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STUDIES ON SYNTHESIS AND RING CLOSURE OF 4-ACETAMIDO-4-DEOXY SUGARS

DISSertation

Presented in Partial Fulfillment of the Requirements for the Degree Doctor of Philosophy in the Graduate School of The Ohio State University

By

Ahmed Emad El-Din El-Ashmawy, B.S., M.Sc.

The Ohio State University 1966

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Adviser
Department of Chemistry
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VITA

I, Ahmed Emad El-Din El-Ashmawy, the son of Abd El-Wahab El-Ashmawy was born in Egypt on May 5, 1933. I was graduated at the University of Cairo, U.A.R. in June 1955 with a Bachelor of Science degree. I worked as chemist in the Egyptian Sugar Company at Kom Ombo, U.A.R. for one year, from February 1954 till February 1955. The same month I joined the National Research Center at Cairo. I received the Master of Science degree in November 1958 from Faculty of Science, Cairo University, U.A.R. The thesis, supervised by Dr. Yehia A. Fahmy, of the National Research Center of U.A.R. and Prof. Dr. H. Fahim of Faculty of Science, Asyout University, was entitled "Studies on some Egyptian Raw Materials for Production of Viscose Pulps". I have been granted a graduate fellowship by the U.A.R. Government to study in the United States. In September 1960, I began work on the Doctor of Philosophy degree at The Ohio State University.
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INTRODUCTION

1. Occurrence of Amino Sugars in Nature and Their Role in Biological Systems

Amino sugars are sugars in which one or more of the hydroxyl groups other than the hemiacetal hydroxyl group have been replaced by amino groups. Sugars having the hemiacetal hydroxyl substituted by an amino group are termed glycosylamines.

Amino sugars are present as component residues of a number of important biological macromolecules chiefly in the so-called mucoid substances (Table 1), and are also involved in certain low molecular-weight soluble metabolites (Table 2).

Amino sugars in macromolecules

Mucoid substances include mucopolysaccharides, glycoproteins, and glycolipids, depending on whether the substance is mainly polysaccharide, or contains a major protein or lipid moiety. The commonly encountered amino sugars include the hexosamines, 2-amino-2-deoxy-D-glucose, 2-amino-2-deoxy-D-galactose, and also N-acetyleneuraminic acid (I) and muramic acid (II), and may be accompanied in the macromolecule

![Chemical structures](I) and (II)
# Table 1

## Amino Sugars in Biological Macromolecules

<table>
<thead>
<tr>
<th>Amino Sugar</th>
<th>Macromolecule</th>
<th>Tissue Source, or Microorganism</th>
</tr>
</thead>
<tbody>
<tr>
<td>2-Amino-2-deoxy-D-glucose</td>
<td>Chitin</td>
<td>Exoskeleton of many invertebrates</td>
</tr>
<tr>
<td></td>
<td>Heparin</td>
<td>Mast cells of circulatory system, especially in liver, muscle, lung, heart.</td>
</tr>
<tr>
<td></td>
<td>Hyaluronic acid</td>
<td>Connective tissue, intercellular spaces, joint fluids, cornea, bone, heart valves, mammalian plasma, and bacterial cell wall</td>
</tr>
<tr>
<td>2-Amino-2-deoxy-D-galactose and its 4- or 6-sulfate</td>
<td>Chondroitin</td>
<td>Mammalian cornea, cartilage, blood group substances</td>
</tr>
<tr>
<td></td>
<td>Chondroitin sulfates</td>
<td>Dermatan sulfate</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Spleen, brain, skin, heart valves, aorta</td>
</tr>
<tr>
<td>2-Amino-2,6-dideoxy-D-galactose</td>
<td>Lipopolysaccharide</td>
<td>Chromobacterium violaceum</td>
</tr>
<tr>
<td>(D-Fucosamine)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2-Amino-2,6-dideoxy-L-galactose</td>
<td>Polysaccharide</td>
<td>Pneumococcus type V</td>
</tr>
<tr>
<td>(L-Fucosamine)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2-Amino-2,6-dideoxy-L-talose</td>
<td>Polysaccharide</td>
<td>Pneumococcus type V</td>
</tr>
<tr>
<td>(Pneumosamine)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2-Amino-2-deoxy-D-galacturonic acid</td>
<td>Vi antigens</td>
<td>Escherichia coli</td>
</tr>
<tr>
<td>Amino Sugar</td>
<td>Macromolecule</td>
<td>Tissue Source, or Microorganism</td>
</tr>
<tr>
<td>----------------------------------------------------</td>
<td>----------------------</td>
<td>--------------------------------------------</td>
</tr>
<tr>
<td>2-Amino-2-deoxy-glucuronic acid</td>
<td>Polysaccharide</td>
<td>Haemophilus influenza</td>
</tr>
<tr>
<td>2-Amino-2-deoxy-mannuronic acid</td>
<td>Polysaccharide</td>
<td>Micrococcus lysodeikticus</td>
</tr>
<tr>
<td>4-Amino-4,6-dideoxy-D-glucose (viosamine)</td>
<td>Lipopolysaccharide</td>
<td>Chromobacterium violaceum</td>
</tr>
<tr>
<td>2-Amino-3-O-(D-1-carboxyethyl)-2-deoxy-D-glucose (Muramic acid)</td>
<td></td>
<td>Cell walls of bacteria and actinomycetes</td>
</tr>
<tr>
<td>N-acetylneuraminic acid</td>
<td>Colominic acid</td>
<td>E. coli (as colominic acid) aorta, cartilage, gangliosides</td>
</tr>
</tbody>
</table>
### Table 2

**Amino Sugars in Low Molecular-weight Soluble Metabolites**

<table>
<thead>
<tr>
<th>Amino Sugar</th>
<th>Occurrence in antibiotics</th>
</tr>
</thead>
<tbody>
<tr>
<td>2-Amino-2-deoxy-D-glucose (Glucosamine)</td>
<td>Kanamycin C</td>
</tr>
<tr>
<td></td>
<td>Paromomycin</td>
</tr>
<tr>
<td>2-Deoxy-2-methylamino-L-glucose</td>
<td>Mannosidostreptomycin</td>
</tr>
<tr>
<td></td>
<td>Hydroxystreptomycin</td>
</tr>
<tr>
<td></td>
<td>Streptomycin</td>
</tr>
<tr>
<td>2-Amino-2-deoxy-D-gulose</td>
<td>Streptolin B</td>
</tr>
<tr>
<td></td>
<td>Streptothricin</td>
</tr>
<tr>
<td>3-Amino-3-deoxy-D-glucose (Kanosamine)</td>
<td>Kanamycin A</td>
</tr>
<tr>
<td></td>
<td>Kanamycin B</td>
</tr>
<tr>
<td></td>
<td>Kanamycin C</td>
</tr>
<tr>
<td>3-Amino-3-deoxy-D-ribose</td>
<td>Puromycin</td>
</tr>
<tr>
<td>3-Dimethylamino-3,4,6-trideoxy-D-xylo-hexose</td>
<td>Erythromycin</td>
</tr>
<tr>
<td>(Desosamine)</td>
<td></td>
</tr>
<tr>
<td>3,6-Dideoxy-3-dimethylamino-D-glucose (Mycaminose)</td>
<td>Carbomycin</td>
</tr>
<tr>
<td></td>
<td>Formacidines A, B and C</td>
</tr>
<tr>
<td></td>
<td>Leucomycin</td>
</tr>
<tr>
<td>3-Amino-3,6-dideoxy-D-mannose (Mycosamine)</td>
<td>Amphotericin B and</td>
</tr>
<tr>
<td></td>
<td>Nystatin, pimaricin</td>
</tr>
<tr>
<td>3-Dimethylamino-2,3,6-trideoxy-L-lyxo-hexose</td>
<td>Cinerubin A and B</td>
</tr>
<tr>
<td>(Rhodosamine)</td>
<td>Pyrromycin</td>
</tr>
<tr>
<td></td>
<td>Rhodomycin A and B</td>
</tr>
<tr>
<td></td>
<td>7-rhodomycins</td>
</tr>
</tbody>
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Table 2 (continued)

<table>
<thead>
<tr>
<th>Amino Sugar</th>
<th>Occurrence in antibiotics</th>
</tr>
</thead>
<tbody>
<tr>
<td>4,6-Dideoxy-4-dimethylamino-D-glucose (Amosamine)</td>
<td>amicetin</td>
</tr>
<tr>
<td>4-Dimethylamino-2,3,4,6-tetradeoxyhexose</td>
<td>formacidines A, B and C</td>
</tr>
<tr>
<td>6-Amino-6-deoxy-D-glucose</td>
<td>Kanamycin A</td>
</tr>
<tr>
<td>2,6-Diamino-2,6-dideoxy-L-idose (Neosamine B, paromose)</td>
<td>Neomycin B Paromomycin Zygomycin A</td>
</tr>
<tr>
<td>2,6-Diamino-2,6-dideoxy-D-glucose (Neosamine C)</td>
<td>Neomycin B Neomycin C Zygomycin A</td>
</tr>
</tbody>
</table>
by residues of other sugars, hexuronic acids, amino acids, and lipid components.

Mucopolysaccharides, sometimes called glycosaminoglycans or glycosamino-glycuronoglycans, when the chemical structure is well established, have been found extensively in nature.

Chitin is the major organic component of the exoskeleton of arthropods and many other invertebrates. It is found also in the cell walls of most fungi. It is a linear, β-D(1→4)-linked condensation polymer of 2-acetamido-2-deoxy-D-glucose residues (1). The first amino sugar to be reported was "glykosamin", obtained by hydrolysis of lobster shells (2), and it was shown soon thereafter to be either 2-amino-2-deoxy-


D-glucose hydrochloride or the 2-epimer. The D-gluc configuration was not firmly established until in 1939 (3).


2-Amino-2-deoxy-D-galactose ("chondrosamine") was first isolated in 1913. It is a component of the mucoproteins of cartilage and tendon (4) and its configuration was unequivocally established (5) in 1945.

(4) P. A. Levene and F. B. LaForge, J. Biol. Chem., 18, 123 (1914).
Hyaluronic acid is found in the ground substance of many connective tissues where it seems to play an important physiological role (6). It binds water in intercellular spaces and serves as a lubricant in moving joints. Its presence has been reported in cornea, heart valve, and tumor fluids. It is a linear copolymer of two alternating sugar units, D-glucuronic acid, β-D-(1→3) linked to 2-acetamido-2-deoxy-D-glucose (N-acetylglucosamine) unit, which in turn is β-D-(1→4) linked to a second D-glucuronic acid residue (7, 8).

Chondroitin 4-sulfate is one of the major constituents of cartilage. It is a sulfated, linear polysaccharide having alternating 2-acetamido-2-deoxy-D-galactose and D-glucuronic acid residues and the repeating unit is shown (III). Nearly half of the total glycosaminoglycan in bovine
Another chondroitin sulfate isolated from human umbilical cord, tendon, cartilage, heart valve, skin and other sources is chondroitin 6-sulfate, which differs from chondroitin 4-sulfate only in the placement of the sulfate group on the 2-acetamido-2-deoxy-D-galactose residue.

Dermatan sulfate (earlier known as chondroitin sulfate B) has also been separated from the mixture of mucopolysaccharides obtained from skin, tendon, heart valves, and other tissues. It differs from chondroitin 6-sulfate and chondroitin 4-sulfate in that the uronic acid moiety is L-iduronic acid, the 5-epimer of D-glucuronic acid (10). The sulfate groups are believed to be at C-4 of the hexosamine moiety (6).

Heparin (the blood anticoagulant factor) has been shown (11) to be
an α-D-(1→4) linked polymer of D-glucuronic acid and 2-amino-2-deoxy-D-glucose; the amino group and some of the hydroxyls are sulfated.

There also exists in nature a family of amino sugars (nonulosaminic acids or sialic acids) containing 9 carbon atoms in the principal chain. The members of this group may be regarded as acyl derivatives of neuraminic acid (5-acetamido-3,5-dideoxy-D-glycero-D-galacto-nonulosonic acid). Neuraminic acid itself has not been isolated from a natural source, but its N-acetyl derivative, and various O-acetyl derivatives thereof are found widely distributed throughout the animal kingdom (9, 12).

They are found in aorta, aqueous humor, cartilage, corneal stroma, dental pulp, gangliosides of nervous tissue, kidney, liver, salivary glycoprotein, and synovial fluid.

Nonulosaminic acids have not been detected in the free state in appreciable concentration, but only in combined form. Colominic acid is a polysaccharide obtained from the culture medium in which *Escherichia coli* K-235 has been grown (13). It is a linear (2→8) linked polymer of N-acetylneuraminic acid residues (14). The linkage between the polymer
The glycolipids that occur in the ganglion cells of the nervous system are termed gangliosides. By applying chromatography on cellulose, Svennerholm (15) succeeded in separating gangliosides of human brain into two main fractions; N-acetylneuraminic acid and 2-acetamido-2-deoxy-D-galactose were detected.

Herring and Kent (16) isolated from adult bovine cortical bone a glycoprotein that contains sialic acid (sialoprotein). The sialoprotein is characterized by a high content of N-acetylneuraminic acid and hexoses (galactose, glucose and mannose), and amino sugars (2-amino-2-deoxy-D-glucose and 2-amino-2-deoxy-D-galactose).

The Vi antigens of certain strains of bacteria are strongly acidic polymers and upon hydrolysis yield 2-amino-2-deoxy-D-galacturonic acid as the principal product (17). A potent immunizing polysaccharide which has been designated SPA (staphylococcal polysaccharide antigen), has been


(15) Ref. 9, Chapter 36.


isolated from *Staphylococcus aureus* (18). This antigen was shown to possess active immunizing properties against a variety of staphylococcal strains in mice, dogs and man. The staphylococcal polysaccharide antigen has been shown to consist of equimolar proportions of 2-acetamido-2-deoxy-D-glucoronic acid and 2-[(N-acetylalanyl) amino]-2-deoxy-D-glucuronic acid linked by β-(1→4) linkages (19, 20).


Hexosamines and sialic acid (9) are also present in blood-group specific substances, which occur in erythrocytes (red blood cells). These substances are antigens, that is, they can induce the formation of antibodies (21).


**Amino sugars in metabolites of low molecular weight**

Several amino sugars have been isolated from antibiotic substances (22) and other soluble metabolites of cultures of microorganisms (Table 2).

(22) J. D. Dutcher, Advan. Carbohydrate Chem., 18, 259 (1963).
Folkers and co-workers (23) discovered 2-deoxy-2-methylamino-L-glucose as a moiety of the antibiotic streptomycin. The elucidation of the structure of streptomycin has been reviewed (24).


The amino sugars present in antibiotic substances are generally linked glycosidically to the remainder of the molecule. The antibiotic puromycin is a nucleoside of 3-amino-3-deoxy-ribose, and it has an amino acid residue at the 3-amino group. It was discovered by Waller and co-workers in 1953 (25), and was synthesized by Baker and Schaub (26);


its full structure is 6-dimethylamino-9-(3-p-methoxy-L-phenylalanylamino-3-deoxy-β-D-ribofuranosyl)purine. Puromycin has been shown to have tumor-inhibiting activity.

The antibiotic amicetin, discovered in 1953 as a metabolic product of several Streptomyces species (27), has been shown to be a derivative

of a pyrimidine nucleoside, and it contains an amino sugar moiety. The latter sugar (amosamine) has been shown by synthesis to be 4,6-dideoxy-4-dimethylamino-D-glucopyranose (28) (IV).


Incorrect structures had previously been assigned to amosamine through incorrect interpretations of data from degradation studies. Thus in a brief communication (29), the structure of a 3,4-dideoxy-3-dimethylamino hexose was first proposed, later (30) the structure was considered to be a 3,6-dideoxy-3-dimethylaminoaldehydhexose. The free 4-amino-4,6-dideoxy-D-glucose is termed viosamine, it is one of the components of the lipopolysaccharide of Chromobacterium violaceum (28).
2,6-Diamino-2,6-dideoxy-L-idose (paromose) is a component of the antibiotic paromomycin (31). The structures of paromomycin and some related antibiotics are shown in Figure (1).

Neomycin was first obtained by Waksman (32) from the culture fluid of Streptomyces fradiae. Later, it was shown to consist of the closely related antibiotics termed neomycins B and C. Both substances consist of three C₆ units and one C₅ unit, linked glycosidically. The diaminohexose component (neosamine C) of neomycin C has been shown to be 2,6-diamino-2,6-dideoxy-D-glucose (33, 34, 35) by comparison with


synthetic material and the diaminohexose (neosamine B) of neomycin B was shown by Rinehart et al. (36) to be 2,6-diamino-2,6-dideoxy-L-idose

(also known as paromosone). This structure was later confirmed also by synthesis (37, 38). Rinehart and co-workers (39) gave the complete structure for neomycins B and C shown in Figure (1).


The amino sugars have been the subject of a number of comprehensive and specialized reviews (40 - 44) and tables of derivatives of amino


sugars has been published (45).


Neomycin B (R=H, R'=R''=CH₂NH₂)
Neomycin C (R=R''=CH₂NH₂, R'=H)
Paromomycin I (R=H, R'=CH₂NH₂, R''=CH₂OH)
Paromomycin II (R=CH₂NH₂, R'=H, R''=CH₂OH)

Fig. (1)

2. Cyclic Forms of Sugars

The chemical reactions of most monosaccharides indicate that these compounds do not exist to a significant extent in the free aldehyde or aldehydrol form. For example, most aldoses do not give a positive Schiff test. Treatment with alcohols in acid solution leads not to acyclic acetals as in the case of simple aldehydes, but to mixtures of isomeric
cyclic substances (glycosides) having a single alkyl group. Treatment of aldoses with acetic anhydride normally leads to isomeric, cyclic, acetylated derivatives, and not to acyclic derivatives. Furthermore, the free sugars usually exhibit mutarotation in hydroxylic solvents. Infrared and ultraviolet spectroscopy confirm that the free carbonyl group is absent. Intramolecular, cyclic hemiacetal formation takes place between the carbonyl group and the hydroxyl on the γ or δ carbon atom to form a five-membered (furanose) or six-membered (pyranose) ring. These cyclic hemiacetals possess an additional asymmetric carbon atom at C-1 and each is capable of existing in two stereoisomeric forms, known as anomers. When the anomeric hydroxyl is in cis relationship with the hydroxyl of the highest numbered asymmetric center of the sugar chain in Fischer projection it is called an α-anomer. If the relation between these two hydroxyls is trans it is called a β-anomer.

A reducing sugar in solution represents an equilibrium mixture of the free aldehyde, the aldehydrol, the two furanoses, two pyranoses, and possibly also the two septanose forms, in proportion determined by their relative thermodynamic stabilities.

In the presence of alcohol and catalytic amount of an acid, sugars give mixtures of alkyl α- and β-furanosides and pyranosides, and the composition of this mixture remains unchanged once the acid has been removed, the system is particularly suitable for the study of the formation of cyclic acetals. The reaction of D-xylose with methanolic hydrogen chloride under controlled conditions (46) has been studied in detail by gas-liquid partition chromatographic analysis of the products.

Samples were removed after various times, neutralized with sodium methoxide to stop the reaction, and evaporated to dryness. The residues were methylated by the Kuhn (47) procedure and the fully methylated methyl glycosides were analyzed. The components were identified by comparison with authentic samples of each of the four possible glycosides. The furanosides were the first compounds formed in the methanalysis of xylose. The following sequence of reactions occurs in the formation of methyl xylosides, xylose→furansides, →anomerization of furanosides, furanosides→pyranosides,→anomerization of pyranosides. It appears, therefore, that the furanose forms are the products of kinetic control, whereas the pyranose forms are the thermodynamically more stable products. Similar results (48) were obtained in case of other pentoses.

(47) R. Kuhn, H. Trischmann, and I. Löw, Angew. Chem. 67, 32 (1955)

Conformation of the pyranose forms

Hassel and Otter (49) suggested in 1947 that the facts then recently established for the conformation of the cyclohexane system could also be applied to sugars in their pyranose forms. Reeves, (50) by studying


the complexes formed from sugars and their derivatives in cuprammonium solution, deduced that the pyranoid sugars exist in chair conformations.
Reeves introduced the symbols C\text{I} and C\text{I}C to describe the two possible forms of pyranoses.

\begin{center}
\begin{tikzpicture}
\node at (0,0) {\includegraphics[width=0.4\textwidth]{diagram.png}};
\end{tikzpicture}
\end{center}

The chair conformations of cyclic sugars have been verified directly by X-ray crystallographic measurements in several examples, notably \(\alpha\)-D-glucose (51) and 2-amino-2-deoxy-\(\alpha\)-D-glucose hydrobromide (52, 53).

\begin{itemize}
\end{itemize}

R. U. Lemieux and his co-workers (54) studied the n.m.r. spectra (at 40 Mc.p.s.) of acetylated cyclitols and sugars and found that, in general, equatorial hydrogen atoms give rise to signals at lower field (by about 25 c.p.s.) than similar but axial hydrogen atoms.

\begin{itemize}
\end{itemize}
More important was the observation by Lemieux (54) that the first order spin-spin coupling constant for the hydrogens on neighboring carbon atoms is 2-3 times longer when the hydrogens are both axial (dihedral angle 180°) than when one or both of the hydrogens is equatorial (60°). This observation led to the development of the Karplus equation (55) which gives the relation between dihedral angle and coupling constant.

The proton on the anomeric carbon atom of the aldoses is deshielded by two oxygen atoms and gives rise to a signal at lower field than any of the other protons, it is therefore easily identified and studied.

The skew-boat, or twist, form of cyclohexane is less stable than the chair form, its energy has been calculated (56, 57) to be

\[ 1.6 \text{ k-cal.}/\text{mole} \text{ less than that of the true boat or } 5.3 \text{ k-cal.}/\text{mole} \text{ more than that of the chair form}. \]

It has been proposed (58, 59) that 1,2-alkyldene acetals of arabinopyranose, glucopyranose, and some related derivatives, adopt a skew form as the favored conformation.

In an undistorted chair, the distance between vicinal equatorial hydroxyl groups (\textit{trans}) is the same as that between an axial and an
equatorial hydroxyl group (cis). However, formation of the dioxolane ring, which cannot readily incorporate a dihedral angle of 60°, causes distortion of the chair. The energy required for such distortion is less in the case of cis than in that of trans-hydroxyl groups (60).


In their study, Coxon and Hall (58) measured the n.m.r. spectra of various 1,2-alkylidene acetics. From the vicinal coupling constants obtained, they concluded that the pyranose ring in these derivatives adopts a conformation essentially that of a skewed boat (VI).

\[
\begin{align*}
J_{1,2} & = 5.0 \\
J_{2,3} & = 2.7 \\
J_{3,4} & = 2.5 \\
J_{4,5} & = 9.0 \\
J_{5,6} & = 4.2
\end{align*}
\]

"1,2-alkylidene acetics of α-D-glucopyranoses"

It is immediately obvious from inspection of the coupling constants, that these compounds are unlikely to have the expected conformation (V) since this would require H-2, H-3, H-4 and H-5 to have axial orientation and hence \( J_{2,3} \approx J_{3,4} \approx J_{4,5} > 9 \) c.p.s. The H-4 - H-5 coupling constant has a value consistent with (V) but the small values found for the
H-2 — H-3 and H-3 — H-4 couplings are characteristic of ring hydrogens having approximately "gauche" orientation rather than "dixial" orientation. This indicates that some fundamental change of conformation must have accompanied the attachment of the 1,2-acetal ring.

Substituent orientation

Monosubstituted cyclohexanes may exist in two alternative chair conformations. Flipping of the chair interchanges the equatorial and axial substituents, and the molecule may be expected to exist predominantly in the conformation in which the substituent group is equatorial.


to non-bonded interaction (repulsion) between axial substituents in the syn-dixial (1, 3) positions, substituents are normally more stable in the equatorial position.

The substituent may be stabilized in the axial orientation by various factors such as hydrogen bonding, interaction of dipoles, and solvent effects. For example, it was found that methyl 2-deoxy-α-D-
erythropyranoside changes its conformation as the solvent is changed (63); in water it occurs preponderantly in the Cl conformation, in chloroform in the IC conformation. Presumably the latter is stabilized, in the non-hydroxylic solvent, by an internal hydrogen bond. (Fig. 2)

Figure 2.
Table 3

Experimental Free-Energy Differences between Equatorial and Axial Substituents (61, 62)

<table>
<thead>
<tr>
<th>Group</th>
<th>$-\Delta F$, kcal./mole</th>
<th>Group</th>
<th>$-\Delta F$, kcal./mole</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Range reported</td>
<td>Average value</td>
<td></td>
</tr>
<tr>
<td>OH</td>
<td>0.25-1.25;</td>
<td>0.70</td>
<td>NH$_2$</td>
</tr>
<tr>
<td>OAc</td>
<td>0.40-1.50;</td>
<td>0.70</td>
<td>NH$_3^+$</td>
</tr>
<tr>
<td>OTs</td>
<td>0.60-1.70;</td>
<td>0.70</td>
<td>N(CH$_3$)$_2$</td>
</tr>
<tr>
<td>OCH$_3$</td>
<td>0.60-0.75;</td>
<td>0.70</td>
<td></td>
</tr>
<tr>
<td>OC$_2$H$_5$</td>
<td>0.90-1.00;</td>
<td>0.90</td>
<td>CH$_3$</td>
</tr>
<tr>
<td>F</td>
<td>0.25</td>
<td></td>
<td>CO$_2$H$_2$</td>
</tr>
<tr>
<td>Cl</td>
<td>0.40</td>
<td></td>
<td>CO$_2$Et$_2$</td>
</tr>
<tr>
<td>Br</td>
<td>0.50</td>
<td></td>
<td>CN</td>
</tr>
<tr>
<td>I</td>
<td>0.40</td>
<td></td>
<td></td>
</tr>
<tr>
<td>SH</td>
<td>0.90</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Conformational and configurational studies by n.m.r. spectroscopy of a series of poly-O-acetylated aldopyranosyl halides revealed that the stable anomer in every case has the halogen atom axial when the molecule concerned is in its favored conformation (64). This preference for a halogen atom (or other polar group) at C-1 to adopt the axial orientation, as a thermodynamically more stable arrangement than the equatorial orientation, contrary to the usual order of conformational stabilities predicted from steric considerations, has been termed the anomeric effect (65, 63).


It has been attributed to the dipole-dipole interaction between the carbon-oxygen bonds of the ring and the bond from the anomeric carbon to the substituent (Fig. 3).

Anomeric effect (dipole moments)

Fig. 3
The furanose forms

Six membered carbocyclic rings are more stable than five-membered ones; the excess enthalpy of cyclopentane is about 6.5 kcal./mole higher than that of cyclohexane (65). Replacement of one methylene group by an oxygen atom would be expected to lessen the difference between stabilities of the five and the six-membered ring systems by reducing the eclipsed interaction of the hydrogen atoms in the former; the difference in stability between tetrahydrofuran and tetrahydropyran has been estimated, from measurements of the heats of combustion, as 2.9 kcal./mole (66). The chair form of a six-membered ring, having a fully staggered arrangement of hydrogen atoms throughout, can accommodate substituents with less strain than the five-membered ring, which necessarily has some partially eclipsed interactions. The difference in the stability between the two forms of cyclic compounds in the sugar derivatives is not large, and substituent groups, if suitably placed, may reverse this order of stability (63).

The conformations of furanoid sugars are believed to be similar to those of cyclopentane. The ring is not planar, and two conformations of low energy are possible, the envelope (C₅), having four atoms in a plane and one out of the plane, and the twist (C₂), having three atoms in plane, one above and one below.
The term twist is probably the best since terms such as skew (67) and half chair (68) already have accepted meanings in six-membered rings.


The symbols $C_s$ and $C_2$ (68) refer to the existence of a plane of symmetry and a two-fold symmetry in these respective forms. It is important to note that the puckering is not necessarily fixed but can be pictured as rotating around the ring by an up and down motion of the five methylene groups in what has been termed a "pseudorotation" (69).

Specific conformations, based upon n.m.r. spectroscopy, have been proposed for D-ribofuranose in purine and pyrimidine nucleosides (70) in thymidine (71), in certain deoxyribonucleosides (72),


nucleotides (73) and in 1,3,5-tri-O-benzoyl-α-D-ribofuranose, methyl


2,3-anhydro α-D-ribofuranoside, and its β-D-anomer (74). Hall (74)


reported the spin-spin coupling constants of the adjacent hydrogens (J values) of 1,3,5-tri-O-benzoyl-α-D-ribofuranose (VII). The coupling constants were related to the corresponding angles by using the modified Karplus equation.

\[ J = J_0 \cos^2 \theta - 0.28 \]

where \( J_0 = 9.26 \) for \( 0^\circ \leq \theta \leq 90^\circ \)

and \( J_0 = 10.36 \) for \( 90^\circ \leq \theta \leq 180^\circ \).

The calculations gave these angle values, angle H-1, H-2 = 48°, H-2 → H-3 = 32°; and H-3 → H-4 = 115° as the most probable values. Exact bond angles calculated from Karplus-type relationships must, however, be interpreted with caution.
In a planar 5-membered ring the projected valency angle between cis hydrogens is 0° and between trans hydrogens it is 120°. Since the H₃-H₄ is 115°, this section of the ring must be virtually unaltered by any deformation of the molecule. Hence C-2, C-3, C-4 and the ring oxygen must be essential coplanar, with C-1 deformed out of this plane. The molecule adopts, therefore, the so-called "envelope" conformation.

3. The Acyclic Forms

Solutions of pentoses and hexoses at equilibrium contains only very minor proportions of the open chain aldehyde forms (0.0036% reported in case of glucose) (75). The proportions of aldehyde are somewhat larger when all conformations of the pyranose and the furanose forms are unfavorable. Solutions of ribose at equilibrium appear to

contain an unusually high concentration (8.5%) of the aldehydo form (76). This suggests that hexoses that have an axial secondary hydroxyl group in the more stable chair conformation have a higher content of free aldehyde than does glucose. D-Idose has been reported (77) to give a positive Schiff reaction for aldehydes, unlike other sugars.

Steric effects may stabilize the aldehyde form in special cases; thus 3,6-anhydro-2,4-di-O-methyl-D-galactose (78) appears to exist in solution mainly in the free aldehyde form.

All aldehydo sugars mutarotate in alcohols due to the formation of hemiacetals. The hemiacetal of 2,3,4,5,6-penta-O-acetyl-aldehydo-D-galactose has been isolated in crystalline form (79). In water aldehydo sugars form aldehydrol (Fig. 4).


4. Pyranose-Furanose Interconversion

At equilibrium, most sugars exist preferentially in the pyranose rather than the furanose form. When a crystalline sugar (which usually consists of one tautomer only) is dissolved in water, the optical rotation of the solution changes until equilibrium is reached, this phenomenon is known as mutarotation. The relative free-energy content of the various tautomeric forms determines the proportion of each form present at equilibrium. The pattern of mutarotation can fall into one of two types (63). Sometimes the mutarotation may be described by the equation for a first-order reversible reaction, whether approached from the $\alpha$- or from the $\beta$-pyranose (simple mutarotation). In this case the equilibrium mixture consists principally of the two pyranose forms. This is the case for sugars such as xylose (VIII) and glucose (IX), which in the pyranose form can have all of the large
substituents in the favored equatorial orientation by adopting the favored chair conformation, but in the furanose form these sugars will possess both the 2- and 3-hydroxyl groups in axial orientation in the $C_5$ conformation. For these compounds, the furanose-to-pyranose equilibrium is far to the right, and the observed mutarotation is normal (80, 81).

(80) Ref. 65, p. 714.

Other sugars show "complex mutarotation" with an initial fast change, followed by a slower one, not necessarily in the same direction; more than two components are then involved in the equilibrium. The initial fast mutarotation is ascribed to a pyranose-furanose conversion, and the slow one to the anomerization of pyranose forms. The occurrence of an initial fast mutarotation can be taken as an indication that a substantial proportion of furanoses is present at equilibrium. This is
the case with arabinose, galactose, ribose and talose. In these sugars, the relative free-energy difference between the furanoid and pyranoid ring forms is less than that in the case of sugars that show simple mutarotation.

Formulas X and XI show that in the furanose forms, galactose and arabinose can have the 2,3 and 4 substituents in the equatorial orientation, whereas in the favored conformation of the pyranose form, the 4-hydroxyl is in the axial orientation.

5. Cyclic Forms of Amino Sugars

Amino sugars having the amino group at C-2 and C-3 resemble the simple sugars in that they can exist in the normal pyranoid or furanoid forms. The amino group, especially when it is at C-2, may influence the properties of the sugar. The glycosides of 2-amino-2-deoxy sugars are unusually stable toward acid hydrolysis. The low reactivity of the
glycosides has been related to the electrostatic repulsion of the 
\[ + \text{NH}_3 \] group toward hydronium ions approaching the glycosidic center (82).


The suggestion has been made that an ammonium group at C-2 that is axial in the favored conformation reduces the rate of protonation of the ring oxygen atom to such an extent that mutarotation becomes too slow to be observed (65). This viewpoint has been used to explain why mutarotation is not observed with 2-amino-2-deoxy-D-mannose hydrochloride. An opposite viewpoint has been advanced (83) to explain this lack of observed mutarotation in water, the ammonium ion is considered to act as a very effective proton donor to the ring oxygen atom, with the result that anomerization is fast, and equilibrated mixture of anomers is already present when the first polarimetric measurement is made after dissolution of the sample. This was proved by the fact that n.m.r. measurements showed the presence of both pyranose anomers in solution. It has also been shown that 2-amino-3,6-anhydro-2-deoxy-D-mannose hydrochloride does not exhibit observed mutarotation, but more than one tautomeric form is present in solution (84).

It has been also shown that, relative to a 2-hydroxyl group, an acetamido or ammonium group at C-2 exerts a stabilizing effect on a cis-related hydroxyl group at C-1 (85).


Amino sugars having the amino group at C-4 or C-5 are of particular interest because ring closure involves a possible competition between nitrogen and oxygen atoms. This can lead to a furanose or pyranose sugar having nitrogen as the ring hetero atom. A similar situation has been encountered in 5-thio sugars (86 - 89) and 4-thio sugars (90, 91),


where the ring hetero atom is sulfur. Results to date indicate that ring closure with sulfur is favored somewhat over oxygen. The position regarding nitrogen is not yet clear.
BACKGROUND OF THE PROBLEM

Interest in sugar derivatives containing nitrogen in the ring has grown rapidly since the first example was reported in 1962. These sugars are of interest from the standpoint of the configurational and conformational aspects of ring closure, their reactivity in comparison with conventional sugars, and their behaviour toward enzymes.

Synthesis of Pyranoid Sugars Having Nitrogen in the Ring

Pyranoid sugars in which the ring oxygen atom is replaced by nitrogen are now well known. They are generally obtained, as the stable N-acetyl derivatives, from 5-acetamido-5-deoxyaldoses. Equilibration of the various possible tautomeric forms in solution generally gives a preponderance of the six-membered ring form.

The first sugar having nitrogen in the ring was announced nearly simultaneously, from two laboratories. Jones and Turner (92) prepared


a 5-acetamido-5-deoxy-L-arabinopyranose. The other report, a communication by Paulsen (93) concerned the synthesis of 5-acetamido-5-deoxy-D-xylopyranose

(93) H. Paulsen, Angew. Chem. 74, 901 (1962).

by a nearly identical procedure.

5-Acetamido-5-deoxy-L-arabinose

This compound was required as a model for a study of microbiological
oxidation. It was prepared (92) by hydrolyzing the isopropylidene acetal ring of 5-acetamido-5-deoxy-1,2-0-isopropylidene-β-L-arabinose.

\[
\begin{align*}
1. & \text{MeOH/} \text{NH}_3 \\
2. & \text{Ac}_2\text{O-H}_2\text{O} \\
3. & \text{H}^+ \\
\end{align*}
\]

The compound was found to exist in two forms (XIII and XIV). At equilibrium in aqueous solution, the furanose form (XIII) was favored by 4 to 1 (see Fig. 5). The two forms were chemically and physically distinct, and were separable by cellulose-column chromatography. Both compounds gave the same crystalline phenylosazone and on reduction both forms gave 5-acetamido-5-deoxy-L-arabinitol. The furanose structure of compound (XIII), which was obtained as a sirup, was assigned on the basis of its infrared spectrum and by periodate
Figure 5.
oxidation. It showed bands at 3.03 \(\mu\) (OH and NH), the amide carbonyl absorption (Amide I band) at 6.13 \(\mu\) and the amide NH absorption (Amide II band) at 6.43 \(\mu\). All amides show a carbonyl absorption known as the amide I band. Simple open chain secondary amides absorb near 1640 cm\(^{-1}\) (6.10 \(\mu\)) while the carbonyl absorption of tertiary amides occurs in the range 1670-1630 cm\(^{-1}\) (5.99 - 6.14 \(\mu\)). Secondary amides display an amide II band in the region of 1570-1515 cm\(^{-1}\) (6.37 - 6.60 \(\mu\)) \(\text{(94)}\).


The pyranose form was obtained as a white amorphous powder, which showed no infrared absorption band for the NH group. Both compounds were stable at room temperature, but addition of a drop of ammonia to the aqueous solution resulted in rapid equilibration to give a 2:1 mixture of (XIII) and (XIV). Equilibration was also catalyzed by acetic acid, but much less effectively. The equilibrium mixture was also formed when an aqueous solution of either compound was heated.

5-Acetamido-5-deoxy-L-arabinopyranose (XIV) and 5-acetamido-5-deoxy-L-arabinofuranose (XIII) have also been prepared \(\text{(95)}\) by another route.


Treatment of 5-0-\(\text{p}\)-tolylsulfonyl-L-arabinose diethyl dithioacetal with an excess of sodium azide in methyl sulfoxide for 12 hrs. at
80-85° afforded crystalline 5-azido-5-deoxy-L-arabinose diethyl dithioacetal in 80% yield. Reduction of the latter compound with lithium aluminum hydride in ether followed by N-acetylation of the product yielded crystalline 5-acetamido-5-deoxy-L-arabinose diethyl dithioacetal. Demercaptalation of this dithioacetal with mercuric chloride and cadmium carbonate in aqueous solution gave a mixture of compounds (XIII) and (XIV). The n.m.r. spectrum of (XIV) showed two peaks at \( \tau 7.41 \) and 7.37 ascribed by the authors (95) to the N-acetyl methyl groups of the two possible anomeric structures of the pyranose ring. There was a doublet at \( \tau 3.95 \) assigned to the C-1 axial hydrogen and another doublet at \( \tau 3.52 \) assigned to the C-1 equatorial hydrogen. The spectrum of (XIII) showed the N-acetyl methyl group as singlet at \( \tau 7.53 \).

5-Acetamido-5-deoxy-D-xylose

The synthesis of this compound was noted briefly (93) and subsequently described in full by Paulsen (96). The key step involved

\[ (\text{96}) \text{ H. Paulsen, Ann., 670, 121 (1963).} \]

hydrolysis of 5-acetamido-1,2-\( \text{O} \)-cyclohexylidene-5-deoxy-D-xylofuranose with 0.1 N hydrochloric acid for 2 hours at 70° followed by neutralization. The pyranose form having nitrogen in the ring was crystalline, and was favored over the sirupy furanose form by a ratio 2:1. In the xylo configuration, all substituents in the pyranose ring are equatorial, whereas substituents at C-3 and C-4 are cis to each other in the five-membered ring. In the arabino configuration the pyranose-furanose ratio was 1:4. The furanose form appears to be favored, because there...
is one axial substituent in the pyranose ring whereas all substituents are \textit{trans} in the furanose ring-form. (See Fig. 5)

5-Acetamido-5-deoxy-D-xylose has also been prepared by Hanessian and Haskell (95) by essentially the same procedure. Hydrolysis of 5-acetamido-5-deoxy-1,2-\textit{o}-isopropylidene-D-xylofuranose was effected in dilute sulfuric acid (pH 1.3 - 1.6) for 3 or 4 days at room temperature. Aqueous solutions of both pyranose and furanose forms did not mutarotate at room temperature during 72 hours. A change in optical rotation was observed in acidified solutions and the equilibrium was attained in 24 hours in case of the pyranose form and 32 hours in case of the furanose form. Equilibration in aqueous ammonia was much faster (2 hours). The pyranose ring form, which has an acetylated nitrogen atom as the hetero atom, gave an n.m.r. spectrum in deuterium oxide that showed two distinct peaks for the N-acetyl methyl group at \(\tau\) 7.37 and 7.33, which were attributed to the two possible anomeric structures. Although evidence indicating the formation of anomers was not secured from mutarotation studies, the presence of anomers was suggested by the appearance of a doublet at \(\tau\) 3.95 attributed to the C-1 axial hydrogen and \(\tau\) 3.55 attributed to the C-1 equatorial hydrogen.

Szarek, Wolfe, and Jones (97) gave another interpretation of the n.m.r.


They observed that at 80° the two resonances for the C-1 (anomeric) proton were merged, and the two signals for the N-acetyl group were collapsed to a single peak. This evidence suggests that
5-acetamido-5-deoxy-D-xylopyranose exists in two rotational isomers because of restricted rotation about the CO-N-bond, and the observed spectrum was not that of a mixture of anomers. The doublet pattern is restored on cooling to room temperature. This behavior is analogous to that of several N-methylamides (98, 99).


Hindered internal rotation in
"5-acetamido-5-deoxy-D-xylopyranose"

Jones and Szarek (100) independently prepared the two isomers of 5-acetamido-5-deoxy-D-xylose by the method of Hanessian and Haskell.

except that hydrolysis of the isopropylidene ring was accomplished by using 20% aqueous acetic acid at 90° for 1.5 hours.

Lamchen and Whistler (101) reported an alternative synthesis from


1,2-0-isopropylidene-α-D-glucofuranose. The procedure involved 5,6-glycol cleavage, oximation of the resultant aldehyde, followed by reduction, N-acetylation, and acid hydrolysis of the isopropylidene group.

5-Acetamido-5-deoxy-D-ribose (95)

Treatment of 2,3-0-benzylidene-5-0-γ-tolylsulfonyl-β-D-ribofuranose with sodium azide in methyl sulfoxide afforded crystalline 5-azido-2,3-0-benzylidene-5-deoxy-β-D-ribofuranose. This was treated with benzyl alcohol containing 1% hydrogen chloride, and thus sirupy benzyl 5-azido-2,3-0-benzylidene-5-deoxy-D-ribofuranoside was obtained. Reduction of the latter with lithium aluminum hydride in ether followed by N-acetylation gave benzyl 5-acetamido-2,3-0-benzylidene-5-deoxy-D-ribofuranoside as a sirup. Reduction of this sirupy compound with hydrogen and a palladium catalyst in ethanol afforded a mixture of 5-acetamido-5-deoxy-D-ribopyranose and 5-acetamido-5-deoxy-D-ribofuranose. In contrast to the corresponding derivatives of the xylose and arabinose series, the six-membered form in the ribose series was formed in very small proportion (Fig. 5). The furanose ring structure was ascertained by the detection of an amide II band in the infrared spectrum.
5-Acetamido-5-deoxy-L-xylose.

Haskell and Hanessian (31) studied the structure of the antibiotic paromomycin. Methyl paromobiosaminide dihydrochloride (XV), a product obtained from methanolysis of the antibiotic, was treated with ethanethiol in the presence of fuming hydrochloric acid to yield paromose diethyl dithioacetal dihydrochloride. Conventional acetylation followed by 0-deacetylation gave crystalline N, N'-diacetyl paromose diethyl dithioacetal. Oxidation of the latter with peroxypropionic acid in aqueous methanol at -10° afforded crystalline N, N'-diacetyl-1,1-bis (ethylsulfonyl)-1-deoxy-paromitol (XVI). Alkaline degradation of (XVI) afforded 5-acetamido-5-deoxy-L-xylofuranose (XVIII) and 5-acetamido-5-deoxy-L-xylopyranose (XVII). This degradation scheme follows the conditions of the MacDonald-Fischer method for chain descent in the sugars (102 - 104).

Compounds XVII and XVIII were separated by cellulose chromatography as a colorless sirup and crystalline solid. Solutions of XVII or XVIII
were stable in water. Traces of acids and basic impurities, or heating

(102) D. L. MacDonald and H. O. L. Fischer, J. Am. Chem. Soc.,
74, 2087 (1952).

(103) D. L. MacDonald and H. O. L. Fischer, Biochim. Biophys.
Acta, 12, 203 (1953).

(104) R. Barker and D. L. MacDonald, J. Am. Chem. Soc., 82, 2297
(1960).
solutions of pure XVII or XVIII, caused the other isomer to appear revealed by paper chromatography. This investigation prompted the subsequent synthesis of model compounds in the Parke-Davis laboratories.

Transformations of 5-Amino Sugars Into Pyridine Derivatives

In a preliminary communication (105), Paulsen reported an attempt


to obtain a free 5,6-diamino sugar by acid hydrolysis of the blocked derivative (XIX). It was found, however, that three moles of water were spontaneously eliminated, and a pyridine derivative (XX)

was obtained. A sugar derivative having a nitrogen atom in the ring is evidently involved as intermediate species. A detailed paper on the preparation of 5,6-diamino-5,6-dideoxy-D-glucose and L-idose and their

\[
\begin{align*}
(XIX) & \quad \xrightarrow{\text{Acid Hydrolysis}} \quad (XX) \\
\text{Acid Hydrolysis} & \\
\end{align*}
\]
transformation into pyridine derivatives has been also published (106).


Whistler and co-workers (107) subsequently reported the synthesis

(107) R. E. Gramera, R. M. Bruce, S. Hirase, and R. L. Whistler,

of methyl 5-amino-5-deoxy-L-idofuranoside. The compound contains a free
amine group at C-5. It is the only form obtained under the conditions
employed for methyl glycoside formation. 3-O-Benzyl-1,2:5,6-di-O-
isopropylidene-α-D-glucofuranose was hydrolyzed selectively to remove
the 5,6-O-isopropylidene group. Subsequent tritylation gave
crystalline 3-O-benzyl-1,2-O-isopropylidene-6-O-trityl-α-D-glucofuranose.
P-Toluenesulfonation of the latter compound afforded the 5-p-tolylsulfonyl
derivative which on hydrazinolysis in absolute butanol gave a smooth
N\textsubscript{2} displacement of the 5-p-tolylsulfonyloxy group with the formation
of the 5-deoxy-5-hydrazino-L-idofuranose derivative. Hydrogenolysis of
the latter with Raney nickel did not remove completely the benzyl and
trityl groups, therefore the resulting sirup was further reduced with
da palladium on carbon to produce 5-amino-5-deoxy-1,2-O-isopropylidene-
β-L-idofuranose. Methanolysis of the 5-amino derivative furnishes
sirupy methyl 5-amino-5-deoxy-α, β-L-idofuranoside. The furanose ring
structure of this compound was proved by periodate oxidation. It consumed
two moles of periodate per mole with the release of one mole of
formaldehyde. A positive 5-nitrosalicylaldehyde test indicated that
the product contained a free amino group.
Acid hydrolysis of 5-amino-1,2-0-cyclohexylidene-5-deoxy-D-xylofuranose hydrochloride XXI (Fig. 6) gave 3-hydroxypyridine, XXXI, together with 1,5-dideoxy-1,5-imino-D-threo-pentulose hydrochloride hydrate XXX (108, 109). The latter product presumably arises from


the intermediate XXIV by the Amadori rearrangement. The proportion of the two products depends on the hydrolysis conditions. Thus when XXI in HCl hydrochloric acid was kept for 6 days at 20°, neutralization dilution with ethanol, and evaporation gave crystalline XXX in 40% yield, and no XXXI was found in the mother liquor. On the other hand, hydrolysis of XXI in 0.1N hydrochloric acid for 2.5 hrs. at 70° gave XXX in 16% yield, the mother liquor gave XXXI, m.p. 108-110° in 56% yield.

However, treatment of the blocked 5-amino sugar derivative XXI with absolute methanol containing 0.5% hydrogen chloride under strictly anhydrous conditions lead to cleavage of the O-cyclohexylidene group and the formation of crystalline methyl 5-amino-5-deoxy-β-D-xylofuranoside hydrochloride XXII as the major product. The minor product was the α-D-glycoside, which remained as sirup. Traces of 3-hydroxypyridine, also present in the reaction mixture, were believed to arise by the presence of some moisture in the methanolic solution. The course of the methanolysis reaction, to give the furanoside and none of the nitrogen-containing six-membered ring-form is predictable; the protonated amino group would have little or no tendency to act as a nucleophile in reactions at C-1.
Figure 6.
The β-D-configuration was assigned to compound XXII on the basis of its optical rotation and n.m.r. spectrum. The coupling constant $J_{1,2}$ was 0.3 c.p.s., a value that corresponds to a dihedral angle between H-1 and H-2 approximately of 90°. The furanoside XXIII was assigned the α-D-configuration. It had a high positive rotation and the $J_{1,2}$ value was 4.2 c.p.s. which corresponds to a dihedral angle of about 40°. It is known that the negligible coupling ($J \leq 1$ c.p.s.) is only possible between trans protons at C-1 and C-2 of the D-ribo-furanose ring (that is, β-D-configuration at C-1). (110).


Paulsen prepared 5-benzylamino-1,2-0-cyclohexylidene-5-deoxy-L-idofuranose. Hydrolysis of the cyclohexylidene ring with 2 N hydrochloric acid for 2 hours at 70°, lead to the formation of N-benzyl-5-hydroxy-2-hydroxymethyl-pyridinium chloride salt (111).


The first report of a sugar having an unsubstituted NH group in the ring is the preparation of 5-amino-5-deoxy-D-xylopyranose XXXII obtained as sirup by Paulsen and co-workers by scheme shown (Fig. 7) (112).


This compound is very sensitive to acid but is stable to alkali; $N$-acetylation gave the known crystalline 5-acetamido-5-deoxy-D-xylopyranose.
The same compound XXXII has been prepared also by Hanessian (113)


by hydrogenation of 5-azido-5-deoxy-D-xylose.

Figure 7.

Conclusions
1. Interest in sugar derivatives containing nitrogen as the hetero atom is rapidly growing in connection with their use as model compounds in various chemical reactions, and with their behavior as unnatural sugars in the study of some biological functions.
2. The existing methods for the synthesis of all these "heteroses" involve the generation of the aldehyde group by mild deblocking methods in the presence of an acetamido group. The product thus obtained consists of a mixture of pyranose and furanose forms, the proportion of which depends on conformational factors. (See Fig. 5).

3. A free amino group at C-5 interacts with the free aldehyde of the sugar to give a 6-membered piperidine-type ring which, under acidic conditions, either loses 3 moles of water to give a pyridine derivative or undergoes the Amadori rearrangement. (Fig. 6).

4. The 5-amino-5-deoxy sugars are only stable under alkaline conditions. Under acidic conditions dehydration occurs to give a pyridine derivative.

**Synthesis of 4-Amino Sugars**

**4-Amino-4-deoxy-D-glucose**

Baker and co-workers (114) tried to synthesize 4-amino-4-deoxy-D-

(114) E. J. Reist, R. R. Spencer, B. R. Baker and L. Goodman,

glucose by a route involving direct displacement of a 4-(methylsulfonyl) oxy group by azide ion in a blocked D-galactose derivative, and the product was isolated as the crystalline methyl α-D-pyranoside. Their attempts to hydrolyze methyl 4-amino-4-deoxy-D-glucoside gave inconclusive results. The amorphous material obtained showed two components by paper chromatography and gave a poor analysis for 4-amino-4-deoxy-D-glucose hydrochloride. The authors stated that possible involvement of the 4-amino group in ring formation, to give a pyrrolidine type
molecule, may have been partially responsible for the difficulties encountered in the hydrolysis.


of derivatives of L-amino-4-deoxy-D-glucose. The acid hydrolysis step was avoided by using the baselabile acetyl group at C-1, and 4-azido-4-deoxy-D-glucose XXXIV was obtained as shown.

\[
\begin{align*}
\text{XXXIII} & \quad \text{Deacetylation} \quad \text{XXXIV} \\
\end{align*}
\]

Hydrogenation of the azide XXXIV yielded a sirup that did not reduce Benedict or aniline citrate reagents. An n.m.r. spectral analysis of the sirup and its acetate indicated a hydroxylated pyrrolidine structure. The authors suggested the following sequence of transformations:
The loss of reducing character was attributed to the fact that the product contained no potential carbonyl group. It was proposed that, during hydrogenolysis, the 4-amino-4-deoxy-D-glucose first formed subsequently underwent formation of a cyclic Schiff base (1-pyrroline A). The latter then underwent rapid hydrogenation to the pyrrolidine B, a compound which gives a negative test with the reagents used for detecting reducing sugars. Acetylation of the crude hydrogenation product (presumably B) gave a substance whose infrared and n.m.r. spectra were compatible with the structure C. Thus the i.r. spectrum showed an amide I band at 6.0 μ but no amide II band at 6.50 μ, indicating that the amide was tertiary. The n.m.r. spectrum showed the presence of 15 acetate methyl protons at τ 7.88 - 7.98.

Ammonolysis of 1,6-anhydro-2,4-di-O-p-tolylsulfonyl-β-D-glucopyranose
gave one diamino and two monoamino derivatives (116). The structure of the first product was established as a 2,4-diamino-2,4-dideoxy derivative of D-glucose. One of the two monoamino compounds is presumably a derivative of 4-amino-4-deoxy-D-glucose. The anhydro ring could be cleaved most successfully by acetylation followed by acetalolysis, and crystalline derivatives isolated included 4-acetamido-4-deoxy-β-D-glucopyranose, which gave negative Morgan-Elson reaction (117), indicating that it was not a 2-acetamido-2-deoxy sugar. However, attempts to obtain 4-amino-4-deoxy-D-glucose hydrochloride in crystalline form failed.

4-Amino-4,6-dideoxy-D-glucose (viosamine)

Stevens et al. (118) reported the complete identification and synthesis of viosamine, 4-amino-4,6-dideoxy-D-glucose, isolated from the lipopolysaccharide of Chromobacterium violaceum. This amino sugar also occurs in extracts of Escherichia coli strain B in the form of its thymidine 5-pyrophosphate.
Viosamine has been also isolated from *Streptomyces plicatus* and *S. vinaceus-drappus*. (119).


The starting material for synthesis of viosamine hydrochloride was methyl 2,3-di-\(\beta\)-benzyoyl-4,6-di-\(\beta\)-tolylsulfonyl-\(\alpha\)-D-galactopyranoside. The latter was heated at 110° with 1.5 equivalent of sodium iodide in acetone, and alumina chromatography of the product gave the crystalline 6-iodo-compound in 32% yield, and 20% of the starting material was recovered. A third reaction product was isolated as an oil, and its physical properties and elemental analysis indicated that it was a dideoxy-dideo sugar derivative. The high yield of this compound (45%) indicates that the primary and secondary \(\beta\)-tolylsulfonyloxy groups, at C-6 and C-4, are of comparable reactivity toward nucleophilic displacement. The relative reactivity at C-6 and C-4 in the gluco configuration (120) is in fair agreement with the ≈ 100-fold difference in reactivity between primary and secondary alkyl bromides in simple systems (121). Hydrogenolysis of the 6-iodo group with Raney


nickel was facile and gave the 6-deoxy derivative.

Treatment of methyl 2,3-di-O-benzoyl-6-deoxy-4-O-2-tolylsulfonyl-α-D-galactopyranoside XXXVI with lithium azide in N,N-dimethylformamide at 125° for 20 hours followed by reduction with platinum catalyst followed by saponification with barium hydroxide, gave methyl 4-amino-4,6-dideoxy-α-D-glucopyranoside XXXVII.

\[
\text{XXXVI}
\]

\[
\text{XXXVII}
\]

Many attempts to hydrolyze the methyl α-D-glycoside XXXVII failed to give a crystalline product. N-acetylation of XXXVII followed by hydrolysis with 2.5 N hydrochloric acid for 6.5 hr. at 100° gave crystalline viosamine hydrochloride which was identical with the natural product.

Viosamine has been prepared too by a route involving double inversion at C-4, (Fig. 8) (122, 119). This is a superior method which

used the readily available methyl α-D-glucopyranoside.

Methyl 2,3-di-O-benzyl-6-deoxy-4-O-(methylsulfonyl)-α-D-glucopyranoside XXXVIII was treated with sodium benzoate in N,N-dimethylformamide and the product was saponified to give methyl 2,3-di-O-benzyl-6-deoxy-α-D-galactopyranoside. Mesylation of the latter gave methyl 2,3-di-O-benzyl-6-deoxy-4-O-(methylsulfonyl)-α-D-galactopyranoside XXXIX. The methylsulfonyloxy group at C-4 in XXXIX was displaced with inversion by an azido group by treatment with sodium or lithium azide in N,N-dimethylformamide and the azido group was then reduced with lithium aluminum hydride to give oily methyl 4-amino-2,3-di-O-benzyl-4,6-dideoxy-α-D-glucopyranoside which was characterized as its hydrochloride XL. The benzyl ether groups were removed by hydrogenation to give methyl 4-amino-4,6-dideoxy-α-D-glucopyranoside XLI. Compound XLI was N-acetylated and then hydrolysed (2.5 N hydrochloric acid at 98-100° for 6.5 hrs.) to give viosamine.

Figure 8.
4-6-Dideoxy-4-dimethylamino-D-glucose (Amosamine)

The N-dimethylation of methyl 4-amino-4,6-dideoxy-α-D-glucopyranoside was performed by the Clark-Eschweiler procedure to give an 87% yield of the product, and hydrolysis of the latter gave amosamine. (119).

4,6-Dideoxy-4-methylamino-D-glucose (Bamosamine) (119)

Methyl 4-amino-2,3-di-O-benzyl-4,6-dideoxy-α-D-glucopyranoside was treated with ethyl chloroformate, and the resultant N-carbethoxy derivative was reduced with lithium aluminum hydride in refluxing dioxane. Hydrogenolytic cleavage of the benzyl groups from the product was smoothly accomplished in the presence of acid and afforded methyl 4-methylamino-4,6-dideoxy-α-D-glucopyranoside. Attempts to hydrolyse this product, to give bamosamine (a component moiety of the antibiotic bamicetin) were not successful.

4-Amino-4,6-dideoxy-D-galactose (123, 120)


Methyl 4,6-O-benzylidene-α-D-glucopyranoside was benzylated to give the dibenzyl derivative. Mild hydrolysis gave methyl 2,3-di-O-benzyl-α-D-glucopyranoside. Mesylation gave the crystalline methyl 2,3-di-O-benzyl-4,6-di-O-(methylsulfonyl)-α-D-glucopyranoside. Sodium iodide in butanone at reflux selectively displaced the primary mesyloxy group to give methyl 2,3-di-O-benzyl-6-iodo-4-O-methylsulfonyl-α-D-glucopyranoside. The primary iodo group could be smoothly reduced to the
6-deoxy derivative by treatment with lithium aluminum hydride in tetrahydrofuran. The resulting methyl 2,3-di-O-benzyl-6-deoxy-4-methylsulfonyl-α-D-glucopyranoside was heated with lithium azide in N,N-dimethylformamide to give methyl 4-azido-2,3-di-O-benzyl-4,6-dideoxy-α-D-galactopyranoside. This compound was reduced with lithium aluminum hydride in refluxing dioxane to methyl 4-amino-2,3-di-O-benzyl-4,6-dideoxy-α-D-galactopyranoside. The benzyl groups were cleaved by catalytic hydrogenation over palladium on carbon in the presence of two moles of hydrochloric acid, to give the crystalline hydrochloride of methyl 4-amino-4,6-dideoxy-α-D-galactopyranoside. Attempted hydrolysis of the latter with dilute acid failed to give a crystalline product. Examination of the reaction mixture by paper chromatography showed some reducing material along with large quantities of the starting material. Under vigorous hydrolytic conditions extensive decomposition occurred. It was claimed that methyl 4-acetamido-4,6-dideoxy-α-D-galactopyranoside, on heating for 3 hr. in 3N hydrochloric acid at 100° removed only the N-acetyl group, and the glycoside group was not cleaved. (Identical conditions with the gluco isomer were successful for hydrolysis of the glycoside). An alternative approach for hydrolysis of the glycoside was by peracetylation with acetic anhydride and pyridine, followed by acetylation with acetic anhydride and sulfuric acid. Chromatography of the product afforded 4-acetamido-1,2,3-tri-O-acetyl-4,6-dideoxy-α-D-galactopyranose, which on hydrolysis with 3N hydrochloric acid at 55° for 5 hours gave the free sugar, isolated as an amorphous solid, in 31% yield. The 4-monomethylamino glycoside, and the 4-dimethylamino derivative of the free sugar have been also prepared.
Acetolysis of methyl 4-acetamido-2,3-di-O-benzyl-4,6-dideoxy-\(\alpha\)-D-galactopyranoside (120) gave a mixture of products. One of the major components (30\%) gave a positive bromine test for unsaturation but it failed to reduce Fehling or Benedict's reagents. No amide II band was observed in the i.r. spectrum, and the compound was tentatively assigned the structure (XLIII).

\[
\text{Ac} \\
\text{N} \\
\text{OH} \\
\text{OH} \\
\text{CH}_3 \\
\text{OCH}_2\text{Ph}
\]

\text{XLIII}

Derivatives of 4-amino-4-deoxy-D-(and L)-mannose (124)


The 1,6-anhydro derivative of 4-amino-4-deoxy-\(\beta\)-D-mannose compound formed the major product in the ammonolysis of 1,6:3,4 dianhydro-\(\beta\)-D-talo-pyranose XLIII. It is known that the opening of sugar epoxides with ammonia or amines lead to the formation of two possible isomers. If the epoxide is part of a rigid six-membered ring system, the product that results from trans diaxial ring opening is preponderant (Furst-Plattner rule) (125).

Methyl 4,6-dideoxy-4-dimethylamino-α-L-mannopyranoside formed
the minor product of the reaction of methyl 3,4-anhydro-6-dideoxy-α-L-
talopyranoside with dimethylamine (126).

(126) J. Jary, K. Capek, and J. Kovar, Collection Czechoslov.

Derivatives of 4-amino-4-deoxy-D-talose

Oxidation of 1,6-anhydro-2,3-O-isopropylidene-β-D-mannopyranose
(XLIV) with ruthenium tetroxide in carbon tetrachloride, or with methyl
sulfoxide - acetic anhydride gave 1,6-anhydro-2,3-O-isopropylidene-
D-lyxohexopyranose-4-uloose (XLV) (127, 128). Oximation of the latter,


followed by reduction with hydrogen using platinum oxide as a catalyst, in the presence of hydrochloric acid, afforded the hydrochloride of 1,6-anhydro-4-amino-4-deoxy-β-D-talopyranose XLVII, which was N-acetylated to give the corresponding 4-acetamido compound.

Methyl 4-acetamido-4,6-dideoxy-α-D-talopyranoside was prepared by oxidation of methyl 6-deoxy-2,3-α-isopropylidene-α-D-mannopyranoside to the 4-ulo derivative, followed by stereoselective reduction of the oxime, with lithium aluminum hydride, N-acetylation, and selective hydrolysis of the α-isopropylidene group (129).

Derivatives of 4-amino-4-deoxy-D-gulose (130)


Methyl 4-O-acetyl-6-deoxy-2,3-di-O-(p-tolylsulfonyl)-α-D-glucopyranoside was treated with base, and the resultant 3,4-anhydro ring was opened by azide ion. Catalytic hydrogenation of the azide, followed by reductive desulfonation and N-acetylation gave the methyl 4-acetamido-4,6-dideoxy-α-D-gulopyranoside.

4-Amino-2,7-anhydro-4-deoxy-β-D-heptulopyranoses (131)


When the dialdehyde XLVIII (obtained by periodate oxidation of 2,7-anhydro-β-D-altro-heptulopyranose) was condensed in methanolic solution with one molecular equivalent of nitromethane in the presence of one molecular equivalent of sodium methoxide, a mixture of three stereoisomeric aci-nitro sodium salts was obtained. Deionization gave the free 2,7-anhydro-4-nitro-4-deoxy-β-D-heptulopyranoses (L). Catalytic hydrogenation of the nitrodeoxyheptulosans L afforded the corresponding 2,7-anhydro-4-amino-4-deoxy-β-D-heptulopyranoses, which were isolated as their crystalline hydrochlorides LI.

The formation of the aci-nitro sodium salts during the cyclization reaction involves the creation of two asymmetric centers at position 3 and 5, and four isomers are possible. Acification generates a new asymmetric center at C-4, and eight isomers of L are possible.
However, Richardson and Fischer (132), having investigated the
course of the nitromethane condensation with the homologous dialdehyde
from levoglucosan, suggested that the nitro substituent would adopt
almost exclusively the more stable equatorial position. This is due
to the large steric influence which an axial NO₂ would encounter from
the anhydride bridge. Similarly, Baer suggested that the three products
I (and hence II too) carry equatorially disposed nitrogen atoms, that
is, they possess any three of the D-gulo, D-altr o, D-allo and D-ido configuration.

The configurations of the three isomers that were isolated have been assigned by n.m.r. (133), by a study of the spectra of the fully acetylated amino sugar derivatives (See table 4).

**Table 4**

Chemical shifts of some heptulosan derivatives (τ values)

<table>
<thead>
<tr>
<th>Compound (Configuration)</th>
<th>H₂</th>
<th>H₅</th>
<th>1-OAc</th>
<th>3-OAc</th>
<th>NHAc</th>
<th>5-OAc</th>
<th>NH</th>
</tr>
</thead>
<tbody>
<tr>
<td>I (D- allo)</td>
<td>4.77</td>
<td>5.13</td>
<td>7.91</td>
<td>7.84</td>
<td>8.02</td>
<td>7.81</td>
<td>4.38</td>
</tr>
<tr>
<td>II (D-gulo)</td>
<td>4.71</td>
<td>4.89</td>
<td>7.94</td>
<td>7.88</td>
<td>8.11</td>
<td>7.94</td>
<td>4.11</td>
</tr>
<tr>
<td>III (D-altr o)</td>
<td>4.89</td>
<td>4.97</td>
<td>7.93</td>
<td>7.93</td>
<td>8.08</td>
<td>7.83</td>
<td>4.03</td>
</tr>
</tbody>
</table>

Although the assignments of the H-5 and H-9 hydrogens are unequivocal in all cases, the remaining assignments are only tentative, and are based on the widths calculated from the splitting of the H-5 and H-5 multiplets. Lemieux et al. (134) found that, in carbohydrates of pyranose structure, axial acetoxy substituents usually resonate at lower field (τ value) than equatorial substituents.

One of the compounds (LII) (Fig. 9) showed acetoxy resonances at τ 7.81, 7.84 and 7.91, which indicated that it had two axial and one
equatorial acetoxy groups and hence that it had the D-allo configuration. Both of the other compounds exhibited one axial and two equatorial acetoxy resonances so that they were apparently the D-altro and D-gulo isomers. It was possible to distinguish between the isomers LIII and LIV (Fig. 9) by a study of the ring-hydrogen resonances. It was

\[
\begin{align*}
\text{D-\,allo} & \\
\text{III} \\
\text{D-\,gulo} & \\
\text{III} \\
\text{D-\,altro} & \\
\text{LIV}
\end{align*}
\]

\text{Figure 9.}

assumed that the acetamido substituents at C-4 was equatorial in these compounds, and thus H-4 was axial. The D-gulo isomer LIII, bearing an axial acetoxy at C-3, showed a doublet (H-3) at \( \tau \) 4.71, \( J_{3-4} \) 4.6 c.p.s.
and three lines of a partially concealed quartet assigned to H-5, at $\tau$ 4.89 ($J_{5,6}$ 3.5, $J_{5,4}$ 10.4 c.p.s.). The isomer assigned as D-altro showed the H-3 doublet at $\tau$ 4.89 ($J_{3,4}$ 9.9 c.p.s.) and a partially concealed quartet for H-5 at $\tau$ 4.97 (splittings 2.1 and 4.4 c.p.s.), all of which is consistent with the assigned configuration (LIV). In case of the D-allo isomer, the H-3 signal was observed as a poorly resolved doublet at $\tau$ 4.77 ($J_{3,4}$ 4 c.p.s.).

Synthesis of specifically deuterated derivatives, and direct comparison by n.m.r. of the deuterated and non-deuterated compounds, provides a completely unambiguous method for assigning specific signals in n.m.r. spectra. It has been shown that the signal of the N-acetyl methyl group is not the highest-field acetyl group signal in some pyranose pentaacetates derivatives of amino sugars, especially when an aryl group is present e.g., trityl (135).


Methyl 4-amino-4-deoxy-α-D-lyxoside

Methyl 4-amino-4-deoxy-α-D-lyxoside has been prepared by Overend et al. (136). N-Substituted derivatives of this glycoside were obtained


by heating methyl 3,4-anhydro-β-L-riboside in a sealed tube for 24 hours at 100° with aqueous or alcoholic solutions of the appropriate amine. Attempts were made to obtain the amino sugars from their glycosides by
hydrolysis with 1.0 and 0.1 N hydrochloric acid. 4-Deoxy-4-dimethyl-
amino-D-lyxose was obtained as the hydrochloride, but the other experi-
ments were unsuccessful and only a black tar was obtained after heating
compounds for 10 minutes in the acid. Paper chromatographic examination
of the tar indicated the presence of pyrrole-like substances. At
least two reducing spots and several ninhydrin-sensitive materials
producing both yellow and mauve spots were detected. Treatment with
2 dimethylamino[n]benzaldehyde in hydrochloric acid (Ehrlich's reagent)
(117) produced faint pink spots. The formation of red pigments with
Ehrlich's reagent in hydrochloric acid solution is a characteristic of
several classes of compound including α and β-substituted pyrroles (40,41).
The behavior of these glycosides resemble that of the glycosides of
N-acetylneuraminic acids (137) in dilute acids, they produce large

(137) Gottschalk, "The Chemistry and Biology of the Sialic acids"
1960, Cambridge University Press.

amounts of tar and pyrrole-like substances.

Conclusions

The various methods for synthesizing 4-amino sugars can be summarized
as:

1. Direct S\textsubscript{N}2 displacement of a 4-(methylsulfonyl)oxy or (p-tolylsulfonyl)-
oxy group by azide ion in a suitably blocked sugar derivative. The azide
group can be reduced to the amino group by hydrogen and Raney nickel,
by hydrogenation over palladium or platinum catalyst, or by hydride
ion (lithium aluminum hydride).
2. Cleavage of an epoxide ring of a 3,4-anhydro sugar derivative by ammonia, amine or azide ion.

3. Oximation of a 4-ULOse sugar followed by reduction with hydrogen over palladium or lithium aluminum hydride.

4. Condensation of a suitable dialdehyde with one mole of nitromethane and separation of the isomers produced.

The pyranose-pyrrolidine isomerization in 4-aminohexoses has been observed and this offers difficulties in the hydrolysis step of the corresponding glycosides into their free sugars, except in the case of the 4-dimethylamino sugars, which undergo hydrolysis without complication. The formation of pyrrole derivatives is evidently associated with the presence of a free hydrogen atom on the nitrogen atom at C-4.
STATEMENT OF THE PROBLEM

At the outset of this investigation, pyranoid ring systems having nitrogen as the ring hetero atom had been described in D-xylose, D-arabinose, D-ribose and L-xylose series. None had been described having the nitrogen atom in a furanoid ring. Few satisfactory synthetic approaches to 4-amino-4-deoxy sugars had been described.

The object of the investigation was to synthesize a 4-amino-4-deoxy pentose derivative having a free hydroxyl group at C-5, and one having no free C-5 hydroxyl, and to determine the behavior of these derivatives under ring-closure conditions, to determine the relative stabilities of the aldehyde, aldehydrol, furanose (having nitrogen in the ring), and pyranose (having oxygen in the ring) in a common configurational series. The examples selected for synthesis were 4-acetamido-4-deoxy-L-xylose and its 5-deoxy analog.

The proposed synthetic route sets out, in each case, from the readily available 2-amino-2-deoxy-D-glucose, and involves reduction of the C-1 function and cleavage of the 5,6-carbon-carbon bond.
DISCUSSION OF RESULTS

PART I

Part I is concerned with the synthesis of 4-acetamido-4,5-dideoxy-L-xylofuranose.

Synthesis of 4-Acetamido-4,5-dideoxy-L-xylofuranose

The route of synthesis is outlined in Chart I.* The essential features of the synthesis involve conversion of the potential aldehyde group at C-1 in 2-acetamido-2-deoxy-α-D-glucose into a methyl group, followed by glycol cleavage at the 5,6-position in a protected derivative, and removal of the protecting groups. The route involves no reactions at the secondary alcohol groups in the final product, and hence the synthesis is stereochemically definitive.

The starting material for the synthesis, 2-amino-2-deoxy-D-glucose hydrochloride (I), which is readily available commercially, was N-acetylated by the method described by Horton (138). 2-Acetamido-


2-deoxy-α-D-glucose II was obtained in 94% yield. Mercaptailation of the latter by the procedure of Wolf from and Anno (139) gave a 72% yield

of 2-acetamido-2-deoxy-D-glucose diethyl dithioacetal (III). Treatment of III with acetone and catalytic amount of sulfuric acid under the conditions described by Yoshimura and Sato (140) gave the crystalline


3,4; 5,6-diisopropylidene derivative (IV). Partial hydrolysis of this product with 1N sulfuric acid for 40 min. at 40° gave selective removal of the terminal acetal residue, to give 2-acetamido-2-deoxy-3,4-o-isopropylidene-D-glucose diethyl dithioacetal (V) in 60% yield. This product was reported by Yoshimura and Sato (140) to have m.p. 136-137°, [α]D^28 -6.7° in methanol. In this work the product was found to have m.p. 137-138°, [α]D^21 -26.0.

Compound V was smoothly desulfurized with excess Raney nickel in hot ethanol (141) to give 2-acetamido-1,2-dideoxy-3,4-o-isopropylidene-


D-glucitol (VI), in high yield, as a chromatographically homogeneous sirup whose infrared and nuclear magnetic resonance (n.m.r.) spectra were in accord with the assigned structure.

Oxidation of the free diol VI with one equivalent of aqueous periodic acid caused rapid cleavage of the C-5 to C-6 bond to give 4-acetamido-4,5-dideoxy-2,3-o-isopropylidene-aldehydo-L-xylose (VII) as a chromatographically homogeneous sirup, characterized as the crystalline benzylphenylhydrazone.

Hydrolysis of the blocked aldehyde sugar VII in aqueous acetic acid cleaved the isopropylidene group to give 4-acetamido-4,5-dideoxy-L-xylose (VIII) as a sirup.
Discussion of the steps involved in the synthesis

Treatment of a sugar with a thiol in the presence of a strong acid usually leads to the formation of an acyclic dithioacetal.

The dithioacetal, which is rapidly formed at temperatures near 0° may be isolated in high yield if it separates from the reaction by crystallization (142), or by rapid neutralization of the acid (143, 144).

(142) E. Fischer, Ber., 27, 673 (1894).


(144) M. L. Wolfrom and F. B. Moody, ibid, 62, 3456 (1940).

At longer reaction times or at higher temperatures, a subsequent, slower reaction occurs to give a mixture of products from which fair yields of 1-thioglycosides are obtained. It would seem that, under mercaptalation conditions, dithioacetal formation is a fast, kinetically controlled reaction, followed by a slower reaction which, at equilibrium, gives a distribution of products, including free aldose, thioglycosides and dithioacetal, according to their thermodynamic stabilities in the system (145, 146).


2-Acetamido-2-deoxy-D-glucose is converted into the diethyl dithioacetal in 81% yield (139) by treatment with ethanethiol and concentrated hydrochloric acid for 24 hours at 0°, but, at room temperature, a mixture of products was isolated (147), including


the dithioacetal (24%), starting material (12%), and 2-amino-2-deoxy-D-glucose hydrochloride (29%), together with the anomeric ethyl 2-acetamido-2-deoxy-1-thio-α (and β) D-glucopyranosides (9% and 17% respectively). In the present work, a large-scale adaptation of the procedure of Wolf from and Anno was employed for the conversion of II into III.

The proposed glycol-cleavage step required protection of the C-3 and C-4 hydroxyls. The isopropylidene acetal was chosen because it can readily be formed, and can also be removed under relatively mild conditions. Moreover, the acetal group is unattacked by such reducing agents as hydrogen in presence of Raney nickel, and it is stable towards the oxidizing agents commonly employed in carbohydrate chemistry.

Yoshimura and Sato (140) have reported that treatment of 2-acetamido-2-deoxy-D-glucose diethyl dithioacetal with cupric sulfate and boron trifluoride etherate in acetone produced the 5,6-mono-O-isopropylidene derivative. With sulfuric acid as catalyst, a di-O-isopropylidene derivative was formed. One of the O-isopropylidene groups could be selectively hydrolyzed, leaving a 2-acetamido-2-deoxy-mono-O-isopropylidene-D-glucose diethyl dithioacetal. Evidence was presented that this product was the 3,4-isopropylidene acetal. In this work the procedure
of Yoshimura and Sato was followed.

Desulfurization of 2-acetamido-2-deoxy-3,4-0-isopropylidene-D-glucose diethyl dithiocetal

The simultaneous desulfurizing and reducing action of Raney nickel, was discovered by Bougault, Cattelian and Chabrier (148) and extended


by Mozingo and co-workers (149). Thiocarbonyl compounds, thiol,


disulfides, heterocyclic sulfur, sulfides, sulfoxides, and sulfones can be reduced by Raney nickel. It was shown by Wolfrom and Karabinos to afford a simple and direct route to the 1-deoxy alditols from dithioacetals.

Reductive desulfurization of the dithioacetal (V) was accomplished by refluxing the dithioacetal with neutral Raney nickel in absolute ethanol for 6 hours, to give 2-acetamido-1,2-dideoxy-3,4-0-isopropylidene-D-glucitol VI, in 90% yield, as a chromatographically homogeneous sirup whose infrared and nuclear magnetic resonance (n.m.r.) spectra were in accord with the assigned structure. This compound was characterized as the crystalline 5,6-diacetate, which was isolated by distillation.

The n.m.r. spectrum of the latter showed the signal of the C-1 methyl group at $\tau$ 8.77 as a doublet $J_{1,2} 7.0$ c.p.s. the signal of the isopropylidene methyl group as a 6-proton singlet at $\tau$ 8.59, and the
acetyl group signals as 3-proton singlets at \( \tau 8.00 \), \( 7.97 \), and \( 7.89 \). The absence of multiplets characteristic of ethyl groups in the n.m.r. spectra of VI or its diacetate indicated that the desulfurization reaction had gone to completion.

Attempts were made to obtain 2-acetamido-1,2-dideoxy-3,4-\(\beta\)-isopropylidene-D-glucitol by desulfurization of compound IV, followed by partial hydrolysis of 2-acetamido-1,2-dideoxy-3,4:5,6-di-\(\beta\)-isopropylidene-D-glucitol. The desulfurization step was successfully accomplished by refluxing the dithioacetal IV with neutral Raney nickel in absolute ethanol for 6 hours. The crystalline 2-acetamido-1,2-dideoxy-3,4:5,6-di-\(\beta\)-isopropylidene-D-glucitol (IX) was obtained in 90% yield. Treatment of IX with 50% aqueous acetone solution of IN sulfuric acid at 40° for 40 minutes—conditions used successfully in the partial hydrolysis of IV to V—gave a sirup that on thin layer chromatography showed two components, a faster one corresponding to the mono-\(\beta\)-isopropylidene derivative, VI, and a slower one which is presumably the completely hydrolysed product.
Synthesis of 4-Acetamido-4,5-dideoxy-2,3-O-isopropylidene-
aldehyde-L-xylose

Oxidation of the diol VI with one equivalent of aqueous periodic acid caused rapid cleavage of the C-5 to C-6 bond to give 4-acetamido-4,5-dideoxy-2,3-O-isopropylidene-aldehyde-L-xylose VII as a chromatographically homogeneous sirup, characterised as the crystalline benzylphenylhydrazone. The infrared spectrum of VII showed carbonyl absorption at 5.80 μ. The sirup gave a positive Schiff test, as did a concentrated aqueous solution of VII. The n.m.r. spectrum of VII in chloroform-d showed a one-proton singlet at τ 0.67. This is in the region characteristic of formyl protons (150). These data clearly show


that VII is not intermolecularly combined or intramolecularly cyclized. Intramolecular cyclization of VII would lead to a trans-fused bicyclo \[3.3.0\] structure, a strained (151) system that has not yet been observed in carbohydrate structures.

(151) Ref. 61, p. 273

Synthesis of 4-acetamido-4,5-dideoxy-L-xylose

4-Acetamido-4,5-dideoxy-L-xylose, obtained by hydrolysis of VII with aqueous acetic acid was a sirup that was homogeneous by paper
pyridine-ethyl acetate-water-acetic acid) and it was well resolved from the starting material VII.

**Tautomeric structure of 4-acetamido-4,5-dideoxy-L-xylose**

The acetamido sugar reduced Fehling solution, but it did not restore the color to Schiff reagent. Its infrared spectrum showed negligible absorption for the aldehyde group near 5.8 μ; it showed amide carbonyl absorption at 6.12 μ but negligible amide NH absorption near 6.50 μ. These data fully support formulation of the product as the 5-membered cyclic structure, 4-acetamido-4,5-dideoxy-α, β-L-xylo-furanose (VIII). Paper chromatography of VIII in a 40:11:19 butanol-ethanol-water system revealed the product as an incompletely-separated double zone, Rₐ 0.49 and 0.54. Excision of each zone, extraction with the chromatography solvent and rechromatography gave two separate components. When an aqueous solution of either component was heated for 1 hour at 90°, chromatography revealed the re-formation of the original double zone. This indicates a slow interconversion between two two tautomeric forms of VIII, and since little of the aldehyde form can be present, the two tautomers must be the α-L and β-L anomers of the furanose form. The zones Rₐ = 0.49 and 0.54 had approximate relative intensities 2:3, and since the zone Rₐ 0.54 was more dextrorotatory than the equilibrated mixture it was considered to be the β-L anomer. The formation of a single zone when VIII was subjected to chromatography...
in the Fischer-Nebel solvent system may be ascribed to rapid interconversion of anomers in this more-polar solvent system.

The n.m.r. spectrum of VIII in deuterium oxide solution showed a three-proton multiplet at \( \tau \) 8.84 assigned to the C-5 methyl group, a three-proton multiplet at \( \tau \) 7.85 assigned to the N-acetyl methyl group, a three-proton multiplet at \( \tau \) 5.87 assigned to the ring protons at C-2, C-3, and C-4, and a pair of multiplets, \( \tau \) 4.82 and 4.68, corresponding to one proton, assigned to the anomeric proton. The fine structure of the spectrum was interpreted to indicate that VIII was a mixture of the \( \alpha \)-L and \( \beta \)-L anomers, and that rotation about the nitrogen-acetyl bond was restricted. A small proportion of the acyclic form appeared to be present.

The multiplets at \( \tau \) 4.82 and \( \tau \) 4.68 were assigned to H-1 of the \( \beta \)-L and \( \alpha \)-L anomeric forms respectively. These signals show multiplicity greater
than simple doublets, attributable (153, 154) to virtual coupling due

to the small difference in chemical shift between the H-2 and H-3


signals, and to rotational isomerism about the nitrogen-acetyl bond.
The multiplet signal of the 5-methyl group at $\tau \ 8.84$ is resolved when
the temperature is raised to 86°, into two doublets, $\tau \ 8.76$ and 8.90,
$J_{4,5} = 7.0$ c.p.s., in the approximate intensity ratio 3:2. These
doublets were assigned on the basis of their intensities to the $\beta$-L
and $\alpha$-L anomers, respectively. Signals of acetyl methyl groups
appeared at $\tau \ 7.82$, 7.85, 7.88 and 8.01, the last constituting about
5% of the total. When the sample was heated the signals at $\tau \ 7.85$
and 7.88 coalesced and the intensity of the signal at $\tau \ 7.82$ decreased;
the reverse process occurred on cooling. This behavior is analogous
to that of several $N$-methylamides (98, 99). This is due to resonance
conjugation between the $p$-orbital on nitrogen and the $p$-orbital of the
$\pi$-electron system so that two formal dipolar structures VIIIa and
VIIIb can be written (97). The n.m.r. spectrum observed at room
temperature is the result of a superposition of the peaks arising from
the two possible anomeric forms, each in two slowly interconverting
conformers resulting from restricted rotation about the bond between
the C atom of the CO group and the nitrogen atom. The low-intensity
singlet at $\tau \ 8.01$, is unaffected by change of temperature, and it is
probably due to small proportion of the aldehydrol form of VIII in
equilibrium with the furanose forms. This work has been published (155).


It can be concluded that the acetamido sugar adopts the cyclic structure, with nitrogen as the hetero atom, rather than the acyclic structure (X) or its hydrated form.

\[
\begin{align*}
&\text{CHO} \\
&\text{HOCH} \\
&\text{HCOH} \\
&\text{AcHN\textsuperscript{\textbullet}}\text{CH} \\
&\text{CH}_{3} \\
&\text{X}
\end{align*}
\]

Comparison of 4-acetamido-4,5-dideoxy-L-xylofuranose and its D-enantiomorph

In a brief communication, Hanessian (156) has reported a synthesis

(156) S. Hanessian, Carbohydrate Res., 1, 178 (1965).

of 4-acetamido-4,5-dideoxy-D-xylofuranose, the enantiomorph of VIII. Starting from L-arabinose, by a series of transformations, 5-deoxy-2,3-O-isopropylidene-4-O-(\text{\textbeta}-nitrobenzenesulfonyl)-L-arabinose diethyl dithioacetal was synthesized. Treatment of the latter with sodium azide in \textit{N},\textit{N}-dimethylformamide gave 4-azido-4,5-dideoxy-2,3-O-isopropylidene-
D-xylose diethyl dithioacetal. Reduction of this product, followed, by \( \text{\(N\)} \)-acetylation, gave the 4-acetamido derivative. Removal of the acetal group afforded 4-acetamido-4,5-dideoxy-D-xylose diethyl dithioacetal, which on demercaptalation gave a sirup, \([\alpha]_{D}^{24} + 11.2^\circ\) (at equil. in methanol). Paper chromatography of the sirupy product showed a double spot having \( R^f \) 0.65 and 0.72 in an approximate ratio 1:1 by using butanol-ethanol-water, 3:1:1 as a solvent system. The author stated that the two components, the \( \alpha \) and \( \beta \) anomers, were separated, but, surprisingly, stated that the optical rotational data of the separated components were not unambiguously discriminating for the two anomers. The slower-moving component showed signals in the n.m.r. of \( \tau \) 4.72 (1 proton multiplet, H-1); \( \tau \) 7.82, 7.85, 7.88, 7.92 (acetyl hydrogens); \( \tau \) 8.88, 8.97 (H-5 main peaks). A peak at \( \tau \) 8.03 (acetyl hydrogens) was attributed to the acyclic form. The acyclic form was considered to be present (8\%), in the original mixture. The faster moving component showed peaks almost at the same \( \tau \) values of the slower moving component except that H-1 gave a multiplet at \( \tau \) 4.84.

The n.m.r. spectrum of 4-acetamido-4,5-dideoxy-L-xylofuranose and the data reported by Hanessian for the D-enantiomorph are closely similar. The specific rotations at equilibrium of the reported D enantiomorph (\([\alpha]_{D}^{24} + 11.2^\circ\)) is of approximately equal magnitude but of opposite sign, to that of the L-enantiomorph (\([\alpha]_{D}^{21} - 10^\circ\)). This fact, and the fact that compound (VIII) was synthesized, in this work, by a stereochemically definitive route, confirms the structure of the D-enantiomorph synthesized by Hanessian.
The tentative anomeric assignments made by Hanessian, based on the appearance of the H-1 signals of the separated anomers, are in agreement with those established by rotatory data in the present work.

4-Acetamido-4,5-dideoxy-D-xylofuranose has been synthesized from 6-deoxy-L-mannose (L-rhamnose) (157). This was converted into


1,2:3,4-di-O-isopropylidene-5-O-p-tolylsulforyl-L-rhamnitol. On treatment the latter with sodium azide in N,N-dimethylformamide it yielded 2-azido-1,2-dideoxy-3,4:5,6-di-O-isopropylidene-L-glucitol. Reduction of the azide with lithium aluminum hydride, followed by N-acetylation, afforded 2-acetamido-1,2-dideoxy-3,4:5,6-di-O-isopropylidene-L-glucitol. The 5,6-di-O-isopropylidene group was selectively hydrolyzed and the resulting diol was cleaved with one equivalent of periodic acid to give 4-acetamido-4,5-dideoxy-2,3-O-isopropylidene-aldehydo-D-xylose. Hot aqueous acetic acid converted this into 4-acetamido-4,5-dideoxy-D-xylose, isolated as a sirup, [α]_D^{28} + 12 ± 2° (water).

4-Amino sugars with a nitrogen in a five-membered ring

During the course of this work, several papers have been published regarding the synthesis of furanose sugars having nitrogen as the ring heterocycle.

Methyl 4-acetamido-4-deoxy-L-erythrofuranoside (158), the first

five-membered ring sugar having nitrogen as the ring hetero atom, was synthesized by selective oxidation of 1-acetamido-1-deoxy-D-ribitol (XI) by using limited proportion of sodium metaperiodate. The product (XII) was isolated from a mixture of components as a sirup, whose structure was confirmed by i.r. and periodate oxidation. The n.m.r. spectrum of XII showed two peaks at $\tau$ 7.92 and 7.88 (NAC), two peaks at $\tau$ 6.64 and 6.59 (OCH$_3$), two doublets at $\tau$ 5.05 and 4.96 (H-1 proton).

It was suggested that either the compound possesses internal hindered rotation or the substance may be an approximately 1:1 mixture of $\alpha$ and $\beta$ anomers. An evidence has been presented that substantiates the former hypothesis (96), thus, at an elevated temperature (80°) the two anomeric resonances were broadened and the O-methyl and N-acetyl absorptions collapsed to single peaks.

4-Acetamido-4-deoxy-D-threofuranose (159) was obtained as a sirup

by lead tetraacetate oxidation of 6-acetamido-6-deoxy-D-galactose XIII by using two moles of oxidant. The n.m.r. spectrum of compound XIV showed a great similarity to that of 4-acetamido-4,5-dideoxy-L-xylofuranose, prepared in the present work. There were 5 peaks in the region $\tau 7.85 - 8.04$, assigned to the $\text{N}$-acetyl group, and the signals of the anomeric protons appear as a multiplet at $\tau 4.76 - 4.57$. This was attributed to the presence of $\alpha$ and $\beta$ anomers with restricted rotation about the CO-N bond. The synthesis of 4-acetamido-4-deoxy-L-erythrofuranose was reported in this paper.

Acetolysis of methyl 4-acetamido-4-deoxy-\text{-}$\alpha$-D-ribopyranoside XV gave the sirupy 4-acetamido-1,2,3,5-tetra-O-acetyl-4-deoxy-D-ribofuranose XVI (160). Compound XVI showed by n.m.r. the presence of five acetyl groups having chemical shifts between $\tau 7.88$ and $8.02$. There was

no NH absorption at 6.5 μ in the infrared, and the pentaacetate was therefore assigned the furanose structure. The contraction of the 4-acetamido-4-deoxy ribopyranosides into the furanose ring under acid conditions is noteworthy. It is also interesting to note that 5-acetamido-5-deoxy-D-ribose showed a greater tendency toward furanose formation than did the analogous xylose and arabinose derivatives (95).
DISCUSSION OF RESULTS

PART II

Synthesis of 4-acetamido-4-deoxy-L-xylopyranose

4-Acetamido-4-deoxy-L-xylose was synthesized by a stereochemically definitive route involving C-5 to C-6 cleavage of a 2-amino-2-deoxy-D-glucitol derivative. The sequence of conversions is outlined in Chart II.

Reduction of 2-acetamido-2-deoxy-D-glucose with hydrogen over Raney nickel (161) gave 2-acetamido-2-deoxy-D-glucitol in 91% yield.


Hydrolysis of the acetyl group was effected by heating the alditol in 6 N hydrochloric acid for 1 hour at 100° to give 2-amino-2-deoxy-D-glucitol hydrochloride in 85% yield. Treatment of the latter with aqueous sodium hydrogen carbonate and 1-fluoro-2,4-dinitrobenzene (162)


afforded 2-deoxy-2-(2,4-dinitroanilino)-D-glucitol XVII in 69% yield.

Acetonation of 2-deoxy-2-(2,4-dinitroanilino)-D-glucitol XVII gave the 3,4:5,6-diisopropylidene acetal XVIII, which was converted by mild acid hydrolysis into the 3,4-monoisopropylidene acetal XIX, followed by mild hydrolysis of the oxidation product XX gave 4-deoxy-4-(2,4-dinitroanilino)-L-xylopyranose XXI which on condensation with acetone gave the 1,2-0-isopropylidene derivative XXII. Treatment of
XXII with Dowex-1 (OH\textsuperscript{-}) ion exchange resin smoothly removed the
N-substituent and gave crystalline 4-amino-4-deoxy-1,2-\textbeta-isopropylidene-
\alpha-L-xylopyranose XXIII.

Compound XXIII was acetylated to give 4-acetamido-3-\textbeta-acetyl-
4-deoxy-1,2-\textbeta-isopropylidene-\alpha-L-xylopyranose XXIV. \textbeta-Deacetylation
of XXIV, followed by mild hydrolysis of the product XXV gave 4-acet-
amido-4-deoxy-L-xylose XXVI as a chromatographically homogeneous,
crystalline product.

The acetamido sugar XXVI could also be prepared directly, in one
step, from 4-amino-4-deoxy-1,2-\textbeta-isopropylidene-\alpha-L-xylopyranose XXIII,
by treatment of XXIII in aqueous solution with acetic anhydride;
selective N-acetylation took place and the acetic acid formed by hydrolysis
of the excess acetic anhydride caused cleavage of the \textbeta-isopropylidene
group.

The yields of all steps in the synthesis were at least 50\% and most
were considerably higher. The overall yield in the conversion of
2-acetamido-2-deoxy-D-glucose into 4-acetamido-4-deoxy-L-xylose XXVI
was about 10\% when purification at some of the intermediate stages was
omitted.

Discussion of the steps involved in the synthesis
2-Deoxy-2-(2,4-dinitroanilino)-D-glucitol XVII

This compound was prepared in two steps from the readily available
2-acetamido-2-deoxy-D-glucose. The acetamido sugar has been reduced to
the corresponding alditol by hydrogenation over Raney nickel (161), or
by treatment with aluminum amalgam (163), or by sodium borohydride (162).


In this work, a large-scale adaptation of the catalytic hydrogenation method was employed, and the product was then hydrolyzed to 2-amino-2-deoxy-D-glucitol hydrochloride. The procedure gave the latter in better yield than by the one-step reduction of 2-amino-2-deoxy-D-glucose hydrochloride (161). The alditol hydrochloride was converted into compound XVII by treatment with sodium hydrogen carbonate and 1-fluoro-2,4-dinitrobenzene, by a modification of the procedure of Leskowitz and Kabat (162).

2-Deoxy-2-(2,4-dinitroanilino)-3,4:3',6'-di-O-isopropylidene-D-glucitol (XVIII)

Yoshimura and co-workers (164) prepared 2-acetamido-2-deoxy-3,4:


5,6-di-O-isopropylidene-D-glucitol by stirring 2-acetamido-2-deoxy-D-glucitol with dry acetone, in the presence of sulfuric acid and copper sulfate for 70 hrs. at 30 - 40°. The sirup obtained was separated by high vacuum distillation into two fractions. The first fraction was found to be 2-amino-2-deoxy-3,4:5,6-di-O-isopropylidene-D-glucitol and the second fraction contained two components, the major one was 2-acetamido-2-deoxy-3,4:5,6-di-O-isopropylidene-D-glucitol and another minor product of unknown structure. Attempts to acetonate 2-amino-2-deoxy-D-glucitol
hydrochloride, for 100 hrs. at 30 - 45° gave a very low yield (less than 10%) of the corresponding di-0-isopropylidene derivative. It appears probable that acetonation may be inhibited by electrostatic factors caused by the protonated amino group, and therefore synthesis of the title compound by dinitrophenylation of the di-0-isopropylidene derivative of the amino alditol was not a practicable route for large-scale synthesis.

In this work, shaking of 2-deoxy-2-(2,4-dinitroanilino)-D-glucitol with dry acetone, in presence of sulfuric acid and anhydrous cupric sulfate for 21 hours at room temperature gave the crystalline 2-deoxy-2-(2,4-dinitroanilino)-3,4:5,6-di-0-isopropylidene-D-glucitol XVIII in 63% yield. The position of substitution of the 0-isopropylidene groups were assigned by analogy with the known 2-acetamido analog (164) and verified by subsequent conversions. The compound XVIII was further characterized as it crystalline 1-0-(p-tolylsulfonyl) derivative. The n.m.r. spectrum of XVIII in chloroform-d showed a 1-proton triplet at τ 7.39, J 6 c.p.s., which disappears on deuteration; this was assigned to the OH proton of a CH₂OH group. One-proton, broadened doublet at τ 0.90, not exchanged on simple deuteration, was assigned to the NH proton. The acetal substituents were, therefore, located at 0-3, 4, 5, and 6.

Alternative approach for preparation of 2-deoxy-2-(2,4-dinitroanilino)-3,4:5,6-di-0-isopropylidene-D-glucitol (XVIII)

Mercaptalation of 2-amino-2-deoxy-D-glucose hydrochloride with ethanethiol under forcing conditions (saturation with hydrogen chloride gas at 0°) for 20 hr. gave 2-amino-2-deoxy-D-glucose diethyl dithioacetals.
hydrochloride XXVII in 85% yield. This compound has been reported by Whitehouse, Kent, and Pasternak (165) who gave a yield of 70%, and by Hough and Taha (166) who obtained a 70% yield of sirup which crystallizes slowly. Dinitrophenylation of the dithioacetal by the procedure of Wolfson, Garg and Horton (167) gave the sirupy 2-deoxy-2-(2,4-dinitro-


anilino)-D-glucose diethyl dithionacetal XXVIII.

Acetonation of XXVIII afforded crystalline 2-deoxy-2-(2,4-dinitro-
anilino)-3,4;5,6-di-O-isopropylidene-D-glucose diethyl dithioacetal XXIX in a 52% yield. Treatment of the latter with bromine, by adaptation of the general procedure of Defaye (168) gave 2-deoxy-2-(2,4-


dinitroanilino)-3,4;5,6-di-O-isopropylidene-aldehyde-D-glucose (XXX) as a homogeneous sirup whose i.r. spectrum showed absorption band for the aldehyde group at 5.8 μ. Attempts to convert the aldehydo sugar into the glucitol derivative XVIII (Chart II) by reduction with sodium borohydride led to the cleavage of the 2,4-dinitrophenyl group, and the attempt to prepare 2-deoxy-2-(2,4-dinitroanilino)-3,4;5,6-di-O-
isopropylidene-D-glucitol by this route was therefore abandoned.
CHART III

XXXVII

XXXVIII

XXXIX

XXX

NaBH₄ Liberation of 2,4-dinitrophenol

DNP = 2,4-dinitrophenyl
2-(2,4-dinitroanilino)-3,4-O-isopropylidene-D-glucitol (XIX)

Optimum conditions for partial acid hydrolysis of the diisopropylidene acetal XVIII were established by t.l.c. of the reaction solution at various intervals of time. Under these conditions there was obtained, beside the desired compound (slow moving spot), a trace of the di-O-isopropylidene compound XVIII (faster moving spot) and a trace of the completely hydrolyzed compound XVII (slower moving spot).

2-Deoxy-2-(2,4-dinitroanilino)-3,4-O-isopropylidene-D-glucitol could be crystallized either from ether or from ethyl acetate-hexane. Substance XIX was encountered as the orange, anhydrous form, m.p. 128-129°, and also as a stable, yellow form, m.p. 95-96°, that was solvated with chloroform. The latter was converted into the former by heating to 110°, but it could be recrystallized unchanged from chloroform. Acetylation of either form gave the 1,5,6-triacetate of XIX whose n.m.r. spectrum shown in Fig. 3. Periodate oxidation of XIX showed consumption of one mole of oxidant within 5 min. with the formation of a pentose derivative. This experiment showed that the acetal group removed was at the 5,6-position, and also provided complete verification of the structure of substance XVIII.

Removal of the N-substituent from XIX by treatment with Dowex-1 (OH⁻) ion-exchange resin, with subsequent acetylation, gave crystalline 2-acetamido-1,5,6-tri-O-acetyl-2-deoxy-3,4-O-isopropylidene-D-glucitol. The yield was low, probably because of adsorption of the product to resin due to the presence of three free hydroxyl groups. It has been shown that, the separation of mixtures of nucleosides on Dowex-1 (OH⁻), the adenine derivatives are eluted in the order: 2'-3'-isopropylidene-adenosine, 2'-deoxyadenosine, 3'-deoxyadenosine, adenosine; the ease of
elution is related to the number of sugar hydroxyl groups and the extent of their dissociation (169). It was therefore decided to postpone the removal of the dinitrophenyl blocking group to a later stage in the synthesis.

\[ \begin{align*}
\text{4-deoxy-4-(2,4-dinitroanilino)-L-xylopyranose and acetonation to} \\
\text{4-deoxy-(2,4-dinitroanilino-1,2-O-isopropylidene-\(\alpha\)-L-xylopyranose} \\
\end{align*} \]

Preparative, periodate oxidation of XIX (either form) gave 4-deoxy-4-(2,4-dinitroanilino)-2,3-O-isopropylidene-aldehydo-L-xylose XX as a glass, which showed absorption for the aldehyde group in the infrared spectrum. Cyclization of this derivative is presumably prevented by the fact that a trans-fused 2,3-O-isopropylidene group would result. Hydrolysis of the O-isopropylidene group with aqueous acetic acid gave crystalline 4-deoxy-4-(2,4-dinitroanilino)-L-xylopyranose XXI. The infrared spectrum of XXI showed no carbonyl absorption. The ring size of the product was indicated by the fact that acetylation with acetic anhydride-pyridine gave a sirupy triacetate that showed absorptions for the NH group in the infrared and n.m.r. spectra. A furanose form would have given a tetraacetate having no NH group, and an acyclic form of the free sugar would have exhibited carbonyl absorption.

Acetonation of XXI in the presence of sulfuric acid and copper (II) sulfate gave a crystalline mono-O-isopropylidene in high yield, which was shown to be 4-deoxy-4-(2,4-dinitroanilino)-1,2-O-isopropylidene-\(\alpha\)-L-xylopyranose XXII; acetylation gave the crystalline 3-acetate of XXII. The structure assigned to XXII was based on the fact that the substance
was nonreducing, it was different from XX, and removal of the N-substituent from XXII gave a nonreducing 4-amino-4-deoxypentose derivative XXIII that consumed one mole of periodate. Further, concordant data were provided by the n.m.r. spectra of some of the transformation products. No further acetonation of XXII was observed when treatment with acetone was prolonged, even though a 1,2:3,5-di-O-isopropylidene derivative (XXXI) of the furanose form might be considered possible.

The hydroxyl groups of C-2, C-3 and C-4 of the favored conformation of α-D(or L)-xylopyranose are situated equatorially and are trans-disposed to each other. Cyclic acetals only of the corresponding furanose are known (170). The interpretation given was, that in the


case of the cis-diol, a decrease of the dihedral angle between the hydroxyl groups (initially at 60°) to an angle of about 50° (required for an unstrained dioxolane ring), is comparatively unhindered, whereas, for the trans-diol, the movement is such as to cause considerable
repulsive non-bonded interaction in the molecule. The fact that a 1,2-\(\text{Q}\)-isopropylidene derivative of the pyranose form was obtained in the present work may be ascribed to the reluctance of the nitrogen atom to enter into ring formation, because of its negligible nucleophilic character when substituted with the dinitrophenyl group.

**4-Acetamido-4-deoxy-L-xylopyranose**

Removal of the \(N-(2,4\text{-dinitrophenyl})\) group from (XXII) gave crystalline 4-amino-4-deoxy-1,2-\(\text{Q}\)-isopropylidene-\(\alpha\)-L-xylo-
pyranose (XXIII) in good yield; the product consumed one mole of periodate rapidly (5 min.). The oxidant consumption remained constant for several hours, and slow overoxidation was subsequently observed.

Acetylation of compound XXIII followed by O-deacetylation gave the corresponding sirupy 3-hydroxy derivative XXV. Removal of the \(\text{Q}\)-isopropylidene group from XXV by mild acid hydrolysis gave crystalline 4-acetamido-4-deoxy-L-xylose (XXVI), m.p. 155-157°, \([\alpha]_D^{24} = 53 \rightarrow -49^\circ\) (water). The acetamido sugar (XXVI) was also prepared directly, in one step by treatment of XXIII in aqueous solution with acetic anhydride. This one-step procedure involves initial N-acetylation followed by hydrolysis of the \(\text{Q}\)-isopropylidene group by the acetic acid formed by hydrolysis of the acetic anhydride.

**Tautomeric structure of 4-acetamido-4-deoxy-L-xylose**

The infrared spectrum of the crystalline acetamido sugar XXVI showed absorptions typical of the amide carbonyl and amide NH groups. A solution of the sugar in deuterium oxide, at room temperature and at mutarotational equilibrium, showed a narrow doublet in the n.m.r. spectrum,
at $\tau$ 4.78, $J_{1,2}$ 3 c.p.s., assigned to the equatorial H-1 of the $\alpha$-L pyranose anomer of XXVI. The signal of the axial H-1 of the $\beta$-L-anomer was not observable at room temperature because of interference by the HOD signal. The latter signal was shifted upfield (171) in the spectrum (171) R. U. Lemieux and J. D. Stevens, Can. J. Chem., 44, 249 (1966).

measured at 80° (Fig. 1), and the axial H-1 signal of the $\beta$-L pyranose anomer was clearly observable at $\tau$ 5.44 as a wide doublet, $J_{1,2}$ 7.6 c.p.s. and the total integral of both H-1 signals corresponded to one proton. The magnitudes of the observed $J_{1,2}$ couplings leave no doubt that the anomic pyranoses are involved, and the fact that the chemical shifts of the H-1 signals of the anomers correspond closely to those reported (171, 172) for the anomic D-xylopyranoses provides further confirmation. (172) L. D. Hall, Tetrahedron letters, 23, 1457 (1964).

Integration of the spectrum indicated that the $\alpha$-L and $\beta$-L anomers of XXVI are present in 2:3 proportion at equilibrium. The signal of the NAc protons appeared as 3-proton singlet at $\tau$ 7.99.

The small amount of mutarotation observed with XXVI, in relation to the anomic composition at equilibrium in aqueous solution, suggests that the crystalline product is a co-crystallized mixture of anomers.

A solution of XXVI in methyl sulfoxide-$d_6$, a solvent in which mutarotation is normally slow (173, 174), showed signals for H-1 of (173) B. Casu, M. Reggiani, G. G. Gallo, and A. Vigevani, Tetrahedron letters, 2839 (1964); 2253 (1965).

Fig. 1

The 60-Mc.p.s. n.m.r. spectrum of 4-acetamido-4-deoxy-L-xylose (XXVI) at equilibrium in deuterium oxide, at 80°.
both pyranose anomers, supporting the proposal that crystalline sugar was a co-crystallized mixture of anomers.

Conformation of 4-acetamido-3-0-acetyl-4-deoxy-1,2-0-isopropylidene-$\alpha$-L-xylopyranose (XXIV)

The n.m.r. spectrum of 4-acetamido-3-0-acetyl-4-deoxy-1,2-0-isopropylidene-$\alpha$-L-xylopyranose (XXIV), measured in chloroform-$d$ showed (Fig. 2) a broad, one proton signal at low yield ($\tau$ 3.60) which was assigned to the NH proton, since it disappeared on deuteration. This observation provided independent evidence that compounds XXII, XXIII, XXIV and XXV did not have the nitrogen in the ring. A two-proton multiplet at $\tau$ 4.91 in the spectrum of XXIV (in chloroform-$d$) was assigned to H-1 and H-3, the two protons most strongly deshielded of those attached to carbon. The overlap of these two signals were well separated when the spectrum was measured in benzene (Fig. 2). A sharp doublet at $\tau$ 4.96 was assigned to H-1 ($J_{1,2}$ 2.5 c.p.s.), and the narrow signal at $\tau$ 4.72 (total width 10 c.p.s.) was assigned to H-3. The fact that the H-3 signal was not observed as a wide (18-20
The 60-Mc.p.s. n.m.r. spectrum of \( \text{N-acetamido-3-O-acetyl-4-deoxy-1,2-O-isopropylidene-\( \alpha \)-L-xylopyranose} \) (XXIV).

Fig. 2
c.p.s.), symmetrical triplet indicated that H-2, H-3, and H-4 were not trans-diaxial [in the 1C conformation (A)], and the observed data accord with formulation of XXIV in a skew conformation (B). It has been proposed (58, 59) that 1,2-O-alkylidene acetals of arabinopyranose, glucopyranose, and related derivatives, adopt a skew form as the favored conformation.

N.m.r. spectra of the 2,4-dinitroanilino derivatives

The 2,4-dinitroanilino derivatives studied in this work all showed signals at low field for the three aryl protons and the NH proton. None of the signals disappeared when the samples were deuterated in the usual way, even during several days, indicating that the NH proton could not be exchanged by deuterium oxide alone. However, the signal of the NH proton could be assigned definitively by adding a few drops of tributylamine to the prepared sample in chloroform-d containing deuterium oxide; this caused immediate exchange of the NH proton. A typical example is provided by the spectrum of 1,5,6-tri-O-acetyl-2-deoxy-2-(2,4-dinitroanilino)-3,4-O-isopropylidene-D-glucitol, shown (Fig. 3) before and after addition of tributylamine in deuterium oxide. The broadened doublet at τ 1.01, which disappears on treatment with base, may clearly be assigned to the NH proton. The narrow doublet at lowest field (τ 0.85), the doublet of doublets (τ 1.60), and the wide doublet (τ 2.68) can thus be assigned unambiguously to H-3, H-5 and H-6, respectively of the 2,4-dinitrophenyl group. N.m.r. data on N-methyl 2,4-dinitroaniline (175) and N-(2,4-dinitrophenyl) serine

The 60-Mc.p.s. n.m.r. spectrum of 1,5,6-tri-O-acetyl-2-deoxy-2-(2,4-dinitroanilino)-D-glucitol in chloroform-\textit{d}, and in chloroform-\textit{d} containing tributylamine in deuterium oxide.
derivatives (176) are in agreement with the present work.


During the course of the work, a paper appeared by Dick and Jones
(177) in which synthesis of 4-acetamido-4-deoxy-L-xylopyranose by a
different route was described. The route was not stereochemically
unambiguous, but it was deduced on mechanistic grounds and from data
of periodate oxidation that the product had the assigned structure XXVI.
Treatment of methyl 2,3,4-tri-O-(methylsulfonyl)-α-D-xylopyranoside
with sodium azide in N, N-dimethylformamide gave an azido-deoxy-di-O-
(methylsulfonyl) derivative. The azide ion was considered to attack
selectively at C-4 in an S$_2$ displacement reaction to yield 4-azido-4-
deoxy-2,3-di-O-(methylsulfonyl)-β-L-arabinopyranoside (XXXII), by
analogy with work of Goodman and co-workers (178), in which the selective
replacement of the 4-O-p-tolylsulfonyloxy group of methyl 2-O-benzyl-
3,4-di-O-p-tolylsulfonyl-β-L-arabinopyranoside by azide ion was observed.
Treatment of XXXII with methanolic potassium hydroxide gave the
2,3-anhydro derivative (XXXIII). Opening of this epoxide with aqueous
potassium hydroxide gave a mixture containing preponderantly methyl
4-azido-4-deoxy-β-L-xylopyranoside (XXXIV, together with a smaller pro-
portion of methyl 4-azido-4-deoxy-β-L-arabinopyranoside (XXXV). The mixture was separated by fractional crystallization. Reduction of XXXIV followed by N-acetylation gave methyl 4-acetamido-4-deoxy-β-L-xylopyranoside. Mild acid hydrolysis of the latter gave 4-acetamido-4-deoxy-L-xylopyranose which melted at 157-158° and had $[\alpha]_D = -22$ mutarotating to $-16°$ (in water). It consumed two moles of periodate with liberation of 1 mole of formic acid. Infrared spectral data indicated that the favored ring form of the acetamido sugar is pyranoid, with oxygen as the hetero atom in the ring.

Although the m.p. of 4-acetamido-4-deoxy-L-xylopyranose reported by Dick and Jones was in close agreement with that found in the present work, there is a wide disparity in the observed rotation ($-53 → -49°$) and that reported by Dick and Jones ($-22 → -16°$). A sample of the material provided by Professor Jones proved to have an X-ray and m.p. identical with that obtained in the present stereochemical definitive
synthesis. In a personal communication, Jones reported that the infrared spectrum of the two samples of 4-acetamido-4-deoxy-L-xylopyranose is identical. He also reported a revised value for the specific rotation of his compound, \([\alpha]_D^\circ = -43^\circ\) (equilibrium), in reasonable agreement to the value (-49°) found in the present work. The present work has been briefly reported (179) and the details are in press (180).


A degradative route to derivatives of 4-amino-4-deoxy-L-xylose, by way of uronic acid intermediates, was reported in June 1966 (181).


2-Benzamido-2-deoxy-D-glucuronic acid was treated with sodium borohydride, and the resultant 5-benzamido-5-deoxy-L-gulonolactone was converted into 5-benzamido-5-deoxy-L-gulonamide by methanolic ammonia. Peracetylation followed by dehydration of the amide with benzenesulfonyl chloride in pyridine gave the corresponding L-gulonitrile XXXVI. Treatment of XXXVI with sodium methoxide (Zemplén- Pacsu) gave 4-benzamido-4-deoxy-L-xylopyranose XXXVII, m.p. 204-205° (dec.), \([\alpha]_D^{20} = 0.

The i.r. spectrum showed the amide I band at 6.08 μ and amide II band at 6.53 μ indicating that the sugar exists in the pyranoid form. Since none of the derivatives corresponded to any of those in the present
synthesis, direct comparison of the products could not be made.

It is noteworthy that, in the case of 4,5-acetamido-4,5-dideoxy-L-xylose (XXXVIII), which has been reported as a sirup (182) and also in crystalline form (183), the more favorable, pyranose ring-form is adopted in aqueous solution; nitrogen is the hetero atom in the ring.

(182) S. Hanessian, Carbohydrate Res. 1, 178 (1965).

It can therefore be concluded that the sugars having the xylo configuration favor the pyranose structure, where all of the bulky substituents are in the more stable equatorial position, whether the hetero atom in the ring is oxygen or nitrogen.
EXPERIMENTAL

PART I

Melting points were determined with a Hershberg-type apparatus. Optical rotations were measured with a 2-dm tube. Infrared spectra were measured with a Perkin-Elmer "Infracord" spectrometer. N.m.r. spectra were measured with a Varian A-60 n.m.r. spectrometer (Varian Associates, Palo Alto; California) operating at 60 Mc.p.s., equipped with a Varian V-6040 variable-temperature probe. Tetramethylsilane ($\tau = 10.00$) and sodium 4,4-dimethyl-4-silapentane-1-sulfonate ($\tau = 10.000$) were used as internal standards for spectra determined in deuteriochloroform and deuterium oxide, respectively. The recorded first-order coupling constants are the measured peak spacings and are considered accurate ± 0.5 c.p.s. Unless otherwise stated, the spectra were measured at about 40°. Deuteration was performed by adding one drop of deuterium oxide to the prepared sample. Microanalytical determinations were made by W. N. Rond. X-ray powder diffraction data give interplanar spacings, $\AA$, for Cu $K_\alpha$ radiation. Camera diameter was 114.59 mm. Relative intensities were estimated visually: s, strong; m, moderate, w, weak; v, very. The strongest lines are numbered (1, strongest). Thin-layer chromatography was performed with Desaga equipment, by using Silica Gel G (E. Merck, Darmstadt, Germany) activated at 110°, with the solvent noted in parentheses, and with indication by sulfuric acid. Paper chromatography was effected by the descending method with Whatman No. 1 paper, and either 40:11:19 butyl
alcohol—ethanol—water (solvent A) or 5:5:3:1 pyridine—ethyl acetate—water—acetic acid (solvent B) (152) as eluant, and indication by the silver nitrate-sodium hydroxide method (184).


Preparation of 2-acetamido-2-deoxy-α-D-glucose (II)

This compound was made by following essentially the method described by Horton (158). Sodium (23 g., 1 mole) was dissolved in ice-cold methanol (1 l). This solution was brought to room temperature and powdered 2-amino-2-deoxy-α-D-glucose hydrochloride (220 g., 1.02 moles) was added. The mixture was gently stirred for 4-5 min. for thorough mixing and was then filtered through a Buchner funnel with gentle suction. The filter was washed with four 100-ml portions of methanol. The combined filtrate was treated without delay with acetic anhydride (120 ml., 1.26 mole) and the stoppered flask was cooled under the tap for a few minutes to moderate the initial reaction. The solution was set aside at room temperature overnight, and then refrigerated for a few hours to complete crystallization. The product was collected by filtration and dried by suction; yield 208 g (94%), m.p. 203-205° (dec.), $[\alpha]_D^0 + 75 \rightarrow + 41 \, (c \, 1.0, \, \text{water})$. The product was analytically pure.

Preparation of 2-acetamido-2-deoxy-D-glucose diethyl dithioacetel (III)

This compound was prepared by an adaptation of the procedure described by Wolfrom and Anno (139). 2-Acetamido-2-deoxy-α-D-glucose
(II, 30 g) was dissolved in concentrated hydrochloric acid (120 ml) and stirred mechanically with ethanethiol (120 ml) for 24 hr. at 0°. The reaction mixture was poured into a vigorously agitated mixture of methanol (1000 ml) and an excess of basic lead carbonate (800 g). After the addition of a further 500 ml. of methanol, the solids were removed by filtration and washed with 1000 ml. of methanol. The filtrate and washings were evaporated under reduced pressure (temp. 40-50°) to a sirupy residue that was evaporated twice with absolute ethanol, and the residue was crystallized from methanol—ether, yield 32 g (72%), m.p. 130-131° (lit. 139) m.p. 129.5-130.5°), \( R_f \) 0.81 (t.l.c., methanol).

Preparation of 2-acetamido-2-deoxy-3,4:5,6-di-O-isopropylidene-D-glucose diethyl dithioacetal (IV)

This compound was prepared by the procedure of Yoshimura and Sato (140). Into a suspension of 2-acetamido-2-deoxy-D-glucose diethyl dithioacetal (III, 29 g) in acetone (500 ml), was dropped concentrated sulfuric acid (20 ml). The suspension became clear on addition of the acid. After having been stirred for 3 hr. at 25-30°, the reaction mixture was poured into an excess of cold, aqueous, sodium hydrogen carbonate to neutralize the acid. The solution was evaporated until a sirup or oil appeared, and then it was extracted with three 100 ml. portions of chloroform. The extract was washed with water, dried over magnesium sulfate, and evaporated in vacuo to a sirup. The resulting sirup was distilled, b.p. 155-160°/0.05 mm Hg, yield 24.7 g (69%). The sirup was left in evacuated desiccator for 2-3 days whereupon it
crystallized. Recrystallization from hexane gave analytically pure product m.p. 67-68°. Yoshimura and Sato (140) gave m.p. 66-67°.

**Preparation of 2-acetamido-2-deoxy-3,4-O-isopropylidene-D-glucose diethyl dithioacetal (V)**

This compound was prepared by the procedure of Yoshimura and Sato (140). The diisopropylidene derivative (IV, 8 g) was dissolved in acetone (100 ml). To this solution 1 N sulfuric acid (100 ml) was added, and the solution was kept for 40 min. at 40°. The solution was cooled, neutralized with sodium hydrogen carbonate, and after evaporation of the acetone, extracted with chloroform. The extract was washed with water, dried over magnesium sulfate, filtered and evaporated to a sirup that readily crystallized. It was recrystallized from methanol-ether; yield 4.3 g (60%), m.p. 137-138°, \([\alpha]_D^{21}\) 26.0 ± 1° (c 2.35, methanol); \(\lambda_{\text{KBr}}^{\text{max}} 2.91 \mu\) (OH), 3.10 \(\mu\) (NH), 6.05, 6.49 \(\mu\) (NHAC), 7.30 \(\mu\) (CMe₂); X-ray powder diffraction data: 8.54 s (1), 7.25 w, 6.15 s (2), 5.70 w, 5.03 s (3), 4.36 m, 3.98 m, 3.48 m, 3.62 m, 3.41 w; homogeneous by thin layer chromatography, \(R_f 0.81\) (methanol), \(R_f 0.19\) (1:1 ether—ethyl acetate).

Yoshimura and Sato (140) gave m.p. 136-137°, \([\alpha]_D^{28}\) 6.7 (c 2.1, methanol) for this compound.

**2-Acetamido-1,2-dideoxy-3,4-O-isopropylidene-D-glucitol (VI)**

To a solution of 2-acetamido-2-deoxy-3,4-O-isopropylidene-D-glucose diethyl dithioacetal (V, 0.92 g) in absolute ethanol (100 ml)
was added an excess (10-15 g) of Raney nickel (185) and the mixture was

(185) Raney Nickel Catalyst No. 28, The Raney Catalyst Division of
the W. R. Grace Co., Inc., Chattanooga, Tennessee, U. S. A.

refluxed for 6 hr. The catalyst was filtered, washed several times with
hot ethanol, and the filtrate and washings were evaporated to a glass,
yield 0.56 g (90%), [α]_D^{19} = 17° (c 1, ethanol), λ_{max}^{film} 3.05 μ (OH, NH),
6.10, 6.45 μ (NHAc), 7.30 μ (CMC₂); n.m.r. data (deuteriochloroform):
τ 8.77 (3-proton doublet J \_1,\_2 = 7.0 c.p.s., 1-CH₃); τ 8.62 (6-proton
singlet, CMC₂); τ 8.00 (3-proton singlet NAc). The product was homo-
geneous by thin layer chromatography, R_f 0.65 (methanol), R_f 0.10
1:1 ether—ethyl acetate).


2-Acetamido-5,6-di-O-acetyl-1,2-dideoxy-3,4-O-isopropylidene-D-glucitol
To a solution of 2-acetamido-1,2-dideoxy-3,4-O-isopropylidene-
D-glucitol (VI, 0.48 g) in pyridine (5 ml) at 0° was added acetic
anhydride (1 ml) and the mixture was maintained for 1 hr. at 0°. After
a further 24 hr. at room temperature, the mixture was stirred
with ice, and extracted with chloroform. The extract was washed
successively with cold dilute hydrochloric acid and aqueous sodium
hydrogen carbonate, and then dried (magnesium sulfate), and evaporated.
The residue was distilled, and the distillate crystallized spontaneously,
yield, 0.392 g (62%); m.p. 74-76°, b.p. 175° (bath temp.) 0.1 mm Hg;
[α]_D^{25} + 36 ± 1° (c 0.6, chloroform), λ_{max}^{film} 3.04 μ (NH), 5.74 μ (OAc),
6.09, 6.55 μ (NHAc), 7.30 μ (CMC₂); n.m.r. data (deuteriochloroform):
τ 8.77 (3-proton doublet, $J_{1,2} = 7.0$ c.p.s., 1-CH$_3$); τ 8.59 (6-proton singlet CMe$_2$); τ 8.00 (3-proton singlet, NAc); τ 7.97 (3-proton singlet, OAc); τ 7.89 (3-proton singlet, OAc). The product was homogeneous by thin layer chromatography, $R_F$ 0.75 (methanol) $R_F$ 0.34 (1:1 ether—ethyl acetate).

Anal. Calcd. for C$_{15}$H$_{25}$NO$_2$: C, 54.38; H, 7.55; N, 4.23. Found: C, 54.52; H, 7.44; N, 4.20.

2-Acetamido-1,2-dideoxy-3,4,5,6-di-O-isopropylidene-D-glucitol (IX)

To a solution of 2-acetamido-2-deoxy-3,4,5,6-di-O-isopropylidene-D-glucose diethyl dithiaoacetal (IV, 2.44 g) in absolute ethanol (100 ml) was added an excess (20-25 g) of Raney nickel and the mixture was refluxed for 6 hr. The catalyst was filtered, washed several times with hot ethanol, and the filtrate and washings were evaporated to a sirup which crystallized on standing, yield 1.53 g (90%), m.p. 84-5°.

Anal. Calcd. for C$_{14}$H$_{25}$NO: C, 58.53; H, 8.71; N, 4.88. Found: C, 58.57; H, 8.84; N, 4.92.

4-Acetamido-4,5-dideoxy-2,3-O-isopropylidene-aldehydo-L-xylose (VII)

To a solution of 2-acetamido-1,2-dideoxy-3,4-O-isopropylidene-D-glucitol (VI, 1.474 g) in ethanol (10 ml) was added 0.9 N aqueous periodic acid (11.1 ml). The mixture was kept for 15 min. in the dark, excess barium carbonate was added, the neutralized solution was filtered, and the solution was evaporated. The residue was dissolved in ethanol, the solution was re-evaporated, the sirup was dissolved in benzene, and the solution was centrifuged. The supernatant was evaporated to give the product as a clear sirup, yield 1.05 g (83%); $[\alpha]_{D}^{20} - 15° (15$ min) → -12.6 ± 1.0° (c 1, ethanol); $\lambda_{\text{max}}^\text{film} 3.10 \mu$ (NH), 5.81 \mu
(CHO), 6.10, 6.50 μ (NHaC), 7.30 μ (CMe2); n.m.r. data (deuteriochlorofo-
form): τ 8.73 (3-proton doublet, J4.5 = 6.5 c.p.s., 5-CH3); τ 8.60
(6-proton singlet, CMe2); τ 8.00 (3-proton singlet, NAc); τ 0.67
(1-proton singlet, CHO). The product was homogeneous by thin-layer
chromatography, Rf 0.74 (methanol), Rf 0.24 (1:1 ether—ethyl acetate),
and by paper chromatography, Rf 0.85 in solvent A. It recolorized
Schiff reagent and gave a positive Fehling test.

Anal. Calcd. for C15H27NO4: C, 55.81; H, 7.90; N, 6.51. Found:
C, 55.56; H, 8.01; N, 6.03.

4-Acetamido-4,5-dideoxy-2,3-O-isopropylidene-L-xylose benzylphenylhydrazone

A solution of 4-acetamido-4,5-dideoxy-2,3-O-isopropylidene-
aldehyde-L-xylose (VII, 315 mg) in 95% ethanol (4 ml) was mixed with a
solution of sodium acetate-3H2O (1.039 g) in water (4 ml), and benzyl-
phenylhydrazine hydrochloride (412 mg) was added. The mixture was re-
fluxed for 2.5 hr. with addition of a few drops of ethanol to maintain
a homogeneous solution. The solvent was evaporated under a stream of
nitrogen, water (4 ml) was added to the residue, and the product was
extracted with chloroform. The extract was washed with water, dried
(sodium sulfate), evaporated, and the residue was crystallized from
ether, yield 200 mg (36%); m.p. 151-152°, [α]22D +32 ± 2° (c 0.5
methanol);λKBr max 3.10 μ (NH), 6.10, 6.42 μ (NHaC), 6.28, 6.70, 6.90 μ
(aryl C=C), 7.30 μ (CMe2), 14.35 μ (substituted benzene); n.m.r. data
(deuteriochloroform): τ 8.79 (3-proton doublet, 5-CH3); τ 8.67, 8.59
(6 protons, CMe2); τ 8.05 (3 protons, NAc); τ 2.75 (10 protons, aryl);
X-ray powder diffraction data 12.72 w, 8.30 m, 7.56 m, 6.83 s (1),
6.19 m, 5.54 w, 5.26 m, 4.74 s (2), 4.46 m, 4.24 w, 4.01 s (3),
3.74 w, 3.59 vw, 3.50 vw, 3.39 vw, 3.28 m.

Anal. Calcd. for C H N O : C, 69.87; H, 7.34; N, 10.63. Found:
C, 69.76; H, 7.68; N, 10.59.

4-Acetamido-4,5-dideoxy-α, β-L-xylofuranose (VIII)

A solution of 4-acetamido-4,5-dideoxy-2,3-O-isopropylidene-aldehyde-
L-xylose (VII, 0.36 g) in a mixture of acetic acid (1.0 ml) and water
(1.0 ml) was heated for 2.5 hr. at 95°, and the solution was evaporated.
Paper chromatography (solvent A) revealed the presence of two coalescent
zones, R 0.49 and 0.54, together with a small proportion of a component,
F 0.83, corresponding to starting material (VII). The sirupy product
was extracted with benzene, and the extract was evaporated to a sirup
(20 mg), which was found to be chromatographically homogeneous starting
material. The benzene-extracted sirup was found by chromatography to
be free from VII; it gave a single zone, R 0.78 in solvent B, two
coaalescent zones R 0.49 and 0.54, relative proportion 2:3 in solvent A,
and was formulated as VIII, yield 0.18 g (67%); [α] D 21 -10 ± 2°
(c 0.5 water); λ max 3.02 μ (OH), 6.12 μ (NAC), slight trace of ab-
sorption at 5.80 μ (CHO) and 6.50 μ (NHAC), no absorption at 7.30 μ
(CMe 2 ); n.m.r. data (deuterium oxide): τ 8.84 (3-proton multiplet,
5-CH 3 ); τ 8.01, 7.88, 7.85, 7.82 (3 protons, singlets, approximate
relative intensities 1:3:8:8, NAC); τ 5.87 (3-proton multiplet, H-2,
H-3, H-4); τ 4.82, 4.68 (1 proton, two multiplets, H-1, β-L and α-L
anomers respectively). The spectrum showed little change when the
probe temperature was lowered to 0°, but when it was raised to 86° the
τ 8.84 signal was resolved into a pair of doublets, J 4,5 = 7.0 c.p.s.,
at τ 8.90 and 8.76, approximate intensity ratios 2:3 (5-CH₃ of α-L and β-L anomers, respectively). At 86° the τ 8.01 signal remained unchanged at about 5% of the total NAc signal intensity; the τ 7.88 and 7.85 signals coalesced, and the τ 7.82 signal was diminished in intensity.

The sirupy VIII was resolved by preparative paper chromatography in solvent A. Excised strips from the center of each zone, Rₖ 0.49 and 0.54, were eluted with the chromatography solvent and the separated components were rechromatographed. The two components migrated as single zones, Rₖ 0.44 and 0.52. The faster-moving zone was more dextrorotatory than the original mixture. Aqueous solutions of each separated component were heated for 1 hr. at 90°, evaporated, and rechromatographed. Each component gave a coalescent double zone indistinguishable from that of the original anomeric mixture (VIII).
EXPERIMENTAL

PART II

General methods were the same as those in Part I, except that melting points were determined with a Thomas-Hoover "Unimelt" apparatus (Arthur H. Thomas Co., Philadelphia, Pennsylvania). Unless otherwise stated, ethyl acetate was used as the developer for t.l.c. experiments in this section.

Preparation of 2-acetamido-2-deoxy-D-glucitol (161)

2-Amino-2-deoxy-D-glucose hydrochloride (100 g) was N-acetylated (138) and the resultant, crystalline 2-acetamido-2-deoxy-α-D-glucose (95-100 g), dissolved in 20% aqueous ethanol (1 l), was shaken in a 2-l autoclave with Raney nickel (40 g) for 36 hr. at 100° under an atmosphere of hydrogen at 1000 lb. in⁻² pressure. The catalyst was filtered from the cooled solution, the filtrate was concentrated, and the product, which crystallized on standing, was filtered and washed with a little ethanol; yield 92 g (91%), m.p. 150-151°, [α]²¹°d -9 ± 1° (c 1.3, water), [lit. (161) m.p. 153°, [α]²¹°d -11° (water)]; λmax KBr 3.00 μ (OH, NH), 6.05, 6.30 μ (NHAc), no absorption in the range 11.7-12.1 μ; X-ray powder diffraction data: 16.06 m, 10.16 m, 8.93 vw, 6.70 vs (1,1), 4.92 vs (1,1), 4.69 m, 4.41 m, 4.27 m, 4.06 s (2,2), 3.91 s (2,2), 3.46 s (3).

Preparation of 2-amino-2-deoxy-D-glucitol-hydrochloride

2-Acetamido-2-deoxy-D-glucitol (30 g) was heated with 6N hydrochloric acid (400 ml) for 1 hr. at 100°, the solution was evaporated
(codistillation with propyl alcohol) and the crystalline residue was recrystallized from methanol—ether to give 2-amino-2-deoxy-D-glucitol hydrochloride; yield 25 g (85%), m.p. 160-162°. Lit. (161) m.p. 160-161°, $[\alpha]_D^{23} -2.7 \pm 0.6$° (c 1.8, water); ninhydrin positive; $\lambda_{\text{max}}^{\text{KBr}}$ 3.0 μ (OH), 3.30, 4.90, 6.18 μ (NH$_3^+$); X-ray powder diffraction data: 7.47 m, 5.65 m, 5.29 m, 4.53 w, 4.33 vs (1,1), 4.15 vs (1,1), 3.74 vs (1,1), 3.63 s (1,1), 3.44 w, 3.35 m, 3.26 s (2), 3.13 m, 3.00 m.

**Alternative preparation of 2-amino-2-deoxy-D-glucitol hydrochloride**

2-Amino-2-deoxy-D-glucose (30 g), dissolved in water (150 ml) was shaken in an autoclave with Raney nickel (20 g) in an autoclave for 48 hr. at 110° under an atmosphere of hydrogen at 1500 lb. in$^{-2}$ pressure. The catalyst was removed by filtration, the filtrate was concentrated down to a sirup which crystallized from methanol—ether; yield 20 g (66%).

**Preparation of 2-deoxy-2-(2,4-dinitroanilino)-D-glucitol (XVII)**

To a solution of 2-amino-2-deoxy-D-glucitol hydrochloride (20 g) in 50% aqueous ethanol (240 ml) was added sodium hydrogen carbonate (15.4 g) and 1-fluoro-2,4-dinitrobenzene (17.0 g), and the mixture was stirred for 18 hr. at room temperature. The yellow solid that separated was filtered off, washed with small amounts of water and ethanol, and then washed thoroughly with ether; yield 22 g (69%). Recrystallization from methanol gave XVII as fine yellow needles, m.p. 162-163°. Lit. (162) m.p. 163-164°, $[\alpha]_D^{24} + 95$° (c 1, methanol); $R_F$ 0.12; $\lambda_{\text{max}}^{\text{KBr}}$ 3.0 μ (NH, OH), 6.14, 6.30, 6.70 μ (aryl C=C), 7.44 μ (NO$_2^-$), 13.45 μ (substituted benzene); X-ray powder diffraction data: 9.11 w, 8.19 m, 7.19 m, 6.23 w, 5.40 vw, 4.74 s (2,2), 4.62 m, 4.21 s (2,2), 3.98 m, 3.83 s (2,2), 3.61 vw, 3.38 vs (1).
2-Deoxy-2-(2,4-dinitroanilino)-3,4:5,6-di-O-isopropylidene-D-glucitol (XVIII)

A solution of XVII (11 g) in dry acetone (200 ml) was shaken with concentrated sulfuric acid (2 ml) and anhydrous cupric sulfate (15 g) for 21 hr. at room temperature. The mixture was filtered, the filtrate was poured into an excess of aqueous sodium hydrogen carbonate, the acetone was evaporated, and the resultant solution was extracted with 3 X 100-ml portions of chloroform. The combined extract was washed with water, dried (sodium sulfate), evaporated, and the crystalline residue was re-crystallized from ether; yield 8.5 g (63%), m.p. 164-165°, [α]_D^{20} + 103 ± 1° (c 0.7, chloroform); Rf 0.9; χ_{max}^{KBr} 2.91 μ (OH), 3.08 μ (NH), 6.20, 6.32, 6.70 μ (aryl C=C), 7.30 μ (CMe₂), 7.55 μ (NO₂), 13.48, 13.80 μ (substituted benzene); n.m.r. data (chloroform-d); τ 0.85 (1-proton doublet, J 2.8 c.p.s., H-3'), τ 0.90 (1-proton broadened doublet, unchanged on deuteration, disappears after addition of 0.01 ml of tributylamine, NH), τ 1.72 (1-proton quartet, J 9.5 c.p.s., H-5'), τ 2.79 (1-proton doublet, H-6'), τ 5.48-6.52 (8-proton multiplet, H-1,2,3,4,5,6), τ 7.39 (1-proton triplet, J 6 c.p.s., disappears on deuteration, OH), τ 8.48, 8.57, 8.65, 8.68 (3-proton singlets, CMe₂); X-ray powder diffraction data: 8.84 m, 7.86 w, 6.88 vs (1), 6.60 vw, 6.02 m, 5.60 s (2), 5.14 w, 4.72 m, 4.15 m, 3.97 m, 3.86 m, 3.76 m, 3.59 w.

Anal. Calcd. for C_{18}H_{25}NO₉: C, 50.58; H, 5.85; N, 9.83. Found: C, 50.88; H, 5.73; N, 9.89.

2-Deoxy-2-(2,4-dinitroanilino)-3,4:5,6-di-O-isopropylidene-1-O-(p-tolylsulfonyl)-D-glucitol

A solution of XVIII (0.6 g) in pyridine (5 ml) was treated at 0° with
of both forms 53%), m.p. 128-129°, \( [\alpha]_D^{25} +78 \pm 2° \) (c 1, acetone); \\
R_F 0.22; \( \lambda_{\text{KBr}}^\text{max} 3.00 \mu \) (OH, NH), 6.18, 6.30, 6.60 \( \mu \) (aryl C=C), 7.20 \( \mu \) \\
\( \text{CMe}_2 \), 7.50 \( \mu \) (NO\(_2\)), 13.48 \( \mu \) (substituted benzene); n.m.r. data (acetone-
\( \delta \_6 \)): \( \tau 0.98 \) (1-proton doublet, \( J \) 2.7 c.p.s., H-3'), \( \tau 1.00 \) (1-proton
broad doublet, NH), \( \tau 1.82 \) (1-proton quartet, H-5'), \( \tau 2.71 \) (1-proton doublet, \( J_{5',6'} 9.5 \) c.p.s., H-6'); \( \tau 8.51, 8.60 \) (3-proton singlets, \( \text{CMe}_2 \)); X-ray powder diffraction data: 11.26 m, 9.99 w, 8.42 vw, 6.86 vs (2), 6.15 vw, 5.86 vw, 5.54 vs (1), 4.79 m, 4.48 m, 4.07 m, 3.78 m, 3.53 vw, 3.37 s (3), 3.27 s.

**Anal.** Calcd. for \( \text{C}_n \text{H}_m \text{N}_o \text{O} \): C, 46.51; H, 5.42; N, 10.85. Found: C, 46.30; H, 5.49; N, 11.06.

The preparation was repeated 20 times, and although the total yield of both forms was approximately the same each time, the proportion of the solvated form was frequently much higher than that described, and the ratio of the two forms appeared to depend on minor variations in experimental procedure. Recrystallization of the solvate from chloroform gave fine yellow needles, m.p. 95-96° (with effervescence, solidifying at higher temperature and remelting at 126-127°); \( R_F 0.22 \); X-ray powder diffraction data: 12.27 m, 9.40 w, 8.11 w, 6.81 m, 6.32 w, 5.90 w, 5.57 s (2), 5.12 vs (1).

**Anal.** Calcd. for \( (\text{C}_n \text{H}_m \text{N}_o \text{O}) \cdot \text{CHCl}_3 \): C, 43.12; H, 5.03; N, 9.84. Found: C, 42.72; H, 4.92; N, 10.34.

A sample of the solvate, kept for 2 hr. at 110° over phosphoric oxide, lost 10% of its weight (calcd. 9.4%) and gave the solvent-free form, m.p. and mixed m.p. 126-127°.

**Anal.** Calcd. for \( \text{C}_n \text{H}_m \text{N}_o \text{O} \): C, 46.51; H, 5.42; N, 10.85. Found: C, 46.33; H, 5.27; N, 11.11.

The solvate showed the same molar uptake of periodate as the non-solvated form, and the n.m.r. spectra of the two forms were identical except for the fact that the solvate showed an additional signal at \( \tau 2.80 \)
(singlet, 1/3 proton, CHCl\textsubscript{3}). The i.r. spectra (KBr disc) of the two forms were very similar, but were not completely superposable.

Periodate oxidation of 2-Deoxy-2-(2,4-dinitroanilino)-3,4-O-isopropyldiene-D-glucitol (XIX)

The Fleury-Lange method (186) was used.


To a solution of 2-deoxy-2-(2,4-dinitroanilino)-3,4-O-isopropyldiene-D-glucitol (XIX, 82.8 mg., 0.214 mmoles) in water (20 ml) was added 0.025 M sodium metaperiodate solution (25 ml), and the solution was made up to 50 ml. A blank determination was similarly prepared, but without sample. Aliquots (5 ml) were taken at varying time intervals, to which were added saturated sodium hydrogen carbonate solution (10 ml), 0.01 N sodium arsenite (20 ml), and 20% potassium iodide solution (2 ml). The solutions were stored in the dark for 15 min. at 0°, and then the excess sodium arsenite was titrated at 0° with 0.01 N iodine solution, using a starch indicator.

Blank = 7.9 ml.

Sample-Blank = A (ml)

\[ \text{Moles of NaIO}_4 \text{ consumed} = \frac{1}{214} \times A \times \frac{50 \text{ ml}}{5 \text{ ml}} \times 0.00107 \]

\[ \text{moles of sample} = 2.14 \times 10^{-4} \]
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<tr>
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2-Acetamido-1,5,6-tri-0-acetyl-2-deoxy-3,4-0-isopropylidene-D-glucitol

A solution of XIX (8 g) in acetone (280 ml) and water (120 ml) was passed slowly through a column (35 X 3.5 cm) of Dowex-1 (OH⁻) ion-exchange resin which had been pre-washed with 7:3 (v/v) acetone—water. The column was washed with 7:3 (v/v) acetone—water until the effluent gave a negative ninhydrin reaction, and the combined effluent was evaporated. The dried residual sirup (4.2 g) was dissolved in pyridine (25 ml), acetic anhydride (45 ml) was added, and the solution was kept for 18 hr. at room temperature. The solution was poured into ice and water (200 ml), and the mixture was extracted with 3 X 75-ml portions of chloroform. The extract was washed successively at 0° with dilute sulfuric acid, aqueous sodium hydrogen carbonate, and water, the dried (sodium sulfate) solution was evaporated, and the residue was crystallized from ether, yield 2.21 g m.p. 93-94°, [α]²⁵ D +31 ± 1° (c 1.7, chloroform); Rₚ 0.42; λ max 3.08 μ (NH), 5.73 μ (OAc), 6.08,
6.53 µ (NHAc), 7.28 µ (CMe₂); n.m.r. data (chloroform-d): τ 3.89 (l-proton doublet, J 8.5 c.p.s., NH), τ 4.80 (l-proton octet, width 16 c.p.s., H-5), τ 5.30-6.30 (7-proton multiplet, H-1,2,3,4,6), τ 7.89, 7.95 (singlets, 3 and 9 protons, acetyls), τ 8.59, 8.61 (3-proton singlets, CMe₂); X-ray powder diffraction data: 7.69 s (1), 6.97 w, 5.71 m, 5.43 m, 4.64 m, 4.25 s (2), 3.81 m.

Anal. Calcd. for C₂₁H₂₇NO₉: C, 52.4; H, 6.94; N, 3.60. Found: C, 52.52; H, 6.89; N, 3.84.

1,5,6-Tri-0-acetyl-2-deoxy-2-(2,4-dinitroanilino)-3,4-0-isopropylidene-D-glucitol

A solution of XIX (430 mg) in pyridine (4 ml) was treated with acetic anhydride (2.5 ml) at room temperature, and after 24 hr. the solution was poured into water. The product was extracted with dichloromethane and processed in the usual way to give the triacetate of XIX as a yellow glass, yield 435 mg (76%), Rf 0.54 (1:3 ethyl acetate—benzene); λmaxfilm 3.02 µ (NH), 5.77 µ (OAc), 6.20, 6.30, 6.60 µ (aryl C=C), 7.34 µ (CMe₂), 13.25 µ (substituted benzene); n.m.r. data in chloroform-d (See Fig. 3): τ 0.85 (l-proton doublet, J 2.7 c.p.s., H-3'), τ 1.01 (l proton, broad, NH), τ 1.60 (l-proton quartet, H-5'), τ 2.68 (l-proton doublet, J 9.5 c.p.s., H-6'), τ 4.78 (l-proton octet, width 16.5 c.p.s., H-5), τ 5.26-6.30 (7-proton multiplet, H-1,2,3,4,6), τ 7.85, 7.87, 7.98 (3-proton singlets, acetyls), τ 8.40, 8.50 (3-proton singlets, CMe₂).

The signal at $\tau$ 1.01 was not affected by deuterium, even after 57 hr. at room temperature, but the signal disappeared (Fig. 1) rapidly when 0.01 ml of tributylamine was added to the prepared sample, and a 1-proton singlet appeared at $\tau$ 5.31 (HOD).

Acetylation of the yellow solvate of XIX gave a product whose n.m.r. spectrum was identical to that recorded above.

4-Deoxy-4-(2,4-dinitroanilino)-2,3-O-isopropylidene-aldehydo-L-xylose (XX)

A solution of sodium metaperiodate (1.81 g, 1.1 molar equiv.) in water (30 ml) was added to a solution of XIX (3.0 g) in 1:4 ethanol—water (75 ml). The mixture was kept for 25 min. in the dark at room temperature, and was then evaporated at 30°. Anhydrous sodium sulfate (3 g) was added to the residue, and the solid mixture was extracted repeatedly with ether. Evaporation of the extract gave XX as a yellow glass, yield 2.4 g (87%); $R_F$ 0.40; $\lambda_{\text{film}}$ max 2.95 $\mu$ (OH), 3.05 $\mu$ (NH), 3.50, 5.80 $\mu$ (CHO), 6.20, 6.30, 6.65 $\mu$ (aryl C=C), 7.35 $\mu$ (CMe$_2$), 7.55 $\mu$ (NO$_2$), 12.25, 13.2 $\mu$ (substituted benzene).

Anal. Calcd. for C$_{14}$H$_{17}$N$_3$O$_8$: C, 47.32; H, 4.79; N, 11.83. Found: C, 47.18; H, 5.46; N, 11.77.

4-Deoxy-4-(2,4-dinitroanilino)-L-xylopyranose (XXI)

A solution of XX (3.29 g) in 50% aqueous acetic acid (16 ml) was heated for 2.5 hr. at 95°, and then evaporated. Addition of ethanol to the residual sirup gave the crystalline product, yield 2.54 g (84%), m.p. 194-195° (dec.), $[\alpha]_{D}^{23} + 37 \pm 1^o$ (c 1, acetone); $R_F$ 0.15; $R_F$ 0.69 (papergram, 40:11:19 butyl alcohol—ethanol—water); $\lambda_{\text{KBr}}$ max 2.9 $\mu$ (OH), 3.05 $\mu$ (NH), 6.20, 6.30, 6.60 $\mu$ (aryl C=C), 7.52 $\mu$ (NO$_2$), 13.50,
14.0 μ (substituted benzene); X-ray powder diffraction data: 10.46 m, 6.02 m, 5.08 vs (1), 4.44 m, 4.06 m, 3.44 s (3), 3.20 s (2), 3.06 vw, 2.91 vw.

Anal. Calcd. for C_{11}H_{13}N_{0.8}: C, 41.90; H, 4.21; N, 13.33.
Found: C, 41.42; H, 4.38; N, 13.59.

1,2,3-Tri-O-acetyl-4-deoxy-4-(2,4-dinitroanilino)-L-xylopyranose

To a solution of XXI (0.5 g) in pyridine (6 ml) was added acetic anhydride (3 ml). The mixture was kept for 24 hr. at room temperature, and then processed in the usual way to give the product as a yellow, distillable glass; yield 0.35 g (50%); R_f 0.60 and 0.70 (1:1 ethyl acetate—benzene); λ_{film}^{max} 3.05 μ (NH), 5.70 μ (OAc), 6.15, 6.30, 6.60 μ (aryl C=O), 13.40 μ (substituted benzene); n.m.r. data (chloroform-d): τ = 0.88, 1.0-1.2, 1.68, 2.78 (multiplets, 4 protons, H-3',5',6', and NH, of anomers), τ 7.77, 7.81, 7.91, 7.95 (singlets, 9 protons, acetyls).

Anal. Calcd. for C_{17}H_{19}N_{1.1}: N, 9.52. Found: N, 9.74.

4-Deoxy-4-(2,4-dinitroanilino)-1,2-O-isopropylidene-α-L-xylopyranose (XXII)

A solution of XXI (1.84 g) in acetone (400 ml) was shaken with anhydrous cupric sulfate (5 g) and conc. sulfuric acid (0.5 ml) for 1 day at room temperature. The mixture was filtered, the filtrate was neutralized with aqueous sodium hydrogen carbonate, acetone was evaporated, and the product was extracted with 3 X 75-ml portions of ethyl acetate. The extract was washed with water, dried (sodium sulfate) and evaporated, and the crystalline residue was recrystallized from ethyl acetate, yield 1.64 g (79%), m.p. 220-222° (dec.), [α]_D^{25} +
175 ± 2° (c 1.1, acetone); R 0.70; λ max

\[ \lambda_{\text{max}} \approx 3.00 \mu \text{ (OH, NH), 6.20, 6.32, 6.62 \mu (aryl C=C), 7.22, 7.30 \mu (CMe}_2\), 7.56 \mu (NO}_2\), 13.42, 14.10 \mu \text{ (substituted benzene); n.m.r. data (pyridine):} \]

\[ \tau 4.39 \text{ (1-proton doublet,} J_1,2 \text{ 2 c.p.s., H-1), } \tau 5.24-6.13 \text{ (5-proton multiplet, H-2,3, 4,5), } \tau 8.23, 8.60 \text{ (3-proton singlets, CMe}_2\); X-ray powder diffraction data: 13.29 w, 8.97 w, 7.86 w, 6.70 m, 5.57 vs (1), 5.15 s (2), 4.79 s (3,3), 4.53 s (3,3), 4.27 s (3,3), 4.09 s (3,3), 3.77 s (3,3), 3.63 s (3,3).

**Anal. Calcd. for C H N O:** C, 47.32; H, 4.82; N, 11.83. **Found:** C, 47.55; H, 5.09; N, 12.23.

Only one product could be detected by t.l.c. in the above preparation, and no difference in the yield of XXII was observed when the time of reaction was extended to 4 days.

3-O-Acetyl-4-deoxy-4-(2,4-dinitroanilino)-1,2-O-isopropylidene-α-L-xylopyranose

A solution of XXII (175 mg) in pyridine (2 ml) and acetic anhydride (3 ml) was kept 1 day at room temperature, and then poured into ice and water. The precipitated solid was filtered off, washed with water, dried, and recrystallized from methanol; yield 130 mg (66%), m.p.

171-172°, [α]_D^{21} + 190 ± 2° (c 1, chloroform); R 0.84; λ \[ \lambda_{\text{max}} \approx 3.02 \mu \text{ (NH), 5.77 \mu (OAc), 6.18, 6.30, 6.60 \mu (aryl C=C), 7.30 \mu (CMe}_2\), 13.45, 14.10 \mu \text{ (substituted benzene); n.m.r. data (chloroform-d):} \]

\[ \tau 0.84 \text{ (1-proton doublet,} J_0,5,5' \text{ 2.7 c.p.s., H-3'), } \tau 0.88 \text{ (1-proton, broad, disappears on addition of tributylamine in deuterium oxide, NH),} \]

\[ \tau 1.66 \text{ (1-proton quartet, H-5'), } \tau 2.66 \text{ (1-proton doublet,} J_5',6' \]
9.5 c.p.s., H-6), τ 4.69 (2-proton multiplet, width 10 c.p.s., H-1, H-3), τ 5.92-6.38 (4-proton multiplet, H-2,4,5), τ 7.78 (3-proton singlet, OAc), τ 8.33, 8.62 (3-proton singlets, CMe₂).

Anal. Calcd. for C₁₆H₁₉NO₄: C, 48.36; H, 4.78; N, 10.58. Found: C, 47.75; H, 4.61; N, 10.72.

4-Amino-4-deoxy-1,2-0-isopropylidene-α-L-xylopyranose (XXIII)

A solution of XXII (0.72 g) in acetone (60 ml) and water (20 ml) was stirred with Dowex-1 (OH⁻) ion-exchange resin, added in small portions at 45-50° until the solution became colorless. The mixture was filtered, and the resin was washed with hot methanol (500 ml).

The filtrate and washings were evaporated to a colorless sirup which crystallized from ether, yield 0.247 g (63%), m.p. 131-132°, [α]D²⁴ + 32.5 ± 1° (c 1, methanol), λmaxBr 3.4, 6.3 μ (NH), 7.25 μ (CMe₂);

X-ray powder diffraction data: 7.97 vs (2), 6.23 w, 5.79 vw, 5.27 vs (1), 5.01 m, 4.77 m, 4.39 s (3), 4.00 m, 3.64 m, 3.14 vw, 3.01 vw, 2.85 m.


Periodate oxidation of 4-amino-4-deoxy-1,2-0-isopropylidene-α-L-xylopyranose (XXIII)

The Fleury-Lange method (186) was used.

To a solution of 4-amino-4-deoxy-1,2-0-isopropylidene-α-L-xylopyranose (XXIII, 0.4 mg., 0.214 mmoles) in water (20 ml) was added 0.025 M sodium metaperiodate solution (25 ml), and the solution was made up to 50 ml. A blank determination was similarly prepared, but
without sample. Aliquots (5 ml) were taken at varying time intervals, to which were added saturated sodium hydrogen carbonate solution (10 ml), 0.01 N sodium arsenite (20 ml), and 20% potassium iodide solution (2 ml). The solutions were stored in the dark for 15 min. at 0°, and then the excess sodium arsenite was titrated at 0° with 0.01 N iodine solution, using a starch indicator.

Blank = 7.9 ml.

Sample-Blank = A (ml)

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<th>Time</th>
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<th>Sample-Blank A (ml)</th>
<th>Moles of NaIO₄ Moles of sample</th>
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4-Aacetamido-3-O-acetyl-4-deoxy-1,2-O-isopropylidene-α-L-xylopyranose (XXIV)

A solution of XXIII (590 mg) in pyridine (5 ml) and acetic anhydride (5 ml) was kept for 1 day at room temperature, and then poured into ice and water. The product was extracted with 3 X 60-ml portions of dichloromethane, the extract was washed with aqueous sodium hydrogen
carbonate, dried (sodium sulfate) and evaporated (codistillation with toluene). The resulting sirup crystallized on storage, and re-crystallization from chloroform gave XXIV as fine needles, yield 596 mg (70%), m.p. 109-110°, \([\alpha]_{D}^{22} + 68 \pm 1°\) (c 1, chloroform); \(R_{F}^0\) 0.40; \(\lambda_{KBr}^{max} \) 3.04 \(\mu\) (NH), 5.73 \(\mu\) (OAc), 6.03, 6.52 \(\mu\) (NHAc), 7.22 \(\mu\) (CMe\(_2\)); n.m.r. data in chloroform-d (see Fig. 2): \(\tau\) 3.60 (1-proton broad doublet, \(J_8\) c.p.s., disappears on deuteriation, NH), \(\tau\) 4.91 (2-proton multiplet, H-1,3), \(\tau\) 5.85-6.50 (4-proton multiplet, H-2,4,5), \(\tau\) 7.90, 8.03 (3-proton singlets, acetyl), \(\tau\) 8.38, 8.63 (3-proton singlets, CMe\(_2\)); in benzene (see Fig. 2): \(\tau\) 4.72 (1-proton multiplet, width 10 c.p.s., H-3), \(\tau\) 4.96 (1-proton doublet, \(J_{1,2}\) 2.5 c.p.s., H-1), \(\tau\) 5.70-5.95 (1-proton multiplet) and \(\tau\) 6.20-6.50 (3-proton multiplet, H-2,4,5), \(\tau\) 8.33, 8.37 (3-proton singlets, acetyl), \(\tau\) 8.51, 8.82 (3-proton singlets, CMe\(_2\)); X-ray powder diffraction data: 8.88 m, 7.47 vs (3), 6.23 m, 5.54 vs (2), 5.15 s, 4.72 m, 4.37 vs (1), 4.08 m, 3.92 m, 3.58 m, 3.43 w, 3.18 m, 2.98 m.

\[\text{Anal. Calcd. for } C_{12} H_{19} NO_{6} \]: C, 52.74; H, 6.96; N, 5.12. Found: 52.79; H, 6.86; N, 5.05.

4-Acetamido-4-deoxy-1,2-0-isopropylidene-\(\alpha\)-L-xylopyranose (XXV)

A solution of XXIV (0.5 g) in anhydrous methanol (10 ml) was treated with a very small piece of metallic sodium, and after 2 hr. at room temperature the solution was neutralized with Amberlite IR-120 (H\(^+\)) ion-exchange resin. Evaporation of the solution gave XXV as a colorless sirup, yield 0.36 g (85%); \(R_{F}^0\) 0.15; \(\lambda_{film}^{max} \) 2.85-3.10 \(\mu\) (OH, NH), 6.10, 6.50 \(\mu\) (NHAc), 7.30 \(\mu\) (CMe\(_2\)).
4-Acetamido-4-deoxy-L-xylopyranose (XXVI)

(a) From 4-acetamido-4-deoxy-1,2-0-isopropylidene-α-L-xylopyranose (XXV). A solution of XXV (350 mg) in water (10 ml) was stirred with Amberlite IR-120 (H⁺) ion-exchange resin (1.5 g) for 2.5 hr. at 60°. The resin was filtered, washed with methanol, and the filtrate was evaporated to a chromatographically homogeneous syrup which crystallized after trituration with ethanol and ethyl acetate; yield 152 mg (53%). After recrystallization from methanol—ether the product had m.p. 155-157°, [α]D²⁴" -53 → -49° (c 3.3, water); R D Rhamnose 0.85 (papergram, 3:1:1 butyl alcohol—ethanol—water); λmax 3.00 μ broad (NH, OH), 6.18, 6.44 μ (MeAc); X-ray powder diffraction data: 9.30 m, 7.86 vw, 6.02 s (3,3), 5.71 vs (2), 5.09 w, 4.67 vs (1), 4.33 s (3,3), 4.03 s (3,3), 3.80 m, 3.64 m, 3.37 vw, 3.18 m, 3.05 m, 2.93 s.

Anal. Calcd. for C₁₇H₁₃NO₅: C, 43.97; H, 6.85; N, 7.32. Found: C, 43.67; H, 6.91; N, 7.69.

For this compound, prepared by a different route, Dick and Jones reported (177) m.p. 157-158°, [α]D²²" -22 → -16° (c 1, water) and R D Rhamnose 0.81 (papergram, 3:1:1 butyl alcohol—ethanol—water).

(b) From 4-amino-4-deoxy-1,2-0-isopropylidene-α-L-xylopyranose (XXIII). To a solution of XXIII (200 mg) in methanol (1 ml) and water (4 ml) was added acetic anhydride (1 ml), and the mixture was kept for 2 hr. at room temperature. Examination by t.l.c. (1:2 isopropyl alcohol—benzene) revealed that the starting material XXIII, Rf 0.17) was completely converted into XXV (Rf 0.72). Water (2 ml) was added and the solution was heated for 3 hr. at 90°, by which time con-
version of XXV into XXVI (R, 0.07) was complete. Evaporation of the
solution (codistillation with toluene), and crystallization of the
residue from ethanol—ether gave XXVI, yield 120 mg (60%), m.p.
155-157°, identical by mixed m.p. and i.r. spectrum with the product
prepared by procedure (a).

N.m.r. spectrum of 4-acetamido-4-deoxy-L-xylose

The n.m.r. spectrum of XXVI (Fig. 1), measured at 80° in deuterium
oxide with an equilibrated solution gave the following data: $\tau \, 4.78$
(doublet, $J_{1,2} \, 3.0$ c.p.s., H-1 of $\alpha$-L anomer) and $\tau \, 5.44$
(doublet, $J_{1,2} \, 7.6$ c.p.s., H-1 of $\beta$-L anomer) (total integral, 1 proton, relative
intensities 2:3), $\tau \, 5.89$-$6.28$ (5-proton multiplet, H-2,3,4,5), $\tau \, 7.99$
(3-proton singlet, NAc). The signal at $\tau \, 5.44$ was partially obscured
by the HO-D signal when the spectrum was measured at 40°.

A spectrum of XXVI, measured 2 min. after dissolution in deuterium
oxide, showed the signal at $\tau \, 4.78$. The relative intensity of the latter
was not appreciably different from its intensity in the spectrum of
the equilibrated solution. The spectrum of XXVI, measured 30 min.
after dissolution in methyl sulfoxide-$d_6$, showed signals at $\tau \, 4.79$
(H-1 of $\alpha$-L pyranose anomer) and $\tau \, 5.46$ (H-1 of $\beta$-L pyranose anomer),
a singlet at $\tau \, 8.00$ (NAc), and a broad signal, $\tau \approx 4.7$ which dis-
appeared on deuteration (NH). The spectrum of the deuterated sample,
measured at 80°, showed the H-1 signals as sharp doublets at $\tau \, 4.79$
($J_{1,2} \, 2.8$ c.p.s.) and $\tau \, 5.46$ ($J_{1,2} \, 7.2$ c.p.s.), total integral 1 proton,
in approximately 2:3 proportion. The H-2,3,4,5 signals were observed
as a 5-proton multiplet, $\tau \, 5.95$-$6.86$. 
Preparation of 2-amino-2-deoxy-D-glucose diethyl dithioacetal hydrochloride (XXVII) (165, 166)

2-Amino-2-deoxy-D-glucose hydrochloride (I, 30 g) was dissolved in concentrated hydrochloric acid (120 ml) that has been saturated at 0° with hydrogen chloride gas, and the mixture was stirred mechanically with ethanethiol (120 ml) for 20 hr. at 0°. The reaction mixture was processed by the method used in preparation of the 2-acetamido analog III; yield 38 g (83%), m.p. 82-83°. Lit. m.p. 79-80° (166).

Preparation of 2-deoxy-2-(2,4-dinitroanilino)-D-glucose diethyl dithioacetal (XXVIII) (167)

A mixture of 2-amino-2-deoxy-D-glucose diethyl dithioacetal hydrochloride (XXVII, 6.4 g), water (30 ml), sodium hydrogen carbonate (3.3 g), and ethanol (30 ml) was stirred for 10 min., 1-fluoro-2,4-dinitrobenzene (3.72 g) was added, and stirring was continued for 20 hr. at room temperature. The solution was evaporated at 40°, the residue was extracted with ethyl acetate, and the extract was washed with water, then dried (magnesium sulfate). Evaporation of the solvent gave 2-deoxy-2-(2,4-dinitroanilino)-D-glucose diethyl dithioacetal as a sirup which was dried in a vacuum desiccator to a glass; yield 7.9 g (88%).

2-Deoxy-2-(2,4-dinitroanilino)-3,4:5,6-di-O-isopropylidene-D-glucose diethyl dithioacetal (XXIX)

The glass from the preceding preparation was dissolved in dry acetone (150 ml), concentrated sulfuric acid (5 ml) was added, and the reaction mixture was stirred for 3 hr. at room temperature. The
acid was neutralized by pouring the reaction mixture in cold, aqueous, sodium hydrogen carbonate. The solution was evaporated to remove acetone, and was then extracted with three 100-ml portions of chloroform. The extract was washed with water, dried over magnesium sulfate and evaporated in vacuo to a sirup, which was crystallized from ether—petroleum ether; m.p. 94-95°; \( R_f \) 0.92 (9:1 benzene—methanol); \( \lambda_{\text{max}}^{KBr} \), 3.02 \( \mu \) (NH); 6.20, 6.32, 6.65 \( \mu \) (aryl C=C); 7.30 \( \mu \) (CMe₂); 7.51 \( \mu \) (NO₂); 13.4 \( \mu \) (substituted benzene).

Anal. Calcd. for C₁₈H₁₄N₂O₁₀S₂: C, 49.71; H, 6.21; N, 7.90; S, 12.05. Found: C, 49.83; H, 6.18; N, 7.87; S, 11.45.

2-Deoxy-2-(2,4-dinitroanilino)-3,4:5,6-di-O-isopropylidene-aldehydo-D-glucose (XXX)

An adaptation of the procedure of Defaye (168) was followed.

2-Deoxy-2-(2,4-dinitroanilino)-3,4:5,6-di-O-isopropylidene-D-glucose diethyl dithiacetal (XXIX, 0.531 g) was dissolved in ether (12 ml) and water (8 ml) was added, together with an excess of calcium carbonate. To the vigorously stirred mixture, a solution of bromine (0.08 ml) in ether (7 ml) containing water (0.25 ml) was added slowly. After the addition was complete, the calcium carbonate was removed by filtration, the ether extract was washed with sodium hydrogen carbonate, then with water, dried over sodium sulfate, in the presence of some calcium carbonate, filtered and evaporated to a sirup; yield 0.225 g (53%). The product was homogeneous on thin-layer chromatography, \( R_f \) 0.52 (9:1 benzene—methanol); \( \lambda_{\text{max}}^{\text{film}} \), 3.01 \( \mu \) (NH); 5.80 \( \mu \) (CHO); 6.18, 6.30, 6.64 \( \mu \) (aryl C=C); 7.3 \( \mu \) (CMe₂), 3.14 \( \mu \) (substituted benzene).
Attempted synthesis of 2-deoxy-2-(2,4-dinitroanilino)-3,4:5,6-di-o-isopropylidene-D-glucitol by reduction of XXX

The sirupy product (XXX, 0.2 g) was dissolved in methanol (10 ml). On addition to that solution of one drop of sodium borohydride solution in methanol, the reaction mixture turned deep red in color due to the cleavage of the 2,4-dinitrophenyl group.