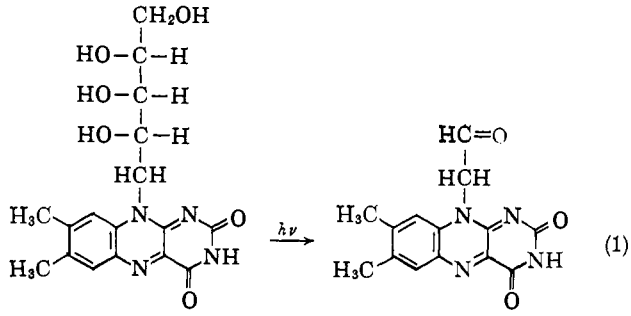
alloxazine (Fig. 4). Upon treatment (in the dark) with 2 *N* NaOH both compound X and authentic 6,7-dimethyl-9-formylmethylisoalloxazine are converted to a compound corresponding to lumiflavin in R_f value (Fig. 4). Furthermore, both compound X and the authentic 9-formylmethyl flavin undergo a similar anaerobic photobleaching which is much more rapid than that of riboflavin. The reoxidation product from this photobleaching is exclusively lumichrome (in acid or neutral aqueous solution).

Protonation of the isoalloxazine ring of flavins results in a distinct spectral change by means of which the pK_a of the conjugate acid maybe determined. The pK_a value for authentic 6,7-dimethyl-9-formylmethylisoalloxazine is 3.5, three pK units higher than those of other flavins.[19] We have determined the pK_a for compound X spectrophotometrically as 3.46 ± 0.08, providing further proof of its identity to 6,7-dimethyl-9-formylmethylisoalloxazine.

Discussion

The results of microbiological assays and thin layer chromatography verify that anaerobically photobleached riboflavin is not dihydroriboflavin (leucoflavin) but a mixture of flavins including riboflavin itself, lumichrome, and two unknown compounds. The major unknown component which accounts for about 10% of the total flavin has been identified as 6,7-dimethyl-9-formylmethylisoalloxazine (eq. 1).



riboflavin (1)

This finding is in agreement with the conclusions of Kocent[20] that glyceraldehyde and glycolaldehyde are the products of side-chain cleavage. He also postulated the existence of the 9-formylmethyl flavin as an intermediate giving rise to glycolaldehyde and lumichrome during flavin photolysis. This compound meets the description of "deuteroflavin," the riboflavin oxidation product postulated by Kuhn as the precursor of lumiflavin in basic solution.

The very slow bleaching of lumiflavin alone and its rapidity in the presence of alcohol together with the microbiological assays and chromatographic evidence lend credence to the idea that the initial photochemical reaction of riboflavin consists of the oxidation of one of the alcohol groups of the ribityl side chain. Whether or not this results in a single step in cleavage to yield glyceraldehyde and 6,7-dimethyl-9-formylmethylisoalloxazine remains uncertain. These results do not offer any support for the proposal that water is cleaved during the photoreduction.

The presence of a certain amount of genuine dihydroriboflavin in the photobleached riboflavin solution is evident from both the microbiological assays and the chromatography. This can be understood if we assume that any reduced flavin with modified side chain, $F'H_2$, could react rapidly by an exchange reaction with riboflavin, F.

$$F'H_2 + F \longrightarrow F' + FH_2 \qquad (2)$$

Dihydroriboflavin so produced is "trapped" in the reduced state and is no longer as sensitive to light. Depending upon the equilibrium constant for the exchange reaction, a greater or lesser amount of dihydroriboflavin could be trapped in the reduced state.

Work is in progress in an attempt to identify the second minor photolysis product and to elucidate the mechanisms of the photolysis reactions.

Acknowledgments.—We gratefully acknowledge the able assistance of Martha L. McDonald.

(19) C. H. Suelter and D. E. Metzler, *Biochim. Biophys. Acta*, **44**, 23 (1960).

(20) A. Kocent, *Chem. Listy*, **47**, 195 (1953).

[CONTRIBUTION FROM THE RESEARCH LABORATORIES OF THE UPJOHN CO., KALAMAZOO, MICH.]

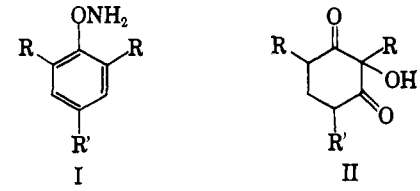
Dihydroazepinone Chemistry. II.[1a] Mechanistic Considerations of the Formation and Acid Hydrolysis of the 1,3-Dihydro-2H-azepin-2-ones

BY LEO A. PAQUETTE[1b]

RECEIVED MAY 3, 1963

The one-step preparation of 1,3-dihydro-2H-azepin-2-ones by ring expansion of appropriate sodiophenoxides with chloramine and methylchloramine is discussed. Evidence is presented to suggest that the ring enlargement proceeds by initial C-alkylation of the ambident phenoxide ion to give an amino dienone which undergoes thermal rearrangement to give the observed products. Acid hydrolysis of the dihydroazepinones affords novel dihydro-2(3H)-furanones. The mechanisms of these reactions are discussed.

Because of our recent activity in various aspects of hydroxylamine chemistry, including the preparation and rearrangement of aminooxy compounds,[2] we noted with more than casual interest the recently published communications of Theilacker and co-workers[3-5] describing the preparation of O-arylhydroxylamines (I) *via* the reaction of sodio-2,6- and 2,4,6-alkyl-substituted phenoxides with chloramine. The resulting O-arylhydroxyamines (I) were described as colorless, stable, highly crystalline solids, capable of distillation without change,



but incapable of condensation with aromatic aldehydes; in addition, they were not acetylatable and, in the

(1) (a) For a preliminary account of this work see L. A. Paquette, *J. Am. Chem. Soc.*, **84**, 4987 (1962); (b) Department of Chemistry, The Ohio State University, Columbus 10, O.

(2) L. A. Paquette, *Tetrahedron Letters*, No. **11**, 485 (1962), and other papers in this series to be published.

(3) W. Theilacker and E. Wegner, *Angew. Chem.*, **72**, 127 (1960); for a correction of their earlier structural assignments, see W. Theilacker, K. Ebke, L. Seidl, and S. Schwerin, *ibid.*, **75**, 208 (1963).

(4) W. Theilacker, *ibid.*, **72**, 498 (1960).

(5) K. Ebke, Ph.D. Dissertation, University of Hannover, Germany, 1959.

presence of mineral acids, were presumably transformed into hydroxycyclohexanediones (II).

These results, although based on sound analogy with the reaction of sodium alkoxides with chloramine to produce O-alkylhydroxylamines,[3] were suspect for several reasons.

First, examination of the literature reveals that the N-monosubstituted hydroxylamines are, as a general rule, appreciably more stable, in a relative sense, than the isomeric aminooxy compounds. Relevant to the problem is the fact that N-arylhydroxylamines are rather unstable substances which, in addition, are well known to suffer transformation to *o*- and *p*-aminophenols under a variety of mild conditions,[6] including in one instance simple dissolution of the base in water at room temperature.[7] Therefore, the O-arylhydroxylamines (I) would be expected, in light of the above evidence, to be substantially more prone to ready decomposition.[10] The predicted instability properties of compounds of type I were at variance with the data reported by the German workers.[3-5]

Secondly and of equal significance is the fact that O-alkyl- and o-aralkylhydroxylamines are readily acetylatable under standard conditions and do undergo smooth condensation with aromatic aldehydes.[11,12]

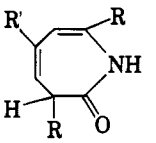
Thirdly, it has been well documented[13] that solutions of phenolic salts in molten phenols undergo extensive carbon alkylation in contrast to preponderant oxygen alkylation in the more commonly used solvent systems. Exclusive O-alkylation in the chloramine reaction would therefore appear incompatible with previous observations.

Finally, there exists no precedence for the transformation I → II.

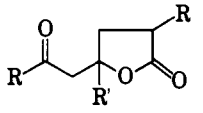
In view of our interest in hydroxylamine chemistry[2] we undertook to reinvestigate the chloramine–phenol reaction. In our hands, the reaction led to products which, although they were identical in physical properties with the compounds reported previously,[3-5] were not O-arylhydroxylamines (I) but rather 1,3-dihydro-2H-azepin-2-ones (III),[1a] resulting from ring enlargement of the phenoxide moiety. In repeating the addition of cold ethereal chloramine to a hot stirred solution of sodio-2,4,6-trimethylphenoxide in excess 2,4,6-trimethylphenol we obtained a fluffy white solid having infrared absorption at 3200 and 1695 cm.$^{-1}$. The ultraviolet absorption spectrum showed a single absorption at 252 mμ (6050). The elemental analyses were in agreement with a $C_9H_{13}NO$ formulation. The n.m.r. spectrum was fully compatible with the 3,5,7-trimethyl-1,3-dihydro-2H-azepin-2-one (IIIa) structural assignment.[1a] Catalytic reduction of IIIa over platinum oxide in ethanol proceeded with the uptake of two moles of hydrogen to give 3,5,7-trimethylhexahydroazepin-2-one, m.p. 73–75°. An authentic sample of this material was prepared by the Beckmann rearrangement of 2,4,6-trimethylcyclohexanone oxime,[14] and the lower melting isomer from this reaction was found to be identical with respect to spectral and melting point properties to the hydrogenation product.

Although a scarcity of model compounds made prediction of the ultraviolet spectrum of IIIa uncertain, Vogel and Erb[15] have recently reported a five-step synthesis of the previously unknown dihydroazepinone ring system (III, R = R′ = H) and have reported a similar ultraviolet absorption.

This reaction therefore represents a facile one-step synthetic entry into an otherwise difficultly obtainable heterocyclic system.[15] Furthermore this procedure is the third reported example[16] of the ring enlargement of a benzenoid system by a nitrogen-containing species and also is one of the few examples of aromatic ring expansion by a noncarbenoid entity.[17]

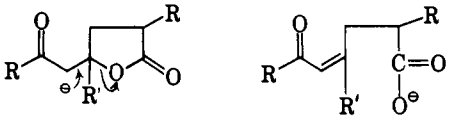


IIIa, R = R′ = CH_3
b, R = CH_3, R′ = H
c, R = CH_2CH_3, R′ = H



IVa, R = R′ = CH_3
b, R = CH_3, R′ = H

Acid hydrolysis of compounds of type III have resulted in the synthesis of the dihydro-2(3H)-furanones (IV). Worthy of mention is the fact that these ketolactones hydrolyze so readily that a direct titration at slow speed is possible.[18] This phenomenon is observed because of the inherent capability of this system to undergo facile retro-Michael reaction according to the scheme



Mechanism of the Ring Enlargement.—The ambident character of phenoxide ions has been clearly established by several investigators.[19] In fact, the inherent capability of the phenoxide anion for covalent bond formation at either carbon or oxygen has found utility as a direct method for the synthesis of cyclohexadienones and phenoxyethers, respectively.[19b] More recent studies have examined the effect of heterogeneity,[19c] hydro-

(6) (a) E. Bamberger, *Ber.*, **27**, 1349, 1522 (1894); **33**, 3600 (1900); **34**, 229 (1901); (b) C. K. Ingold, "Structure and Mechanism in Organic Chemistry," Cornell University Press, Ithaca, N. Y., 1953, pp. 621–624, discusses this example of an aromatic nucleophilic rearrangement.

(7) 2-Hydroxyaminopyridine, a heterocyclic counterpart, is known to decompose to a dark brown resin simply on standing in air.[8,9]

(8) G. T. Newbod and F. S. Spring, *J. Chem. Soc., Suppl.*, **No. 1**, S133 (1939).

(9) E. G. Kovach and D. E. Barnes, *J. Am. Chem. Soc.*, **76**, 1176 (1954).

(10) After completion of this work, strong support of this contention was demonstrated by two groups of workers [(a) C. L. Bumgardner and R. L. Lilly, *Chem. Ind.* (London), 559 (1962); (b) J. S. Nicholson and D. A. Peak, *ibid.*, 1244 (1962)] who reported the preparation of O-phenylhydroxylamine (I, R = R′ = H) and noted it to be a distillable liquid which, however, colored rapidly and decomposed at room temperature. This substance underwent ready acetylation.

(11) E. L. Schumann and R. V. Heinzelman, *J. Med. Chem.*, to be published.

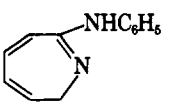
(12) Note especially the final sentence of ref. 10.

(13) N. Kornblum, P. J. Berrigan, and W. J. LeNoble, *J. Am. Chem. Soc.*, **82**, 1257 (1960).

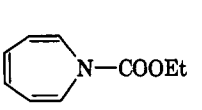
(14) H. E. Ungnade and A. D. McLaren, *ibid.*, **66**, 118 (1944); *J. Org. Chem.*, **10**, 29 (1945).

(15) E. Vogel and R. Erb, *Angew. Chem.*, **74**, 76 (1962).

(16) To the author's knowledge the only previous benzenoid ring enlargements by nitrogenous entities are: (a) the report that the decomposition of phenyl azide in aniline results in the formation of the amidine i [R. Huisgen, D. Vossius, and M. Appl, *Ber.*, **91**, 1 (1958); R. Huisgen and M. Appl, *ibid.*, 12 (1958)]; (b) the report that the action of carbethoxynitrene, generated either by the photolysis of ethyl azidoformate [K. Hafner and C. Koenig, *Angew. Chem.*, **75**, 89 (1963)] or by the α-elimination of *p*-nitrobenzenesulfonic acid from its N-hydroxyurethan ester [W. Lwowski, T. J. Maricich, and T. W. Mattingly, Jr., *J. Am. Chem. Soc.*, **85**, 1200 (1963)], on benzene gives rise to N-carbethoxyazepine (ii).



i



ii

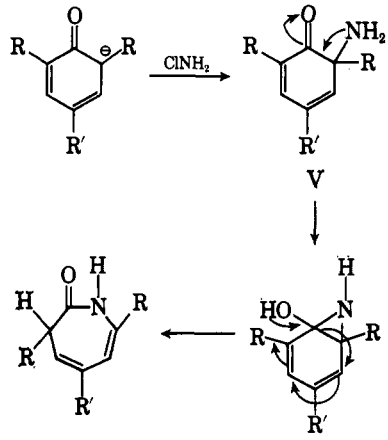
(17) For examples of carbenoid ring expansions of phenols see G. L. Closs and L. E. Closs, *J. Am. Chem. Soc.*, **83**, 599 (1961), and references cited therein.

(18) The author is indebted to D. J. Weber for these data.

(19) (a) L. Claisen, F. Kremers, F. Roth, and E. Tietze, *Ann.*, **442**, 210 (1925); L. Claisen, *Z. angew. Chem.*, **36**, 478 (1923); (b) D. Y. Curtin and D. H. Dybvig, *J. Am. Chem. Soc.*, **84**, 225 (1962), and earlier papers; (c) N. Kornblum and A. P. Lurie, *ibid.*, **81**, 2705 (1959).

gen bonding capacity of the solvent,[20] and dielectric constant of the medium[21] as factors in the alkylation of phenoxide ions. These investigations have demonstrated that when solutions of the salts of phenols are alkylated in a wide variety of solvents, oxygen alkylation is overwhelmingly preferred and the sole product is the ether. However, when the same reactions are conducted in water, excess phenol, or fluorinated alcohols, substantial quantities of C-alkylation products result. Attention has been directed only to alkyl, alkenyl, and aralkyl halides as alkylating agents; granting that these halides are excellent substrates for the studies described above, we saw no reason to limit the applicability of the method.

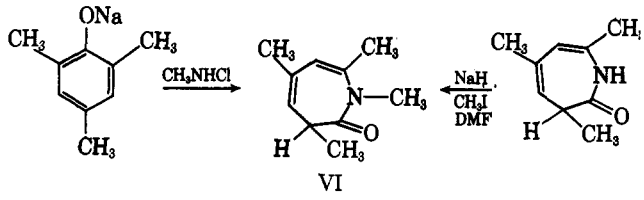
Since the dihydroazepinones are produced in excess phenol as solvent, it is entirely reasonable that the reaction likewise proceeds by nucleophilic displacement on chloramine by the cyclohexadienone anion to produce the aminoketone V. At the elevated temperatures



V

(90–150°),[22] rearrangement can occur with a remarkably small readjustment of bond angles[23] to give an hydroxyaziridine which can undergo further rearrangement and concomitant ring enlargement to produce the dihydroazepinone.

Corroborative evidence for the mechanism advanced above was obtained by examining the alkylation of sodio-2,4,6-trimethylphenoxide with N-methylchloramine and N,N-dimethylchloramine under the conditions previously outlined. Utilization of N-methylchloramine gave rise to 1,3-dihydro-1,3,5,7-tetramethyl-2H-azepin-2-one (VI) in 38% crude yield. Alternatively, VI was prepared by methylation of the sodium salt of 1,3-dihydro-3,5,7-trimethyl-2H-azepin-2-one in



VI

dimethylformamide. From an intense band at 1660 cm.$^{-1}$ in the infrared spectrum of VI the presence of an unconjugated tertiary cyclic amide can be inferred. The ultraviolet absorption at 256 mμ (ε 4,950) lends further support to the 1,3-dihydroazepin-2-one formulation. Finally the n.m.r. spectrum of VI was fully compatible with the proposed structure.

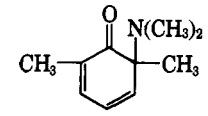
(20) N. Kornblum, P. J. Berrigan, and W. J. LeNoble, *J. Am. Chem. Soc.*, **85**, 1141 (1963).

(21) N. Kornblum, R. Seltzer, and P. Haverfield, *ibid.*, **85**, 1148 (1963).

(22) It has been observed that at lower temperatures the yield of the dihydroazepinones diminishes very drastically. The author is indebted to P. E. Marlatt for calling this to his attention.

(23) When a Dreiding model of VI was constructed, the proximate location of the amino nitrogen to the carbonyl carbon was apparent.

Treatment of sodio-2,6-dimethylphenoxide with N,N-dimethylchloramine, on the other hand, produced a voluminous black tar. This substance most certainly results from the rapid polymerization of the aminocyclohexadienone VII, which cannot undergo rearrange-



VII

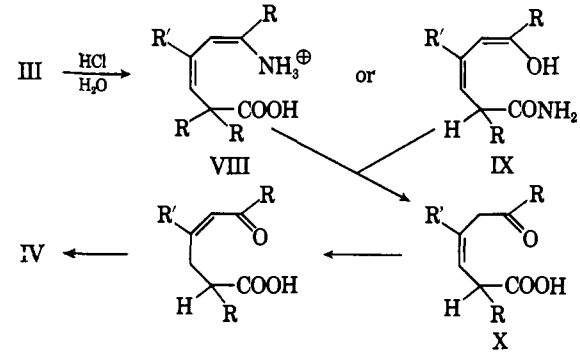
ment at the elevated temperatures.

This mechanism also explains the fact that phenols which are not 2,6-disubstituted afford aminophenols when submitted to the conditions of this reaction.[3,4]

Of the large number of alternative mechanistic possibilities which may be advanced to account for the formation of the dihydroazepinones (III), all but the one described in detail above have not been found plausible for at least two reasons. First, many were inherently incapable of stereoselectively inserting nitrogen into a position ultimately vicinal to the phenolic hydroxyl. Secondly, of the remaining proposed mechanisms which were capable to a degree of achieving the goal of proper nitrogen insertion, all had to be discarded because they lacked precedent and violated established rules governing electronic considerations.

Mechanism of the Acid Hydrolysis.—It has been shown[1a] that hydrolysis of the dihydroazepinones in dilute hydrochloric acid produces the γ-lactones IV. A mechanistic rationalization of the formation of IV involves the initial hydrolysis of III to one of two entities, VIII or IX (see Chart I). Either of these sub-

CHART I



stances can undergo further hydrolysis to the β,γ-unsaturated keto acid X which can further be isomerized to the α,β-unsaturated isomer. Cyclization of this conjugated keto acid in a Michael-type reaction produces IV.

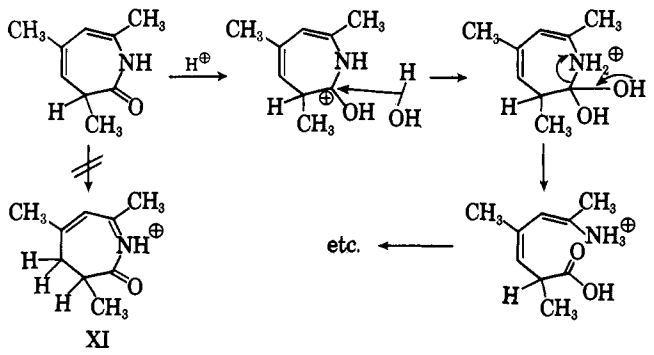
It was of interest to determine the initial mode of hydrolysis of III, for this result could have far-reaching implications in eneamide chemistry.[24] To gain further insight into the nature of the hydrolytic sequence, a sample of 1,3-dihydro-3,5,7-trimethyl-2H-azepin-2-one (IIIa) in 3 *N* hydrochloric acid was heated for 10 min. on a steam bath with occasional swirling.[25] On appropriate work-up[25] there was obtained an oil whose infrared spectrum was identical with that of 5-acetonyldihydro-3,5-dimethyl-2(3H)-furanone previously prepared by vigorous acid hydrolysis of the same dihydroazepinone. This result is strongly suggestive that the eneamino acid VIII is the initial intermediate, because if IX had formed it presumably would have been con-

(24) It can be easily recognized that the dihydroazepinones III are cyclic dieneamides, a class of compounds which remains to date rather elusive.

(25) These reaction conditions are known not to cause amide hydrolysis: A. Brossi, H. Besendorf, B. Pellmont, M. Walter, and O. Schnider, *Helv. Chim. Acta*, **43**, 1459 (1960).

verted to an unsaturated keto amide under these reaction conditions. In addition, it seems entirely reasonable that the initial protonation of the dihydroazepinone molecule occurs preferably at the amide carbonyl, rather than at one of the olefinic sites as in XI.[26] The hydrolytic ring cleavage may then be represented as proceeding by the route shown in Chart II.

CHART II



XI

Experimental[27]

1,3-Dihydro-3,7-dimethyl-2H-azepin-2-one (IIIa). A 300-g. (2.46 moles) sample of 2,6-dimethylphenol was heated to 100°. With stirring, 13.8 g. (0.60 mole) of sodium metal was slowly added at such a rate that the temperature did not exceed 120°. The dark mass was heated to 140° and treated with a cold (−70°) solution of approximately 0.50 mole of chloramine in 500 ml. of ether, the temperature being maintained at 100–140° throughout the addition. The dark mixture was allowed to stand overnight at room temperature.

The dark reaction mixture was distilled at 85–95° (8 mm.) to remove most of the phenol. The contents of the distillation flask were allowed to cool and were treated with 500 ml. of ether and 500 ml. of water. The layers were separated and the aqueous phase was again extracted with 500 ml. of ether. The combined ether layers were dried, filtered, and evaporated to give a red-brown residue. Distillation of this material gave a pale orange crystalline fraction, b.p. 138–147° (5 mm.), 38.0 g. (55.4%). Two recrystallizations of this material from ligroin afforded white cubes, m.p. 121–122°; ν^{Nujol} 3180 (–NH) and 1675 cm.$^{-1}$ (amide carbonyl), λ_{max}^{EtOH} 253 mμ (6100).

Anal. Calcd. for $C_8H_{11}NO$: C, 70.04; H, 8.08; N, 10.21. Found: C, 69.83; H, 7.98; N, 10.56.

1,3-Dihydro-3,5,7-trimethyl-2H-azepin-2-one (IIIb).—2,4,6-Trimethylphenol (334 g., 2.46 moles), sodium metal (13.8 g., 0.60 g.-atom), and 0.50 mole of chloramine in 250 ml. of cold ether solution were allowed to react at 120–150° as described above. The major portion of the unreacted phenol distilled at 100–104° (8–14 mm.). The residue was worked up as described above to give a fraction, b.p. 127–157° (11 mm.). Recrystallization of the crude distillate from ligroin gave, after thorough drying, 38.5 g. (51.0%) of pale yellow solid, m.p. 116–124°. Two additional recrystallizations from ligroin gave pure product as fluffy white needles, m.p. 132°, ν^{Nujol} 3200 (N–H) and 1695 cm.$^{-1}$ (amide carbonyl), λ_{max}^{EtOH} 252 mμ (6050).

Anal. Calcd. for $C_9H_{13}NO$: C, 71.49; H, 8.67; N, 9.26. Found: C, 71.68; H, 8.47; N, 9.16.

3,7-Diethyl-1,3-dihydro-2H-azepin-2-one (IIIc).—2,6-Diethylphenol (370 g., 2.46 moles), sodium metal (13.8 g., 0.60 mole), and 0.50 mole of chloramine in 250 ml. of cold ether solution were allowed to react at 140–150° as described above. The major portion of the unreacted phenol distilled at 85–115° (10 mm.). The residue was worked up as described above to give a fraction (crystallized), b.p. 142–172° (12 mm.), 27.1 g. (32.8%). Two recrystallizations of the crude product from ligroin gave pure azepinone as large white prisms, m.p. 88.5–89.0°, ν^{Nujol} 3200 (N–H) and 1665 cm.$^{-1}$ (amide carbonyl), λ_{max}^{EtOH} 253 mμ (6300).

Anal. Calcd. for $C_{10}H_{15}NO$: C, 72.69; H, 9.15; N, 8.48. Found: C, 72.56; H, 9.03; N, 8.36.

Hexahydro-3,5,7-trimethyl-2H-azepin-2-one.—A solution of 14.6 g. (0.0967 mole) of 1,3-dihydro-3,5,7-trimethyl-2H-azepin-2-one in 130 ml. of glacial acetic acid was hydrogenated over platinum oxide (700 mg.). The major portion of the solvent was removed under reduced pressure. Water was added and the solution was rendered alkaline with concentrated ammonium hydroxide. The mixture was extracted with three 50-ml. portions of methylene chloride and the combined organic layers were dried, filtered, and evaporated. The residual pale yellow oil soon crystallized and it was recrystallized from hexane to give, after drying, 10.8 g. (72.0%) of white solid, m.p. 71–73°. Pure lactam was obtained as a fluffy white solid from hexane; m.p. 73–75° (lit.[14] m.p. 73.5–75°); ν^{Nujol} 3300, 3220 (N–H), and 1670 cm.$^{-1}$ (amide carbonyl).

Anal. Calcd. for $C_9H_{17}NO$: C, 69.63; H, 11.04; N, 9.02. Found: C, 69.57; H, 10.82; N, 8.84.

1,3-Dihydro-1,3,5,7-tetramethyl-2H-azepin-2-one (VI). **A. Ring Expansion with Methylchloramine.**—A 150-g. (1.1 moles) sample of 2,4,6-trimethylphenol was heated to 100° and to this molten solid was added 6.9 g. (0.30 g.-atom) of sodium metal in small portions at such a rate that the temperature of the stirred mixture did not exceed 150°. A solution of 0.25 mole of methylchloramine[28] in about 250 ml. of ether cooled to −70° was added in a thin stream to the phenoxide–phenol mixture previously heated to 140°. The temperature was maintained at 120–140° during the addition by external heating. When the addition was completed, the hot mixture was stirred for an additional 15 min., and the excess phenol was then removed under reduced pressure at 100–117° (12 mm.). The residue was cooled and treated with ether and water. The layers were separated and the aqueous phase was re-extracted with ether. The combined organic layers were dried over magnesium sulfate, filtered, and evaporated; the residue was distilled to give, after a small forerun of phenol, 14.5 g. (38.4%) of a pale yellow liquid, b.p. 118–122° (12 mm.). Redistillation of this material gave a fraction, b.p. 121–122° (12 mm.), $n^{25}D$ 1.5262, whose infrared spectrum was identical with that of the material prepared by method B, except for some slight contamination with residual phenol.

B. **Alkylation of the Dihydroazepinone.**—To a solution of 29.0 g. (0.192 mole) of 1,3-dihydro-3,5,7-trimethyl-2H-azepin-2-one in 150 ml. of dimethylformamide was added 9.0 g. of 51.5% sodium hydride–oil dispersion (0.192 mole). After heating the mixture at 50° for 1 hr., it was cooled and 42.6 g. (0.30 mole) of methyl iodide was added in two portions. Sodium iodide separated immediately. After stirring the slurry at room temperature for 1 hr., ether (250 ml.) was added and the precipitated solid was separated by filtration. The filtrate was evaporated and the residual oil was distilled to give 29.5 g. (93.0%) of colorless liquid, b.p. 115–120° (11 mm.). This material was redistilled to give pure product, b.p. 121.5° (13 mm.), $n^{24}D$ 1.5198.

Anal. Calcd. for $C_{10}H_{15}NO$: C, 72.69; H, 9.15; N, 8.48. Found: C, 72.32; H, 9.26; N, 8.59.

1,3-Dihydro-1,3,7-trimethyl-2H-azepin-2-one.—The same procedure as that described in section A above on 150 g. (1.23 moles) of 2,6-dimethylphenol was employed. There was obtained 7.1 g. (18.8%) of a pale yellow oil, b.p. 109–115° (13 mm.). Purification of this material by preparative scale gas chromatography[29] and redistillation (Hickman still) gave a colorless fluid liquid.

Anal. Calcd. for $C_9H_{13}NO$: C, 71.49; H, 8.67; N, 9.26. Found: C, 71.04; H, 8.71; N, 8.98.

5-Acetonyldihydro-3,5-dimethyl-2(3H)-furanone (IVa).—A mixture of 3.0 g. (0.0198 mole) of 1,3-dihydro-3,5,7-trimethyl-2H-azepin-2-one and 25 ml. of 4 *N* hydrochloric acid was refluxed for 2 hr. during which time solution was effected. The cooled solution was extracted with two 50-ml. portions of methylene chloride and the combined organic layers were dried, filtered, and evaporated. The residual liquid was distilled to give 3.1 g. (83.8%) of colorless lactone, b.p. 89–90° (0.125 mm.), $n^{24.5}D$ 1.4528, $\nu^{pure\ liquid}$ 1770 (lactone carbonyl) and 1720 cm.$^{-1}$ (ketone carbonyl).

Anal. Calcd. for $C_9H_{14}O_3$: C, 63.51; H, 8.29; equiv. wt., 170. Found: C, 63.32; H, 8.51; equiv. wt., 173.

5-Acetonyldihydro-3-methyl-2(3H)-furanone (IVb).—A mixture of 6.3 g. (0.046 mole) of 1,3-dihydro-3,7-dimethyl-2H-azepin-2-one and 50 ml. of 4 *N* hydrochloric acid was refluxed for 3 hr. and allowed to stand overnight at room temperature. The solution was extracted with two 50-ml. portions of methylene chloride and the combined organic layers were dried, filtered, and evaporated. Distillation of the residue afforded 6.6 g. (92.0%)

(26) For an excellent summary of the evidence pertaining to the protonation of amides see A. R. Katritzky and R. A. Y. Jones, *Chem. Ind.* (London), 722 (1961).

(27) Boiling points are uncorrected. The author is indebted to Dr. Robert Rinehart and his associates of the Physical and Analytical Chemistry Department of the Upjohn Co. for the analytical and spectral data.

(28) G. H. Coleman, *J. Am. Chem. Soc.*, **55**, 3001 (1933).

(29) An F and M Model 500 gas chromatograph was employed. The column consisted of a 0.25-in. stainless steel tubing 4 ft. in length containing 20% by weight of Polyester A on firebrick. The collection of samples was performed at a column temperature of 200° with a helium flow rate of approximately 200 ml. per minute. The only impurity was 2,6-dimethylphenol.

of colorless liquid, b.p. 105–106° (3.0 mm.), $n^{24}D$ 1.4556, $\nu_{max}^{pure\ liquid}$ 1775 (lactone carbonyl) and 1720 cm.$^{-1}$ (ketone carbonyl).

Anal. Calcd. for $C_8H_{12}O_3$: C, 61.52; H, 7.75; equiv. wt., 156. Found: C, 61.39; H, 7.96; N, 0.00; equiv. wt., 155.

Mild Acid Hydrolysis of 1,3-Dihydro-3,5,7-trimethyl-2H-azepin-2-one.—A slurry of 3.0 g. (0.020 mole) of 1,3-dihydro-3,5,7-trimethyl-2H-azepin-2-one in 10 ml. of 3 *N* hydrochloric acid was heated on a steam bath for 10 min. with frequent swirling. The solid was converted to an oil. The solution was neutralized by the cautious addition of solid potassium carbonate and the mixture was extracted with methylene chloride. The combined organic layers were dried over magnesium sulfate, filtered, and evaporated to give a colorless oil. The infrared spectrum of this material was superimposable on that of pure 5-acetonyldihydro-3,5-dimethyl-2(3H)-furanone.

[CONTRIBUTION FROM THE DEPARTMENT OF BIOLOGICAL CHEMISTRY, UNIVERSITY OF ILLINOIS, COLLEGE OF MEDICINE, CHICAGO 12, ILL.]

Phosphonic Acid Analogs of Nucleoside Phosphates. I. The Synthesis of 5′-Adenylyl Methylenediphosphonate, a Phosphonic Acid Analog of ATP[1,2]

BY TERRELL C. MYERS, KANAME NAKAMURA, AND JAMES W. FLESHER

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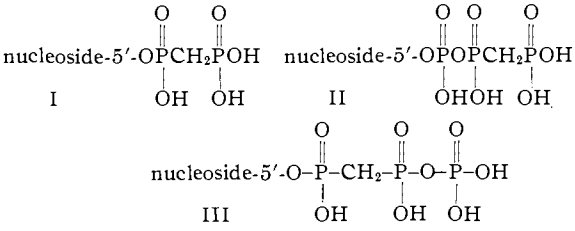
The synthesis of 5′-adenylyl methylenediphosphonate (IV) has been accomplished by the reaction of adenosine 5′-phosphoramidate with methylenediphosphonic acid and by the condensation of AMP with methylenediphosphonic acid in the presence of excess dicyclohexylcarbodiimide.

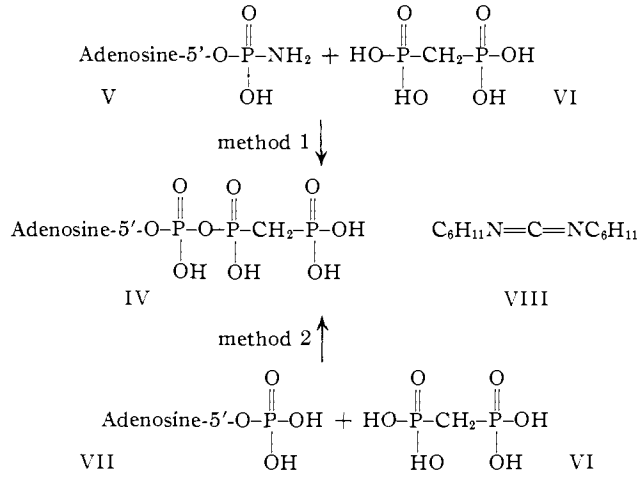
The role of the nucleoside polyphosphates in metabolism is such that studies on the metabolic effects of their analogs may provide information as to their functions. It seems especially important to investigate analogs in which the polyphosphate portion of the molecule has been altered. In particular, it would be of interest to investigate nucleoside polyphosphate analogs which have structures closely related to the polyphosphate groupings, but in which cleavage cannot readily occur, since so many of the metabolic reactions in which the nucleoside polyphosphates participate involve liberation or transfer of ortho- or pyrophosphate groups. Such analogs include compounds in which one or more of the pyrophosphate oxygens are replaced by methylene bridges. Some possible variations are depicted below for analogs of nucleoside 5′-diphosphates (I) and nucleoside 5′-triphosphates (II and III)



Since compounds of types I, II, and III are phosphonic acid derivatives, they would be expected to have physical and chemical properties similar to those of parent polyphosphates except that the C–P bonds of the analogs would be resistant to cleavage.[3] The ease of cleavage of the pyrophosphate moiety of nucleotides is well known.

We have therefore undertaken the synthesis of a series of such phosphonic acid analogs of nucleoside polyphosphates. The present paper describes the synthesis of 5′-adenylyl methylenediphosphonate (abbreviated AMP-PCP)[3] (IV). Assuming that the P–O–P linkage of this analog would be susceptible to enzymic cleavage or group transfer but the P–C–P bonds would not, AMP-PCP might act as: (1) an inhibitor of ATP in processes involving cleavage of the terminal P–O–P bond of ATP; (2) a metabolic substitute for ATP in processes involving cleavage of the P–O–P bonds of the second pyrophosphate oxygen of ATP; (3) a metabolic substitute for ATP in processes involving complexing or binding actions of ATP that are not accompanied by pyrophosphate bond cleavage.

AMP-PCP (IV) was synthesized by two independent methods: (1) the reaction of adenosine 5′-phosphoramidate (V) with methylenediphosphonic acid (VI)



and (2) the reaction of AMP (VII) with methylenediphosphonic acid using dicyclohexylcarbodiimide (VIII) as the condensing agent.

These methods are modifications, in which methylenediphosphonic acid is used in place of orthophosphoric or pyrophosphoric acid, of established syntheses of ATP and ADP.[4]

In method 1 the reaction between the phosphoramidate and the phosphonic acid (three molar excess) was carried out in a solution of *o*-chlorophenol and pyridine. The nucleotide derivatives in the reaction mixture were adsorbed on charcoal at pH 2, freed from excess methylenediphosphonic acid by washing the charcoal with water, and eluted with aqueous ethanolic ammonium hydroxide. AMP-PCP was purified by ion-exchange chromatography and isolated as the tetralithium salt in 22% yield.

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(2) The following abbreviations are used: AMP-PCP, 5′-adenylyl methylenediphosphonate; ATP, adenosine 5′-triphosphate; ADP, adenosine 5′-diphosphate; AMP, adenosine 5′-phosphate; DCC, dicyclohexyl carbodiimide.

(3) G. M. Kosolapoff, "Organophosphorus Compounds," John Wiley and Sons, Inc., New York, N. Y., 1950, Chapter 7.

(4) For procedures analogous to those employed in method 1 see (a) R. W. Chambers and H. G. Khorana, *J. Am. Chem. Soc.*, **80**, 3749 (1958); (b) D. Kessler, B. Mossand, and R. W. Chambers, *Biochem. Biophys. Acta*, **38**, 549 (1960); (c) K. Tanaka, M. Honjo, Y. Sanno, and H. Moriyama, *Chem. Pharm. Bull.* (Tokyo), **10**, 220 (1962); to those employed in method 2 see (d) H. G. Khorana, *J. Am. Chem. Soc.*, **76**, 3517 (1954).