catalyzed iodination of pyrazole[2a] is given as $k_0 = 1.5 \times 10^{-13}$ l.^{2} mole^{-2} sec^{-1}, corrected for first-order dependence upon the *stoichiometric* concentration of pyrazole, [Py], and for [H$^+$] and [I$^-$]^2. The ratio of the relative second-order rates of iodination of Py$^-$ and Py would therefore be

$$\frac{k_2^{Py^-}}{k_2^{Py}} = \frac{1.5 \times 10^{-13}/K_a}{(2 \times 10^{-12} \text{ to } 5 \times 10^{-9})} = 3 \times 10^9 \text{ to } 7 \times 10^{12} \quad (1)$$

where K_a is the acid dissociation constant of pyrazole. This result is meaningful if both Py$^-$ and Py undergo iodination by a common iodinating agent.

The assumption that both substrates undergo iodination by the same iodinating agent is not necessarily valid. The rate laws for the iodination of pyrazole and 1-alkylpyrazoles are compatible with iodination by either molecular I_2 or the hydrated iodinium ion IOH_2^+.[2a,8] The ratio of the molar concentrations of these possible iodinating agents in aqueous solution[15] is given by

$$\frac{[I_2]}{[IOH_2^+]} \sim [I^-] \times 10^{11}$$

It is just as reasonable to assume that anion substrates undergo attack by molecular iodine and molecular substrates by IOH_2^+ than to assume the substrates are attacked by *either* I_2 or IOH_2^+. In justification, we note that the probability of encounter between anion substrates and IOH_2^+ will be very small, whereas the probability of encounter between molecular substrates and IOH_2^+ will be comparatively large, since the substrate concentration here is the stoichiometric concentration. If the latter assumption were made, we must conclude that the ratio of reactivities would be even greater than the limits expressed by eq 1.

Theoretical Considerations. The great difference in the reactivities of the anion and molecular forms of pyrazole, and in the reactivities of aromatic heterocycles in general, provides an interesting and difficult theoretical problem. The complexity of the problem is illustrated by the following considerations. In phosphate-buffered iodinations of 1-alkylpyrazoles, both HPO_4^{2-} and $H_2PO_4^-$ were catalytically active (Table III). The ratio k_A/k_B is about 10 for 1-methylpyrazole and also for 1-isopropylpyrazole. In the iodination of aniline,[8] this ratio is about 100 and in the iodination of pyrazole[2] HPO_4^{2-} was catalytically active but $H_2PO_4^-$ was not detectably active. Clearly, the degree of participation of a base in the transition state of the proton-removal step is quite sensitive to the identities of the substrate and the base. Quantum theoretical interpretation of the relative magnitudes of rate constants should therefore utilize models of the transition states that include the base explicitly. This argument applies also to the "uncatalyzed" rate as well, since water presumably is the catalyst in this case. Brønsted correlations between catalytic rates and base strengths would be germain to this problem, since the sensitivity of the rate to the base strength can be interpreted as a measure of the degree of proton removal in the transition state.[16] We are presently making base catalytic studies of heterocyclic iodinations, which we plan to report later, together with theoretical interpretation of the results.

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Kinetics of Nucleophilic Substitution on 6-Chloropurine Ribonucleoside in Aqueous Solution[1]

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Abstract: Nitrogen, sulfur, and oxygen compounds displace chloride from 6-chloropurine ribonucleoside in water at room temperature at rates which are proportional to the first power of the free base of the attacking nucleophile in all cases. When analogs of various amino acid side chains are compared in their rate of reaction with this compound, which is a substrate for the enzyme adenosine deaminase, thiols are found to be the dominant nucleophiles at pH 7 by four orders of magnitude; imidazole is relatively unreactive. Successive addition of methyl groups to ammonia strongly enhances its nucleophilicity for 6-chloropurine ribonucleoside, 1-fluoro-2,4-dinitrobenzene, and *p*-nitrophenyl acetate. However, attack by trimethylamine seems to be sterically hindered except in the purine substrate, yielding a quaternary amine which is readily hydrolyzed by alkali.

The action of a number of hydrolytic enzymes involves nucleophilic displacement yielding an enzyme derivative which is easily hydrolyzed. Recent evidence suggests that purine aminohydrolases may form purinyl–enzyme intermediates in a reaction analogous to nucleophilic aromatic substitution.[2-4] Nu-

(1) Supported by Research Grant GM-12725 from the National Institutes of Health, U. S. Public Health Service, and by a National Science Foundation fellowship held by B. T. Walsh. Inquiries should be addressed to R. Wolfenden.

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(4) A recent report [L. G. Howell and I. Fridovich, *Federation Proc.*, **26**, 448 (1967)] that yeast adenine aminohydrolase catalyzes an apparent exchange of hypoxanthine into 6-chloropurine tends to contradict formation of a purinyl–enzyme intermediate in the case of this enzyme.

cleophilic aromatic substitution has received little study in aqueous solution, except for rates of reaction of some amino acids with 1-fluoro-2,4-dinitrobenzene and with triazines[5] and some primary and secondary amine reactions with *p*-nitrophenyl phosphate.[6] The extensive information accumulated, mainly by Bunnett's group,[7] for nucleophilic aromatic substitutions in organic solvents, is difficult to extrapolate with confidence to aqueous solution, because of differences in solvation energy and occasional complications arising from prototropic equilibria between solvent and nucleophile. In water, the solvent of interest for enzyme action, the individual concentrations of acid and base species may be accurately determined by use of buffers with the glass electrode.

This paper compares the effectiveness of various nucleophiles, analogs of amino acid side chains, in attacking the three kinds of unsaturated carbon present in 6-chloropurine ribonucleoside, 1-fluoro-2,4-dinitrobenzene, and *p*-nitrophenyl acetate in aqueous solution. The purine substrate derives special interest from the fact that it is also a substrate for mammalian[8] and fungal[2] adenosine deaminases (adenosine aminohydrolase EC 3.5.4.4).

Experimental Section

Materials. Reagent grade salts and solvents were used without further purification. Phenol, 2-mercaptoethanol, amines, dimethyl sulfoxide, and water were redistilled before use. 6-Chloropurine ribonucleoside, 1-fluoro-2,4-dinitrobenzene, *p*-nitrophenyl acetate, glycine, and glutathione were obtained from Sigma Chemical Co. and used without further purification. Authentic samples of 6-substituted purine ribonucleosides were prepared by published procedures[9] or kindly provided by the Drug Development Branch of the Cancer Chemotherapy National Service Center, Bethesda, Md. Glutathione was standardized by amperometric titration of solutions with mercuric chloride.[10]

Kinetic Measurements. Rates of the various reactions of 6-chloropurine ribonucleoside were followed by measuring the change in absorption of water solutions, at the wavelength of maximum absorption of the product in question (Table I), with a Zeiss Model PMQ II spectrophotometer equipped with a thermostated cuvette compartment maintained at 25°. In each case the concentration of nucleophile, which also served as the basic component of the buffer system, was in large excess over the concentration (5×10^{-5} *M*) of nucleoside. Ionic strength was maintained at 0.50 by appropriate addition of KCl. Buffer bases or acids, adjusted with HCl or KOH, were used in large excess so that the pH, which was measured at the beginning and the end of reaction with the glass electrode of a Radiometer Model 4 pH meter, was never found to change. No attempt was made to use the glass electrode in experiments with dimethyl sulfoxide–water mixtures; the approximate concentration of free base present in such mixtures was calculated from the pH of the same buffer at the same dilution in water.

Reactions were followed in duplicate, with at least ten optical density readings over at least four half-times except in the case of reaction with ammonia and imidazole. Reaction with these compounds was so slow that it was necessary to estimate the rate

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Despite the probable similarity of the enzymes, we have been unable to find any trace of exchange of inosine into 6-chloropurine ribonucleoside in the presence of mammalian or fungal adenosine aminohydrolases. This discrepancy has been resolved, since Dr. Fridovich writes that the apparent exchange reactions were due to the presence of a radioactive impurity in the hypoxanthine-8-C^{14} used.

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Table I. Ultraviolet Absorption of Reactants and Products[a]

	6-X-Substituted purine ribonucleosides			2,4-Dinitro-1-X-substituted benzenes	
X	λ_{max} (product)	ϵ_{max} (product)	ϵ (reactant)[b]	$\epsilon_{241\ m\mu}$ (product)	$\epsilon_{241\ m\mu}$ (reactant)[c]
OH	253	1.17[d]	0.67	0.82[d]	1.46
NH_2	260	1.49	0.89	0.76	
Glycine	267	1.80	0.90		
$(CH_3)NH$	269	1.74	0.83	0.80	
$(CH_3)_2N$	275	1.85	0.52	0.93	
$(CH_3)_3N^+$	265	0.84	0.91	0.95	
$(C_2H_5)NH$	270	1.78	0.82		
$(C_2H_5)_2N$	275	1.81	0.52		
CH_2OHCH_2S	290	1.84	0.021		
Glutathione	290	1.94	0.021		
C_6H_5O	255	1.10	0.76		

[a] Wavelengths are expressed as mμ; extinction coefficients are expressed as the true value multiplied by 10^{-4}. [b] The extinction of 6-chloropurine ribonucleoside at λ_{max} of the product in question. [c] 1-Fluoro-2,4-dinitrobenzene. [d] Determined in alkaline solution above pH 10.

Table II. Second-Order Rate Constants for Reactions of Nucleophilic Reagents with 6-Chloropurine Ribonucleoside, 1-Fluoro-2,4-dinitrobenzene, and *p*-Nitrophenyl Acetate at 25° [a]

Compound	pK_a[a]	6-Chloropurine ribonucleoside[b]	1-Fluoro-2,4-dinitrobenzene[c]	*p*-Nitrophenyl acetate[c]
Imidazole	7.1	0.00008		29[d]
Ammonia	9.5	0.0001	0.016	21, 16[e]
Glycine	9.8	0.052	16.7[f]	155[e]
Methylamine	10.9	0.53	60	3100
Dimethylamine	10.9	16	640	3000
Trimethylamine	10.1	55	0.66	10, 9.9[d]
Ethylamine	10.8	0.14		
Diethylamine	11.1	0.25		
Triethylamine	10.7	0.005		
Glutathione	8.75[h]	8.6		383[g]
2-Mercaptoethanol	10.0	9.6		700[g]
Phenol	10.1	0.08		105[g]
Hydroxide ion	15.7	0.016	13	850[d]

[a] Rate constants for conjugate base expressed as l. mole^{-1} min^{-1}; apparent pK_a values for conjugate acid determined at 25° at ionic strength 0.50, except for water. Concentration range tested in this investigation (pH range in parentheses) for 6-chloropurine ribonucleoside, 1-fluoro-2,4-dinitrobenzene, and *p*-nitrophenyl acetate, in that order (NT means that the compound was not tested in this investigation): imidazole 0.25 (7.1), NT, NT; ammonia 0.25 (9.4), 0.049–0.20 (9.5), 0.0044–0.023; glycine 0.1–0.25 (9.8), NT, NT; methylamine 0.025–0.25 (10.9–11.8), 0.0005–0.0062 (9.7–10.4), 3.0×10^{-5} to 2.2×10^{-4} (8.3); dimethylamine 0.0025–0.025 (10.1–10.9), 8×10^{-5} to 4×10^{-4} (9.9), 1.5×10^{-5} to 5.5×10^{-5} (8.1–8.3); trimethylamine 1.6×10^{-4} to 2.5×10^{-3} (8.5–10.0), 0.014–0.17 (10.0–10.2), 3.4×10^{-3} to 2.7×10^{-2} (8.7–8.9); ethylamine 0.047–0.25 (10.8), NT, NT; diethylamine 0.045–0.25 (11.1), NT, NT; triethylamine 0.20–0.45 (10.7) NT, NT; glutathione 7.6×10^{-5} to 1×10^{-2} (6.2–11.5), NT, NT; 2-mercaptoethanol 0.005–0.01 (10.0), NT, NT; phenol 0.025–0.05 (10.0), NT, NT; hydroxide ion 0.01–1.0 (12–14), 0.001–0.01 (11–12), NT. [b] Reactions at ionic strength 0.50. [c] Reactions at ionic strength 1.0. [d] See ref 11. [e] W. P. Jencks and J. Carriuolo, *J. Am. Chem. Soc.*, **82**, 1778 (1960). [f] See ref 5. [g] J. W. Ogilvie, J. T. Tilden, and B. S. Strauch, *Biochemistry*, **3**, 754 (1964). [h] pK_a determined from pH dependence of reaction with 6-chloropurine ribonucleoside (Figure 2). Rates for this reaction have not been analyzed in terms of microscopic dissociations constants of glutathione since these are not available for the present experimental conditions; the given rate constant represents the maximum rate of reaction of 6-chloropurine ribonucleoside with glutathione sulfur (Figure 2).

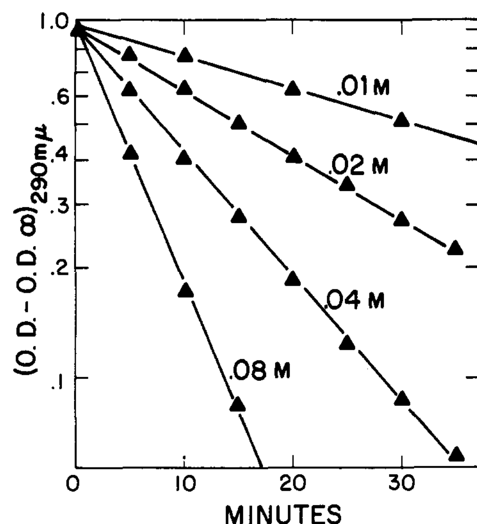

Figure 1. Time course of reaction of 6-chloropurine ribonucleoside with various concentrations of glutathione buffer, pH 8.33, at 25°, ionic strength 0.50.

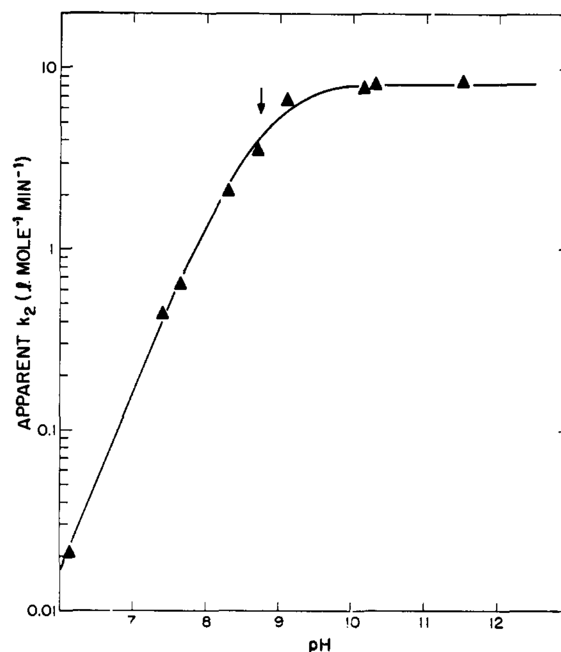

Figure 2. Observed second-order rate constants for reaction of 6-chloropurine ribonucleoside with glutathione, as a function of pH, at 25°, ionic strength 0.50. The solid line is calculated for a reaction involving an acid with pK_a = 8.75, as indicated by the arrow.

from the first 25–50% reaction. In the reaction with phenol, where the nucleophile itself contains a strong chromophore, the reaction mixture was made 10^{-2} *M* in nucleoside and the course of the reaction was followed spectrophotometrically in samples removed at intervals and diluted 200-fold. Extinction coefficients of the reactant and the products at the various wavelengths at which reactions were followed are listed in Table I. Similar procedures were adopted for 1-fluoro-2,4-dinitrobenzene, reactions being followed by the decrease in absorption at 241 mμ, the wavelength of maximum absorption of this substrate, at ionic strength 1.0 (Table I). Aminolysis of *p*-nitrophenyl acetate was followed by the increase in absorption at 400 mμ, the wavelength of maximum absorption of *p*-nitrophenol,[11] at ionic strength 1.0. In other respects procedures were the same as those used for the purine substrate.

Good first-order kinetics, as exemplified in Figure 1, were obtained in all cases. Pseudo-first-order rate constants were calculated from the equation $k = 0.693/t_{1/2}$. Pseudo-first-order rate constants obtained at several buffer concentrations between 10^{-4} and 0.2 *M*, at a single pH value, were plotted against buffer concentration, yielding a line whose slope gave the second-order rate constant for reaction with buffer at that pH. These second-order rate constants, obtained at two or more pH values above and below the apparent p*K* of the buffer under the experimental conditions, were then corrected for the actual concentration of free base of the buffer, in order to obtain the true second-order rate constant. Figure 2 shows the pH dependence of the uncorrected second-order rate constant for glutathione attack on 6-chloropurine ribonucleoside.

Product Analysis. Reactions of 6-chloropurine ribonucleoside with hydroxide ion, ammonia, methylamine, and dimethylamine gave rise to inosine, adenosine, 6-methylaminopurine ribonucleoside, and 6-dimethylammoniumpurine ribonucleoside, identified by spectral comparison with authentic samples of the known compounds. The reaction of 6-chloropurine ribonucleoside with hydroxide ion gave ring opening as well as hydrolysis.[12] Descending paper chromatography on Whatman No. 40 paper in isopropyl alcohol–ammonia–water (85:1.3:15), followed by quantitative elution of the spots, showed that the yield of inosine in this reaction remained constant, at 11.5% of the total 6-chloropurine ribonucleoside which had disappeared, at pH 12, 13, and 14. In this chromatographic system, inosine (R_f 0.11) is well separated from 6-chloropurine ribonucleoside (R_f 0.64) and the product of ring opening (R_f 0.69). The rate constant given for this reaction in Table II is therefore 11.5% of the observed second-order rate constant for the disappearance of 6-chloropurine ribonucleoside in alkali. Reaction of 6-chloropurine ribonucleoside with trimethylamine resulted in a single ultraviolet-absorbing product which migrated with a positive charge upon paper electrophoresis in 0.01 *M* ammonium acetate buffer, pH 5.4 (100 v/cm). In 0.1 *M* potassium carbonate buffer, pH 10.7, this compound was hydrolyzed at room temperature to inosine and trimethylamine with a half-time of 50 min; at this pH the other 6-amino derivatives of adenosine are completely stable. Reactions of 6-chloropurine ribonucleoside with glycine, ethylamine, and diethylamine yielded in each case a product resembling in spectrum those derived from the other amines discussed above; wavelengths of maximum absorption of the products are listed in Table I. Reactions with triethylamine and with imidazole were slow and were followed by the appearance of inosine, the sole ultraviolet-absorbing product; chromatography of reaction mixtures at various times during the course of reaction showed that the expected proportions of starting material remained; it thus appears that the addition products are formed slowly but hydrolyzed rapidly. Reactions with 2-mercaptoethanol and with glutathione each gave rise to a product with an ultraviolet absorption maximum at 290 mμ, almost identical with that of an authentic sample of 6-methylmercaptopurine ribonucleoside. The glutathione product, purified by adsorption on charcoal and eluted with ethanol, was found to react with ninhydrin and to undergo rapid hydrolysis (half-time = 4 min) in 1 *M* KOH at room temperature to 6-mercaptopurine ribonucleoside, which was identified with an authentic sample by ultraviolet spectrum in alkaline and neutral solutions and by paper chromatography in *n*-propyl alcohol–tetrahydrofurfuryl alcohol–0.08 *M* ammonium acetate, pH 3.02 (2:1:1). This confirmed identification of the original product as a thioether. Reaction of 6-chloropurine ribonucleoside with phenol gave rise to a single product, isolated by paper chromatography (R_f 0.84) in *n*-propyl alcohol–2 *N* ammonia (70:30), which separated it from 6-chloropurine ribonucleoside (R_f = 0.72). This product, which resembled 6-methoxypurine ribonucleoside in spectrum, was hydrolyzed to inosine and phenol (half-time = 400 min) in 1 *M* KOH at room temperature and was identified as the 6-phenoxy ether.

Reaction of 1-fluoro-2,4-dinitrobenzene with ammonia and with hydroxide ion gave the expected aniline and phenol, identified by spectral comparison with the known compounds.[11,13] Reaction with methylamine, dimethylamine, and trimethylamine gave products with molar extinctions as noted in Table I. The trimethylamine product, like that obtained with the purine substrate,

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was found to migrate with a positive charge upon paper electrophoresis at pH 5.4 under the conditions noted above.

Results

6-Chloropurine ribonucleoside was found to react with a variety of nucleophiles at reasonable rates at room temperature; in each case the reaction rate was observed to increase with pH, approaching a plateau above the apparent pK_a of the conjugate acid of the buffer nucleophile under the experimental conditions. Figures 1 and 2 show the dependence on pH and on nucleophile concentration for the reaction of 6-chloropurine ribonucleoside with glutathione. Rate constants in column 2 of Table II are calculated from similar data for other nucleophiles. There was no evidence in any of the purine reactions for a greater than first-order dependence on the concentration of nucleophile. There was also no evidence of specific base catalysis in the pH range studied.

Table II also includes second-order rate constants obtained for 1-fluoro-2,4-dinitrobenzene and *p*-nitrophenyl acetate in the present investigation, as well as some second-order rate constants compiled from earlier studies.

Table III shows the second-order rate constants for reaction of 6-chloropurine ribonucleoside with primary, secondary, and tertiary methylamines in water–dimethyl sulfoxide mixtures.

Table III. Rates of Reaction of 6-Chloropurine Ribonucleoside with Amines in Water–Dimethyl Sulfoxide Mixtures[a]

Nucleophile	100% water	10% water	1% water
Methylamine	0.53	0.62	0.65
Dimethylamine	16	9.6	9.0
Trimethylamine	55	3.8	1.7

[a] Second-order rate constants at 25°, expressed as $l.\ mole^{-1}\ min^{-1}$. Water % expressed as w/v. KCl was omitted from water–dimethyl sulfoxide mixtures. Concentration ranges tested (pH of buffer, determined separately in water, in parentheses): 10% water–methylamine 0.009–0.045 (10.9), dimethylamine 0.008–0.040 (10.9), trimethylamine 0.004–0.040 (10.1); 1% water–methylamine 0.0079 (10.9), dimethylamine 0.0078 (10.9), trimethylamine 0.0077 (10.1).

Discussion

6-Chloropurine ribonucleoside is distinguished from 1-fluoro-2,4-dinitrobenzene and from *p*-nitrophenyl acetate by its susceptibility to attack by trimethylamine (Table II). Its rate of reaction increases by almost six orders of magnitude in the series ammonia, methylamine, dimethylamine, trimethylamine. Reactivity of the tertiary amine is sharply reduced in the ethylamine series and in reactions of the methylamine series with the nitrobenzene and carboxylic acid derivatives. Since base catalysis is absent, it is reasonable to assume that the purine substitution reactions proceed by the same general mechanism, with rate-limiting formation of a tetrahedral intermediate.[6,14] Examination of space-filling models suggests less crowding of transition states for formation of a tetrahedral intermediate in trimethylamine attack on 6-chloropurine ribonucleoside than in attack on the nitrobenzene or carboxylic acid derivatives. Such comparison may, of course, be invalid if distances between atoms in transition states for reaction with the three substrates are markedly different. It is noteworthy that the amide, which is presumably formed during nucleophilic catalysis by trimethylamine of the hydrolysis of *p*-nitrophenyl acetate, has not been isolated;[11] its implied instability is probably shared by the transition state for its formation.

Reactivity has been found to increase markedly from ammonia to trimethylamine (though the range is less extreme than in the present case) in a number of aqueous reactions where steric effects appear to be small.[15] Rates of attack in the methylamine series may be largely explained by decreasing strength of solvation of the nucleophile and by the increasing availability of its lone pair electrons for reaction with Lewis acids as methyl groups are added. Thus, the basicity of ammonia, measured with indicators in nonaqueous systems where specific solvation effects are reduced or abolished, increases sharply with successive methyl substitution[16] and partition coefficients between water and nonpolar solvents increase more than 100-fold in favor of the nonpolar solvent in the series from ammonia to trimethylamine.[17] The solvation effect may account for the leveling effect of increasing proportions of dimethyl sulfoxide (Table III) on the relative rate constants for attack in the methylamine series; this could also be explained by changes in transition state structure with changes in solvent composition.

The diversity of 6-substituted purine derivatives which serve as substrates for adenosine deaminase, together with their similar rates of hydrolysis in the case of the enzyme from takadiastase, have suggested formation of a purinyl–enzyme intermediate whose decomposition is rate limiting.[2,18] As models for the reaction producing this intermediate, nucleophiles resembling common amino acid side chains may be compared in their rate of attack on the purine substrate. From the data in Table II it may be calculated that at pH 7 glutathione sulfur is at least 20,000 times more effective than imidazole, phenol, or methylamine, though considerably slower than an enzyme. This contrasts with the behavior of *p*-nitrophenyl acetate, which is most rapidly attacked by imidazole. The special dominance of sulfur nucleophiles in aromatic substitution has been previously recognized in nonaqueous systems and ascribed to the high polarizability of sulfur.[19] It is interesting to note that fungal and mammalian adenosine deaminases are specifically inactivated by sulfhydryl reagents. In these enzymes the binding of substrate and the binding of mercurials have been found to be mutually exclusive; furthermore the reactive group or groups on the enzyme remain masked in the transition state for the rate-limiting

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step.[20] The present findings strengthen the possibility of a direct role for sulfur in catalysis by adenosine deaminase.

Deficiencies of the sulfur nucleophiles, which could be satisfied by the intervention of other catalytic groups, are their relatively slow rates of reaction compared with enzymes and the direction of cleavage of the glutathione derivative in alkali. This latter reaction results in a net transfer of sulfur from glutathione to 6-mercaptopurine ribonucleoside, a reaction which may provide a mechanistic analogy for the enzymatic transfer of sulfur from cysteine to the sulfur-containing nucleotides of soluble ribonucleic acid.[21]

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The Mechanism of the Acid-Catalyzed Hydration of Phenylpropiolic Acid[1,2]

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Abstract: In 62–70% sulfuric acid at 25° phenylpropiolic acid is hydrated at a convenient rate to benzoylacetic acid, which decarboxylates more slowly to acetophenone. Rate constants, obtained spectrophotometrically and corrected for decarboxylation, for *p*-methoxy-, *p*-methyl-, unsubstituted, *p*-chloro-, *m*-chloro-, and 3,4-dichlorophenylpropiolic acids show strict proportionality to h_0, plots of log k *vs.* H_0 having essentially unit slope. Hydration rates are quite sensitive to the presence of electron-donating substituents in the phenyl ring; rates for the first five compounds extrapolated to 50% H_2SO_4 at 25° correlate with σ^+ with a ρ of -4.77 ± 0.07. The first four acids hydrate some four times faster in H_2O–H_2SO_4 than in D_2O–D_2SO_4 of the same mole fraction sulfate at 25°. Rates determined at 25 and 44° for *p*-chlorophenylpropiolic acid correspond to an activation energy of 23.7 ± 1.2 kcal/mole and an entropy of activation of -23 ± 4 eu. These results are most consistent with a mechanism involving rate-determining protonation of the α-carbon of phenylpropiolic acid to give a benzyl vinyl cation. A mechanism involving the formation of phenyltrihydroxyallene by 1,4 addition of water, followed by rate-limiting carbon protonation of the allene, is not in agreement with these experimental observations, but is not rigorously excluded by them.

In 1882, Baeyer reported that ethyl phenylpropiolate was converted by cold, concentrated sulfuric acid to another ester, which could be hydrolyzed to an acid.[4] He suggested that the new acid was benzoylacetic acid, formed by hydration of the triple bond. This was confirmed by Perkin, who also prepared benzoylacetic acid directly from phenylpropiolic acid.[5]

Phenylacetylene had been reported to undergo hydration under similar conditions,[6] and Baeyer suggested that this was a general reaction of the triple bond.[4]

Jacobs and Searles[7] studied the rate of hydrolysis of alkyl ethynyl ethers, which hydrolyze more rapidly than alkyl vinyl ethers. Jacobs and Searles suggested that the hydrolysis is initiated by the rate-determining addition of a proton to the triple bond. This conclusion was completely substantiated by the very careful work of Drenth[8–11] and co-workers on alkyl ethynyl ethers and thioethers.

Very recently, Bott, Eaborn, and Walton concluded that the hydration of substituted phenylacetylenes in acetic acid–aqueous sulfuric acid proceeded by the same mechanism.[12]

Two general mechanisms for the acid-catalyzed hydration of α,β-unsaturated carbonyl compounds can be considered: 1,4 addition of water (eq 1–5) or 3,4 addition of water (eq 6–8).

Steps 1, 3, and 5 involve proton transfers to oxygen and are fast equilibrium steps.[13] Steps 2 and 4 may both be rate limiting. In the second general mechanism, (6) is rate determining in aqueous acid, although (7) can conceivably become rate limiting if the activity of water in the medium is low.

Extension of these mechanisms to α,β-acetylenic

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