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Kinetic Salt Effect in Pharmaceutical Investigations

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Abstract □ The use of the kinetic salt effect in kinetic investigations has been widespread since its original derivation by Brønsted and Bjerrum in 1925. Its derivation is tied in with the Debye-Hückel theory, and it would be expected that the concentration range of applicability of the kinetic salt effect would be confined to that of the corresponding Debye-Hückel expression, *i.e.*, less than 0.01 *M*. A review of the pharmaceutical literature shows that applications may be extended to higher concentration ranges. Deviations from the Debye-Hückel expressions by the charged reactants and by the transition complex may be of the same magnitude and sign, and this may be the cause for the concentration range extension.

Keyphrases □ Kinetic salt effect—pharmaceutical solutions □ Ionic strength effect—high ionic concentrations □ Hydrolysis—kinetic salt effect □ Degradation, drug—kinetic salt effect

Properly conducted kinetic studies always, directly or indirectly, take into account the so-called kinetic salt effect. By varying the ionic strength by addition of an inert electrolyte (*e.g.*, NaCl) while keeping other concentrations constant, the rate constants for reacting species will either increase, remain constant, or decrease. Accordingly, the effect is denoted positive, absent, or negative. As shall be seen in the following discussion, the sign or the absence of the kinetic salt effect is a valuable aid in interpretation of mechanisms. Strictly quantitative relations between rate constants and ionic strength are only theoretically valid at exceedingly low concentrations. Since pharmaceutical investigations are most often conducted at ionic strength ranges higher than the theoretical limits, a review of findings from kinetics of pharmaceutical model systems might throw light on the actual range to which the kinetic salt effect can be extended.

THEORY

It can be shown (1) by means of transition-state theory that for a reaction in solution:

$$A + B \rightarrow [AB^{\dagger}] \rightarrow \text{products} \qquad \text{(Eq. 1)}$$

the rate constant, k, is related to the activity coefficients, γ, of the reactants (A and B) and the transition complex ($[AB^{\dagger}]$) by:

$$\log k = \alpha + \log [\gamma_A \gamma_B / \gamma_{[AB^{\dagger}]}] \qquad \text{(Eq. 2)}$$

The charges, z, of the three species are related to one another by $z_A + z_B = z_{[AB^{\dagger}]}$. Applying this and the Debye-Hückel limiting law:

$$\log \gamma = -Q \cdot z^2 \sqrt{\mu} \qquad \text{(Eq. 3)}$$

Table I—Values of $2Q = 3.65 \cdot 10^6 \cdot [\rho/\epsilon^3 T^3]^{0.5}$ at Various Temperatures

Temperature	$2Q^a$
20	1.008
25	1.018
30	1.026
35	1.036
40	1.046
45	1.057
50	1.068
55	1.079
60	1.092
70	1.117
80	1.145
90	1.174
100	1.198

[a] ϵ-values used in the computation are from *Reference 43*, and ρ-values are from *Reference 44*.

to Eq. 2 for a solution of overall ionic strength, μ, leads to the well-known Brønsted-Bjerrum equation (2–5):

$$\log k = \alpha + 2 \cdot Q \cdot z_A \cdot z_B \cdot \sqrt{\mu} \qquad \text{(Eq. 4)}$$

where $2Q = 1.018$ for aqueous solutions at 25°. Since $Q = 1.825 \cdot 10^6 \cdot [\rho/\epsilon^3 T^3]^{0.5}$, where ϵ is the dielectric constant, ρ is density, and T is the absolute temperature, the coefficient to $\sqrt{\mu}$ changes with temperature. A list of values of $2Q$ is given in Table I.

The Debye-Hückel equation is usually only obeyed in ionic strength ranges up to 0.01 (6). Figure 1 is an example of this showing the mean activity coefficients of hydrochloric acid in potassium chloride solutions of varying ionic strength (7). Equation 4 is, therefore, only strictly applicable up to this concentration.

The modified Debye-Hückel equation for higher concentrations is

$$\log \gamma_{\pm} = \frac{z^2 \cdot Q \cdot \sqrt{\mu}}{1 + \beta\sqrt{\mu}} \qquad \text{(Eq. 5)}$$

and holds up to an ionic strength of about $\mu = 0.1$ (8). Using this in the development outlined for Eq. 4 yields

$$\log k = \alpha + 2 \cdot Q \cdot z_A \cdot z_B \cdot \frac{\sqrt{\mu}}{1 + \beta\sqrt{\mu}} \qquad \text{(Eq. 6)}$$

It is recalled that μ is the overall ionic strength, but β depends on the ionic diameter of the reacting species, and this usually is unknown. Linearity and slopes, however, are not very sensitive to the magnitude of β, which is always close to unity; it is the practice of some authors (9) to test kinetic salt effects with this assumption, *i.e.*,

$$\log k = \alpha + 2 \cdot Q \cdot z_A \cdot z_B \cdot \frac{\sqrt{\mu}}{1 + \sqrt{\mu}} \qquad \text{(Eq. 7)}$$

The slope of such a plot should be close to the value of $2 \cdot Q \cdot z_A \cdot z_B$ (which is not necessarily an integer, all depending on tem-

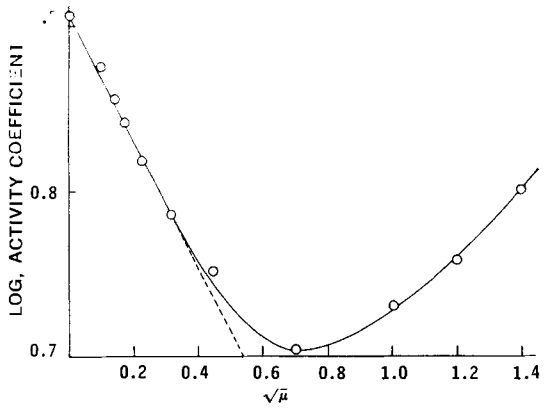

Figure 1—*Logarithm of the activity coefficient of hydrochloric acid as a function of the square root of ionic strength (2).*

perature) and should allow evaluation of the integer value of the product of z_A and z_B.

In the case of higher ionic strengths, an ionic strength effect is possible even if one or both reacting molecules are uncharged. At high ionic strength, if A was charged and B not, then reaction $A + B \rightarrow [AB^{\dagger}]$ would dictate that $z_{\dagger} = z_A$. At high ionic strength, the following relation (10) often holds:

$$-\log \gamma_A = -\log \gamma_{\dagger} = \frac{z^2 \cdot \alpha \cdot \sqrt{\mu}}{1 + \beta\sqrt{\mu}} - b \cdot \mu \qquad \text{(Eq. 8)}$$

At high ionic strength (11), the activity coefficient of the uncharged species adheres to

$$\log \gamma_B = b_B \mu \qquad \text{(Eq. 9)}$$

which, by a treatment similar to the one leading to Eq. 7, yields

$$\log k = \log k_0 + [b_A + b_B - b_{\dagger}] \cdot \mu \qquad \text{(Eq. 10)}$$

i.e., the logarithm of the rate constant should be proportional to the ionic strength. In the case of two uncharged molecules, application of Eq. 9 by itself would lead to an equation of the type of Eq. 10.

Aside from these so-called primary salt effects, a secondary salt effect at times is important. In buffer-catalyzed degradations, an ionized species B^- may be catalytic, whereas the corresponding acid, HB, may not be. The ionization constants of the acid are affected in a manner similar to that described by Eqs. 4 and 10. Increasing ionic strength can affect the concentration of B^- and, in this indirect manner, influence the value of the rate constant.

EXAMPLES

Zero Effect—One of the values of the kinetic salt effect as an experimental tool is that if a decomposition does not exhibit a kinetic salt effect, then reactions of two charged species can be eliminated from consideration, since either z_A or z_B would have to be zero. Examples of this are hydrolysis of the following compounds in aqueous solution: streptozotocin (12), streptovaricin (13), diethylaminoethylsalicylate hydrochloride (14), acetylsalicylic acid anhydride (15), procaine hydrochloride (16, 17), chloramphenicol (18), *N*-acetyl-*p*-aminophenol (19), chlorobutanol (20), ascorbic acid (21, 22), phenobarbital (23), idoxuridine (24), penicillin at low pH (25), morphine (26), cycloserine (27), and hydroxocobalamin (28).

Integer Effects—Where kinetic salt effects do exist, they imply that charged species are of identical or opposite charge. Since even model pharmaceutical systems by necessity are in concentration ranges above 0.01 ionic strength, Eq. 4 cannot be expected to hold. In all of the cases to be cited (29–38), however, linearity prevails, but the slopes differ from $2 \cdot Q \cdot z_A \cdot z_B$, and other evidence has been employed by the respective authors to support proposed mechanisms.

Brooke and Guttmann (29) used kinetic salt effect data in investigating the complex formation between 3-carbomethoxy-1-methylpyridinium cation (NME) and 8-chlorotheophyllinate anion (CT). Figure 2 shows their data plotted both according to Eqs. 4 and 7 and (in spite of the scatter) implies the applicability of Eq. 7

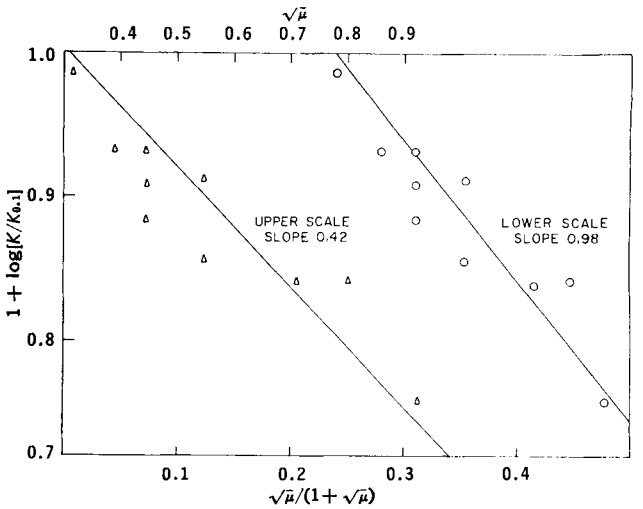

Figure 2—*Kinetic salt effect on degradation of 3-carbomethoxy-1-methylpyridinium cation (29) at 30°, plotted according to Eq. 4 (△, upper scale) and according to Eq. 7 (○, lower scale).*

up to an ionic strength of 1.2. When plotted according to Eq. 7, the slope is close to the theoretical.

Hussain *et al.* (30), in studying the degradation of echothiophate in aqueous solution, supported their proposed mechanism by a negative kinetic salt effect and found linearity up to an ionic strength of 0.02 when plotting was performed according to Eq. 4; the slope (0.9) is close to the theoretical as shown in Fig. 3. It is seen from Fig. 3, however, that use of Eq. 7 extends the useful range to an ionic strength of 0.2 (but not 0.5), still yielding a slope (0.91) close to the theoretical.

Felmeister *et al.* (31) employed the kinetic salt effect to support their contention that dismutation of the semiquinone free radical of chlorpromazine ($C\cdot$) took place *via* a reaction of the type $C\cdot^{++} + H^+ \rightarrow [HC\cdot^{+++}]$. However, they point out that at pH 1.96, the species $H_2C\cdot^{+++}$ might also react with H^+ to form a transition complex. The former type, at 25°, should yield a positive slope of 2.036, as opposed to the value of 1.6 found when plotting was performed according to Eq. 4. As shown in Fig. 4, plotting according to Eq. 7 yields a slope of 2.3, *i.e.*, somewhat closer to the theoretical value, and may be implying that $H_2C\cdot^{+++} + H^+$ plays a part in the overall scheme. The useful range would appear to be up to $\mu = 0.2$, but tabulated data (31) would imply linearity according to Eq. 7 (but not Eq. 4) up to $\mu = 0.5$.

Finholt *et al.* (25) demonstrated a positive kinetic salt effect in penicillin degradation at alkaline pH. Their data are shown in Fig. 5, plotted according to both Eqs. 4 and 7. At pH 6.8, the positive effect is used to postulate penicillin (P) degradation according to the scheme $P^- + HPO_4^= \rightarrow$ products, and the slope of 2 resulting from Eq. 7 would support such a view. The probable mechanism at pH

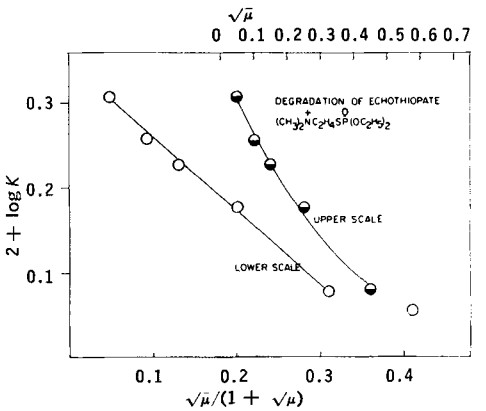

Figure 3—*Kinetic salt effect on degradation of echothiophate at 22° (30), plotted according to Eq. 4 (◒, upper scale) and according to Eq. 7 (○, lower scale).*

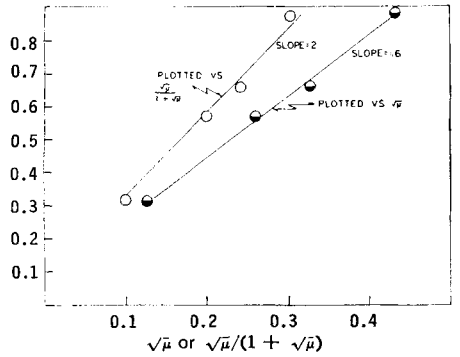

Figure 4—*Kinetic salt effect on degradation of chlorpromazine at 25° (31), plotted according to Eq. 4 (◒) and according to Eq. 7 (○).*

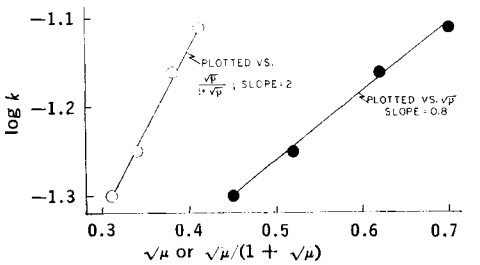

Figure 5—*Kinetic salt effect on degradation of penicillin in phosphate buffer at pH 6.8 and 60° (25), plotted according to Eq. 4 (○) and according to Eq. 7 (●). Plots in borate buffer at pH 8.75 parallel the lines shown in the figure.*

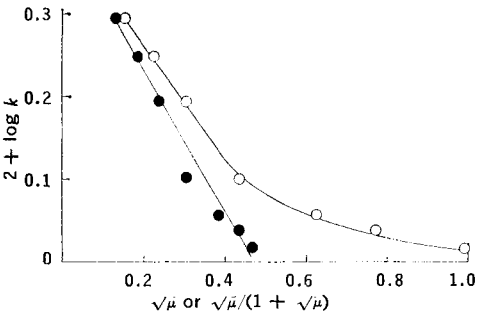

Figure 6—*Kinetic salt effect on degradation of thiamine at pH 6.40 and 96.4° (34), plotted according to Eq. 4 (○) and according to Eq. 7 (●).*

8.75 would be a reaction of the type $P^- + HBO_3^= \rightarrow$ products. This is not in conflict with the reported pH profile since, well below the pK_2 of boric acid, the logarithm of the concentration of $HBO_3^=$ is proportional to pH. In this case, use of Eq. 7 appears valid to an ionic strength of 0.5.

Koshy and Mitchner (32) studied the hydrolysis of 2-(4-phenyl-1-piperazinylmethyl)cyclohexanone (X) at 60°. At pH 5 the kinetic salt effect yields a slope of −0.61 when plotted according to Eq. 4 and a slope of −1.29, *i.e.*, close to the theoretical, when plotted according to Eq. 7. This strongly supports the authors' contention (32) that $X^+ + B^- \rightarrow$ products is the predominant reaction. In this case, both Eqs. 4 and 7 yield linearity up to $\mu = 0.4$.

Fractional Effects—The theoretical slope values, of course, can only be expected when the possibility of parallel reactions can be eliminated (unless both of the same ionic makeup). Szulczewski *et al.* (33) studied the isomerization and hydrolysis of 4,6-diamino-1-(3,5)-dichlorophenyl-1,2-dihydro-2,2-dimethyl-1,3,5-triazine (I) at 65° and a pH of 1.85 and found it to involve both IH^+ and I. When plotted according to Eq. 4, their data yield a slope of 0.5, whereas use of Eq. 7 results in the value 0.86, *i.e.*, still somewhat removed from the theoretical value. Equations 4 and 7 here hold up to $\mu = 0.16$.

A similar situation exists in the degradation of thiamine (34). Here, again, plotting (Fig. 6) according to Eq. 7 extends the useful range of ionic strength to $\mu = 0.22$, but the slope (0.87) is not close to the theoretical. As pointed out by the authors (34), secondary salt effects may be of significance in their system.

The positive kinetic salt effect reported for acid hydrolysis of niacinamide (N) at 89.4° by Finholt and Higuchi (35) yields a slope value of 0.1 if plotted according to Eq. 4 and a slope of 0.6 if plotted according to Eq. 7. Therefore, as pointed out by the authors (35), both $N^+ + H^+$ and $N + H^+$ must be involved, with rate constants of the same order of magnitude.

Other examples of fractional effects are the study on barbital degradation by Goyan *et al.* (36) and the degradations of diethylaminoacetylsalicylate hydrochloride (37) and methylpyrrolidylacetylsalicylate hydrochloride (38) reported by Garrett.

Behavior at High Ionic Strength—At ionic strengths above 1, Eq. 10 may be expected to hold, but the *b*-values are mostly of such low magnitude that absence of ionic strength effect would result in the cases to which it applies. There are three cases in recent pharmaceutical literature where Eq. 10 applies: (*a*) Schroeter's investigation of salicyl alcohol interaction with sulfites (39); (*b*) the investigation of Siegel *et al.* (40) of the hydrolysis of methyl DL-α-phenyl-2-piperidylacetate; and (*c*) Hou and Poole's investigation of degradation of ampicillin at low pH (41).

DISCUSSION

The only case in recent pharmaceutical literature where the extended Debye-Hückel equation has been used is the (nonkinetic) study by Zografi *et al.* (42). The intents of this communication have been to advocate the advantages of using Eq. 7 as a plotting procedure when kinetic salt effects are under study, and to point to the fact that the concentration range in which it may be used may be as high as unity ionic strength, *i.e.*, well beyond the range of both Eqs. 3 and 5.

Table II lists, in tabulated form, the slope values and ionic strength ranges extracted from this survey. It is quite obvious that the range of applicability is considerably higher than what would be expected from the limiting μ-ranges for Eqs. 3 and 5. Only in the case of echothiophate is the maximum allowable ionic strength definitely less than 0.5. Since Eq. 8 holds for ionic strength ranges above $\mu = 0.1$, an equation of type 7 can be derived to yield:

$$\log k = \log k_0 + \frac{2 \cdot z^2 \cdot Q\sqrt{\mu}}{1 + \beta\sqrt{\mu}} - [b_A + b_B - b\dagger] \cdot \mu \quad \text{(Eq. 11)}$$

Table II—Slope Values and Ionic Strength Ranges for the Kinetic Salt Effect in Degradations Reported in Pharmaceutical Literature

Substance	Reference	Slope			Maximum Ionic Strength, μ	Temperature
		Eq. 4	Eq. 7	2Q		
3-Carbomethoxy-1-methylpyridinium cation	29	0.42	1.2	1.03	$\mu_{max.} > 1.2$	30
Echothiophate	30	0.9	0.9	1.01	$0.2 < \mu_{max.} < 0.5$	22
Chlorpromazine	31	1.6	2.3	2.04	$\mu_{max.} > 0.5$	25
Penicillin	25	0.8	2.0	2.18	$\mu_{max.} > 0.5$	60
2-(4-Phenyl-1-piperazinylmethyl)cyclohexanone	32	−0.61	−1.29	−1.09	$\mu_{max.} > 0.4$	60
4,6-Diamino-1-(3,5)-dichlorophenyl-1,2-2,2-dimethyl-1,3,5-triazine	33	0.5	0.86	1.10	$\mu_{max.} > 0.16$	65
Thiamine	34	−0.37	−0.87	−1.19	$\mu_{max.} > 0.22$	96.4
Niacinamide	35	0.1	0.6	1.17	$\mu_{max.} > 0.8$	89.4

An explanation for the extended range could be that, in most cases, the factor $[b_A + b_B - b_\dagger]$ is close to zero. Both b_A and b_B can be determined experimentally by measuring activity coefficients as a function of ionic strength for the reactants A and B. Of course, $b_\dagger$ is not subject to direct experimental determination.

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In Vitro and *In Vivo* Chlorpromazine Availability from Flocculated Polysalt Complex Systems

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Abstract □ An insoluble drug–polysalt complex of chlorpromazine hydrochloride, sodium carboxymethylcellulose, and protamine sulfate was selected as a model to evaluate the effects of these macromolecular constituents on the *in vitro* and *in vivo* availability of the interacted drug. The *in vitro* liberation of drug from the polysalt complex was studied in simulated gastrointestinal fluids as a function of particle size, pH of formation of the complex flocculate, and presence and absence of enzymes in the medium. The *in vitro* drug-release studies conducted under these varying conditions suggested that the product possessed prolonged-release properties. In contrast, the *in vivo* studies with rats revealed a promoted bioavailability of the drug in the presence of the polysalt complex. Protamine sulfate, a known pinocytotic inducer, was observed to be specifically implicated in this phenomenon.

Keyphrases □ Flocculated complex systems, polysalt—chlorpromazine-^{35}S availability □ Chlorpromazine-^{35}S-polysalt complex systems—*in vivo–in vitro* availability □ Release rates—chlorpromazine-^{35}S-polysalt complex □ pH effect—chlorpromazine-^{35}S-polysalt complex stability □ Scintillometry—analysis

The physical form of a polyelectrolyte salt complex (polysalt) is a function of the charge densities of the interacting species. Macromolecules with high charge densities tend to form precipitates (flocculates), while those with lower charge densities tend to form a gel (coacervate) or quasiliquid product (1). The use of complex coacervates as a means of microencapsulating drugs has been claimed in a number of patents (2–7). Luzzi and Gerraughty (8, 9) have noted that complex coacervates of gelatin and acacia with pentobarbital may be employed to control the liberation of the encapsulated material into gastrointestinal fluids. They studied the release rate of the drug as a function of pH, temperature, ratio of solid to encapsulating material, quantity of hardening agent (formaldehyde), and addition of surfactants. In contrast, polysalt flocculates have received relatively little attention in the pharmaceutical literature.

The present drug–polysalt system was chosen on the basis of preliminary studies. It was found that a polysalt of sodium carboxymethylcellulose (NaCMC) and protamine sulfate interacted with appreciable quantities of