

EXAMINATION OF THE CONSTITUENTS OF JUTE SEEDS

Part II.—Identification and Characterization of a Glycoside and Aglycone from the Seeds

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A glucoside of β sitosterol has been isolated from jute seeds. The constitution of the glucoside has been decided by its reactions and ultimate synthesis from β -sitosterol and aceto-bromoglucose.

In Part I of this series of papers,¹ a description of the main constituents of jute seeds was recorded. Besides these, a number of other new products have been isolated from these seeds and in this respect both *Corchorus capsularis* and *Corchorus olitorius* appear to behave similarly. A glucoside, corchoraside A, has been described by Reichstein² but in the present work several glucosides have been obtained. Of these glucosides, the two bitter ones bear some resemblance with Reichstein's glucoside but appear to be different from the one described by him and they will be dealt with in a subsequent communication. Besides these, a sterol glucoside has also been isolated from the seeds, which forms the subject matter of this paper. This glucoside has been synthesised and shown to be identical with β -sitosterol-*d*-glucoside.

Sterol-glucosides have been found to occur widely distributed in the different parts of various plants. Of these, special reference may be made to the glucosides from soyabean oil,³ cotton seed oil,⁴ *Impomoea purpurea* Roth.⁵ and *Hevea* rubber.⁶ The 'jute seed oil' on being saponified, yielded a nonsaponifiable fraction (ca 5.7%). This appeared to be a mixture of sterol and some other products. The sterol is identical with β -sitosterol.³ Most probably this is present in the seed in the form of a glucoside or in some other form which has not yet been identified. The defatted seed powder, when extracted with rectified spirit, among other products, yielded a glucoside in a very characteristic crystalline form. Its m.p., optical rotation, and the m.p. and optical rotation of its acetate, together with its elemental analyses appeared to be identical with those recorded for β -sitosterol-*d*-glucoside. In infrared, it showed strong O-H, C=C, C-CH₃, and C-O- absorptions near 3390; 1639; 1458; 1379; 1073 and 1021 cm.⁻¹, respectively, but its acetate had no O-H absorption.

The glucoside was hydrolysed and the sugar moiety was readily identified by paper chromatography as *d*-glucose and calculated as one mole

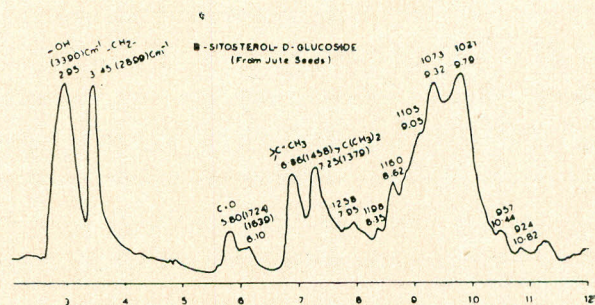


Fig. 1.—I.R. Absorption spectra of β -sitosterol-*d*-glucoside from jute seed.

for each molecule of the glucoside. The aglycone thus obtained readily crystallised and responded to the sterol colour reactions. From the melting point, analysis as well as the optical rotation, it was found to be β -sitosterol and its infrared absorption spectrum was superimposable upon that of an authentic β -sitosterol. Final confirmation was secured by the synthesis of β -sitosterol-*d*-glucoside from β -sitosterol and acetobromoglucose by a modification of the method of Koenigs and Knorr,⁷ possessing the same Rf value. Its mixed m.p. with the isolated compounds was undepressed and an infrared spectrum obtained for it was superimposable.

Experimental

Isolation of β -Sitosterol-d-Glucoside.—Finely powdered seeds of *Corchorus capsularis* L. in batches of 10 kg. were extracted with light petroleum ether (b.p. 60-80°) in the cold and the defatted seeds thus obtained were then extracted with ethanol (95%) as already described in Part I of this series. The total alcoholic extract after concentration to half the volume was refrigerated (-5°C.) when a voluminous white flocculent precipitate was thrown down. This was collected the next morning by filtration and was washed with a little light petroleum ether.

The separated solid (2.6 g., 0.026%) darkened at 220-225° and melted at 273-280°. It crystallised from dilute ethanol into flakes, which darkened at 270-275° and melted at 294-296° (decomp.). It had $[\alpha]^{31}_D - 45.8^\circ$ (*c* 6.1 in pyridine), (Found: C, 73.3; H, 10.7. Calculated for $C_{35}H_{60}O_6$: C, 72.8; H, 10.5%), Mol. wt. found 542.3; required for $C_{35}H_{60}O_6$, 576.8, γ max. (in KCl) 3390 (OH), 1639 (C=C), 1458, 1379 (C-CH₃), 1073, 1021 (C-O) cm^{-1} . It gave a strong Liebermann-Burchard reaction for sterol and the Whitby reaction⁸ for sterol glycoside.

Acetate of β -Sitosterol-d-Glucoside.—A sample (0.4 g.; m.p. 294-296°, decomp.) was refluxed for 1 hr. at 150°C. with pyridine (2 ml.) and acetic anhydride (10 ml.), the mixture was then distilled under reduced pressure, and the dry residue crystallised from ether-ethanol (charcoal) as long needles (0.51 g.), m.p. 170°. It had $[\alpha]^{30}_D - 36.8^\circ$ (*c* 0.8 in pyridine). Found: C, 68.6 per cent; H, 9.1 per cent. Calculated for $C_{43}H_{68}O_{10}$: C, 69.3 per cent; H, 9.2 per cent ν max. No hydroxyl absorption was found, but otherwise the absorption band was identical with the above, except that it has a new strong CO absorption.

Hydrolysis of β -Sitosterol-d-Glucoside.—The glucoside (0.4732 g., m.p. 294.-296°, decomp.) dissolved in alcohol (200 cc.), was heated under reflux with concentrated hydrochloric acid (2 ml.) for 12 hrs. This was concentrated under reduced pressure to 50 ml. and water (150 ml.) was added. The precipitate that formed was washed free of acid and dried to constant weight (0.27 g.), m.p. 135-136°. It crystallised from methanol and melted at 137-138° and had $[\alpha]^{32}_D - 30.93^\circ$ (*c* 0.63 in CHCl_3). (Found: C, 83.6 per cent; H, 12.2 per cent. Calculated for $C_{29}H_{50}O_5$: C, 84.0 per cent; H, 12.1 per cent) and gave a positive Liebermann-Burchard reaction and the same Rf value with that of an authentic sample of β -sitosterol.

The aqueous acid filtrate was then refluxed for 6 hours in order to hydrolyse the ethylglucoside, neutralised with silver carbonate, and filtered. Analysis showed the presence of 0.14 g. (0.95 mole) of sugar calculated as glucose. The sugar, on a paper chromatogram was found to be identical with d-glucose.

Acetylation of the Sterol.—The above isolated sterol (0.2 g.) was refluxed with acetic anhydride (12 ml.) for 2 hours, cooled, and the crystals obtained (0.15 g.) were recrystallised from methanol into lustrous plates, m.p. 132° and had $[\alpha]^{30}_D - 37^\circ$ (*c* 0.23 in CHCl_3) (Found: C, 81.7; H, 11.4. Calculated for $C_{13}H_{22}O_2$: C, 81.5; H, 11.5 per cent).

β -Sitosterol from Jute Seed Oil.—'Jute seed oil' (100 g.) was saponified according to Soliman *et al.*⁹ to give a crude sterol mixture (1.5 g.) which, when crystallised from dilute methanol, produced a crystalline mass (fraction 1, 0.3 g.) m.p. 110-120°, and the remaining mother liquor on concentration yielded crystals (fraction 1, 1.2 g.) m.p. 128-129°. The latter on repeated crystallisation from petroleum ether (charcoal) melted at 136-137° and had $[\alpha]^{28}_D - 32.3^\circ$ (*c* 0.42 in CHCl_3), mixed m.p. with β -sitosterol obtained from the glucoside was undepressed and infrared spectrum superimposable upon that of the same sterol.

Synthesis of β -Sitosterol-d-Glucoside.—For the synthesis, 2:3:4:6-tetraacetyl α -glucosyl bromide (acetobromoglucose) was prepared according to the method of Fischer¹⁰ by allowing β -pentaacetyl glucose (150 g.) to react with dry hydrogen bromide¹¹ in acetic acid at 0°C. to give 115 g. (76%) of a crystalline paste; crystallised from dry ether-petroleum ether mixture when it melted at 88-89°, and had $[\alpha]^{30}_D + 195.2^\circ$ (*c* 4.4 in CHCl_3) (given for acetobromoglucose, m.p. 88-89°, $[\alpha]^{19}_D + 198^\circ$ in chloroform).

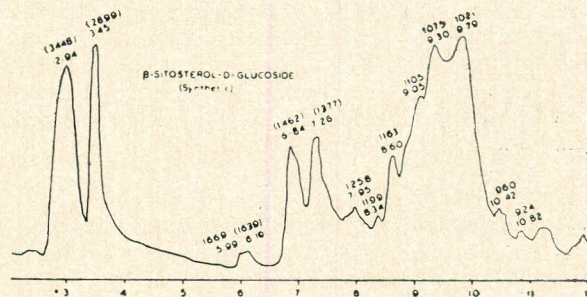


Fig. 2.—I.R. Absorption spectra of β -sitosterol-*d*-glucoside synthetic.

This was next employed as described by Salway¹² for the synthesis. β -sitosterol (2 g., m.p. 137°) was allowed to react with acetobromoglucose (3 g., m.p. 88°) dissolved in dry ether (100 ml.) for 8 hours in the presence of dry silver oxide (3 g.). The product was worked up in the usual way when a crystalline solid (0.56 g., 20%), separated out. This was crystallised from pyridine-ethanol mixture and showed a shrinkage at 270-275°, but melted at m.p. 298°. It had $[\alpha]^{28}_D - 48.6^\circ$ (*c* 0.81 in pyridine) and had the same Rf value. The mixed m.p. with the natural glucoside remained undepressed and the infrared spectrum was superimposable upon the β -sitosterol-*d*-glucoside obtained from jute seeds.

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