[CONTRIBUTION FROM THE BUREAU OF CHEMISTRY AND SOILS, U. S. DEPARTMENT OF AGRICULTURE]

Synthetic Nucleosides—Some 1-Glycosidouracils

BY GUIDO E. HILBERT

Of the two known methods for synthesizing pyrimidine nucleosides, the one most recently introduced,[1] involving the interaction of 2,4-dialkoxypyrimidines and acetobromosugars, has the distinct advantage of actually introducing the sugar into the pyrimidine ring at the 1-position, which is that occupied by ribose in uridine and cytidine. Unfortunately the method seemed not to be as general as first expected. For example, two other types of pyrimidines, namely, 2-methoxy-4-amino-[2] and 2,4-dimethoxy-5-methyl-pyrimidine[3] formed in the presence of acetobromoglucose only methylated derivatives. These results suggest that the success of the method is dependent to a marked extent upon the nature of the substituents attached to the pyrimidine cycle. The following investigation was initiated (a) to test the generality of the reaction between various acetobromosugars and 2,4-diethoxypyrimidine and (b) with the hope of obtaining compounds similar in constitution to the anomalous 4-ethoxy-2-triacetyl-*d*-ribosidopyrimidine (I) obtained as a by-product in the interaction of acetobromo-*d*-ribose and 2,4-diethoxypyrimidine.[4] If such a product could be formed from an easily accessible sugar, there would be no difficulty in the way of obtaining sufficient material for a proof of structure.

Four different sugar derivatives, namely, acetobromo-*d*-galactose, acetobromo-*d*-xylose, acetobromo-*l*-arabinose and acetobromo-*d*-mannose, were used in this work. Each of the first three gave with 2,4-diethoxypyrimidine compounds of the type (II); in none of these reactions could crystal-

(II) Acetyl sugar —N(C(=O)—N=C(COC$_2$H$_5$)—C(H)=C(H))

line analogs of the labile 4-ethoxy-2-triacetyl-*d*-ribosidopyrimidine (I)[4] be isolated. Considering the number of side reactions that are possible (see below), the yields of the 1,2-dihydro-2-keto-4-ethoxy-1-acetylglycosidopyrimidines (II) are quite satisfactory. The products (II) were converted by simultaneous deacetylation and deëthylation with alcoholic hydrogen chloride into the corresponding 1-glycosodouracils, which, like uridine, neither respond to the Wheeler-Johnson color test nor reduce Fehling's solution after acid treatment. It is interesting that the pentose-pyranosidouracils melt considerably higher, are thermally more stable and are much less soluble in water than uridine and the hexose–pyranosidouracils. Evidently the presence of a carbinol side group on a sugar ring affects significantly the properties of the nucleosides.

Of the acetobromosugars investigated, acetobromo-*d*-mannose, because of the similarity of its chemical reactions with those of acetobromo-*d*-ribose ("orthoacetate" formation[5]), was considered to offer the most promise of producing an analog of (I). Unfortunately, the interaction of acetobromo-*d*-mannose and 2,4-diethoxypyrimidine gave a sirup and the lability of the desired product precluded fractionation of the sirup to remove by-products. In view of the sluggishness with which most mannose derivatives crystallize and the complexity of the reaction product, this result was not altogether unexpected. However, presumptive evidence in favor of the formation of an analog of (I) in this reaction was obtained in the following manner. When the sirup was allowed to stand for a considerable length of time, a crystalline material gradually deposited. After separation and purification it proved to be 1,2-dihydro-2-keto-4-ethoxypyrimidine,[6] which had been obtained previously from the mild alkaline hydrolysis of (I). Presumably this was formed by the slow hydrolysis of 4-ethoxy-2-tetraacetyl-*d*-mannosidopyrimidine. Since the formation of a pyrimidine substituted in the 1-position by mannose was of less immediate interest, no effort was made to prepare 1-*d*-mannosidouracil by using a procedure similar to that employed for the isolation of 1-*d*-ribosidouracil.[4]

The 1,2-dihydro-2-keto-4-ethoxy-1-acetylglycosidopyrimidines (II) represented only a minor fraction of the product formed by the interaction

(1) Hilbert and Johnson, THIS JOURNAL, **52**, 4489 (1930).
(2) Hilbert, *ibid.*, **56**, 190 (1934).
(3) Schmidt-Nickels and Johnson, *ibid.*, **52**, 4511 (1930).
(4) Hilbert and Rist, *J. Biol. Chem.*, in press.
(5) Levene and Tipson, *ibid.*, **92**, 109 (1931).
(6) This was also a secondary product of the interaction between acetobromo-*d*-galactose and 2,4-diethoxypyrimidine.

of 2,4-diethoxypyrimidine and acetobromosugars. Considerable ethyl bromide was formed, presumably by the degradation of the intermediate pyrimidinium bromide, and interacted competitively with 2,4-diethoxypyrimidine to give 1,2-dihydro-2-keto-1-ethyl-4-ethoxypyrimidine. This was separated readily from the other products of the reaction and on treatment with hydrogen chloride gave 1-ethyluracil quantitatively. A previous attempt to carry out the graded ethylation of 2,4-diethoxypyrimidine with ethyl iodide[7] was unsuccessful and gave only the end-product, 1,3-diethyluracil. In a few reactions, the by-products, 1,2-dihydro-2-keto-4-ethoxypyrimidine, mentioned above, and uracil, in traces, were also formed. Little is known regarding the other and major side product or products. In one instance, a mixture resulting from the interaction of acetobromo-*d*-glucose with 2,4-diethoxypyrimidine was fractionated and the material that did not crystallize distilled in a high vacuum (0.0001 mm.). Analysis of the distillate which could not be made to crystallize indicates that it is a tetraacetylglucosidoethoxypyrimidine.

From the preceding results and those previously published on 2,4-diethoxypyrimidine, it may be concluded that, although the reaction with acetobromoglycosides is quite complex, a certain amount of the sugar will enter the 1-position of this particular type of pyrimidine. This is of interest because, of the various pyrimidines that could be studied in the above type of reaction, the 2,4-diethoxy derivative is probably the most important, since its use can be directed not only to the preparation of analogs of uridine but also indirectly to those of cytidine.[8] As the synthesis of this type of compound seems to be independent of the kind of acetobromoglycoside used, it is believed that the synthesis of the naturally occurring pyrimidine nucleosides now resolves itself primarily into a sugar problem.

I wish to express my appreciation to Dr. R. T. Milner and Mrs. M. S. Sherman for performing the microanalyses recorded.

Experimental

Acetobromo-*d*-xylose,[9] acetobromo-*d*-mannose and acetobromo-*d*-galactose were prepared from the acetates by the method of Levene and Raymond.[10] Acetobromo-*l*-arabinose[11] was prepared directly from *l*-arabinose according to Hudson's[12] modification of the method of Chavanne.[13] All the acetobromoglycosides used in this work were recrystallized several times, generally from dry ether–petroleum ether. Acetobromo-*d*-galactose, which has rarely been obtained crystalline, melted at 87–88°[14,15]; $[\alpha]^{21}D$ +216° (*c* = 7.44 in U. S. P. chloroform).

The reaction between 2,4-diethoxypyrimidine and the acetobromoglycosides was carried out in an oven (temperature control). Since considerable ethyl bromide is evolved in the reaction it is desirable to connect the flask containing the reactants with a glass tube leading to the outside atmosphere; the portion projecting from the furnace was fitted with a tube containing a drying agent.

1,2 - Dihydro - 2 - keto - 4 - ethoxy - 1 - triacetyl - *d* - xylosidopyrimidine.—A mixture of acetobromo-*d*-xylose (38 g.) and 2,4-diethoxypyrimidine (38 g.) was heated at 65° for eighteen hours. After thirty minutes there was complete solution and in two hours large colorless plates started to separate. The final reaction mixture consisted of a mush of crystals in a pale yellow sirup. It was treated with 15 cc. of dry ether, filtered and the solid washed with ether; yield, 14 g. (31% of the theoretical yield based on the acetobromo-*d*-xylose or 16% based on 2,4-diethoxypyrimidine). The product was recrystallized from 150 cc. of 95% ethyl alcohol and deposited as needles on rapid cooling; m. p. 218° resolidifying at 200°; $[\alpha]^{22}D$ 58.4° (*c* = 5.14 in U. S. P. chloroform). This xyloside was very soluble in cold chloroform, soluble in hot alcohol, slightly soluble in ether and sparingly soluble in water.

Anal. Calcd. for $C_{17}H_{22}O_9N_2$: C, 51.23; H, 5.57; N, 7.04; OC_2H_5, 11.31. Found: C, 51.35; H, 5.51; N, 6.96; OC_2H_5, 11.24.

The original sirup from which the nucleoside had been filtered, after standing for several hours, deposited an additional small quantity of the nucleoside, which was contaminated with a finely divided material. The latter was separated mechanically; it did not melt at 300° and was probably uracil.

1,2 - Dihydro - 2 - keto - 4 - ethoxy - 1 - *d* - xylosidopyrimidine.—A solution of 3 g. of 1,2-dihydro-2-keto-4-ethoxy-1-triacetyl-*d*-xylosidopyrimidine in 50 cc. of warm absolute ethyl alcohol was treated with 20 cc. of alcohol containing 2 cc. of liquid ammonia. On cooling the solution to room temperature, the acetyl compound separated but redissolved gradually within several hours. After two days, the solution was concentrated under diminished pressure; the resulting colorless residue rapidly crystallized when agitated. It was recrystallized from 50 cc. of absolute ethyl alcohol and separated as large stout rods; the yield was practically quantitative. The product sintered at 206° and melted at 208°; it resolidified on

(7) Hilbert and Johnson, THIS JOURNAL, **52**, 2001 (1930).

(8) Hilbert and Jansen, *ibid.*, **58**, 60 (1936).

(9) This was prepared from *d*-xylose which was kindly donated by W. E. Emery, Chief, Organic and Fibrous Materials Division, Bureau of Standards.

(10) Levene and Raymond, *J. Biol. Chem.*, **90**, 247 (1931).

(11) *l*-Arabinose was a gift from Professor J. J. Donleavy of Yale University.

(12) Hudson, Sci. Papers Bur. of Stand. (U. S. Dept. of Commerce) No. 533, 350 (1926).

(13) Chavanne, *Compt. rend.*, **134**, 661 (1902).

(14) All melting points are corrected.

(15) Fischer and Armstrong [*Ber.*, **35**, 838 (1902)] report 82–83°; and Ohle, Marecek and Bourjau [*ibid.*, **62**, 846 (1929)] report 85°.

cooling and then melted again at 208°; $[\alpha]^{23}D$ +47.9° (c = 4.64 in distilled water). This xylosido derivative is very soluble in cold water, slightly soluble in hot alcohol and insoluble in non-polar solvents.

Anal. Calcd. for $C_{11}H_{16}O_6N_2$: C, 48.50; H, 5.93; N, 10.29; OC_2H_5, 16.55. Found: C, 48.58; H, 5.83; N, 10.39; OC_2H_5, 16.17.

1-*d*-Xylosidouracil.—To a solution of 4 g. of 1,2-dihydro-2-keto-4-ethoxy-1-triacetyl-*d*-xylosidopyrimidine in 65 cc. of hot absolute methanol was added 10 cc. of methanol containing 26% by weight of hydrogen chloride. After standing for three days, the clear solution was concentrated under diminished pressure and the residual sirup rapidly crystallized. The solid cake was broken up, triturated with alcohol and filtered; yield 2.35 g. It was recrystallized from 60 cc. of 90% ethyl alcohol, separating as distorted octahedra, which were anhydrous; sintered 243°, m. p. 245°; $[\alpha]^{24}D$ +21.8° (c = 1.46 in distilled water). On another occasion it separated as long silky needles containing 8.5% solvent of crystallization (slowly removed at the boiling point of xylene *in vacuo*). 1-*d*-Xylosidouracil is much less soluble in water than other pyrimidine nucleosides; about 30 cc. of water was required to dissolve 1 g. at room temperature.

Anal. Calcd. for $C_9H_{12}O_6N_2$: C, 44.24; H, 4.95; N, 11.48. Found: C, 44.42; H, 4.74; N, 11.39.

1,2-Dihydro-2-keto-4-ethoxy-1-tetraacetyl-*d*-galactosidopyrimidine.—A solution of 40 g. of acetobromo-*d*-galactose and 40 g. of 2,4-diethoxypyrimidine was heated at 65° for twenty-one hours. The reaction mixture consisted of a pale brown sirup and a small amount (0.1 g.) of uracil. After adding an equal volume of dry ether, it was filtered and the filtrate placed in the ice-chest overnight. A crystalline cake separated, which was broken up, filtered and washed with ether; yield 16.7 g. (the yield based on acetobromo-*d*-galactose was 37%, or based on 2,4-diethoxypyrimidine 15% of the theoretical). It was recrystallized from 95% ethyl alcohol and separated as plates which when desolvated sintered at 156° and melted at 159°; $[\alpha]^{21}D$ +59.2° (c = 5.53 in U. S. P. chloroform). The amount of solvent of crystallization present in the crystals varied with the conditions of crystallization; for example, the loss in weight of air-dried specimens obtained from three different crystallizations was 5.0, 5.6 and 6.3%. The ethoxyl content of the air-dried specimen, which lost 5.6% by weight when heated *in vacuo*, was determined by analysis; found, 12.0, thus indicating that a portion of the solvent of crystallization is ethyl alcohol. After drying at 61° to constant weight it was analyzed.

Anal. Calcd. for $C_{20}H_{26}O_{11}N_2$: C, 51.04; H, 5.57; N, 5.96; OC_2H_5, 9.53. Found: C, 51.26; H, 5.90; N, 6.14; OC_2H_5, 9.47.

1-*d*-Galactosidouracil.—1,2-Dihydro-2-keto-4-ethoxy-1-tetraacetyl-*d*-galactosidopyrimidine was simultaneously deacetylated and deëthylated in the same manner as that described for the preparation of 1-*d*-xylosidouracil. The galactoside was recrystallized from 80% ethyl alcohol and deposited on cooling as thick rhombs; s. p. 235°; m. p. 250–251°; $[\alpha]^{25}D$ +59.9° (c = 5.01 in distilled water). It was soluble in an equal weight of boiling water and crystallized on cooling.

Anal. Calcd. for $C_{10}H_{14}O_7N_2$: C, 43.80; H, 5.11; N 10.22. Found: C, 43.47; H, 5.33; N, 10.21.

1,2-Dihydro-2-keto-4-ethoxy-1-triacetyl-*l*-arabinosidopyrimidine.—Two grams of finely divided acetobromo-*l*-arabinose and 2 cc. of 2,4-diethoxypyrimidine were heated at 70° for nineteen hours. Acetobromo-*l*-arabinose was not very soluble in warm 2,4-diethoxypyrimidine and the mixture was frequently shaken to facilitate solution. The end-product of the reaction was a clear light brown sirup. It was dissolved in an equal volume of ether and placed in the ice-chest for one week as crystallization took place very slowly. The crystalline material was separated and washed well with ether; yield 0.90 g. (38% of the theoretical yield based on the acetobromo-*l*-arabinose). It was dissolved in hot ethyl alcohol, ether added and the solution cooled; the crystals separated without solvent of crystallization; $[\alpha]^{21}D$ +108.8° (c = 4.11 in U. S. P. chloroform).

Anal. Calcd. for $C_{17}H_{22}O_9N_2$: C, 51.23; H, 5.57; N, 7.04. Found: C, 51.35; H, 5.56; N, 7.12.

This arabinose derivative was dimorphous; when its alcoholic solution was chilled rapidly it had a tendency to separate as needles; m. p. 157°. When the solution was cooled very slowly it generally crystallized as huge monoclinic prisms; m. p. 167.5°. Apparently the latter was the stable form as the mixed melting point of the two was 167.5°. The melt sometimes crystallized in the unstable form and at other times in the stable form.

1-*l*-Arabinosidouracil was prepared in an analogous manner to that described for the preparation of 1-*d*-xylosidouracil. The sirup obtained after concentrating the alcoholic hydrochloric acid solution crystallized when treated with alcohol (frequent vigorous stirring); yield, practically quantitative. After crystallization from 90% ethyl alcohol from which it separated as diamond-shaped hexahedra it melted at 251–252°; $[\alpha]^{23}D$ +88.2° (c = 1.21 in distilled water).

Anal. Calcd. for $C_9H_{12}N_2O_6$: C, 44.24; H, 4.95; N, 11.48. Found: C, 44.40; H, 5.00; N, 11.51.

Some By-products Formed in the Interaction of Acetobromoglycosides with 2,4-Diethoxypyrimidine

1,2-Dihydro-2-keto-1-ethyl-4-ethoxypyrimidine was isolated from the reaction mixtures in which the reactants with 2,4-diethoxypyrimidine were acetobromo-*d*-glucose, acetobromo-*d*-xylose and acetobromo-*d*-mannose. The manner in which it was obtained was the following one. After filtering the nucleoside that crystallized from the solution of the reaction sirup in ether, the filtrate was set in a refrigerator. The separation of 1,2-dihydro-2-keto-1-ethyl-4-ethoxypyrimidine was very slow, generally requiring several days. The solid product, after filtering and washing with ether, was quite sticky and was purified by sublimation (long plates); yield 4 g.[16] from 50 g. of acetobromo-*d*-glucose and 50 g. of 2,4-diethoxypyrimidine. It was recrystallized from ether as stout needles of m. p.

(16) This represents only a fraction of the product present in the reaction mixture. 1,2-Dihydro-2-keto-1-ethyl-4-ethoxypyrimidine can be removed quantitatively by extracting the ether solution of the original filtered sirup with water. This procedure was not adopted since it was desired to avoid the hydrolysis of and to isolate if possible "orthoacetates" or products in which the sugar was attached to the 2-position of the pyrimidine ring.

88°. This 1-ethyl derivative is very soluble in cold water, alcohol, and chloroform and slightly soluble in ether; it does not give a Wheeler–Johnson color test.

Anal. Calcd. for $C_8H_{12}O_2N_2$: C, 57.10; H, 7.20; N, 16.67. Found: C, 57.21; H, 7.13; N, 16.58.

1-Ethyluracil.—This was obtained by dissolving the above compound (1.00 g.) in concentrated hydrochloric acid (10 cc.) and concentrating to dryness on a steam-bath. The residue was crystallized from a solution of alcohol and ether and separated as clusters of prisms; m. p. 147.5°; yield 0.53 g. 1-Ethyluracil is very soluble in cold water, alcohol and acetone, slightly soluble in ether and sparingly soluble in hot carbon tetrachloride; it did not give a Wheeler–Johnson color test.

Anal. Calcd. for $C_6H_8O_2N_2$: C, 51.40; H, 5.76; N, 20.00. Found: C, 51.61; H, 5.75; N, 20.14.

1,2 - Dihydro - 2 - keto - 4 - ethoxypyrimidine was isolated from the products formed in the interaction of either acetobromo-*d*-mannose or acetobromo-*d*-galactose with 2,4-diethoxypyrimidine. As the methods of isolating the material in the two cases was quite similar only the details of the procedure, in which the former sugar was used, will be described. A solution of 7 g. of acetobromo-*d*-mannose and 7 g. of 2,4-diethoxypyrimidine was heated at 65° for seventy hours; after cooling a small amount of uracil deposited and this was removed. From the filtrate, 1,2-dihydro-2-keto-1-ethyl-4-ethoxypyrimidine (2 g.) slowly crystallized and after a week it was collected. The sirupy filtrate over a period of a year slowly deposited a crystalline product. The solid was removed by filtration and washed with a small amount of ether. Trituration with chloroform separated the ethoxy derivative from uracil. The chloroformic extract was concentrated and the residue recrystallized from a solution containing 90% benzene and 10% absolute ethyl alcohol; massive aggregates of colorless plates were thus obtained; yield 0.36 g.; m. p. 168°; a mixed melting point with 1,2-dihydro-2-keto-4-ethoxypyrimidine[17] was unchanged. The properties as well as the response to the Wheeler–Johnson color test are identical with those described previously for this compound.

Anal. Calcd. for $C_6H_8O_2N_2$: C, 51.40; H, 5.76; N, 20.00. Found: C, 51.75; H, 5.76; N, 19.95.

Uracil was isolated in traces in all experiments except when acetobromo-*d*-glucose was used. This pyrimidine was easily obtained in the pure state by triturating the crude material with chloroform and then recrystallizing from water; it was identified by its properties, Wheeler–Johnson color test and analysis.

(17) Hilbert and Jansen, THIS JOURNAL, **57**, 552 (1935).

Summary

Acetobromoglycosides, in general, interact with 2,4-diethoxypyrimidine to give 1,2-dihydro-2-keto - 1 - acetylglycosido - 4 - ethoxypyrimidines. Appreciable amounts of the secondary product, 1,2 - dihydro - 2 - keto - 1 - ethyl - 4 - ethoxypyrimidine are also formed. Presumptive evidence indicates that by-products, structurally related to the 4-ethoxy-2-triacetyl-*d*-ribosidopyrimidine which is formed in the interaction of acetobromo-*d*-ribose and 2,4-diethoxypyrimidine, are present in the products of the reaction of 2,4-diethoxypyrimidine with acetobromo-*d*-mannose and with acetobromo-*d*-galactose; however, attempts to isolate these analogs in the crystalline condition have thus far been unsuccessful. Hydrolysis of 1-acetylglycosidopyrimidine derivatives with alcoholic hydrochloric acid produces the 1-glycosidouracils. 1-*d*-Xylosido-, 1-*l*-arabinosido- and 1-*d*-galactosidouracil were prepared in this manner and their chemical properties found to be similar to those of uridine.

WASHINGTON, D. C. RECEIVED NOVEMBER 16, 1936

[CONTRIBUTION FROM THE DEPARTMENT OF CHEMISTRY, STANFORD UNIVERSITY]

Conductivities of One-Molal Mixtures of Alkali Halides and Nitrates

BY PIERRE VAN RYSSELBERGHE AND LEE NUTTING

Introduction

As pointed out by Smith and Gortner,[1] few systematic studies of the conductivity of mixed electrolytes are available for theoretical investigation. Moreover, the available data have not as yet received an entirely satisfactory interpretation. The small but definite departures from the mixture rule observed by Stearn and by Ruby and Kawai[2] in the case of mixed alkali halides have recently been discussed by Van Rysselberghe and Nutting.[3] They showed that these departures could be reduced appreciably if the mixture rule is corrected by means of simple but plausible assumptions concerning the adjustment of mobilities which takes place upon mixing. The modified form of the mixture rule obtained by them involves the transport numbers of the ions in solutions of the pure salts. As these transport numbers are not always known with great accuracy calculations of real significance cannot be carried

(1) Smith and Gortner, *J. Phys. Chem.*, **37**, 79 (1933).

(2) Stearn, THIS JOURNAL, **44**, 670 (1922); Ruby and Kawai, *ibid.*, **48**, 1119 (1926).

(3) Van Rysselberghe and Nutting, *ibid.*, **56**, 1435 (1934).