[CONTRIBUTION FROM THE LABORATORIO DE QUÍMICA ORGÁNICA, FACULTAD DE CIENCIAS EXACTAS, FÍSICAS Y NATURALES, AND THE LABORATORIO DE QUÍMICA BIOLÓGICA, FACULTAD DE CIENCIAS MÉDICAS]

THE REACTION OF AMMONIA WITH SOME ACETYLATED AND BENZOYLATED MONOSACCHARIDES. I. D-GLUCOSE DERIVATIVES AND AN INTERPRETATION

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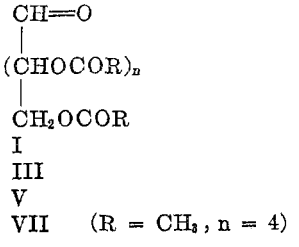
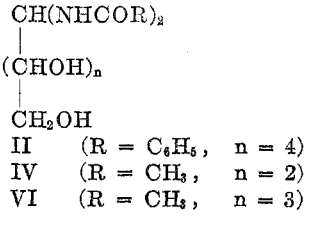
The action of ammonia on acetylated and benzoylated monosaccharides has been the subject of several prior investigations. Brigl, Mühlschegel, and Schinle (1) treated pentabenzoyl-*aldehydo*-D-glucose (I), 3,4,5,6-tetrabenzoyl-*aldehydo*-D-glucose, and 3,5,6-tribenzoyl-D-glucofuranose with ammonia and obtained in each case D-glucose dibenzamide (II). They suggested that this compound was formed by condensation of the aldehyde group with molecules of benzamide produced by ammonolysis. For the reaction of 3,5,6-tribenzoyl-D-glucofuranose they postulated the intermediate formation of the *aldehydo* structure.



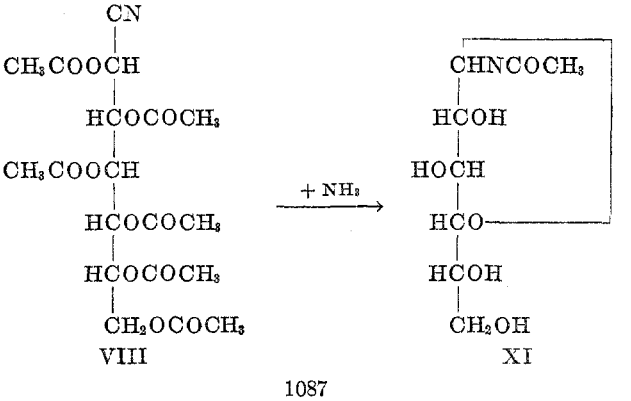


I
III
V
VII (R = CH_3, n = 4)

II (R = C_6H_5, n = 4)
IV (R = CH_3, n = 2)
VI (R = CH_3, n = 3)

Deulofeu (2) obtained L-erythrose diacetamide (IV) by the action of ammonia on triacetyl-*aldehydo*-L-erythrose (III) and Isbell and Frush (3) L-arabinose diacetamide (VI) from tetraacetyl-*aldehydo*-L-arabinose (V). These authors proposed a reaction mechanism, to explain the formation of the "aldose diamide" compounds, that will be discussed later.

Hockett and Chandler (4) found that N-acetyl-D-glucofuranosylamine (XI) is formed either by treatment of pentaacetyl-*aldehydo*-D-glucose (VII) with ammonia or by Wohl-degradation of hexaacetyl-D-gluco-D-*gulo*-heptonic nitrile. Niemann and Hays (5) observed that N-acetyl-D-glucofuranosylamine (XI) is also produced by the action of methanolic ammonia on pentaacetyl β-D-glucopyranose.



VIII XI

Some time ago (6) we found that when pentabenzoyl-α-D-glucose is treated with ammonia, D-glucose dibenzamide (II) is produced in 21% yield. When the β-isomer was submitted to the same reaction, 19% of D-glucose dibenzamide was obtained, showing that spatial modification of carbon atom 1 does not introduce a significant change in the course of the reaction.

We have confirmed and repeated the preparation of N-acetyl-D-glucofuranosylamine (XI) from pentaacetyl-β-D-glucose, following the directions of Niemann and Hays (5), and have found that, if the crude reaction product is acetylated, octaacetyl-di-D-glucosylamine (7) is obtained. The original di-D-glucosylamine is undoubtedly formed by condensation of D-glucosylamine (8) produced in the reaction by the action of ammonia on D-glucose.

We have found that N-acetyl-D-glucofuranosylamine (XI) can also be obtained in 35% yield by treating hexaacetyl-D-gluco-D-*ido*-heptonic nitrile (VIII) with methanolic ammonia. Hexaacetyl-D-gluco-D-*gulo*-heptonic nitrile, that is the epimer of the former, gives under the same conditions (9) a yield of 30%. Hence there is no significant difference in the results obtained by use of epimers (*i.e.*, the configuration of carbon 2 is not a critical factor for the course of the reaction).

The hexaacetyl-D-gluco-D-*ido*-heptonic nitrile was prepared by dehydration of the amide with phosphorus oxychloride. In chloroform it has a low dextro rotation, and does not follow the rule formulated by Deulofeu (10) as to the sign of the rotation of acetylated aldonic nitriles.

The formation of N-acetyl-N-glucofuranosylamine and of D-glucose dibenzamide from pentaacetyl- and pentabenzoyl-D-glucose, presumably follows the mechanism postulated by Isbell and Frush (3), for the formation of "aldose diacetamides" in the Wohl's degradation, or in the reaction of acetylated-*aldehydo*-monosaccharides with ammonia.

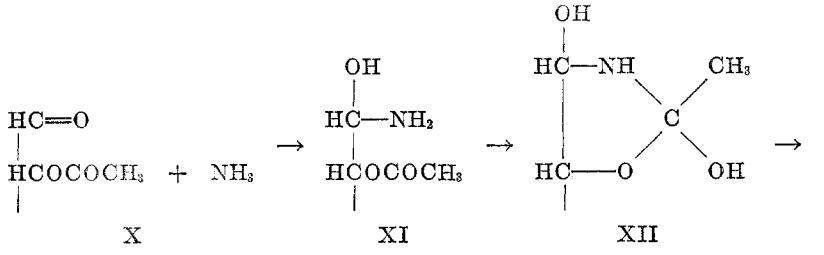
In this theory, the amide groups are derived from acyl groups without separation from the aldose molecule. The presence of a free aldehyde group is necessary (X). Ammonia condenses with this group and compounds of type (XI) are formed. The nitrogen of the amino group then forms a labile orthoester with the neighboring acetyl (XII). This orthoester rearranges with migration of the acetyl group to the nitrogen (XIII), forming the first amide group. The second amide may be formed by addition of ammonia to another acyl group (XIV), a new arrangement produces the final "aldose diacetamide" (XVI).

In the case of the cyclic acetyl and benzoyl derivatives of the monosaccharides, it is necessary to make the acceptable assumption that in a number of molecules, the acyl group in carbon atom 1 is separated by ammonolysis before the other acyls. A potential aldehyde group is then formed, that can pass into an actual one, condense with ammonia, and through the reactions X $\rightarrow$ XVI, produce the diamide compounds.

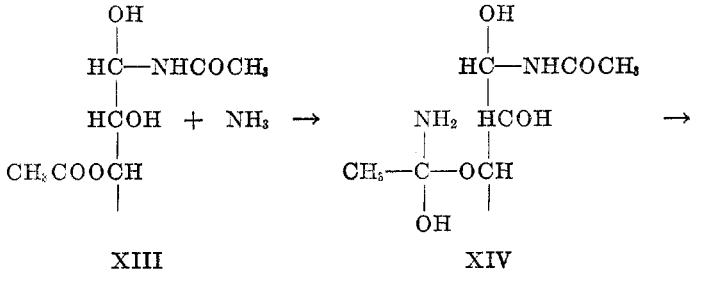
If, when the reaction has arrived at stages XI–XIII, an hydroxyl is already free on a convenient carbon atom of the monosaccharide, because the acyl group united to it has been separated by ammonolysis, it can condense with the hydroxyl present on carbon atom 1 (XIII) and give a stable ring. In this case, monoamide

derivatives can be obtained, and as happens with pentaacetyl-β-D-glucose, N-acetyl-D-glucofuranosylamine is produced.

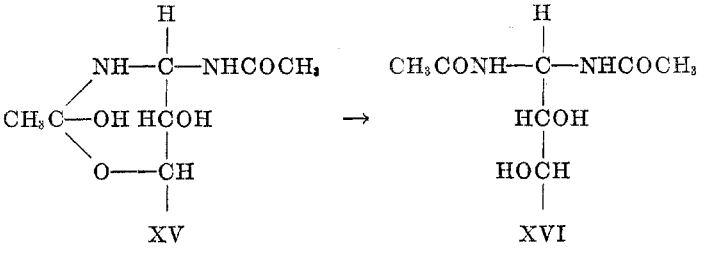
With benzoyl groups ammonolysis is slower than with acetyl groups, and there is a higher probability for the production of diamides by the path XII–XVI, giving in this case D-glucose dibenzamide.



X XI XII



XIII XIV



XV XVI

The production of di-D-glucosylamine isolated as the octaacetate is interpreted as the result of the formation of small amounts of D-glucose by ammonolysis. D-Glucose is known to yield D-glucosylamine and di-D-glucosylamine by reaction with alcoholic ammonia.

Summarizing, it can be stated that because of the complexity of the process, the products obtained from the acylated-D-glucoses and the acylated heptonic nitriles are the result of a series of competitive reactions, amongst which the velocity of ammonolysis, of the different acyl groups, plays an important part.

That amide molecules in solution do not react appreciably to give "aldose amide" compounds, has already been proved by Hockett, Deulofeu, and Deferrari (11), who employed a solution of methanolic ammonia, labelled with N^{15}, and containing acetamide with normal nitrogen, to carry out the reaction.

When tetraacetyl-L-arabonic nitrile was degraded under these conditions, the L-erythrose diacetamide obtained contained N^{15} in an amount indicative that most of the reaction took place by the mechanism formulated by Isbell and Frush.

Deulofeu and Deferrari (9) also found that when an amide, with an acyl group different from that present in the acylated monosaccharide, is added to the ammonia solution, it does not condense with the aldose.

In this paper another example is given. When one molecule of pentabenzoyl-α-D-glucose was treated with methanolic ammonia containing 10 moles of acetamide in solution, D-glucose dibenzamide was isolated in almost the same yield (19%) as when the acetamide was not present. No product of condensation of D-glucose with acetamide could be detected.

EXPERIMENTAL

The methanolic ammonia employed in all experiments contained 16% ammonia. Ethanol, when not specifically stated, was 96%.

D-Glucose dibenzamide from pentabenzoyl-α-D-glucose. Pentabenzoyl-α-D-glucose (10 g.), finely ground, was suspended in 350 ml. of methanolic ammonia (16%) and shaken to dissolution. After 18 hours at room temperature, the solution was evaporated in a vacuum to dryness and the residue was dissolved in 30 ml. of boiling ethanol. By cooling 1.22 g. (21.2%) of D-glucose dibenzamide, m.p. 197–198° were obtained. Recrystallization from ethanol gave m.p. 201–202°; $[\alpha]_D^{25}$ +1.30°, in pyridine.

The material was the same as that prepared from pentabenzoyl-*aldehydo*-D-glucose, by the method of Brigl, Mühlschlegel, and Schinle (1).

Anal. Calc'd for $C_{20}H_{24}N_2O_7$: C, 59.40; H, 5.94; N, 6.93.

Found: C, 59.20; H, 6.12; N, 7.11.

Pentaacetyl-D-glucose dibenzamide. D-Glucose dibenzamide (730 mg.) was dissolved in 25 ml. of a mixture (1:1) of pyridine and acetic anhydride, by shaking at room temperature. After 24 hours it was poured into ice-water. An oil precipitated, that crystallized easily. The crystals (950 mg.) were separated and recrystallized from ethanol. White needles melting at 196° (sintering from 193–194°); $[\alpha]_D^{25}$ −39.7°, in chloroform (*c*, 1.006) were obtained.

Anal. Calc'd for $C_{30}H_{34}N_2O_{10}$: C, 58.68; H, 5.54 N, 4.56.

Found: C, 58.86; H, 5.12; N, 4.85.

This acetate, treated with methanolic ammonia, gave the original D-glucose dibenzamide, m.p. 200–201°.

D-Glucose dibenzamide from pentabenzoyl-α-D-glucose in the presence of acetamide. Pentabenzoyl-α-D-glucose (5 g., 1 mole), was dissolved in 150 ml. of methanolic ammonia containing 4.1 g. (10 moles) of acetamide. After 18 hours at room temperature, the solution was evaporated to dryness and the residue extracted with boiling ethyl acetate. The residue was dissolved in 15 ml. of boiling ethanol and by cooling 400 mg. of D-glucose diacetamide were obtained, m.p. 191°.

The ethyl acetate extract on standing deposited 160 mg. of crystals, m.p. 196–197°, that were found to be also D-glucose dibenzamide. Total yield: 560 mg. (19.4%). Recrystallization from ethanol gave 520 mg. of the pure compound, m.p. 202–203°.

D-Glucose dibenzamide from pentabenzoyl-β-D-glucose. Pentabenzoyl-β-D-glucose (750 mg.) was treated with 23 ml. of methanolic ammonia and the procedure described for the α-isomer was followed. Yield: 70 mg. (18.5%), m.p. 197–198°, which, recrystallized from ethanol, gave pure D-glucose dibenzamide, m.p. 200°.

D-Gluco-D-ido-heptonic lactone. This lactone was obtained from the residues of the preparation of D-gluco-D-*gulo*-heptonic lactone, through the brucine salt. The elimination of the

last traces of barium, employed for the decomposition of the salt, was done by passing the water solution containing the acid through a column of Zeokarb-H resin, which shortens appreciably the time employed in the preparation. M.p. 151–152°; $[\alpha]_D^{25}$ −67.9° [Fisher (12) gives m.p. 151–152°; $[\alpha]_D$ −67.7°, in water].

*Hexaacetyl-*D*-gluco-*D*-ido-heptonic amide.* D-Gluco-D-*ido*-heptonic amide(12 g.) prepared from the lactone according to the method of Hudson and Komatsu (13), was dissolved by shaking with a mixture of 72 ml. of pyridine and 72 ml. of acetic anhydride. After 48 hours at room temperature, 200 ml. of chloroform was added and the clear solution was extracted three times with 150 ml. of 3 *N* sulphuric acid, twice with 150 ml. of saturated sodium bicarbonate solution, and finally with water. The chloroform was then dried with sodium sulphate, evaporated in a vacuum, and the residue crystallized from ethanol. Yield 14.8 g. (62%); m.p. 116–118°. Recrystallized from ethanol as white prisms, m.p. 120–122°; $[\alpha]_D^{14}$ +47.2°, in chloroform (*c*, 1.72).

Anal. Calc'd for $C_{19}H_{27}NO_{13}$: C, 47.75; H, 5.65.
Found: C, 47.67; H, 5.77.

*Hexaacetyl-*D*-gluco-*D*-ido-heptonic nitrile.* To 15 g. of the above acetylated amide, 30 ml. of phosphorus oxychloride was added and the mixture was heated for 30 minutes at 80°. The excess of oxychloride was distilled in a vacuum and the residue was dissolved in water-chloroform. The chloroform layer was separated, and washed with sodium bicarbonate solution and with water. It was then dried, evaporated, and the oily residue dissolved in ethanol, when crystallization took place. Recrystallization yielded 8.65 g. (60%) of small white prisms, m.p. 105–108°; $[\alpha]_D^{22}$ +0.68°, in chloroform (*c*, 4.42); $[\alpha]_D^{20}$ +2.16°, in acetic acid (*c*, 3.47).

Anal. Calc'd for $C_{19}H_{25}NO_{12}$: C, 49.65; H, 5.44.
Found: C, 49.69; H, 5.53.

When the acetylated nitrile was treated with hydrobromic acid in acetic acid solution, it hydrolyzed to the amide. M.p. 120–122°; $[\alpha]_D^{21}$ +47.8°, in chloroform.

*N-Acetyl-*D*-glucofuranosylamine from hexaacetyl-*D*-gluco-*D*-ido-heptonic nitrile.* The nitrile (5 g.) was added to 150 ml. of methanolic ammonia. Dissolution was very rapid and after 24 hours at room temperature the solution was evaporated in a vacuum. The syrupy residue, well dried, was extracted with ethyl acetate, dried again, and treated with careful warming, with 30 ml. of absolute ethanol. Crystallization began easily and after two hours at 0°, the suspension was filtered and the crystals were well washed with absolute ethanol. Yield: 690 mg., m.p. 200–201°. By slow evaporation of the ethanolic mother liquors, a further 150 mg. were obtained, m.p. 202–203°. Total yield: 840 mg. (34.9%). It was recrystallized from 80% ethanol and gave m.p. 201–202°; $[\alpha]_D^{20}$ +86.2°, in water.

The substance was identified by mixture melting point with an authentic sample of N-acetyl-D-glucofuranosylamine.

*Octaacetyl-di-*D*-glucosylamine.* Pentaacetyl-β-D-glucose (5 g.) was dissolved in 150 ml. of methanolic ammonia and left for 24 hours at room temperature. The solution was then evaporated in a vacuum, and the residue, well dried, was extracted with boiling ethyl acetate, dried again, and dissolved by cautious warming in 15 ml. of pyridine. After cooling 15 ml. of acetic anhydride were added. After 24 hours the whole was poured into ice-water and on agitation crystals formed very easily. These were filtered, washed with water, and recrystallized from ethanol. Yield: 560 mg., m.p. 213–214°; $[\alpha]_D^{25}$ +86°; [Brigl and Keppler (7) give m.p. 214°; $[\alpha]$ +85°, in water].

SUMMARY

1. By the action of methanolic ammonia on the α- and β-isomers of pentabenzoyl-D-glucose, D-glucose dibenzamide is obtained in almost the same yield.

2. When hexaacetyl-D-gluco-D-*ido*-heptonic nitrile is treated with methanolic

ammonia, N-acetyl-D-glucofuranosylamine is produced, in a yield similar to that obtained from the epimer, hexaacetyl-D-gluco-D-*gulo*-heptonic nitrile.

3. A discussion is given of the mechanism of the reaction.

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