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# **STUDIES ON CORDIA MYXA**

# Part I. The Monosaccharide and Polysaccharide Components of Fruits of Cordia Myxa

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Abstract. Monosaccharides and polysaccharides were extracted from the fruits of Cordia myxa in a stepwise manner.

The 95% ethanol extract afforded the monosaccharides, while stepwise extraction with cold water, hot water, ammonium oxalate and sodium hydroxide gave the polysaccharides. The monosaccharides were identified as glucose, fructose, xylose and an unidentified sugar was found to be present.

The polysaccharides from all the four extracts were found to have similar uronic acid anhydride (uaa) contents, and their specific rotations were measured. The major constituents of the polysaccharides were galacturonic acid, glucose, arabinose and xylose as a minor constituent.

Our interest in the Pakistani plants used in indigenous medicines led us to the systematic investigations of the chemical constituents of fruits of *Cordia myxa*. People in Indo-Pak subcontinent pickle the fruits of *Cordia myxa* (local name Sapistan). Medically the dried fruit is valued on account of its mucilagenous nature and demulcent properties, it is much used in coughs and chest affections, and also in irritations of the urinary passages. In larger quantities it is given in bilious infections as laxative.<sup>1</sup>

#### **Experimental**

Fresh fruits of *Cordia myxa* were extracted with ethanol to inactivate the enzymes and to remove the soluble sugars and proteins. The residue left was dried and extracted stepwise with cold water, hot water, ammonium oxalate and NaOH to extract the polysaccharides according to the method described by M. Uddin and his co-workers.<sup>2,3</sup>

Monosaccharides. In the present investigation fruits of Cordia myxa were extracted with 1:1 alcohol for 48 hr in a Soxhlet to extract the soluble monosaccharides, The alcoholic extract was then concentrated under reduced pressure.

The concentrated alcoholic extract (20 g) was then passed through Amberlite resin IR  $120(H^+)$  in order to separate the carbohydrates from amino acids if present.

The monosaccharides from the alcoholic extract were subjected to paper chromatography using the following solvent systems : (a) ethyl acetate-pyridine-water (10:4:3); (b) ethyl acetate-acetic acidformic acid-water (18:3:1:4); (c) butanol-ethanolwater (4:1:5) upper layer; and (d) butanol-acetic acid-water (4:1:5) upper layer.

The monosaccharides were identified as glucose, fructose, traces of xylose and an unidentified sugar, probably, sucrose. The identity of these constituent sugars was further confirmed by preparing their osazones and taking their m.p. and mixed m,p. with authentic sample. The analysis of the monosaccharides is given in Table 1.

TABLE 1. MONOSACCHARIDES PRESENT

IN THE FRUITS OF Cordia myxa

(WT. OF PLANT MATERIAL 1000 g).

Dried alcohlic extract taken for identifica- tion (g)	Constituent sugars present	Consti- tuent sugars obtained (g)	Consti- tuent sugars (%)	
20	Glucose	8.75	43.7	
	Fructose	0.74	3.7	
	Xylose	0.10	0.5	
	Unidentified sugar	0.32	1.6	

Extraction With Cold Water. The residue was treated with distilled water  $(2\frac{1}{2} 1)$  at room temperature overnight. The solution was stirred for homogenous mixing and centrifuged. The cold water soluble polysaccharides (0.60081 g) were precipitated with alcohol (1:1: v/v), centrifuged and dried by solvent exchange method.

Extraction with Hot Water. The residue was treated with hot water  $(2\frac{1}{6}1)$  at  $80^{\circ}$ , with stirring for 24 hr and then the solution was centrifuged. The hot water soluble polysaccharides (0.68948 g) were precipitated with alcohol (1:1 v/v), centrifuged and dried by solvent exchange method.

Extraction with Ammonium Oxalate. The residue left was treated with ammonium oxalate 0.5% $(2\frac{1}{2}$  l), heated at 100° with continuous stirring and centrifuged. The ammonium oxalate soluble polysaccharides (0.78166 g) were precipitated with alcohol (1:1: v/v), centrifuged and dried by solvent exchange method.

Extraction with Sodium Hydroxide. The residual material was finally extracted with NaOH  $(2\frac{1}{2})$ 

with continuous stirring and centrifuged. The NaOH soluble polysaccharides (2.77124 g) were precipitated with alcohol (1:1; v/v) centrifuged and dried by solvent exchange method.

The specific rotations of the aqueous solutions of the polysaccharides were measured at room temperature. Uronic acid anhydride (uaa) contents of the samples of polysaccharides were determined by Andersons decarboxylation method.<sup>4</sup> The results of these experiments are given in Table 2. The samples of the polysaccharides obtained by different extractions were examined for the constituent sugars.

The polysaccharide samples (5-10 mg) were hydrolysed in 1N  $H_2SO_4$  (2-5 ml) in seal tubes at 100° for 18 hr.<sup>5</sup>,<sup>6</sup> The hydrolysates were then neutralized with Ba(OH)<sub>2</sub> and BaCO<sub>3</sub>. Insoluble inorganic salts were removed by centrifugation. The supernatant was deionized with Amberlite resin IR-120(H+), concentrated and examined by paper partition chromatography using the solvent systems a, b, c, d and further were subjected to preparative paper chromatography. The constituent sugar of the different samples of polysaccharide obtained by stepwise extraction were identified as galacturonic acid, glucose, arabinose and xylose.

The identities of these constituent sugars were further confirmed by preparing their osazones and taking their m.p. and mixed m.p., with authentic samples.

The analysis of the polysaccharides is given in Table 2.

### TABLE 2. POLYSACCHARIDES PRESENT

# IN THE FRUIT OF Cordia myxa

# (WT. OF PLANT MATERIAL 1000 g).

Sample	Polysacc- haride obtained (g)	Uronic acid anhydrid	Speci rotati e	fic Constituent ion sugars
Polysaccharide extracted with cold water	0.60081	22	+266	Galacturonic acid Glucose+++ Arabinose+++ Xylose (tr)
Polysaccharide extracted with hot water	0.68948	22	+270	Galacturonic acid Glucose++ Arabinose+++ Xylose++
Polysaccharide extracted with ammonium oxalate	0.78166	22	+258	Galacturonic acid Glucose++ Arabinose +++ Xylose (tr)
Polysaccharide extracted with sodium hydroxide	2.77124	22	+265	Galacturonic acid Glucose+++ Arabinose++ Xylose (tr)

#### **Result and Discussion**

The monosaccharides identified in Cordia myxa (Table 1)were glucose, fructose and xylose. Paper chromatography indicated the presence of a disaccharide whose Rf value was similar to that of

sucrose, but this could not be confirmed because the quantity obtained of this sugar was not sufficient to subject it for detailed investigation. The constituent sugars present in the various polysaccharides obtained by methods discussed above were almost identical and were found to be galacturonic acid, glucose, arabinose, and xylose. It is, however, interesting to note that Kassem and his coworkers7 reported the presence of glucose, fructose and galacturonic acid alongwith an unidentified sugar after acid hydrolysis of mucilage of Cordia myxa.

Our investigation confirm the presence of glucose and galacturonic acid, but does not show the presence of fructose. We have obtained arabinose and traces of xylose. To clarify this difference in our observation and that of Kassem and his coworkers, we carried out several paper chromatographic separations of authentic samples of glucose, fructose, arabinose galacturonic acid and xylose, and found that arabinose and fructose have similar Rf values and it is, therefore, not possible to distinguish between them. Arabinose does, however, give a very specific colour (reddish brown) on paper chromatography when sprayed with Aniline Phthalate reagent (fructose give a light brown colour) and heated to 120° to develop the chromatogram. On this basis we are led to believe that one of the constituent sugar in the polysaccharides of Cordia myxa is arabinose and not fructose. This was further confirmed by taking their m.ps. and mixed m.ps. and preparing the osazone derivative of the sugar (arabinose) that was separated. The unidentified sugar as reported by Kassem and his coworkers is probably the xylose as found by us.

On the basis of our results, the polysaccharides of Cordia myxa are probably of polydisperse type, i. e. they comprise a family of polymers all built up on the same general plan with minor differences, such as extent of branching etc. in their fine structure.

We are at present engaged in the investigations of the antifungal and antibacterial properties of Cordia myxa fruits the results of which will be communicated later.

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