704. *The* α- *and the* β-*Form of* 2 : 3 : 4 : 6-*Tetra-acetyl* D-*Galactopyranose Anilide.*

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Acetylation of D-galactose anilide under the appropriate conditions gives the α- and the β-pyranose form of 2 : 3 : 4 : 6-tetra-acetyl D-galactose anilide. It has also been found that the α-form may be made by treatment of 2 : 3 : 4 : 6-tetra-acetyl D-galactose with alcoholic aniline, while the β-form is produced by reaction of 2 : 3 : 4 : 6-tetra-acetyl α-D-galactopyranosyl bromide with aniline. The two isomers are interconvertible, and removal of the aniline group from each of them by the agency of formic acid gives 2 : 3 : 4 : 6-tetra-acetyl D-galactose.

SUGAR anilides and methylated sugar anilides usually display mutarotation (Irvine and McNicoll, *J.*, 1910, **97**, 1450; Sorokin, *J. pr. Chem.*, 1888, **37**, 292). It has been recognised that this phenomenon is probably due to the existence in solution of more than one form of the anilide, but the structural changes which are accompanied by mutarotation have not been ascertained because the isomeric anilide formed during mutarotation has never been isolated. It would, therefore, appear that that form of the anilide produced during mutarotation is stable only in solution. Support and illustration of this follows from the fact that removal of solvent from the solution of an anilide which has attained equilibrium regenerates the original anilide, and unless the anilide is sensitive to traces of water, the mutarotation and regeneration may be repeated indefinitely.

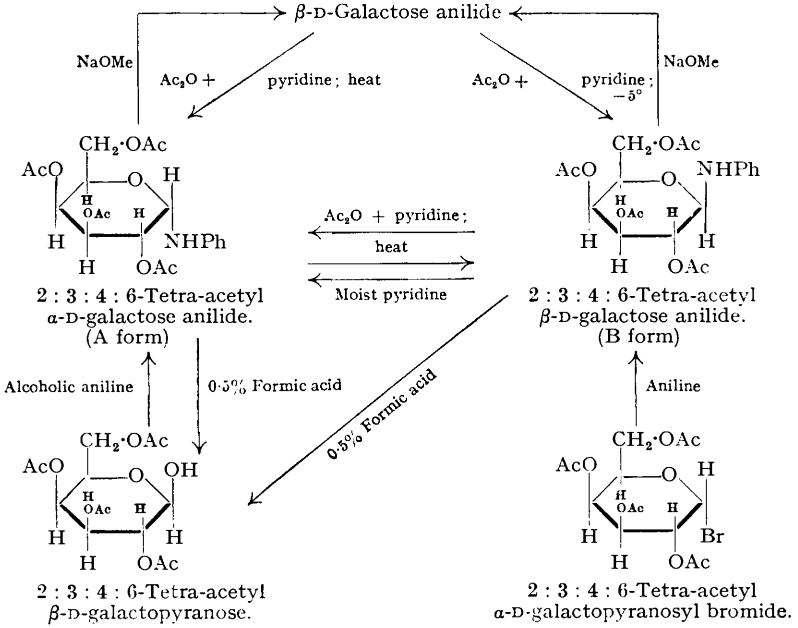
It should be noted, in this connection, that D-ribose affords two anilides, one of which is said to have a pyranose and the other a furanose structure; the former may be converted into the latter in boiling alcohol (Lee, *et al.*, *J. Org. Chem.*, 1946, **11**, 75; U.S.P. 1945, 2,384,102, 2,384,103, 2,384,104; Howard *et al.*, *J.*, 1946, 855). The mutarotation of anilides, however, cannot be due solely to an equilibrium between furanose and pyranose forms, because those anilides which cannot have a furanose structure such as 2 : 3 : 4 : 6-tetramethyl D-galactose anilide also exhibit mutarotation (Haworth and Leitch, *J.*, 1918, **113**, 197).

D-Galactose anilide behaves like its 2 : 3 : 4 : 6-tetramethyl derivative in that it exists only in a single crystalline form which displays mutarotation in suitable solvents and can be recovered from the equilibrium solution as the original crystalline substance. Although conditions have not been ascertained for isolating the second form of galactose anilide present in an equilibrium solution, this work shows that acetylation enables the mutarotation to be arrested and two crystalline forms of tetra-acetyl D-galactose anilide to be separated. The evidence in favour of assigning pyranose structures to these two forms is based upon the following experimental facts: D-galactose anilide (which is probably the β-isomer since it shows an upward mutarotation), prepared by boiling D-galactose with methyl-alcoholic aniline (Irvine and McNicoll, *loc. cit.*), gives, on acetylation with an excess of a mixture of acetic anhydride and pyridine either at room temperature or on warming, a crystalline tetra-acetyl D-galactose anilide, m. p. 176°, $[\alpha]_D^{20}$ +238° (in pyridine), designated the A-form. When the acetylation of D-galactose anilide is carried out with the calculated amount of acetic anhydride in pyridine solution at −5°, there is produced an isomeric crystalline tetra-acetyl galactose anilide, m. p. 121°, $[\alpha]_D^{18}$ −58° (in pyridine), called the B-form. Catalytic deacetylation of either the A- or the B-form of the anilide by Zemplén's method (*Ber.*, 1923, **56**, 1705; 1926, **59**, 1258; 1936, **69**, 1827) gives rise to the original D-galactose anilide.

The B-form may be transformed into the A-form by treatment with a mixture of pyridine and acetic anhydride at room temperature. Furthermore, each of the two forms undergoes mutarotation in moist pyridine to give a mixture of both, which can be separated by fractional crystallisation. These transformations are shown in the scheme below.

When either the A- or the B-form is warmed with 0·5% formic acid in aqueous alcohol or aqueous acetone there is formed in good yield the characteristic crystalline 2 : 3 : 4 : 6-tetra-acetyl β-D-galactopyranose. In the absence of acyl migration, which seems most improbable, this observation indicates that both the A- and the B-form of the anilide have the pyranose configuration. Further evidence for this conclusion follows from the fact that treatment of 2 : 3 : 4 : 6-tetra-acetyl D-galactopyranose with alcoholic aniline yields the A-form of the anilide, while the reaction of 2 : 3 : 4 : 6-tetra-acetyl α-D-galactopyranosyl bromide with aniline gives the B-form. Since acetobromo-sugars normally yield β-glycosides, it is concluded that the B-form must be 2 : 3 : 4 : 6-*tetra-acetyl* β-D-*galactopyranose anilide*, from which it follows that the A-form must be the *α-anilide*.

The transformation of one acetylated anilide into the other is a common phenomenon in the mono- and the di-saccharide series (Frèrejacques, *Compt. rend.*, 1937, **204**, 1480). The facility of the conversion is analogous to the ease with which the α- and the β-form of 3 : 6-anhydromethylglucopyranoside can be transformed into the corresponding α- and β-forms of 3 : 6-anhydromethylglucofuranoside (Haworth, Owen, and Smith, *J.*, 1941, 88); the transformation also recalls the rapid conversion of the α-form of 2 : 4-dimethyl 3 : 6-anhydromethylgalactopyranoside into the β-form of the same sugar which is effected by traces of acids (Haworth, Jackson, and Smith, *J.*, 1940, 620). It seems not unlikely that in all these cases there is an intermediate, unstable, open-chain form in which $C_{(1)}$ carries a positive charge.



While the curious transformation observed in the 3 : 6-anhydro-derivatives of glucose and galactose is suggested to be due in part to the steric effect of the 3 : 6-anhydro-ring and to the formation of a positive carbonium ion at $C_{(1)}$, the new phenomenon of the occurrence of two isomeric anilides with the same ring structure recorded in this paper may be due partly to the influence of the electrophilic aromatic secondary amine group and also to the induced stability of the groups at $C_{(1)}$ due to the steric effect of the acetyl groups.

EXPERIMENTAL.

D-*Galactose Anilide.*—(*a*) A suspension of galactose (10 g.) in methyl alcohol (300 c.c.) containing aniline (10 c.c.) was boiled for 4 hours. During this process the galactose dissolved. Removal of the solvent by distillation in a vacuum followed by recrystallisation of the residue from methyl alcohol gave D-galactose anilide (10·4 g.), m. p. 151—153° (decomp.).

(*b*) A solution of aniline (1 g.) in ethyl alcohol (5 c.c.) was mixed with a solution of galactose (1 g.) in water (10 c.c.), the pH of which had been adjusted to 3—5 by addition of 0·01N-sulphuric acid. After being kept at room temperature for 4 days, the solution deposited crystals of D-galactose anilide, which when filtered off, washed with methyl alcohol, and dried (0·92 g.), had m. p. 157—159° (decomp.).

The anilide showed $[\alpha]_D^{15}$ −114·7° initial value (*c*, 5·4 in dry pyridine) changing in 48 hours to −56°. Removal of the solvent from the equilibrium solution left a colourless syrup which showed $[\alpha]_D^{19}$ −53° initial value (*c*, 5·4 in pyridine) changing to −55·3° in 3 days. Trituration of the syrupy mixture of the α- and the β-form of the anilide with methyl alcohol caused crystallisation of the original β-isomer (yield quantitative), m. p. 147—148° (decomp.), $[\alpha]_D^{19}$ −112° initial value in pyridine changing to −55·5° in 48 hours.

Acetylation of D-*Galactose Anilide.*—(i) (*a*) To a solution of galactose anilide (2 g.) in phosphoric oxide-dried pyridine (20 c.c.) at 0°, acetic anhydride (8 c.c.) was added. After the mixture had been kept for 18 hours at 0° and for 18 hours at room temperature, it was poured with stirring into ice-water (150—200 c.c.). The plastic mass which separated was triturated with water and kept overnight. Recrystallisation of the product from methyl alcohol gave 2 : 3 : 4 : 6-*tetra-acetyl* α-D-*galactose anilide* (1·74 g.), m. p. 175—176°, $[\alpha]_D^{15}$ +202° (*c*, 1·0 in chloroform), +238° (*c*, 1·0 in pyridine) [Found : C, 56·8; H, 6·1; N, 3·35; OAc (by hydrolysis), 39·2. $C_{20}H_{25}O_9N$ requires C, 56·75; H, 5·95; N, 3·31; OAc, 40·67%).

(*b*) To a hot solution of galactose anilide (2 g.) in dry pyridine (20 c.c.), acetic anhydride (8 c.c.) was added and the mixture was kept at room temperature for 18 hours. Isolation and crystallisation of the product in the normal manner gave 2 : 3 : 4 : 6-tetra-acetyl α-D-galactose anilide (0·96 g.), m. p. 175—176° (after recrystallisation from methyl alcohol).

(ii) Acetic anhydride (10 c.c.) was added to a solution of galactose anilide (5 g.) in dry pyridine (40 c.c.) cooled to 0°. The mixture was left at 0° for 18 hours and then poured into ice-water. After trituration of the product with water and crystallisation from methyl alcohol, 2 : 3 : 4 : 6-*tetra-acetyl β*-D-*galactose anilide* was obtained (2·0 g.), m. p. 120—121°, $[\alpha]_D^{15}$ −31·4° (*c*, 1·2 in chloroform), −58° (*c*, 1·0 in pyridine) (Found : C, 57·0 ; H, 6·0 ; N, 3·4 ; OAc, 39·3%).

(iii) To a solution of galactose anilide (3 g.) in dry pyridine (20 c.c.) cooled to −5°, acetic anhydride (5 c.c.) was added and the mixture kept at 0° for 2 days. Isolation of the product in the usual manner, followed by recrystallisation from methyl alcohol, gave both α- and β-forms of tetra-acetyl galactose anilide. Separation of the α-form was effected by flotation in methyl alcohol, and, after recrystallisation from this solvent, both forms were obtained in a pure state (α-form, 0·36 g. ; β-form, 2·53 g.).

Regeneration of D-*Galactose Anilide from the Acetates.*—When a solution of tetra-acetyl β-galactose anilide (0·5 g.) in absolute methyl alcohol (10 c.c.) was treated with a trace of sodium (*ca.* 1 mg.), crystals of D-galactose anilide were deposited after 2 days at room temperature, m. p. and mixed m. p. 153—154°.

A methyl-alcoholic solution (10 c.c.) of the α-form (0·1 g.) also yielded D-galactose anilide when treated in a similar manner.

Interconversion of the α- and the β-Form of Tetra-acetyl D-*Galactose Anilide.*—A 1% solution of either the α- or the β-form in dry pyridine showed little or no change in rotation when boiled for 7 hours. On the addition of 1 drop of water, mutarotation commenced and reached the same constant value. The α-form showed $[\alpha]_D^{20}$ +238° (initial value) changing to +20° in 3 hours. The β-form showed $[\alpha]_D^{18}$ −58° changing to +20° in 9 hours.

The products from each of the equilibrium solutions were isolated by pouring them into water. Fractional crystallisation from methyl alcohol gave in both cases the α- and the β-form of tetra-acetyl D-galactose anilide.

Treatment of the 2 : 3 : 4 : 6-*Tetra-acetyl* D-*Galactose Anilides with Acetic Anhydride and Pyridine.*—(*a*) *α-Anilide.* The acetylation mixture, consisting of tetra-acetyl α-galactose anilide (0·2 g.), dry pyridine (4 c.c.), and acetic anhydride (1·5 c.c.), was kept at 0° for 18 hours. Isolation and crystallisation of the product in the normal manner gave the original material (0·124 g.), m. p. 170—171°. When the treatment was carried out at room temperature, the solution showed : $[\alpha]_D^{19}$ +225° (initial value) ; +192° (2 hours) ; +160° (5 hours) ; +108° (10 hours) ; +80° (15 hours) +65° (20 hours) ; +40° (30 hours) ; +15° (45 hours) ; +6° (70 hours) (constant).

(*b*) *β-Anilide.* When a solution of 2 : 3 : 4 : 6-tetra-acetyl β-galactopyranose anilide (1 g.) in dry pyridine (20 c.c.) was treated with acetic anhydride (8 c.c.) at room temperature, it showed : $[\alpha]_D^{20}$ −58° (initial value) ; −38° (6 hours) ; −31° (10 hours) ; −17·5° (20 hours) ; −13° (25 hours) ; −9·5° (30 hours) ; −0·5° (50 hours) ; +2° (70 hours). After 18 hours a mixture of the α- and the β-form of tetra-acetyl galactopyranose anilide isolated from the reaction mixture (0·5 g.) had m. p. 114—121°. A mixture of the two forms was similarly obtained after the acetylation mixture had been left for 60 hours.

Preparation of Tetra-acetyl β-D-*Galactose Anilide from α-Acetobromo*-D-*galactose.*—A solution of α-acetobromo-D-galactose (8 g.) and aniline (2 c.c.) in dry ether (50 c.c.) was shaken with excess of silver carbonate for 2 days. After filtration, the ethereal solution was dried ($CaCl_2$), and the solvent removed in a vacuum. The residue crystallised spontaneously on addition of light petroleum. The crystals of 2 : 3 : 4 : 6-tetra-acetyl β-D-galactose anilide were filtered off, washed with ethyl alcohol, and dried (yield 5·5 g.) ; m. p. 119—120°. Evaporation of the mother-liquors at room temperature gave a mixture, m. p. 108—135°, of α- and β-tetra-acetyl galactose anilide (0·19 g.).

Preparation of Tetra-acetyl α-D-*Galactose Anilide from Tetra-acetyl β*-D-*Galactose.*—When a solution of 2 : 3 : 4 : 6-tetra-acetyl β-D-galactose (70 mg.) and aniline (20 mg.) in absolute ethyl alcohol (10 c.c.) was boiled for 2 hours and the solvent removed by distillation under reduced pressure, there was obtained a clear, viscous, yellow liquid. Nucleation with the α-form of tetra-acetyl D-galactose anilide resulted in crystallisation. Separation of the crystals by trituration with methyl alcohol followed by recrystallisation from methyl alcohol yielded tetra-acetyl α-D-galactose anilide (44 mg.), m. p. and mixed m. p. 175—176°.

Removal of the Anilide Group.—(*a*) *From* 2 : 3 : 4 : 6-*tetra-acetyl α*-D-*galactose anilide.* A suspension of the anilide (1 g.) was treated for 5 hours in boiling ethyl alcohol (10 c.c.) with 0·5% aqueous formic acid (40 c.c.) in an atmosphere of carbon dioxide. The anilide dissolved and a pale yellow solution resulted. The alcohol was removed by distillation under reduced pressure and the residual solution was extracted three times with chloroform. The combined extracts were washed successively with 0·2N-hydrochloric acid, 5% sodium hydrogen carbonate solution, and water, and dried ($MgSO_4$). Distillation of the solvent in a vacuum gave 2 : 3 : 4 : 6-tetra-acetyl β-D-galactose, which crystallised on treatment with absolute ethyl alcohol and light petroleum. After two crystallisations from acetone–ether–light petroleum, the product (0·35 g.) showed $[\alpha]_D^{24}$ +26° (*c*, 1·0 in chloroform), m. p. and mixed m. p. 125—126°.

(*b*) *From* 2 : 3 : 4 : 6-*tetra-acetyl β*-D-*galactose anilide.* Treatment of a suspension of this anilide (1·0 g.) in acetone (10 c.c.) and 0·5% formic acid (40 c.c.) as in (*a*) gave tetra-acetyl β-D-galactose (0·3 g.), m. p. and mixed m. p. 127—128°, $[\alpha]_D^{20}$ +23° (*c*, 1·0 in chloroform).

When a solution of tetra-acetyl α-D-galactose anilide (0·2 g.) in 2% methyl-alcoholic hydrogen chloride (20 c.c.) was kept at room temperature it became yellow and reached a constant specific rotation after 5 minutes : $[\alpha]_D$ +247° (initial value) ; +36° (3 minutes) ; +35° (5 minutes) ; +35° (10 minutes) ; +35° (13 minutes) ; +32° (40 minutes). After neutralisation with silver carbonate and removal of the solvent, a yellow glass (0·17 g.) was obtained which did not crystallise when nucleated with either tetra-acetyl α- or β-methylgalactopyranoside.

In another experiment, a solution of tetra-acetyl α-galactose anilide (0·2 g.) in dry chloroform (20 c.c.) was saturated with hydrogen chloride at 0°, and kept at room temperature until its rotation was constant, [α]$_D$ +185° (initial value); +36° (40 minutes). It was washed with 0·1N-hydrochloric acid, sodium hydrogen carbonate solution, and water, and dried ($MgSO_4$). The brown syrup which remained after removal of the chloroform under reduced pressure gave crystals of the original material (0·1 g.) when treated with ethyl alcohol–light petroleum.

When a solution of tetra-acetyl β-D-galactose anilide (0·5 g.) in dry chloroform (50 c.c.) was treated as described above for the α-compound it showed: [α]$_D$ −32° (initial value); +23° (16 minutes); +42° (125 minutes) (constant). A small quantity of tetra-acetyl α-D-galactose anilide was isolated from the reaction. Concentration of the ethyl-alcoholic mother-liquors left a dark brown syrup which did not crystallise when nucleated with tetra-acetyl β-D-galactose.

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