NOTE

Analysis of Sugars from the Seeds of Tecoma urgentia

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Polysaccharides like galactose, mannose and reducing sugars have been analysed from the seeds of *Tecoma urgentia*. The seeds have high nutritional value.

Tecoma urgentia belongs to Bignoniaceae family. It is found at an altitude of 1200 m in the outer Himalayas. Bignoniaceae plant extract is used for the treatment of skin diseases, snakebites, malaria, diuretic, liver diseases, etc. Its wood is polishable and durable. The leaves have good nutritional value¹⁻³.

Analysis of Polysaccharides: The seeds of this plant were collected from Shidh Seeds Sales Corporation, Dehradun (U.P.) The powdered seed material (20 mg) of *Tecoma urgentia* was suspended in water (200 mL) for overnight and then heated at 100°C for 4 h. The swollen material was blended further with 300 mL of water. The viscous solution was filtered and centrifuged to remove suspended seed material. The polysaccharide was precipitated by adding large amount of ethanol to this solution. The coarse powder was redissolved in water and saturated Ba(OH)₂ solution was added, when the polysaccharide was precipitated as the barium complex. This was made into paste with water and decomposed by dropwise addition of 2 N acetic acid with vigorous stirring when it was partially dissolved. The insoluble fraction was separated by filtration.

The soluble fraction was recovered by precipitation with ethanol (300 mL). Both the acetic acid soluble fraction as well as the acetic acid insoluble fractions were then successively washed with 50%, 80% and 90% ethanol to remove the acid. The fractions thus obtained did not reduce Fehling's solution. The polysaccharides were obtained in the yield of 2.03% (insoluble) and 4.04% (soluble).

(a) Hydrolysis of Polysaccharide and Identification of the Constituents

The polysaccharide 0.1 g was hydrolysed with 1 N H_2SO_4 (15 mL) at 100°C for 17 h. The resulting solution was neutralized with barium carbonate, filtered and concentrated to a thin syrupy mass. This syrupy solution was subjected to descending chromatography⁴ along with various authentic sugar samples on Whatman No. 1 paper using the solvent system n-butanol: acetic acid: water (4:1:5, upper layer). The chromatogram was sprayed with aniline hydrogen phthalate. Various constituents present in the polysaccharide are reported in Table-1.

TABLE-1

S. No.	Name of sugar	R _f value known	R _f value observed	Optical density	Molarity from graph	Calculated molarity of the sugar
1.	Acetic acid Insoluble fraction					
	(a) Galactose	0.16	0.16	2.52	3.78	72.50
	(b) Mannose	0.20	0.19	0.92	1.43	27.44
2.	Acetic Acid Soluble fraction	-				
	(a) Galactose (b) Mannose	0.16 0.20	0.16 0.20	2.55 0.94	3.83 1.44	72.67 27.32

(b) Quantitative Estimation of Sugars of Polysaccharide

Quantitative estimation was done by Harborne's method. A measured volume of polysaccharide hydrolysate was placed along the starting line of chromatogram and the paper developed with the upper layer of n-butanol: acetic acid: water (4:1:5 v/v), sprayed with aniline hydrogen phthalate and dried. The coloured spots were cut out and each cut spot was eluted with 3 mL methanol containing 1% stannous chloride. These solutions were subjected to UV analysis and absorbances at max. wavelength were recorded on spectrophotometer. The same treatment was followed with glucose solutions of different molar concentrations of references. A graph was plotted between optical density and molarity of glucose solutions, when a straight line was obtained passing through the origin. From the graph concentrations of unknown sugars were calculated and the results are given in Table-1.

Analysis of Reducing Sugars: 10 g of seed powder was refluxed with small quantity of calcium carbonate and 100 mL of distilled water for 1 h. The aqueous extract was separated by decantation and the powder was further refluxed thrice with 50 mL of distilled water each time. The aqueous filtrates were combined and 10% solution of lead acetate was added till the precipitation was complete. It was filtered and H₂S gas was passed through the filtrate. It was again filtered and the filtrate was neutralised with ammonia. This neutral solution was concentrated on water bath till the volume became 100 mL.

Identification of Reducing Sugars: For identification of sugars spots of the concentrated test mixture and authentic sugars were applied on Whatman No. 1 paper and chromatograms were developed in n-butanol: acetic acid: water (4:1: 5, upper layer) solvent system. After developing, the chromatogram was sprayed with anisaldehyde sulphuric acid reagent. The identity of test sugars were confirmed by comparison of their R_f values with those of authentic sugars. The results are given in Table-2.

The amounts of reducing sugars were estimated quantitatively (as glucose) by Fehling's method using methylene blue as indicator. Thus the percentage of reducing sugars found to be present in the solution is 5:69% (as glucose).

Thus the polysaccharides present in the seeds of Tecoma urgentia are galactose

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and mannose in 5:2. The reducing sugars present in the seeds are 5.69% (as glucose) which contain D-arabinose, D-rhamnose, xylose, D-fructose, and maltose.

TABLE-2

S. No.	Name of reducing sugar	R _f value reported	R _f value observed
1.	D-arabinose	0.21	0.22
2.	L-rhamnose	0.37	0.37
3.	Xylose	0.28	0.28
4.	D-fructose	0.23	0.24
5.	Maltose	0.11	0.11

ACKNOWLEDGEMENT

Thanks to Prof. S.P. Banerjee, Head, Dept. of Chemistry, for providing laboratory facilities.

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(Received: 11 September 1999; Accepted: 14 December 1999) AJC-1956