strain in norbornane is +0.38 kcal/mole. Subtracting this amount gives the total calculated strain of 17.1 kcal/mole for norbornane.[23]

Although the calculated value for the energy is in striking agreement with that observed (17.8–18.2 kcal/mole), Allinger's deduction of the geometry of norbornane at minimum energy is regrettably in poor agreement with observation. While the angles he calculated[24] correspond fairly well to our measured ones, all the C–C internuclear distances are smaller by about 0.03 Å, even after adding the 0.006 Å for conversion to the microwave scale.[21] Worse yet, the model calculation predicts $(C_2-C_3) \gtrapprox (C_1-C_2) > (C_1-C_7)$ while the observed distances decrease in the reverse order. Finally, Allinger found that his minimum energy structure had a strain energy of 12 kcal/mole; consequently, correspondence of the value calculated in this paper with the observed magnitude of (17.8–18.2) kcal/mole must be largely fortuitous. It appears that, despite the success of both of Allinger's models in calculating differences in conformational energies of flexible molecules, they do not satisfactorily reproduce the value or the character of strain present in norbornane.

The present analysis suggests a hierarchy of difficulties in testing theory against experiment. All of the strain models in use today appear to be successful in reproducing conformational energy differences. Less accurately, but still acceptably, is the applicability of the models in seeking the correct minimum energy structure. The least reliable results appear to be the calculated strain energies of highly distorted molecules.

(23) This is not entirely correct since the calculated value is for a single linear conformation rather than for the mixture actually present in *n*-heptane, in terms of which the strain energy of norbornane is defined. Properly, the energy of any single species should be weighted over the energies of the different conformations accessible to the system. A calculated energy based on the single most stable species tends to be too negative, the more so as the number of accessible states increases. On the other hand, the parameters of the model have been selected to reproduce heats of isomerization such as that accompanying *n*-butane to isobutane, and thus indirectly the requisite weighting has already (but approximately) been included.

(24) N. L. Allinger, private communication.

Acknowledgments. The authors express their sincere thanks to the National Science Foundation, GP-4291, and the Material Science Center, ARPA-SD-68, for support of the work presented in this report. The structural study of these compounds was initiated by Dr. Mary E. Wilcox, who made preliminary analyses of the electron diffraction photographs. We also thank Dr. Robert K. Bohn for taking the second set of electron diffraction photographs on the Oslo apparatus and for providing us with copies of his reduced intensity scattering patterns.

Dielectric Dispersion and Chemical Relaxation

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Abstract: Starting from Debye's concept of dielectric dispersion, it is shown how chemical relaxation effects can contribute to the frequency dependence of dielectric permittivity. Since for rapidly orienting dipole molecules the chemical effect is nonlinear, it can be observed only if a very strong dc field is superimposed to the alternating field. The chemical-field effect could be identified for dimerization and association processes involving H-bond chelation. The dispersion range was found to be in the MHz region and thus clearly separated from the orientational relaxation in the microwave range. The magnitude of the chemical effect is very small and requires very sensitive methods of detection. The technique opens the possibility of studying the elementary steps of base pairing as involved in the information transfer in nucleic acids.

I. The Debye Curve of Dielectric Dispersion

Among Peter Debye's various achievements, the interpretation of the phenomena of electrical polarization stands up as a landmark in molecular physics. Actually, this subject intrigued him so much, that the only book he wrote[1] was dedicated to this field. It was especially the dynamic behavior of polar molecules on which he centered his interest.

The anomalous dispersion of the dielectric constant of certain substances was already observed by Drude[2] as early as 1895, but it was Debye[3] who could show that this phenomenon was due to the existence of polar molecules, which can be oriented by an electric field. The dielectric dispersion then can be explained by a delay of orientation due to the viscous motion of the molecular dipoles. The temporal delay can be characterized by a time constant τ_{or}. In an alternating field of the (angular) frequency ω ($\approx 1/\tau_{or}$), the polarization will lag behind the orienting field with a phase angle $\varphi = \arctan(\omega\tau_{or})$. At the same time the amplitude of the orientational polarization will decrease, and an energy loss will occur showing up as a contribution to electrical conductivity.

Both amplitude dispersion and energy loss can be described by a complex dielectric permittivity

$$\epsilon_0\epsilon_\infty + \frac{\epsilon_0\epsilon_{or}}{1 + j\omega\tau_{or}} \qquad (1)$$

(1) P. Debye, "Polare Molekeln," S. Hirzel Verlag, Leipzig, 1929; "Polar Molecules," New York, N. Y., 1929.
(2) P. Drude, *Z. Physik. Chem.*, 23, 267 (1897).
(3) P. Debye, *Verhandl. Deut. Phys. Ges.*, **15**, 777 (1913).

leading to a real term (dispersion)

$$\frac{\epsilon_0\epsilon_{or}}{1 + \omega^2\tau_{or}^2} \tag{2}$$

and an imaginary term

$$\epsilon_0\epsilon_{or}\frac{\omega\tau_{or}}{1 + \omega^2\tau_{or}^2} \tag{3}$$

which, multiplied with ω, represents the conductivity increase. For small molecules in liquids the dispersion region is usually found at microwave frequencies.

The above treatment was introduced by Debye more than 50 years ago, and therefore any such frequency terms are nowadays associated with the name of Debye. On the other hand, such terms are not restricted to dielectric processes. They can be found as well in acoustical relaxation phenomena,[4] and several authors[5-7] have used a similar treatment to describe the dispersion and absorption of sound caused by relaxation of chemical equilibration. In this connection it would be of interest to find out whether chemical relaxation phenomena would also contribute similar terms to dielectric polarization. Such studies would be of great value, since dielectric measurements can be performed with great accuracy over a considerable frequency range.

II. The Field Effect of Dipolar Association Equilibria

In the study of rapid chemical recombination processes, we require a molecular parameter which changes sufficiently upon association. In some cases, for instance, in the pair formation by hydrogen bonding, the only useful parameter turned out to be the electric moment. As an example let us consider the pair formation of a purine and pyrimidine base as involved in the double-stranded helical structure of nucleic acids. Because the group moments are partially compensated in hydrogen-bonded chelates (two oppositely oriented H bonds), the electric moment changes quite drastically upon pair formation. This will cause an electric-field dependence of the association equilibrium. If A and B represent the two reaction partners and AB the hydrogen-bonded pair, the field dependence of the pairing equilibrium

$$A + B \rightleftharpoons AB$$

is given by the analog of the van't Hoff relation

$$\frac{\partial \ln K}{\partial E} = \frac{\Delta M}{RT} \tag{4}$$

where K denotes the equilibrium constant, E the electric field strength, and ΔM the "reaction moment," *i.e.* the difference of partial molar moments of reactants and products

$$\Delta M = M_{AB} - M_A - M_B \tag{5}$$

For dilute solutions of polar molecules in a nonpolar solvent, the partial molar moment can easily be calculated from the polarizability and the permanent dipole moment using the treatment of Langevin[8] and Debye.[1]

A more general treatment given by Onsager[9] distinguishes the internal and directing fields (field strength E_i and E_r, respectively) and yields

$$M = N_A\left(\alpha E_i + \frac{\mu^2}{3kT}E_r\right) \tag{6}$$

(N_A being the Avogadro number). It is important to note that M is proportional to the square of the permanent dipole moment rather than the moment itself. This is due to the disorienting Brownian motion which in the absence of the field causes a completely random orientation. The linear dependence of M on E holds only at sufficiently low field strengths, where no saturation of orientation occurs. For small molecules this condition is fulfilled up to field strengths of 10^6 V/cm. The complete expression for ΔM can be obtained from eq 6 using Onsager's[9] theory and the Lorenz–Lorentz approximation for the polarizability

$$\Delta M = N_A\frac{\epsilon^2(n^2 + 2)^2}{2\epsilon^2 + n^4}\frac{(\mu_{AB}^2 - \mu_A^2 - \mu_B^2)}{9kT}E \tag{7}$$

It is certainly an advantage that ΔM is proportional to the difference of the squares of permanent moments. As a consequence, one will usually find a finite difference, even if the permanent moments do not compensate at all. On the other hand, the proportionality of ΔM to E represents a disadvantage. Actually, this is the reason why the chemical effect never has been observed in dielectric dispersion and absorption measurements. Since such measurements are done with small field amplitudes, there is no (linear) influence on the chemical equilibrium as can be seen from Figure 1. Any finite shift of the equilibrium constant will be proportional to E^2 as follows from integration of eq 4.

Similarly any dielectric increment of chemical relaxation will also be proportional to E^2 according to

$$\epsilon_{chem} = \left(\frac{\partial M}{\partial \xi}\right)\frac{\partial \xi}{\partial E} = \frac{\Gamma(\Delta M)^2}{\epsilon_0 VRT}\frac{1}{1 + j\omega\tau_{chem}} \tag{8}$$

with ξ being the extent of reaction and Γ a factor depending on the stoichiometry of the reaction, which in the present case is given by

$$\Gamma = [1/C_A + 1/C_B + 1/C_{AB}]^{-1} \tag{9}$$

(C_i = molar concentrations). τ_{chem} is the relaxation time for chemical equilibration (*cf.* below).

The chemical field effect can be observed most conveniently if a strong dc field is superimposed on a small amplitude alternating field. Then the usual procedures of linearization with respect to the alternating field can be used. Since ϵ_{chem} is a complex quantity, one will find, as shown above for Debye's treatment of orientational relaxation, a dispersion and energy loss (tan δ) term. For practical reasons the effect can be observed more sensitively as an increment of tan δ, which is given by

$$(\tan\delta)_{chem} = \frac{\epsilon_{chem}}{\epsilon_{tot}}\frac{\omega\tau}{1 + \omega^2\tau^2} \tag{10}$$

The chemical field effect was identified recently for solutions of ϵ-caprolactam in benzene and carbon tetrachloride. The results and a more extensive theoretical treatment were given in a paper together with Berg-

(4) K. F. Herzfeld and T. A. Litovitz, "Absorption and Dispersion of Ultrasonic Waves," Academic Press Inc., New York, N. Y., 1959.
(5) A. Einstein, *Sitzber. Preuss. Akad. Wiss. Physik-Math. Klasse*, 380 (1920).
(6) J. Meixner, *Kolloid-Z.*, **134**, 3 (1953).
(7) M. Eigen, G. Kurtze, and K. Tamm, *Z. Elektrochem.*, **57**, 103 (1953).
(8) P. Langevin, *J. Phys.*, **4**, 678 (1905).
(9) L. Onsager, *J. Am. Chem. Soc.*, **58**, 1486 (1936).

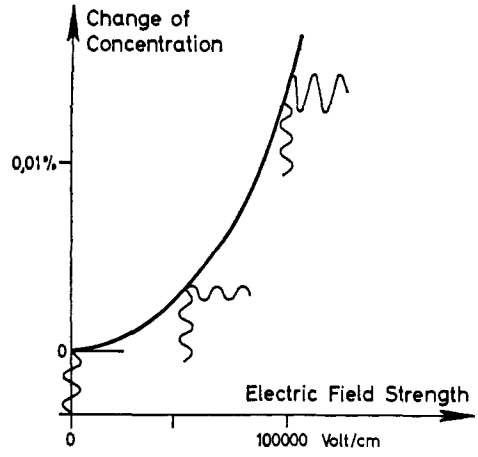


Figure 1. Dependence of chemical concentration changes upon external electric field strength for dipolar association equilibria.

mann.[10,11] The effect is not easily detectable since, even under the extreme conditions of dc fields of 2–3 × 10^5 V/cm, $(\tan \delta)_{chem}$ amounts to only 10^{-4}–10^{-6}. On the other hand, the effect can be most easily distinguished from orientational relaxation because of the quadratic dependence on the dc field strength, the concentration dependence of the relaxation time τ_{chem} (according to a second-order recombination mechanism), and the position of the absorption maximum (tan δ) in the megahertz rather than the microwave region. It provides information about equilibria, rates, and mechanisms of dipolar association reactions like the mentioned process of base pairing (an example will be discussed in the next section).

The above treatment requires the orientation of dipoles to be fast as compared with chemical relaxation. This condition is usually fulfilled for small molecules. On the other hand, large polymeric molecules may remain at a fixed orientation during the process of chemical relaxation. Although the equilibrium then does not shift as long as the molecules remain at a fixed orientation, there is an alternative way of polarization *via* the chemical equilibration. Schwarz[12] has shown that this effect can be found in absence of strong fields since it is linear in E. He used it to measure the kinetics of helix–random coil transition in long-chain polypeptides.

III. Experimental Techniques

An experimental determination of the chemical effects described above requires a very sensitive measurement of small increments in the dielectric permittivity or the dielectric loss upon application of a strong dc field. The field strength that can be used in liquid media is limited by electric breakdown to about 3 × 10^5 V cm^{-1}. Indeed it turned out that field strengths of this order of magnitude are required to raise the effects to a detectable level. At low frequencies ($<\sim$1 MHz) the admittance of a suitable two-terminal element (a capacitor containing the dielectric medium to be investigated) can be compared to standard resistances and reactances in a bridge circuit. At higher frequencies ($<10^8$–10^9 Hz), resonant circuits are used,

(10) K. Bergmann, M. Eigen, and L. De Maeyer, *Ber. Bunsenges. Physik. Chem.*, 67, 819 (1963).
(11) K. Bergmann, *ibid.*, 67, 826 (1963).
(12) G. Schwarz, *J. Phys. Chem.*, 71, 4021 (1967).

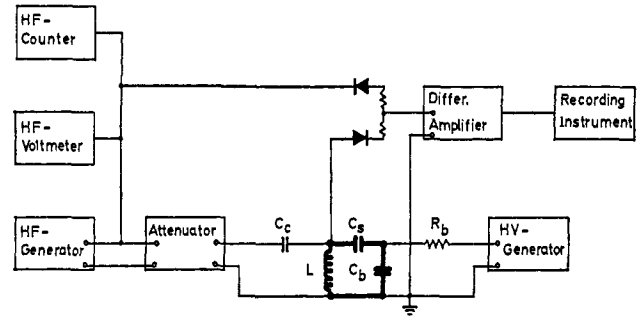


Figure 2. Block diagram of the measuring circuit.

in which the sample cell capacitance is combined with an inductance to give parallel or series resonance. Conductance and susceptance of the sample are then derived from accurate measurements of the resonance frequency and of the width of the resonance curve. At still higher frequencies, resonant circuits with distributed capacitance and inductance are used, and the dielectric properties must be derived from the equations describing electromagnetic wave propagation in these structures.

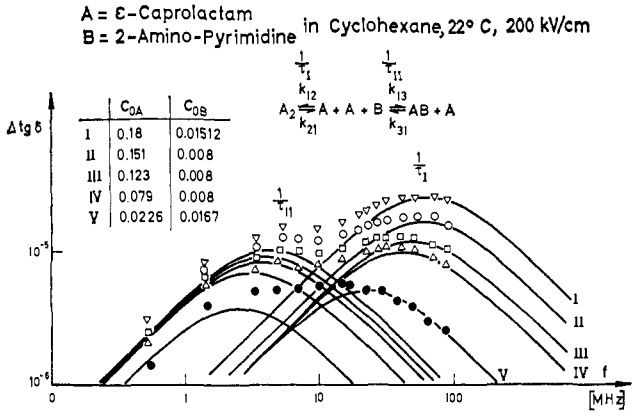
Bimolecular reaction rates in dilute solutions are limited by the diffusion of the reaction partners; at concentrations below 10^{-1} M the relaxation frequency of such reactions can hardly exceed a few hundred megahertz. For this reason the resonance method was chosen to study the reaction kinetics of the formation of hydrogen-bond-associated chelates in nonpolar solvents.

As a first example, the dimerization of ε-caprolactam was studied,[11] since this compound has a relatively large dipole moment ($\sim$4 D), and is quite soluble in nonpolar solvents. Since then, the method has been applied to the study of association reactions of ε-caprolactam with other compounds, and of association reactions between molecules of the purine and pyrimidine structure, as models for the base-pairing reactions in nucleic acids.[13,14]

The experimental technique described earlier[11] has been modified and will briefly be outlined. A schematic diagram of the circuitry used is given in Figure 2. The sample cell capacitor C_s is part of a resonant circuit comprising an exchangeable inductance L and blocking capacitor C_b ($C_b \gg C_s$) that prevents short-circuiting the dc voltage by the inductance. High voltage can be applied to the sample *via* a large resistor R_b, which avoids loading the resonant circuit by the HV-generator. HF energy is coupled into the resonant circuit *via* a small coupling capacitance C_c ($C_c \ll C_s$). The variable frequency generator covers the range of 0.1–100 MHz. Its output is monitored and measured by a HF-voltmeter, and the frequency is determined accurately with a digital counter. The HF voltage across the inductance of the resonant circuit is compared with the generator output by peak-rectifying diodes and a comparison circuit.

With this arrangement the resonance frequency ω_0 and the resonance voltage U_0 at zero field can be determined accurately as well as the incremental values $\Delta\omega_0$ and ΔU_0 after the high dc field has been applied.

(13) J. Suarez, Dissertation, Göttingen, 1967.
(14) R. Hopmann, L. De Maeyer, J. Suarez, and S. Thein, to be published.



	c_{0A}	c_{0B}
I	0.18	0.01512
II	0.151	0.008
III	0.123	0.008
IV	0.079	0.008
V	0.0226	0.0167

Figure 3. Measured increments in the dielectric loss factor upon application of a high dc field as a function of frequency for different concentrations of the reaction partners. The curves are drawn according to the theoretical shape of the Debye equation.

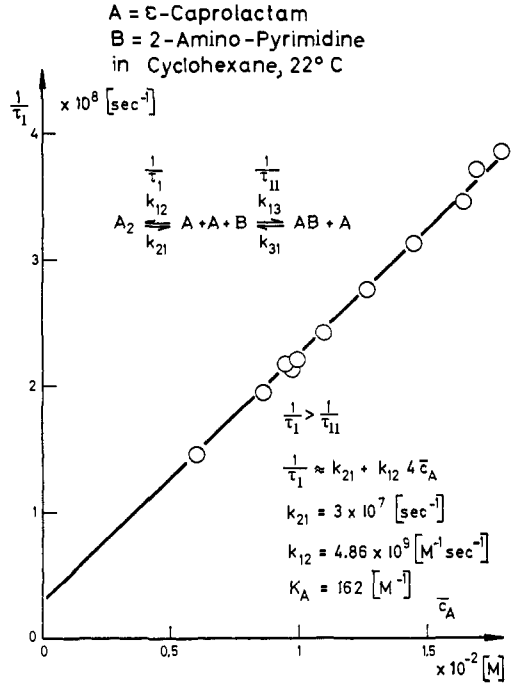


Figure 4. Dependence of the reciprocal relaxation time $1/\tau_I$ as a function of concentration.

From a consideration of the different circuit parameters one obtains

$$(\Delta \tan \delta)_E = \frac{\gamma}{Q_0}\left(\frac{\Delta\omega_0}{\omega_0} - \frac{\Delta U_0}{U_0}\right)_E \qquad (11)$$

The quantity γ depends on parasitic capacitances of the sample cell and is obtained from calibration measurements involving solutions with known increments in dielectric constant. Q_0 (circuit quality) is measured from the width of the resonance curve.

A detailed description of the experimental technique will be given elsewhere.[14] Figure 3 shows an example of the measurements of the chemical relaxation effects in solutions of ε-caprolactam with 2-aminopyrimidine in cyclohexane at a dc field of 200 kV cm^{-1}. Two discrete relaxation processes are observable, attributed to the dimerization of the lactam and to the formation of a 1:1 associate between ε-caprolactam and 2-aminopyrimidine (dimerization of the 2-aminopyrimidine would be detectable only at much higher concentrations as could be shown by independent measurements in solutions containing 2-aminopyrimidine only).

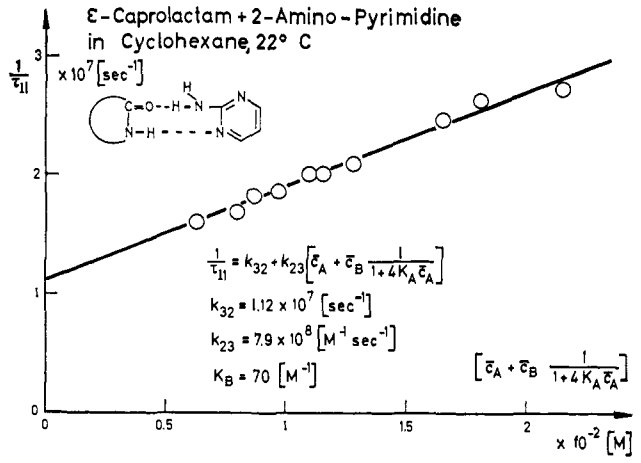
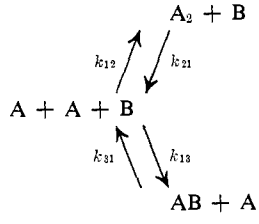


Figure 5. Dependence of the reciprocal relaxation time $1/\tau_{II}$ as a function of concentration.

IV. Kinetics of Base Pairing

The system ε-caprolactam (A) and 2-aminopyrimidine (B) represents an example of base pairing with two competing reactions (eq 12). Such a reaction scheme



(12)

is characterized by two relaxation times. They are usually not simply related to the individual reaction pathways but belong to "normal coordinates" of reaction, and each of them depends, in the general case, upon all the individual rate constants of the coupled reaction scheme.

The fact that the two relaxation times, as seen in Figure 3, differ by about an order of magnitude, allows a considerable simplification of the expression for the relaxation times. A comparison of the measured concentration dependence given in Figure 4 with results obtained with pure ε-caprolactam solutions allowed the identification of the relaxation processes and led to a further simplification of the expression for the relaxation times. The reciprocal relaxation times are given by

$$\frac{1}{\tau_I} = k_{21} + 4k_{12}\bar{c}_A \qquad (13)$$

$$\frac{1}{\tau_{II}} = k_{31} + k_{13}\left(\bar{c}_A + \frac{1}{1+4K'\bar{c}_A}\bar{c}_B\right) \qquad (14)$$

τ_I is the shorter and τ_{II} the longer relaxation time; $\bar{c}_A$ and $\bar{c}_B$ are the equilibrium concentrations of A and B. Since the equilibrium constants K' of the dimerization and K'' of the 1:1 association were known,[15] these

(15) K' is known from measurements with solutions containg no 2-aminopyrimidine; K'' was determined spectrophotometrically by R. Hopmann (unpublished results).

equilibrium concentrations could be calculated from the initial concentrations of A and B. In Figures 4 and 5, $1/\tau_I$ and $1/\tau_{II}$, respectively, are plotted according to the above equations. From those plots one may derive $k_{12} = (4.8 \pm 0.2)10^9\ M^{-1}\ sec^{-1}$; $k_{21} = (3.0 \pm 0.2)10^7\ sec^{-1}$; $K' = 162 \pm 10\ M^{-1}$; $k_{13} = (7.9 \pm 0.2)10^8\ M^{-1}\ sec^{-1}$; $k_{31} = (1.1 \pm 0.2)\ 10^7\ sec^{-1}$; $K'' = 70 \pm 5\ M^{-1}$. From these results it may be concluded that the formation of hydrogen-bonded dimers and molecule pairs is very rapid (diffusion controlled at least in the case of the dimerization of ϵ-caprolactam). The pairing takes place in a single step, which means that the second H bond is formed as fast as or faster than the first H bond is broken up again.

The fast pairing reaction is essential for the fast recognition of complementary bases in genetic code reading.

On the other end, the lifetime of the paired complex is very short, *i.e.*, of the order of 10^{-8}–10^{-7} sec. The dissociation involves the breakage of both H bonds. The high dynamic lability, expressed in the short lifetime, is again essential for fast code reading based upon a "trial and error" mechanism, where wrong combinations must disappear rapidly. Since all combinations of two bases will lead to more or less pronounced H-bond pairing, the number of wrong pairs formed and disrupted is large compared to the number of right combinations. The wrong pair, once formed, may not occupy the code site for too long a time. The stability constants must therefore remain relatively low. All these requirements of course do not favor a high accuracy of the molecular code reading process. The accuracy of such processes as they are observed in living organisms cannot be based upon the differential stability of the complementary base pairing alone. A code-checking mechanism presumably involving enzymic recognition sites is required to establish the small error rate observed in the biological processes of genetic information transfer and readout.

Although the base-pairing model systems presented here were studied in nonpolar media, many properties of the actual base-pairing systems in their biochemical environment are conserved. This follows also from kinetic studies with oligonucleotides carried out in aqueous media.[16] The orders of magnitude for the individual H-bond pairing reactions are in the same range. In aqueous media the lifetimes may be smaller because of the competitive H-bond formation with the solvent.

The above example shows that the dielectric relaxation technique described in this paper is very suitable for a study of the rates of base-pairing reactions and yields information which was not obtainable so far. The results of more extensive studies of this type will be published in forthcoming papers.

(16) M. Eigen, G. Maass, W. Müller, and D. Pörschke, to be published.

Counterions and Micelle Size. II. Light Scattering by Solutions of Cetylpyridinium Salts[1]

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Abstract: Dilute solutions of cetylpyridinium bromide in 0.200 *m* NaX (X = F, Cl, Br, ClO_3, BrO_3, IO_3, NO_3) were examined by light scattering. Counterion ability to promote aggregation was found to increase in the order: $IO_3^- < F^- < Cl^-, BrO_3^- \ll Br^- < NO_3^- < ClO_3^- \ll SCN^-, ClO_4^-, I^-$. Micelles associated with the first four anions are probably spheres, while those associated with Br^- are apparently rod-like. Micelles associated with NO_3^- and ClO_3^- are neither rigid rods nor spheres. Cetylpyridinium thiocyanate, perchlorate, and iodide are only slightly soluble in 0.200 *m* NaSCN, $NaClO_4$, and NaI, respectively. The aggregating power of an anion roughly parallels its ability to disrupt the structure of water.

Hartley and coworkers[2,3] concluded from diffusion, conductivity, and transport number measurements that any dependence of micelle size on counterion nature existing in aqueous solutions of cetylpyridinium salts tends to fade out when a large amount of simple salt is added. The results of some recent light-scattering measurements[4] on solutions of dodecyltrimethylammonium salts indicate that counterion specificity can persist at high supporting electrolyte concentrations. These conflicting implications, as well as our rather limited understanding of the counterion's role in micelle formation, prompted the present investigation. We herein report aggregation numbers—as determined by light scattering—of cetylpyridinium micelles present in aqueous solutions of a number of simple sodium salts.

Experimental Section

Most of the solutions examined were prepared by dissolving weighed samples of cetylpyridinium bromide (CPBr) in 0.200 *m* solutions of NaX (X = F, Cl, Br, NO_3, ClO_3, BrO_3, IO_3). All calculations were based on the assumptions that Br^- and X^- are complexed by the micelles in proportion to their respective stoichio-

(1) A preliminary report was made at the 17th Annual Northwest Regional Meeting of the American Chemical Society in Pullman, Wash., June 1962.

(2) G. S. Hartley and D. F. Runnicles, *Proc. Roy. Soc.* (London), **A168**, 420 (1938).

(3) C. S. Samis and G. S. Hartley, *Trans. Faraday Soc.*, **34**, 1288 (1938).

(4) E. W. Anacker and H. M. Ghose, *J. Phys. Chem.*, **67**, 1713 (1963).