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An Experimental Investigation of the Interaction of Aspirin with Urea in Water

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AN EXPERIMENTAL INVESTIGATION OF THE INTERACTION OF ASPIRIN WITH UREA IN WATER

BY

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ABSTRACT

A study of the interaction of aspirin with urea in water was initiated in an effort to obtain evidence of complex formation between these compounds and to observe possible changes in aspirin stability resulting from complexation. A solubility method was used to detect complexation and consisted of observing changes in the solubility of aspirin in the presence of varying urea concentrations at pH 2.0 and at pH 3.5. The rate of degradation of aspirin in the presence of various urea concentrations was observed at five pH values. All data was obtained at 30.0°C. The results indicated that urea markedly affected the solubility of aspirin. The solubilization was attributed to complex formation and apparent equilibrium constants for one to one and two to one species were obtained from the solubility curves. An analysis of the degradation data indicated that the aspirin-urea interaction caused faster aspirin hydrolysis at pH values lower than 2.6 and inhibited aspirin hydrolysis at pH values from 2.6 to 3.5. This effect could be attributed to changes in the aspirin species, i.e., different charges at different hydrogen ion concentrations, as well as changes in the nature of the aspirin-urea complex. The rate of degradation of aspirin in the complex at pH 2.0 was calculated and was reasonably constant over a wide concentration of urea.

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I. INTRODUCTION

In recent years the application of complexation to pharmaceutical systems has been the subject of several extensive investigations. Lach and Pauli have reviewed most of the pertinent literature (1). More recently, the formation of molecular complexes with urea has been singled out for study by various authors. Bolton has reviewed the literature on the subject of urea as a complexing agent for various pharmaceuticals, including oxytetracycline, benzocaine, sulfonamides, wool fat alcohols, quinoxaline, detergents and barbituric acid derivatives (2).

Since complex formation may affect the solubility and stability characteristics of certain drugs, its importance in pharmaceutics is obvious (1, 3, 4). Some investigators, for example, have pointed out the possibility of using complex formation as an approach to the stabilization of drugs undergoing hydrolytic degradation (3, 5, 6).

The above considerations, as well as reports of complexation between urea and various monohydroxybenzoic acids (2, 7), led to the speculation that aspirin, a drug which is known to undergo hydrolytic degradation (8, 9, 10), might react with urea and that the interaction products might be useful as a means of inhibiting

or slowing the hydrolysis of aspirin.

In this investigation, solubility and degradation studies were used to obtain information concerning the interaction of aspirin and urea. The kinetic studies of the degradation of aspirin in the presence of urea were performed in order to draw some general conclusions on the stability of an aspirin-urea complex.

II. EXPERIMENTAL

Reagents

Acetylsalicylic Acid U.S.P., recrystallized from 95 per cent ethanol, m.p. 133-135°C; Urea N.F., recrystallized from absolute ethyl alcohol, m.p. 132-133°C; Salicylic Acid U.S.P., recrystallized from 95 per cent ethanol, m.p. 158-159°C; formic acid C.P.; hydrochloric acid C.P.; sodium hydroxide 10 N, Fisher reagent; Alcohol U.S.P.

Equipment

Beckman spectrophotometer[1]; Beckman pH meter[2]; constant temperature water bath[3], set at 30.0±0.2°C, fitted with a mechanical shaker consisting of a rotating wheel which can be submerged vertically into the bath thus allowing vials fixed on its perimeter to rotate within the bath.

Procedure

1. Complexing studies:

The procedure used for the study of the interaction of aspirin and urea was essentially the same as that used by Higuchi and Zuck (11). Accurately weighed quantities of aspirin, well in excess of the solubility limits of aspirin at 30.0°C, were placed in 15 ml vials.

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[1]Beckman DU Spectrophotometer with automatic power supply, Beckman Instruments Inc., 2500 Harbor Blvd., Fullerton, Calif.

[2]Beckman Zeromatic pH meter, Beckman Instruments Inc.

[3]Labline Instruments Inc., Chicago, Ill. Cat. No. 3052.

Measured amounts of urea solutions of varying concentrations at a selected pH were introduced into the vials. The vials were placed into a water bath at 30.0°C and the solutions were allowed to equilibrate for five hours. After this period of time, a clear aliquot portion of solution was removed from each vial, diluted to the proper concentration, and the aspirin concentration was determined spectrophotometrically (see page 7, this work).

Complexing studies were made at pH 2.00 and pH 3.50. The amounts of aspirin used per 10 ml of solution were 0.5 Gm at pH 2.00 and 1.0 Gm at pH 3.50. The buffers used were:

1. pH 2.00-hydrochloric acid in water.
2. pH 3.50-formic acid and sodium formate.

One to ten Molar urea solutions at pH 2.00 and at pH 3.50 were prepared by dissolving the correct amount of urea into the appropriate buffer and adjusting the final pH of each solution with hydrochloric acid and formic acid respectively. A period of five hours was established as optimum for the solutions to equilibrate by experimentation. Excess quantities of aspirin were shaken together with urea solutions of varying concentrations at pH 2.00 and a clear aliquot portion of the different solutions removed at intervals of one hour and analyzed for aspirin concentration. The results indicated

no appreciable change in aspirin concentration after five hours. The shortest period of time needed to achieve equilibrium was selected in order to keep the decomposition of aspirin to a minimum. A longer period of time would have resulted in the formation of salicylic acid in appreciable amounts, which would cause difficulties in the determination of aspirin concentrations (i.e., salicylic acid shows some absorbance at the wavelength where aspirin absorbance is at maximum) and possible complications due to a urea-salicylic acid interaction (2).

In order to collect samples free of undissolved solid particles, the pipettes were connected, by means of a small section of neoprene tubing, to a small section of glass tubing into which fine glass wool had been packed.

The pH of all solutions was checked after the completion of each complexing study and no appreciable changes were observed.

2. Kinetic studies:

Kinetic studies of aspirin degradation in urea solutions of concentrations 2, 4 and 8 M were made at pH values of 2.00, 2.50, 2.75, 3.00 and 3.50. All data was obtained at 30.0 ± 0.2°C. The buffers were as follows:

1. pH 2.00 hydrochloric acid in water
2. all other pH's formic acid and sodium formate.

The urea solutions were prepared as described under the procedure for complexing studies.

Accurately weighed 75 mg portions of aspirin were introduced into 50 ml volumetric flasks, dissolved with the aid of two ml of 95 per cent ethanol, and brought to volume with the appropriate buffer and urea solutions. The concentration of 75 mg of aspirin per 50 ml is well below the solubility limits of aspirin at 30.0°C and resulted in clear solutions. The flasks were then introduced into a water bath at 30.0°C and were allowed to come to equilibrium temperature before the zero time reading was taken. Aliquots were withdrawn from the flasks at accurately measured intervals of time, diluted to the proper concentration and the absorbance immediately read on the spectrophotometer.

The pH of all solutions was checked after the completion of each kinetic study and no appreciable changes were observed.

3. Analytical methods:

Since the hydrolysis of aspirin yields salicylic acid in a mole to mole ratio, the rate of degradation could be followed accurately by measuring the amount of salicylic acid formed. The optimum wavelength of absorption for salicylic acid was determined on the Beckman spectrophotometer by varying the wavelength and observing

the absorption values. The peak absorption value for salicylic acid was found to be 302 mμ, which agreed with previously reported observations (9). At this wavelength the absorption of aspirin was found to be essentially zero. A Beer's Law relationship (12) at this wavelength was determined for salicylic acid at all pH values involved. The optical blanks for the spectrophotometric determinations consisted of the appropriate buffers. Similarly, the maximum wavelength of absorption for aspirin was found to be 275 mμ and Beer's Law relationships at this wavelength were determined for aspirin at pH 2.00 and at pH 3.50.

Salicylic acid and aspirin followed a Beer's Law relationship at all pH values involved. Figure 1 shows the Beer's Law plots for aspirin and Figure 2 shows a representative Beer's Law plot for salicylic acid. Molar absorptivity values, calculated from the plots, appear in Table I. The molar absorptivity values obtained compared favorably with the results reported by Edwards (9). All subsequent calculations to determine aspirin and salicylic acid concentrations were made using the calculated molar absorptivity values.

All experiments were performed at least twice. The difference between values obtained in duplicate experiments did not exceed five per cent indicating that the experimental results were reproducible.

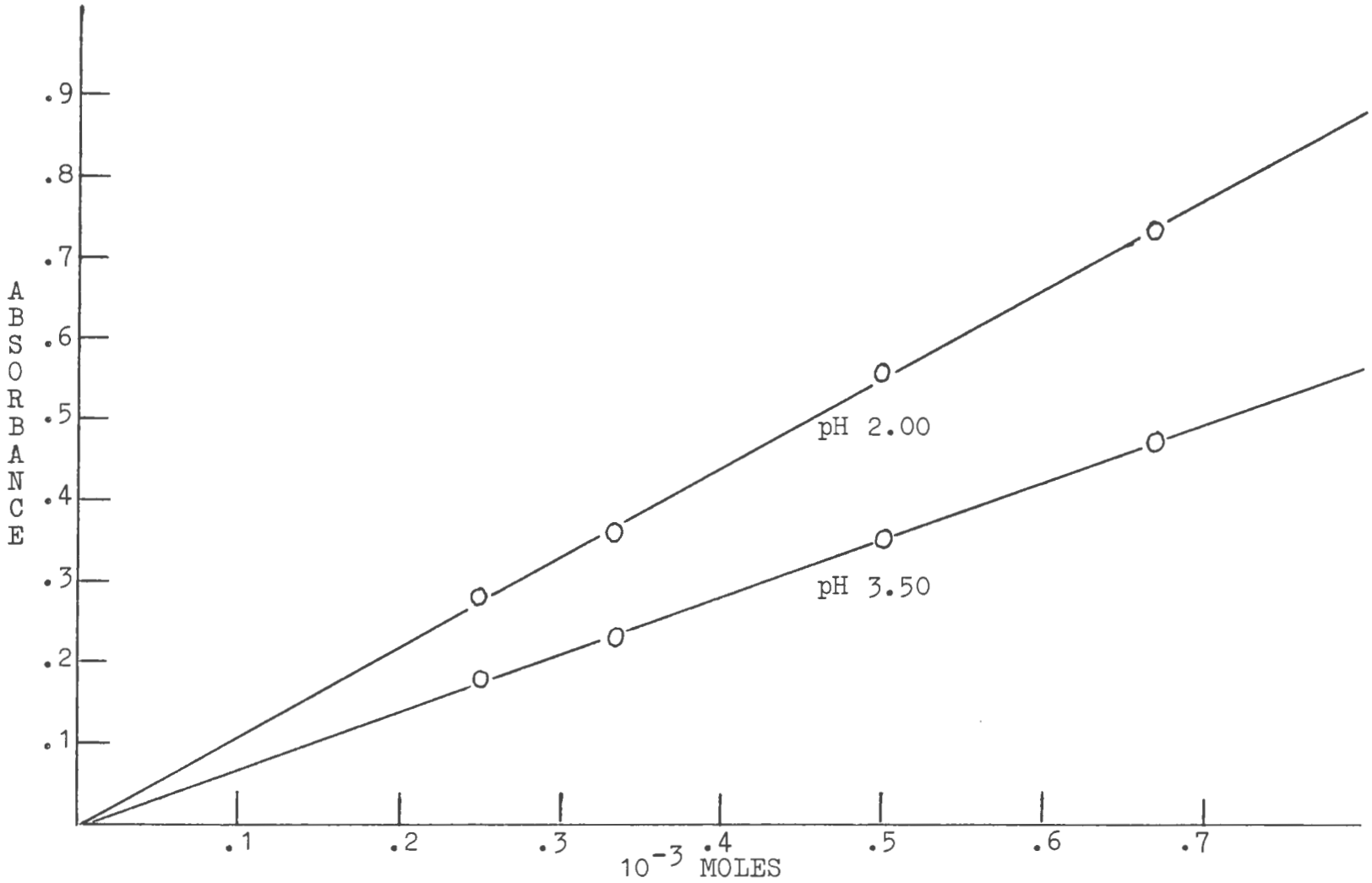

Figure 1. Beer's Law Plots for Aspirin at 275 mμ at pH 2.00 and at pH 3.50.

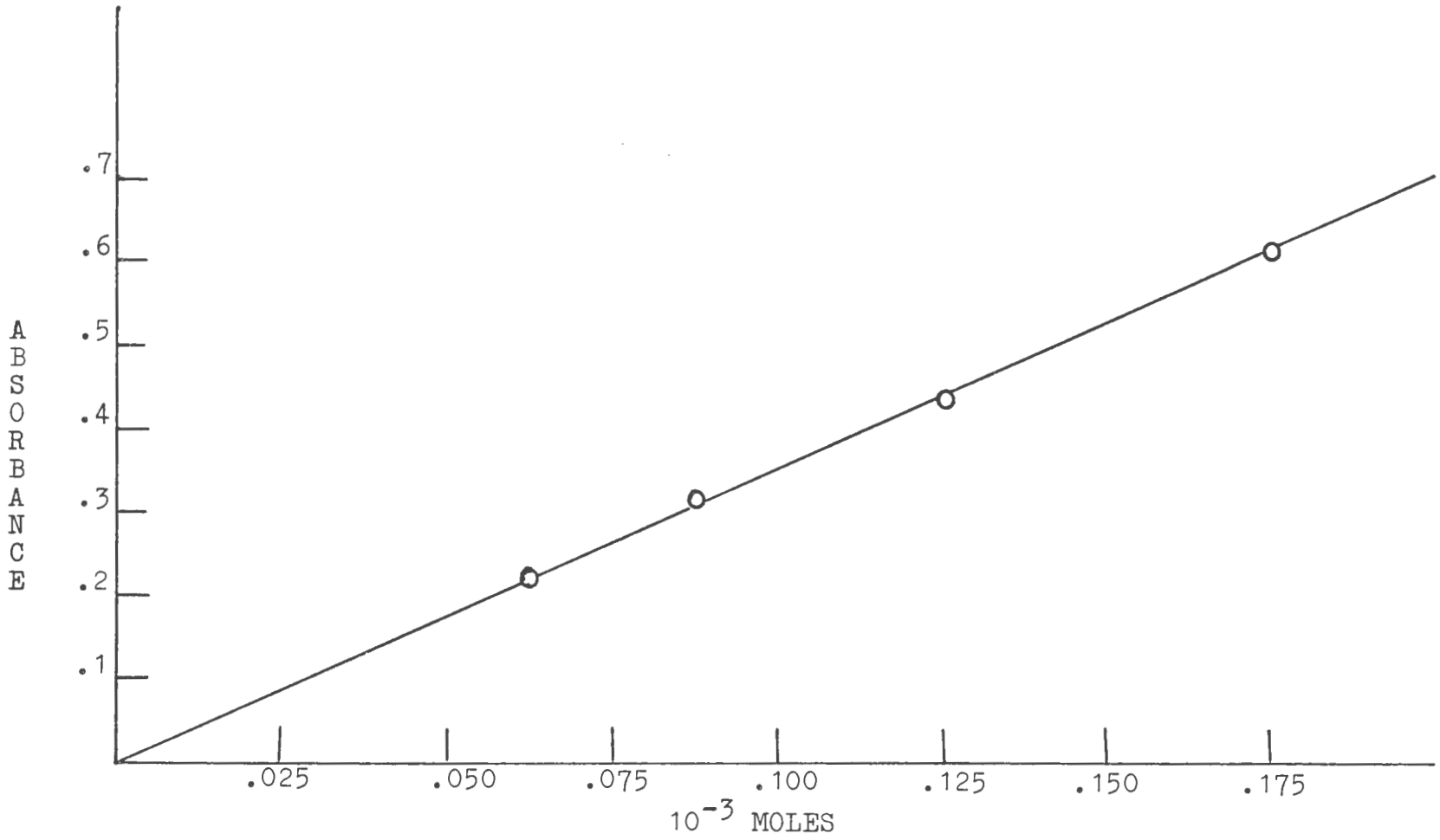

Figure 2. Beer's Law Plot for Salicylic Acid at 302 mμ and pH 2.00.

TABLE I

MOLAR ABSORBANCE OF SALICYLIC ACID AND ASPIRIN

	pH	Molar Absorptivity $x10^3$
Aspirin	2.00	1.095
Aspirin	3.50	0.705
Salicylic Acid	2.00	3.520
Salicylic Acid	2.50	3.500
Salicylic Acid	2.75	3.460
Salicylic Acid	3.00	3.380
Salicylic Acid	3.50	3.260

III. RESULTS

Complexing Curves

Figure 3 represents the interaction of aspirin with urea at pH 2.00 and at pH 3.50. The experimental technique was checked by calculating the concentration of un-ionized aspirin, S_o, at zero urea concentration at pH 2.00 and at pH 3.50, using the equation (13):

$$S=S_o\left[1+\frac{K_a}{(H_3O)^+}\right]$$

It can be seen from Figure 3 that urea decidedly affected the solubility of aspirin at both pH values.

Degradation Curves

Figures 4 through 8 represent the degradation of aspirin in the various buffers alone and in the presence of urea at pH values of 2.00, 2.50, 2.75, 3.00 and 3.50. The concentrations of undegraded aspirin at the various times were determined by subtracting the calculated concentrations of salicylic acid formed from the original concentration of aspirin. In all cases the observed rate of disappearance of aspirin followed a first order rate over at least the first half of the reaction. Rate constants for the observed reactions were obtained from the solpes of the straight lines observed according to

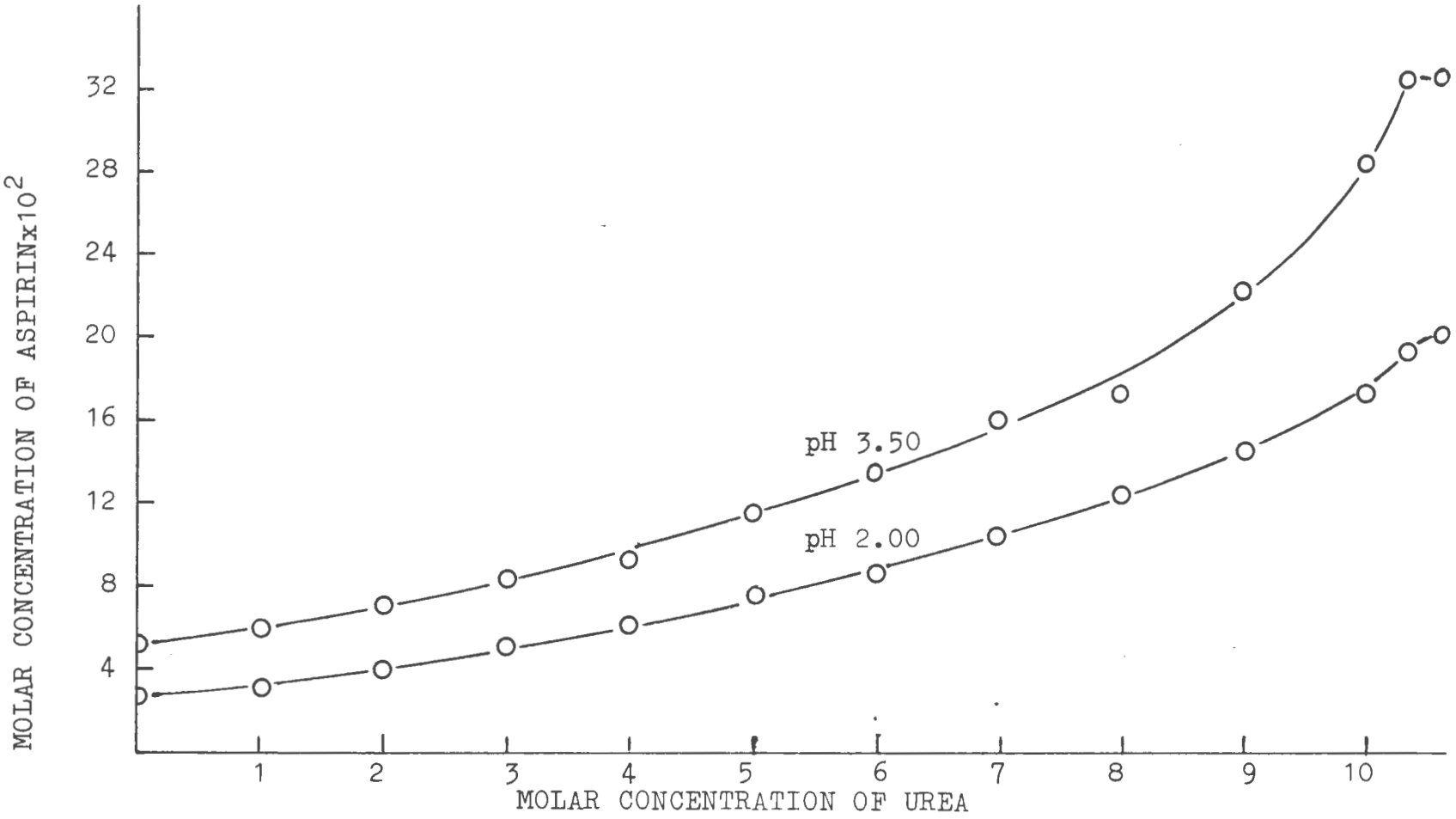

Figure 3. Phase Diagrams Showing the Solubility Behavior of Aspirin in the Presence of Urea in Water at 30.0°C at pH 2.00 and at pH 3.50.

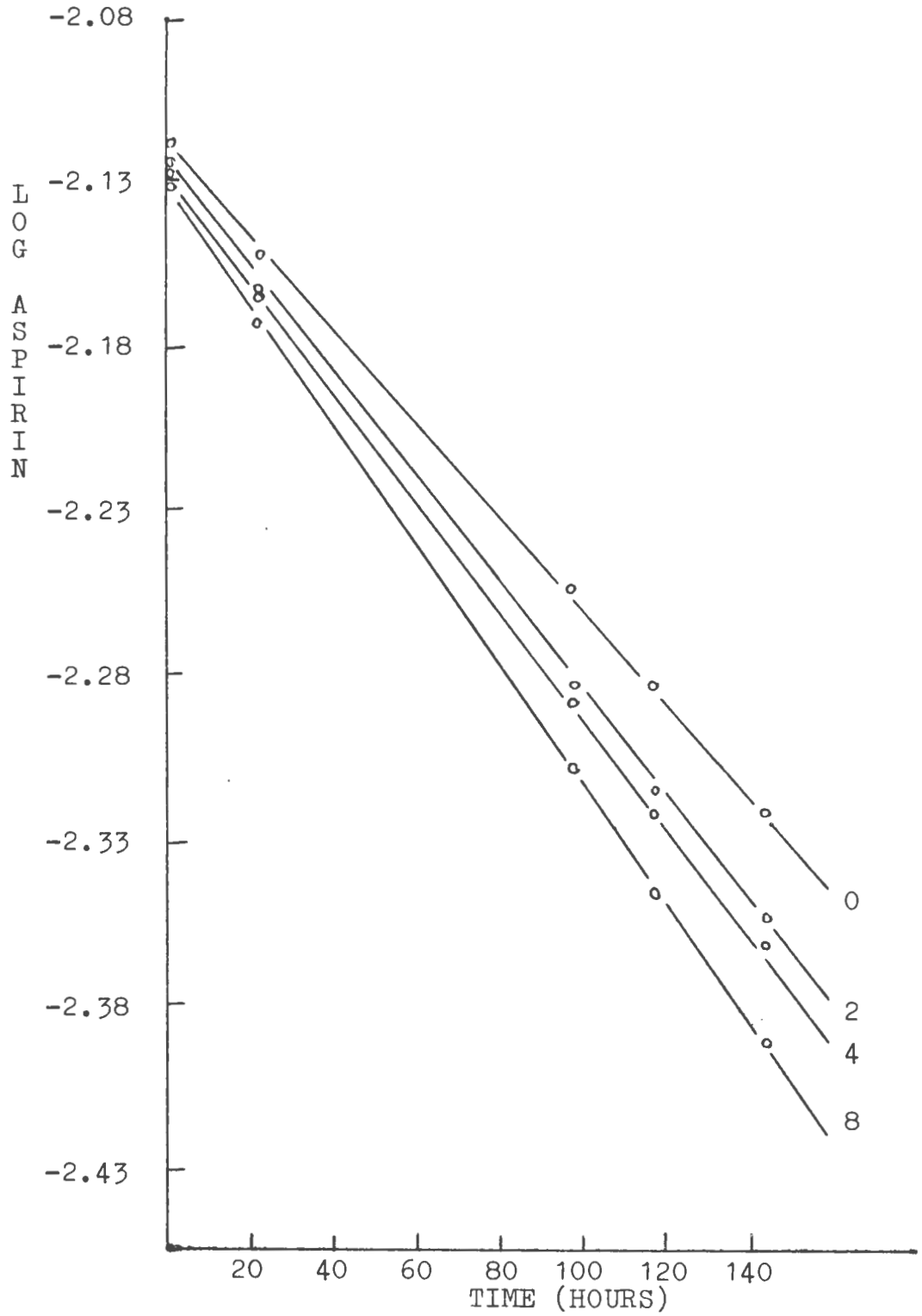

Figure 4. Degradation of Aspirin, Alone and in the Presence of Urea at pH 2.00 and 30°C. O, Aspirin Alone; 2, in 2 Molar Urea; 4, in 4 Molar Urea; 8, in 8 Molar Urea.

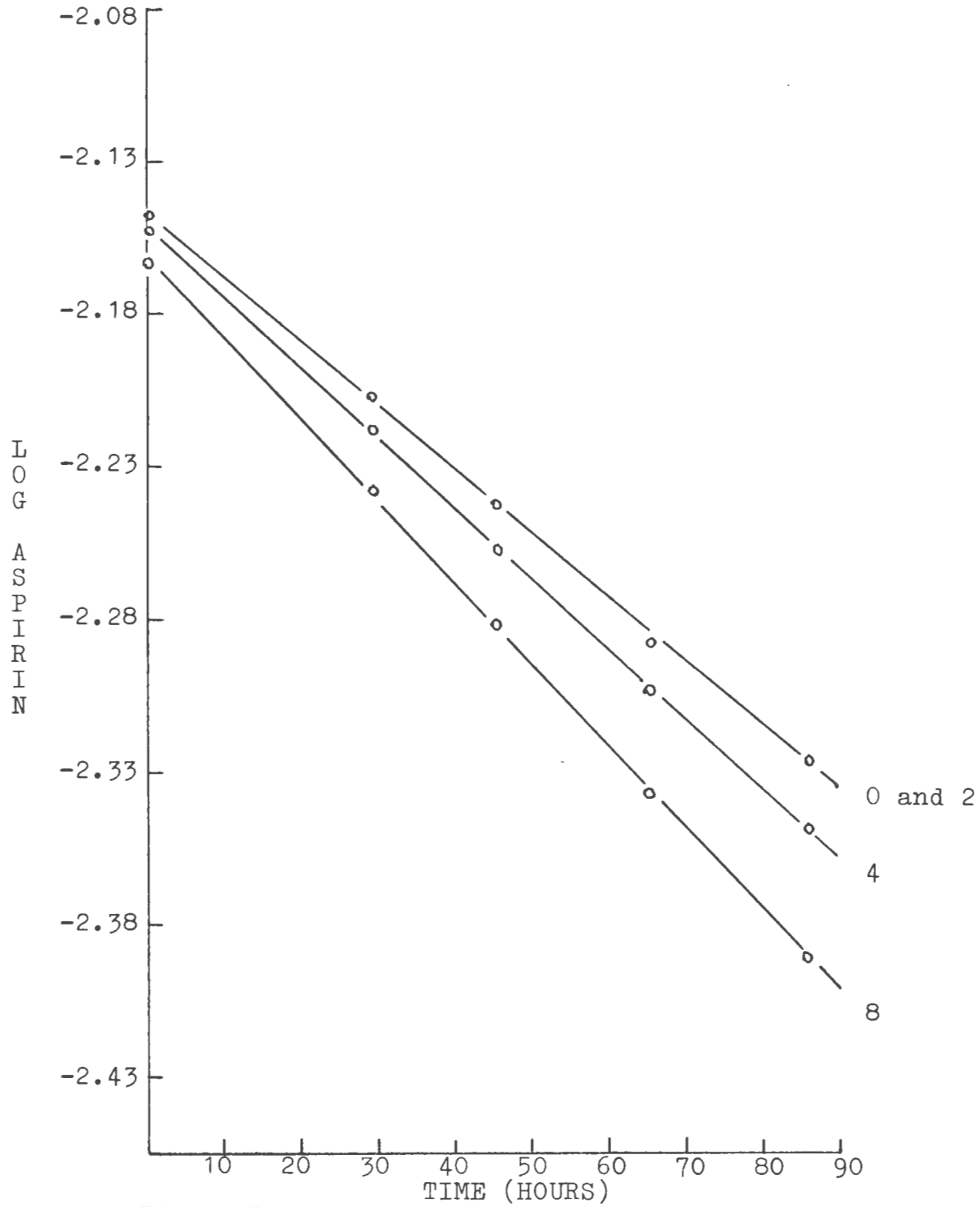

Figure 5. Degradation of Aspirin, Alone and in the Presence of Urea at pH 2.50 and 30.0°C. 0, Aspirin Alone; 2, in 2 Molar Urea; 4, in 4 Molar Urea; 8, in 8 Molar Urea.

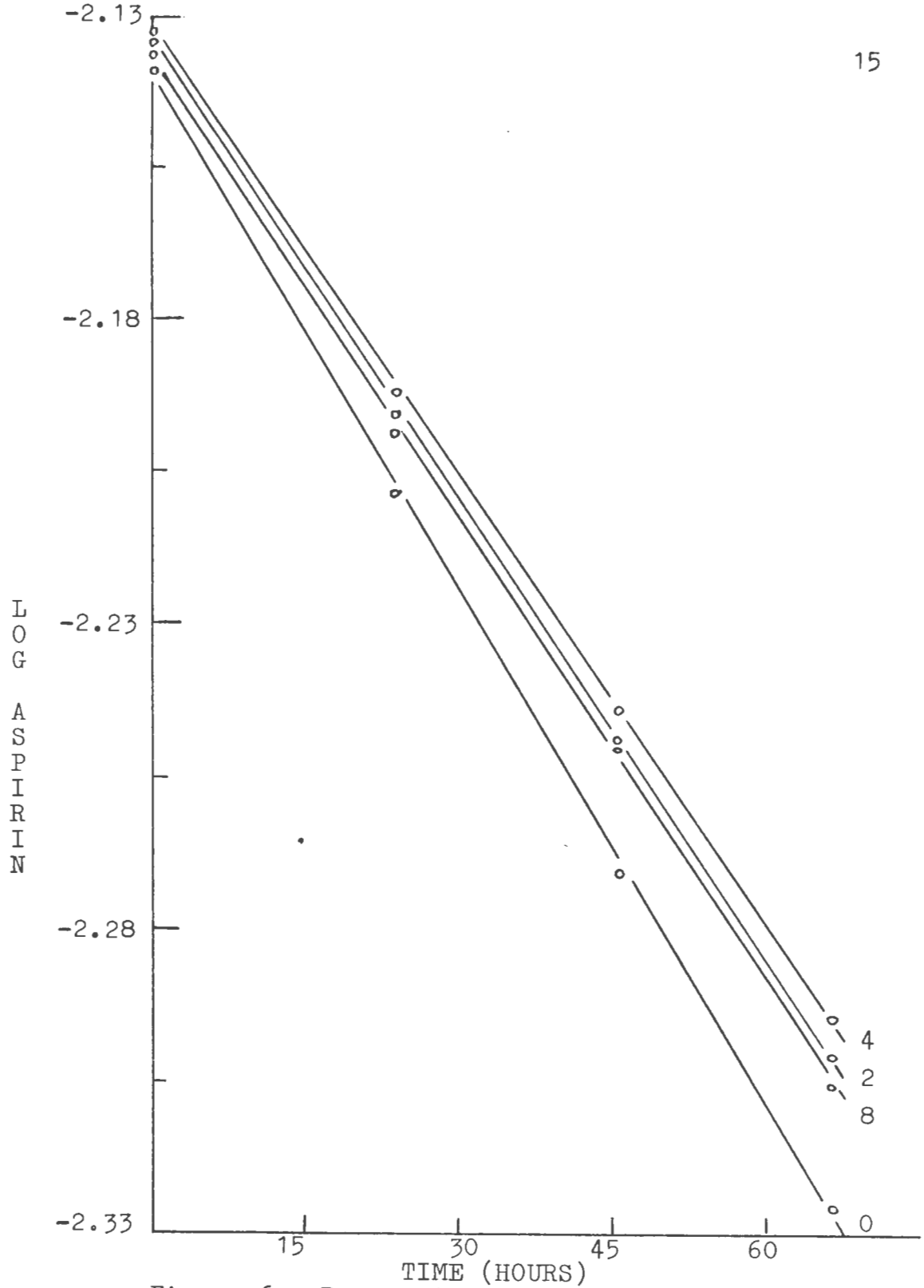

Figure 6. Degradation of Aspirin, Alone and in the Presence of Urea at pH 2.75 and 30.0°C. 0, Aspirin Alone; 2, in 2 Molar Urea; 4, in 4 Molar Urea; 8, in 8 Molar Urea.

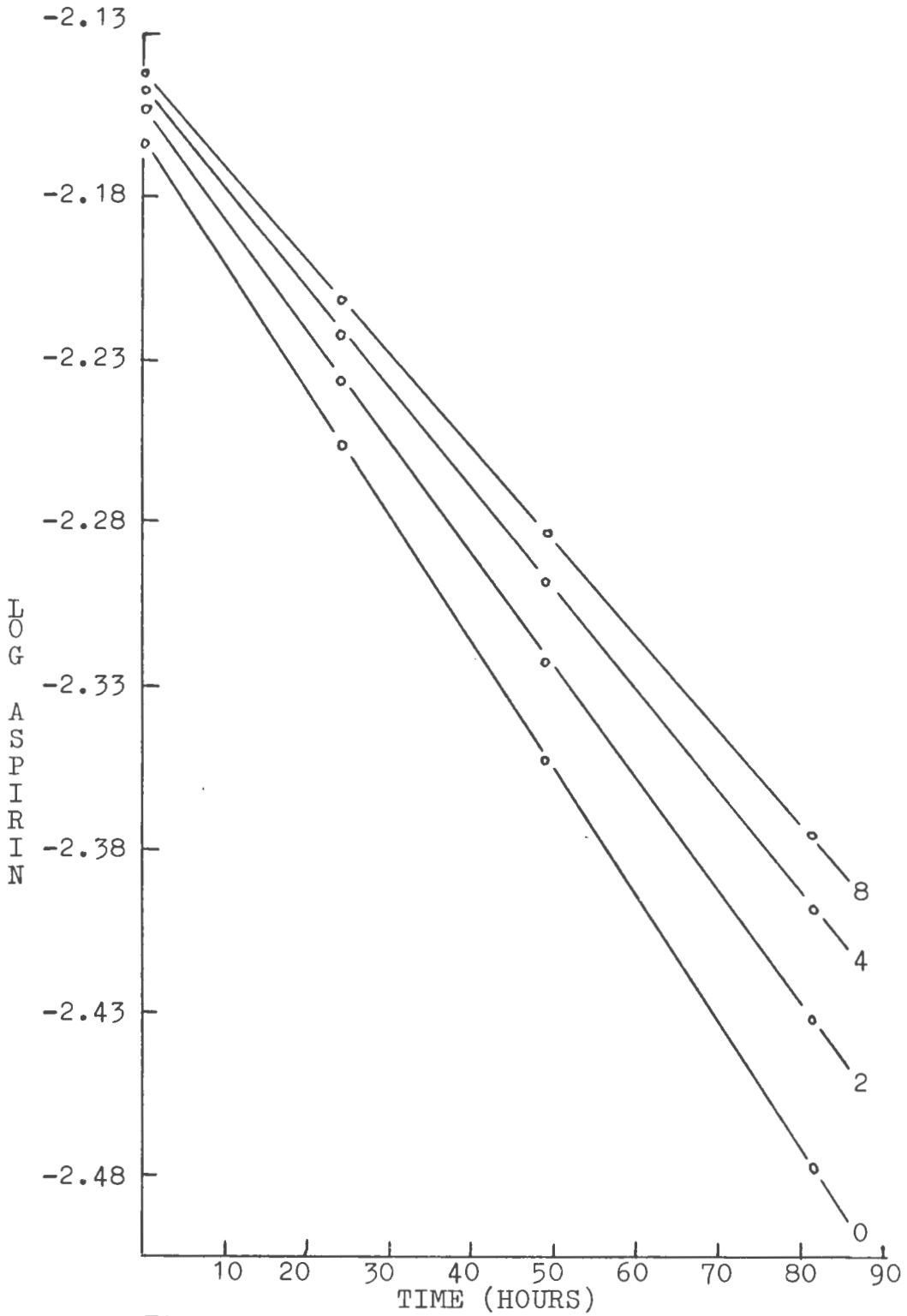

Figure 7. Degradation of Aspirin, Alone and in the Presence of Urea at pH 3.00 and 30.0°C. 0, Aspirin Alone; 2, in 2 Molar Urea; 4, in 4 Molar Urea; 8, in 8 Molar Urea.

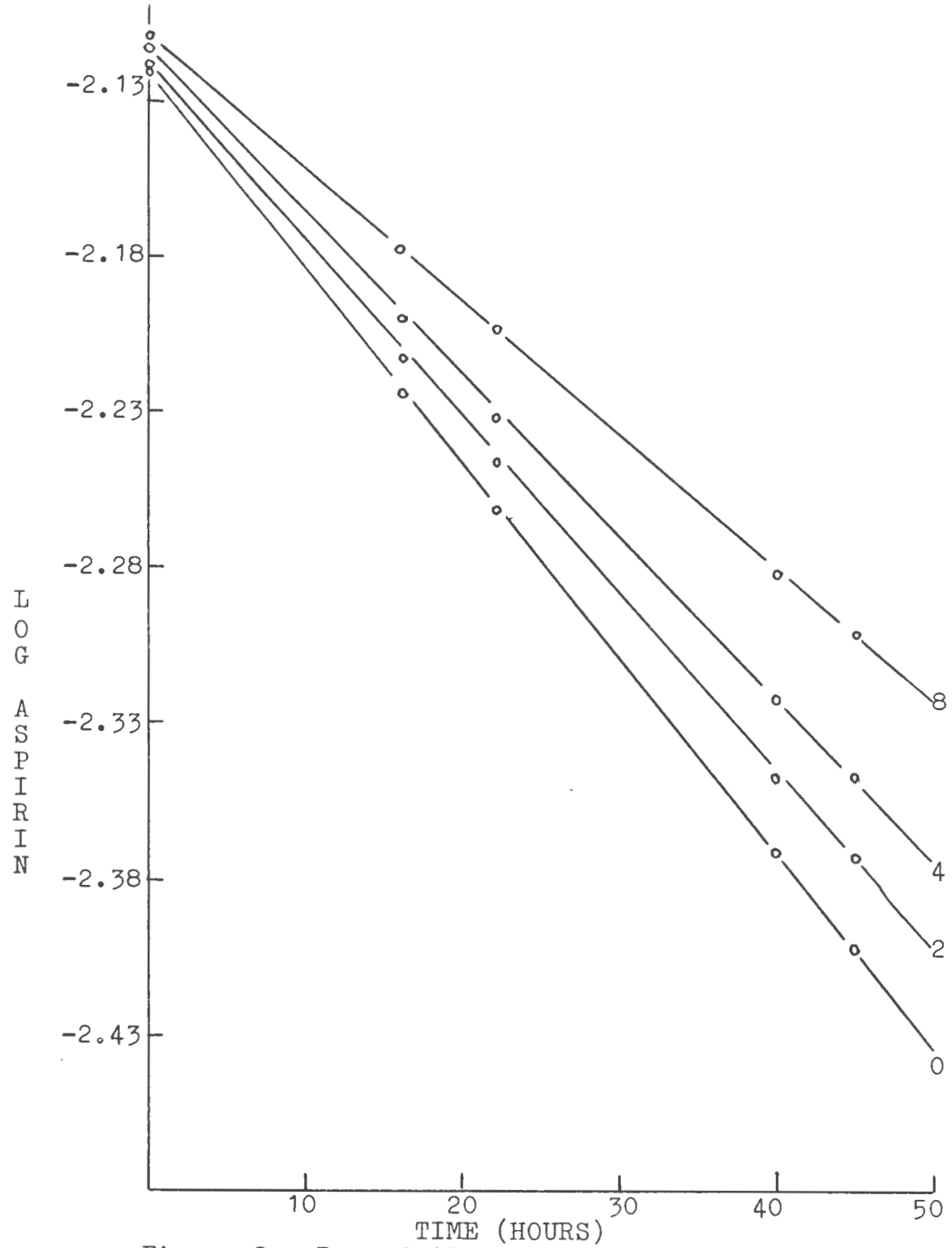

Figure 8. Degradation of Aspirin, Alone and in the Presence of Urea at pH 3.50 and 30.0°C. 0, Aspirin Alone; 2, in 2 Molar Urea; 4, in 4 Molar Urea; 8, in 8 Molar Urea.

the relationship:

$$k=-2.303 \times slope$$

The calculated rate constants, reported in $hours^{-1}$, are shown in Table II.

Analysis of Complexing Curves

If the solubilization of aspirin is attributed to complex formation, the shape of the curves represented in Figure 3 should give an indication of the order of reaction between aspirin and urea. The assumption of the formation of a one to one complex, or a complex containing one mole of urea, would lead to a straight line, but it is obvious in the case represented by Figure 3, that the interaction between the species is more complicated. It was possible, however, to calculate equilibrium constants for the complex formations at the different pH values according to a method previously outlined by Bolton (14):

Considering K_1 and K_2 as constants describing the curves where,

$$(\text{aspirin})+(\text{urea}) \rightleftharpoons (\text{aspirin-urea})$$

$$(\text{aspirin-urea})+(\text{urea}) \rightleftharpoons (\text{aspirin-urea}_2)$$

then,

$$K_1=\frac{(\text{aspirin-urea})}{(\text{aspirin})(\text{urea})} \quad \text{and} \quad K_2=\frac{(\text{aspirin-urea}_2)}{(\text{aspirin-urea})(\text{urea})}$$

TABLE II

RATE CONSTANTS FOR DEGRADATION REACTIONS AT VARYING UREA CONCENTRATIONS AND pH

	$k=hours^{-1} \times 10^{-3}$				
Urea Concentration	pH 2.00	pH 2.50	pH 2.75	pH 3.00	pH 3.50
0 M	3.2	4.8	6.4	8.8	14.5
2 M	3.6	4.8	5.6	7.9	13.3
4 M	3.7	5.2	5.6	7.0	11.9
8 M	4.1	6.0	5.8	6.6	10.0

and, an overall equilibrium constant can be expressed as:

$$K_o = \frac{(\text{total complexed aspirin})}{(\text{aspirin})(\text{urea})} \qquad (1)$$

$$= \frac{(\text{aspirin-urea}) + (\text{aspirin-urea}_2)}{(\text{aspirin})(\text{urea})}$$

$$= \frac{(\text{aspirin-urea})}{(\text{aspirin})(\text{urea})} + \frac{(\text{aspirin-urea}_2)}{(\text{aspirin})(\text{urea})}$$

$$= K_1 + K_2\left(\frac{\text{aspirin-urea}}{\text{aspirin}}\right)$$

and since

$$\left(\frac{\text{aspirin-urea}}{\text{aspirin}}\right) = K_1(\text{urea})$$

then,

$$K_o = K_1 + K_1K_2(\text{urea}) \qquad (2)$$

Since the urea added is in great excess, compared to the aspirin concentration, total urea is essentially equal to the concentration of urea. By substituting the appropriate data from experimental results in equation (1), K_o values can be readily calculated and a plot of K_o versus the concentration of urea should, according to equation (2), result in a straight line with the intercept on the ordinate equal to the K_1 value. The K_2

value can in turn be calculated from the slope of the line. The resulting plots, at pH 2.0 and at pH 3.5, are shown in Figures 9 and 10. These plots were constructed according to the method of least squares (15). The K_1 and K_2 values obtained from the plots in Figures 9 and 10, for the complex formations, are listed in Table III.

Further considerations allowed calculation of K_1 and K_2 values for the interaction of ionized aspirin and urea at pH 3.5. Since, at pH 2.0, aspirin is essentially un-ionized, the equilibrium constants for complex formation calculated at that pH represent the interaction of urea and un-ionized aspirin. On the other hand, at pH 3.5, the equilibrium constants for the reaction probably represent a combination of the interactions of un-ionized aspirin with urea and ionized aspirin with urea. It should be possible, therefore, to calculate equilibrium constants for the interaction of ionized aspirin with urea at pH 3.5 if it is assumed that the same equilibrium constants for un-ionized aspirin operated at pH 2.0 and at pH 3.5. Thus at pH 3.5,

$$\frac{(\underline{\text{total complex}})}{(\text{aspirin})(\text{urea})} = K_o = K_1 + K_1K_2(\text{urea})$$

(total complex) is equal to (complex due to ionized and un-ionized species)

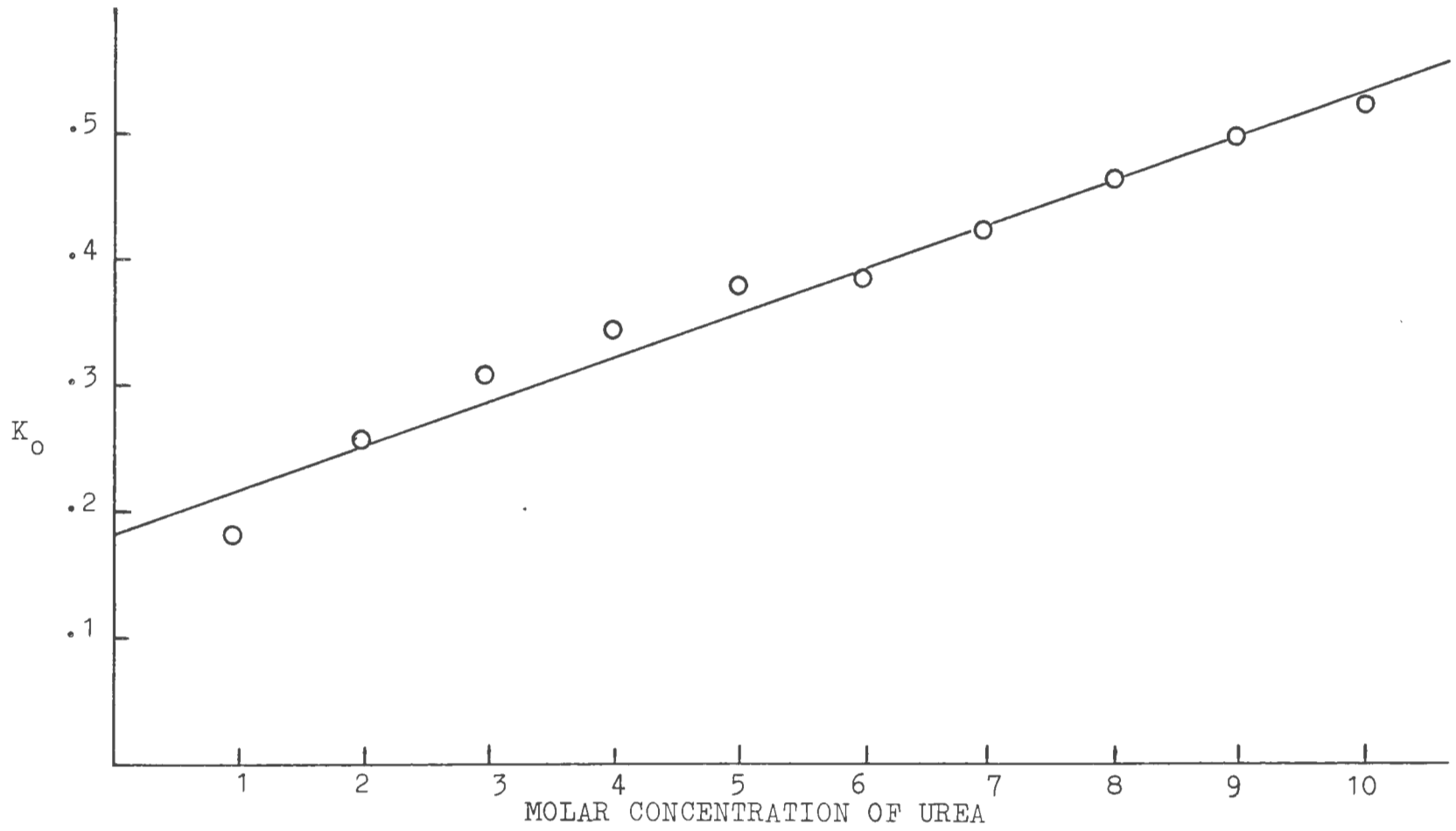

Figure 9. Plot Illustrating the Calculation of Equilibrium Constants, K_1 and K_2, at pH 2.00.

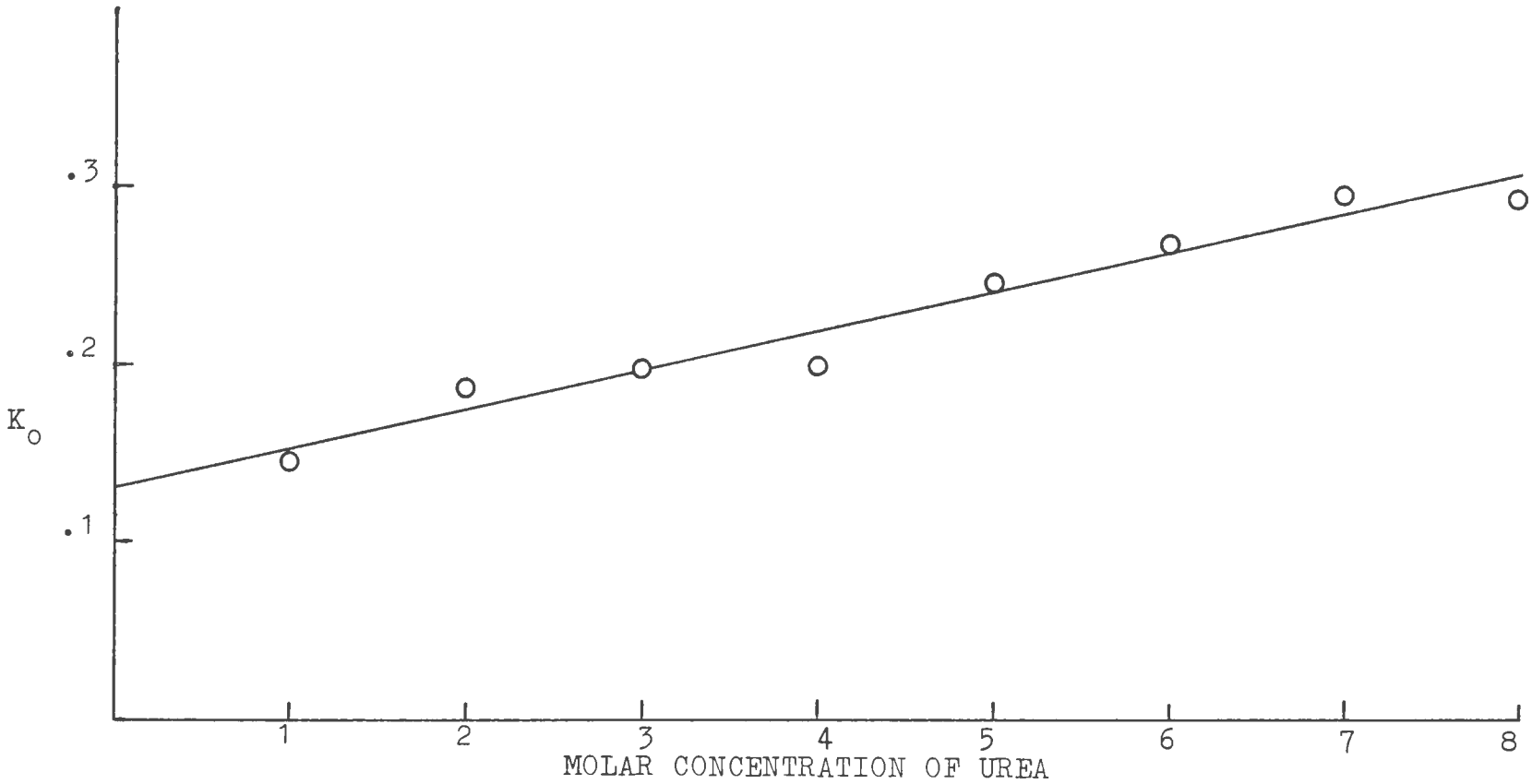

Figure 10. Plot Illustrating the Calculation of Equilibrium Constants, K_1 and K_2, at pH 3.50.

TABLE III

CALCULATED APPARENT EQUILIBRIUM CONSTANTS FOR THE INTERACTION OF ASPIRIN AND UREA

pH	Interaction	Equilibrium Constant	Value
2.00	$A^{\circ}+U \rightleftharpoons A^{\circ}U$	K_1	.186
2.00	$A^{\circ}U+U \rightleftharpoons A^{\circ}U_2$	K_2	.188
3.50	$A_t+U \rightleftharpoons A_tU$	K_1	.130
3.50	$A_tU+U \rightleftharpoons A_tU_2$	K_2	.168
3.50	$A+U \rightleftharpoons AU$	K_1	.072
3.50	$AU+U \rightleftharpoons AU_2$	K_2	.114

Key: A°=un-ionized aspirin
U =urea
A =ionized aspirin
A_t=total aspirin, un-ionized and ionized

$$=K_1(\text{urea})(\text{aspirin})+K_1K_2(\text{urea})(\text{aspirin})(\text{urea})$$

$$=K_1(\text{aspirin})(\text{urea})\left[1+K_2(\text{urea})\right] \qquad (3)$$

Similarly, at pH 2.0,

$$(\text{total complex})=(\text{complex due to un-ionized species})$$

$$=K_1(\text{aspirin})(\text{urea})\left[1+K_2(\text{urea})\right] \qquad (4)$$

Assuming that the un-ionized species interaction operates independently of the ionized species interaction, the total complex due to ionized aspirin can be obtained by subtracting the value obtained in equation (4) from the value obtained in equation (3).

$$\text{Since } K_o=\frac{(\text{complex due to ionized aspirin})}{(\text{aspirin})(\text{urea})},$$

a new set of K_o's representing the interaction of ionized aspirin with urea can be calculated. The values obtained can be plotted versus the urea added, as previously outlined, to arrive at the two equilibrium constants for the reaction.

Table IV shows the pertinent values thus obtained for ionized aspirin at pH 3.50 at the various urea concentrations. Figure 11 shows the plot of K_o versus the concentration of urea. The values of the equilibrium constants, K_1 and K_2, derived from this plot, are listed in Table III.

From inspection of Figure 3, it is evident that, although consistent, the data obtained at pH 3.50 showed

TABLE IV

CALCULATED COMPLEX CONCENTRATIONS DUE TO IONIZED ASPIRIN AT pH 3.50 AT VARIOUS UREA CONCENTRATIONS AND CORRESPONDING VALUES OF OVERALL CONSTANTS

Urea Concentration (molar)	Total Complex ($Mx10^{-2}$)	K_o
1	.207	.081
2	.455	.088
3	.739	.096
4	1.074	.104
5	1.457	.113
6	1.843	.120
7	2.340	.130
8	2.850	.139
9	3.380	.146
10	3.910	.152

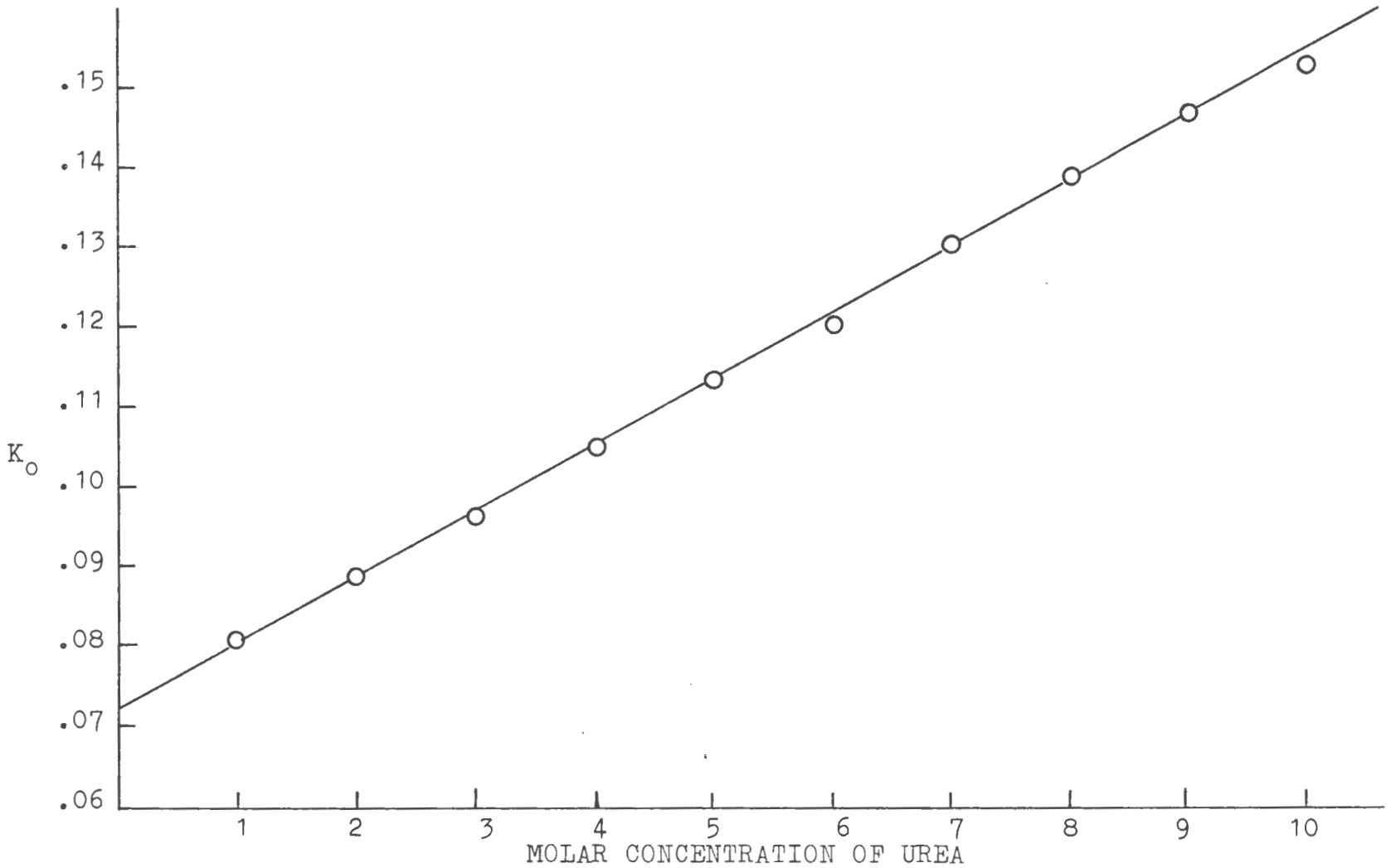

Figure 11. Plot Illustrating the Calculation of Equilibrium Constants, K_1 and K_2, for the Interaction of Ionized Aspirin-Urea.

greater deviations than the data obtained at pH 2.00. This was attributed to difficulties in determining the aspirin concentration in the complexing solutions after the equilibrium period of five hours had elapsed. At the higher pH a small amount of aspirin hydrolysed resulting in a more complicated system due to the formation of a substantial amount of salicylic acid. Because of this consideration, complexing studies at pH values higher than 3.50, where aspirin is highly ionized, were not pursued.

Analysis of Degradation Curves

At pH 2.00, where aspirin is essentially un-ionized, the overall rate constant for the degradation of aspirin can be described as follows (3):

$$k=k_f F_f+k_c F_c \qquad (5)$$

where, k =the apparent rate constant for the degradation of aspirin in presence of urea

k_f=the rate constant for the degradation of aspirin alone

k_c=the rate constant for the degradation of aspirin complexes

F_f=the fraction of aspirin remaining free in solution

F_c=the fraction of aspirin complexed in solution.

F_f and F_c can be determined as follows:

Representing aspirin as A and urea as U,

$$K_1 = \frac{(AU)}{(A)(U)} , \quad K_2 = \frac{(AU_2)}{(AU)(U)}$$

$$(AU) = K_1(A)(U) , \quad (AU_2) = K_2(AU)(U) = K_1K_2(A)(U)^2$$

$$(\text{total complex}) = (AU) + (AU_2) = K_1(A)(U) + K_1K_2(A)(U)^2$$

$$K_f = \frac{A}{A + \text{total complex}} = \frac{1}{1 + K_1(U)\left[1 + K_2(U)\right]} \qquad (6)$$

$$K_c = \frac{\text{total complex}}{A + \text{total complex}} = \frac{K_1K_2(A)(U)^2 + K_1(A)(U)}{A + K_1K_2(A)(U)^2 + K_1(A)(U)} \qquad (7)$$

$$= \frac{K_1(U)\left[1 + K_2(U)\right]}{1 + K_1(U)\left[1 + K_2(U)\right]}$$

substituting equations (6) and (7) into (5) gives:

$$k = k_f \frac{1}{1 + K_1(U)\left[1 + K_2(U)\right]} + k_c \frac{K_1(U)\left[1 + K_2(U)\right]}{1 + K_1(U)\left[1 + K_2(U)\right]} \qquad (8)$$

With a knowledge of the values of k and k_f obtained from the experimental data (see Table III), it is possible to determine a rate constant for the degradation of complexed aspirin. The value of k was calculated from equation (8) for aspirin-urea solutions at pH 2.00 and the results at several urea concentrations are shown in Table V. It can be seen from the Table that the values of k remained constant over the range of urea concentrations used.

TABLE V

CALCULATED RATE CONSTANT FOR THE DEGRADATION OF ASPIRIN COMPLEX AT pH 2.0 AT SEVERAL UREA CONCENTRATIONS

Urea Concentration (Molar)	k_c ($hours^{-1} \times 10^{-3}$)
2	4.5
4	4.2
8	4.4

At pH values higher than 2.00 the use of an equation similar to (5) to analyze the stability of the aspirin-urea complexes would be extremely difficult. In effect, since aspirin ionizes, the interactions should be represented as follows:

$$A^{\circ}+U \rightleftarrows A^{\circ}U$$

$$A^{\circ}U+U \rightleftarrows A^{\circ}U_2$$

$$A+U \rightleftarrows AU$$

$$AU+U \rightleftarrows AU_2$$

where A°=un-ionized aspirin

A^{-} =ionized aspirin

and equation (5) can be rewritten as:

$$k=k_f F_f+k_c^{\circ} F_c^{\circ}+k_c F_c \qquad (9)$$

where k=apparent rate constant for the degradation of aspirin in the presence of urea at the pH involved

k_f=rate constant for the degradation of aspirin alone

k_c°=rate constant for the degradation of un-ionized aspirin-urea complexes

k_c=rate constant for the degradation of ionized aspirin-urea complexes

F_f=fraction of aspirin remaining free in solution

F_c°=fraction of aspirin in unionized aspirin-urea complexes

F_C=fraction of aspirin in ionized aspirin-urea complexes.

Considering the nature of the assumptions and approximations which would be used in order to obtain values to fit equation (9) it was felt such calculations would not be meaningful.

IV. DISCUSSION

The solubilization of aspirin in the presence of urea can be described by two constants, K_1 and K_2, which correspond to the formation of one to one and two to one complexes respectively. It cannot be said with assurance that these constants have the previously designated physical interpretation since unknown solvent effects probably are important in highly concentrated urea solutions. These constants, therefore, can be considered a description of the curves shown in Figure 3 and, for this reason, are referred to, in Table III, as "apparent equilibrium constants". It should be noted that the apparent equilibrium constants calculated at pH 3.50 from equation (2) represent a combination of the reactions of urea with ionized aspirin and un-ionized aspirin. These K values were used to determine the apparent equilibrium constants for the interactions of ionized aspirin and urea at pH 3.50.

The small magnitude of the K values listed in Table III indicates that the interaction between aspirin and urea, at the pH values involved, is relatively weak. The constants obtained for the interaction of un-ionized aspirin and urea are slightly higher than the ones obtained for the ionized aspirin interaction, indicating that the interaction is more favorable at pH values where aspirin

is principally un-ionized. This might be expected because of the weak basic properties of urea. Generally, the values listed in Table III are approximately of the same order of magnitude as those obtained by Bolton for benzoic acid-urea and salicylic acid-urea systems (2), suggesting the possibility of similar solubilization mechanisms in these systems. Because of the limited amount of data in this study, speculation on the nature of the mechanisms involved does not seem to be feasible.

The data of Table II, showing the rates for the degradation of aspirin alone and aspirin in the presence of urea at various pH values, indicates that the aspirin-urea interaction product at pH 2.00 results in decreased aspirin stability. However, as the pH increases, the situation reverses itself and, at pH 3.50, the aspirin complex is more stable than free aspirin at that pH. The pH where the observed rate of degradation of aspirin alone becomes higher than that of aspirin in the presence of urea seems to occur in the vicinity of 2.60. It is most interesting to note that this "crossover" occurs at a pH corresponding to the pH of maximum stability reported by Edwards when rate constants are plotted against pH (9). Thus, this pH may represent a point where a different mechanism is responsible for aspirin decomposition and this could provide an explanation for the contrasting

effects of urea below and above this pH.

Since ester hydrolysis involves steric effects as well as polar effects, some explanation for the behavior described above might be possible from either or both of these two standpoints. Due to the fact that the mechanism of complexation is rather ill defined, the possible explanations of the stability behavior presented below, are, of necessity, only of a very general nature.

Firstly, it is not unreasonable that, at pH 2.00, aspirin in solution may dimerize through hydrogen bonding of the unionized carboxyl hydrogen, thereby offering some steric hindrance to the access of the ester group. Urea, at pH 2.00, in the process of complexation might break the hydrogen bonding and yield an aspirin-urea product where the ester group of the aspirin would be more accessible than in dimerized aspirin. This effect could also be observed if changes in the properties of the solvent caused by the solvation of urea affected intermolecular aspirin bonding. As the pH begins to assume higher values, hydrogen bonding of aspirin may become less prevalent because of ionization and the ester groups might then be more accessible. The corresponding complexed forms of aspirin might then become relatively more stable.

Secondly, it is also possible that urea might attach to un-ionized and ionized aspirin at different sites.

At pH 2.00, for example, urea might attach at the carboxyl group of un-ionized aspirin, resulting in breaking of intermolecular hydrogen bonds as described above. On the other hand, at the higher pH values, urea attachment might affect the ester group, resulting in more stable species.

Lastly, it has been suggested that base catalyzed hydrolysis is more affected by polar effects and less by steric effects than acid catalyzed hydrolysis (16, 17). Since the hydrolysis of aspirin becomes more dependent on $(OH)^-$ species than $(H_3O)^+$ species as the pH increases from 2.00 to 3.50, the polar effects should be more important at pH 3.50. It is possible that urea might attach to aspirin in such a way that a species is formed whose electronic properties are such that it is more susceptible to $(H_3O)^+$ and less susceptible to $(OH)^-$. This could account for the strange stability properties of the complex. For example, if urea attaches to the carboxylate ion at pH 3.50, the effect could be to diminish the electronegative effect of the carboxylate group resulting in an increased stability of aspirin.

The consistant nature of the data in Table V, showing the values obtained for the rate constant of the degradation of the aspirin-urea complex at pH 2.00 at the several

urea concentrations used is evidence that complex formation rather than solvent effects is responsible for the increased solubilization of aspirin. A further study to obtain degradation rate values for the complexed species at pH values where aspirin is mostly ionized would have been most interesting in an attempt to further elucidate the nature of the interaction. This was not feasible however, since, as previously mentioned, it was not possible to obtain apparent equilibrium constants for the aspirin-urea interaction at pH values higher than 3.50.

V. CONCLUSIONS

Aspirin weakly interacts with urea in aqueous solution to yield a soluble complex. The complex formation can be described by two constants corresponding to the formation of one to one and two to one complex species. The interaction is more favorable at low pH values where aspirin is principally in the un-ionized form. At pH values lower than 2.60 complex formation appears to hasten the hydrolysis of aspirin. At pH values higher than 2.60, and at least up to 3.50, the complexed species appears to be more stable than the corresponding free aspirin species.

From a pharmaceutical point of view, although urea markedly enhances the solubility of aspirin in water within the pH range of 2.00 to 3.50, it does not inhibit its decomposition sufficiently to be of much practical use in this regard. A further study of aspirin-urea interaction at pH values higher than 3.50 would be valuable in furnishing additional information about the nature of the complex formation and properties of the complex. The present study, it is hoped, will serve as a starting point for further investigations along these lines that might lead to the development of a stable "liquid aspirin" preparation.

VI. BIBLIOGRAPHY

1. Lach, J.L., and Pauli, W.I., Drug Standards, 27, 104-07 (1959).

2. Bolton, S., J. Pharm. Sci., 52, 1071-4 (1963).

3. Higuchi, T., and Lachman, L., J. Am. Pharm. Assoc., Sci. Ed., 44, 521-6 (1955).

4. Higuchi, T., and Bolton, S., J. Pharm. Sci., 48, 557-64 (1959).

5. Lachman, L., Ravin, L.J., and Higuchi, T., J. Am. Pharm. Assoc., Sci. Ed., 45, 290-5 (1956).

6. Lachman, L., and Higuchi, T., J. Am. Pharm. Assoc., Sci. Ed., 46, 32-6 (1957).

7. Altwein, D.M., Delgado, J.N., and Cosgrove, F.P., J. Pharm. Sci., 54, 603-6 (1965).

8. James, K.C., J. Pharm. Pharmacol., 10, 363 (1958).

9. Edwards, L.J., Trans. Faraday Soc., 46, 723-35 (1950).

10. Garrett, E.R., J. Am. Pharm. Assoc., Sci. Ed., 46, 584-6 (1957).

11. Huguchi, T., and Zuck, D.A., J. Am. Pharm. Assoc., Sci. Ed., 42, 138-45 (1953).

12. Ayres, G.H., Quantitative Chemical Analysis, Harper & Brothers, New York, N.Y., 1958, p. 509.

13. Martin, A. Physical Pharmacy, Philadelphia, Penn., Lea & Febiger Inc., 1960, p. 373.

14. Bolton, S., unpublished Ph.D. Dissertation, University of Wisconsin, 1958.

15. Mode, E.B., Elements of Statistics, 2nd Ed., Prentice Hall, N.Y., 1951, p. 217.

16. Taft, R.W., Jr., J. Am. Chem. Soc., 74, 3120 (1952).

17. Ingold, C.K., Structure and Mechanisms in Organic Chemistry, Cornell University Press, Ithica, 1953, p. 752.