Pentacovalent Phosphorus in the Hydrolysis of Dibenzylphosphoenolpyruvate

Sir:

Clark and Kirby[1] originally observed that either dimethyl- or diphenylphosphoenolpyruvic acid undergoes hydrolysis with loss of methanol or phenol at an accelerated rate relative to trimethyl or triphenyl phosphate at neutral pH. This phenomenon was attributed to intramolecular nucleophilic attack by carboxyl or carboxylate on phosphorus with displacement of alcohol or alkoxide rather than expulsion of the enol or enolate. Alternatively, the hydrolysis can be formulated as general catalysis by the carboxylate or carboxyl group.[2,3]

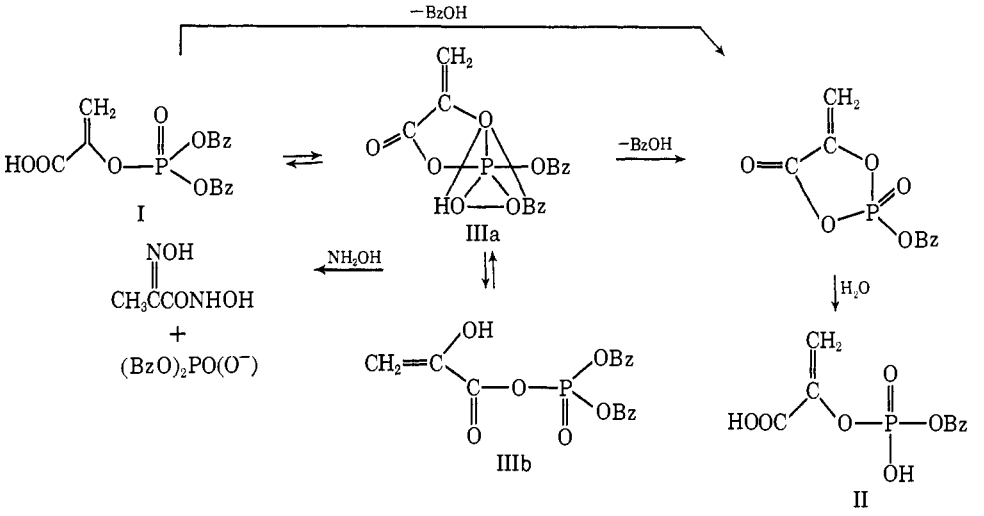
We have investigated the hydrolysis of dibenzylphosphoenolpyruvic acid (I)[4] and monobenzylphosphoenolpyruvic acid (II)[4] which proceed *via* stepwise loss of benzyl alcohol. The pH–rate profiles for release of benzyl alcohol[5] (1 mol from I) reveal that the hydrolytically reactive species for I and II must involve a protonated carboxyl group or its kinetic equivalent. In order to further elucidate the mechanism of this reaction we have conducted experiments in the presence of hydroxylamine, a possible acyl trapping reagent.[6] In Table I the products of hydrolysis in water *vs.* 0.67 *M* hydroxylamine are compared.

Table I. Products of Hydrolysis of I and II in Water *vs.* 0.67 *M* NH_2OH

Compd	Products in water[a]	Products in 0.67 *M* NH_2OH[a]
I	Benzyl alcohol,[b] monobenzylphosphoenolpyruvate	Dibenzyl phosphate,[c] pyruvic acid hydroxamate[d]
II	Benzyl alcohol, phosphoenolpyruvate	Benzyl alcohol, pyruvic acid hydroxamate[e]

[a] Carried out in 50:50 v/v water–dioxane at pH 5.05 maintained by 0.5 *M* acetate buffer (pH region of carboxyl group participation). [b] 95 ± 4% yield. [c] Isolated as dibenzyl hydrogen phosphate; mp 78–79° (lit. mp 78°),[f] 98 ± 2% yield. [d] Isolated as the oxime; mp 152–154° (lit. mp 143°,[f,g] 150–160°,[h] 161°[i]). Infrared spectrum identical with that of authentic sample. [e] Shown to be chromatographically identical in two solvent systems with authentic sample and presumably arises from phosphoenolpyruvate hydroxamate (amyl alcohol–acetic acid–H_2O, 4:1:5; octyl alcohol–formic acid–H_2O 3:1:3). [f] V. M. Clark and A. R. Todd, *J. Chem. Soc.*, 2023 (1950). [g] M. A. Whitely, *ibid.*, 77, 1040 (1900). [h] G. Ponzio, *Gazz. Chim. Ital.*, **55**, 453 (1925). [i] C. Gastaldi, *ibid.*, **54**, 1040 (1924).

Scheme I

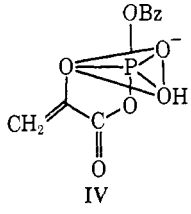


The highly informative observation of hydroxamate products indicates nucleophilic participation by carboxyl to form a reactive acyl phosphate bond in both I and II. Moreover in hydroxylamine solution the course of the reaction of I is altered from that of loss of benzyl alcohol to cleavage of the enol phosphorus bond and formation of dibenzyl phosphate. Therefore, hydroxamate formation cannot simply be trapping of a cyclic tetravalent phosphate as this requires prior release of benzyl alcohol which is not observed. Hydroxylamine must trap an intermediate prior to the cyclic phosphate, most logically the cyclic pentacovalent intermediate IIIa or the acyl phosphate IIIb (see Scheme I). With the present data the existence of IIIb cannot be unequivocally excluded since the calculated ratio of rate constants[7] for hydroxylaminolysis and hydrolysis of the acyl phosphate is 10^8–10^9 and is similar to the ratio of rate constants for the identical nucleophilic reactions on 2,4-dinitrophenyl acetate (10^8).[8]

The intermediacy of IIIa, however, is required to rationalize the formation of IIIb through pseudorotation leading to an axial enolic oxygen (see discussion below) and is consistent with the catalyzed expulsion of benzyl alcohol, since on chemical grounds IIIb would not be anticipated to lose benzyl alcohol more rapidly than I. A possible competing pathway is a direct displacement of benzyl alcohol whose contribution to the hydrolysis scheme cannot be presently assessed.

(1) V. M. Clark and A. J. Kirby, *J. Amer. Chem. Soc.*, **85**, 3705 (1963); for a further example see G. M. Blackburn and M. J. Brown, *ibid.*, **91**, 525 (1969).
(2) T. C. Bruice and S. J. Benkovic, "Bioorganic Mechanisms," Vol. II, W. A. Benjamin, Inc., New York, N. Y., 1966, p 54.
(3) A. J. Kirby and S. G. Warren, "The Organic Chemistry of Phosphorus," Elsevier Publishing Co., New York, N. Y., 1967, p 324.
(4) F. Cramer and D. Voges, *Chem. Ber.*, **92**, 952 (1959).
(5) J. Kumamoto and F. H. Westheimer, *J. Amer. Chem. Soc.*, **77**, 2515 (1955).
(6) F. Lippmann and C. Tuttle, *J. Biol. Chem.*, **159**, 21 (1945).
(7) Based on the observed product distribution, Table I, with the proviso that all products arise according to Scheme I and the assumption that the rate constant for expulsion of BzOH from IIIa is not a function of $[NH_2OH]$.
(8) W. P. Jencks and M. Gilchrist, *J. Amer. Chem. Soc.*, **90**, 2622 (1968)

In the case of II the experiments with hydroxylamine also indicate cyclization but do not require intermediates analogous to IIIa and IIIb. This is in accord with the decreased stability of a monoanionic pentacovalent intermediate (IV) whose zwitterion possesses a greater potential driving force for expulsion of BzOH than the neutral species. The successful trapping with hydroxyl-



IV

amine in the case of I may indicate a higher concentration of IIIa and/or IIIb relative to IV if the latter exists other than as a transition state species. A second consideration influencing the hydroxylamine results is the relative magnitude of the proton-transfer steps leading to the product forming zwitterion from the respective initial species.[9]

Consideration of the recently reviewed[13,14] chemistry of pentacovalent phosphorus indicates that IIIa will be formed by axial attack of the carboxyl with a benzyl group axial and the ring spanning basal and axial positions. The experiments of Dennis and Westheimer[15] and Frank and Usher[12] suggest that the phosphoryl oxygen initially occupies a basal position. Given a pentacovalent intermediate in the hydrolysis of the monobenzyl ester, it is possible that rate of pseudorotation and/or proton transfer to yield the acyl phosphate is competitive with the rate of loss of benzyl alcohol from IV. Consequently, if trapping by hydroxylamine occurs only at the acyl phosphate, the lack of monobenzyl phosphate in the products is explained.

Our observation provides direct verification for the hypotheses of Westheimer[13] and Ramirez[14] and is in accord with the calculations of Boyd,[16] although there is some question as to the distribution of charge in the pentacovalent species.

In view of the observation that monobenzyl PEP monoanion forms a cyclic intermediate we are examining the possibility that an unproductive cyclic phosphate may form with phosphoenolpyruvic acid itself, although as we have shown previously[17] the carboxyl group has no kinetically important role in PEP hydrolysis. Complete details of the mechanism of this reaction system will be published shortly.

(9) That the ionized hydroxyl group prefers an equatorial position is based on the conclusion of Muetterties[10] that the more electronegative groups are axial, the product distribution of methyl ethylenephosphate as a function of pH,[11] and the products of dimethylphosphoacetoin hydrolysis.[12]

(10) E. L. Muetterties and R. A. Schunn, *Quart. Rev.* (London), 20, 245 (1966).

(11) R. Kluger, F. Covitz, E. Dennis, L. D. Williams, and F. H. Westheimer, personal communication.

(12) D. S. Frank and D. A. Usher, *J. Amer. Chem. Soc.*, **89**, 6360 (1967).

(13) F. H. Westheimer, *Accounts Chem. Res.*, **1**, 70 (1968).

(14) F. Ramirez, *ibid.*, **1**, 168 (1968).

(15) E. A. Dennis and F. H. Westheimer, *J. Amer. Chem. Soc.*, **88**, 3431, 3432 (1966).

(16) D. B. Boyd, *ibid.*, **91**, 1200 (1969).

(17) S. J. Benkovic and K. J. Schray, *Biochemistry*, **7**, 4090 (1968).

Acknowledgment. We wish to acknowledge support of this work by the National Institutes of Health and the constructive comments of Dr. D. Usher.

(18) Alfred P. Sloan Fellow, 1968–1970; National Institutes of Health Career Development Awardee.

(19) National Institutes of Health Predoctoral Fellow, 1967–1969.

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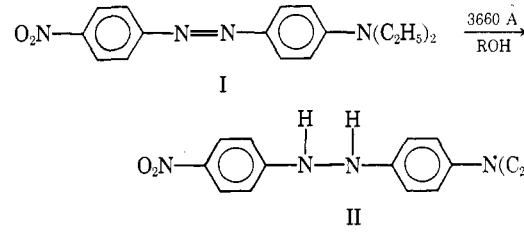
Received April 7, 1969

Photochemistry of Azo Compounds. III. Evidence for an Electron-Transfer Process in Amine Solvents

Sir:

We wish to report our finding that both the products and the kinetics of the photoreduction of 4-(diethylamino)-4′-nitroazobenzene[1] (I) are markedly influenced by the nature of the solvent used. The results from photolyses in alcohols and in butylamine allow a distinction to be made between a mechanism involving hydrogen abstraction by an excited state of I and electron transfer to an excited state of I. We recently found[2,3] that photoreduction of I in alcoholic media occurred with low quantum efficiency to produce 4-(diethylamino)-4′-nitrohydrazobenzene (II) (reaction 1). Sensitizers such as naphthalene were inefficient, thus dem-



I

II

(1)

onstrating that the low quantum yields for reaction I were not a result of inefficient intersystem crossing to a photoreducible triplet.[2] However, benzophenone efficiently sensitized the photoreduction by a mechanism involving transfer of a hydrogen atom from the photochemically generated benzophenone ketyl radical to the azo linkage of I.[3]

Photolyses[4,5] of degassed solutions of I in butylamine (*ca.* 10^{-4} *M*) with 3660-Å light, both with and without ketonic sensitizers present, yielded a product absorbing at 420 nm ($\epsilon \sim 2.6 \times 10^4$) with a shoulder at 460 nm. This product was not formed during photolyses in isopropyl alcohol. When air was admitted to the solution, this product was rapidly converted to a new absorbing species (λ_{max} 507 nm). The second product was identified as 4,4′-bis[[*p*-(diethylamino)phenyl]azo]azoxybenzene (IV): mp 295–298° dec; ir (KBr) 1303 cm^{-1} (s, NO); mass spectrum[6] (70 eV) *m/e* (rel intensity) 548 (25), 532 (100), 148 (38), 44 (27); visible spectrum $\lambda_{max}^{butylamine}$ 507 nm (ϵ 5.2 × 10^4).

(1) E. Baumberger, *Ber.*, **28**, 843 (1895); E. Ziegler and G. Snatzke, *Monatsh. Chem.*, **84**, 610 (1953).

(2) G. Irick, Jr., and J. G. Pacifici, *Tetrahedron Lett.*, 1303 (1969).

(3) J. G. Pacifici and G. Irick, Jr., *ibid.*, 2207 (1969)..

(4) Photolyses were followed spectrophotometrically by methods previously described.[2,3]

(5) $\lambda_{max}^{butylamine}$ 483 nm (ϵ 3.04 × 10^4).

(6) Obtained using a Consolidated Electrodynamics Corp. Model 21-110B mass spectrometer.