2 was refluxed with *N*, *N*-dimethylaniline a mixture of two products was obtained. One compound crystallised from benzene-light petroleum mixture to afford 4-methyl-4′-phenyl-6′ (H)-pyrano (2′, 3′: 6, 7) coumarin (5) as light yellow crystalline product, m.p., 119–20°; R_f 0·45; UV (nm), 226, 280 and 344 (3·92, 3·42 and 3·40 respectively); NMR, 2·43 (s, 3H, CH_3 in 4 position), 4·73 (d, J = 5 Hz, 2H, O–CH_2–CH=), 6·28 (s, 1H, *H*–3), 6·75–7·03 (m, 1H, $-CH_2-CH=$), 7·08 (d, J = 1·5 Hz, H–8), 7·28 (d, J = 1·5 Hz, *H*–5) and 7·20–7·45 ppm (m, 5H, C_6H_5). The second product crystallised from ethyl acetate-light petroleum mixture as colourless crystals to yield 4-methyl-6-hydroxy-7 (1-phenyl allyl) coumarin (3): m.p., 242–43°; R_f 0·38; UV (nm), 225, 245 and 330 (4·24, 4·19 and 3·87); NMR, 2·40 (s, 3H, CH_3 in 4 position), 4·80–5·01 (m, 1H, *H*–A), 5·04–5·30 (m, 1H, *H*–C), 5·30–5·52 (m, 1H, *H*–B), 5·94–6·32 (m, 1H, *H*–X), 6·25 (s, 1H, *H*–3), 6·90 (s, 1H, *H*–8), 7·1 (s, 1H, *H*–5), and 7·12–7·30 ppm (m, 5H, C_6H_5). This formed methyl ether (4), m.p., 180–81°; R_f 0·49; UV (nm), 228 and 250 (4·01 and 3·82 respectively); NMR, 2·42 (s, 3H, CH_3 in 4 position), 3·84 (s, 3H, CH_3O), 4·80–5·0 (m, 1H, *H*–A), 5·04–5·28 (m, 1H, *HC*), 5·29–5·52 (m, 1H, *H*–B), 5·93–6·33 (m, 1H, *H*–X), 6·25 (s, 1H, *H*–3), 6·91 (s, 1H, *H*–8), 7·01 (s, 1H, *H*–5) and 7·28 ppm (br s, 5H, C_6H_5.

The formation of (3) can be explained by a normal Claisen rearrrangement with favoured migration to the 7-position rather than the 5-position, whereas that of (5) can be explained as a result of subsequent oxidative cyclisation to give a six-membered ring. The type of neoflavene ring present in 5 has never been noted earlier in the Claisen rearrangement but is present in some naturally occurring neoflavonoids[7].

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5. UV data were recorded in methanol; figures before parenthesis represent λ_{max} values, whereas those in parenthesis are log ε values.
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STUDIES ON THE MAJOR ACIDIC POLYSACCHARIDE FROM THE SEED MUCILAGE OF *OCIMUM CANUM*

IN continuation of our studies concerning the structure-function relationship in polysaccharides[1, 2], we wish to report the isolation, and the physico-chemical characterization of the major acidic polysaccharide from the seed mucilage of *Ocimum canum*.

The acid-soluble portion of the mucilage of *O. canum*[1] obtained in improved yield (40%), by ethanol-sodium acetate precipitation, on DEAE-cellulose fractionation by stepwise elution successively with water, 0·05, 0·1, 0·2, 0·3, 0·4 and 0·5N sodium hydroxide gave one major and four minor acidic fractions. The nature and composition of these fractions are given in Table I. The compositions of these fractions were determined by the g.l.c. analysis, on 3% SE–52, of the trimethylsilyl ethers of the sugars present in the acid hydrolyzates of the respective fractions.

The major acidic fraction obtained in 55% yield was further purified by repeated DEAE-cellulose chromatography. Ultracentrifugal analysis of 0·4% solution of the polysaccharide in 0·1M sodium acetate-acetic acid buffer, pH 4·8, indicated a single sharp peak (sedimentation coefficient 11·1S). However free boundary electrophoresis of 1·0% solution of the polysaccharide in 0·05M sodium tetraborate, pH 9·26, indicated a trace amount of impurity having lower mobility than the major polysaccharide (electrophoretic mobility $-1·8 \times 10^{-4}$ cm^2 sec^{-1} volt^{-1}). The impurity was found to be strongly associated with the polysaccharide and attempts at further purification using sephadex G_{200} were unsuccessful, and it was found to be excluded through the column indicating its high molecular weight. The molecular weight as determined by light scattering technique was $3·03 \times 10^6$. The polysaccharide had an intrinsic viscosity of 22·6 dl/g and an uronic acid content of 34% as determined by the carbazole-sulfuric acid method[3].

Hydrolysis of the polysaccharide with 0·5M sulfuric acid, separation of the resulting sugars into neutral and acidic portions using Amberlite IR-120 (H^+) and Amberlite IRA–400 ($CO_3^=$), and g.l.c. analysis[4] of the neutral sugars as their alditol acetates on 3% ECNSS–M indicated the presence of xylose, arabinose, rhamnose and galactose in the molar ratio 9·7: 6·0: 7·7: 1·1. Paper chromatographic examination of the acidic sugars using 4 : 1 : 5 *n*-butanol, acetic acid, water (upper

TABLE I

Nature and sugar composition of the fractions obtained by the fractionation of the acid-soluble portion of the mucilage on DEAE-Cellulose

Fraction No.	Designation	Eluant	Yield (mg)/ 300 mg of the acid-soluble fraction	Physical form	Solubility in water	Neutral Sugar Composition*
O. canum						
1	OC0	water	5	white powder	soluble	man, gal, ara, rha. —
2	OC1	0·05N NaOH	165	white shining flakes	soluble	xyl, ara, rha, gal ; 10:6:7·5:1.
3	OC2	0·1N NaOH	50	light brown flakes	soluble	xyl, ara, rha, gal, glc, man; 10·5: 8:7: 2:0·9; 0·8.
4	OC3	0·2N NaOH	21	brown flakes	insoluble	xyl, ara, rha, gal, glc, man; 6:6:9:5: 5:4.
5	OC4	0·5N NaOH	33	pale brown granular powder	insoluble	xyl, glc, man; 1·5 ; 18·6:1; gal (trace)

* Except OC0, all other fractions contain galA and an undentified uronic acid derivative. In addition to these OC1 and OC2 contain a small amount of glcA.

layer) indicated mainly glacturonic acid, a small amount of glucuronic acid and trace unidentified uronic acid (R_{galA} 1·3). These were further confirmed by the g.l.c. analysis, on 3% ECNSS-M, of the corresponding alditol acetates obtained by reducing their methyl ester methyl glycosides with sodium borohydride followed by acid hydrolysis, sodium borohydride reduction and acetylation.

The polysaccharide on periodate oxidation[5] was found to consume 0·91 moles of periodate per mole of hexose unit and most of the xylose units remained unatacked, while all other sugars were destroyed except trace arabinose and rhamnose. Smith degradation studies[6], indicated the presence of a xylan backbone in the polysaccharide. Hakomori methylation[7] of the corresponding carboxyl-reduced polysaccharide followed by hydrolysis and g.l.c. analysis of the resulting partially methylated sugars as their alditol acetates[8] revealed the xylose exclusively as its 2-0-methyl and 3-0-methyl derivative, indicating that the xylan backbone of the polysaccharide has (1 → 4) glycosidic linkages in which some of the xylose units carry branch points at C-2 and some at C-3.

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