3. **g-Values.**[32]—The *g*-values of radical intermediates observed in the dimethylaniline–quinone reactions may be compared with the *g*-values of semiquinones prepared by other methods (Table II). The radicals which we observed in dimethylaniline have *g*-values which are identical to the independently determined *g*-values of the corresponding semiquinones. In fact, it is this comparison which shows that the radical intermediates are indeed semiquinones.

Summary.—Chloranil reacts with N,N-dimethylaniline in N,N-dimethylaniline as solvent to yield a crystal violet salt. Diamagnetic donor-acceptor complexes and paramagnetic semiquinones are two observed intermediates. Four other tetrahalogenated quinones react similarly with dimethylaniline.

Preliminary kinetic data have been obtained from measurements of optical absorption, conductivity and e.s.r. absorption and the mechanism of the oxidation has been discussed. If the transfer of an electron from dimethylaniline to chloranil occurs, the steady-state concentration of ions produced by the electron transfer is very small.

(32) In the late stages of reaction, after the semiquinones have disappeared, a second radical has been observed in the reactions of chloranil and *o*-chloranil with dimethylaniline. The radical in the latter case has $g = 2.0049 \pm 0.0002$. The *g*-value of the secondary radical derived from chloranil is also less than that of the primary radical. This *g*-value was not measured because absorption of the two radicals overlapped.

[CONTRIBUTION FROM THE DEPARTMENT OF BIOCHEMISTRY, COLLEGE OF DENTISTRY AND GRADUATE SCHOOL OF ARTS AND SCIENCES, NEW YORK UNIVERSITY, NEW YORK, N. Y.]

Effect of Urea on Hydrogen Bonding in Some Dicarboxylic Acids[1]

BY MILTON LEVY AND JOHN P. MAGOULAS

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The effect of urea on proteins has been attributed to hydrogen bond "breaking." In the course of testing this hypothesis we have developed a method of calculating the free energies of hydrogen bonding between carboxyl groups in a dicarboxylic acid as well as between carboxyl and carboxylate groups in its mono-anion. This involves comparison of titration constants between hydrogen bonded forms (*e.g.*, maleic acid) and comparable non-bonded compounds (*e.g.* fumaric acid). Titration constants of these acids and of malonic, dimethylmalonic, diethylmalonic and ethylisopropylmalonic aids are recorded in aqueous media containing 0-7 *M* urea and at 7, 25 and 37°. From these data it does not appear that urea has any great effect on the hydrogen bonds. Thus in maleic acid the free energy of the carboxyl–carboxylate bond is about −2300 calories in water and about −2200 calories in 7 *M* urea at 25°.

Proteins are "denatured" in aqueous solutions which contain large concentrations of urea. This result was ascribed by Mirsky and Pauling[2] to an ability of urea to "break" hydrogen bonds and this postulate has been widely accepted. No direct evidence of such an action of urea in aqueous solution has been adduced, unless its action on proteins and certain polymers of amino acid is considered evidence. Kauzman[3] has emphasized intramolecular hydrophobic bonds as stabilizing influences in the specific molecular conformations of proteins and assumes that urea affects these. Klotz and Stryker[4] suggest "solvent–(macromolecular) solute interaction" as a stabilizing influence and further suggest that a perturbation of this interaction by urea results in denaturation. In this hypothesis the action of urea could be more directly with the water than with the protein.[5] It is in connection with the problems of the effect of urea on proteins that one of us began a study of the influence of urea on acid–base behavior.[6] The present paper concerns dicarboxylic acids which contain intramolecular hydrogen bonds in aqueous solutions and the effect of urea upon these bonds.

The existence of hydrogen bonded structures in mono-anions of certain dicarboxylic acids was assumed by Jones and Soper[7] (more recently by Hunter[8] and by McDaniel and Brown[9]) to account for the large ratio K_1/K_2 in such acids. This qualitative idea was formulated in terms of equilibria by Westheimer and Benfey[10] and applied to maleic acid in water. The formulation leads to a hydrogen bonding constant which seemed reasonable. Some uncertainty (a factor of 2) in the theory results from the nature of the models used to estimate the dissociation constants unperturbed by hydrogen bonding. We will use the same formulation of equilibrium but different final equations and models. We believe the formulation below takes maximum advantage of the Westheimer and Benfey equilibrium scheme.

Formulation.—Let MH_2 represent a dicarboxylic acid. Reactions 1 and 2 show its successive ionizations. The lower case letters represent con-

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(2) A. E. Mirsky and L. Pauling, *Proc. Natl. Acad. Sci. U. S.*, **22**, 439 (1936).

(3) W. Kauzmann, *Advances in Protein Chemistry*, **14**, 1 (1959).

(4) I. M. Klotz and V. H. Stryker, *J. Am. Chem. Soc.*, **82**, 5169 (1960).

(5) I. M. Klotz and J. W. Russell, *J. Phys. Chem.*, **65**, 1274 (1961).

(6) M. Levy, *Compt. rend. Lab. Carlsberg Ser. Chim.*, **30**, 291 (1958).

(7) I. Jones and F. J. Soper, *J. Chem. Soc.*, 133 (1936).

(8) L. Hunter, *Chem. and Ind.* (*London*), 155 (1953).

(9) D. H. McDaniel and H. C. Brown, *Science*, **118**, 370 (1953).

(10) F. H. Westheimer and O. T. Benfey, *J. Am. Chem. Soc.*, **78**, 5309 (1956).

centrations (or activities, no distinction will be attempted) of the components symbolized above them in the reactions.

$$\underset{a}{\mathrm{MH_2}} \rightleftarrows \underset{b}{\mathrm{MH^-}} + \underset{h}{\mathrm{H^+}} \quad K_1 = bh/a \qquad (1)$$

$$\underset{b}{\mathrm{MH^-}} \rightleftarrows \underset{c}{\mathrm{M^=}} + \underset{h}{\mathrm{H^+}} \quad K_2 = ch/b \qquad (2)$$

Either or both MH_2 or MH^- may be internally hydrogen bonded and such bonded forms are indicated by a superior bar as in 3 and 4. The concentrations are symbolized by lower case letters with superior bars.

$$\underset{a}{\mathrm{MH_2}} \rightleftarrows \underset{\bar{a}}{\overline{\mathrm{MH_2}}} \quad L_1 = \bar{a}/a \qquad (3)$$

$$\underset{b}{\mathrm{MH^-}} \rightleftarrows \underset{\bar{b}}{\overline{\mathrm{MH}}^-} \quad L_2 = \bar{b}/b \qquad (4)$$

The two branches of the titration curves of dicarboxylic acids are described by the usual equations and the titration constants G_1 and G_2.

These are written in 5 and 6 following rules of conservation and electroneutrality.

$$G_1 = (b + \bar{b})h/(a + \bar{a}) \qquad (5)$$

$$G_2 = ch/(b + \bar{b}) \qquad (6)$$

Combining equation 2, 4 and 6 gives 7 and this with 1, 3 and 5 gives 8.

$$G_2 = K_2/(1 + L_2) \qquad (7)$$

$$G_1G_2 = K_1K_2/(1 + L_1) \qquad (8)$$

The two last equalities in logarithmic "p" form are restated in

$$\log (1 + L_1) = (pG_1 + pG_2) - (pK_1 + pK_2) \qquad (9)$$

$$\log (1 + L_2) = (pG_2 - pK_2) \qquad (10)$$

The application of these equations requires values for pK_1 and pK_2 to be obtained from models which do not have hydrogen bonds but in which other factors affecting dissociation are identical or appropriately estimated. Thus Westheimer and Benfey[10] estimated $K_1 = 2Ke$ where Ke is the titration constant of a half ester of the acid in question. It was also assumed that L_1 (if it is significant) is no greater than $2Le$ (the constant for hydrogen bonding of the carboxyl hydrogen in the half ester with the ester carbonyl group). This leaves a factor between 0.5 and 1 unevaluated and a corresponding uncertainty in the hydrogen bonding constant L_2 results.

We will estimate pK_1 and pK_2 from the behavior of analogous acids in which internal hydrogen bonding appears to be impossible or improbable. Thus in a pair of *cis–trans* isomers such as fumaric and maleic acids only the *cis* form can be internally hydrogen bonded. The titration constants of the *trans* form will be set equal to pK_1 and pK_2. The titration constants of maleic acid become pG_1 and pG_2.

In other cases sufficient freedom of rotation about unhindered single bonds exists to allow the required spacing of the two carboxyl groups, and it will be assumed that such groups will form hydrogen bonds with water rather than to each other. Thus malonic acid will be used as a model for substituted malonic acids with appropriate corrections for inductive effects suggested by the behavior of the half esters.

Experimental

The experiments consist of the potentiometric determination of the two pK of each acid in water and in urea solutions. The method of continuous titration was used. Titrations were carried out in a glass vessel carrying a "Radiometer" glass electrode, a salt bridge terminated by a U shaped tip, a N_2 inlet and the buret tip. The salt bridge led to a saturated calomel electrode immersed in the same constant temperature bath as the titration vessel. A PHM-4 "Radiometer" meter was used and the system was routinely calibrated with pH 6.5 buffer commercially supplied. Calibration with other standard buffers showed the scale of the meter to be linear within reliability. Appropriate blank titrations with standard acid and alkali were run on the solvents. Especially when pK_1 of the acid was small and in urea, which has a large acid blank,[6] it was often convenient to suppress the ionization of the first carboxyl by adding HCl and to calculate pK_1 from this data. The method of correcting for volume changes, blanks, etc., is essentially that outlined previously.[6]

Reagents.—Distilled water was prepared from deionized water by distillation in a two stage all quartz still.

Urea.—A 9 M solution of urea in conductivity water was prepared without raising the temperature above 25° and filtered through medium porosity sintered glass. An equal volume of absolute alcohol was added and the mixture placed at −27° for 2–3 days. The crystalline precipitate was filtered cold and washed with sufficient cold absolute alcohol to cover the crystals. After 3 washings the product was air dried in a porcelain dish in a dust free cabinet. A 7 M solution of this urea contains no more than $10^{-6}M$ titratable acid (NH_4^+) or base (CNO^-).

The compounds to be titrated were obtained commercially and recrystallized if necessary until the m.p. and titration equivalent agreed with the expected values. Half esters were prepared by partial hydrolysis of diesters and judged pure when their titration equivalent was normal and conformed to a mono acid.

Standard KOH was prepared to be carbon-dioxide free[11] and standardized against Bureau of Standards potassium acid phthalate. Standard HCl was prepared from constant boiling HCl and standardized against the potassium hydroxide. Potassium Chloride was A.C.S. grade.

Results

Maleic Acid.—The titration constants obtained for fumaric and maleic acids and their hydrogen ethyl esters are set forth in Table I for three temperatures and three urea concentrations. All solutions contained 0.1 M KCl. Acid concentrations were 0.02 M.

TABLE I

TITRATION CONSTANTS OF MALEIC AND FUMARIC ACIDS AND THEIR HALF ESTERS

Urea, M	Temp., °C.	Maleic acid			Fumaric acid		
		pK_1	pK_2	pK_e	pK_1	pK_2	pK_e
0	7	1.95	5.87	2.71	2.94	4.21	3.31
3.5	7	2.20	6.11	3.08	3.18	4.51	3.64
7.0	7	2.49	6.39	3.40	3.53	4.85	3.97
0	25	1.97	5.91	2.84	3.00	4.25	3.27
3.5	25	2.03	6.13	3.16	3.14	4.48	3.64
7	25	2.15	6.35	3.40	3.39	4.75	3.85
0	37	1.97	5.95	3.01	2.99	4.24	3.31
3.5	37	2.12	6.16	3.16	3.06	4.51	3.50
7.0	37	2.15	6.41	3.36	3.40	4.79	3.75

The constants L_1 and L_2 are to be calculated from the titration constants in Table I by the use of equations 7 and 8. Table II shows the values of L_1 and L_2 so calculated as well as the molar free energies of hydrogen bonding ($-\Delta F_1 = RT \ln L_1$, etc.). L_1 is smaller and subject to considerable greater uncertainty than L_2 because it involves four titration constants. We think the

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[11] J. Powell and M. Hiller, *J. Chem. Ed.*, **34**, 330 (1957).

uncertainty in ΔF_1 is about 300 calories and in ΔF_2 about 100 calories. Moreover the variation in ΔF_2 is fitted within 20 calories by the parameters obtained graphically and shown at the bottom of Table II for the equation $\Delta F = \Delta H - T\Delta S$ where $\Delta H = 700 + 17[\mathrm{U}]$. From these figures the small effect of urea is on the enthalpy of the hydrogen bonding reaction rather than on the entropy. It is interesting to note that the enthalpy of hydrogen bonding is positive and the reaction is therefore endothermic and "driven" by the positive entropy change.

TABLE II

CONSTANTS AND THERMODYNAMIC PARAMETERS OF HYDROGEN BONDING IN MALEIC ACID

Urea, M	Temp. °C.	L_1	L_2	ΔF_1, cal./mole	ΔF_2, cal./mole	ΔF_2 calcd.,[a] cal./mole
0	7	3.6	44.8	−720	−2120	−2100
3.5	7	3.2	38.8	−640	−2040	−2040
7.0	7	2.2	33.7	−440	−1970	−1980
0	25	3.3	44.7	−710	−2240	−2280
3.5	25	2.5	43.7	−560	−2230	−2220
7.0	25	1.3	38.9	−150	−2170	−2160
0	37	3.9	50.4	−840	−2410	−2400
3.5	37	4.1	43.7	−870	−2330	−2340
7.0	37	4.9	40.7	−980	−2280	−2280

[a] From the equation $\Delta F = 700 + 17[\mathrm{U}] - 10T$; [U] is moles per liter and T is the absolute temperature.

Malonic Acids.—In Table III are set forth the titration constants of malonic acid and some derivatives at a series of temperatures and in various concentrations of urea. It is evident from an examination of the data at 0 M urea and 37° that substitution of hydrocarbon groups on the methylene group has two kinds of effects. The regular progression in the half ester series is interpretable as the inductive effect of hydrocarbon substituents and is similar though greater in magnitude than that observed by Larsson[12] for monocarboxylic acids. The dicarboxylic acids show an increasing spread of the two constants, not very great for the dimethyl derivative but quite marked at diethyl. We will assume that the increasing bulk of the substituents (steric hindrance) forces the carboxyl centers closer to each other. At some critical separation the hydrogen bond increases the stability of the conformation and a significant hydrogen bond is formed. In our data this occurs between dimethyl and diethylmalonic acids. We have chosen to assume that the same inductive effects occur in the dicarboxylic acids as in the half esters and have added the differences between pK_e for malonic acid and pK_e for the acid under examination to each of the pK of malonic acid to obtain pK_1 and pK_2 for equation 7 and 8. No correction for electrostatic interaction of the carboxylate groups has been made because the tetrahedral structure of the central carbon atom could not be greatly distorted. Thus we assume the distance of separation of the two carboxyl groups is not significantly different in the various malonic acids. pG_1 and pG_2 are the titration constants given in Table III. From these we have calculated the hydrogen bonding constants and free energies in Table IV.

TABLE III

MALONIC ACID AND DERIVATIVES, pK IN UREA SOLUTIONS

Urea, M	Temp., °C.	Malonic pK_1	Malonic pK_2	Malonic pK_e	Dimethyl malonic pK_1	Dimethyl malonic pK_2	Dimethyl malonic pK_e	Diethyl malonic pK_1	Diethyl malonic pK_2	Diethyl malonic pK_e	Ethyl isopropyl malonic pK_1	Ethyl isopropyl malonic pK_2	Ethyl isopropyl malonic pK_e
0	7	2.75	5.31	3.23	..	..	..	2.06	7.00	3.44	..	..	..
3.5	7	3.00	5.51	3.52	..	..	..	2.56	7.10	3.71	..	..	..
7.0	7	3.32	5.78	3.83	..	..	..	2.81	7.29	4.05	..	..	..
0	25	2.74	5 31	3.25	3.03	5.73	3.49	2.15	7.04	3.54	..	..	..
3.5	25	2.96	5.50	3.49	3.31	5.91	..	2.41	7.16	3.79	..	..	..
7.0	25	3.23	5.84	3.76	3.51	6.11	3.91	2.73	7.32	4.03	..	..	..
0	37	2.79	5.48	3.31	3.06	5.80	3.55	2.21	7.12	3.64	2.18	7.93	3.72
3.5	37	2.93	5.58	3.54	3.35	5.99	3.76	2.25	7.25	3.87	2.35	8.02	3.91
5.0	37	3.11	5.65	3.67	3.38	6.04	..	2.73	7.29	3.99	2.59	8.10	4.07
7.0	37	3.19	5.83	3.74	3.49	6.16	3.93	2.85	7.42	4.16	2.66	8.16	4.19
8.0	37	3.30	5.85	3.86	3.60	6.23	..	2.74	7.43	4.17	2.51	8.33	4.22

Examination of the ΔF_2 values for diethyl malonic acid discloses somewhat irregular changes with temperature. The ΔS values required will be small but sometimes positive and sometimes negative. The enthalpy will be negative and close to the free energy. In this case then the hydrogen bond formation is driven by bonding energy; entropy appears to play a minor role. However, a closer analysis should be reserved for a later time when more data are available.

Table IV shows that intramolecular hydrogen bonding in the malonic acid derivatives increases with the number of carbons in the substituent groups and is somewhat weakened (by about 160 calories in 7 M urea) in urea solutions.

Discussion

Electrostatic Interaction.—The ratio of the two K of a dibasic acid is larger than called for by statistical factors, especially if the two carboxyl groups are close together. Bjerrum[13] suggested electrostatic interaction would cause this effect. The interaction is a function of the distance separating the two potential charges and of the dielectric between them. Gane and Ingold[14] applied this idea to many dicarboxylic acids, assumed the dielectric constant involved to be that of the medium and calculated the distance of separation from the ratio K_1/K_2 as determined by experiment.

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(12) E. Larsson and B. Adell, *Z. Physik. Chem.*, **153**, 352 (1931); **156**, 381 (1932); **157**, 342 (1932); **159**, 306 (1933).

(13) N. Bjerrum, *ibid.*, 106, 219 (1923).

(14) R. A. Gane and C. K. Ingold, *J. Chem. Soc.*, 1594, 2267 (1928); 1691 (1929).

TABLE IV

MALONIC ACIDS

Urea, M	Temp., °C.	Dimethyl malonic L_1	Dimethyl malonic L_2	Dimethyl malonic ΔF_1, cal./M	Dimethyl malonic ΔF_2, cal./M	Diethyl malonic L_1	Diethyl malonic L_2	Diethyl malonic ΔF_1, cal./M	Diethyl malonic ΔF_2, cal./M	Ethyl isopropyl malonic L_1	Ethyl isopropyl malonic L_2	Ethyl isopropyl malonic ΔF_1, cal./M	Ethyl isopropyl malonic ΔF_2, cal./M
0	7	..	..	...	...	2.80	25.3	−570	−1800	...	...	....	
3.5	7	..	..	...	...	4.89	24.1	−880	−1770	...	...	....	....
7.0	7	..	..	...	...	2.63	18.5	−540	−1630	...	...	....	....
0	25	0.48	0.41	+435	+528	2.63	26.5	−570	−1940	...	...	....	....
3.5	25	1.29	.62	−150	+283	2.23	21.9	−470	−1830	...	...	....	....
7.0	25	0.78	.32	+147	+675	1.75	15.2	−330	−1620	...	...	....	....
0	37	0.29	.20	+763	+992	1.51	19.4	−250	−1830	9.47	108	−1380	−2880
3.5	37	1.45	.55	−228	+370	1.14	20.9	− 80	−1870	9.50	104	−1390	−2860
5.0	37	0.26	.19	+122	+370	3.17	19.9	−710	−1840	12.5	111	−1560	−2900
7.0	37	.55	.38	+368	+596	1.57	13.8	−280	−1620	6.95	149	−1190	−2660
8.0	37	.90	.51	+ 65	+414	1.51	17.6	−250	−1770	8.33	130.8	−1300	−3000

These distances are often unreasonably short, but by applying corrections designed to take account of the effect of "radially oriented and attracted solvent molecules surrounding an ion on local electrical conditions"[15] more reasonable distances resulted.[16] Schwarzenbach[17] assumed distances fixed by molecular models and showed that the shorter the distance the lower the apparent dielectric constant had to be to accommodate the data. Kirkwood and Westheimer[18] modified the treatment by placing the interacting groups on a cavity (the volume of the molecule) of low dielectric constant compared to that of the medium. The interaction then became a function of an effective dielectric constant. This number is constructed upon parameters depending on (a) the actual dielectric constant of the cavity (b) the dielectric constant of the medium, (c) the shape of the cavity and its volume and (d) the location of the charge centers within or upon the cavity.[19] Appropriate choice of parameters led Kirkwood and Westheimer to reasonable distances between the carboxyl groups of the dibasic acids. Tanford[22] noted that the Kirkwood and Westheimer[18] dimensions were consistent with the placement of the charges approximately 1 Å. within the Kirkwood–Westheimer cavity and noted the effect this would have on the "effective dielectric constant." This depth can be another adjustable parameter. The availability of the three adjustable and more or less arbitrary parameters for the calculation of expected ratios of K_1/K_2 for dicarboxylic acids inhibited attempts to correct for the effects of the different distances in fumaric acid (6.3 Å.) and maleic acid (4.5 Å.) derived from models by Ashton and Partington.[23] As used the parameters give reasonable distances between charge centers for most acids because they were so chosen. Hydrogen bonding is invoked only if no reasonable choice can be made.[22] Its absence is otherwise the assumed condition. For the malonic acids we feel that little latitude for change of dimensions and placement exists so that no electrostatic correction is needed for the model compound.

Urea solutions have a higher dielectric constant than does water, and there is no reason to expect that *a*, *c* or *d* would differ between water and urea solutions. The effective dielectric constant should therefore increase, and all acids should become stronger in urea solutions than in water. In fact they are weaker[6,24,25] and the differences are dependent upon water activities to an extent which suggests that net increase in hydration on dissociation is the main factor determining the behavior.[6,25] Increasing effective dielectric constant should also diminish the interaction between charge centers. Urea should increase the effective dielectric constant because the medium dielectric constant contributes to it. Since the expectation of increasing strength of monocarboxylic acids in urea solutions is not fulfilled, we will assume that changes in effective dielectric constant are not a controlling factor in the effect of urea on dicarboxylic acids either.

Hydration.—To consider the effect of the changes in water concentrations accompanying the changes in urea concentrations, we write the equilibrium between a non hydrogen bonded form MH^- and a hydrogen bonded form $\overline{MH}^-$ to include water of hydration in each case thus

$$MH(H_2O)_q \rightleftharpoons \overline{MH}^-(H_2O)_n + (q - n)H_2O$$

and the equilibrium equation

$$L_n = \frac{[\overline{MH}(H_2O)_n]\ [H_2O]^{(q-n)}}{[MH(H_2O)_q]}$$

which is equivalent to $L_n = L_2(H_2O)^{(q-n)}$. The

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(15) C. K. Ingold, *J. Chem. Soc.*, 2179 (1931).

(16) R. Gane and C. K. Ingold, *ibid.*, 2153 (1931).

(17) C. Schwarzenbach, *Z. Physik. Chem.*, **176**, 133 (1936).

(18) J. Kirkwood and F. H. Westheimer, *J. Chem. Phys.*, **6**, 506, 513 (1938).

(19) Wynne-Jones and Rushbrooke[20] developed this kind of analysis into a criticism of the Kirkwood and Westheimer treatment, concluding that the treatment did not take sufficiently into account "specific" (*i.e.*, close range) interactions between solvent and solute molecules or ions. In their reply Kirkwood and Westheimer[21] disclaim anything beyond a rough basic model and state that any refinement of theory must indeed be based on models having more details of structure both of solute and solvent and their interactions specified. The critique in the next paragraph is in part an extension of Wynne-Jones and Rushbrooke's thesis.

(20) W. F. K. Wynne-Jones and G. S. Rushbrooke, *Trans. Faraday Soc.*, **40**, 99 (1944).

(21) F. H. Westheimer and J. G. Kirkwood, *ibid.*, **43**, 77 (1947).

(22) C. Tanford, *J. Am. Chem. Soc.*, **79**, 5348 (1957).

(23) H. W. Ashton and J. R. Partington, *Trans. Faraday Soc.*, **30**, 598 (1934).

(24) N. F. Burk and D. M. Greenberg, *J. Biol. Chem.*, **87**, 197 (1930).

(25) M. Levy, *Fed. Proc.*, **17**, 263 (1958).

most convenient way to show this relationship is to plot the equation $\log L_2 = \log L_n - (q - n) \log (H_2O)$. Such plots of our data for maleic acid give positive slopes of 0.5 to 0.8 indicating that n is larger than q but not by more than 1. Whether it is reasonable to expect the hydrogen bonded form to carry more water of hydration than the non-bonded form is moot, but the difference is small and probably permissible. At any rate the effect of urea solutions on hydrogen bonds of this character might be accounted for simply on the basis of differential hydration and the reduction of water concentration. This matter should be tested by measuring hydrogen bonding constants in other mixed solvents of similar water activities.

The Effect of Urea on Hydrogen Bonds.—The effects of urea on hydrogen bonds are given by the changes in the free energies of the bonds resulting from the presence of urea. For maleic acid at the three temperatures the differences between 0 and 7 *M* urea, 130 cal. for carboxyl–carboxylate interactions is in the direction of weakening. For the malonic acids these differences average 90 cal., for carboxyl–carboxyl interactions in the direction of weakening and 240 cal. for carboxyl–carboxylate interactions in the direction of strengthening. The differences are all rather small and not much greater or less than the estimated error (100–200 cal.) from the uncertainties in the pK values. We therefore conclude that urea in aqueous solution has little or no effect on such hydrogen bonds beyond that associated with differences related to the lower water concentrations at high urea concentration.

Our first calculations of hydrogen bonding constants were made following the equation of Westheimer and Benfey[10] and showed an *increase* of the bonding constants as urea concentration increased, and it was on this basis that our preliminary reports were made. The more sophisticated treatment outlined and applied above shows that the hydrogen bonding constants decrease with urea concentration but not sufficiently to alter our conclusion that urea does not have a significant effect on intramolecular carboxyl–carboxyl or carboxyl–carboxylate bonds.

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, UNIVERSITY OF MISSOURI, COLUMBIA, MISSOURI]

The Rates of Formation and Dissociation of 2,2′-Ethylenediamino-bis-(2-methyl-3-butanone)-dioxime Nickel(II) Ions[1]

BY R. KENT MURMANN

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The rate of formation [Ni(EnAO–H]$^+$ follows the law: rate of formation = $k_1[\text{Ni}^{+2}][\text{EnAOH}^+] + k_2[\text{Ni}^{+2}][\text{EnAO}]$ with $k_1 = 2.3 \times 10^2$ and $k_2 = 1.5 \times 10^6$ l. moles^{-1} min.$^{-1}$ at 24.3° and $\mu = 1.00$ with LiCl. The acid dissociation of [Ni(EnAO–H]$^+$ proceeds according to the rate equation: Rate = $k[\text{Ni(EnAO)–H}]^+ + k_1[\text{Ni(EnAO)}^{+2}] + k_2[\text{NiEnAO}^{+2}][\text{H}^+]$: at 24.3° and $\mu = 1.00$, $k = 4.58 \times 10^{-3}$ min.$^{-1}$, $k_1 = 2.65 \times 10^{-2}$ min.$^{-1}$ and $k_2 = 5.8 \times 10^{-2}$ at unit concentration of HCl. Substitution on the amine nitrogen and the loss of one of the chelate rings increases the rate of dissociation in acid. The equilibrium constant for the formation of [Ni(EnAO)]$^{+2}$ from the rate constants is smaller by a factor of $10^{2.5}$ than that obtained by equilibrium measurements. This may be due to the slow Ni(II) catalyzed geometrical rearrangement of the oxime groups or to the stepwise detachment of the ligand in the dissociation.

Introduction

The addition of 2,2′-ethylenediamino-bis-(2-methyl-3-butanone) dioxime (EnAO)[2] to Ni^{+2} in essentially neutral aqueous solution has been shown to lead to the formation of a yellow, planar complex having the structure shown in Fig. 1a. In rapid equilibrium with it is the blue, paramagnetic and presumably octahedral complex (Fig. 1b) which has the hydrogen bond broken by the addition of a hydrogen ion; the other two octahedral positions are probably filled with solvent molecules (not shown).

The kinetics of the formation and decomposition of this type of complex is especially interesting in the light of the work of Wilkins, *et al.*,[3a] on the rate of ligand exchange and the rate of dissociation of C-substituted ethylenediamine nickel(II) complexes in which the rates of dissociation were acid dependent and slower with increased alkyl size. [Ni(EnAO)]$^{+2}$ lends itself to a kinetic study because (a) both the forward and reverse rates are measurably slow, (b) the ligand is a tetradentate which removes the difficulties of varying ligand/metal ratio during the measurements and (c) most of the thermodynamic data necessary for evaluation of the rate expression have previously been measured.[1] Of special interest is the hydrogen ion dependence of the rate of formation and hydrolysis for the light it may shed on the mechanism of this and other complex ion reactions.

In this paper are reported the results of a study of the rate of formation and the rate of acid hydrolysis of [Ni(EnAO)-H]$^+$ at an ionic strength of 1.00 (LiCl) and 24.3°.

Experimental

[Ni(EnAO)-H]ClO$_4$ and EnAO were prepared by methods previously described.[1]

Anal. of [Ni(EnAO)-H]ClO$_4$. Calcd. for $NiC_{12}H_{25}N_4O_6Cl$: Ni, 14.14; N, 13.49; Cl, 8.54. Found: Ni, 14.23; N, 13.28; Cl, 8.57.

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(1) For previous papers on this topic see R. K. Murmann, *J. Am. Chem. Soc.*, **80**, 4147 (1958); **79**, 521 (1957).

(2) The following abbreviations will be used in this paper: (EnAO) = 2,2′-ethylenediamino-bis-(2-methyl-3-butanone)-dioxime, [Ni(EnAO)]$^{+2}$ = 2,2′-ethylenediamino-*bis*-(2-methyl-3-butanone) dioxime nickel(II) ion (Fig. 1b), [Ni(EnAO)-H]$^+$ is meant to express the product of the ionization of an hydrogen ion from [Ni(EnAO)]$^{+2}$ in the formation of a hydrogen bond. (MeEnAO) = 2,2′-propylenediamino-bis-(2-methyl-3-butanone)-dioxime. All other abbreviations are given in earlier papers.

(3) (a) R. G. Wilkins, *J. Chem. Soc.*, 4521 (1957). (b) F. P. Dwyer and A. Bryson. *J. Proc. Roy. Soc. N. S. W.*, **74**, 471 (1940).