} is fit using all of the data, and k_{2r} is computed from mass balance equations. A summary of fitted and computed parameters is given in Table I. The fitted curves for total tacrine dissolution and the measured curves agree well with collected data having correlation coefficients near 0.99 (Fig. 4).

Equilibrium aqueous solubilities of *TS* decrease as the hydrophilicity (HLB) of the organic ion decreases, and show a significant "salting in" effect in PBS solutions (Table I). The effect of hydrophobicity on both solubility and ion exchange rates provides for dramatic differences in overall dissolution profiles. At saturation conditions, this results in large changes in the ratio of HIP-tacrine to total tacrine. Tacrine dodecylsulfate dissolution does not reveal a measurable difference between the HIP-tacrine concentration and the total tacrine concentration. Combining this observation with the extremely fast k_{2r} computed suggests that the dissolution of tacrine dodecylsulfate could be just as well described by a single kinetic dissolution rate constant. This hypothesis is verified by the lack of a biphasic dissolution profile observed in Fig. 5a.

Unlike tacrine dodecylsulfate, tacrine tetradecylsulfate and tacrine octadecylsulfate demonstrate biphasic behavior in Fig. 5 b and c. As the linear organic ion becomes more hydrophobic the "reverse" ion-pairing reaction must be included in the dissolution model to obtain agreement. The visible deviations observed in Fig. 5c are generated by the very low drug concentrations in solutions thus more representative of measurement uncertainty than poor modeling. After the initial dissolution to the *TS* equilibrium solubility limit, the release rate of the drug into solution becomes dominated by the ion-exchange kinetics. Increasing the hydrophobicity of the organic ion slows the subsequent dissolution.

This effect of hydrophobicity on dissolution rates may be of particular relevance to pharmaceutical applications. In preliminary work, we have found that HIP complexes cross cell walls more readily than the hydrophilic parent compound, and HIP antibiotics show lower minimum inhibitory concentrations than the unpaired compounds (unpublished results). Control of the ion exchange process through organic counterion characteristics is likely to provide a mechanism to control pharmacokinetics and pharmacodynamics *in vivo*. As seen in Fig. 4 and Table I, decreasing the hydrophobicity of the pairing counterion increases the quantity of drug dissolved or becoming available for circulation within salt solutions compared to organic ions that exhibit higher hydrophobicity. The actual time duration for release *in vivo* is expected to depend upon the type of organic counterion used, the circulation rate characteristic of the administration site and the pH of the local environment.

Importantly, we demonstrated that hydrophobic ion-paired molecules do not instantaneously undergo reverse ion exchange when placed an ionic aqueous environment. Rather, the dissolution rate of a hydrophobic ion-paired complex is governed by the ionic strength of the aqueous environment and the hydrophobic character of the organic ion. The more hydrophobic the organic counterion the greater the influence the ion-exchange process has on the overall dissolution kinetics of a solid HIP complex.

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