[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY OF THE OHIO STATE UNIVERSITY]

Acyclic Sugar Nucleoside Analogs[1]

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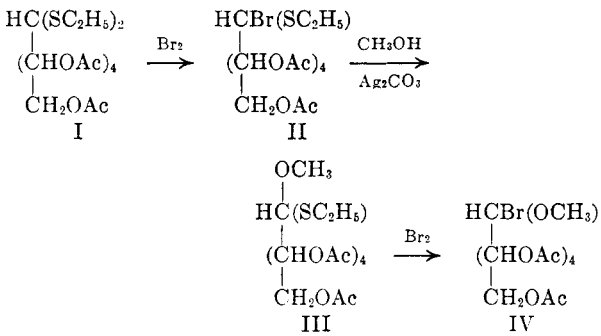
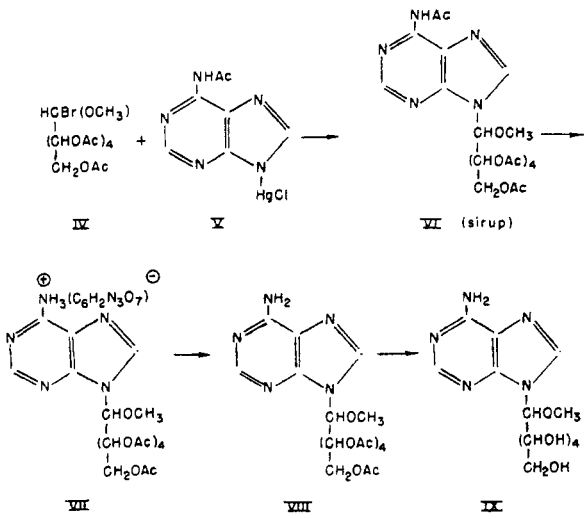
The preparation and characterization of a pair of 1-epimers of 1-(9-adenyl)-1-deoxy-1-*O*-methyl-*aldehydo*-D-galactose aldehydrol (IX) and of 9-β-D-galactopyranosyladenine are described.

The hydroxyl groups of the aldehydrol form of the acyclic aldose acetates readily undergo derivatization[2] to form esters, acetals, hemiacetals, and esters (including halides) of such hemiacetals. Penta-*O*-acetyl-1-chloro-1-deoxy-1-*O*-methyl-*aldehydo*-D-galactose aldehydrol[3] (compare IV), penta-*O*-acetyl-1-*O*-methyl-1-thioethyl-*aldehydo*-D-galactose aldehydrol (III)[3] and penta-*O*-acetyl-1-bromo-1-thioethyl-*aldehydo*-D-galactose aldehydrol (II)[4] were first described by Wolfrom and coworkers, but a better method of synthesis (I → IV) for the bromo analog (IV) is the Gauthier bromination method[5] as adapted by Weygand and associates.[6] Compounds II, III, and IV may exist in C-1 epimeric forms but so far only one of each has been encountered.



We wish to report herein the use of this active bromo derivative of *aldehydo*-D-galactose to prepare acyclic analogs (IX) of a nucleoside. The corresponding 1-chloro derivative[3] was also employed but its use resulted in poorer yields. Penta-*O*-acetyl-1-bromo-1-deoxy-1-*O*-methyl-*aldehydo*-D-galactose aldehydrol (IV) was condensed with 6-acetamido-9-chloromercuripurine (V) by a procedure analogous



to that described by Davoll and Lowy[7] for the synthesis of cyclic nucleosides. Ethanolic picric acid removed the *N*-acetyl group from the sirupy product VI, in a manner similar to the *N*-debenzoylation by Parikh, Wolff, and Burger,[8] resulting in the formation of crystalline penta-*O*-acetyl-1-(9-adenyl picrate)-1-deoxy-1-*O*-methyl-*aldehydo*-D-galactose aldehydrol (VII). Removal of the picrate group gave crystalline penta-*O*-acetyl-1-(9-adenyl)-1-deoxy-1-*O*-methyl-*aldehydo*-D-galactose aldehydrol (VIII). The sharpness of its melting point indicates that this material is probably a single substance. Deacetylation of VIII with a boiling solution of *n*-butylamine in methanol produces two products. This observation leads to the conclusion that the penta-*O*-acetyl-1-(9-adenyl)-1-deoxy-1-*O*-methyl-*aldehydo*-D-galactose aldehydrol is either a mixture of 1-epimers or that a partial inversion of carbon one occurs during the deacetylation resulting in a pair of 1-epimers. We designate these compounds: First Form, m.p. 201–202.5°, $[\alpha]_D^{25}$ +24.4° (water); and Second Form, m.p. 131–134°, $[\alpha]_D^{25}$ −23.6° (water). These substances are a new type of nucleoside analog. They are derivatives of an acyclic hemi-

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(2) M. L. Wolfrom in "Organic Chemistry," H. Gilman, editor, John Wiley and Sons, New York, 2nd edition, 1943, Vol. II, pp. 1575–1581.

(3) M. L. Wolfrom and D. I. Weisblat, *J. Am. Chem. Soc.*, **62**, 878 (1940).

(4) M. L. Wolfrom, D. I. Weisblat, and A. R. Hanze, *J. Am. Chem. Soc.*, **62**, 3246 (1940).

(5) C. Gauthier, *Ann. pharm. franç.*, **12**, 281 (1954).

(6) F. Weygand, H. Ziemann, and H. J. Bestmann, *Chem. Ber.*, **91**, 2534 (1958).

(7) J. Davoll and B. A. Lowy, *J. Am. Chem. Soc.*, **73**, 1650 (1951).

(8) J. R. Parikh, M. E. Wolff, and A. Burger, *J. Am. Chem. Soc.*, **79**, 2778 (1957).

acetal while the nucleosides are derivatives of cyclic hemiacetals.

We also wish to report the synthesis and characterization of 9-β-D-galactopyranosyladenine which was obtained by the condensation of tetra-*O*-acetyl-α-D-galactopyranosyl bromide with 6-acetamido-9-chloromercuripurine (V). The product was purified through 9-(tetra-*O*-acetyl-β-D-galactopyranosyl)-adenine and its picrate, both crystalline substances. Removal of the picrate and acetyl groups resulted in crystalline 9-(β-D-galactopyranosyl)adenine, characterized further by its crystalline picrate.

EXPERIMENTAL

Penta-O-acetyl-1-(9-adenyl picrate)-1-deoxy-1-O-methyl aldehydo-D-galactose aldehydrol (VII). A mixture of 12.7 g. of 6-acetamido-9-chloromercuripurine (V),[9] 14.0 g. of cadmium carbonate, 5 g. of Celite,[10] and 425 ml. of xylene was azeotropically dried by distillation of 100 ml. of the xylene. To this hot suspension, 15 g. of penta-*O*-acetyl-1-bromo-1-deoxy-1-*O*-methyl-*aldehydo*-D-galactose aldehydrol (IV)[6] was added with stirring.[7] The mixture was refluxed 3.5 hr., filtered hot, and the filtrate concentrated to a sirup. The residue and filter cake were extracted with hot chloroform. The chloroform solution was washed with 30% potassium iodide solution, with water, dried with anhydrous sodium sulfate, and concentrated under reduced pressure to a glassy solid (VI); yield 13.5 g. (67%). The corresponding 1-chloro derivative[3] of IV was also employed but its use resulted in much lower yields.

A solution of 4 g. of the glassy product (VI) in 80 ml. of ethanol was boiled with 11 ml. (1.1 moles) of a 10% ethanol solution of picric acid[8] for 1 min. and the solution was cooled. The resulting yellow precipitate was filtered and washed with a little absolute ethanol; yield 3.77 g. (71%). This material was chromatographed, in two 0.7-g. portions, on a column[11] (100 × 44 mm.) of Micro-Cel C.[10] Development with 150 ml. of benzene-ethanol (5:1 by vol.), produced one main yellow zone. The sectioned zone was eluted with acetone and the product obtained on solvent removal was crystallized from ethanol by evaporation at room temperature to produce bright yellow crystals; yield 250 mg. of penta-*O*-acetyl-1-(9-adenyl picrate)-1-deoxy-1-*O*-methyl-*aldehydo*-D-galactose aldehydrol (VII), dec. 287–290°.

Anal. Calcd. for $C_{28}H_{32}N_8O_{18}$: N, 14.57. Found: N, 15.61.

Penta-O-acetyl-1-(9-adenyl)-1-deoxy-1-O-methyl-aldehydo-D-galactose aldehydrol (VIII). A solution of 2.4 g. of penta-*O*-acetyl-1-(9-adenyl picrate)-1-deoxy-1-*O*-methyl-*aldehydo*-D-galactose aldehydrol (VII) in 500 ml. of 50% aqueous acetone was passed through a 30 × 420 mm. column of Dowex-1 (carbonate form) anion exchange resin[12] and the column was washed with 700 ml. of the solvent. The solution was evaporated to 350 ml. and extracted thrice with 75 ml. of chloroform. The extract was dried with anhydrous sodium sulfate and concentrated to a sirup. The material crystallized from absolute ethanol; yield 1.11 g. Two recrystallizations from 95% ethanol produced fine needles, m.p. 191–192°, $[\alpha]_D^{25}$ −25° (*c*, 0.8 chloroform); absorption spectra data[13]: $\lambda_{max}^{C_2H_5OH}$ 260 mμ, λ_{max}^{KBr} 3.00, 3.15 μ (NH_2, NH), 5.70 μ (ester carbonyl), 5.98, 6.10, 6.22, 6.80 μ (NH_2, NH, and purine ring); 7.30 μ (methyl hydrogen); 8.22–8.38 μ (broad C—O—C of acetates), 9.70 μ (C—O—C); x-ray powder diffraction data[14]: 13.55 m, 12.22 w, 9.21 m, 7.08 s (2), 6.77 m, 6.08 vs (1), 5.61 vw, 5.34 s (3), 5.06 m, 4.87 vw, 4.71 vw, 4.61 vw, 4.39 m, 4.20 vw, 4.00 s, 3.87 vw, 3.73 vw, 3.62 vw, 3.51 m, 3.36 m, 3.22 vw, 3.12 w, 3.06 w.

Anal. Calcd. for $C_{22}H_{29}N_5O_{11}$: C, 48.97; H, 5.42; N, 12.98. Found: C, 48.42; H, 5.42; N, 12.71.

1-(9-Adenyl)-1-deoxy-1-O-methyl-aldehydo-D-galactose aldehydrol, first form (IX). Penta-*O*-acetyl-1-(9-adenyl)-1-deoxy-1-*O*-methyl-*aldehydo*-D-galactose aldehydrol (VIII, 1.3 g.) was refluxed in 60 ml. of absolute ethanol containing 5 ml. of *n*-butylamine.[15,16] The solution was concentrated to a sirup by repeated evaporation, under reduced pressure, of its absolute methanolic solution. The dry residue was extracted with chloroform to remove unreacted material, and dissolved in water, decolorized with carbon, evaporated under reduced pressure, and dried by repeated evaporation of its methanolic solution. The dried solid was triturated with warm ethanol and filtered. The filtrate upon evaporation produced a solid; yield 530 mg. (67%). Recrystallization from methanol produced 1-(9-adenyl)-1-deoxy-1-*O*-methyl-*aldehydo*-D-galactose aldehydrol (IX), First Form; yield 360 mg., m.p. 201–202.5°, $[\alpha]_D^{25}$ + 24.4° (*c*, 0.5 water); absorption spectra data[13]: $\lambda_{max}^{H_2O}$ 260 mμ; λ_{max}^{KBr} 2.85, 2.95, 3.10 μ (OH, NH), 6.00, 6.10, 6.20, 6.36, 6.78 μ (NH_2, NH, and purine ring); 7.30 μ (methyl hydrogen); 8.34 μ (C—O—C of methyl ether); 8.90, 9.35, 9.70 μ (C—O—C, C—OH); x-ray powder diffraction data[14]: 7.23 s, 6.09 m, 5.66 m, 5.13 s, 4.83 m, 4.48 vs (1), 4.30 w, 4.12 vw, 3.93 vw, 3.64 s (2), 3.54 m, 3.47 m, 3.38 vw, 3.28 vs (3), 3.16 vw, 2.93 vw, 2.84 vw, 2.65 w, 2.58 w, 2.47 vw, 2.39 vw, 2.32 w.

Anal. Calcd. for $C_{12}H_{19}N_5O_6$: C, 43.77; H, 5.82; N, 21.27; OCH_3, 9.43. Found: C, 43.99; H, 5.77; N, 21.69; OCH_3, 10.18.

These data indicate the substance to the higher rotating epimer of 1-(9-adenyl)-1-deoxy-1-*O*-methyl-*aldehydo*-D-galactose aldehydrol (IX) which we designate the First Form.

1-(9-Adenyl)-1-deoxy-1-O-methyl-aldehydo-D-galactose aldehydrol, second form (IX). Penta-*O*-acetyl-1-(9-adenyl)-1-deoxy-1-*O*-methyl-*aldehydo*-D-galactose aldehydrol (VIII, 1.1 g.) was deacetylated as described above in the preparation of the First Form. The dry residue was redissolved in methanol, decolorized with carbon, and the colorless filtrate was allowed to evaporate at room temperature. The initial fraction of crystals was separated; yield 200 mg. (30%). Recrystallization from ethanol gave 1-(9-adenyl)-1-deoxy-1-*O*-methyl-*aldehydo*-D-galactose aldehydrol (IX), Second Form, m.p. 131–134°, $[\alpha]_D^{25}$ −23.6° (*c*, 0.6 water); absorption spectra data[13]: $\lambda_{max}^{H_2O}$ 260 mμ; λ_{max}^{KBr} 2.92, 3.10 μ (OH, NH); 6.00, 6.10, 6.35, 6.75 μ (NH_2, NH, and purine ring); 7.25 μ (methyl hydrogen); 8.30 μ (C—O—C); 9.10, 9.40, 9.70, 9.82 μ (C—O—C, C—OH); x-ray powder diffraction data[14]: 12.95 vw, 7.90 m, 7.25 s, 6.99 m, 6.43 m, 6.04 m, 5.75 vw, 5.58 w, 5.28 w, 4.98 m, 4.66 w, 4.40 w, 4.09 m, 3.92 vs (1), 3.80 s, 3.67 w, 3.53 vs (2), 3.36 vs (3), 3.24 vw.

Anal. Calcd. for $C_{12}H_{19}N_5O_6$: C, 43.77; H, 5.82; N, 21.27; OCH_3, 9.43. Found: C, 43.55; H, 5.93; N, 21.09; OCH_3, 9.10,

(9) B. R. Baker, Kathleen Hewson, H. Jeanette Thomas, and J. A. Johnson, Jr., *J. Org. Chem.*, **22**, 954 (1957).

(10) A product of Johns-Manville Co., New York, N. Y.

(11) W. H. McNeely, W. W. Binkley, and M. L. Wolfrom, *J. Am. Chem. Soc.*, **67**, 527 (1945).

(12) A product of the Dow Chemical Co., Midland, Mich.

(13) The ultraviolet absorption analyses were made on a Cary Recording Spectrophotometer, Model 10, Applied Physics Corp., Pasadena, Calif. The infrared spectral data were obtained with an Infrared Recording Spectrophotometer, Model B, Baird Associates, Inc., Cambridge, Mass. Structural assignments were made following W. B. Neely, *Advances in Carbohydrate Chem.*, **12**, 13 (1957) and B. R. Baker and associates.[9]

(14) Interplanar spacing, Å, CuKα radiation. Relative intensity, estimated visually: s, strong; m, medium; w, weak; v, very; three strongest lines numbered in order of increasing strength.

(15) L. Goldman, J. W. Marsico, and R. B. Angier, *J. Am. Chem. Soc.*, **78**, 4173 (1956).

(16) E. J. Reist and B. R. Baker, *J. Org. Chem.*, **23**, 1083 (1958).

These data indicate that this substance is the lower rotating 1-epimer of 1-(9-adenyl)-1-deoxy-1-*O*-methyl-*aldehydo*-D-galactose aldehydrol (IX), herein designated as the Second Form.

9-(Tetra-O-acetyl-β-D-galactopyranosyl)adenine picrate. A mixture of 28.0 g. of 6-acetamido-9-chloromercuripurine (V),[9] 28.0 g. of cadmium carbonate, 5 g. of Celite,[10] and 425 ml. of xylene was dried azeotropically by distillation of 100 ml. of the xylene. To this mixture was added, at 90° and with stirring, a solution of 28.0 g. of tetra-*O*-acetyl-α-D-galactopyranosyl bromide[17] in 30 ml. of xylene. The mixture was refluxed for 3 hr., essentially according to the procedure of Davoll and Lowy,[7] filtered hot, and the filtrate was concentrated under reduced pressure. The filter cake and residue were extracted with hot chloroform, the chloroform solution was washed with 30% aqueous potassium iodide, water, dried with anhydrous sodium sulfate, and concentrated under reduced pressure to a sirup; yield 24.0 g. of crude 6-acetamido-9-(tetra-*O*-acetyl-β-D-galactopyranosyl)purine. The crude product (13.1 g.) was converted to the picrate by boiling for 1 min. with 67 ml. of 10% picric acid in ethanol.[8] The picrate of 9-(tetra-*O*-acetyl-β-D-galactopyranosyl)adenine readily crystallized upon cooling; yield 12.4 g. (63%). The material was further purified by column chromatography[11] by placing 0.8-g. portions of material on (100 × 44 mm.) columns of Micro-Cel C[10] using 150 ml. of benzene-ethanol (5:1 by vol.) as developer. The material in the bright yellow zone was eluted with acetone and crystallized from ethanol as deep yellow needles, dec. 284–287°.

Anal. Calcd. for $C_{25}H_{26}N_8O_{16}$: N, 16.14. Found: N, 15.90.

9-(Tetra-O-acetyl-β-D-galactopyranosyl)adenine. A mixture of 9-(tetra-*O*-acetyl-β-D-galactopyranosyl)adenine picrate (7.4 g.) was converted to the nucleoside by treatment with Dowex-1 as described above for the acyclic derivative. The resulting sirup (3.7 g., 75%) was concentrated under reduced pressure twice from ethanol, dissolved in 25 ml. of ethanol, and allowed to evaporate from an open beaker at room temperature. Crystallization occurred in 24 hr. The material was filtered and washed with cold methanol; yield 1.13 g. (26%) of 9-(tetra-*O*-acetyl-β-D-galactopyranosyl)adenine. Recrystallization from methanol produced fine white needles; m.p. 212–213.5°, $[\alpha]_D^{24}$ +7.3° (*c*, 2.6 chloroform); absorption spectra data[13]: $\lambda_{max}^{C_2H_5OH}$ 248 mμ, λ_{max}^{KBr} 2.88, 3.05 μ (NH, NH_2), 5.65 μ (C=O of acetates), 5.95, 6.15, 6.70, 6.96 μ (NH_2 and purine ring), 7.28 μ (methyl hydrogens), 8.05–8.22, 9.15, 9.45 μ (C—O—C); x-ray powder diffraction data[14]: 11.07 vs (1), 9.72 vw, 8.69 vw, 7.86 m, 6.76 m, 6.38 w, 6.04 w, 5.65 m, 5.10 s (2), 4.82 m (3), 4.31 m, 4.19 m, 3.94 w, 3.85 vw.

Anal. Calcd. for $C_{19}H_{23}N_5O_9$: C, 49.02; H, 4.98; N, 15.15. Found: C, 48.95; H, 4.40; N, 15.52.

9-β-D-Galactopyranosyladenine. A solution of 24.0 g. of crude 6-acetamido-9-(tetra-*O*-acetyl-β-D-galactopyranosyl)-purine in 225 ml. of dry methanol and 35 ml. of *n*-butylamine was refluxed for 5 hr. The solution was concentrated to 50 ml. under reduced pressure and held at 0° until precipitation occurred. The precipitate was filtered and washed with cold methanol, chloroform, and ether; yield 9.6 g. (67.5%). The solid was dissolved in water, decolorized with carbon and concentrated to a sirup which crystallized. The mass was triturated with methanol, filtered, and dried; m.p. 192–195°. Further purification was effected by treatment with hot 10% ethanolic picric acid to form a crystalline 9-(β-D-galactopyranosyl)adenine picrate; m.p. 230–232°.

Anal. Calcd. for $C_{17}H_{18}N_8O_{12}$: C, 38.79; H, 3.45; N, 21.29. Found: C, 38.83; H, 3.65; N, 19.98.

Regeneration of the nucleoside from the picrate with Dowex-1 (carbonate form)[12] anion exchange resin by the method described above for the acyclic picrate derivative gave fine colorless needles, m.p. 198–200°, $[\alpha]_D^{23}$ +95.5° (*c*, 0.5 water); absorption spectra data[13]: $\lambda_{max}^{H_2O}$ 260 mμ; λ_{max}^{KBr} 2.90, 2.98 μ (OH, NH), 6.02, 6.24, 6.32, 6.78 μ (NH_2, NH, and purine ring), 9.20–9.50, 10.0 μ (C—O—C, C—OH); x-ray powder diffraction data[14]: 7.65 w, 7.00 vw, 6.59 m, 6.35 m, 5.88 w, 5.20 vw, 4.88 vs (1), 4.60 w, 4.32 vw, 4.18 w, 4.08 vw, 3.97 w, 3.60 m, 3.50 m (3), 3.42 w, 3.36 w, 3.26 s, (2), 3.17 w, 3.07 vw, 2.94 w, 2.89 w.

Anal. Calcd. for $C_{11}H_{15}N_5O_5$: C, 44.43; H, 5.08; N, 23.54. Found: C, 43.85; H, 5.36; N, 23.31.

Identical material was also obtained on deacetylation, with boiling methanolic *n*-butylamine, of 9-(tetra-*O*-acetyl-β-D-galactopyranosyl)adenine by the same procedure used to deacetylate VIII.

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(17) H. Ohle, W. Marecek, and W. Bourjau, *Ber.*, **62**, 833 (1929).

[CONTRIBUTION FROM THE DEPARTMENT OF AGRICULTURAL BIOCHEMISTRY, HAWAII AGRICULTURAL EXPERIMENT STATION UNIVERSITY OF HAWAII]

The Constitution of a Galactomannan from the Seed of *Leucaena glauca*[1]

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A galactomannan, composed of 57% mannose and 43% galactose, was isolated in 25% yield from the seed of *Leucaena glauca*. The polysaccharide had an average D.P. of 150 and gave highly viscous aqueous solutions at low solute concentrations. Periodate degradation revealed that some mannose residues and a smaller number of galactose residues were not attacked. Hydrolysis of the methylated gum showed that galactose occupied mainly terminal, nonreducing positions and the considerable quantity of 2,3,4,6-tetra-*O*-methyl-D-mannose found indicated the presence of some major branch-points. Further evidence that some 1,3-linkages were present was obtained.

DISCUSSION

The seed of *Leucaena glauca* (better known as koa hoale in the Hawaiian Islands) has an interesting chemical composition. Among the nitrogenous constituents found in the seed, a poisonous amino acid known as mimosine[3] is present in relatively large quantities. The isolation of this amino acid which

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