Table II. Oxygen Evolution during the Decomposition of Potassium Bromate in 68% Sulfuric Acid Solution

run	gas evolved,[a,b] mL	atmospheric pressure, KpA	mmol of gas
1	297	96.4	11.7
2	302	98.2	12.1
3	296	98.2	11.9
4	297	99.1	12.1
5	297	98.8	12.0
			av 12.0

[a] Gas obtained by displacement of a volume of mineral oil. [b] 0.01 mol of $KBrO_3$ dissolved in 7 mL of H_2O and reacted with 8 mL of concentrated H_2SO_4 at 0 °C.

Table III. Identification of Products of Decomposition of $KBrO_3$ in 68% Sulfuric Acid

run[a]	Br_2,[b] mmol	HOBr,[c] mmol	BrO_3^-,[d] mmol
1	2.35		
2	2.55		
3		4.45	0.38
4		4.25	0.45
5		4.15	0.48
av	2.45	4.28	0.44

[a] Reaction of 0.01 mol of $KBrO_3$ with 68% sulfuric acid solution at 0 °C. [b] Treatment of CCl_4 extract with excess NaI followed by titration with 0.1 N $Na_2S_2O_3$. [c] Treatment of aqueous phase as in *b* after CCl_4 extraction and after treatment with 0.1 N phenol. [d] Difference before and after 0.1 N phenol treatment calculated as BrO_3^-.

1.2 g of acetophenone (10 mmol). To this was added 1.67 g of potassium bromate (10 mmol) slowly in portions. The reaction mixture became warm and, after addition was complete, was left stirring at room temperature overnight. A total of 1.4 g of product was obtained, 70% yield. Analysis by GC on a 2-ft UCW 952 column at 100 °C indicated that the mixture consisted of *o*- and *m*-bromoacetophenone in a ratio of 1:6 (GC comparison with authentic sample).

Preparation of Bromobenzene. To a solution of 3.33 g of concentrated sulfuric acid and 6.66 g of water was added 0.78 g of benzene. Then 1.67 g of potassium bromate was added in a single portion. The temperature rose slightly from 26 to 30 °C and the mixture was stirred overnight. The product was then diluted with water (50 mL) and extracted with ethyl ether. Analysis on a 2-ft UCW 982 column indicated that bromobenzene in 97% yield was produced (GC comparison with authentic sample).

Reaction of Potassium Bromate with Sulfuric Acid. To 1.67 g of potassium bromate (0.01 mol) in 7 mL of water at 0 °C under nitrogen was added concentrated sulfuric acid dropwise via syringe. After about 5 mL had been added, gas evolution commenced and decomposition of the potassium bromate occurred. A total of 8 mL of H_2SO_4 was added. After the mixture was stirred for 1 h, the total gas volume was measured (Table II) and the gas was anlyzed by mass spectrometry. A total of 12 mmol of oxygen gas was produced which was 98% pure by mass spectrometry. To the remaining solution was then added water (50 mL) and the solution was extracted with carbon tetrachloride (3 × 20 mL) to remove the bromine which produced (Table II). The resulting aqueous solution was then diluted to 100 mL with water and was analyzed for hypobromous acid by treatment with phenol followed by titration.[16] These results are reported in Table III.

A similar experiment was carried out with nitrobenzene present (0.01 mol). The evolved gas (0.0056 mol) was analyzed by mass spectrometry and found to consist of 82% carbon dioxide and 18% oxygen. Identification of the CO_2 was confirmed by reaction with aqueous barium hydroxide solution to form the insoluble barium carbonate.

Acknowledgment. We gratefully acknowledge the technical expertise of Ted Tabacchi.

Registry No. Nitrobenzene, 98-95-3; benzoic acid, 65-85-0; phthalic acid, 88-99-3; acetophenone, 98-86-2; benzene, 71-43-2; *m*-bromonitrobenzene, 585-79-5; 3-bromobenzoic acid, 585-76-2; 4-bromophthalic acid, 6968-28-1; *o*-bromoacetophenone, 2142-69-0; *m*-bromoacetophenone, 2142-63-4; bromobenzene, 108-86-1; potassium bromate, 7758-01-2.

(16) E. Billmann and E. Rimbert, *Bull. Soc. Chim. Fr.*, **33**, 1465, (1923).[17]

(17) This method is not totally precise due to the slow reaction of iodine with phenol.[18]

(18) T. Anderson and H. E. L. Madsen, *Ana. Chem.*, **37** 49 (1965).

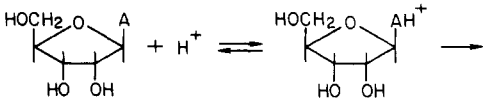
Effect of the Structure of the Glycon on the Acid-Catalyzed Hydrolysis of Adenine Nucleosides

J. Lyndal York

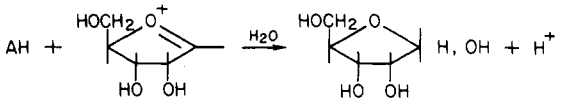
Department of Biochemistry, University of Arkansas for Medical Sciences, Little Rock, Arkansas 72205

Received November 14, 1980

Recent studies[1-6] of the acid-catalyzed hydrolysis of the purine nucleosides support an A1 mechanism, in which the



A = adenine



protonated nucleoside dissociates in the rate-controlling step to a glycosyl carbonium ion and free purine. The glycosyl carbonium ion then extracts a hydroxyl from water to form the sugar. The alternative bimolecular mechanism,[7,9] involving the intermediate formation of a Schiff base, does not have strong experimental support.

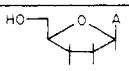
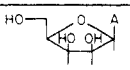
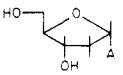
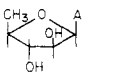
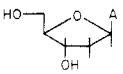
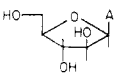
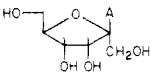
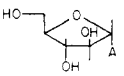
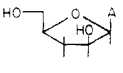
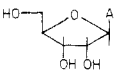
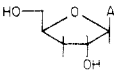
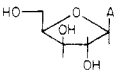
The effect of changes in the nucleoside base on the rates of acid-catalyzed hydrolysis has been thoroughly investigated, but a systematic study of the effect of the sugar structure on the lability of the glycosyl–purine bond has not been reported. The acid solvolyses of the β-arabinoside, riboside, and xyloside of adenine have been reported[4] as has that of the 2-deoxyriboside, the dideoxyriboside, and the psicofuranoside.[10] In this paper we report a comprehensive evaluation of the effect of structural changes in the glycon on the rates of hydrolysis of adenosine nucleosides.

In agreement with the reports of others,[4,11] removal of

(1) J. A. Zoltewicz, D. F. Clark, T. W. Sharpless, and G. Grahe, *J. Am. Chem. Soc.*, **92**, 1741 (1970).
(2) J. A. Zoltewicz and D. F. Clark, *J. Org. Chem.*, **37**, 1193 (1972).
(3) R. D. Panzica, R. J. Rosseau, R. K. Robins, and L. B. Townsend, *J. Am. Chem. Soc.*, **94**, 4708 (1972).
(4) E. R. Garrett and P. J. Mehta, *J. Am. Chem. Soc.*, **94**, 8532 (1972).
(5) L. Hevesi, E. Wolfson-Davidson, J. B. Nagy, O. B. Nagy, and A. Bruylants, *J. Am. Chem. Soc.*, **94**, 4715 (1972).
(6) R. Romero, R. Stein, H. G. Bull, and E. H. Cordes, *J. Am. Chem. Soc.*, **100**, 7620 (1978).
(7) G. W. Kenner in "The Chemistry and Biology of Purines", G. E. W. Wolstenholme and C. M. O'Connor, Eds., Little, Brown and Co., Boston, MA, 1957, p 312.
(8) C. A. Dekker, *Annu. Rev. Biochem.*, **29**, 453 (1960).
(9) F. Micheel and A. Heesing, *Chem. Ber.*, **94**, 1814 (1961).
(10) E. R. Garrett, *J. Am. Chem. Soc.*, **82**, 827 (1960).
(11) H. Venner, *Z. Physiol. Chem.*, **339**, 14 (1964).

0022-3263/81/1946-2171$01.25/0

Table I. Apparent Second-Order Rate Constants for the Acid-Catalyzed Solvolysis of Adenine Nucleosides[a]

compd	configuration, C-1′-C-3′	k_2, L/mol/min	compd	configuration, C-1′-C-3′	k_2, L/mol/min
1		35	6		9.6×10^{-3}
α-2		1.37	7		7.7×10^{-3}
β-2		0.96	β-8		1.9×10^{-3}
3		0.53	α-8		1.6×10^{-3}
β-4		0.057	9		0.82×10^{-3}
β-5		0.012	10		0.29×10^{-3}
			11		0.046

[a] All compounds were pentose furanosides of adenine except 11 which was a ribopyranoside. The reaction temperature was 40.0 °C and the ionic strength 0.89. $k_2 = k_1/[HCl]$, calculated from the slopes of the plot of k_1 vs. [HCl] at 40.0 °C. A = adenine.

the 2′- or 3′-hydroxyl enhances the rate of hydrolysis. In the β anomer, removal of the 3′-hydroxyl increases the rate 15 times (**9** vs. β-**5**, Table I) while removal of the 2′-hydroxyl increases the rate 1200 times (**9** vs. β-**2**). Removal of both 2′- and 3′-hydroxyls increases the rate 4×10^4 times (**9** vs. **1**). The hydroxyl is thought to decrease the tendency for C–N bond cleavage and formation of the C-1′ carbonium ion by virtue of a –I effect,[1,12] therefore, removal of the hydroxyl group increases the rate of hydrolysis. The 5′-OH also has a stabilizing influence on the rate of hydrolysis through a –I effect via the ring oxygen on C-1′ carbonium ion formation. Removal of the 5′-OH increases the rate 4-fold (**7** vs. β-**8**).

The data suggest the presence of a reverse anomeric effect.[13] In the β anomers the adenine lies in a quasi-equatorial position and when protonated will develop a dipole opposed to the dipole of the ring oxygen. This effect could be expected to result in an increased stability of the β relative to the α anomer. This expectation is supported by a significant decrease in the rate of hydrolysis of the β-xyloside (**10**) when compared to that of the α-arabinoside (α-**8**). In both these compounds, the 2′-OH is trans and the 3′-OH is cis to the adenine; consequently, the only variable in structure relative to these two compounds is the configuration at the anomeric carbon.

The glycosyl–purine bond is labilized in those compounds in which the adenine and the hydroxyl on C-2′ are cis. This can be seen by a comparison of the relative rates of hydrolysis of compounds β-**4** vs. β-**5** and β-**8** vs. α-**8** (Table I). The lability is enhanced further if the C-3′ hydroxyl is also cis to the C-2′ hydroxyl and to the adenine; i.e., a 12-fold increase in hydrolysis rate is observed in **6** relative to **9**. The steric strain of the cis configuration accounts for the increased hydrolysis rate. Garrett[4] has predicted that the lyxoside (**6**) would be the most labile pentoside. Apparently 2′,3′-*cis*-dihydroxyls lead to some steric strain which is relieved in the trans arrangement of the 2′,3′-dihydroxyls (**9** vs. **10**). A similar effect is also observed in the α series of methyl aldofuranosides[14] where compounds having cis substituents at C-1′ and C-2′ are hydrolyzed at a greater rate.

Hydrogen bonding of the sugar hydroxyls with the adenine ring nitrogens has been considered[15] to be pertinent to the question of the effects of sugar structure on hydrolysis rates. This consideration now appears to be unimportant since most nucleosides.[16,17] are in the anti conformation where hydrogen bonding of sugar and purine ring is impossible.

A 650-fold increase in rate is observed in **3** relative to **9**. In **3** carbonium ion formation would be favored because of the relative increased stability of a tertiary carbonium ion compared to a secondary carbonium ion. These results support the A1 mechanism where formation of the glycosyl carbonium ion is rate limiting.

The rate of hydrolysis of 2′-deoxyadenosine in 99% D_2O and 0.11 M DCl was increased 2.34-fold for the β and 2.04-fold for the α anomer over the rate in H_2O. This reverse solvent isotope effect strongly supports the A1 mechanism of nucleoside hydrolysis. A reverse isotope effect also has been observed in the glycosides.[18] In the A1 mechanism, the rate depends upon the concentration of protonated nucleoside and since deuteroacids[19] are weaker acids than protoacids, a higher concentration of the nucleoside dication will exist in D_2O, resulting in a greater rate of hydrolysis. It has been shown that in 0.1 M acid hydrolysis occurs via the dication of adenosine.[4,6] In the A2 mechanism[7] participation of water in the transition state would be expected to result in a decrease in k_D/k_H rather than an increase as observed. The A2 mechanism is also ruled out on the basis of the entropy of activation, $\Delta S^{\pm}$, which was calculated[20] from the Ar-

(12) A. Streitweiser, Jr., "Solvolytic Displacement Reactions", Wiley, New York, 1962, pp 112–113.
(13) R. U. Lemieux and A. R. Morgan, *Can. J. Chem.*, **43**, 2205 (1965).
(14) I. Augestad and E. Berner, *Acta Chem. Scand.*, **10**, 911 (1956).
(15) E. R. Garrett, J. K. Seydel, and A. J. Sharpen, *J. Org. Chem.*, **31**, 2219 (1966).
(16) I. Ekiel, M. Remin, E. Darzynkiewicz, and D. Shugar, *Biochim. Biophys. Acta*, **562**, 177 (1979).
(17) A. E. V. Haschemeyer and A. Rich, *J. Mol. Biol.*, **27**, 369 (1967).
(18) W. G. Overend, C. W. Rees, and J. S. Sequeira, *J. Chem. Soc.*, 3429, 1962.
(19) C. A. Bunton and V. J. Shiner, Jr., *J. Am. Chem. Soc.*, **83**, 42 (1961).

rhenius activation energy. E_a and $\Delta S^{\pm}$ were 22 kcal mol^{-1} and +1.2 eu for 9-β-D-2′-deoxyribofuranosyladenine and 25 kcal mol^{-1} and +4.4 eu for 9-α-D-2′-deoxyribopyranosyladenine. Bimolecular reactions are characterized by $\Delta S^{\pm}$ values about 20 eu more negative than observed for unimolecular mechanisms.[18,21] Small positive values of $\Delta S^{\pm}$ have been reported for pyrimidine[22] and purine[1] nucleoside as well as for glycopyranoside[18] hydrolysis. Our data are consistent with these observations.

In this, the first systematic study of the effect of sugar structure on the acid-catalyzed hydrolysis of adenine nucleosides, new data are presented which suggest that the variations in hydrolysis rates observed in a homologous series of adenine furanosides can be explained on the basis of the degree of steric interaction between the adenine and the 2′- and/or 3′-hydroxyls. In contrast, the increase in hydrolysis rate of the 2′-, 3′-, or 5′-deoxyfuranoside can be attributed to removal of the electron-withdrawing inductive effect of the hydroxyls. Also identified is a reverse anomeric effect which has not been reported previously for the nucleosides. The reverse solvent isotope effect and the entropies of activation are consistent with the unimolecular mechanism for nucleoside hydrolysis.

Experimental Section

The nucleosides used in this study have been previously identified as to their source and characterization.[23,24] **11** was a gift from Dr. L. B. Townsend of the University of Utah.

The rate of hydrolysis of the adenine nucleosides was followed by using the differential spectrometric method described by Garrett.[4] The rate of change in absorbance at 255 nm was recorded. Pseudo-first-order rate constants (k_1') were determined at HCl concentrations of 0.11, 0.32, 0.62, and 0.89 M at 40 °C. Second-order rate constants, k_2, were determined from the slopes of the plots of k_1' vs. HCl concentration. The rate constants increased with increasing ionic strength; consequently, the ionic strength was maintained at 0.89 by the addition of appropriate amounts of KCl. The temperature dependence of k_1' was determined for **β-2** and **11** in 0.11 M HCl and an ionic strength of 0.89 at temperatures of 25, 40, 49, and 60 °C. The Arrhenius activation energy, E_a, was calculated from the slope of plots of log k_1' vs. $1/T$.

Registry No. **1**, 4097-22-7; **α-2**, 3413-66-9; **β-2**, 958-09-8; **3**, 1874-54-0; **β-4**, 6998-75-0; **β-5**, 73-03-0; **6**, 4005-33-8; **7**, 4152-76-5; **α-8**, 3228-71-5; **β-8**, 5536-17-4; **9**, 58-61-7; **10**, 524-69-6; **11**, 17434-50-3.

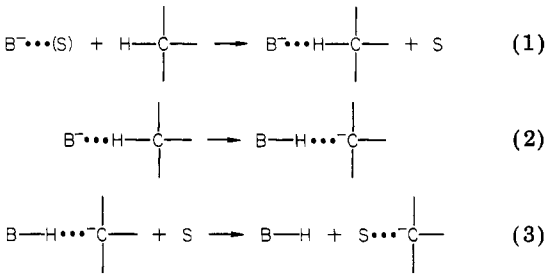
(20) K. J. Laidler, "Chemical Kinetics", 2nd ed., McGraw Hill, New York, 1965, p 89.
(21) F. A. Long and M. A. Paul, *Chem. Rev.*, **57**, 935 (1957).
(22) R. Shapiro and M. Danzig, *Biochemistry*, **11**, 23 (1972).
(23) G. A. LePage and I. G. Junga, *Cancer Res.*, **25**, 46 (1965).
(24) J. L. York and G. A. LePage, *Can. J. Biochem.*, **44**, 331 (1966).

Protonic Charge Densities in Kinetic Acidities of Carbon Acids

Carl D. Slater[1]

Department of Chemistry, Memphis State University, Memphis, Tennessee 38152, and Department of Physical Sciences, Northern Kentucky University, Highland Heights, Kentucky 41076

The study of carbon acids has provided fertile ground for the development of chemical theory because proton transfer is one of the simplest chemical reactions for which both rates and equilibrium positions can be measured.[2] In particular, the acidities of nitroalkanes, carbonyl compounds, and fluoroalkanes have been examined intensively. Nitroalkanes have attracted interest because they give anomalous rate–equilibrium correlations based on the Brønsted relationship.[3] Modified Marcus theory[4] has been utilized to interpret this behavior and Agmon has recently argued that under certain conditions kinetic acidities depend largely on the work term in Marcus theory.[5] In this view proton transfer is considered to occur via a three-stage mechanism as outlined in eq 1–3. Step 1 involves the



formation of a contact complex in which a solvent molecule in the solvation shell of the base is replaced by the carbon acid. The free-energy change in this stage is referred to as the work term, denoted W_R. It receives its principal contributions from solvent reorganization, steric effects in the reagents, and the difference in hydrogen-bonding strengths in the interactions shown. The second stage of the reaction is the actual proton transfer of eq 2 for which the activation energy is G_a. The specific rate of the forward process is thus given by eq 4, in which Z is the frequency factor.

$$k = Z\exp[-(W_R + G_a)/RT] \quad (4)$$

For reactions involving a series of structurally similar carbon acids with a common base in the same solvent, steric interactions and solvent reorganization should play a nearly constant role. Accordingly, differences in the strength of hydrogen bonding between the base and the carbon acids should be the predominant variable, and Agmon suggests that in these instances the W_R and G_a terms may vary systematically, leading to abnormal Brønsted α values.

Hydrogen bonding has been the subject of numerous theoretical treatments, and Del Bene has concluded that for hydrogen-bonded species having water or substituted water molecules as proton donors the primary factor responsible for stabilizing the complex is the electrostatic interaction between the proton and the lone pair of electrons from the acceptor.[6] If this finding applies generally to other donor–acceptor combinations, a measure of the abilities of carbon acids to hydrogen bond to a negatively charged base can be developed by considering the acids as charged particles bearing the same charge as the interacting protons. If specific solvent interaction is neglected, the free-energy change associated with bringing two particles of charge Z_A and Z_B from infinite separation to interaction distance d_{AB} in a medium of dielectric constant ϵ is given by eq 5, in which e is the electronic charge.[7]

$$\Delta G_{el} = Z_A Z_B e^2/\epsilon d_{AB} \quad (5)$$

(1) Correspondence should be sent to the second address.
(2) An excellent review of this work is given in: Reutov, O. A.; Beletskaya, I. P.; Butin, K. P. "C-H Acids"; Pergamon Press: Oxford, England, 1978.
(3) (a) Bordwell, F. G.; Boyle, W. J., Jr.; Hautala, J. A.; Yee, K. C. *J. Am. Chem. Soc.* **1969**, *91*, 4002. (b) Bordwell, F. G.; Boyle, W. J., Jr.; Yee, K. C. *Ibid.* **1970**, *92*, 5926.
(4) Marcus, R. A. *J. Am. Chem. Soc.* **1969**, *91*, 7224.
(5) Agmon, N. *J. Am. Chem. Soc.* **1980**, *102*, 2164.
(6) Del Bene, J. *J. Am. Chem. Soc.* **1973**, *95*, 5460.
(7) Frost, A. A.; Pearson, R. G. "Kinetics and Mechanism"; John Wiley and Sons, Inc.: New York, 1953; p 133.

0022-3263/81/1946-2173$01.25/0