Intra- and Inter-molecular General Base Catalysis in the Aminolysis of Benzylpenicillin

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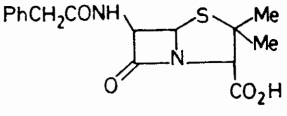
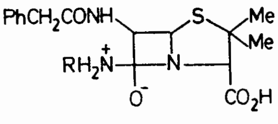
Summary The rate law for the aminolysis of benzylpenicillin is obtained and Brønsted β-values indicate that the general-base-catalysed reaction occurs with a stepwise proton-transfer step; intramolecular general base catalysis occurs in the reaction of ethylenediamine with penicillin.

The aminolysis of penicillin (I) is of interest because the principal antigenic determinant in penicillin allergy is the penicilloyl group bound by an amide linkage to amino-groups on proteins.[1] However, it is not known whether these penicilloyl compounds are formed by direct aminolysis[2] or by aminolysis of the highly reactive degradation product, penicillenic acid.[3]

The susceptibility of penicillins to attack by nucleophiles has been attributed to strain in the β-lactam ring[4] and to the nonplanarity of the system which inhibits the usual amide resonance[5] and which may cause addition to penicillins to resemble more closely that to ketones rather than that to normal amides.



(I)



(II)

The observed pseudo first-order rate constants, k_{obs}, for the aminolysis of the sodium salt of benzylpenicillin in water at 30·0 °C were obtained spectrophotometrically at 235 nm and may be described by equation (1). k_0 is the

$$k_{\text{obs}} - k_0 = k_1[\text{amine}] + k_2[\text{amine}]^2 + k_3[\text{amine}]\,[\text{OH}]^- \qquad (1)$$

first-order constant for the hydrolysis reaction and was normally less than 15% of k_{obs}. This rate law is similar to that found for the related aminolysis of *N*-aryl β-lactams[6] and the individual rate constants were separated as described previously.[7] The relative importance of the terms in equation (1) depends on the basicity and the concentraton of amine. For strongly basic amines the amino-(k_2) and the hydroxide ion-(k_3) catalysed reactions contribute most to aminolysis, the k_2 term, of course, being more important with increasing concentration of amine. For weakly basic amines, where the hydroxide ion concentration is low, aminolysis occurs mainly through the amine-catalysed pathway (k_2) and the uncatalysed pathway (k_1). The results are shown in the Table.

The Brønsted β-value for the general-base-catalysed aminolysis reaction (k_2) is 1·0 and that for the hydroxide ion-catalysed pathway is 1·1. These values may be compared with those obtained from the aminolysis of the β-lactam 1-*p*-nitrophenylazetidin-2-one, 0·97 and 0·75, respectively.[6] It may have been expected that lower values would be obtained in this work because inhibition of amide resonance in penicillin increases the electrophilicity of the carbonyl carbon and would thus, perhaps, make proton transfer less important compared with that in the *N*-aryl β-lactam. However, this is not the case and the β-values are indicative of a stepwise rather than a concerted mechanism for general-base catalysis and this reaction thus represents another example of such a pathway involving proton transfer.[8] The β-values near unity indicate that the reaction behaves as if approximately one positive charge has been developed in the transition state that is distributed between the nucleophile and the catalyst. The simplest mechanism consistent with this involves pre-equilibrium formation of a tetrahedral intermediate (II), exhibiting a β-value of 1·0,[8] followed by diffusion of the catalysing base into the same solvent cage as the intermediate which, for strongly basic catalysts, would be the rate-limiting step and show a β-value of 0. Support for this mechanism also comes from the ratio of the rate constants for base catalysis by hydroxide-ion and by amine, k_3/k_2, which is 65 ± 30. The rate of encounter-controlled proton transfer involving hydroxide-ion is often 10–100 times faster than that predicted by extrapolation of data for nonsolvent bases.[9]

TABLE. Rate constants[a] for the aminolysis of benzylpenicillin at 30 °C in water, ionic strength 0·2 M.

Amine	pK_a	$10^2\,k_1$ / l mol^{-1} s^{-1}	$10^2\,k_2$ / l^2 mol^{-2} s^{-1}	k_3 / l^2 mol^{-2} s^{-1}
$PrNH_2$	10·79	<1·0	105(7%)	48·0(4%)
$HOCH_2CH_2NH_2$	9·73	0·835(8%)	7·75(7%)	6·50(6%)
$MeOCH_2CH_2NH_2$	9·66	0·140(7%)	3·58(4%)	3·56(6%)
$H_2NCH_2CH_2NH_2$	10·07	8·07(3%)	52·8(6%)	16·7(15%)

[a] First-order observed rate constants were calculated by means of a generalised least-squares method and the values of k_1, k_2, and k_3 derived using a linear least-squares method. Estimated standard deviations (%) are given in parentheses.

The rate constant, k_1, for the reaction of ethylenediamine with benzylpenicillin is at least 30-fold (statistically corrected) larger than those for the reactions of primary monoamines of the same basicity. The exact rate enhancement is difficult to obtain since the uncatalysed aminolysis represents such a small contribution to the total reaction. This rate acceleration is interpreted as evidence for intramolecular general-base catalysis of aminolysis by the second nitrogen atom in ethylenediamine. Although the absolute rate increase caused by intramolecular general-base catalysis relative to the 'water' reaction is considerable because of the importance of general-base catalysis in these reactions, the contribution of intramolecularity itself is small. The second-order rate constant for the reaction of ethylenediamine with penicillin may be divided by the third-order constants, k_2, for intermolecular catalysis of aminolysis by a second molecule of amine of similar basicity to give an 'effective molarity' of the adjacent amine in the intramolecular reaction.[10] The value obtained is small, 1 M, presumably because the entropy of low-frequency vibrations in the loose transition-state involved offsets the large loss of translational and rotational entropy that normally occurs in intermolecular reactions.[10]

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