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Note

The mucilage of *Opuntia ficus-indica* Mill.

EL S. AMIN, OLFAT M. AWAD, AND M. M. EL-SAYED
Department of Chemistry, Alexandria University, Alexandria, Egypt (U. A. R.)
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The Indian fig (*Opuntia ficus-indica* Mill.) is a plant found in many regions of the world. In Egypt, its fruits are widely used as human food. The aim of this work was to study the mucilage which was found in Opuntia leaves.

The mucilage was isolated by extraction of the leaves with 0.1M hydrochloric acid, and precipitation with acetone. It is a creamy white, amorphous substance, having $[\alpha]_D^{23}$ − 170° (*c* 0.05, water), that is very difficultly soluble in water but is more soluble in aqueous alkali; its copper complex is soluble in water. Hydrolysis with acid, followed by paper chromatography of the hydrolyzate indicated the presence of 37.5% of an arabinose, 35.7% of a galactose, 15.5% of a xylose, and 11.5% of a rhamnose, and no uronic acids. Complete methylation of the mucilage with dimethyl sulfate and sodium hydroxide, and then with methyl iodide and silver oxide in *N*,*N*-dimethylformamide, gave a product having $[\alpha]_D^{16}$ −133° (*c* 0.1, chloroform) which, on hydrolysis and paper chromatography, showed two major components, namely, a 2,3,5-tri-*O*-methylarabinose (indicating that arabinose residues are present in the mucilage as a terminal group) and a 2,3,6-tri-*O*-methylgalactose [indicating (1→4)-linkage], and two minor components which were found to be a 2,4-di-*O*-methylgalactose (indicating a branched structure) and a 3,4-di-*O*-methylrhamnose [indicating (1→2) linkage]. The results of assay of end groups by periodate oxidation showed that the mucilage molecule is composed of ~55 sugar residues.

EXPERIMENTAL

Isolation of the mucilage. — The Opuntia leaves, harvested in November, 1966, at Alexandria, Egypt, were cut into thin slices and minced in 0.1M hydrochloric acid (2 ml/g) for 24 h. The aqueous extract was passed through a sieve (mesh 2 mm), and centrifuged at room temperature, and the clear, supernatant liquor was poured into acetone (3 ml/ml) to precipitate the mucilage. Purification[1] was effected by dissolving the precipitate in M sodium hydroxide (500 ml), acidifying the solution with acetic acid, and precipitating the mucilage with acetone (3 ml/ml). It was a creamy-white, amorphous substance, difficultly soluble in water, but more soluble in aqueous alkali. (Found: ash, 0.06; N, 0.08%). Attempted fractionation by use of 25% aqueous cupric chloride gave no precipitate.

Acid hydrolysis of the mucilage. — A portion (0.5 g) of the mucilage was heated in a sealed tube with 0.5M sulfuric acid (30 ml) for 12 h at 100°. The resulting mixture was filtered, to remove a small amount of flocculent material, and the filtrate was rendered neutral with 0.05M barium hydroxide. Barium sulfate was removed, and the solution was evaporated to dryness. The residue was extracted several times with boiling ethanol, and the ethanol extracts were combined, de-ionized with a mixture of Amberlite IR-120 (H^+) and IRA-400 (OH^-) ion-exchange resins, and evaporated to a yellow syrup. After the extraction, the brown residue was tested for uronic acids with the benzidine reagent or the Lefèvre–Tollens test[2]; it was found that the residue contained no uronic acids, and that it was a barium salt.

Chromatography and estimation of sugar in the mucilage. — A portion of the acid hydrolyzate was examined on a paper chromatogram by use of 5:4:1 butyl alcohol–water–ethanol, and *p*-anisidine hydrochloride as the spray agent; the major components were an arabinose and a galactose, and a xylose and a rhamnose were minor components. No uronic acid could be detected with the benzidine reagent or with the Lefèvre–Tollens test[2]. After separation of the sugars on the chromatogram, quantitative estimation by the method of Mayer, Noelting, and Bernfeld[3] indicated 37.5% of an arabinose, 35.7% of a galactose, 15.5% of a xylose, and 11.5% of a rhamnose. Another portion of the syrup was separated on a paper chromatogram, and the individual components were extracted therefrom with methanol. The first spot was converted into galactaric acid (m.p., and mixed m.p., 210°); the second spot was converted into arabinose benzoylhydrazone (m.p., and mixed m.p., 200°); and the third spot was converted into xylose benzoylhydrazone (m.p., and mixed m.p., 167°).

Acetylation of the mucilage[4]. — The dry, powdered mucilage (1.0 g) was heated with pyridine (40 ml) for 2 h at 70°, and the mixture was cooled, and kept overnight at room temperature. Acetic anhydride (20 ml) was then added dropwise during 30 min, and the mixture was kept for 3 days at 52°. The resulting, clear solution was diluted with acetic acid (40 ml) and poured, with stirring, into ethanol. The precipitate was dried, and the product was re-acetylated, giving 0.5 g of a white material, $[\alpha]_D^{20}$ −100° (*c* 0.1, chloroform).

Methylation of the mucilage. — The powdered mucilage (2 g) was suspended in water (30 ml) and allowed to swell for 5 h at 5°. It was then methylated[5] eight times with dimethyl sulfate and sodium hydroxide solution under an atmosphere of nitrogen, the mixture was dialyzed, and the product was methylated twice with methyl iodide and silver oxide in *N*,*N*-dimethylformamide. The product (isolated by extraction with chloroform) was a brown, brittle substance (yield 0.92 g). It had $[\alpha]_D^{18}$ −133° (*c* 0.1, chloroform).

Methanolysis and hydrolysis of the methylated mucilage. — The methylation product (0.5 g) was heated for 12 h at 100° with methanolic hydrogen chloride (3%), and the mixture was made neutral with cold, ethereal diazomethane[6]. The solution was concentrated at room temperature, and a solution of the product in 4% hydrochloric acid was boiled for 7 h under reflux. The acid was neutralized with silver

carbonate, and the filtrate was de-ionized with a mixture of Amberlite IR-120 (H^+) and IRA-400 (OH^-) ion-exchange resins, and evaporated to a syrup (0.51 g).

Chromatography of the hydrolysis product of the methylated mucilage. — Examination of the hydrolysis product on a paper chromatogram, with the upper layer of 5:1:4 butanol–ethanol–water as the mobile phase and *p*-anisidine hydrochloride as the spray reagent, showed four components, corresponding to a 2,3,5-tri-*O*-methylarabinose (R_G 0.95), 2,3,6-tri-*O*-methylgalactose (R_G 0.71), 2,4-di-*O*-methylgalactose (R_G 0.41), and 3,4-di-*O*-methylrhamnose (R_G 0.84).

Periodate oxidation of the mucilage. — A series of samples (50 mg) of the mucilage were dissolved in M ammonia, and each solution was rendered neutral with dilute hydrochloric acid, made up to 5 ml with 3% aqueous sodium chloride, and oxidized with 0.37M sodium metaperiodate (2 ml) at room temperature. The excess of periodate was decomposed with ethylene glycol, and the formic acid liberated was titrated with 0.05M barium hydroxide. The results obtained (5-h intervals) indicated chains having ~55 sugar residues.

REFERENCES

1 EL S. AMIN, *J. Chem. Soc.*, (1956) 828.
2 K. U. LEFÈVRE AND B. TOLLENS, *Ber.*, 40 (1907) 4513.
3 K. H. MEYER, G. NOELTING, AND P. BERNFELD, *Helv. Chim. Acta*, 31 (1948) 103.
4 E. L. HIRST, J. K. N. JONES, AND A. J. ROUDIER, *J. Chem. Soc.*, (1948) 1779.
5 C. E. BALLOU AND E. G. V. PERCIVAL, *J. Chem. Soc.*, (1952) 1054.
6 L. HOUGH, J. K. N. JONES, AND W. H. WADMAN, *J. Chem. Soc.*, (1952) 3393.