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Research Article

OPTIMIZATION AND CHARACTERIZATION OF PURIFIED GUMMY POLYSACCHARIDE ISOLATED FROM *AEGLE MARMELOS* FRUIT PULP AS A NOVEL PHARMACEUTICAL EXCIPIENT

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ABSTRACT

Objective: Natural polysaccharides having an immense amount of attention in the recent years as pharmaceutical excipients in the pharmaceutical industry. Gummy polysaccharides, a as naturally occurring polymers are abundance in natural sources and less costly which makes them an ideal choice for use in food and pharmaceuticals. The fruits of Aegle *marmelos* are edible and have been recommended in ancient system of medicine for its various medicinal uses. In the present study the main objective was to illustrate the physicochemical, thermal and functional properties of polysaccharide obtained from *Aegle marmelos (AMPS)*.

Methods: A purified component of polysaccharide was isolated from *Aegle marmelos* fruit pulp *(AMPS)* by DEAE –Sephadex-50. Physicochemical, micrometrics and microbiological properties are determined. Elemental analysis, Scanning electron microscopy (SEM), X-ray diffraction spectroscopy (XRD), Differential scanning microscopy (DSC), Fourier-transformed infrared (FT-IR) and CHN Analysis Techniques were used to characterize the polysaccharide.

Results: Physicochemical, micrometrics and microbiological properties are significant as other natural excipients. Analytical data indicated that the major neutral sugar were α-D-glucose, β -D-glucose, Galactose shown in Osazone formation test of hydrolyzed sample . Elemental analysis of the polysaccharide has shown the contents of carbon, hydrogen and nitrogen contents to be 27.20, 4.07 and 0.62 (w/w %) respectively. SEM analysis suggests that the polysaccharide has irregular particle size, mostly seen in aggregates and fibrous in nature. The XRD pattern of the polysaccharide indicates both crystalline and amorphous structure. A DSC study of crude polysaccharide also shows amorphous and crystalline behavior of the polysaccharide.

Conclusion: The experimental work provides enough evidence to exploit this neutral biopolymer in food and pharmaceutical industry.

Keyword: *Aegle marmelos* polysaccharide (*AMPS)*, Bulk density, Angle of repose, Kawakita analysis, Scanning electron microscopy (SEM).

INTRODUCTION

In recent times, focus on plant research has increased all over the world and a large body of evidence has collected to show immense potential of plants used in various pharmaceutical application. [1] Plant derived polysaccharides have evoked tremendous interest due to their diverse pharmaceutical applications such as diluents, binder, disintegrants in tablet formulation, thickeners in oral liquids, protective colloids in suspensions, gelling agents in gels and bases in suppository. [2] These polymers of monosaccharide are inexpensive and available in a variety of structures with a variety of properties. They are highly stable, safe, non-toxic, non-carcinogenic and hydrophilic and gel forming in nature. Natural polysaccharides serve as an alternative to synthetic products because of local accessibility, environment friendly nature and lower prices compared to imported synthetic products. [3] Polysaccharides are also generally non-polluting renewable sources for the sustainable supply of cheaper pharmaceutical products. They have also been found useful in formulating immediate and sustained release preparations. [4] The extraction and characterization of non starch polysaccharide is an essential step in establishing their suitability as pharmaceutical excipients. *Aegle marmelos* Linn.(family: Rutaceae) is a perennial tree, wild in the Sub- Himalaya tract, central and South India, a plant of Indian origin having tremendous therapeutic potential is not fully utilized. [5, 6] The neutral oligosaccharides of the fruit pulp was characterized as 3-0-beta-Dgalactopyranosyl- L-arabinose, 5-0-beta-D-galactopyranosyl-L-arabinose, and 3-0-beta-Dgalactopyranosyl-D-galactose and the acidic oligosaccharides uronic acid.[5, 7] Literature survey reveals that comprehensive physicochemical characterization and evaluation of the purified polysaccharides isolated from fruit pulp of *Aegle marmelos (AMPS)* as pharmaceutical excipients has not been performed. Therefore, the objective of this study was to isolate and purify the polysaccharide from *Aegle marmelos* and examines the various pharmaceutical properties to assess its functionality as a pharmaceutical excipient. Specifically the study was performed to evaluate the physicochemical properties including flow characteristics, rheological properties, elemental composition and thermal properties. The isolated polysaccharide may provide an alternative to other natural polysaccharide or their synthetic counterparts to design and formulate suitable novel drug delivery system.

MATERIALS AND METHODS

Materials

Ripen *Aegle marmelos* fruits were obtained from the local market of Guwahati, Assam, India. DEAE- Sephadex A-50, Dextran was purchased from Hi-media, Mumbai, India. Glucose, d-glucoronic acid, was purchased from S.D fine chemicals, Mumbai, India. All other chemicals and solvents used were of analytical grade.

Extraction, isolation and purification of polysaccharides

Ripen fruit of *Aegle marmelos* were collected in the month of March, were cut into two after breaking the hard pericarp. Fruit pulp was scraped out, seeds and the large fibrous derbies of the fruit pulp were separated manually and dried at 50°C, then powdered and pass through Sieve no 20#. Then dried and stored in airtight container at room temperature.[8]Dried powdered fruit pulp (1 kg) was first defatted with petroleum ether and then extracted with methanol to remove saponins.[9]Then the marc were soaked in double distilled water (10 times) for 5-6 h, at 80°C untilled slurry was formed .The slurry was cooled and kept in refrigerator (4°C) overnight so that most of the un-dissolved portion was settled out. Material was squeezed in a muslin bag to remove marc from filtrate and then filtered through Whatman filter paper. The filtrate was concentrated in a rotary evaporator under reduced pressure at 60°Cuntil the volume reduced to one third of its original volume and centrifuged at 3000 rpm 15 minutes .Solution was cooled down to the room temperature and was poured into thrice the volume of acetone by continuous starring. The precipitate was washed three times with acetone and dried at 50 °C under vacuum (% of yield is 9.6% or 96 gm). The dried material was powdered and kept in a desiccators (30°C & 45% relative humidity. 1 g crude polysaccharide was dissolved in 100 ml distilled water, then deproteinated by using 5% trichloracetic acid (TCA) and obtained deproteinated polysaccharide

(*AMPS*) was dissolved with distilled water and scanned from 190 to 440 nm to evaluate the effect of deproteination. the polysaccharide was free of proteins as determined by scanning the UV Spectra at 260 nm and 280 nm. Then crude polysaccharide was subjected to DEAE- Sephadex A-50 column chromatography, washed with H_2O and eluted with 1.0 M NaCl solution**.** Most of the pigments were absorbed in the column. The elutes collected were concentrated under reduced pressure to an appropriate volume, and then dialyzed against distilled water. The retained portion was lyophilized to afford the total purified polysaccharide (yield 22.5 g).[10, 11, 12, 13,14]

Composition of the polysaccharide

Total sugar content was estimated by the phenol-sulfuric acid analysis using glucose as standard. The nature of the carbohydrate was confirmed by Molisch tests, Fehling's test and Iodine test. [15]Identification of monosaccharide unit was done by Osazone formation test of the hydrolyzed sample of polysaccharide. [16] The protein content was estimated by UV-VIS spectra and as per the method described by Bradford test using bovine serum albumin as standard**.**[17] Uronic acid content was determined by the carbazole-sulfuric acid method using d-glucoronic acid as standard**.**[18]

Physical characterization of the polysaccharide

Solubility test

The purified polysaccharide *(AMPS)* was evaluated for solubility in water, acetone, chloroform, and ethanol in accordance with the B.P specification.[19]

Loss on Drying

500 mg of *AMPS* powder was weighed and placed in a clean and neat china dish. It was kept in hot air oven at 105°C until a constant weight was obtained. The china dish was removed from the oven and again the weight of the polysaccharide powder was determined. [22]The moisture content was then determined as the ratio of weight of moisture loss to weight of sample expressed as a percentage.

Total ash and insoluble ash

2 g of *AMPS* was weighed accurately in a previously ignited and tarred silica crucible. The material was then ignited by gradually increasing the heat to 500-600°C until it appeared white indicating absence of carbon. It is then cooled in a desiccators and total ash in mg per gm of air dried material is calculated. To the crucible containing total ash, 25ml of 2M HCl was added and boiled gently for 5minutes and then about 5ml of hot water was added and transferred into crucible. The insoluble matter was collected on an ash less filter paper. This was then washed with hot water until filtrate is neutral and the filter paper along with the insoluble matter was transferred into crucible and ignited to constant weight. The residue was then allowed to cool and then weighed. [23] The percentage of acid insoluble ash was calculated from the weight of the sample taken.

Sulphated Ash Value

A silica crucible was heated to red for 10 min. and was allowed to cool in a desiccators and weighed. A gram of substance was accurately weighed and transferred to the crucible. It was ignited gently at first, until the substance was thoroughly charred. Then the residue was cooled and moistened with 1 ml of concentrated sulfuric acid, heated gently until white fumes are no longer evolved and ignited at 800 o C ± 25 o C until all black particles have disappeared. The ignition was conducted in a place protected from air currents. The crucible was allowed to Cool. A few drops of concentrated sulfuric acid were added and heated. Ignited as before and was allowed to cool and weighed. The operation was repeated until two successive weighing do not differ by more than 0.5 mg. [22]

Determination of pH

This was done by shaking a 1 % w/v dispersion of the sample in water for 5 min and the pH determined using a digital pH meter. The data present here is for triplicate determination. [23]

Micromeretic property of polysaccharide:

Angle of Repose

The *AMPS* powder (10 g) was accurately weighed and carefully introduced into a funnel clamped to a stand with its tip 10 cm from a plane paper surface. The powder was allowed to flow freely unto the paper surface. The height of the cone, *H* formed after complete flow and the radius of the cone, *R* were measured and used to calculate the angle of repose using the following equation:

$$\text{Angle of repose } (\tan\theta) = \frac{H}{R}$$

Bulk and tapped densities

The *AMPS* powder (10 g) was accurately weighed into a 100 ml measuring cylinder and without disturbing the cylinder the volume of powder was read to give the bulk volume. The measuring cylinder was then clamped to the USP I tapper of a USP tap density tester (Electro lab, model ETD-1020).The volume of the powder was read after every 50 taps up to a total of 300 taps when volume of powder was constant. This represents the tapped volume of the powder. The bulk density and tapped density was calculated using the following equations.

$$\text{Bulk Density } (\rho b) = \frac{\text{Weight of the Sample}}{\text{Bulk Volume}}$$

$$\text{Tapped Density } (\rho t) = \frac{\text{Weight of the Sample}}{\text{Tapped Volume}}$$

Compressibility index

The compressibility index was calculated as follows

$$\text{Compressibility}(C\%) = \frac{\text{Tapped density} - \text{Bulk density}}{\text{Tapped desity}} \times 100$$

Hausner ratio

Hausner ratio is a measure of flowability of the *AMPS* and is calculated using the following equation. A low Hausner ratio means that the polysaccharide powder has a high flowability. Hausner ratio above 1.25 indicates poor flow.

$$\text{Hausner ratio}(H) = \frac{\text{Tapped density}}{\text{Bulk desity}}$$

Kawakita analysis

The Kawakita equation is described by using the following equation. This equation describes the relationship between the degree of volume reduction of the powder column and the applied pressure. [24] The basis for the Kawakita equation for powder compression is that particles subjected to a compressive load in a confined space are viewed as a system in equilibrium at all stages of compression, so that the product of the pressure term and the volume term is a constant.

$$\frac{P}{C} = \frac{P}{a} + \frac{1}{ab}$$

Where C is the degree of volume reduction of a powder compact at pressure P (no of tapping). 'C' can be expressed as $C = V_0 - V_\infty / V_0$, where V_0 is the initial volume before tapping and V_∞ is the final tapped volume after 'n' number of tapping. The constants (a and b) can be evaluated from a plot of P/C versus P. A value of a is indicative of the total volume reduction for the powder bed, and b is a constant that is inversely related to the yield strength of the particles. The data from this study were modeled via the Kawakita equation in an attempt to evaluate the relationship between the volume reduction and applied pressure for under studied isolated polysaccharide as pharmaceutical binder.

Swelling Index

This was done by taking 1.0 g quantity of *AMPS* in a 15 ml plastic centrifuge tubes and the volume occupied was noted. Ten milliliters of distilled water was added to it and the content was mixed on a

vortex mixer (Lab line Equipments, India) for 2 min. The mixture was allowed to stand for 10 min and immediately centrifuged at 1000 rpm on a bench centrifuge (Remi, India). The supernatant was carefully decanted and the volume of sediment was measured. [25] The swelling index was computed using the following equation.

$$S=\frac{(V2-V1)}{V1} \times 100$$

Where S is the % swelling capacity, V2 is the volume of the hydrated or swollen material and V1 is the volume of the material prior to hydration. The experiment was repeated by using 0.1 N HCl and Phosphate buffer 7.4 in water.

Viscosity and Effect of ageing on viscosity of polysaccharide

AMPS sample was dissolved in distilled water (1% w/v) by stirring on magnetic stirrer (for 3 h and then centrifuged (C-24 BL, REMI Elektrotechnik Limited) for 25 min at 25 ◦ C at a speed of 2500 rpm to remove insoluble matter. Effect of shear rate (rpm) was investigated on viscosity using spindle S62 by varying the shear rate between 0 to 30 rpm. The change in viscosity was recorded and the viscosity-shear rate profile was plotted. The viscosity of a 1.0% w/v dispersion of the *AMPS* read at shear rates between 0 to 30 reciprocal seconds and at 23°C using a Brookfield viscometer (LVDV-E) (Brookfield Engineering Labs, Stoughton-USA). Spindle 62 was used and 3 minutes was allowed for stabilization of the readings before the viscosity was read. The experiment was repeated by using 0.1 N HCl, normal saline (0.9% NaCl) and Phosphate buffer 7.4 in water.

To study the effect of ageing on viscosity of *AMPS,* the above was stored in a controlled humidity and temperature (75 % RH and 40°C) environment for 180 days after which the viscosity experiment was repeated and the average results recorded. [26]

Microbial quality

The total aerobic viable count and fungal count of AMPS was determined by the pour-plate method. [27, 28] In determining the presence of pathogenic bacteria and fungus in the polysaccharide, 0.1 g of the powder was dissolved separately in 10 ml of sterile water and 1ml of the solution was inoculated into a previously stabilised casein soya bean digest agar and Sabauraud plus streptomycin agar respectively. Four specific pathogens viz. *Escherichia coli, Staphylococcus aureus, Pseudomonas aeruginosa*, *Salmonella spp*. were checked for their presence along with total aerobic bacterial count and combined yeast and mould count. The inoculated agars were incubated at 37°C for 48 hours and growth of specific organisms depending on the selective media that was used was read as present or not present. All the experiments were done in triplicate.

Elemental Analysis

Elemental analysis of carbon, hydrogen and nitrogen was carried using a Perkin Elmer 2400 Semis II CHN analyzer. Accurately weighed 0.5 mg of sample was heated to 1150 °C and the corresponding element was determined by using thermal conductivity detector. [29]

Scanning Electron Microscopy (SEM)

The morphological features of the *AMPS* were studied with HITACHI, S-3600 N Scanning Electron Microscope. The dried sample was mounted on a metal stub and sputtered with gold in order to make the sample conductive, and the images were taken voltage of 20 KV at 500x magnification.

Differential Scanning Calorimetry (DSC)

A differential scanning calorimetry (JADE DSC, Perkin Elmer, and U.S.A) was used to study the thermal properties of the *AMPS*. The polysaccharide was scanned in the temperature range of 50-220°C under an atmosphere of nitrogen. The heating rate was 20°C/min, followed by a cooling cycle back to 30°C at the same rate.

X-ray powder diffraction (XRD)

X-ray diffraction (XRD) patterns of the *AMPS* were analyzed using a Siemens D5000 X-ray diffractometer (Siemens, Munich, Germany). Powder sample, packed in rectangular aluminium cells, illuminated using CuKα radiation (λ = 1.54056 Å) at 45 kV and 40 mA. Samples were scanned between diffraction angles of 5° to 65°C 2θ. Scan steps of 0.1 were used and the dwell time was 15.0 Sec. A nickel filter was used to reduce the Kβ contribution to the X-ray signal. The'd' spacing was computed according to Bragg's law of diffraction.

Fourier transform-infra red (FTIR)

The Fourier transform-infra red (FTIR) spectrum of the sample was recorded in an IR spectrometer (Bruker Alpha FTIR). Triplicate measurements were made, and the spectrum with the clearest identifiable peaks was chosen.

Statistical analysis

Data were expressed as mean ± standard deviations of three replicated determinations.

RESULT & DISCUSSION

It is well known that the polysaccharide is mixture of a number of different macromolecular substances and the yield and composition of polymer can vary depending on the methods of isolation. [30]

The yield of total isolated purified polysaccharide (*AMSP)* was 2.25 % of the raw material. AMPS showed negative Fehling's reagent and iodine-pottassium iodide reactions, indicating that it did not contain reducing sugar and starch type polysaccharide. The total carbohydrate polysaccharide content was estimated to be 93±1.3 %. Monosaccharide units was α-D-glucose, galactose seen in the osazone formation reaction.

The UV-Vis spectra showed that the polysaccharide had an absorption peak at 190 nm only, which is the characteristic UV absorption peak for a polysaccharide. There was no absorption at 260 and 280 nm, indicating that the polysaccharides contained no protein or polypeptide. Bradford test was confirmed the absence of protein in AMPS. The uronic acid content in AMPS was found to be 4.3%.

The polysaccharide is sparingly soluble in water and practically insoluble in ethanol, acetone and chloroform. The results of the swelling characteristics show that *AMPS* has high swelling index suggesting that the polysaccharide may perform well as binder/disintegrant/ matrixing agent. The swelling was highest in 0.9% NaCl & water followed by phosphate buffer and least in 0.1 N HCl. The results indicate that the polysaccharide is a pH responsive polymer and may find application in controlled release formulations. [31]

The moisture content of *AMPS* was low. This knowledge is essential for designing and optimizing many process involved in production using these polysaccharide like drying, packing and storing. Given suitable temperature moisture will lead to the activation of enzymes and the proliferation of micro organisms, thereby affecting the shelf life of most routine formulations. Therefore, the low moisture content value of *AMPS* indicates its suitability in formulations containing moisture sensitive drugs.[32]

The total ash and acid insoluble ash value of *AMPS* was found to be 0.83±0.025% and 0.085±0.03%w/w respectively. Ash values reflect the level of adulteration or handling of the drug. Adulteration by sand or earth is immediately detected as the total ash is normally composed of inorganic mixtures of carbonates, phosphates, silicates and silica. Therefore, the low values of total ash and acid insoluble ash obtained in this study indicate high level purity.

Knowledge of the pH of an excipient is an important parameter in determining its suitability in formulations since the stability and physiological activity of most preparations depends on pH. A 1% w/v solution of *AMPS* in water gave a pH of 6.3. The neutral pH of *AMPS* implies that when used in uncoated tablets, it may be less irritating to the gastrointestinal tract. It may find useful application in formulation of acidic, basic and neutral drugs.

The bulk and tapped density gave an insight on the packaging and arrangement of the particles and the compaction profiles of the material. Compressibility value upto 15% usually result in good to

excellent flow properties and indicate desirable packing characteristics. Compressibility index above 25% are often sources of poor tabletting qualities. Between these two value less than optimum performance might be anticipated and require modification of the formulation during process development. Results in the table 1 of *AMPS* would be accepted to have the moderate flow and compressibility property.

Kawakita plot is used to analyze the behavior of powder from the bulk density state to the tap density state. The constants 'a' and 'b' of Kawakita plot were determined from the slope and intercept of graph of n/c versus number of tapping. The value of 'a' indicated compressibility or densification due to tapping and 'b' as rate of achieving final packing. The high value of 'a'(0.341) and small value of 'b'(0.023) indicated poor flowability and higher cohesiveness. Hence, from the results of Kawakita plot, *AMPS* polysaccharide powder had a higher compactability. According to the published results, the compactability and cohesiveness values obtained indicate fair flowability and moderate cohesiveness. [33, 34]

Physicochemical properties of the *AMPS* are summarized in Table 1.

Table 1: Physicochemical properties of purified polysaccharide of *AMPS*

Parameters	Results (Mean ± SD)
Solubility	Slightly soluble in cold water, soluble in hot water, insoluble in ethanol, acetone and chloroform.
Melting point	Decomposed over 245°c
Swelling Index (% v/v)	
Water	525±16.7%
0.1 N HCl	253.6±18.2%
Phosphate buffer (pH 7.4)	404.2±9.55%
In 0.9% NaCl Solution	564.6±9.55
Loss on Drying (% w/w)	3.5 ± 0.03
Total ash (% w/w)	0.83±0.025%
Acid insoluble ash (% w/w)	0 .085±0.001%
Sulphated Ash Value	0.25±0.05%
Total carbohydrate content	67.8±2.27
Total polysaccharide content	93±1.3%
pH	6.4 ± 0.01
Angle of repose (degree)	35.34±2.07
Bulk density (g/ml)	0.71±0.05
Tapped density (g/ml)	0.86±0.004
Compressibility index (%)	17.06±0.53
Hausner ratio	1.2±0.01
Kawakita plot	Slope a b r
	2.936 0.341 0.023 0.99
Microbial quality	1.22 ± 0.21
Total viable aerobic count (cfu/gm)	0.42×10^3
Total fungi count	< 100
Escherichia coli	Absent
Staphylococcus aureus	Absent
Pseudomonas aeruginosa	Absent
Salmonella Spp	Absent

The flow behaviour and the effect of ageing on the viscosity of a 1.0 % w/v aqueous dispersion of the polysaccharide powder are shown in Figure 1. The viscosity of the polysaccharide dispersion decreases with an increase in shear rate. This is indicative of pseudoplastic or shear thinning behaviour. At high shear rates, the decrease in viscosity can be attributed to a decreasing number of chain entanglements. [35]Viscosity of the polysaccharide also changes with reference to the pH of the solution, saline and ageing. There is no very significant difference in the viscosity after the colloidal solution of the polysaccharide powders stored in humidity chamber.

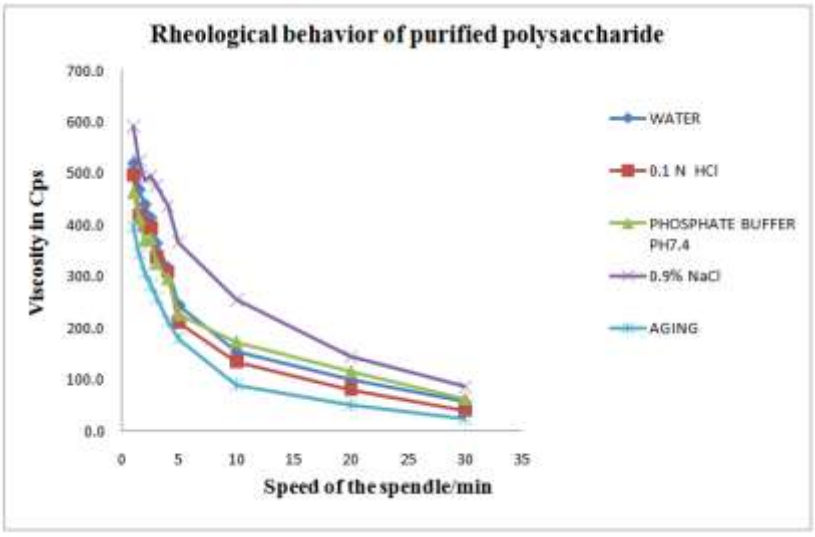

Fig. 1: Flow behavior of a 1% w/v dispersion of *AMPS* powder in distilled water, 0.1 N HCl, Phosphate buffer pH 7.4 at room temperature and the effect of ageing on viscosity (n=3, mean ± SD)

The microbial quality of the polysaccharide was assessed to determine their acceptability for use as pharmaceutical excipients in oral formulations. The total viable aerobic count and fungal count of the *AMPS* was lower than that of the prescribed limit of European pharmacopoeia. The purified polysaccharide did not contain *E. coli, Pseudomonas aeruginosa, Salmonella spp.* and pathogenic Staphylococci. Microbial quality of purified polysaccharide was satisfactory and met the European Pharmacopoeia. [22, 36]

Scanning electron micrographs (SEM) of *AMPS* are shown in Figure 2, 3. The micrographs of the *AMPS* provide the surface morphology of the polysaccharide. The particles are mostly seen in aggregates of irregular shapes and dimensions which are fibrous in, gum like mass devoid of crystalline structure. The shape and structure or surface topology of the polysaccharide may be affected by the method of extraction and purification or preparation of the product. [37]

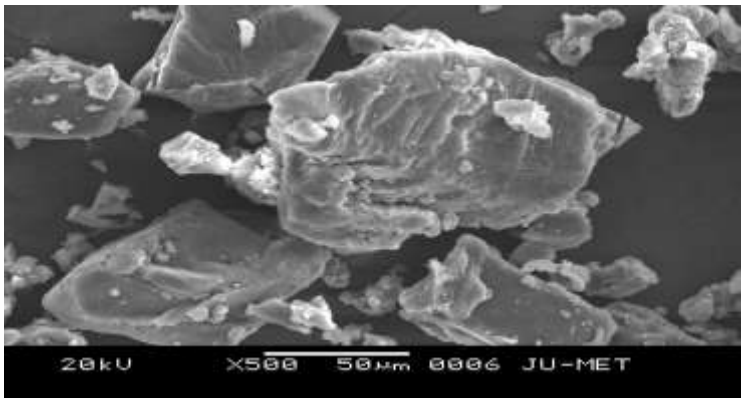

Fig. 2: Scanning Electron Microscopy of the purified of *AMPS.*

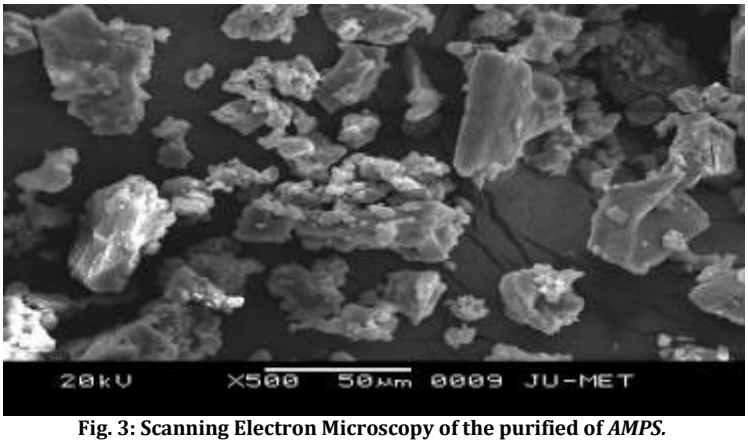

Fig. 3: Scanning Electron Microscopy of the purified of *AMPS.*

Differential Scanning Calorimetry (DSC) was used to measure the occurrence of exothermal or endothermal changes with an increase in temperature. Figure- 4 shows the DSC curve of the polysaccharide. Glass transition temperature 220°C and there was no melting peak. The endothermic transition is indicative of moisture loss in the sample. The onset peak and conclusion temperature of phase transition were observed to be 229.91°C. The weight loss onset (representing the onset of oxidation or decomposition) of 250ºC suggests that the polysaccharide has good thermal stability. Peak is relatively sharp indicating relative purity of the sample [38] the result implies that AMPS may structurally be stable and good thermal stability.

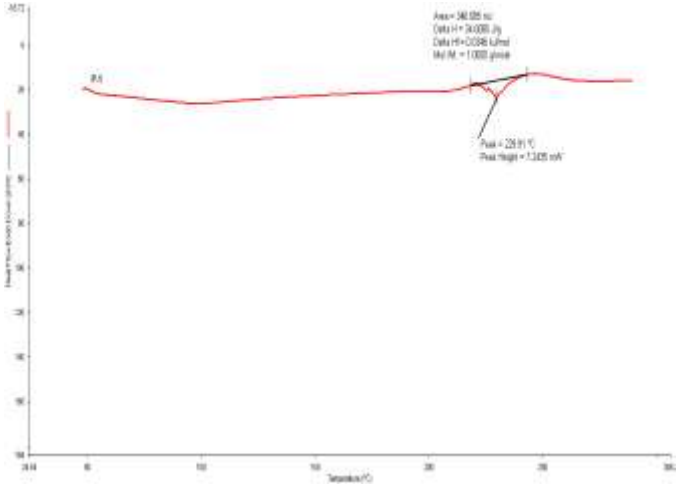

Fig. 4: DSC of purified polysaccharide of *Aegle marmelos* (*AMPS)*

The X-ray diffraction pattern of *AMPS* is shown in Figure 5. The sample shows peaks at approximately 28º, 32º, 42º and 45º 2θ. However, other peaks are very weak and unresolved or are shoulders on more intense peaks. The result of the XRD confirms that of the DSC which shows that, *AMPS* exhibits both crystalline and amorphous portion. [39]

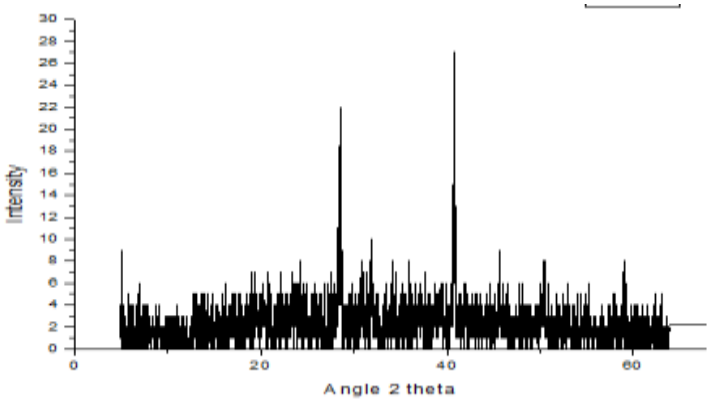

Fig. 5: X-ray diffraction of *AMPS.*

The IR spectrum is shown in Figure 6. Peaks at 3246.85 cm^{-1}due to -OH stretching of primary alcohol. The absorption peaks at 2976 cm^{-1} and 2889 cm^{-1} are indicative of –CH stretching vibration of methyl group. The absence of significant aromatic stretches in the 1739 cm^{-1} region and the weakness of the stretches imply that there is modest amount of cross linking by peptides. The bands at 1607 cm^{-1} is characteristic of C=O of aldehyde. Peak at 1374cm^{-1} is due to symmetrical deformation of -CH_2 and C-OH group. Weak bond at 769.38 cm^{-1} due to the r contribute to the ring stretching and ring deformation of α-D-(1-4) & α-D-(1-6) linkage This is all consistent with a polysaccharide structure that is neither starch nor cellulose, but has some peptide cross links and some amino sugars[40,41,42].

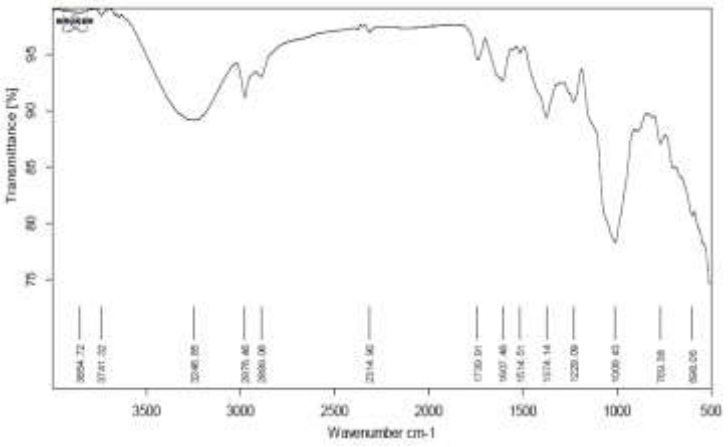

Fig. 6: FTIR of *Aegle marmelos* polysaccharide *(AMPS)*

Analysis of elemental (C, H, N) composition of the purified polysaccharide indicated C, H, and N contents to be 27.20, 4.07 and 0.62 (w/w %) respectively. The ratio of carbon to hydrogen indicating the polysaccharide composition as per the earlier report . [43]

CONCLUSION

The results obtained in this study were established for the first time regarding the physicochemical & structural characteristics of the purified polysaccharide from the fruit of *Aegle marmelos (AMPS).* Studies indicated that the purified polysaccharide with such properties have therefore been used as polymer in novel drug delivery system to prolong the drug release such as in the formulation of mucoadhessive, gatroretentive drug delivery system. The high thermal stability of the polysaccharide indicates that it can be used as pharmaceutical excipient even under conditions of high thermal stress. The relative abundance and easy availability of *AMPS* may serve as an alternative to present available pharmaceutical excipients.

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