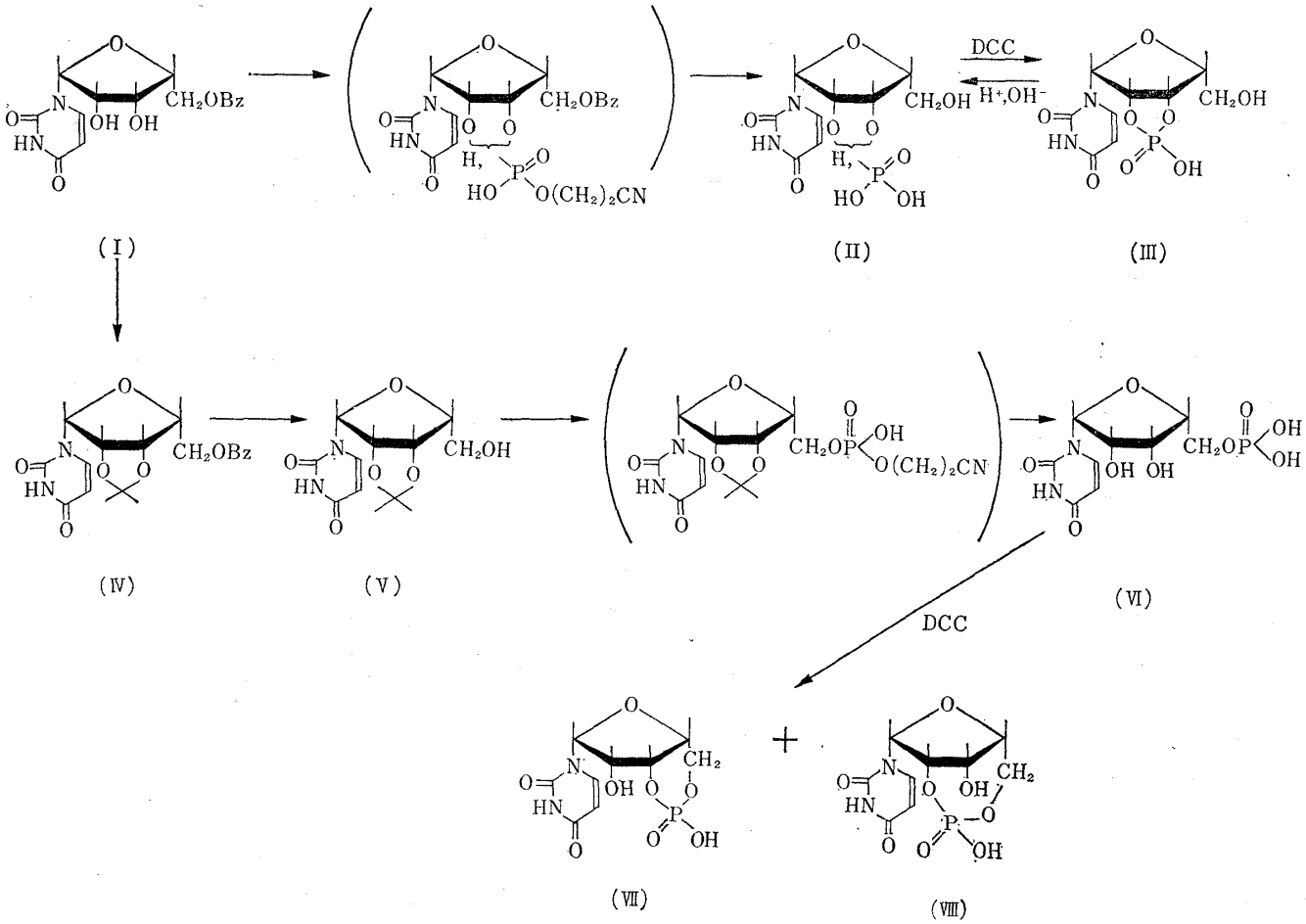
UDC 547.456'118.5 : 547.964

155. Tyunosin Ukita and Hikoya Hayatsu: Organic Phosphates. XVIII.*2,*3 Syntheses of Lyxouridine 2',3'-Cyclic Phosphate and Related Compounds.

(*Faculty of Pharmaceutical Sciences, University of Tokyo**1)

Several investigations on the syntheses of esters of pyrimidine nucleoside 3'-phosphates, pyrimidine nucleoside 2',3'-cyclic phosphates, and their analogs to test their availability as the substrate for bovine pancreatic ribonuclease (RNase) have been reported.[1~3] The present author synthesized methyl α-D-ribofuranoside 2',3'-cyclic phosphate, its β-type isomer, ribothymidine and 5-bromouridine 2',3'-cyclic phosphates, and reported that the last two compounds were hydrolyzed by the RNase,[2] while the former two were inert to this enzyme.[3] Almost all of the above-mentioned researches, however, have been designed in the direction of modifying the structure of the substituent attached at



(I) (II) (III) (IV) (V) (VI) (VII) (VIII)

Chart 1.

*1 Hongo, Tokyo (浮田忠之進, 早津彦哉).
*2 Part XVII. T. Ukita, H. Hayatsu: J. Am. Chem. Soc., in press.
*3 From the thesis of Hikoya Hayatsu for the degree of Doctor of Pharmaceutical Sciences, University of Tokyo.

1) Recent reviews: *a*) H.G. Khorana: "The Enzymes," Vol. V, 79 (1960). Academic Press Inc., New York; *b*) H. Witzel: Ann., **635**, 191 (1960); *c*) C. A. Dekker: Annual Review of Biochemistry, **29**, 453 (1960).
2) Part XI. T. Ukita, M. Irie: This Bulletin, **9**, 217 (1961).
3) Part X. T. Ukita, M. Irie: *Ibid.*, **9**, 211 (1961).

the C-1 position of the sugar moiety of the substrate ribonucleotides.

In the present series of work, 1-β-D-lyxofuranosyluracil 2′,3′-cyclic phosphate (lyxouridine 2′,3′-cyclic phosphate) (III) was prepared which has a structure similar to natural substrate, uridine 2′,3′-cyclic phosphate, except the configuration of the hydroxyl groups at 2′- and 3′-positions of pentafuranose moiety, and which should be a useful compound for further studies on the substrate specificity of RNase-IA.

This paper deals with the chemical synthesis of (III) and several of its isomers necessary for elucidation of the structure of (III).

Recently, Fecher, *et al.*[4] reported the syntheses of the derivatives of lyxouridine, including 1-(5′-O-benzoyl-β-D-lyxofuranosyl)uracil (I). In the present work (I) was phosphorylated with 2-cyanoethyl phosphate[5] in the presence of dicyclohexylcarbodiimide (DCC) and the product was treated with alkali to give lyxouridine 2′(and 3′)-phosphate (II), which was isolated as its barium salt. (II) proved to be homogeneous in paper chromatography and electrophoresis. The spot of this substance showed a similar Rf value and mobility to those of uridine 2′(and 3′)-phosphate, and was negative to periodate spray test.[6]

In the course of the reaction from (I) to (II), during alkali treatment, there is a possibility of migration of the phosphoryl group from 3′-(and/or 2′-) to 5′-position to produce lyxouridine 5′-phosphate*4 (VI), but this possibility was completely excluded by comparison of the Rf value with that of (VI) which was, as will be described below, synthesized and showed a remarkably smaller Rf value than that of (II).

By a general procedure[7] used for the synthesis of ribonucleoside 2′,3′-cyclic phosphate from ribonucleoside 2′(and 3′)-phosphate, (II) was dehydrated with DCC to furnish lyxouridine 2′,3′-cyclic phosphate (III), and (III) was isolated as its barium salt. (III) gave a single spot both in paper chromatography and electrophoresis with Rf value and mobility closely resembling those of uridine 2′,3′-cyclic phosphate. The behavior of (III) to acid and alkali was found to be typical of five-membered cyclic phosphate.[8] (III) was completely hydrolyzed to the parent phosphomonoester (II) by keeping its pH 1 solution for 5 minutes or 0.5*N* sodium hydroxide solution for 10 minutes, at room temperature.

From these results, there can be little doubt about the structure of (III). Differing from uridine, the two hydroxyl groups at 2′- and 3′-positions of lyxouridine are situated in *cis*-configuration to the hydroxymethyl group at 4′-position, and hence, there still remains a possibility of a cyclization of the phosphoryl group to produce lyxouridine 3′,5′-cyclic (VII) and/or lyxouridine 2′,5′-cyclic phosphate (VIII) during dehydration of (II) to (III).

Although this possibility could almost be excluded by the absence of 5′-phosphate (VI) in the hydrolysate of (III),*5 (VII) and (VIII) were prepared via (VI) for further confirmation of (III).*6

*4 1,2-O-Isopropylidene-D-xylofuranose 3-phenyl hydrogenphosphate is known to yield its 3- and 5-phosphate derivatives upon treatment with alkali (J.G. Moffatt, H.G. Khorana: J. Am. Chem. Soc., **79**, 1194 (1957)).

*5 It is known that, on treatment with alkali, 1,2-O-isopropylidene-D-xylofuranose-3,5 cyclic phosphate gives a mixture of its 3- and 5-phosphate derivatives. See ref. in the footnote *4.

*6 To confirm the 2′,3′- cyclic phosphate structure of (III), the simplest way will be the tritylation of this compound to 5′-trityl-2′,3′-cyclic phosphate. Trial of this reaction, however, failed because of the instability of (III) in hydrous pyridine.[7] During conversion of the barium salt of (III) into its pyridinium salt by passing its aqueous solution through a column of Dowex 50 (pyridinium form) resin and subsequent removal of water by codistillation with anhydrous pyridine, a part of (III) was degraded to (II).

4) R. Fecher, J.F. Codington, J.J. Fox: J. Am. Chem. Soc., 83, 1889 (1961).
5) G.M. Tener: *Ibid.*, 83, 159 (1961).
6) J.G. Buchanan, C.A. Dekker, A.G. Long: J. Chem. Soc., **1950**, 3162.
7) M. Smith, J.G. Moffatt, H.G. Khorana: J. Am. Chem. Soc., **80**, 6204 (1958).
8) H.G. Khorana, G.M. Tener, R.S. Wright, J.G. Moffatt: *Ibid.*, **79**, 430 (1957).

On treatment of (I) with acetone and *p*-toluenesulfonic acid, 1-(5′-O-benzoyl-2′,3′-O-isopropylidene-β-D-lyxofuranosyl)uracil*[7] (IV) was obtained. Debenzoylation of (IV) with methanolic sodium methoxide afforded 1-(2′,3′-O-isopropylidene-lyxofuranosyl)uracil (V).

Phosphorylation of (V) with 2-cyanoethyl phosphate and DCC, and subsequent treatment of the product with alkali and acid gave lyxouridine 5′-phosphate (VI), which was isolated as its barium salt in a good yield. (VI) gave a single spot both in paper chromatography and electrophoresis.

(VI) was reacted with DCC in hydrous pyridine for 24 hours at room temperature, according to the method reported by Khorana, *et al.*[8] for the syntheses of propane-1,3-diol and butane-1,4-diol cyclic phosphates. With this reaction condition, (VI) was completely converted into two products, (A) and (B), which gave respective spots with Rf_1 0.49 and 0.41, and showed uridine-type absorption spectra in the ultraviolet range. These spots gave positive reaction to phosphorus spray test[9] and were negative to periodate spray.*[8] In paper electrophoresis at pH 9, they showed a movement typical to phosphodiesters. These properties of the product indicated that (A) and (B) should be represented by a structure none other than lyxouridine cyclic phosphates. Quantitative estimation from their absorption at 261 mμ indicated that (A) and (B) were produced in a ratio of 2.9:1. The products were eluted from sheets of paper-chromatograms and the ammonium salt of (A) was obtained as a powder, that of (B) as colorless needles. Phosphorus content determination of these salts also supported the proposed structures.

An interesting behavior was observed for these products upon acid and alkali hydrolysis. They both resisted hydrolysis under mild reaction conditions sufficient to hydrolyze (III) completely. This behavior was in agreement with the known stability of six- and seven-membered cyclic phosphates.[8] By heating in 0.5*N* hydrochloric acid, (A) was immediately and completely converted to (B), and such a conversion also occurred partially (44%) after one hour's treatment of (A) in hot 0.5*N* sodium hydroxide. The conversion of (A) to (B) by acid and alkali could be explained by an intramolecular alcoholysis of lyxouridine 2′,5′- (VIII) to 3′,5′-cyclic phosphate (VII) or *vice versa*, though which of the (A) and (B) is to be represented by one of these two structures is not confirmed at the present stage of the investigation.

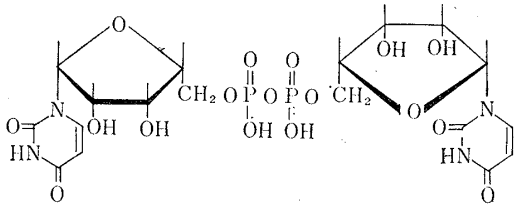
On treatment in a hot 0.5*N* hydrochloric acid solution, (B) was gradually hydrolyzed into (II) and (VI) with concomitant production of uracil. No isomerization between the two hydrolysis products, (II) and (VI), was observed under this condition.

The compound (A) could easily be distinguished from (III) by its different Rf value in paper chromatography and, though the Rf value of (B) is extremely close to that of (III), possibility of the contamination of (B) in (III) can be excluded by the entirely different behavior of these compounds to acid and alkali hydrolysis.

The properties of lyxouridine 2′,3′-cyclic phosphate (III) as a substrate for RNase-IA was reported.[10]

*[7] This compound has been synthesized by Fecher, *et al.*[4] by another route.

*[8] The latter fact excludes the possibility of a pyrophosphate structure like (IX)[8] for (A) and (B).



(IX)

9) R. S. Bandurski, B. Axelrod: J. Biol. Chem., **193**, 405 (1951).
10) T. Ukita, H. Hayatsu, K. Waku: J. Biochem. (Tokyo), **50**, 550 (1961).

Experimental*8

Methods——Paper chromatography was carried out ascendingly on Toyo Roshi No. 53. The following solvent systems were employed: (1) *iso*-PrOH-conc. NH_4OH-H_2O (7:1:2); (2) BuOH-AcOH-H_2O (4:1:5); (3) BuOH-EtOH-H_2O (50:15:35). The Rf value for these solvent systems are represented by Rf_1, Rf_2, and Rf_3, respectively.

Paper electrophoresis was performed by a method described previously[2] using buffer solutions of pH 5.9 and 9.0, and mobilities of the compounds tested are represented by M_1 and M_2, respectively, taking that for uridine 5′-phosphate as standard.

The spots on the paper were located by ultraviolet rays and the P-containing spots detected by a usual method.[9] Total phosphorus content was determined by Allen's method.[11]

Lyxouridine 2′(and 3′)-Phosphate (II)——To a solution of 174 mg. (0.5 mmole) of 1-(5′-O-benzoyl-β-D-lyxofuranosyl)uracil[4] in 5 cc. of pyridine, 1 cc. of hydr. pyridine solution (1 mmole/cc.) of 2-cyanoethyl phosphate[5] was added. The solution was evaporated *in vacuo*, 10 cc. of anhyd. pyridine was added to the residual oil, and the solution concentrated to dryness. The process was repeated, and to a solution of the residue in 5 cc. of pyridine, 0.6 g. of DCC was added, and the mixture was set aside for 66 hr. at room temperature with adequate protection from moisture. After addition of 1.5 cc. of water, the solution was left for 1.5 hr. at room temperature. Pyridine was removed *in vacuo*, the residue was taken up in 5 cc. of water, and filtered to remove dicyclohexylurea, which was washed with 5 cc. of water. The filtrate was combined with the washings, 10 cc. of 1.0*N* NaOH added, and refluxed for 45 min. to hydrolyze the 2-cyanoethyl and benzoyl residues. After cool, the solution was decationized by passing through a column of Dowex-50 (H^+) resin and the column was washed with water until the effluent was neutral (ca. 10 cc.).

The pH of the effluent combined with the washings was adjusted to 7.4～7.8 with aqueous $Ba(OH)_2$ solution and barium phosphate that appeared was separated by centrifugation and washed with 5 cc. of water. The combined supernatant and washings were concentrated *in vacuo* to ca. 10 cc. and two volumes of EtOH was added to the concentrate to precipitate barium lyxouridine 2′ (and 3′)-phosphate.*9 The product was washed with EtOH and Et_2O successively, and dried *in vacuo* over $CaCl_2$. Yield; 189 mg(ca. 65%). The product was homogeneous both in chromatography and electrophoresis, and gave negative reaction with periodate spray.[6] Rf_1, 0.17 (equal value to that for uridine 2′ and 3′-phosphates). Rf_2, 0.24. Rf_3, 0.41. M_1, 1.0.

For analysis, the product was further purified by reprecipitation from H_2O-EtOH. *Anal.* Calcd. for $C_9H_{11}O_9N_2BaP \cdot 7H_2O$: N, 4.78; P, 5.29. Found: N, 4.75; P, 5.62. UV mμ(ε): $\lambda_{max}^{pH\,2}$ 261(8920); $\lambda_{min}^{pH\,2}$ 230 (2320); $\lambda_{max}^{pH\,12}$ 261 (7070); $\lambda_{min}^{pH\,12}$ 241 (4930).

Lyxouridine 2′,3′-Cyclic Phosphate (III)——A solution of 230 mg. of the barium salt of (II) in 3 cc. of water was passed through a column of Dowex-50 (H^+) resin and the decationized effluent was concentrated to dryness *in vacuo*. The residue was taken up in 1.2 cc. of 2*N* NH_4OH and to the solution were added 1.2 cc. of formamide and a solution of 500 mg. of DCC in 3 cc. of *tert*-BuOH. The solution was refluxed for 2.5 hr. After cooling to room temperature and removal of *tert*-BuOH *in vacuo*, 10 cc. of water was added to the residual formamide solution and the mixture was extracted with 5 cc. of Et_2O. Dicyclohexylurea that separated was removed by filtration and the aqueous layer was further extracted twice with Et_2O. The aqueous solution was evaporated first in a reduced pressure and then by suction with an oil pump (bath temperature: 100°). To a solution of the residual glass in 0.5 cc. of water, 15 cc. of Me_2CO was added and the slightly turbid mixture was filtered. A solution of BaI_2 in Me_2CO was added in small portions to the filtrate until the precipitation of barium salt of (III) was complete. The resulting precipitate was collected by centrifugation and washed four times with 15-cc. portions of Me_2CO and finally with 5 cc. of dehyd. Et_2O. The product was dried over P_2O_5 *in vacuo* at room temperature to give 95 mg. (50%) of barium lyxouridine 2′,3′-cyclic phosphate as hexahydrate. The material was homogeneous in paper chromatograpy (Rf_1, 0.40; Rf_2, 0.21; Rf_3, 0.33) and paper electrophoresis (mobility, M_2, 0.90, was equal to that of uridine 2′,3′-cyclic phosphate). *Anal.* Calcd. for $C_9H_{10}O_8N_2Ba_{1/2}P \cdot 6H_2O$: C, 22.43; H, 4.60; N, 5.81; P, 6.43. Found: C, 22.57; H, 4.26; N, 5.66; P, 6.19.

1-(5′-O-Benzoyl-2′,3′-O-isopropylidene-β-D-lyxofuranosyl)uracil (IV)——To a solution of 3.1 g. of *p*-toluenesulfonic acid in 70cc. of dry Me_2CO, 700 mg. (2 mmoles) of 1-(5′-O-benzoyl-β-D-lyxofuranosyl)-uracil[4] was added and the suspension was stirred at room temperature with exclusion of moisture. The reagents gradually went into solution and the solution became clear within 45 min. After 18 hr.'s stirring, 5 cc. of water was added and the solution was neutralized with powdery $NaHCO_3$.

*8 All melting points are uncorrected.

*9 Barium benzoate was removed by this treatment, as it is easily soluble in hydr. EtOH.

11) R. J. L. Allen: Biochem. J., 34, 858 (1940).

The solutinon was filtered and the filtrate was evaporated *in vacuo* to leave a crystalline product, which was collected and washed successively with water, EtOH, and Et_2O. The product weighed 715 mg. (93%) and melted at 245~246°. Recrystallization from EtOH afforded colorless needles melting at 247~248°(reported[4] m.p. 248.5~250°). *Anal.* Calcd. for $C_{19}H_{20}O_7N_2$: C, 58.76; H, 5.19; N, 7.21. Found: C, 58.71; H, 5.16; N, 7.55.

1-(2′,3′-O-Isopropylidene-β-D-lyxofuranosyl)uracil (V)——A solution of 585 mg. of (IV) in 35 cc. of dehyd. MeOH containing 1 mmole of MeONa was refluxed for 3 hr. under anhydrous conditions. The solution was evaporated *in vacuo*, the residue was dissolved in 5 cc. of water, and washed three times with Et_2O. The aqueous solution was passed through a column of Dowex-50 (H^+) resin to remove Na ion. The neutral effluent was evaporated *in vacuo*. By addition of EtOH and repeated evaporation *in vacuo*, crystallization was effected. The product weighed 396 mg. (91.5 %) and melted at 180~182°. Recrystallization from EtOH-Et_2O afforded colorless needles melting at 185~186°. $[\alpha]_D^{32}$ +175°(c=0.515, H_2O). *Anal.* Calcd. for $C_{12}H_{16}O_6N_2$: C, 50.70; H, 5.67; N, 9.86. Found: C, 50.54; H, 5.56; N, 9.90.

Lyxouridine 5′-Phosphate (VI)——A solution of 1 mmole of (V), 2 mmoles of 2-cyanoethyl phosphate, and ca. 6 mmoles of DCC in 10 cc. of dehyd. pyridine was prepared as in the case of the synthesis of (II). The solution was kept at room temperature for 48 hr. After addition of 3 cc. of water, the solution was set aside for 1 hr. and then evaporated to dryness *in vacuo*. The residue was taken up in 10 cc. of water, dicyclohexylurea that appeared was removed by filtration, and the urea was washed with 10 cc. of water. To the combined filtrate and washings 20 cc. of 1.0*N* NaOH was added and the mixture was refluxed for 45 min. to hydrolyze the 2-cyanoethyl residue. After cool, the solution was decationized by passing through a column of Dowex 50 (H^+) resin and the column was washed with ca. 20 cc. of water. The effluent was combined with the washings and the acidic solution (pH 1.4~1.6) was heated on a boiling water bath for 20 min.*10 The pH of the solution was adjusted to 7.4~7.6 with $Ba(OH)_2$, barium phosphate that appeared was removed by centrifugation, and washed with 10 cc. of water. The combined supernatant and washings were concentrated *in vacuo* to ca. 15 cc. and added with two volumes of EtOH to settle the barium salt of (VI), which was washed with EtOH and Et_2O and dried *in vacuo* over $CaCl_2$ to furnish 405 mg. (ca. 80%)of the product. The product was homogeneous both in paper chromatography and electrophoresis, and gave positive reaction to periodate spray test. Rf_1 0.10 (equal to that of uridine 5′-phosphate), Rf_2 0.22, M_1 1.0. Reprecipitation from H_2O-EtOH, and drying *in vacuo* over P_2O_5 at room temperature gave analytically pure material. *Anal.* Calcd. for $C_9H_{11}O_9N_2BaP \cdot 2\frac{1}{2}H_2O$: C, 21.43; H, 3.20; N, 5.55; P, 6.14. Found: C, 20.98; H, 3.23; N, 5.38; P, 6.18. UV mμ(ε): $\lambda_{max}^{pH\,2}$ 261 (9500), $\lambda_{min}^{pH\,2}$ 230 (2170); $\lambda_{max}^{pH\,12}$ 261 (7970), $\lambda_{min}^{pH\,12}$ 246 (5480).

Reaction of Lyxouridine 5′-Phosphate (VI) with DCC——An aqueous solution of 200 mg. of barium lyxouridine 5′-phosphate was passed through a column of Dowex-50 (pyridinium form) resin and the effluent was evaporated *in vacuo* to leave an oily pyridinium salt of (VI). To a solution of the oil in 5 cc. of pyridine containing 10% (v/v) of water, 0.6 g. of DCC was added and the mixture was kept at room temperature for 24 hr. Paper chromatography at this stage revealed that the reaction was complete. The mixture was diluted with 8 cc. of water, dicyclohexylurea which separated was removed by filtration, and the urea was washed with 5 cc. of water. The filtrate was combined with the washings, extracted with three 5-cc. portions of Et_2O, and the aqueous solution was submitted to paper chromatography using seven sheets of Toyo Roshi No. 26 (14×40 cm.) with solvent system 1. Each band with Rf_1 0.49 (compound (A)) and with Rf_1 0.41 (compound (B)) was respectively eluted with water and the aqueous solutions were separately evaporated to dryness in a diminished pressure. (A) was obtained as a highly hygroscopic powdery ammonium salt and (B) as a crystalline ammonium salt (25 mg.). The ammonium salt of (A) was purified by reprecipitation from EtOH-Me_2CO to 37 mg. of slightly hygroscopic powder. The product was homogeneous both in chromatography and electrophoresis. Further purification by reprecipitation gave an amorphous powder which was no longer hygroscopic and the powder was submitted to analysis. The ammonium salt of (B) was purified by recrystallization from EtOH to colorless needles which melted at 242~243°(decomp.). *Anal.* Calcd. for $C_9H_{10}O_8N_2P \cdot NH_4$: P, 9.60. Found (for the ammonium salt of (A)): P, 9.14; (for that of (B)): P, 9.63. Rf_2: (A), 0.25; (B), 0.21. Rf_3: (A), 0.37; (B), 0.31. M_2: (A), 0.90; (B), 0.90.

Both spots of (A) and (B) on the chromatogram were negative to periodate spray test. On periodate oxidation of (A) in a titratable scale, according to the procedure of Khorana, *et al.*,*4 no consumption of the reagent was observed within 3 hr.

Hydrolytic Behavior of the Cyclic Phosphates——Products of the hydrolysis were detected by paper chromatography (solvent system 1). Quantitative estimation was made by extracting the spot

*10 It is known that heating a solution of pH ca. 1.5, at 100° for 10 min. is sufficient to remove the isopropylidene group. See ref. in the footnote *4.

with water and subsequent measurement of optical density of the aqueous extract at 261 mμ. An equal area of the paper free from ultraviolet-absorbing material was extracted with water to use the extract as a blank in the spectrophotometric determination.

(1) Lyxouridine 2′,3′-cyclic phosphate (Ⅲ) : (a) Acid hydrolysis : In a solution of pH 2.0, at room temperature, (Ⅲ) was fairly stable. Thus, at this pH, within 30 min., practically no hydrolysis, and after 33 hr., 40% hydrolysis of (Ⅲ) to (Ⅱ), was observed. At pH 1.0, (Ⅲ) was so labile as to be decomposed completely to (Ⅱ) within 5 min.

(b) Alkali hydrolysis : In 0.5*N* NaOH solution, hydrolysis of (Ⅲ) to (Ⅱ) was essentially complete within 10 min.

(2) Compound (A) : (a) Acid hydrolysis : In a solution of pH 2.0, at room temperature, (A) was stable for 33 hr. At pH 1.0, it was gradually converted to (B); on keeping a solution of (A) at pH 1.0 for 48 hr., 33% of (A) was converted to (B). When a solution of (A) in 0.5*N* HCl was kept on a boiling water bath, (A) was completely isomerized into (B) within 5 min., and, on subsequent additional heating, (B) thus produced was further decomposed into lyxouridine 2′(and/or 3′)-phosphate (Ⅱ), lyxouridine 5′-phosphate (Ⅵ), and uracil.

(b) Alkali hydrolysis : In 0.5*N* NaOH solution, at room temperature, (A) was stable for 24 hr., but, upon heating the solution on a boiling water bath, 44% of (A) was converted to (B) within 1 hr. In this case, however, (Ⅱ) or (Ⅵ) could not be detected on the chromatogram.

(3) Compound (B) : In a solution of pH 1.0, at room temperature, (B) was completely stable for 48 hr. When a solution of (B) in 0.5*N* HCl was heated on a boiling water bath, formation of (Ⅱ), (Ⅵ), and uracil was observed. On heating the solution for 7.5 hr., 79% of (B) was converted to (Ⅱ) (37%), (Ⅵ) (24%), and uracil (18%). Half-life of (B) under this condition was ca. 4 hr. By treatment of (Ⅱ) or (Ⅵ) under the same condition for 7.5 hr., 14% of the former and 38% of the latter was degraded to uracil, without any isomerization between (Ⅱ) and (Ⅵ).

The authors are indebted to Dr. K. Nagasawa of Seikagaku Kenkyusho Co. Ltd., for his valuable discussions and to Dainippon Vitamin Pharmaceutical Co. Ltd. for his gift of the starting materials for the present syntheses. Thanks are also due to Mr. D. Ōhata of the Tokyo Research Laboratory, Yoshitomi Pharm. Ind., Ltd., for carrying out the microanalyses.

Summary

Lyxouridine 2′,3′-cyclic phosphate (Ⅲ), which is a stereoisomer of uridine 2′,3′-cyclic phosphate, a natural substrate of bovine pancreatic ribonuclease, was synthesized. The structure of (Ⅲ) was verified by its comparison with lyxouridine 3′,5′- (Ⅶ) and 2′,5′-cyclic phosphate (Ⅷ) synthesized via lyxouridine 5′-phosphate (Ⅵ).

(Received September 16, 1961)