Kinetics and Mechanism of the Alkaline Hydrolysis of Guanidine, Hexamethylguanidinium Perchlorate, and Tetramethylurea

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The kinetics of the alkaline hydrolysis of tetramethylurea and hexamethylguanidinium perchlorate show terms second order in hydroxide ion. The mechanism probably involves the hydroxide ion catalysed decomposition of a tetrahedral intermediate. The rate of the alkaline hydrolysis of guanidine reaches a plateau at alkali concentrations >1M. Mechanisms involving either attack of hydroxide ion on the guanidinium cation or unimolecular decomposition of the free base are consistent with the kinetic data but unimolecular decomposition of the guanidinium anion, as observed for some carbamic acid esters and a guanidine derivative, is not. The attack of hydroxide ion on the guanidinium cation is thought unlikely as it cannot explain the 750-fold increase in rate over the hexamethyl derivative. This rate difference can be accounted for if hydrolysis proceeds through the free base as this route is inaccessible to the hexamethyl derivative. A unimolecular decomposition of guanidine free base would result in the production of cyanamide but this could not be detected, consequently a mechanism involving the attack of water on the free base is preferred.

THE hydrolysis of carbamic acid derivatives has not been as extensively studied as that of their carboxylic acid counterparts although it exhibits some novel features. It has been demonstrated [1] that *p*-nitrophenylcarbamates with at least one proton on nitrogen are very much more susceptible to alkaline hydrolysis than *p*-nitrophenyl esters of carboxylic acids through the operation of an elimination mechanism of which the first steps are shown in equation (1). A related

$$R^1NHCO_2R^2 \overset{OH^-}{\rightleftharpoons} R^1\bar{N}CO_2R^2 \xrightarrow{slow} R^1NCO + {}^-OR^2 \quad (1)$$

mechanism, but with an intramolecular proton transfer has been proposed for the uncatalysed term in the hydrolysis of urea.[2] However, little work has been reported on the hydrolysis of guanidines for which such a mechanism may also operate.

The early work on the nature of the products produced by the action of alkali on guanidines was reviewed by Bell [3] who concluded that the initial products were urea and ammonia with traces of melamine detected in some experiments. Kinetic studies of the alkaline hydrolysis of simple guanidines have been reported by Eloranta,[4] who concluded that no urea was produced, and Warner [5] who showed that the alkaline hydrolysis of arginine proceeded at a pH independent rate above the pK_a of the guanidinium group. Warner favoured a mechanism in which hydrolysis proceeded through a pH independent reaction of guanidine rather than attack by hydroxide ion on the guanidinium ion. The present investigation attempts to establish which ionic form of guanidine is the reactive species and the subsequent mechanism of hydrolysis in alkaline solutions.

EXPERIMENTAL

Guanidinium chloride (Biochemical grade) and tetramethylurea were obtained from B.D.H. Hexamethylguanidinium perchlorate was synthesised from tetramethylurea, oxalyl chloride, and dimethylamine according to the published method; [6] it had $\lambda_{max.}$ 223 nm (ε 17 100 in water).

Kinetic runs were contained in sealed glass ampoules or tightly capped polypropylene bottles. The extent of hydrolysis of tetramethylurea and hexamethylguanidinium perchlorate was determined spectrophotometrically, after neutralisation with perchloric acid, as described in the Results section. Guanidine was estimated colorimetrically with the diacetylnaphthol reagent.[7] Rate constants were determined from first-order plots; standard deviations are included in Tables 1—4. Absorbance measurements were made on a Cary 14 spectrophotometer.

Chromatograms of the guanidine reaction mixtures were run on plates of binder free cellulose with methanol–3N-ammonia (60 : 75 v/v) as solvent,[8] ammoniacal silver nitrate in acetone being used for detection. Cyanamide and urea, R_F *ca.* 0.8, could not be resolved under our conditions but chromatography of part hydrolysed guanidine showed the absence of melamine, R_F 0.32. Cyanamide was determined colorimetrically through its reaction with the pentacyanoammineferrate(II) ion.[9]

RESULTS

The hydrolysis of tetramethylurea at 60° in sodium hydroxide (0.42—2.1M; μ 3.0) was followed by measuring the disappearance of the substrate at 210 nm. The low absorbance of the final products after acidification was consistent with these being carbon dioxide and dimethylamine. A first-order plot is shown in the Figure and the rate constants are given in Table 1. The apparent order in

TABLE 1

Observed first-order rate constants for the hydrolysis of tetramethylurea (4.17×10^{-4}M) at 60° and μ 3.0

$[OH^-]$/M	$10^7 k_{obs}$/s^{-1}
2.08	42 (2.0) [a]
1.67	30 (0.7)
1.46	21 (1.0)
1.04	11 (0.8)
0.42	3.0 (0.1)

[a] Standard deviation.

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hydroxide ion concentration is 1.8 and consequently the data were fitted to equation (2). The rate constants

$$k_{obs} = k_0 + k_1[OH^-] + k_2[OH^-]^2 \quad (2)$$

obtained are listed in Table 2 where estimates of uncertainty obtained from the limits of the slopes and intercepts of the plots involved are also given.

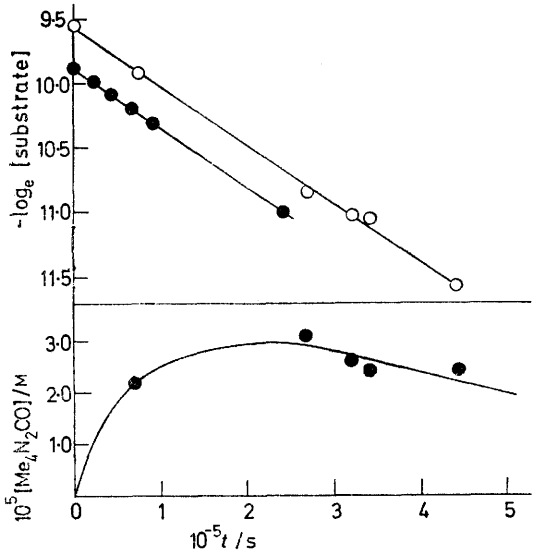
TABLE 2

Calculated rate constants for alkaline hydrolysis at 60° and μ 3.0

Substrate	10^8k_0/s^{-1}	10^8k_1/l mol^{-1} s^{-1}	10^7k_2/ l^2 mol^{-2} s^{-1}
Hexamethylguanidinium	0 ± 1	20 ± 4	7 ± 0.5
Tetramethylurea	5 ± 2	10 ± 2	9.5 ± 1
Urea [a]	20.9 ± 1.5	43 ± 3	4.14 ± 0.2
Guanidinium		$k' = 1.5 \times 0.1 \times 10^{-4}$ l mol^{-1} s^{-1}	
Guanidine		$k'' = 5.2 \pm 0.4 \times 10^{-5}$ s^{-1}	

[a] Data from ref. 13.

The spectra of partially hydrolysed samples of hexamethylguanidinium perchlorate, after neutralisation with perchloric acid revealed the presence of a contribution



Upper part: first-order plots for the disappearance of hexamethylguanidinium perchlorate (○) and tetramethylurea (●) in 2.1M-NaOH, μ = 3.0 ($NaClO_4$), 60°. Tetramethylurea concentrations were determined from the absorbance at 210 nm and hexamethylguanidinium perchlorate concentrations from equation (4). Lower part: tetramethylurea concentrations (●) during the above hydrolysis of hexamethylguanidinium perchlorate determined from equation (5). The line is the theoretical curve for the time dependence of the tetramethylurea concentration for the reaction scheme of equation (3) with rate constants taken from Tables 1 and 3

from a product having an absorption maximum at 210 nm characteristic of tetramethylurea, thus suggesting that this was one of the initial products in accordance with equation (3). As in the case of tetramethylurea hydrolysis the final absorbance was negligible. The concentrations of

$$C(NMe_2)_3^+ + OH^- \longrightarrow \underset{+\ HNMe_3}{Me_2NCONMe_2} \xrightarrow{H_2O} \underset{+\ CO_2}{2HNMe_2} \quad (3)$$

hexamethylguanidinium ion and tetramethylurea during the kinetic runs were obtained from the absorbance at 220 and 230 nm (A_{220} and A_{230}) by using equations (4) and (5) respectively. In these equations the molar extinction

$$[\text{Hexamethylguanidinium ion}] = \frac{5\,625\,A_{230} - 1\,836\,A_{220}}{14\,560 \times 5\,625 - 17\,045 \times 1\,836} \quad (4)$$

$$[\text{Tetramethylurea}] = \frac{14\,560\,A_{220} - 17\,045\,A_{230}}{14\,560 \times 5\,625 - 17\,045 \times 1\,836} \quad (5)$$

coefficients at 220 nm are 17 045 and 5 625 and at 230 nm are 14 560 and 1 836 for the hexamethylguanidinium ion and tetramethylurea respectively. The concentrations of hexamethylguanidinium ion obtained from equation (4) gave good first-order plots; an example is shown in the Figure where the tetramethylurea concentrations during the same run, determined from equation (5), are also shown. These are compared in the Figure with the tetramethylurea concentrations calculated from the theory of consecutive reactions and the rate constants in Tables 1 and 3. The agreement is quite good supporting the pathway of equation (3) for the hydrolysis of the hexamethylguanidinium ion. The values of the rate constants for the disappearance of hexamethylguanidinium ion given in Table 3 are close to

TABLE 3

Observed first-order rate constants for the hydrolysis of hexamethylguanidinium perchlorate (2.11 × 10^{-4}M) at 60° and μ 3.0

[OH$^-$]/M	10^7k_{obs}/s^{-1}
2.1	44 (3) [a]
1.75	23 (0.7)
1.25	14.3 (0.8)
1.00	8.8 (0.3)
0.50	3.0 (0.1)

[a] Standard deviation.

those for tetramethylurea and show a similar dependence on hydroxide ion concentration. Consequently they were fitted to equation (2) and the resultant rate constants are collected in Table 2.

The hydrolysis of guanidine was followed by estimating its concentration colorimetrically,[7] the first-order rate constants for its disappearance in sodium hydroxide solutions (0.01—2.91M, μ 3.0) at 60° (Table 4) reaching a

TABLE 4

Observed first-order rate constants for the hydrolysis of guanidine (5.0 × 10^{-4}M) at 60° and μ 3.0

[OH$^-$]/M	10^5k_{obs}/s^{-1}	[OH$^-$]/M	10^5k_{obs}/s^{-1}
2.91	4.38 (0.13) [a]	0.19	1.84 (0.02)
1.94	4.26 (0.08)	0.15	1.64 (0.03)
0.97	4.03 (0.07)	0.10	1.20 (0.07)
0.58	3.30 (0.13)	0.05	0.70 (0.02)
0.39	2.90 (0.07)	0.01	0.19 (0.01)
0.29	2.38 (0.07)		

[a] Standard deviation.

limiting value in the highest alkali concentrations. Rate profiles of this type, where substrate ionisation with dissociation constant K_a is suspected, can be fitted to equation (6) where k is a composite constant discussed

$$k_{obs} = k[OH^-]/(K_w + K_a[OH^-]) \quad (6)$$

below and K_w is the ionic product of water. A theoretical curve constructed with k 1.5 × 10^{-19} mol l^{-1} s^{-1} and pK_a

12.53 fitted the data well. An Arrhenius plot over the range 15—78° in 0.97M-NaOH, $\mu = 3.0$ yielded $E_a = 20.4$ kcal mol^{-1}.

Chromatographic investigation of partly hydrolysed samples of guanidine gave no evidence of any product except urea, melamine not being detected. Quantitative colorimetric analysis of reaction mixtures (2.6×10^{-3}M-guanidine; 1.0M-NaOH; 60°) for cyanamide revealed a trace ($< 5 \times 10^{-5}$M) at the commencement of the reaction which diminished as the reaction proceeded.

DISCUSSION

The presence of a second-order term in hydroxide ion in the alkaline hydrolysis of tetramethylurea and hexamethylguanidinium perchlorate strongly suggests that the rate-determining step under these conditions is the hydroxide ion catalysed breakdown of a tetrahedral intermediate formed by the addition of hydroxide ion to the substrate in a fast equilibrium.[10] Such a mechanism is known to operate for the alkaline hydrolysis of anilides [11] and amidines [12] and with these substrates a change in the rate-determining step, from the breakdown of the tetrahedral intermediate to its formation, has been inferred from a reduction in the order in hydroxide ion at high concentrations of the latter, but this could not be observed here. Lynn [13] has proposed a similar mechanism for the alkaline hydrolysis of urea. A comparison of the rate constants for the two ureas (Table 2) shows that the second-order term in hydroxide ion is relatively more important than the first-order term in the case of tetramethylurea reflecting the greater importance of hydroxide ion catalysis in the compound with the poorer leaving group. The three compounds urea, tetramethylurea, and hexamethylguanidinium perchlorate, are hydrolysed at similar rates under the conditions reported here. Any electrostatic enhancement of the reaction of the hexamethylguanidinium cation with hydroxide ion is more than counterbalanced by the inherent stability of this symmetrical, resonance stabilised cation.

Four mechanisms will be considered for the alkaline hydrolysis of guanidine, one involving the anion, one the cation, and two the free base of guanidine. First consider a slow decomposition of the guanidine anion to the cyanamide anion and ammonia [equation (7)] which is equivalent to equation (1) for carbamate

$$(NH_2)_3C^+ \overset{K_a}{\rightleftharpoons} HN{:}C(NH_2)_2 \overset{K_{a_2}}{\rightleftharpoons} + H^+$$

$$[(HN)_2{:}CNH_2]^- \overset{\text{slow}}{\longrightarrow} [HNC{:}N]^- + NH_3 \quad (7)$$
$$+ H^+$$

ester hydrolysis. This mechanism could account for the lower rate of hydrolysis of hexamethylguanidinium ion where such ionisations would be impossible; however, both the pH dependence and products observed preclude this mechanism. If the mechanism of equation (7) was followed the plateau in the pH–rate profile would be associated with the second ionisation K_{a2} but the data show that the observed pK_a, 12.53 at 60°, is in accord with the first ionisation of guanidinium ion (pK_a 13.6 at 25°) when allowance is made for the temperature difference [14] and much too low for the second pK_a. In addition no cyanamide was produced during the hydrolysis. This is discussed further below. It is interesting that an elimination mechanism leading to a cyanamide has recently been shown to operate for the alkaline hydrolysis of the much more acidic guanidine, 1-(*N*-benzoylamidino)-3,5-dimethylpyrazole.[15] In that case the leaving group was the 3,5-dimethylpyrazole anion (pK_a of conjugate acid 15) which is a much better leaving group than the amide anion would be if guanidine hydrolysis followed this mechanism.

The other three mechanisms are kinetically equivalent and have neutral transition states in agreement with the kinetic data. They involve either (i) the guanidinium cation in an attack by hydroxide ion [equation (8)] as

$$C(NH_3)_3^+ + OH^- \overset{k'}{\longrightarrow} \text{products} \quad (8)$$

has been proposed for the first step for the hexamethyl derivative, in this case k [equation (6)] $= k'K_w$; or (ii) the free base in a unimolecular decomposition [equation (9a)] or in an attack by water [equation (9b)].

$$HN{=}C(NH_2)_2 \overset{k''}{\longrightarrow} HN{=}C{=}NH \longleftrightarrow H_2N{-}C{\equiv}N \overset{H_2O}{\longrightarrow} H_2CONH_2 \quad (9a)$$
$$+ NH_3$$

$$HN{=}C(NH_2)_2 + H_2O \overset{k''}{\longrightarrow} H_2NCONH_2 + NH_3 \quad (9b)$$

In these cases k [equation (6)] $= k''K_a$. The values of k' and k'' are given in Table 2. It was in an attempt to distinguish between mechanisms involving the cation in equation (8) and the free base in (9) that the hydrolysis of hexamethylguanidinium ion, which cannot readily lose a proton, was examined. The comparison is complicated by the importance of the second-order hydroxide ion term in the hexamethylguanidinium ion hydrolysis which was not detected for guanidine, but if first-order processes in hydroxide ion are compared (k_1 and k', Table 2) the guanidinium ion reacts 750 times faster than its hexamethyl derivative. Such a difference is difficult to accommodate in mechanism (8), methyl groups having a very small influence on the rates of alkaline hydrolysis of urea and tetramethylurea. On the other hand the mechanisms involving the free base could readily accommodate this difference in rates as these mechanisms are not available to the hexamethylguanidinium cation. A distinction between (9a) and (9b) can be made by examination of the reaction products. Reaction (9a) involving an intramolecular proton

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transfer of the type postulated for urea [2] and recently discussed by Williams and Jencks [16] would be expected to lead to carbodi-imide which would tautomerise rapidly to cyanamide [17] before hydrolysis to urea. In reaction (9b) the attack of water on guanidine to give a tetrahedral intermediate would be followed by proton transfers to yield urea and ammonia and no cyanamide would be formed.

The rate of hydrolysis of cyanamide to urea becomes pH independent above 0.5M-NaOH with a rate constant of 1.49×10^{-5} s^{-1} at 60°.[18] At this temperature the rate constant for guanidine hydrolysis increases from 3.1×10^{-5} to 5.2×10^{-5} s^{-1} as the alkali concentration increases above 0.5M. Thus cyanamide is hydrolysed less rapidly than guanidine and its concentration would increase during the initial stages of the reaction if the mechanism of equation (9a) was followed. Calculations using these rate constants and the equations for consecutive reactions predicted that in 1M-alkali at 60° the cyanamide concentration should have reached a maximum of 56% of the initial guanidine concentration after 11 h. In a reaction under these conditions very little cyanamide was detected (<2%) and this decreased during the reaction. Additional evidence that the formation of urea from the hydrolysis of guanidines does not proceed through cyanamide can be obtained from the work of Warner [5] who studied the alkaline hydrolysis of arginine by following the rates of urea and ammonia production. His data are very similar to ours for guanidine. In 1M-alkali at 60° (interpolated) the rate constant for the production of urea was 3.6×10^{-5} s^{-1} which again is greater than the rate constant for the production of urea from cyanamide. These observations rule out a unimolecular process as the slow step in the decomposition of guanidine unless the hydration of carbodi-imide to urea competes favourably with the rate of its tautomerisation to cyanamide.

We agree with Warner [5] that the alkaline hydrolysis of guanidine can best be represented as a pH independent reaction of the free base because of the evidence presented here on the effect of substitution on the rate. In the absence of cyanamide as a product it appears likely that the reaction proceeds by attack of water on the guanidine free base [mechanism (9b)] and not through a zwitterionic intermediate as proposed for urea [16] nor *via* a guanidine anion as has been observed for a substituted guanidine.[15]

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