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The Oxidation of Glycosides

VI. Oxidation of Methyl 4,6-*O*-Ethylidene-β-D-glucopyranoside with Chromium Trioxide

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Methyl 4,6-*O*-ethylidene-β-D-glucopyranoside was oxidised with chromium trioxide in acetone and, after removal of the ethylidene groups, oxo-glucosides were isolated; the principal one being the 3-oxo-glucoside. Fractionation on a charcoal column yielded, *inter alia*, methyl 2-oxo-glucoside admixed with methyl glucoside. Those components were separated by chromatography on a carbon column using an irrigant containing sodium hydrogen sulphite. Small amounts of D-arabinose, D-erythrose and D-erythrono-γ-lactone were also isolated. The oxidation of methyl β-D-glucopyranoside was studied in an analogous experiment.

Amongst the products formed on the oxidation of the anomers of methyl 6-*O*-trityl-glucopyranosides with chromium trioxide are the corresponding 2-oxo-, 3-oxo- and 4-oxo-derivatives (Part V[1]). The present investigation was undertaken to simplify the preparation of methyl β-D-2-oxo-glucopyranoside and of methyl β-D-3-oxo-glucopyranoside. The hydroxyl group at C-4 and that at C-6 were protected by an ethylidene group prior to the oxidation. After removal of the ethylidene groups the mixtures of neutral oxidation products were fractionated on carbon columns and subfractionated by chromatography and by electrophoresis on thick filter papers. When 1.5 moles of oxidant per mole of sugar were used, the yield of 3-oxo-glucoside (5.4 %) was only slightly higher than that obtained (4.7 %) under similar conditions from methyl 6-*O*-trityl-β-D-glucopyranoside. The ethylidene derivative is prepared more easily than is the trityl derivative and the former gives a less complex mixture of products (in the present work no 4-oxo- or 6-oxo-glucosides were detected). The present procedure is then to be preferred to that used earlier. The yield of oxo-glucosides was not increased when a higher proportion of oxidant to sugar was used.

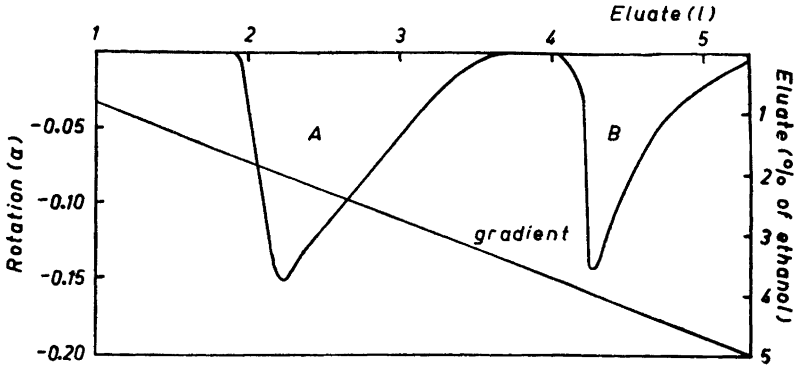

Fig. 1. Separation of methyl β-D-2-oxo-glucopyranoside (A) from methyl β-D-glucopyranoside on a carbon-Celite column using an irrigant containing sodium hydrogen sulphite.

The 2-oxo-glucoside was eluted from the carbon column together with the methyl glucoside. These two substances are easily separated by paper electrophoresis in sulphite buffer [3]. Barker *et al.*[4] used an irrigant containing borate for the carbon column chromatography of some sugars which were difficult to separate by ordinary carbon column chromatography, but which had different electrophoretic mobilities in borate buffer. In the present work the analogous use of sodium hydrogen sulphite in the irrigant resulted in the separation of methyl glucoside and methyl 2-oxo-glucoside (1.2 %) (see Fig. 1).

Many other components were chromatographically indicated to be present and the principal ones were isolated. They included D-glucose (principally from the hydrolysis of part of the methyl glucoside), D-arabinose (0.05 %), D-erythrose (0.13 %) and D-erythrono-γ-lactone (0.20 %) and a trace of a compound chromatographically indistinguishable from ribulose. Small amounts of a neutral and reducing compound (A) were isolated, which gave colour reactions with *p*-anisidine hydrogen chloride and with resorcinol-hydrochloric acid. It had a high electrophoretic mobility in hydrogen sulphite and on reduction with borohydride it gave mainly erythritol.

Another compound (B) giving a characteristic red-violet coloration with resorcinol was isolated. Some properties are given in the experimental part. A compound having similar properties was formed when methyl β-glucoside or its 6-*O*-trityl ether [1,2] was oxidised.

The arabinose and erythrose may have been formed *via* oxoaldonic acids. Whistler *et al.* recently showed that D-erythrono-γ-lactone was formed in relatively large amount when starch was oxidised with hypochlorite [5,6].

Methyl-β-glucoside on oxidation by dichromate in the presence of oxalic acid, yielded 6-oxo- and 3-oxo-glucosides as the principal neutral products [2]. In the present work methyl-β-glucopyranoside was oxidised with trioxide using conditions similar to those above and the following substances were isolated: arabinose (0.1 %), glucose (0.25 %) and the 2-oxo- (0.5 %), 3-oxo- (3.3 %), 4-oxo- (0.3 %) and 6-oxo- (1.3 %) derivatives of methyl glucoside.

Those results were unlike those obtained when an aqueous solution of chromate and oxalic acid was used as the oxidant [2]. In that case there was obtained a larger amount of aldehydo-glucoside than of keto-glucosides. The final yields will of course depend not only on the rate of formation of oxo-groups, but also upon any secondary reactions. The results obtained in the present and in an earlier investigation [1] showed that the 3-oxo-glucoside is the predominating neutral product obtained on oxidation with chromium trioxide. It is unlikely that under the oxidation conditions used one oxo-compound could be transformed into another by enolisation. No chromatographically detectable formation of 3-oxo-glucoside from 2-oxo-glucoside took place during the treatment of the oxidation products prior to fractionation. Recrystallisation of 2-oxo-glucoside from hot *n*-propanol, however, resulted in the formation of 3-oxo-glucoside and of unidentified compounds.

A small amount of methyl glucoside and, in a separate experiment, of methyl 4,6-ethylidene glucoside were oxidised with *t*.-butyl chromate in a mixture of acetone and *t*.-butanol. The yields of oxo-glucosides were lower, and that of the acids higher, than when chromium trioxide in acetone was used as oxidant.

EXPERIMENTAL

Melting points are corrected. All distillations were carried out under reduced pressure (bath temperature $<40°$) or by lyophilisation. Papers and spray-reagents used in the paper chromatography and electrophoresis were as given in Part. V[1].

Irrigants and buffers used:

A. Ethyl acetate-acetic acid-water, 3:1:3 (the upper phase).
B. Butanol-pyridine-water, 3:1:1.5.
C. Hydrogen sulphite buffer pH 4.7, 0.1 M[3].
D. Borate buffer pH 10.0, 0.1 M.

Oxidation of methyl 4,6-*O*-ethylidene-β-D-glucopyranoside with chromium trioxide and fractionation of the product

The methyl 4,6-*O*-ethylidene-β-D-glucopyranoside was made by a slight modification of the method of Helferich and Appel [7].

The oxidation and fractionation procedures were essentially as described in Part V[1] and accordingly only an abbreviated description is given. The percentage yields quoted are based upon the original weight of the starting-material calculated as methyl-β-glucoside.

Methyl 4,6-ethylidene-glucoside (80.0 g) was oxidised by boiling it under reflux, for 30 min, with acetone (4 l) and chromium trioxide (55.0 g). The brown chromium oxides were filtered off from the oxidation mixture and boiled with successive volumes of acetone with intermediate filtrations. During the evaporation of the combined filtrate, crystals of pure starting-material (31.0 g, m. p. 187—188°) separated and were filtered off. To the filtrate there were added ethanol (100 ml) and dilute sulphuric acid to give an 0.3 N solution (600 ml). The solution was kept at 100° for 30 min and then extracted with chloroform. The acidity was reduced to pH 5 by the addition of barium carbonate, the mixture being stirred and the temperature kept below 10°. After filtration, the solution was deionised and the solvent distilled off. The product, a light-yellow syrup (20.2 g), was fractionated on a carbon-Celite column (48 × 6 cm) using the linear-gradient elution technique (ethanol: 0—15 %, 10 l and 50 % 4 l). The following main fractions were obtained:

Fraction I (1.8 g) contained glucose and smaller amounts of other components. The material was sub-fractionated by paper chromatography and electrophoresis on thick filter papers, and the following components were obtained: D-erythrono-γ-lactone (0.20 %), D-arabinose (0.05 %), D-ribulose (<0.02 %), D-erythrose (0.13 %) and an unidentified compound A (0.1 %). The first three were chromatographically and electrophoretically indistinguishable from reference substances. The compound A was neutral and was rapidly oxidised by the alkaline silver reagent and also gave a yellow coloration with *p*-anisidine hydrogen chloride and a faint pink coloration with resorcinol-hydrochloric acid. Compound A had: $R_{Glucose}$ = 2.03 (irrigant A), $R_{Glucose}$ = 1.28 (irrigantB), M_V [3] (buffer C at 50°) = 1.70 and M_G (buffer D) = 0.66. On reduction with borohydride it gave erythritol which had m. p. and mixed m. p. 119—120°.

D-*Erythrose*. The amorphous sugar had $[\alpha]_D^{22}$ −14° (*c*, 2.2 in water), $R_{Glucose}$ = 2.35 (irrigant A), M_V = 0.18 (Buffer C at 50°) and M_G = 0.80 (buffer D). A sample of the sugar was reduced with borohydride. After deionisation and recrystallisation of the product from ethanol it had m. p. 120—120.5°, not depressed on admixture with erythritol. It was paper chromatographically and electrophoretically indistinguishable from erythritol. It was acetylated in the usual way and the product, after recrystallisation from ethanol, had m. p. 84—85°, not depressed on admixture with erythritol tetra-acetate.

D-*Arabinose*. The specific rotation of the amorphous sugar was $[\alpha]_D^{22}$ −100 (*c*, 0.8 in water).

D-*Erythrono-γ-lactone*. It was recrystallised from ethyl acetate and had m. p. 104—105° (*Lit.*[5] 104—105°) and $[\alpha]_D^{21}$ −74° (*c*, ? in water), *Lit.*[5] −72). The equivalent weight by titration was 114 (calc. 116).

Fraction II (11.5 g) consisted mainly of methyl β-glucoside and methyl 2-oxo-glucoside. After recrystallisation from aqueous ethanol, pure methyl glucoside (8.7 g) was obtained. The components in the mother liquor were fractionated on a carbon-Celite column (40 × 3 cm) using aqueous ethanol (0—10 %, 4 l and 50 %, 2 l) to remove both traces of glucose and of products which had been formed in small amounts from the labile 2-oxo-glucoside during the crystallisation of the methyl glucoside. A mixture (2.15 g) of methyl 2-oxo-glucoside and methyl glucoside was eluted with *ca.* 4 % aqueous ethanol. It was refractionated on the same column which had been thoroughly washed with 0.1 M sodium hydrogen sulphite. The column was irrigated with 0—5 % ethanol (5 l) using 0.1 M aqueous sodium hydrogen sulphite solution instead of water. The fractionation was followed polarimetrically (2 dm tube, Fig. 1) and by electrophoretic examination using buffer D. The methyl 2-oxo-glucoside was completely separated from the methyl glucoside and, after deionisation and evaporation of the solutions, the two components (0.85 g and 0.96 g, respectively) were obtained chromatographically and electrophoretically pure. Part of the 2-oxo-glucoside crystallised (for data *cf.* part V[1]). Paper chromatographic examination of the mother liquor showed that some rearrangement had taken place during the crystallisation.

Fraction III (4.0 g) consisted mainly of 3-oxo-glucoside which crystallised on removal of the solvent. It was recrystallised from *n*-propanol and had m. p. 130—131°, $[\alpha]_D^3$ −63 (*c*, 2 in water), The yield of this material plus that recovered from the mother liquors was 5.3 %.

Fraction IV (1.6 g) contained an unidentified neutral compound (B), isolated in amorphous state (0.2 %). It gave a distinctive red-violet coloration with resorcinol-hydrochloric acid, a yellow coloration with *p*-anisidine hydrogen chloride and was acid sensitive but alkali-stable. It had $R_{Glucose}$ = 2.98 (irrigant A) and 1.48 (irrigant B), M_V = 0 (Buffer C) and M_G = 0.42 (buffer D). It had OCH_3 *ca.* 14 % and $[\alpha]_D^{21}$ −29° (*c*, 1.4 in water).

The fraction also contained other components most of which could be chromatographically correlated to substances obtained in the previous fractions, while others were probably condensation products formed from the oxo-glucosides.

Oxidation of methyl β-D-glucopyranoside with chromium trioxide and fractionation of the product

The oxidation and fractionation procedures were essentially as described previously, but the reduction of the chromium compounds in aqueous solution by the ethanol was carried out at room temperature and after slight acidification with sulphuric acid. Methyl β-glucoside (20.2 g) was oxidised with chromium trioxide (15.6 g) in acetone (1.20 l) and the product was fractionated on a carbon-Celite column. After sub-fractionation by chromatography and electrophoresis on thick filter paper the following substances were obtained: arabinose (0.1 %), glucose (0.25 %) and the 2-oxo- (0.5 %), 3-oxo- (3.3 %), 4-oxo- (0.3 %) and 6-oxo- (1.3 %) derivatives of methyl β-glucoside. The substance B (see earlier) was obtained in 0.15 % yield.

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REFERENCES

1. Theander, O. *Acta Chem. Scand.* **11** (1957) 1557.
2. Lindberg, B. and Theander, O. *Acta Chem. Scand.* **8** (1954) 1870.
3. Theander, O. *Acta Chem. Scand.* **11** (1957) 717.
4. Barker, S. A., Bourne, E. J. and Theander, O. *J. Chem. Soc.* **1955** 4276.
5. Whistler, R. L., Linke, E. G. and Kazeniac, S. *J. Am. Chem. Soc.* **78** (1956) 4704.
6. Whistler, R. L. and Schweiger, R. *J. Am. Chem. Soc.* **79** (1957) 6460.
7. Helferich, B. and Appel, H. *Chem. wer.* **64** (1931) 1841.

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