[Chem. Pharm. Bull. 12 (3) 357 ~ 361]

UDC 547.857'128 : 6123.98.145

53. Takuzo Nishimura and Issei Iwai : Studies on Synthetic Nucleosides. II.*1 Novel Synthesis of Pyrimidine Glucosides.

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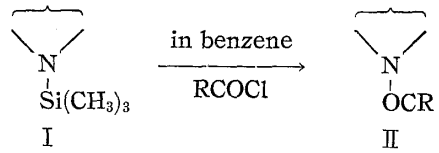
The synthesis of pyrimidine and purine nucleosides has been the subject of numerous investigations for several decades. In 1914, Fischer and Helferich[1] synthesized glucosyladenine by treating the silver salt of 2,8-dichloroadenine with acetobromoglucose in boiling xylene. However, this method was not successfully applied to the syntheses of pyrimidine nucleosides.[2,3] First development in pyrimidine nucleoside was accomplished by Hilbert and Johnson[4] who prepared N-alkyluracil by reaction of 2,4-diethoxypyrimidine with alkylhalide. Subsequently, by the introduction of various acylhalogeno derivatives of hexose and pentose[5~7] to Hilbert-Johnson's method, a number of pyrimidine hexosides and pentosides were synthesized. On the other hand, Davoll, *et al.*[8] further improved Fischer-Helferich's method by using chloromercury salts of purines instead of the corresponding silver salts to prepare adenosine and guanosine. The successful application of this method to the synthesis of pyrimidine nucleosides[9,10] showed it to be the only widely used method to date for the preparation of both pyrimidine and purine nucleosides.

In the hope of development of the simpler method for preparing pyrimidine and purine nucleosides, the authors investigated the reaction of acylhalogeno sugars with trimethylsilyl derivatives of pyrimidine and purine bases which were prepared according to the procedure reported.*2

The present paper deals with the synthesis of pyrimidine glucosides; 1-β-D-glucopyranosyl-uracil, -thymine, and -cytosine.

Birkofer, *et al.*[11] reported that N-acyl derivatives of N-heterocyclic compound (II) were easily obtained from trimethylsilyl-N-heterocyclic compound (I) and acylchloride in nonpolar solvent on account of reactivity of N-Si linkage of these silyl derivatives.



I II

Chart 1.

However an attempt to prepare pyrimidine nucleosides by the interaction of N-trimethylsilylpyrimidines with acylhalogeno sugars failed, because trimethylsilylpyrimidines were not N-silyl but O-silyl derivatives. The fusion of these O-silyl derivatives with acetobromoglucose at 180~195° resulted in 1-N-glucosides of pyrimidines in good yields.

1-β-D-Glucopyranosyluracil (IX)

The preparation of such compound has been previously reported by Hilbert, *et al.*[4]

*1 Part I. T. Nishimura, I. Iwai : This Bulletin, **12**, 352 (1964).

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1) E. Fischer, B. Helferich : Ber., **47**, 210 (1914).
2) E. Fischer : *Ibid.*, **47**, 1377 (1914).
3) P. A. Levene, H. Sobotka : J. Biol. Chem., **65**, 469 (1925).
4) G. E Hilbert, T. B. Johnson : J. Am. Chem. Soc., **52**, 4489 (1930).
5) G. E. Hilbert, E. F. Jansen : *Ibid.*, 58, 60 (1936).
6) G. E. Hilbert : *Ibid.*, **59**, 330 (1937).
7) G. A. Howard, B. Lythgoe, A. R. Todd : J. Chem. Soc., **1947**, 1052.
8) J. Davoll. B. A. Lowy : J. Am. Chem. Soc., **73**, 1650 (1951).
9) J. J. Fox, N. Yung, J. Davoll, G. B. Brown : *Ibid.*, **78**, 2117 (1956).
10) J. J. Fox, N. Yung, I. Wempen, J. L. Doerr : *Ibid.*, **79**, 5060 (1957).
11) L. Birkofer, P. Richter, A. Ritter : Chem. Ber., **93**, 2804 (1960).

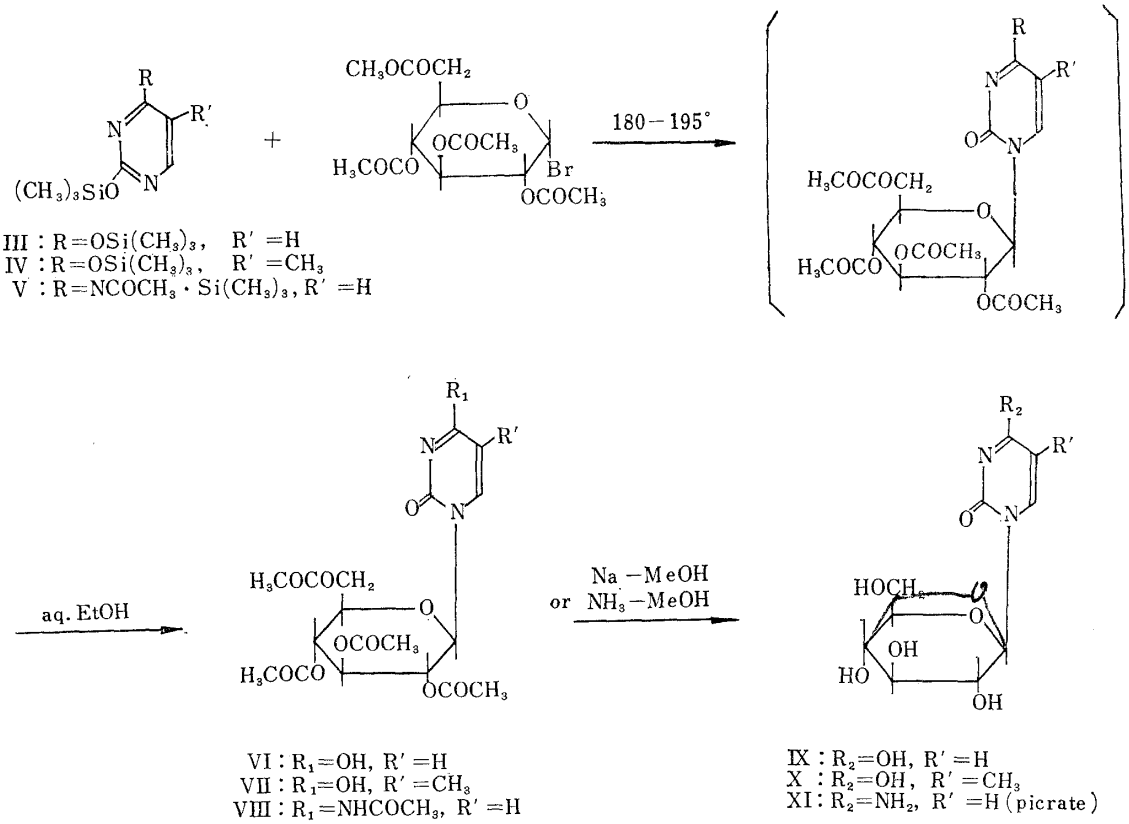
via 4-methoxy-1-tetraacetylglucopyranosyl-2(1*H*)-pyrimidinone by reaction of 2,4-dimethoxypyrimidine with acetobromoglucose. Davoll, *et al.*[12] have shown that thus obtained glucoside has the same configuration as that of uridine in glycosidic center, by the result of periodated oxidation, namely, β-configuration.

Recently, chloromercury method was successfully applied to the synthesis of pyrimidine nucleosides by Fox, *et al.*[10] who prepared glucopyranosyluracil by treating chloromercury-4-ethoxy-2(1*H*)-pyrimidinone with acetobromoglucose in boiling xylene.

The authors prepared 1-(2,3,4,6,-tetra-O-acetyl-β-D-glucopyranosyl) uracil (Ⅵ) by the condensation of tetra-O-acetyl-α-D-glucopyranosyl bromide[13] with O-bis (trimethylsilyl))uracil (Ⅲ) which was obtained by a procedure described in preceding paper. In this case the reactants were simply fused at 180~195° to eliminate trimethylsilyl bromide. After cleavage of second trimethylsilyl group by treating with aqueous ethanol, it was observed that the reaction mixture contained unidentified other by-product as determined by thin layer chromatographic analysis. By treatment of the reaction mixture with ethanol the pure 1-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl)uracil (Ⅵ) was separated in crystalline state in 42% yield.[4] The product was readily deacetylated with sodium methoxide-methanol to give 1-β-D-glucopyranosyluracil (Ⅸ) in 65% yield. The physical constants of nucleoside thus obtained were in good agreement with those of reported nucleoside.[4,12]

1-β-D-Glucopyranosylthymine (X)

When Visser, *et al*[14] treated 5-methyl-2,4-diethoxypyrimidine with acetobromoglucose



III : R=OSi(CH₃)₃, R′ =H
IV : R=OSi(CH₃)₃, R′ =CH₃
V : R=NCOCH₃ · Si(CH₃)₃, R′ =H

VI : R₁=OH, R′ =H
VII : R₁=OH, R′ =CH₃
VIII : R₁=NHCOCH₃, R′ =H

IX : R₂=OH, R′ =H
X : R₂=OH, R′ =CH₃
XI : R₂=NH₂, R′ =H (picrate)

Chart 2.

12) J. Davoll, B. Lythgoe, A.R. Todd : J. Chem. Soc., **1946**, 833.
13) E.C Horning : "Organic Synthesis," Collective Vol. Ⅲ, 11 (1955), John Willey & Sons, Inc., New York.
14) D.W. Visser, I. Goodman, K. Dittmer : J. Am. Chem. Soc., **70**, 1926 (1948).

at 50° for seven days, following the hydrolysis of intermediate with hydrogen chloride in methanol, 1-D-glucosylthymine was obtained in 46% yield. Further, Fox, *et al.*[9] synthesized the same compound *via* 1-(tetra-O-acetyl-β-D-glucopyranosyl)thymine, from dithyminylmercury and acetobromoglucose in boiling toluene. The melting point of the glucoside agreed with that of the sample prepared by Hilbert-Johnson's procedure. They decided the structure of this product to be 1-β-D-glucopyranosylthymine because ultraviolet absorption spectrum was unaffected at various pH values and the configuration at glycosidic center of glucopyranosyluracil analogously produced by Hilbert-Johnson's method would be *beta*.

The crystalline 1-(2,3,4,6-tetra-O-acetyl-β-D-glucopyranosyl)thymine (Ⅶ) was obtained in 52% yield from bis(trimethylsilyl)thymine and α-acetobromoglucose in a same manner as tetraacetylglucosyluracil, m.p. 154.5~155.5°, $[\alpha]_D$ −10.4°, UV : λ_{max} 260 mμ. The product has undepressed melting point on admixture with the sample prepared from dithyminylmercury and acetobromoglucose. Deacetylation of the compound was easily effected to give 1-β-D-glucopyranosylthymine (X) in 83% yield, m.p. 269~271°. The configuration of the product in glycosidic center was established by nuclear magnetic resonance analysis which will be described later.

1-β-D-Glucopyranosylcytosine (XI)

Glucosylcytosine (XI)[15] was synthesized *via* pentaacetylglucosylcytosine (Ⅷ) prepared from bis(trimethylsilyl)-N-acetylcytosine (V) and α-acetobromoglucose by the procedure mentioned above. The thin layer chromatographic analysis showed the presence of considerable amounts of the unidentified by-products in the reaction mixture and the yield of Ⅷ was relatively low. Hilbert, *et al.*[5] synthesized 1-D-glucosylcytosine *via* 1-(tetraacetylglucosyl)-4-ethoxy-1,2-dihydro-2(1*H*)-pyrimidinone. The hydrolysis of nucleoside acetate with ethanol-ammonia afforded solvated crystals of free nucleoside which was identified by its conversion to a known picrate.[5,12]

The physical properties of thus obtained nucleosides and their acetyl derivatives are listed in the Table I.

Table I. Physical Properties and Yields of the Synthetic Pyrimidine Glucosides and their Acetates

Compound	m.p. (°C)	$[\alpha]_D$	UV λ_{max} mμ (log ε)	Yield[a] (%)
Tetraacetylglucosyluracil	149~151	− 9.3 ($CHCl_3$)	254 (4.04)	42
Glucosyluracil	204~206	+21.9 (H_2O)	258 (4.03)	65
Tetraacetylglucosylthymine	154.5~155.5	−10.4 ($CHCl_3$)	261 (4.02)	52
Glucosylthymine	269~271	+18.3 (H_2O)	264 (4.03)	83
Pentaacetylglucosylcytosine	223~225	+43 ($CHCl_3$)	248.5 (4.26)	15
Glucosylcytosine picrate	208~210	—	—	43

a) based on reacted pyrimidines. Melting points are corrected.

Discussion

Acetobromoglucose is relatively unstable against heat, therefore, condensation reaction is to be carried out at lower temperature as much as possible. However our attempt to condense the silylpyrimidines and acetobromoglucose in boiling benzene or xylene was unsuccessful as determined by thin layer chromatographic result and almost quantitative amounts of starting pyrimidine bases being recovered.

15) J.J. Fox, I. Goodman : J. Am. Chem. Soc., 73, 3256 (1951).

On heating the reactants at 180° without solvent, condensation occured and was accompanied with the evolution of trimethylbromosilane (b.p. 82°).

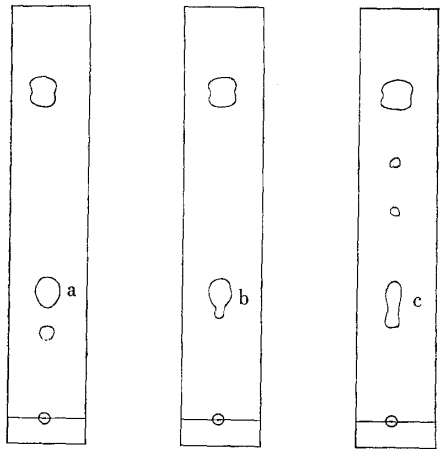


Fig. 1. Thin Layer Chromatograms of the Mixtures of Condensation Reactions

solvent: A, B; AcOEt-$CHCl_3$ (6:4)
C; AcOEt
a : 1-(tetraacetyl-β-D-glucopyranosyl)uracil
b : 1-(tetraacetyl-β-D-glucopyranosyl)thymine
c : 1-(tetraacetyl-β-D-glucopyranosyl)-N-acetylcytosine

Furthermore, it was observed that O-silyl derivatives readily reacted with acetobromoglucose on fusion to give N-glucosides. Ulbricht[16,17] reported that O-alkylpyrimidine readily rearranged to the corresponding N-derivative by heating at 100° with sodium iodide in acetylacetone. Recently Birkofer, *et al.*[18] synthesized 3-N-ribofuranosyluric acid using tetrakistrimethylsilyluric acid and tribenzoylribosyl chloride. From the above results, during the fusion the rearrangement of sugar residue from oxygen to nitrogen atom to form N-glucoside is suggested. The further investigation on this problem are in progress.

After condensation, the treatment of the reaction mixture with aqueous ethanol gave tetraacetylglucosylpyrimidines, which were readily deacetylated with sodium methoxide or ammonia in methanol to afford free glucosylpyrimidines. The preparation of pyrimidine glucosides by this procedure proved to be more convenient than that of Hilbert-Johnson's procedure in which drastic condition is required for hydrolysis.

Experimental

1-(2,3,4,6-Tetra-O-acetyl-β-D-glucopyranosyl)uracil (VI)——Bis(trimethylsilyl)uracil (1.28 g.) was placed in 20 ml. round bottom flask and heated at 170° in dry N_2 stream. Then crystals of freshly prepared α-acetobromoglucose (2.05 g.) were slowly added to the hot syrup during 15 min. After addition of acetobromoglucose the temperature was raised to 185~190°. The additional 15min. heating was continued while the syrupy mixture somewhat darkened. After cooling, the mixture was dissolved in hot 95% EtOH, and treated with charcoal, the solvent was then removed *in vacuo*. To the residue was added $CHCl_3$, and insoluble matter (uracil, 0.17 g.) was filtered off and washed with $CHCl_3$. Filtrate and washings were collected. After removal of solvent, the residue was washed three times with Et_2O. Et_2O insoluble product was crystallized with 99% EtOH. Recrystallization from EtOH gave pure tetraacetylglucosyluracil; m.p. 149~151°(corr.), yield 0.7 g. Analytical sample was recrystallized twice from EtOH. $[\alpha]_D^{28}$ −9.3 (c=1.5, $CHCl_3$). UV : λ_{max}^{EtOH} 254 mμ (log ε 4.04). *Anal.* Calcd. for $C_{18}H_{22}O_{11}N_2 \cdot \frac{1}{2}H_2O$: C, 47.89; H, 5.14; N, 6.21. Found : C, 48.01; H, 4.98; N, 6.70.

1-β-D-Glucopyranosyluracil (IX)——The solution of the acetate (500 mg.) and MeONa (200 mg.) in abs. MeOH was heated under reflux for about 15 min. After cooling, Na ions were removed by addition of wet Dowex 50 (H^+ form). The solution was evaporated to dryness, the residue was dissolved in minimum amounts of H_2O, abs. EtOH was then added. After standing in refrigerator the crystals deposited. Two recrystallization from EtOH gave pure IX, m.p. 204~206°(corr.), yield 200 mg. $[\alpha]_D^{28}$ +21.9°(c=2.43, H_2O). UV : $\lambda_{max}^{H_2O}$ 258 mμ (log ε 4.03). *Anal.* Calcd. for $C_{10}H_{14}O_7N_2 \cdot \frac{1}{2}H_2O$: C, 42.40; H, 5.34; N, 9.89. Found : C, 42.35; H, 5.48; N, 9.87.

1-(2,3,4,6-Tetra-O-acetyl-β-D-glucopyranosyl)thymine (VII)——Bis(trimethylsilyl)thymine (1.4 g.) and α-acetobromoglucose (2.0 g.) was condensed together as in the above experiment. The reaction mixture was cooled and dissolved in hot EtOH. After cooling and filtration, the crude crystals were obtained. Then the crude product was dissolved in $CHCl_3$ and the insoluble matter (thymine, 0.2 g.) was filtered off. After evaporation of $CHCl_3$, crystallization from EtOH gave tetraacetylglucosylthymine as colorless

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17) *Idem* : Angew. Chem. International Edt., **1**, 476 (1962).
18) L. Birkofer, A. Ritter, H. P. Kühlthau : Aagew. Chem., **75**, 209 (1963).

prisms; m.p. 154.5~155.5°(corr.), yield, 0.85 g. Undepressed on admixture with authentic sample which was prepared from dithyminylmercury and α-acetobromoglucose.[9] $[\alpha]_D^{25}$ −10.4°(c=4.84, $CHCl_3$). UV: λ_{max}^{EtOH} 261 mμ (log ε 4.02). *Anal.* Calcd. for $C_{19}H_{24}O_{11}N_2 \cdot \frac{1}{2}H_2O$: C, 49.03; H, 5.41; N, 6.01. Found: C, 48.67; H, 5.39; N, 6.03.

1-β-D-Glucopyranosylthymine (X)——Tetraacetylglucosylthymine (0.5 g.) was treated with MeONa-MeOH by the same manner as described above. The crude product was recrystallized twice from EtOH, m.p. 269~271°(corr.); yield, 0.26 g. $[\alpha]_D^{26}$ +18.3°(c=3.85, H_2O). UV: $\lambda_{max}^{H_2O}$ 264 mμ (log ε 4.03). *Anal.* Calcd. for $C_{11}H_{16}O_7N_2$: C, 45.83; H, 5.60; N, 9.72. Found: C, 45.52; H, 5.64; N, 9.57.

1-(2,3,4,6-Tetra-O-acetyl-β-D-glucopyranosyl)-N-acetylcytosine (VIII)——Bis(trimethylsilyl)-N-acetylcytosine and α-acetobromoglucose was condensed by the similar procedure as described above. The reaction mixture extensively darkened, and was treated with aq. EtOH (charcoal). After evaporation of solvent to dryness, the residue was dissolved in $CHCl_3$ and the insoluble matter (N-acetylcytosine, 0.25 g.) was filtered off. The solution was chromatographed on a column (2.9 cm. internal diam. × 2.0 cm. length) of silica gel. Employing $CHCl_3$ as eluant, 70 ml. fractions were collected. Fractions. 2 to 8 were collected, and evaporated to dryness. The residue was crystallized with EtOH. The crude pentaacetylglucosylcytosine was recrystallized twice from EtOH, yield 0.25 g. Further recrystallization from EtOH gave pure product which melted at 223~225°(corr.). $[\alpha]_D^{28}$ +43°(c=1.51, $CHCl_3$). UV λ_{max}^{EtOH} mμ (log ε): 248.5 (4.26), 297 (3.83). *Anal.* Calcd. for $C_{20}H_{25}O_{11}N_3 \cdot \frac{1}{2}H_2O$: C, 48.79; H, 5.50; N, 8.53. Found: C, 48.92; H, 5.50; N, 8.76.

1-β-D-Glucopyranosylcytosine Picrate (XI)——To the solution of pentaacetylglucosylcytosine (0.65 g.) in 70 ml. of abs. MeOH, dry NH_3 gas was passed at 0°. After saturation with NH_3, the stoppered flask was set aside in refrigerator for 2 days. The mixture was then evaporated to dryness. To the EtOH solution of the residue was added picric acid (0.3 g.) in EtOH. On cooling the picrate was separated which was recrystallized three times from MeOH-EtOH. Glucopyranosylcytosine picrate (0.29 g.) was obtained, m.p. 208~210°(corr.), undepressed on admixture with the sample prepared with previously known procedure.[10,15] *Anal.* Calcd. for $C_{16}H_{18}O_{13}N_6$: C, 38.23; H, 3.61; N. 16.74. Found: C, 38.24; H, 3.86; N, 16.46.

The authors wish to express their gratitude to Mr. M. Matsui, the director of this laboratory, and Professor K. Onodera, Kyoto University. Acknowledgement is also made of valuable advice given by Dr. S. Shimizu during the course of this investigation.

Summary

A new synthetic method is developed for the synthesis of pyrimidine glucosides by the fusion of trimethylsilylpyrimidines and α-acetobromoglucose followed by mild alkaline treatment. 1-β-D-Glucopyranosyl-uracil, -thymine, and -cytosine (picrate) were obtained in good yields. The physical constants of thus obtained nucleosides and their acetyl derivatives show good agreement with those of reported nucleosides.

(Received October 10, 1963)