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Extraction and physicochemical characterization of water soluble galactomannans from Dichrostachys cineria seeds

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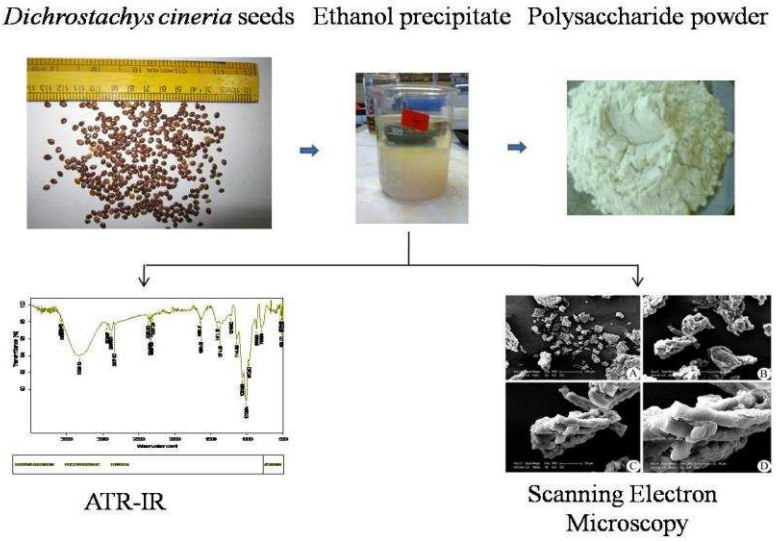
A B S T R A C T

Water soluble seed polysaccharide from Dichrostachys cineria seeds were extracted, yielded ~21% and characterized by several techniques such as SEM, GC, Attenuated total reflection Fourier Transform Infrared spectroscopy (ATR-IR), TGA, XRD and viscosimetry. GC/methylation analysis confirmed the linkage profile of galactomannan a 4-linked mannose polymer with galactose side chains attached at the C6 position. It has an estimated average degree of polymerization of 450 and a degree of branching of 0.65. The powder morphology was visualized using SEM. ATR-IR analysis showed the complexity the polysaccharide. The XRD confirmed the crystallinity and amorphous nature of polysaccharide. TGA showed the presence of three peaks weight loss events and decomposition patterns of the polysaccharide. The polysaccharide had an intrinsic viscosity of 3.42 dl/g. These results showed that the extracted polysaccharides are typically amorphous, thermally stable and these galactomannans might be expected to have wide applications in food and pharmaceutical industries.

Keywords: ATR-IR; Crystallinity; Galactomannan; Thermal stability; Viscosity; XRD



Graphical abstract of the polysaccharide extraction and characterization from the seeds of Dichrostachys cineria

1. Introduction

Galactomannans are the heterogeneous polysaccharides, mostly extracted from the legume endosperm (Suvakanta et al., 2014; Edwin & Andrade, 2002; Kapoor, 1992). Due to their viscous nature in aqueous media, emulsifying and gelling property, they dominate the food industry. Galactomannans are cheap, eco-friendly and non-polluting while production and in several applications. They are also using in different ways for human consumption (Srivastava & Kapoor, 2005; Smith & Montgomery, 1959). Galactomannas are used in food industries in various purposes of adhesives, food coating agents, preparation of ice creams and food processing industries (Stephen & Churns, 1995).

Galactomannans are varies in chemical structure, in the degree of galactose and mannose ratio, which are commonly formed by a linear chain of β-1, 4-D-mannopyranosides branched with single unit of D-galactopyranosides through α-1,6 linkages (Dey, 1978). This ratio depends on the source of seed and varies species to species of which influence the solubility and the chain stiffness (Wu et al., 2012; Vieira et al., 2007; Winter, Chien, & Bouckris, 1984; Dea & Morrison, 1975). The galactomannans are considered as multipurpose macromolecules basically neutral polysaccharides, pH changes would not affect them (Reid & Bewley 1979).

The galactomannans are largely used in thermoplastic, rubber industries and preparation of food industry, pharmaceutical industry as a drug carrier and emulsifier, cosmetics, pastes, toiletries industries, thickener in toothpastes, conditioner in shampoos, denture fixture powders, and textile industries. Also used for the sizing, finishing in printing industries, in crude oil drilling and explosives (Vendruscolo et al., 2009; Sharma et al., 2008; Schneider & Soster-Turk, 2003; Williams & Phillips, 2003), used in the preparation of nano-particles for drug delivery (Soumya, Swapankumar, & Abraham, 2010).

Legume (Leguminosae/ Fabaceae) provides three notable galactomannans which play an important role as commercial gums, namely guar gum (GG, Cyamopsis tetragonolobo, M/G ratio: 2:1), tara gum (TG, Caesalphinia spinosa, M/G ratio: 3:1) and locust bean gum (LBG, Ceratonia siliqua, M/G ratio: 3.5:1) (Dakia et al., 2008). Presently the international trends demands alternative source of seed gums (Joshi & Kapoor, 2003) and it is therefore important to introduce an alternative source and study their physicochemical properties for its applications in industrial fields. In this study we reported the yield of extraction, monosaccharide composition,

mannose/galactose ratio, intrinsic viscosity, thermal characterization was performed by thermogravimetric analysis (TGA), Attenuated total reflection Fourier Transform Infrared spectroscopy (ATR-IR) and X-ray diffraction (XRD).

2. Materials and methods

2.1. Materials

The pods of D. cineria were collected from the University of Hyderabad Campus, Hyderabad, India, in January, 2013. The collected plant material was properly identified using standard Floras, e-Floras. The processed specimens were mounted on herbarium sheet at University of Hyderabad Herbarium (UH), Hyderabad, for further reference. The seeds were manually separated and kept in a cool, dry place (Fig. 1).

2.2. Polysaccharide extraction and purification

Whole seeds were weighed and soaked in water (1:5 ratio). The seed coat, germ and endosperm was separated carefully and left to air dry, calculated the percentages of whole seed weight. A known weight of sample (~40 g) was dispersed in water (6 g/L) for 24 hr, fully hydrated sample was blended in a kitchen blender then spin at at 10000 x g for 30 min. The supernatant solution was kept for 4 h at 4^{o}C then filtered through 1.2 µm nylon membrane filters. The polysaccharide solution was precipitated by addition of absolute ethanol up to a final concentration of 36% ethanol (v/v). Precipitated polysaccharide was washed thoroughly with 70%, 80%, 90% and finally absolute ethanol then and left to dry under vacuum at room temperature, then calculated the yield and total sugars concentration (Dubois et al., 1956).

2.3. Scanning Electron Microscopy

The morphological features of powders were analyzed using a Philips/ESEM XL-30 scanning electron microscope (Amsterdam, Netherlands). The polysaccharide powder was mounted on stubs using double - sided stick tape and then mounted on the stub. The samples were

sputter coated to approximately 10µm thickness with a gold target in a vacuum coating apparatus. Scanned with an accelerating beam voltage of 1 Kv.

2.4. Methylation and GC-MS analysis

The Extracted polysaccharides were activated with Dry NaOH and methylated with 200 µl CH_3I (Ciucanu and Kerek, 1984; Isogai, Ishizu, and Nakano, 1985). 2-5 mg of powdered sample was dispersed in 2 ml of dried DMSO. NaOH (100Mg) powder was added to the solution. Sonicated the sample for 90 min and allowed to stand for 90 min. The methylated solution was dissolved in 3 ml of water and extracted with 4 ml of chloroform. The organic layer was washed with 3 ml of water, repeated 3 times, evaporated and remethylated to achieve complete methylation of all free OH groups. Methylated sample was then hydrolyzed with 2M trifluoroacetic acid (1 ml) at 121 ^{0}C for 1 hr, cooled, and evaporated at 40 ^{0}C. Partially Methylated sugars were then dissolved in dichloromethane (3-5 ml). The dichloromethane phase was washed with water and evaporated to dryness, and silylated with silylation reagent (15 µl) and pyridine (5 µl) and analyzed by GC-MS. GC-MS analysis was performed in Agile technologies gas chromatograph and Bruker, (Germany) mass spectrometer.

2.5. Attenuated total reflection Fourier Transform Infrared spectroscopy (ATR-IR)

The ATR-IR spectra of the polysaccharides were measured using a Bruker Platinum FTIR instrument (USA), with single reflection diamond crystal in reflective with gold coated optics. About 3-5 mg of powdered sample placed on the sample holder and then compressed under pressure to form a pellet. The Spectrums of the powdered form of galactomannans were measured in the wave number range of 4000-500 cm^{-1} using 32 scans and after measurements the spectrum baseline was corrected.

2.6. Thermo-gravimetric analysis (TGA)

TGA of galactomannan were performed using a METTLER Toledo DSC1, TGA/DSC1 thermogravimetric analyzers (Switzerland). About 4-6 mg of samples in ceramic crucible and then

sealed. Experiments were conducted under nitrogen atmosphere, at a heating rate of 10 ^{o}C min^{-1} over a temperature range of 10-500^{o}C. Thermal analysis was carried out under atmosphere at 10 mL/min flow rate. TGA curves are recorded.

2.7. X-ray diffraction

The crystalline structure of the purified galactomannans was obtained by X-ray diffraction analysis, patterns were obtained using Philips/PW1830 powder X-Ray diffractometer (Amsterdam, Netherlands) with Ni- filtered Cu Kα radiation, operating voltage at 40 kv and 25 mA. The spectra measurements were carried out in the angular range of 5-120^{o} 1^{o} (2θ)/min. To determine the crystalline, the total diffracted area and the area under the crystallinity peaks were determined by integration after correcting the data in absorption. The ratio of the crystalline area which account for that of the total was taken as the relative crystallinity.

2.8. Intrinsic Viscosity

The highest possible concentrations of gum solution were prepared by dissolving in 5 mM NaCl, 5 mg/ml. Stirred under magnetic stirrer for overnight and the galactomannan solutions allowed to hydrate slowly in the minimum volume. Later the clear solution was obtained and it was further filtered through membrane (0.2 µm) filter. The solutions were stored at 4^{0} for viscosity measurements. The intrinsic viscosity [η] of galactomannan solutions was measured as average values of four runs for each samples at 25^{o}C using an AMVn automated rolling ball micro-viscometer (Anton paar, Ostfildern, Germany) with programmable tube angle based on the principle of rolling ball time. It was expressed as η_{rel} with respect to water. All measurements were carried out at angles of 50^{o}.

$$\eta_{rel} = \eta/\eta_s$$

$$\eta_{sp} = (\eta - \eta_s) = \eta_{rel} - 1$$

$$\eta_{sp}/c = [\eta] + k'[\eta]^2 c \qquad 1$$

$$(\ln \eta_{rel})/c = [\eta] + k^n[\eta]^2 c \qquad 2$$

$$[\eta] = \{2(\eta_{sp} - \ln \eta_{rel})\}^{1/2}/c \qquad 3$$

The intrinsic viscosities of galactomannan samples were determined in water from experimental measurements of the dynamic viscosity of solutions with different concentrations of each component by the joint extrapolation of the Huggins (Eq. 1), Kraemer (Eq. 2) and single point (Eq. 3) standard relationships (Bohdanecky & Kovar, 1982). Where η and η_s are the viscosities of the solution and the solvent η_{rel} and η_{sp} are parameters of relative and specific viscosity, c is concentration and k' and k^n are constants.

3. Results and Discussions

3.1. Extraction and Purification

Extraction was carried out with ethanol precipitation and purification with gradient purification with ethanol. D. cineria seeds consists 44.6% seed coat, 30.03% endosperm and 21.37% germ (Table 1). The polysaccharide yield was 21.01.

3.2. Structural analysis with SEM

The structure of polysaccharide may be influenced by different extraction methods, purification and preparation of sample (Nep & Conway, 2010). Photographs were taken for each sample at various magnifications at x100, x500, x1000, x2000 and 50 µm scale. Samples having different sizes, distributed with smaller, smooth, fiber surfaced granules and exhibited tubular to irregular shaped particles having compact surfaces with numerous small pores separated each other due to their low density. The structure was observed similar with guar gum (Gong et al., 2012). The particles ranged from 10 to 100 µm. Earlier it has been reported that particle size and specific surface area influence the hydration behavior of gums, also influence their molecular weight and intrinsic viscosity (Qi, Ellis, & Ross-Murphy, 2006). The SEM results suggest that the galactomannan granules are having low density which influences the intrinsic viscosity of the gum (Fig. 2.)

3.3. Methylation and GC-MS analyses

Partially methylated alditol acetates analyzed by GC-MS revealed a polysaccharide total composition. The Mannose and galactose are the major monosaccharides present in the extracted polysaccharide. The total content of mannose and galactose are 63.2% and 29.6%. The polysaccharide containing nonreducing terminal units of Manp with 0.14% and Galp with 36.03 as well as 4-Manp with 26.65%, 4,6-Manp 37.76% and 4-Galp 2.98 (Table 2a). The methylation analysis results confirm the structure of the galactomannans as a 1-4 mannose linkages with galactose side chains attached at C6 position. The galactomannan contain minor amounts of other monosaccharides such as arabinose (Ara), glucose (Glc), rhamnose (Rha) and xylose (Xyl) (Table 2b). These minor components present in the galactomannan could be attributed to a more complex polysaccharide composition. The degree of polymerization of mannose residues for the galactomannan is within the range usually found for polysaccharide structures above 100 (Izydorczyk, 2005) showing of 450 and degree of branching of 0.65.

3.4. ATR-IR analysis

The ATR-IR spectra of D. cineria polysaccharide are presented in Fig. 3. A weak absorption peaks at 550-750 cm^{-1} was attributed to protein absorption which has small amount of protein or polypeptide (Xu et al., 2009). The major peaks appeared in three regions 3700-3000 cm^{-1} was attributed to hydroxyl (O-H) stretching vibration. The peak at 3000-2800 cm^{-1} could be attributed to -C-H stretching vibration of alkane. The absorption of an amino group was confirmed by peaks at 1650-1350 cm^{-1} (Anderson, 1949). The peak present at 1150-950 $cm^{-1}$was representing the - CO & C-O-Hstretching. The peaks at 870-810 cm^{-1} are due to skeletal stretching vibrations of polysaccharide and the characteristic absorption of mannose (Kato, Nitta, & Mizuno, 1973).

3.6. Thermogravimetric analysis (TGA)

TGA was performed to study the decomposition pattern and the thermal transition occurring in the course of heating under inert atmosphere of polysaccharides. Various thermal effects and enthalpy changes of polysaccharide was exhibited early endothermic event located between 90-100^{o}C attributed to water evaporation. The second weight loss region located between

290-330ºC is attributed to the degradation of the galactomannans (Fig. 4). Higher mannose/galactose ratio influences the thermal stability of the galactomannan (Ceraqueira et al., 2011).

3.5. X-ray diffraction (XRD)

X-ray diffraction pattern of seed galactomannan shown in Fig. 5. It has been observed by other workers that galactomannans has two scattering peaks at about 20º and 44º. (Cunha, De Paula, & Feitosa, 2007) which was well similar with our results. The X-ray diffraction patterns of polysaccharide sample having 4 major peeks noticed at 2θ - 20.02, 44.05, 52.05 and at 73.01. The broad peak at 2θ - 20.02 indicates that the powder samples are amorphous materials at room temperature (Joana et al., 2012; Pal, Mal, & Singh, 2007) which has a large degree of crystallinity in all samples and the reduction of water content.

3.7. Intrinsic viscosity

Intrinsic viscosity of D. cineria galactomannan was determined by Huggins, Kraemer and Single-point extrapolations to intrinsic viscosity (Morris, 1984) in 5 mM NaCl. The intrinsic viscosity $[\eta]$ of D. cineria galactomannan obtained at neutral pH by combined Huggins and Kraemer extrapolation (Fig. 6) was 3.42 ± 0.06 dl/g. The low intrinsic viscosity values of D. cineria polysaccharide indicated a high value of C which indicates a significant effect on the viscosity. It indicated that the galactomannan solutions were Newtonian in this concentration range and dissociation of intermolecular 'hyperentanglements' capable to form intermolecular associations. Due to its viscous nature, it could be an effective compound in food industry.

4. Conclusions

D. cineria seed galactomannan yielded 21.37% with high content of Man and Gal, being the M/G ratio 1.05, 4-linked mannans backbone to galactose side chains linked at the C6 position. Powder structure of polysaccharide may be influenced by different extraction methods, purification and preparation of sample. ATR-IR suggested that these galactomannas having a

small amount of protein or polypeptide. TG-DTA analysis the thermogram explained the thermal stability, water evaporation and enthalpy changes of galactomannans. XRD analysis confirmed the galactomannans having 4 major peeks, it indicates that the large degree of crystallinity, amorphous nature of the compound and reduction of water content. Due to its viscous behaviour it could be an effective and excellent alternative to the commercial galactomannans available in the market.

Acknowledgements

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References

Anderson, E. (1949). Endosperm mucilages of legume. Ind. Eng. Chem. Res. 41, 2887-2890.

Bohdanecky, M., & Kovar, J. (1982). Viscosity of Polymer Solutions. Elsevier, Amsterdam, The Netherlands.

Ceraqueira, M. A., Souza, B.W.S., Simoes, J., Teixeira, J.A., Domingues, M.R.M., Coimbra, M.A., & Vicente, A.A. (2011). Structural and thermal characterization of galactomannans from non-conventional sources, Carbohydarate Polymers, 83, 179-185.

Cunha, P. L. R., De Paula, R. C. M., & Feitosa, J. P. A. (2007). Purification of guar gum for biological applications, International Journal of Biological Macromolecules, 41, 324-231.

Dakia, P. A., Blecker, C., Roberta, C., Watheleta, B., & Paquoata, M. (2008). Composition and physicochemical properties of locust bean gum extracted from whole seeds by acid or water de-hulling pre treatment. Food Hydrocolloids, 22, 807-818.

Dea, I.C.M., & Morrison, A. (1975). Chemistry and interactions of seed galactomannans. Advances in Carbohydrte Chemistry and Biochemistry, 35, 241-312.

Dey, P.M. (1978). Biochemistry of plant galactomannans, Advances in Carbohydrte Chemistry and Biochemistry, 35, 341-376.

Dubois, M., Gilles, K. A., Hamilton, J. K., Rebers, P. A., & Smith, F. (1956). Colorimetric method for determination of sugars and related substances. Analytical Chemistry, 28(3), 350-356.

Edwin, G. A., & Andrade, T. C. (2002). Testing procedures for galactomannan purification, Polymer Testing. 21, 551-556.

Gong, H., Liu, M., Chen, J., Han, F., Gao, C., & Zhang, B. (2012). Synthesis and characterization of carboxymethyl guar gum and rheological properties of its solutions. Carbohydrate Polymers, 88, 1015-1022.

Joana T. Martins, Miguel A. Cerqueira, Ana I., Bourbon, Ana C. Pinheiro., Bartolomeu W.S. Souza., & António A. Vicente. (2012). Synergistic effects between k-carrageenan and locust bean gum on physicochemical properties of edible films made thereof. Food Hydrocolloids, 29, 280-289.

Joshi, H., & Kapoor, V. P. (2003). Cassia grandis Linn. f. seed galactomannan: Structural and crystallographical studies. Carbohydrate Research, 338, 1907–1912.

Kapoor, V.P. (1992). In: K.G. Mukerjee (pp. 87-114) (eds.), Current Concept in Seed Biology, Naya Prokash, Calcutta.

Kato, K., Nitta, M., & Mizuno, T. (1973). Infrared spectroscopy of some mannans. Agricultural and Biological Chemistry, 37, 433.

Morris, E. R. (1984). Rheology of hydrocolloids. In Gums and Stabilizers for the Food Industry 2, eds. G. O. Phillips, D. J. Wedlock & P. A. Williams. Pergamon Press, Oxford, 1984, 57-78.

Nep, E.I., & Conway, B.R. (2010). Characterization of grewia gum, a potential pharmaceutical excepient. Journal of experiments and Food chemicals, 1(1), 30-40.

Pal, S., Mal, D., & Singh, R.P. (2007). Synthesis and characterization of cationic guar gum: A high performance flocculating agent. Journal of Applied Polymer Science, 105(6), 3240-3245.

Qi, W. Ellis, P.R., & Ross-Murphy, S. B. (2006). Dissolution kinetics of guar gum powders-III. Effect of particle size, Carbohydrate Polymers, 64, 239-246.

Reid, J.S.G., & Bewley, J.D. (1979). A duel role for the endosperm and its galactomannan reserves in the germinative physiology of fenugreek (Trigonella foenum-graecum L.), an endospermic leguminous seed. Planta, 147, 145-50.

Schneider, R., & Sostar-Turk, S. (2003). Good quality printing with reactive dyes using guar gum and biodegradable additives. Dyes and Pigments, 57, 7-14.

Sharma, B.R., Dhuldhoya, N.C., Merchant, S.U., & Merchant, U.C. (2008). A Glimpse of Galactomannans. Sci. Tech Enterpr.

Smith, F., & Montgomery, R. (1959). The chemistry of plant gums and mucilages and some related polysaccharides. Reinhold Pub. Corp., New York

Soumya, R.S. Swapankumar, G., & Abraham, E.T. (2010) Preparation and characterization guar gum nanoparticles. International Journal of Biological Macromolecules, 46, 267-269.

Srivastava, M., & Kapoor, V.P. (2005). Seed galactomannans: an overview. Chemistry and Biodiversity, 2, 295-317.

Stephen A.M., & Churns, S.C. (1995). Food polysaccharides and their application. pp. 1-18. Marcel Dekker, New York.

Suvakanta, D., Narsimha, M. P., Pulak, D., Joshabir, C., & Biswajit, D. (2014). Optimization and characterization of purified polysaccharide from Musa sapientum L. as a pharmaceutical excipient. Food Chemistry, 149, 76-83.

Vendruscolo, C.W., Ferrero, C., Pineda, E.A.G., Silveira, J.L.M., Freitas, R.A., & Jimenez-Castellanos, M.R. (2009). Physicochemical and mechanical characterization of galactomannan from Mimosa scabrella; effect of dying method. Carbohydrate Polymers, 76, 86-93.

Vieira, I. G. P. V., Mendes, F. N. P., Gallao, M. I., & de Brito E. S. (2007). NMR study of galactomannans from the seeds of mesquite tree (Prosopis juliflora (Sw) DC). Food Chemistry, 101, 70-73.

Williams, P.A., & Phillips, G.O. (2003). Introduction to food hydrocolloids. pp. 1–19 In: G.O. Phillips, and P.A. Williams (eds.), Handbook of Hydrocolloids. CRC Press, New York.

Winter, W.T., Chien, Y.Y., & Bouckris, H. (1984). Structural aspects of food galactomannans. pp. 535-539 In: Gum and Stabilisers for the Food Industry, Vol. 2. G.O. Phillips, D.J. Wedlock, and P.A. Williams, (eds.), Pergamon, Oxford, U.K.

Wu, Y., Li, W., Cui, S.W., & Goff, H.D. (2012). A molecular modeling approach to understand conformation–functionality relationships of galactomannans with different mannose/galactose ratios. Food Hydrocolloids, 26, 359-364.

Xu, H. S., Wu, Y. W., Xu, F. S., Sun, H. X., Chen, F. Y., & Yao, L. (2009). Antitumor and immunomodulatory activity of polysaccharide from the roots of Actinidia eriantha. Journal of Ethanopharmacology, 125, 310-317.

Table 1

Physical composition of Dichrostachys cineria seed and polysaccharide.

Parameter	Range	Mean ± SD
Seed coat (%)	36.6-49.4	44.6 ± 7.15
Endosperm (%)	27.9-31.5	30.03 ± 2.15
Germ (%)	18.4-31.6	25.37 ± 6.35
Yield (%)		21.37 ± 2.1

Table 2

a) Glycosidic-linkage analysis of Dichrostachys cineria polysaccharide.

Linkage	Relative abundance (%)
Terminal-Manp	0.14 ± 0.12
4-Manp	26.65 ± 0.54
4,6-Manp	37.76 ± 9.68
DPm	450
Terminal-Galp	36.03 ± 8.10
4-Galp	2.98 ± 0.27

b) Monosaccharide composition (% mol) and total M/G of Dichrostachys cineria polysaccharide.

Man	Gal	Rha	Ara	Xyl	Glc	Total Man + Gal	Total (ug/mg)	M/G
63.2±0.5	29.6±0.9	1.1±0.2	4.1±1.1	1.2±0.3	0.8±0.2	760.6	814	1.05±0.05

Figures

Fig. 1. Seeds of Dichrostachys cineria

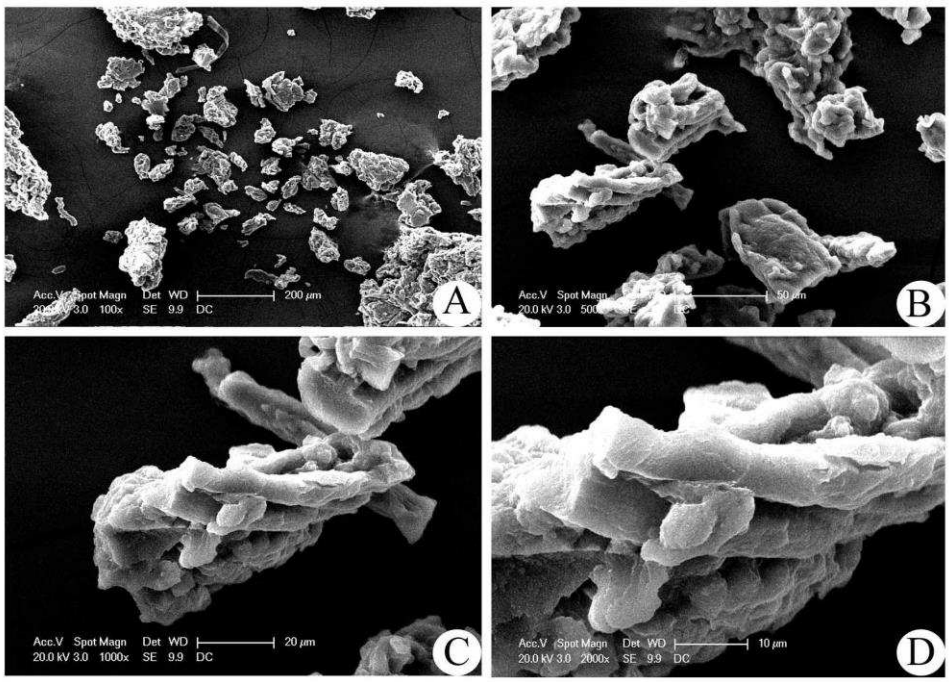

Fig. 2. SEM images of Dichrostachys cineria galactomannan powder A) x100, (scale 200 µm) B) x500 (scale 50 µm) and picture of individual granules C) x1000 (scale 20 µm) & D) x2000 (scale 10 µm) magnifications.

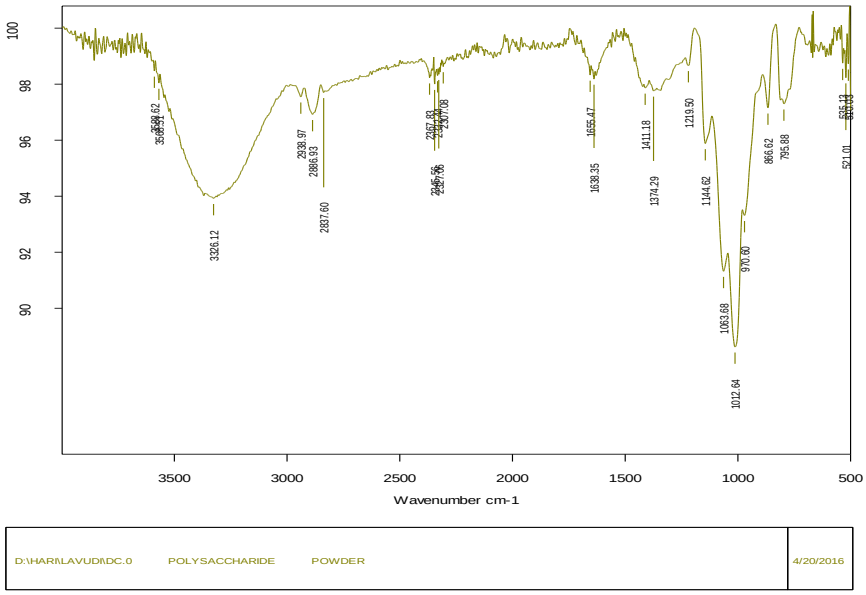

Fig. 3. FT-IR spectra of Dichrostachys cineria galactomannans showing the various absorption peaks.

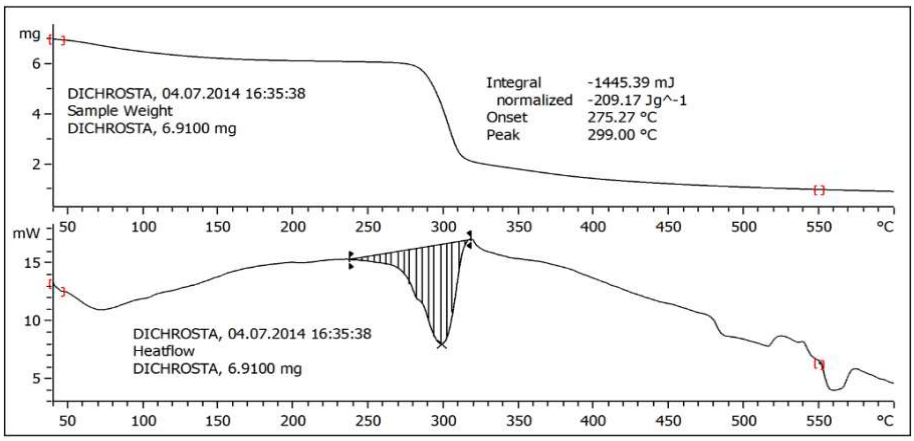

Fig. 4. TG-DTA Weight loss curve of Dichrostachys cineria galactomannan under inert atmosphere at a heating rate of 10 ^{0}C/min.

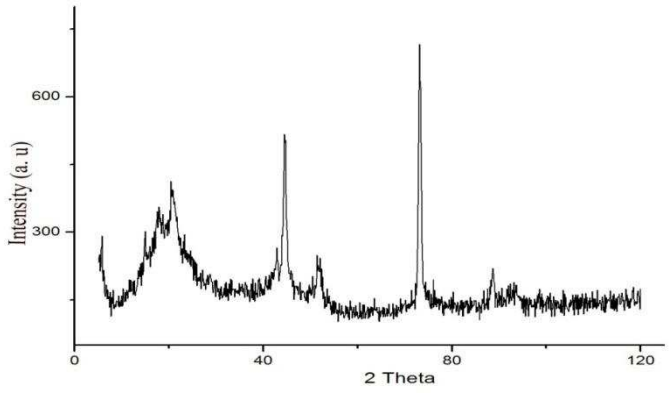

Fig. 5. XRD patterns of Dichrostachys cinerea galactomannan powder.

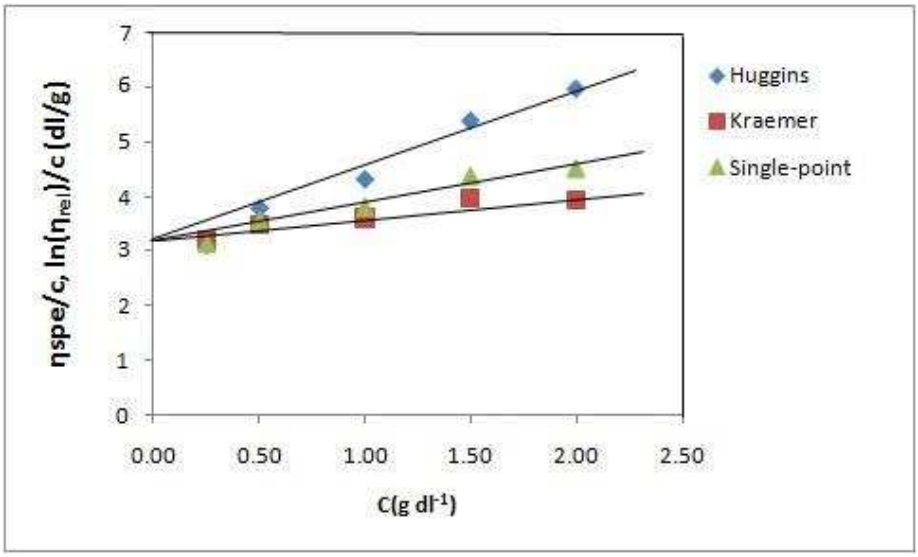

Fig. 6. Determination of intrinsic viscosity for Dichrostachys cineria galactomannan by combined Huggins (◊), Kraemer (■) and single-point (▲) extrapolation of respectively, η_{sp}/c, $\ln(\eta_{rel})/c$, and $\{2(\eta_{sp} - \ln \eta_{rel})\}^{1/2}/c$ to zero concentration.