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Note

Structure of the carbohydrate chains of panaxosides B^I and C from *Panax ginseng* C. A. Meyer

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Gas–liquid chromatography (g.l.c.) has been widely applied in the quantitative determination of carbohydrates in biological materials. The Sweeley method[1] for obtaining volatile derivatives of monosaccharides has been used to analyse the sugar composition of natural glycosides, including derivatives of flavones[2], triterpenes[3], and steroids[4].

Trimethylsilyl (TMS) ethers and alditol acetates are usually preferred to methyl ethers for the quantitative determination of the monosaccharide composition of oligo- and poly-saccharides. However, g.l.c. of methyl ethers is of great value in the study of the structure of oligo- and poly-saccharides by methylation methods.

In the present study, these methods were used to study the carbohydrate portion of panaxosides B^I and C, which are glycosides isolated from the methanol extract of ginseng roots[5].

EXPERIMENTAL

G.l.c. conditions. — The gas chromatograph used was Model "Zvjet-2" (Dzerjinsk, U.S.S.R.) equipped with a differential recorder and a dual-flame detector system. A stainless-steel column (100 × 0.3 cm) filled with 10% SE-30 on Chromosorb W (60–80 mesh) was used for TMS ethers of reduced monosaccharides. Methylated monosaccharides methyl glycosides were determined on a similar column filled with 5% neopentylglycol succinate on Chromosorb W (60–80 mesh) deactivated with hexamethyldisilazane (HMS).

Monosaccharide TMS ethers were analysed with a temperature gradient of 155–200° at a heating rate of 2°/min and an evaporator temperature of 290°. Methyl glycosides of methylated monosaccharides were determined with a temperature gradient of 110–190° at a heating rate of 10°/min and an evaporator temperature of 270°. The gas flow for each analysis was nitrogen, 33 ml/min; hydrogen, 33 ml/min; and air, 270 ml/min.

Preparation of TMS ethers. — A mixture of monosaccharides obtained after complete hydrolysis of 15 mg of panaxoside B^I (C) was dissolved in water (1 ml), sodium borohydride (3 mg) was added, and the mixture was maintained at room

temperature for 1 h. The solution was neutralised with Amberlite IR-120 (H^+) resin and evaporated *in vacuo*, and methanol was distilled from the residue to remove boric acid. The dry residue was treated with HMS and chlorotrimethylsilane in pyridine[1]. The mixture was filtered and evaporated, and the residue was subjected to g.l.c.

myo-Inositol was selected[7,8] as the internal standard. Linear calibration curves were obtained in the usual manner for each sugar by g.l.c. of various amounts of sugar with a constant amount of the internal standard.

Preparation of methyl glycosides. — The methylated panaxoside B^I (Ref. 9 and 10) or panaxoside C (Ref. 10 and 11) (15 mg) was heated with methanolic hydrogen chloride (3%, 1 ml) for 10 jh, and the products were analysed by g.jl.c.

Reference mixtures of 2,3,4-tri-*O*-methyl-L-rhamnose, and 2,3,4,6-tetra-*O*- and 3,4,6-tri-*O*-methyl-D-glucose (1:1:1 and 1:2:1) were treated as described above.

RESULTS AND DISCUSSION

The carbohydrate portion of panaxoside B^I was shown to contain only glucose by g.l.c. of the TMS ethers of the hydrolysed and reduced glycoside. Methylation of panaxoside B^I by successive application of the Hakomori[9] and Purdie[11] methods, followed by methanolysis, gave, in the carbohydrate fraction, derivatives of 2,3,4,6-tetra- and 3,4,6-tri-*O*-methylglucose, in the ratio 2:1. This result is in agreement with the elemental, analytical data for panaxoside B^I and indicates that two carbohydrate residues are attached to the aglycon; one of these residues is glucose and the other is 2-*O*-glucopyranosyl-glucose.

The reduced monosaccharides from panaxoside C were rhamnitol and glucitol in the ratio of 1:2 (g.l.c. of TMS ethers). Together with the elemental, analytical data, panaxoside C is thereby shown to contain three sugar residues.

G.l.c. of the products obtained after methylation (Purdie[11] and Kuhn[10] methods) and methanolysis of panaxoside C revealed methyl glycosides of 2,3,4-tri-*O*-methyl-rhamnose, 2,3,4,6-tetra-*O*-methyl-glucose (α and β isomers), and 3,4,6-tri-*O*-methyl-glucose (α and β isomers) in equimolar amounts.

Thus, two carbohydrate chains are attached to the aglycon in panaxoside C, and the above data are consistent with the alternative arrangements (1) rhamnopyranose and 2-*O*-glucopyranosyl-glucose, or (2) glucopyranose and 2-*O*-rhamnopyranosyl-glucose.

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